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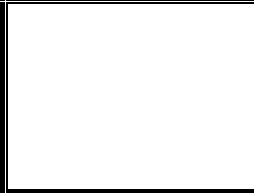
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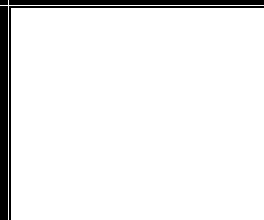
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RESEARCH ARTICLE

A REVIEW ON IMPLEMENTATION OF MPPT BASED PHOTOVOLTAIC CONVERTER SYSTEM WITH PSO.

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Abstract

Maximum power point tracking (MPPTs) play a vital role in photovoltaic (PV) power systems as it maximize the output power & efficiency of PV systems. MPPTs are used to find and maintain operation at the maximum power point. Title presents in detail implementation of PSO MPPT using buck-boost converter. Some results such as current, voltage and output power for various conditions can be recorded. The simulation is possible to carried out in MATLAB/SIMULINK software. The results can be obtained and compared with the theoretical operation which confirm the reliability and performance of the proposed model. It is more efficient, accurate, used rapid and low cost technique without need for complicated mathematical operations. This paper presents Particle Swarm Optimization and Perturbation & Observation techniques to find the optimum operating parameters of a solar photovoltaic panel under varying atmospheric conditions. The terminal voltage, current and corresponding duty-cycle, at which the DC/DC converter should be switched to obtain maximum power output, are determined. Simulation of maximum power point tracking of a solar panel is done in MATLAB. It is observed from the results that the particle swarm optimization based algorithm can track the maximum power point for the whole range of solar data (irradiance and temperature) and has high conversion performance.

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Introduction:-

Solar panels collect the sun's energy and convert it to electrical energy. Unfortunately, the solar output is not consistent throughout the day due to clouds and the sun's angle relative to the position of the solar panel. Addition to this, the intensity of the sun varies according to season and geographical location. Moreover the characteristic curve of a solar cell shows a nonlinear voltage-current relationship [1]. Therefore, a controller i.e. the maximum power point tracker (MPPT) is an crucial part of a photovoltaic (PV) system, to find automatically the maximum power operating point at all environmental conditions and then power the PV system to operate at the point (MPP), to ensure the optimum use of the available solar energy [1-2]. This function is realized by suitably controlling the power processing circuit that is exactly always used as an interface between the PV generator and the load or the

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energy accumulator. Peak Power Tracking algorithms provide the theoretical means to achieve the MPP of solar panels, these algorithms can be realized in very effective manner. In the last few years, several MPPT techniques have been technologically advanced to maintain the PV arrays operating at their MPPs and have been suggested various examples such as the Perturb and Observe (P&O) method, the Incremental Conductance (IC) method, the Artificial Neural Network method, the Fuzzy Logic method [3-5], etc.... It became clear that perturb and observe (P&O) technique was widely used for its ease of implementation and tracking efficiency [6]. This paper present an adaptive perturb and observe MPPT for a photovoltaic module connected to the buck-boost converter (DC-DC converter). The DC-DC converter is able to draw maximum power from the PV module for a given solar radiation level and environment temperature by modifying the duty cycle of DC-DC converter. This paper offers the method to track the MPP for a PV module using Perturb & Observe (P&O) and Particle Swarm Optimization Algorithm (PSO) technique at varying irradiation and temperature. The PSO is a swarm intelligence-based algorithm used to find the global optimum solutions. The reasons why PSO has added the popularity is because it has only a very few parameters that need to be modified. Although PSO is still in its early stages, it has been used across a wide range of applications. On the other hand P&O method has simple structure and high reliability. The assumed techniques find the optimal electrical operating points and corresponding duty cycle at which the maximum power can be transferred. The performance of the proposed MPPT methods is tested by simulation at different irradiation and temperature using MATLAB. Solar panel specifications are used for simulation. To focus the proposed system performance, irradiation and temperature variations were applied in steps. The results are analyzed and compared.

Generalised Block Diagram:-

A. System Design

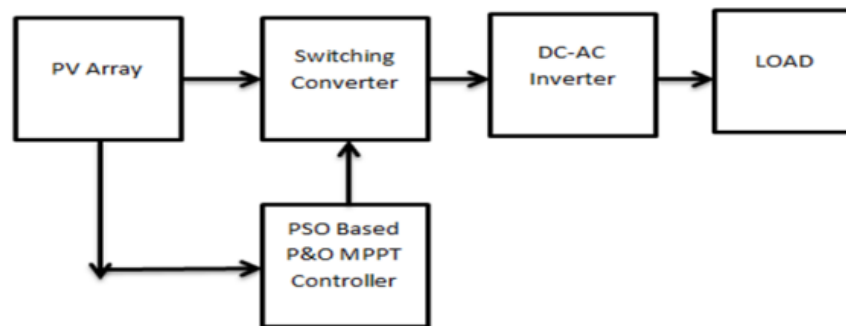


Fig.1:- System Design

The basic design for the photovoltaic system peak power tracker consists of the solar Panel, DC-DC converter (Buck- Boost) connected in a series, the MPPT controller, Voltage and current sensors and the load. This model is known to have better accuracy when the irradiance varies slowly that allows for a more accurate prediction of PV system performance. The Peak Power Tracker is a controlled DC/DC buck-boost converter used by a photovoltaic power system. The microprocessor tries to maximize the output power from the solar panel by controlling the conversion ratio of the DC/DC converter to keep the solar panel operating at its MPP [13]. A DC-DC converter acts as an interface between the load and the module. The Buck-Boost mode DC/DC converter is the most important type of switching regulator. In this converter, the buck and boost topologies are combined into one. The buck-boost converter steps the voltage down when the duty cycle is less than 50% and steps it up when the duty cycle is greater than 50%. The Peak Power Tracker uses an iterative approach to finding this constantly changing MPP [13].

Modelling of Solar Cell:-

In this study the five parameter model (The single diode model for solar cells figure 2) is a very proper choice. Firstly, because most times the results have a high degree of uniformity with data, secondly, as they are not too complex, it is relatively simple to implement and analyze them [7-8]. The equivalent circuit consists of a photocurrent, a diode, a parallel resistor expressing a leakage current, and a series resistor describing an internal resistance to the current flow, as shown in Figure 2.

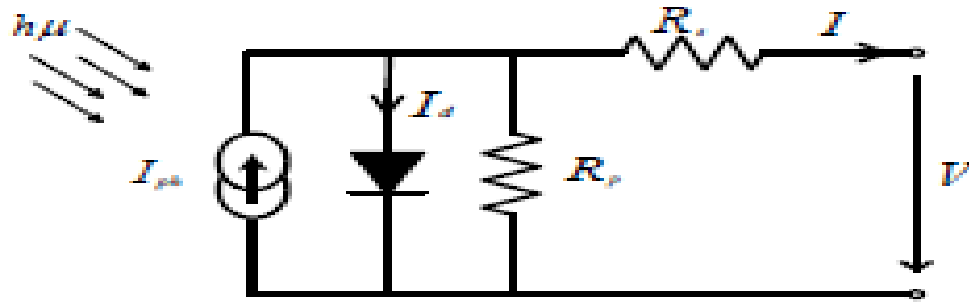


Fig. 2:- The equivalent circuit of practical PV cell.

The characteristic equation I-V is given by:

$$I = I_{ph} - I_0 \left[\exp \left(\frac{V + IR_s}{AV} \right) - 1 \right] - \frac{V + IR_s}{R_p} \quad (1)$$

Where,

I_{ph} : Photo generated current.

I_0 : Saturation current of diode.

R_s : Cell series resistance.

R_p : Cell parallel resistance.

A : Diode quality factor.

V : Thermal voltage.

Here, the PV model parameters; I_{ph} , I_0 , R_s and R_p are determined based on electrical parameters; I_{sc} , I_{mp} , V_{mp} , V_{oc} and A . The aim is to find the model parameters such that the resulted I-V curve accurately matches the experimental curve. These parameters are obtained by solving the fundamental equation (1) for the key points. The values for I_{ph} , I_0 , R_s and R_p are then determined through an iterative procedure [9-10].

Characteristics of Solar Cell:-

Several Cells are connected in series forms a PV panel/module whereas several PV modules are linked together to form PV arrays. If panels are in series current through the cell is constant and voltage across it increases while on the other hand when panels are in parallel current through the cell increases and voltage remains constant.

The I-V and P-V curve of a module is shown in the Fig. For a certain value of voltage and current the panel provides a maximum output. The output power of the panel increases as the module voltage increases, it reaches to a peak (called as peak power or maximum power point (MPP) in the module) and drops as the voltage approaches to the open circuit voltage. The peak power or MPP is defined at standard test condition (STC) of irradiation of 1000 W/m² and 25 °C module temperature, but these condition does not exist for the most of the time, due to which, normally, the module output power will be less than the peak output power.

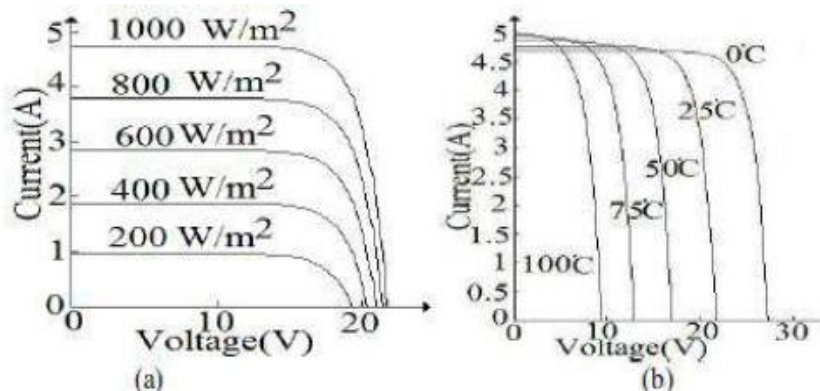


Fig.3:- Waveforms showing the effect of (a) Radiation and (b) Temperature on I-V characteristics

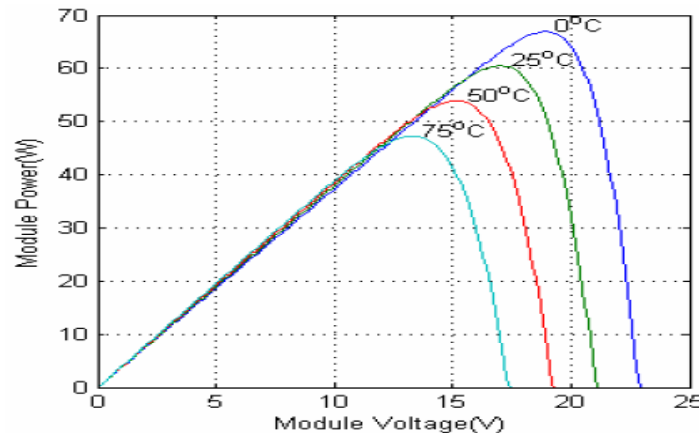


Fig.4:- PV curves at various temperatures and Constant irradiation.

In addition to this, the output current and the voltage are affected by the variations in the irradiation and temperature. Fig.4 and Fig.5 shows the waveforms for the effect of irradiation and temperature on I-V characteristics of a solar cell.

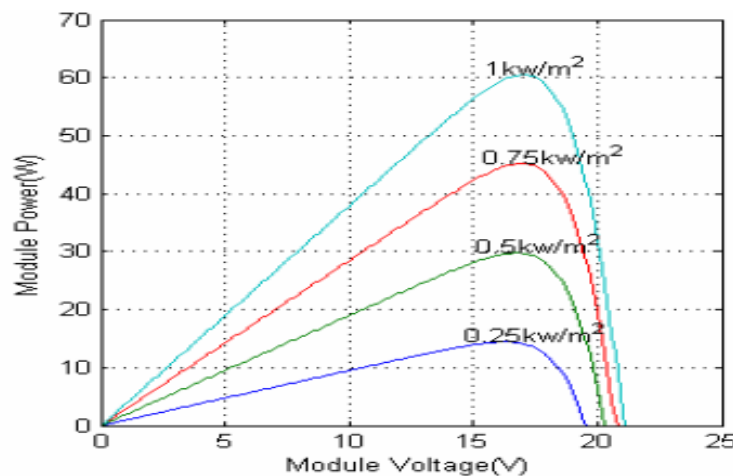


Fig.5:- PV curves at various irradiation levels and constant temperature

Under different conditions, there is a unique point on the curve, called the maximum power point (MPP), at which the photovoltaic cell operates with maximum efficiency and produces maximum output power.

MPPT Using Perturb and Observe Method (P&O):-

The perturb & observe (P&O) algorithm, also known as the “hill climbing” method, is very popular and the most commonly used in practice because of its simplicity in algorithm and the ease of implementation. The concept behind the “perturb and observe” (P&O) method is to modify the operating voltage or current of the photovoltaic panel until you obtain maximum power from it. For example, if increasing the voltage to a panel increases the power output of the panel, the system continues increasing the operating voltage until the power output begins to decrease. Once this happens, the voltage is decreased to get back towards the maximum power point.

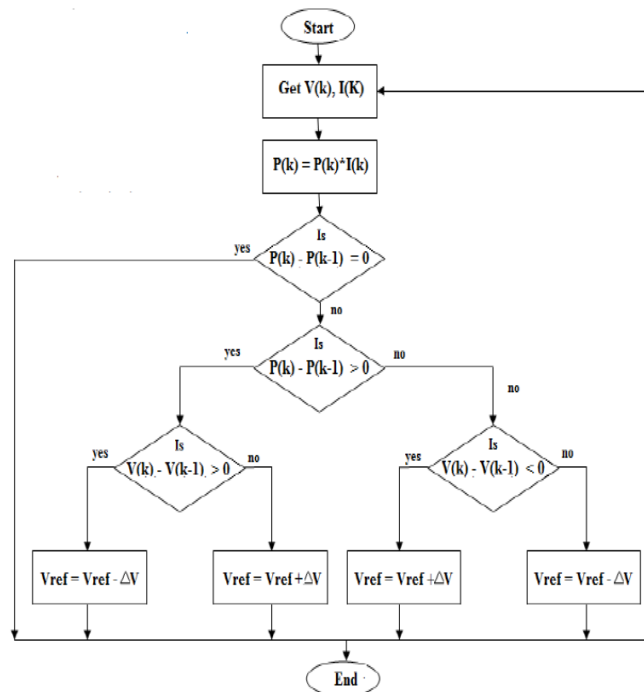


Fig.6:-Flow Chart for P&O MPPT Technique.

This perturbation continues indefinitely. Thus, the output power oscillates around a maximum power point and never stabilizes. The P&O technique has been widely used due to the simple feedback structure, reduced number of parameters and thus can be implemented quickly. In this algorithm the array terminal voltage and current were sensed and processed and power output is calculated. The present PV output power is compared with the power of previous perturbation cycle as shown in the flow chart. The PV voltage and current is perturbed periodically after comparing. If the PV operating voltage varies and change of power is greater than zero $dP/dV > 0$, the algorithm moves the operating point in the same direction, and if the change of power is less than zero $dP/dV < 0$, the algorithm moves the operating point in the opposite direction and if the change of power is equal to zero $dP/dV = 0$ that represents the condition of maximum power point.

The advantages of P&O algorithm is previous knowledge of the PV array characteristics is not required and its simple procedure. But the oscillation of the operating point around MPP under steady state conditions and inability to respond for the rapid changing atmospheric conditions are the drawbacks of this system.

Particle Swarm Optimization Application to MPPT Control:-

Particle Swarm optimization is an optimization method that tries to improve the particle solution with respect to given measure of quality. These particles moves around the search space according to position and velocity. Each particle is influenced by its own best position and velocity.

In PSO, particles are randomly initialized at different positions followed by position update based on new velocity, previous best positions and distance to Pbest to Gbest. Search process is continued till optimum value is obtained. As the particles, approach to MPP they get closer to Gbest position. Correspondingly, Pbest factor and Gbest factor in velocity term moves towards zero.

Eventually, zero velocity is obtained and duty cycle remains almost unchanged under this condition PV system reaches MPP. PSO is useful to track MPP in partial shading conditions. PSO can be used in conjunction with P&O MPPT technique to improve the tracking efficiency. PSO method;

- 1) Used to find out optimal duty cycle
- 2) PSO search starts with random initial guess and it is possible to explore the search region with continuous updation of duty cycle.

- 3) Reduction in steady state oscillations around MPP.
- 4) PSO alongwith MPPT technique responsible for direct duty cycle control.

Main feature is that absence of steady state oscillations at MPP and ability to track MPP for extreme environmental conditions i.e. effective in locating MPP. Drawback is its initial duty cycle selection. If duty cycle values are not within well defined limits and algorithm takes more no. of iterations to obtain the global maximum. This causes reduction in power output. Therefore it is necessary to specify limits of initial duty cycle.

A) Algorithm for PSO Implementation:

Step 1- Set the number of particles and searching parameters along with the limit for position and velocity

Step 2- Randomly initialize Position and velocity of each particle.

Step 3- Compute the fitness value of each particle.

Step 4- The particle having the best fitness value is set as Gbest (Global Best).

Step 5- Update the position and velocity of each particle with respect to the Gbest.

Step 6- Repeat Step 3 & 4 till the optimum solution is reached.

Step 7- Gbest at the end of the last iteration gives the optimized value.

Step 8- Compute the Duty-cycle using the given formula.

$$D = \frac{1}{1 + \sqrt{\frac{R_{in}}{R_{out}}}}$$

DC-DC Converters:-

They are power electronic circuits that convert a dc voltage to a different voltage level. Generally switched mode DC-DC converters are used i.e. the use of a switch or switches for the purpose of power conversion can be regarded as an SMPS. In order to perform power conversion with highest possible efficiency. Converter switches can be implemented using power BJT, power MOSFET, GTO thyristor, IGBT. There are different types of DC-DC converters such as Buck, Boost, Buck-Boost, Cuk converters etc.

One of the basic switched mode converter is the buck-boost converter. The output of the buck-boost converter can be either higher or lower than the input voltage. Assumption made about the operation of this circuit are;

Analysis of the boost converter begins by making these assumptions:-

- 5) The circuit is operating in the steady state.
- 6) The inductor current is continuous (always positive).
- 7) The capacitor is very large, and the output voltage is held constant at voltage V_o .
- 8) This restriction will be relaxed later to show the effects of finite capacitance.
- 9) The switching period is T , the switch is closed for time DT and open for time $(1-D)T$, The components are ideal.

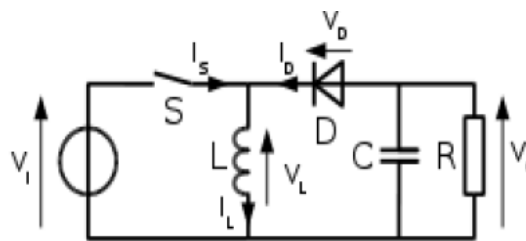


Fig.7:- Schematic of a buck–boost converter

The two operating states of a buck–boost converter: When the switch is turned-on, the input voltage source supplies current to the inductor, and the capacitor supplies current to the resistor (output load). When the switch is opened, the inductor supplies current to the load via the diode D.

Conclusion:-

The aim of this work is to propose a design of PV system peak power tracker and to realize a simple digital controller capable of optimizing the amount of power recovered from a solar panel over a range of environmental conditions. The basic design of the peak power tracker is to read the voltage and current levels at the solar panel simulator output, process these values using the P&O algorithm, and then adjust the voltage in order to obtain

maximum power. The results can be obtained and compared with theoretical operation which confirm the effectiveness of peak power tracking technique. Simulation of Maximum Power Point Tracking of a Solar PV panel using Particle Swarm Optimization and Perturbation & Observation techniques is responsible for direct duty cycle control. If Comparison of SPV panel output with and without optimization is carried out then the results show that particle swarm optimization technique given better results. Hence PSO technique can be employed for MPPT of Solar PV panels. This system is not only able to track MPP for extreme environmental conditions but also it reduces steady state oscillations at MPP which results in stability of the system.

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RESEARCH ARTICLE

FORMULATION DEVELOPMENT OF MUCOADHESIVE BILAYER TABLET WITH METFORMIN HYDROCHLORIDE.

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Mucoadhesion, gastroretentive, sustained release, kinetic model, ex-vivo study.

Abstract

In the present study, an attempt was made to design and evaluate bilayer gastroretentive drug delivery system exhibiting a combination of an immediate release layer and a sustained release mucoadhesive layer as second layer to prolong residence time in stomach using Metformin HCl as a model drug to reduce frequency of administration and to overcome bioavailability problems. A total of 5 formulations of bilayer tablets containing the drug were prepared by direct compression method using 5 different mucoadhesive polymers in 5 different formulations (F₁-F₅) respectively to impart mucoadhesion. Formulated bilayer tablets were evaluated by different parameters such as; *ex vivo* mucoadhesion strength, *ex vivo* mucoadhesion time, *In vitro* drug release study. *In vitro* drug release data were fitted to various kinetic models (Zero order, 1st order, Higuchi model, Korsmeyer- Peppas model). Upon R² value analysis it was observed that release from F₁, F₃, F₅ followed Higuchi model and F₂, F₄ followed Korsmeyer- Peppas kinetic model. Mechanism of drug release was found to be Fickian diffusion (value of n < 0.45). F₄ with Na CMC was found to be more promising in *ex vivo* mucoadhesion study showing residence time of 4.08 hrs. This study concludes that formulated bilayer tablets were satisfactory in providing prompt release from 1st layer then maintained sustained release effect with promising mucoadhesion behavior in stomach.

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Introduction:-

The new drug delivery system with better efficacy and safety with reduced dosing frequency and improved patient compliance is the current area of research by formulation development scientists. Tablet being most preferred dosage form for its ease of manufacturing and patient convenience is always a first choice of dosage form. The single layer tablets leads to frequent dosing and unpredicted plasma level for drugs with shorter half-lives. Number of diseases require immediate release of drug for instant effect and then plasma concentration has to be maintained for prolong effect of the drug. So, in order to deliver drugs in a predictable time frame, multilayered oral controlled release delivery systems are designed. These systems enhance the efficacy, minimize the adverse effects and increase the bioavailability of drugs.

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Mucoadhesive drug delivery is a topic of interest in the design of drug delivery systems to prolong the residence time of the dosage form at the site of application or absorption and thereby to facilitate the intimate contact of dosage form, thus to improve and enhance the bioavailability.

Dosage forms designed for mucoadhesive drug delivery should be small and flexible enough to be acceptable for patients and should not cause irritation. Other desired characteristics of a mucoadhesive dosage form include high drug loading capacity, controlled drug release (preferably unidirectional release), good mucoadhesive properties, smooth surface, tastelessness, and convenient application. Erodible formulations can be beneficial because they do not require system retrieval at the end of desired dosing interval. A number of relevant mucoadhesive dosage forms have been developed for a variety of drugs (Boddupalli et al., 2010).

The mechanism of adhesion of certain macromolecules to the epithelium of a mucous tissue is understood. They are characterized with an epithelial level whose surface is protected by mucus. The mucus contains glycoproteins, lipids, inorganic salts and 95% water by mass, making it a highly hydrated system. Mucin is the significant glycoprotein of mucus and is responsible for its structure. The principal functions of mucus are covering and lubricating the epithelium and some other functions based on the epithelium protection. Mucus width can change from 50-450 μm in the stomach to less than 1 μm in the oral cavity (Smart, 2005).

The mucous area, majorly used for the drug administration and absorption, is gastrointestinal mucus. The mucoadhesion ought to extend over the substrate to initiate the close connection, enhance the surface contact, and in turn increase the diffusion of its chains inside the mucus. The mechanism of mucoadhesion is usually separated in two stages, the contact stage and the consolidation stage. The first stage is characterized by the contact between the mucoadhesion and the mucous membrane, with covering and swelling of the formulation and initiating its deep contact with the mucous level (Hägerstrom and Edsman, 2003).

The development of sustain release dosage form can achieve the aim of releasing the drug slowly for a long period but this is not sufficient to get sustained therapeutic effect. They may be cleared from the site of absorption before emptying the drug content. Instead, the mucoadhesive dosage form will serve both the purposes of sustain release and presence of dosage form at the site of absorption.

In the recent times, multi-layer matrix tablets are gaining importance in the design of oral controlled drug delivery systems. Bi-layer tablets are novel drug delivery systems where combinations of two or more drugs are given in a single unit. They are preferred for the following reasons: To co-administer two different drugs in the same dosage form, to minimize physical and chemical incompatibilities, for staged drug release, IR and SR in the same tablet, for chronic condition requiring repeated dosing.

In the present research, an attempt was made to develop oral mucoadhesive bilayered tablet of Metformin HCl (MH) using Xanthan gum, NaCMC, HPMC 100CPS, HPMC (K-100LV) and Methocel K4M polymers. Metformin HCl, chemically N,N-Dimethyl imido dicarbonimidic diamide, is a biguanide anti hyperglycemic agent used for treating non-insulin-dependent diabetes mellitus (NIDDM). It improves glycemic control by decreasing hepatic glucose production, decreasing glucose absorption and increasing insulin-mediated glucose uptake. Metformin is the only oral anti hyperglycemic agent that is not associated with weight gain. Metformin may induce weight loss and is the drug of choice for obese NIDDM patients. When used alone, metformin does not cause hypoglycemia; however, it may potentiate the hypoglycemic effects of sulfonylurea's and insulin. The half-life of metformin is 6.2 hrs (Kadhe and Arasan, 2002; Desvergne and Wahli, 1999; Berger and Moller, 2002). It is freely soluble in water, slightly soluble in alcohol, practically insoluble in acetone and in methylene chloride (Chawla et al., 200; Chinetti et al., 2000; Willson et al., 2001).

It is a hydrophilic drug and is slowly and incompletely absorbed from the gastrointestinal tract, and the absolute bioavailability of a single 500 mg dose is reported to be 50% to 60%. This indicates the need to develop dosage forms that can retain the drug in the stomach for better absorption. An obstacle to more successful use of metformin therapy is the high incidence of concomitant gastrointestinal symptoms, such as abdominal discomfort, nausea, and diarrhea that especially occur during the initial weeks of treatment. The compound also has relatively short plasma elimination half- life (1.5 - 4.5hrs). Sustained release formulations may be administered once or twice daily. By considering above parameters, mucoadhesive bilayer tablet of metformin may be utilized.

This study describes the preparation of bilayer device comprising Metformin HCl as API and mucoadhesive layer of polymer to prolong its duration of action and improve patient compliance.

Materials and methods:-

Materials:-

Metformin HCl was received as a gift sample from Square Pharmaceutical Ltd, Bangladesh.; Methocel was purchased from Colorcon; Hydrochloric acid was obtained from Merck, Germany; and Xanthan gum, NaCMC, HPMC 100CPS, HPMC (K-100LV), Lactose, Magnesium stearate, Talc and Aerosil were obtained from local market. All other ingredients were of analytical grade.

Methods:-

Preparation of bilayer tablet:-

A total of 5 different types of formulations were prepared in 2 stages using a 13 mm- diameter die on an infrared hydraulic press with 5N compression force for a total of 1 minute 30 seconds. Initially the mucoadhesive layer was prepared according to Formulation plan (Table-1) for 30 seconds by simple thumb pressure then the 2nd layer (immediate release layer) was prepared by final compression force of 5N for 1 minute.

Table 1:- Formulation plan:

Ingredients (mg/ tablet)	Immediate release layer	Mucoadhesive sustained release layer				
	(F ₁ – F ₅) Composition is same for all formulations	F ₁	F ₂	F ₃	F ₄	F ₅
Metformin HCl	150	250	250	250	250	250
HPMC100cps	-	200	-	-	-	-
HPMC(K-100LV)	-	-	200	-	-	-
NaCMC	-	-	-	200	-	-
Methocel K4M	-	-	-	-	200	-
Xanthan gum	-	-	-	-	-	200
Lactose	100	50	50	50	50	50
Magnesium stearate	1	1	1	1	1	1
Talc	1	1	1	1	1	1
Aerosil	1	1	1	1	1	1
Total weight	253	503	503	503	503	503

Evaluation of physical and biological parameters of formulated bilayer tablets:-

In vitro drug release study for mucoadhesive bilayer tablet:-

In vitro release profile for each formulated bilayer tablet was performed using USP type II dissolution apparatus (Electrolab, Mumbai, India). Dissolution studies were carried at rotation speed of 50 RPM, in 900 ml of 0.01 N HCl of pH 2.0 at $37 \pm 0.5^\circ\text{C}$. Aliquot of 5 ml was withdrawn upto 8 hrs. The withdrawn volume was replaced with same volume of dissolution medium in order to keep the total volume constant. The absorbance of sample was measured by an UV spectrophotometer at 233 nm after suitable dilution if necessary. After filtration, the amount of drug release was determined from the standard calibration curve of pure drug (Singh et al., 2012).

Kinetic modeling of *in vitro* drug dissolution profile:-

In vitro drug release data of all the bilayer tablet formulations of Metformin HCl was subjected to goodness of fit test by linear regression analysis according to zero order, first order kinetics and according to Higuchi's and Korsmeyer-Peppas models to ascertain mechanism of drug release.

Measurement of *ex-vivo* mucoadhesive strength:-

Bioadhesive strength of the tablets was measured on modified physical balance. A piece of goat stomach mucosa was pasted to a petri-dish with cyanoacrylate adhesive and the mucus membrane was wetted with 2-3 drops of 0.01 N HCl media. The tablets were tied with thread and attached with the mucous membrane. Another end of thread tied with one side of the physical balance. The weight required to detach the tablet from the mucosal surface was taken as the measure of mucoadhesive strength. Force of adhesion was calculated from the mucoadhesive strength as per following equation:

Force of adhesion (N) = (mucoadhesive strength \times 9.81) \div 1000:

Measurement of *ex-vivo* residence time:-

The *ex-vivo* residence time was determined using a locally modified USP disintegration apparatus. A segment of goat stomach mucosa was glued to the surface of the beaker, vertically attached to the apparatus. The mucoadhesive tablet was then pasted to the mucosal membrane by applying a light force with a finger trip for 30 seconds. Then the tablet and mucosa was hydrated by the medium of 800 ml 0.01 N hydrochloric acid of pH 2.0, allowing the apparatus to move up and down maintaining $37^{\circ}\text{C} \pm 0.5^{\circ}\text{C}$; monitoring the tablet adhesion for 8 hours to see retention time and correlate with sustaining property of formulation. The time for the tablet to detach from the goat stomach mucosa was recorded as the mucoadhesion time (Umarji et al., 2012).

Results and Discussion:-

The main goal of this work was to develop new mucoadhesive bilayer tablets of metformin HCl; consisting of an immediate release and a of sustained release layer. The double layered structure design was expected to provide immediate release of metformin HCl from the outer layer to produce a prompt pharmacological action then sustained release from second layer to provide a sustained release of enough drug over an extended period of time and retain in stomach for an extended period of time.

A total of 5 formulations of mucoadhesive bilayer tablets were prepared and evaluated for biological and physical parameters. The composition of final bilayer formulation had been chosen from the previous single sustained release layer formulation which gives the best average percent release and best fitted with kinetics model of dissolution release pattern. Different amounts (150 mg, 180 mg, 200 mg, 220 mg) of 5 different mucoadhesive polymers (HPMC 100 cps, HPMC K-100LV, Xanthun gum, NaCMC, Methocel K4M) were used to prepare single sustained release layer. The study indicated that increase of concentration of polymer decreases *In vitro* drug release and gives larger $T_{50\%}$ value; which indicates more sustained release effect. Thus the middle amount of polymers (200 mg) was selected for the final bilayer formulation.

According to work plan, the formulated bilayer tablets were evaluated for their *in vitro* drug release profile study, *ex vivo* mucoadhesive strength, *ex vivo* residence time, $T_{50\%}$, $T_{25\%}$, $T_{70\%}$ determination.

Physical appearance:-

The appearance of bilayer tabs was smooth and uniform on physical examination.

Kinetic modeling of *in vitro* drug dissolution profile:-

In vitro drug release studies were conducted and fitted to different kinetic models. Cumulative percent of drug release from the formulations upto 8 hrs is shown in the Figure-1; release rate kinetic constants and R^2 values obtained from different kinetic models are tabulated in the following Table 2.

Table 2:- Interpretation of release rate constants and R^2 values for different release kinetics of formulated bilayer tablets:

Formulations	Zero order		First order		Higuchi		Korsmeyer	
	K_0	R^2	K_1	R^2	K_H	R^2	N	R^2
F ₁	5.935	0.873	-0.040	0.940	19.25	0.978	0.426	0.956
F ₂	7.084	0.839	-0.059	0.950	23.36	0.971	0.328	0.988
F ₃	6.614	0.864	-0.050	0.940	21.48	0.970	0.370	0.962
F ₄	6.648	0.883	-0.048	0.953	21.46	0.980	0.414	0.964
F ₅	6.851	0.854	-0.054	0.948	22.42	0.974	0.354	0.978

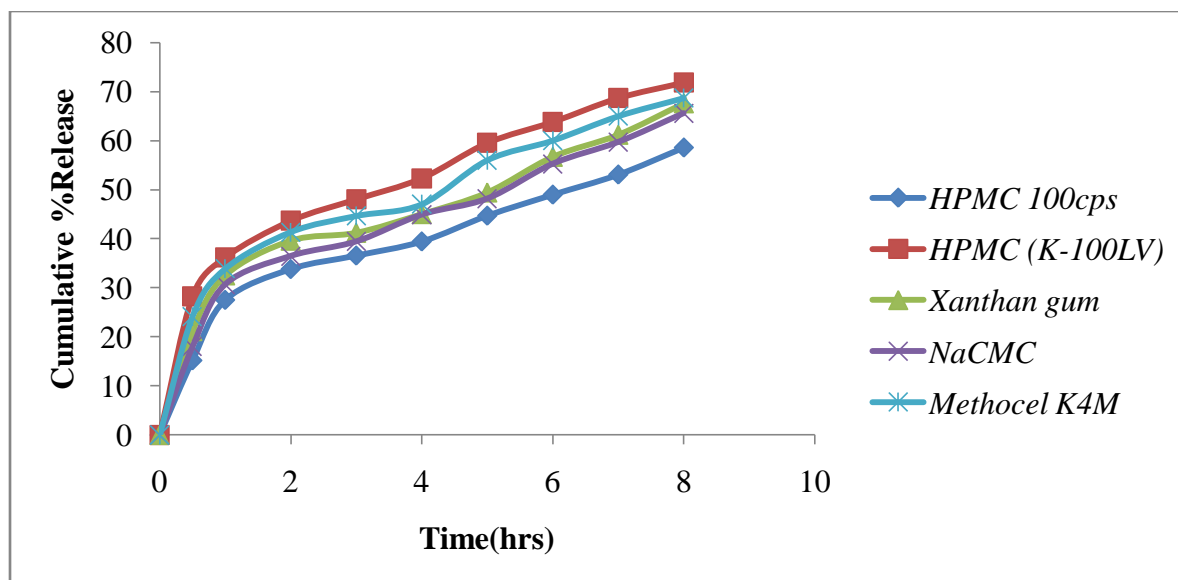


Fig 1:- *In vitro* release profile of formulated bilayer tablets containing different types of polymers. Here, used mucoadhesive polymer indicated the formulations. F₁ is formulated with HPMC 100 cps, F₂ is formulated with HPMC (K-100LV), F₃ is formulated with Xanthun gum, F₄ is formulated with NaCMC, F₅ is formulated with Methocel K4M.

The *in vitro* release data are treated with different model of release kinetics in the above Table 2 and the best fitted model was explained by the help of R² value in Table 3 given below. Evaluated, T_{25%}, T_{50%}, T_{70%} were also tabulated in the Table 3 and n values were also mentioned for drug transport mechanism explanation. Fig 2 illustrates the evaluated values of successive fractional dissolution time of formulated bilayer tablets.

Table 3:- Drug release kinetics and dissolution parameters of formulated bilayer tablets:

Formulations	Best Fit Model	R ²	n	K _H	T _{25%}	T _{50%}	T _{70%}
F ₁	Higuchi	0.978	0.426	19.25	1.18	5.98	18.03
F ₂	Korsmeyer	0.988	0.328	-	0.35	2.93	12.27
F ₃	Higuchi	0.970	0.370	21.48	0.66	4.32	15.37
F ₄	Korsmeyer	0.978	0.414	-	0.88	4.69	14.6
F ₅	Higuchi	0.980	0.354	22.42	0.51	3.62	13.67

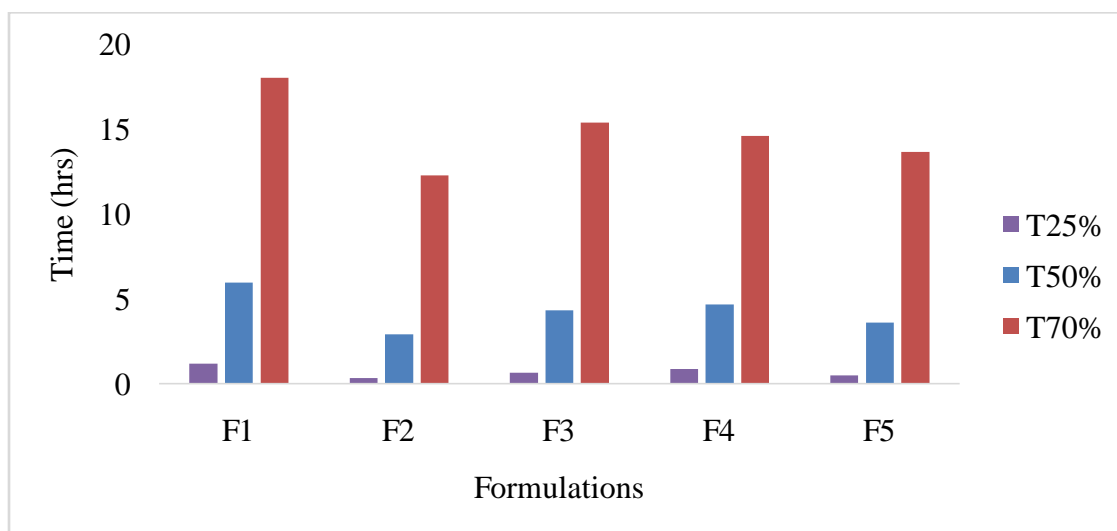


Fig 2:- Successive fractional dissolution time of formulated bilayer tablets.

$T_{50\%}$ values of formulations manifest the drug release rate of dosage form and the retarding efficacy of polymers. Higher $T_{50\%}$ value indicates a higher drug retarding ability of polymer and vice versa. Fig - 2 indicates better sustained release effect was found in F_1 and F_3 compared to other formulations.

Diffusional release mechanism of the formulations was interpreted from *in vitro* drug release data by analyzing the n values by fitting in Korsmeyer Peppas kinetic model and results are tabulated in Table 4.

Table 4:- Interpretation of diffusional release mechanism from *in vitro* drug release data from formulated bilayer tablets:

Formulation	n (Release exponent)	Drug Transport Mechanism
F_1	0.426	Fickian diffusion
F_2	0.328	Fickian diffusion
F_3	0.370	Fickian diffusion
F_4	0.414	Fickian diffusion
F_5	0.354	Fickian diffusion

Ex-vivo study for evaluation of mucoadhesive behavior of bilayer tablets:-

The *Ex- vivo* mucoadhesive strength of the tablets was determined for different formulations using goat stomach mucosa. *Ex vivo* residence time was evaluated by conducting a study for 8 hrs by recording the time for complete detachment of tablet from mucosal surface. The evaluation results are tabulated in Table 5 and illustrated in Fig 3, Fig 4.

Table 5:- *Ex- vivo* evaluation of mucoadhesive behavior of formulated bilayer tablets:

Formulations	Detachment force (N)	Residence time (hrs)
F_1	0.661	2.22
F_2	0.621	1.4
F_3	0.6	3.07
F_4	0.916	4.08
F_5	0.523	2.02

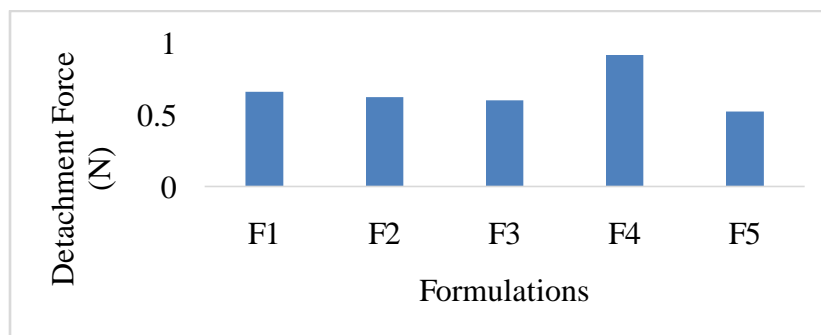


Fig 3:- Detachment force of different formulation containing different mucoadhesive polymer.

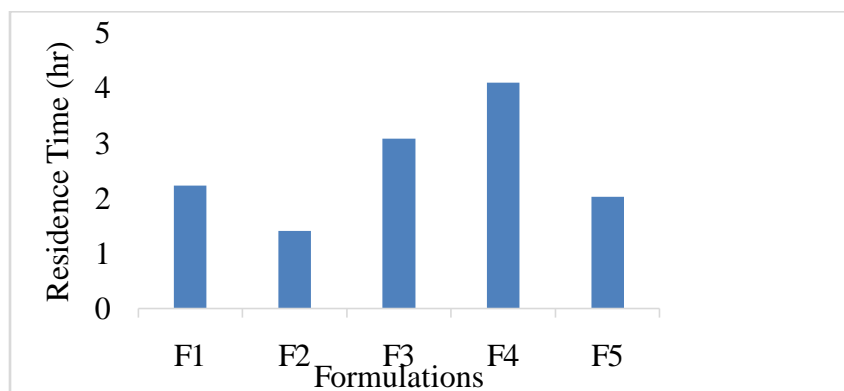


Fig 4:- Ex vivo residence time of different formulation containing different ratio of mucoadhesive polymer.

From the Table 4, Fig 3 and Fig 4; it was observed that the highest mucoadhesive strength was observed with the formulation showing highest detachment force. F₄ containing NaCMC showed highest mucoadhesive strength. Tablet containing an increased concentration of polymer showed higher mucoadhesion with long residence time (4.08 hrs). This finding is owing to the hydrophilic nature of NaCMC; it is hydrated easily with less contact time and forms a strong interpenetrating network with mucin molecules because of the polymer's long linear chain structure. A linear increase in mucoadhesion was observed with an increase in contact time.

Conclusion:-

In the present study, 5 different formulations of mucoadhesive bilayer tablet of metformin HCl (F₁ – F₅) were prepared by using 5 different Polymers by direct compression method. The results of *in-vitro* drug release profile of bilayered tablets were satisfactory. But, F₁ and F₄ provided better controlled and sustained *In vitro* drug release compared to other three formulations. The T_{50%} values of these five formulations which is an important parameter for knowing the retarding ability of drugs were also found satisfactory. Results from *ex vivo* evaluation of mucoadhesive behavior of these five formulations were also good which provides increased residence time of drug in GIT and gave clear understanding about the bioadhesion properties of the bilayer tablet. However, an appropriate balance between various levels of the five polymers is needed to acquire proper release and mucoadhesion. Finally, it can be concluded that by formulating mucoadhesive bilayer tablets of metformin HCl, its complete release can be ensured prior to absorption and hence the problem of incomplete drug release and erratic absorption of pure metformin HCl can be solved by increasing the retention time of drug in GIT for a longer period with the help of mucoadhesive polymers.

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RESEARCH ARTICLE

PROBLEMS AND PROSPECTS IN SOUTH ASIA; INDO-CHINA'S EFFORTS OF COOPERATION.

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Abstract

This paper attempts to analyse the problems and prospects in south Asia by using various indicators based on data from secondary sources. The study reveals that South Asia a land of natural beauty and also known as paradise on Earth, and Switzerland of Asia. But the countries of South Asia are not able to evolve co-operative relations and their relations are characterizing by numerous conflicts. The type and level of conflicts which take place in the sub-continent however vary with changing circumstances, both internal and external, which affect the region. Of late, of course the countries of South Asia realize the need of cooperation and make some progress in this direction. Indo-china both are the most powerful nations of region performing their role in various direction for maintaining the peace, cooperation and integration in the region.

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Introduction:-

The term South Asia is used for the countries lying south of Himalayas and surrounded by the Indian Ocean from three sides. The countries of this region include India, Pakistan, Bangladesh, Sri Lanka, Nepal, Maldives and Bhutan. Apart from Nepal and Bhutan, the rest of the South Asian region has been subject to colonial rule. These countries gained independence in 1947; Pakistan came into existence as a result of partition of India in 1947; Sri Lanka got independence in 1948. Bangladesh emerged as an independent sovereign state after breaking away from Pakistan in 1971. Though the countries differ from each other with regard to climate, race, religion and history, they constitute a single region and possess some common features. Firstly, most of the countries are very poor and majority of their people live below poverty line. Secondly, religion is a predominant factor in these states. In fact, some of the states like Pakistan and Bangladesh were created on religious basis. Thirdly, these countries are faced with problems of unemployment, illiteracy, over population, etc. Fourthly, the countries are agriculturally and industrially backward. However, India is an exception and has made considerable industrial progress. Fifthly, most of the countries are faced with the problem of political instability. Finally, most of the countries (except Pakistan) do not have close military links with super powers. In contrast with the external boundaries of the region, the political geography within the region does not follow any neat pattern. The three most important rivers-Indus, Ganga and Brahmaputra cut across the boundaries of the principal countries in the Indian sub-continent. In spite of differences in physical appearance, complexion, stature and other ethnological features, a common cultural and ethnic outlook unifies the people of the entire region of South Asia. However, the South Asian countries have sharp differences in their perception and approaches on the details of major global and regional issues of peace, security and stability. The pattern of these differences has remained somewhat consistent over a long period except for some occasional minor shift in their strategies.

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South Asia has become an area in which India and China have been seeking to gain political and economic influence over many years. Therefore, it seems appropriate to analyse their relationship with South Asian countries through the lens of the regional power debate. Both countries have claimed regional power status, i.e. a dominant position, and have invested in different ways to shape their respective regions according to their political interests. India has used different bi- and multilateral strategies to pursue its regional leadership role in South Asia. In contrast, China has concentrated on bilateral relations rather than on multilateral structures, in order to expand its traditional regional focus from North East and South East Asia to South Asia¹.

Conflicts in south asia:-

The harsh geo-political realities point towards greater potential for conflict rather than peace. These may be well routed in geo-strategic factors and in the strategies of the leading powers of the world to influence these factors in their own interests. Some of the geo-strategic factors are:

Military Stand Off in the Region- Whereas India has tended to find greater convergence in its perceptions with Russian position on many issues; however other South Asian states have reflected towards west towards their world views.

Emergence of China as a Reckonable Power- China's growing power and its assertive approach in dealing with other nations can only create apprehensions and a sense of insecurity in the countries of South Asia excepting those like Pakistan who have sought a quasi-alliance relationship with it. Similarly, China's growing capabilities of power projection could pose a major challenge to peace and security in the days ahead.

Situation in Afghanistan and Middle East- Afghanistan and Middle East are the two conflict areas where local indigenous conflictual factors have been entangled in great power strategies and interests. The continuation or resolution of the conflict in the two areas will have far-reaching influence on the peace and security in the region.

Growth of Extra Regional Military Forces in the Indian Ocean- There has been a massive growth of extra regional military forces in the Indian Ocean in the recent years. The presence of extra regional forces only helps to exacerbate conflictual responses and aggravate the problem of peace and security in the region. Strategically too the Indian Ocean region is important for global nuclear warfare and thus it has become a victim of US and European game plan. The success of US Coalition Strategy to undertake military responses outside NATO areas in conflict situation during the second half of 1987 in Persian Gulf Arabian Sea is noteworthy².

Cooperations:-

Religious and Cultural Interrelation:-

South Asian region has uniqueness in shared history, heritage, literature, culture, religion and language. The commonality of culture and religion provides a strong motive for regional cooperation as it has similar lifestyle, fashion and clothing trends, music, food or even cinema. Even the people have family relations across borders that were formed before partition and have sympathy towards them. So this cross-religious, cross-cultural pattern also demands cooperation at regional-level to provide better access and opportunity to people for harmony and religious beliefs. South Asia develops greater economic and political cooperation it is also possible that the notion of pan-South Asian identity will emerge at local and global level and trickle down to the people. That is why AmartyaSen (2006) gives notion of cultural identity where people could have multiple identities.

Economic Acquisitions:-

In regional cooperation processes trade must be a means not an aim per se to increase the productive power of a state. A long-term economic strategy of any developing state must have diversion in trade destinations including its neighbour destinations, diversity in trade pattern/products including both labour intensive and capital intensive. Most states in this region lack skills, research capacity, financial and technical resources for research and education, in this context establishment of South Asian University and signing of South Asia Free Trade Area agreement (SAFTA) are significant step (Mehta and Kumar 2004; Banik 2006; Ratna and Sidhu 2008; Taneja et al. 2011). 2 The share of intra-regional trade is very low in South Asia with comparison to intra Association of Southeast Asian Nations (ASEAN) or intra-EU trade for intraregional trade of SAARC nations; also for SAARC trade with rest of the World. The share of SAARC trade has remained same ranging from 4-6 percent between 2000 and 2013. This low intra-regional trade has been on account of positive list based approach on exchange of tariff preferences, small product coverage, narrow margins of preferences and inability to address non-tariff barrier³.

political integration, for instance the promotion of democratic values, were only used selectively and remained contingent on India's security concerns *vis-à-vis* the neighbours. In 2009, the South Asian Association for Regional Cooperation (SAARC) agreed on a Charter of Democracy to strengthen democratic structures in South Asia. The Charter becomes fully operational, it can become an important instrument that allows India to support democratic institutions in neighbouring countries. But it is also an ambivalent instrument because this could be seen as interference in the internal affairs of countries, which India itself rejects. The Indian Technical & Economic Cooperation Programme (ITEC) has trained many bureaucrats from South Asia and has helped New Delhi to increase its political clout in the region. In 2013–2014 more than 80 per cent of India's total loan and aid budget was allocated to its South Asian neighbours, especially Bhutan, Bangladesh, and Afghanistan.

India's efforts

INDIA seems to be a regional power in South Asia by default. Governments in New Delhi have used a variety of instruments to further their interests in the region: wars have India been fought and won (for instance with Pakistan); militant groups have been supported in their fight against authoritative regimes (East Pakistan) and democratic governments (Sri Lanka); trade sanctions and border blockades have been imposed; secret arrangements have been negotiated (Nepal); unilateral economic and political concessions (SAARC) have been made; development aid has been provided (Afghanistan, Bhutan, Nepal); and successful (Nepal) and unsuccessful (Sri Lanka) attempts for mediation in civil wars have been made

Since its independence in 1947, India has been perceived as a regional power or hegemon in South Asia. Due to the asymmetry of its territorial and demographic size, its military dominance, and its economic potential, India seems to be a regional power by default. But a closer look reveals that India's superior economic and military resources have hardly resulted in the achievement of its foreign policy goals.

For many years, India perceived its neighbouring countries as a major part of its national security and its strategic sphere of influence. India's policy *vis-à-vis* the Himalayan states has always been determined by its relations with China and it has always aimed to reduce China's influence on its northern border. The friendship agreements with the Himalaya kingdoms Bhutan (1949), Sikkim (1950), and Nepal (1950) gave India a great involvement in the internal affairs of its neighbours. Prime Minister Jawaharlal Nehru, who is often blamed for being an 'idealist' in international affairs, appears to have had a much more 'realistic' perspective with regard to India's neighbours in the Himalayas⁴.

Indira Gandhi's South Asia doctrine saw the region as part of India's national security. This became the basis for various political and military interventions during the 1970s and 1980s. But many of the interventions did not bring about the desired long-term results for India. The military victory over Pakistan in 1971 was not followed by a permanent solution of the Kashmir issue. India supported Bangladesh after its independence in 1971 but could not prevent Bangladesh's economic and political realignment after the military coup in 1975. India's attempts to mediate in the Sri Lankan civil war in the late 1980s ended in political and military disaster.

After the economic liberalisation in 1991, economic issues became more and more important for India's foreign policy at both the regional and the global levels. In the mid-1990s, the Gujral doctrine highlighted the concept of non-reciprocity: India was now willing to make unilateral, political, and economic concessions to weaker states. South Asia was no longer seen as a part of India's national security, but as part of its economic development. The new focus on economics gained more importance *vis-à-vis* the traditional security concerns. Prime Minister Manmohan Singh moved economic relations to the centre of India's foreign policy, promoted economic development, gave greater importance to India's relations with the great powers, and tried to establish friendly relations with the neighbouring countries.

China's efforts:-

China's relations with India and South Asia have also undergone several changes. The good relationship it had with India in the 1950s was overshadowed by the boundary dispute. After the border war in 1962, China deepened its relations with Pakistan. As part of its ideological foreign policy, China supported various militant movements in South Asia, as for instance rebel groups in India's northeast.

China's strong economic and military support for Pakistan since the 1960s and its later efforts to develop its economic relations with India's neighbours have always raised apprehensions in India, that China's South Asia policy was aimed at pressuring India. Following its economic liberalisation in the late 1970s, China has also shifted its focus *vis-à-vis* South Asia. Its security interests *vis-à-vis* the region are dominated by the domestic challenges in Xinjiang and Tibet. China's South Asia policy has been characterised as 'multi-layered and complicated and not necessarily compatible and complementary'. China's attempts to create new access to the Indian Ocean as part of its 'One Belt, One Road' (OBOR) initiative by building land corridors through Pakistan and Bangladesh will increase the strategic importance of South Asia in the long-term.

In the field of security, China has the upper hand with regard to military hardware and strategic investment, whereas India has stronger military-to-military relations with its neighbours. In 2011, India's National Security Advisor Shiv Shankar Menon pointed out that India has to expand its role as security provider for South Asia as well as for the Indian Ocean region in order to counter the growing Chinese influence⁵. Prime Minister Modi also emphasised this point during his various visits to the SAARC countries and the island states of the Indian Ocean in spring 2015⁴.

Conclusion:-

It seems that despite its superior material resources, India has hardly ever managed to establish itself as regional power in the long-term perspective. Nehru's and Indira Gandhi's attempts to portray the region as part of India's national security and to secure the country's foreign political interests through military, economic, and political interventions have proven counterproductive in two ways. First, the process of nation-building in the neighbouring countries prevented a close approximation with India. Because of common religious, linguistic, and ethnic ties, nation-building was about delimitation of the dominant neighbour. Second, all neighbours have used the strategy of internationalising their bilateral disputes with India, sometimes more and sometimes less successfully. Pakistan is the most obvious case, but Bangladesh, Nepal, and Sri Lanka have also played the 'China card' at various times. Since its liberalisation in 1991, India has put its South Asia policy on a new basis with the Gujral and the Manmohan doctrines. South Asia is regarded more as a market that can contribute to India's economic development and therefore the focus of bilateral and multilateral initiatives is on regional public goods, like better connectivity as well as on unilateral economic concessions, in order to expand intra-regional trade.

In contrast to India, China enjoys a comfortable position. Politically, China is regarded as a 'neutral' player in South Asia. China has never been part of the discourse on nation-building and its relations with the region are not marred by the baggage of history. Economically, China is a more attractive trade and investment partner for South Asian countries. In the field of security, China is an important partner for the acquisition of military hardware. However, India has also improved its security collaboration with many South Asian countries in recent years.

India seems to be caught in a catch-22 situation in South Asia. On the one hand, religious, linguistic, and ethnic ties bind India with the region. On the other hand, those ties do separate India from its neighbours with regard to nation-building. Hence, India will scarcely be able to overcome its resentments in the neighbouring countries and to counter the advantages that China enjoys in South Asia with regard to politics, economics, and security. However, China's growing influence in South Asia does not imply a claim for any regional power status. It pursues its national interests and does not seem to be interested in providing regional public goods. It remains arguable whether China's activities within the region serve its own economic development, or how far it pursues a policy of 'encirclement' against India. It should not be overlooked that India and China are also cooperating in joint regional initiatives like the Bangladesh-China-India-Myanmar and that China has also promised to make large-scale infrastructure investment in India. Despite its disadvantageous position, India should continue its efforts to provide regional public goods through both bilateral and multilateral initiatives. This will foster regional cooperation in South Asia, so that the question of 'who is the regional power' may even become superfluous in the long time

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RESEARCH ARTICLE

EFFECT OF HEATING RATES AND Zn-ADDITION ON THE THERMAL PROPERTIES OF Pb-Sn ALLOY

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Key words:-

Thermal properties; Differential Thermal Analysis (DTA); Enthalpy of fusion; Activation energy.

Abstract

The variations of thermal properties with temperature for Pb-Sn alloys were measured using a heat flow apparatus. According to present experimental results, the thermal parameters (T_{onset} , T_{end} , T_m) of Pb-Sn alloys increase with increasing heating rates, also the results showed that the small amount of zinc improved significantly these parameters. Furthermore the change in enthalpy of fusion and activation energy of fusion for different heating rates were studied experimentally by the differential thermal analysis (DTA). The apparent activation energies of Pb-Sn alloys have been determined based on the Kissinger equation.

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Introduction:-

Pb-Sn alloys are the basis of many engineering materials [1]. An Extensive theoretical and experimental study of the nature of the relationships among microstructure, physical properties and solidification processing parameters has been created [2-5]. Pb-Sn solders are one of the most familiar materials used for various microelectronic connections in computer industry. Different compositions of Pb-Sn solders have considerable potential for advanced structural and electronic applications. Pb-Sn alloys have several attractive attributes: low cost, low eutectic temperature (183°C), good wettability on many common workpiece metals, and good thermal and electrical conductivities, and a choice between eutectic freezing or a wide freezing range which produces "pasty" solid-liquid mixtures[5-8]. Also, a great deal of efforts has been carried out on Pb-Sn eutectic alloys, which is currently used in computer industry for various microelectronic connections [9-12]. Although Pb-Sn solders are still widely used for many applications (e.g., body panel soldering on automobiles). Pb-Sn alloys make excellent solders for electrical and mechanical joining[13]. One of the important benefits of the thermal analysis experiment is its usefulness in selecting appropriate temperatures for the heat treatment cycles of the samples as well as for the investigation of the phase development[14, 15].

Most studies have dealt with the thermal properties of Pb-Sn solders with different compositions at room temperature, but a few papers have been focused on the dependency of thermal properties of these alloys on the temperature and composition of alloy. Thus, the purpose of present work was to determine the following parameters: temperature of onset of melting (T_{onset}), temperature of offset of melting (T_{offset}), heat of fusion, and the activation energy occurring during melting, besides the effects of different heating rates and Zn addition on melting temperature, heat of fusion, and the activation energy of the alloy samples.

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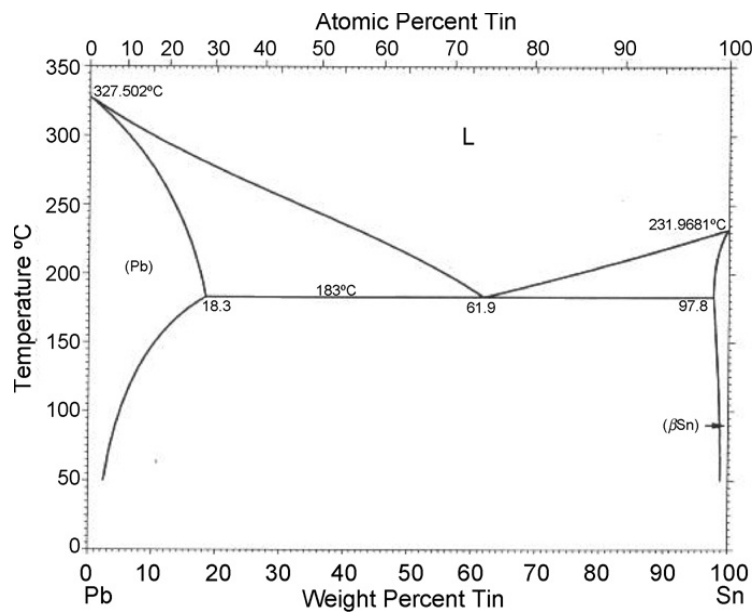


Figure 1:- Phase Diagram of Pb–Sn System

Phase diagram of Pb–Sn system has been evaluated as shown in Figure 1. Solubility of Pb in solid Sn and Sn in solid Pb at the eutectic temperature (456 K) are 2.2 wt.% Pb and 18.3 wt.% Pb, respectively [16]. Thus, the compositions of Pb–Sn alloys were chosen to be Pb–61.9 wt% Sn, Pb–61.9 wt% Sn–2 wt% Zn to investigate the dependence of the thermal measurements on the temperature and the effect of addition of Zn in the Pb–Sn alloys.

Experimental Details:-

Material Preparations:-

The two solder alloys, Pb-61.9wt%Sn and Pb-61.9wt%Sn-2wt% Zn were prepared from high purity Pb, Sn and Zn of purity 99.99%. The appropriate weights of the elements for the binary and ternary alloys were well mixed with CaCl_2 flux to prevent oxidation in a graphite mold. Casting in rod form was performed in a 15 x 1 x 1 cm graphite mold. The casting rod was annealed at 438K for 50 hours. The ingots were rolled into wires of diameter 1mm. In this study, the samples were annealed at 443K for 4h and then slowly cooled to room temperature at cooling rate $T = 2 \times 10^{-2} \text{ K s}^{-1}$.

Mechanical Tests:-

Differential thermal analysis (DTA) measurements were carried out on 40 mg of small pieces of alloy samples using a Labsys Evo_TG DTA thermal analyzer. DTA experiments were performed by heating the samples in a Pt crucible with Al_2O_3 as the reference material. Five different heating rates (10, 15, 20, 25 and $30 \text{ }^\circ\text{C min}^{-1}$) were examined during the measurements.

Results and Discussion:

Melting Temperature (T_m) of Alloys:-

The melting temperature is a critical alloy characteristic because it determines the maximum operating temperature of the system. Figure 2 represents the results of DTA analysis of the Pb-61.9wt%Sn and Pb-61.9 wt%Sn-2wt%Zn alloys measured at the heating rates of 10, 15, 20, 25 and $30 \text{ }^\circ\text{C min}^{-1}$, from room temperature to $450 \text{ }^\circ\text{C}$.

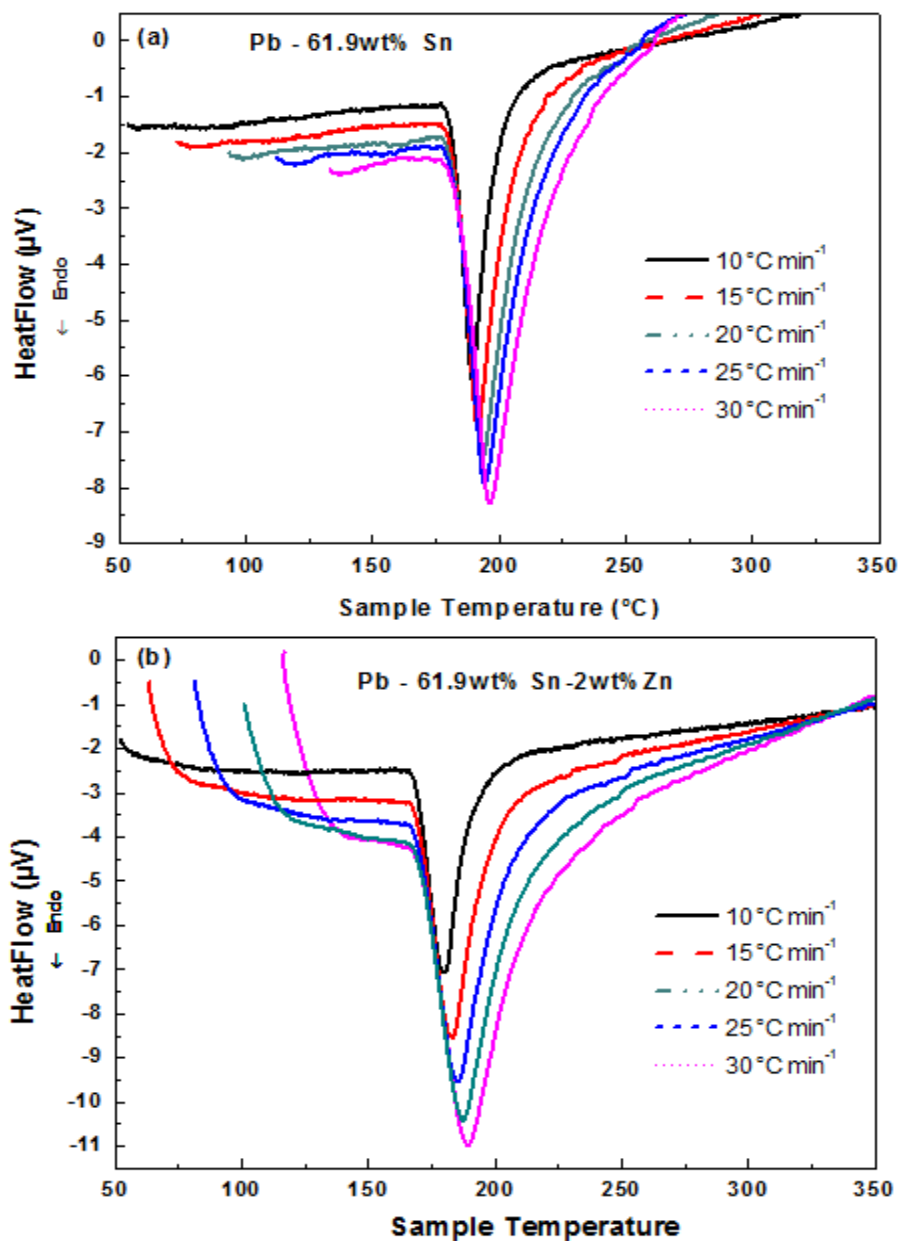


Figure 2:- Different Thermal Analysis (DTA) Curves for (a) Pb-61.9wt%Sn and (b) Pb-61.9 wt%Sn-2 wt% Zn Alloys Taken at Different Heating Rates.

It is shown that there is only one main endothermic peak for the samples during the measurements observed in the temperature range 30 – 450 °C, namely the melting which is indicated by the endothermic peak. The endothermic peak appears in DTA curve has two regions of heat extraction that are indicative of the melting temperature. The melting points were considered in support of probable industrial applications. The temperature of onset of melting, T_{onset} , (the onset point of heat absorption during heating) is defined by the extrapolated beginning of the curve, being defined by the point of intersection of the tangent with the point of maximum slope, on the principal side of the peak with the base line extrapolated [17, 18].

From Figure 2 (a, b) and Table 1, it can also be seen that the heating curves of the Pb-61.9wt%Sn and Pb-61.9 wt%Sn-2wt% Zn alloys are shifted towards higher temperature with the increasing rate of heating. The first effect of the heating rate can be explained by the decrease of the precipitated Zn atoms amount because of its increased solid

solubility at the increased precipitation temperatures when using higher heating rate [19, 20]. However the second effect is explained by the diffusive nature of the precipitation reactions.

Figure3 shows a schematic of the heat flow curve versus the temperature of Pb-61.9 wt%Sn-2 wt% Zn alloy obtained at the heating rate of $10\text{ }^{\circ}\text{C min}^{-1}$ and how the DTA curve is formed from the data recorded. The graph shows that the sample presents an onset temperature for melting of $169.809\text{ }^{\circ}\text{C}$ with $\Delta H = 8.425\text{ }(\mu\text{V.s/mg})$ and a melting temperature of $180.246\text{ }(^{\circ}\text{C})$.

The obtained values of the temperature of onset of melting (T_{onset}), the temperature of offset of melting (T_{offset} or T_{end}) and melting temperature (T_m) are listed in Table 1 and marked on the DTA curves as shown in Figure3 for Pb-61.9 wt%Sn-2 wt% Zn alloy obtained at a heating rate of $10\text{ }^{\circ}\text{C/min}$ (as example).

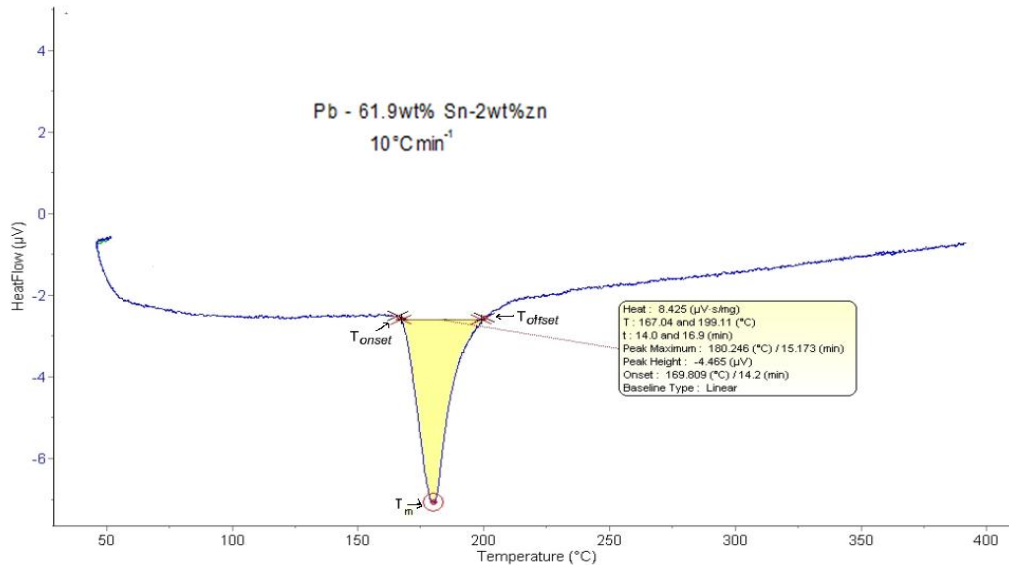


Figure 3:- Schematic of Heat Flow Curve Versus the Temperature for Pb-61.9 wt%Sn-2 wt% Zn Alloy at a Heating Rate of $10\text{ }^{\circ}\text{C/min}$.

Figure: 4 shows the comparison between the values of the melting temperature T_m for Pb-61.9 wt% Sn and Pb-61.9 wt%Sn-2 wt% Zn alloys at a heating rate of $10\text{ }^{\circ}\text{C/min}$. Addition of Zn increased the T_{onset} , and T_{end} as illustrated in Figure4 whereas, the Zn addition caused a further depressed of the melting temperature T_m of Pb-61.9wt%Sn alloy for different heating rate, which makes this alloy interesting for technical applications. In addition the melting range (temperature range between solidus and liquids points according to DTA) is extended as zinc metal addition.

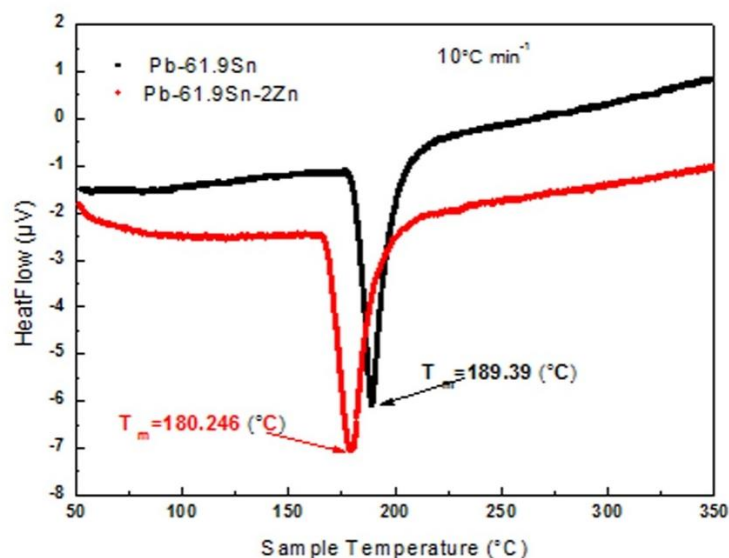


Figure 4:- Heat Flow Curve Versus the Temperature for Pb-61.9 wt% Sn and Pb-61.9 wt%Sn-2 wt% Zn Alloys at a Heating Rate of 10 °C/min.

Table 1:- Comparison of Onset Temperature (T_{onset}), Offset Temperature (T_{end}), Melting Temperature (T_m), and Heat of Fusion (ΔH) for Pb-61.9wt%Sn and Pb-61.9wt%Sn-2wt%Zn Alloys.

Alloys	Heating rate (°C min ⁻¹)	T_{onset} (°C)	T_{end} (°C)	T_m (°C)	ΔH (μV.s/mg)	E_a (kJmol ⁻¹)
Pb-61.9Sn	10	183.04	204.4	189.39	6.612	47.7
	15	182.569	212.3	191.66	6.494	
	20	183.787	217.7	193.489	6.204	
	25	183.67	220.4	194.216	5.637	
	30	184.935	224.9	196.269	5.676	
Pb-61.9Sn-2Zn	10	169.809	193.4	180.246	8.425	31.5
	15	170.896	205.1	183.366	7.728	
	20	170.844	211.2	185.343	7.396	
	25	172.07	219.9	187.344	7.673	
	30	171.611	225.6	189.422	8.031	

Determination of activation energy (E_a) of fusion:-

To determine the activation energy E_a for fusion of alloys by Kissinger methods, which determine the activation energy of the samples starting from the change in the temperature that corresponds to the maximum of the endothermic peak T_m according to the heating rate β as shown in Figure2 (a,b).

The activation energy of the Pb-61.9wt%Sn and Pb-61.9 wt%Sn-2wt%Zn alloys under continuous heating condition can be calculated by the Kissinger equation[21-23]:

$$\ln \left[\frac{\beta}{T_m^2} \right] = -\frac{E}{RT_m} + C, \quad (1)$$

Or it can be written as

$$\frac{d \ln \left(\frac{\beta}{T_m^2} \right)}{d \left(\frac{1}{T_m} \right)} = -\frac{E_a}{R} + C, \quad (2)$$

Where β is the heating rate, E_a is the activation energy of fusion and R is the gas constant (8.314J/mol K), T_m denotes the maximum melting endothermic peak, and C is a constant.

If the fusion mechanism remains constant with the heating rate (β), the plot of $\ln (\beta/T_m^2)$ vs. $1000/T_m$ gives a straight line and E_a is calculated from the slope of this line. Figure5 shows the Kissinger plot for the Pb-61.9Sn-2Zn alloy.

Table 1 lists the calculated activation energies for fusion. The calculated value of E_a decreased from 47.7 kJmol^{-1} for Pb-61.9wt%Sn alloy to 31.5 kJmol^{-1} for Pb-61.9 wt%Sn-2wt%Zn alloy. It is clear that a small amount of Zn-addition enhances the activation energies of fusion.

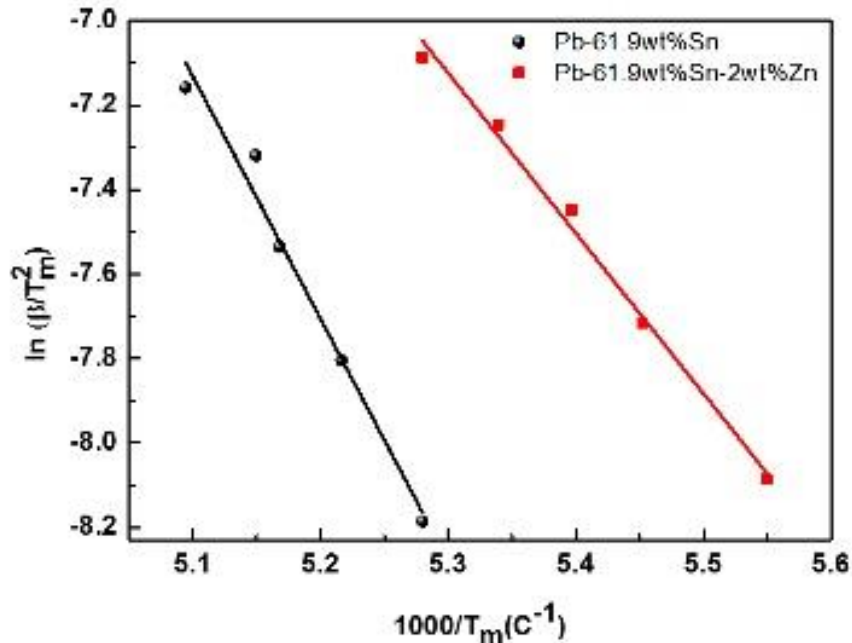


Figure 5:- Kissinger plot of Pb-61.9wt%Sn and Pb-61.9wt%Sn-2wt%Zn

Measuring the enthalpy and specific heat of fusion:-

The heat of fusion is calculated by integrating the area under the melting peak between the peaks start T_{onset} and end T_{end} temperatures. The heat of fusion, ΔC_p , can be determined by Eq. (3) for each material[24]:

$$\Delta C_p = \frac{KA}{m}, \quad (3)$$

Where K is a constant that depends on crucible shape and regards as a constant in the DAT system, m is the mass of the sample, and A is the area under the endothermic peak.

At the melting point, the heat supplied to system will be used supplying the enthalpy of fusion (latent heat of fusion) that is required to convert solid to liquid. Latent heat of fusion is the energy absorbed in a material during its phase change from solid to liquid. For a transformation from solid state to liquid state, enthalpy of fusion can be expressed as:

$$\Delta H \approx \Delta C_p T_m, \quad (4)$$

The enthalpy of fusion (ΔH) and the specific heat change (ΔC_p) were determined because of they are very important parameters for industrial applications. The values of T_{onset} , T_{end} , and ΔH for the two alloys are listed in Table 1. It is obvious that, the fusion heat of Pb-61.9wt%Sn-2wt%Zn is larger than that of the Pb-61.9wt%Sn alloy.

Summary and Conclusions:-

In this work, a detailed analysis of the effects of different heating rates and Zn addition on the melting temperature, heat of fusion, and the activation energy have been investigated. Both Zn addition and heating rates significantly affect the alloys behavior. DTA curves indicate that a small amount of Zn-addition enhances the transition temperature and also melting temperature decrease significantly.

The obtained data on the thermal properties measurements have shown an increase on T_{onset} , T_{end} and T_m values of the samples when the heating rates was increased. The maximum peak in Figure2 is shifted to the higher temperature with increasing heating rates.

In conclusion, this study demonstrated that the heating rate had a significant effect on the values obtained for alloy samples prepared, since the values of T_{onset} , T_{end} and T_m diminish as the heating rate increases. The results showed

that the addition of small amount Zn in Pb–Sn alloy leads to a decrease of thermal parameters (T_{onset} , T_{end} and T_m) and activation energy of fusion.

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Disclosure:-

The authors declare that there is no conflict of interest regarding the publication of this paper.

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RESEARCH ARTICLE

LEAF CLASSIFICATION USING IMAGE PROCESSING: A REVIEW.

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Leaf recognition, Image processing, FFT

Abstract

Earth consists of unicellular and multicellular organisms. Unicellular organisms like bacterium and multi cellular organisms like flora and fauna. Since plant species cannot survive on their own from artificial disasters, it is the responsibility of human beings to protect plant species from such calamities. In order to meet that goal, it is necessary to classify the different plant species and prevent them from extinction. Even though there are huge numbers of databases existing for plant species, there are some species which are still unknown to humans. So it is essential to distinguish the different plant species and classify them into proper categories and protect them. Earlier, manual databases were there for getting this job done. But as technology progressed, automation has its effect in almost all the fields. Classification of plant species through automation is possible through methods like leaf recognition technique. Leaf recognition has its own advantages; apart from the classification plant species, it also helps in recognizing various diseases which can result in their extinction.

Much research works happened in the field of leaf recognition. Most of the researchers used extraction techniques involving spatial or frequency domain. Only few researchers used features extracted using both frequency and spatial domain. In this paper we are trying to come up with a reason for less researches being conducted in the field of leaf database in India.

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Introduction:-

Earth is a huge planet consisting of number of known and unknown species. It is necessary to stand by each other and co-operate with each other and share the resources available in order to lead a happy and prosperous life. Unnecessarily interfering in each other's space may lead to extinction of particular species. [1]Moreover plant species has some special abilities which benefits both flora and fauna. The oxygen has to be circulated in cycles in order to balance the natural consumption. So the cycle is maintained from time to time for the purpose of balancing the life process. The temperature level of atmosphere is also balanced by plant species by the process of transpiration. Leaf recognition is very useful for biodiversity.

Classification is very important when it comes to taxonomy. As a result, such categories of species can be grouped or clustered to a particular location like a park or a dedicated garden. [2]Identification of plant species has many advantages including identification of medicinal plants used in Ayurveda as well as poisonous plants. This process is tedious because human senses are limited and hence identification becomes very difficult especially when there are

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large number of species. So it is necessary to train the computer system using algorithms and develop a system in such a way that it can identify the species by capturing the images of different parts of the plants.

Leaf recognition technique works by identifying the patterns present on the leaf surface and based on the patterns the system can identify the species of the plants. The categorization of both medicinal and poisonous species of plants will help to conserve the medicinal species under extinction and keep away from poisonous plant species and thereby saving lives.

Leaf recognition is based on different characters present on the leaf surface. These characters will determine the category to which a particular plant species belong to. The leaf recognition technique has been followed by botanists to categorize particular plant species. Leaf recognition is limited to factors like climate and plant health because the external surface of the leaf can have change in color due to diseases or climate changes. This makes it even more difficult to proceed with the process.

The climate of a particular region is a key factor which determines the leaf characteristics. The plant has to adopt to different climatic conditions. Thus the plant structure has to be in tune with the particular region in which it is growing. In tropical regions there is high availability of rains and moist winds. Thus the barks and other parts will be thin and the leaves should be capable of enduring strong winds and heavy rains. Desert plants are thornier in nature because loss of water should be prevented. As compared to tropical and desert region of the world, databases on Indian leaf species and South Indian in particular are limited to very small numbers. So my intention is to bring this crisis under attention of the researchers. So that, in future more databases will be generated in South Indian regions. This will in turn help the future researchers to do more study on South Indian leaf classification. Unlike, developed countries, India has limited research facility and resources for students and researchers to work upon. As young researchers require more references, more databases should be made available for further studies. Thus more and more researchers will come forward to invest time and energy in this field of research.

Literature Review:-

There is lot of surveys conducted on leaf recognition from a very long time. Most of the works are concentrated on multiple features of same plant. As time progressed the size of the database started growing and the number of plant species known to mankind also increased considerably. As the number of surveys increases the size of plant database will also increase. The increase in information will result in increase of knowledge regarding plant species. Discovery of more medicinal herbs can cure day to day changing human conditions. Discovery of more poisonous plant species can save lives as human beings and animals can keep away from them. The gap between man and nature can be bridged and reduced day by day.

[3] As part of his contribution, has concentrated on fruit/leaf analysis. He used the technique of wavelet based analysis for classifying the obtained images. This classification helps in identifying the damage caused by pests on fruit/leaf without the involvement of human beings by manually inspecting the fruit/leaf. The method suggested by him has more than 90% accuracy. The method involves neural network for classifying the diseases or damages caused by pests. His method was successful using neural network up to a great extent. Since time consumption is also reduced due to limited human interference in the process.

[4] Proposed a method which is less complicated yet effective in terms of computation using leaf recognition. This method was dedicated for plants with flat leaves which were two dimensional in nature. The base point on the leaf and reference point were decided by the user and with reference to these base points the leaf shape is generated followed by a binary image. Set of features are collected by segmenting the leaves across major axis at the same time segmented across parallel minor axis. The next step is to normalize the feature points. These inputs are taken to neural network.

[5] Here twelve features are used for the purpose of classification. In order to have a faster processing the dimensions are reduced i.e., twelve dimensions are reduced into five dimensions. Probabilistic Neural Network (PNN) technique was used for the above mentioned purpose. Features including leaf area, psychological width, smooth processing, psychological length, perimeter ratio factor, aspect ratio, narrow factor, rectangularity, form factor, perimeter ratio diameter, perimeter ratio of psychological length and psychological width, vein features. Even though there are different sizes of leaves of same plant a standard size should be used, otherwise features of the leaf may vary.

[6] Developed a method for identifying 60 types of leaves. The identification was based on color patterns on the leaves. Even if the plant leaves have the same shape the color on the leaf surface will be different for the leaves. Gray-level co-occurrence matrix (GLCM) and geometric features, Zernike moments were used for the identification of the leaves feature. There were two approaches used for identification. The first being distance measure and the second being Probabilistic Neural Network (PNN).

[7] The proposed technique is based on hybrid image segmentation. A histogram is developed based on the obtained image segmentation. The result of the above operation contains PSNR (peak to signal ratio) and calculation of energy mean square. Based on these two parameters the quality of segmentation is obtained.

[8] Technique of feature extraction and shape analysis is used for leaf recognition. They are given as input for the neural network. The neural network is based on the algorithm with multilayer propagation architecture. The result is ranging between 90 and 94 in its feature category. The error range is between 0-0.7. As a conclusion it is very clear that the accuracy rate is about 94%.

[9] Used 12 features of morphology in which 5 principal variables where is used. In order to classify 32 types of plants by using 1800 leaves probabilistic neural network system was used. The input is obtained using scanners or digital cameras. The following step is to select the base points from the obtained leaves.

[10] Proposed leaf classification based on ontology. Vein recognition is extracted using combined neural network approach as well as thresholding. This will result in improved detailing of leaves. Here automation of the system is carried out by machine learning techniques. For better performance, modified Fourier descriptor and curvature scale space techniques are employed. In order to identify the basic shapes, CCD code system is proposed. Since plant leaf shape is considered to be 2D, an efficient method should be devised to obtain proper characterization of plant species.

[11] Came up with radial basis probabilistic neural network which uses orthogonal least square algorithm as well as optimized recursive square algorithm. The partially damaged leaves are identified using plant recognition. The plant recognition technique is optimized using Fourier descriptors. The final result reveals RBPNN is more efficient when compared to BP neural network and radial basis function neural network.

[12] Proposed a feature extraction technique using natural image. The method comes handy for research students. For obtaining information about leaves, a multidirectional Curvelet transform is employed. The orientation of leaves is not considered, which will enhance the rate of accuracy. SVM classifier uses the coefficients for the purpose of categorization of the obtained results.

[13] Introduced move median centers (MMC) hypersphere classifier for the leaf classification. During classification, pre-processing of data occurs. While doing so it was found that not only storage space is saved but also classification time is reduced. This method is more efficient when compared to contour features because it is very difficult to spot the curvature points.

[14] The approach by them utilized PCA based surface order and complex calculation for the shape highlight delivered a consequence of 89.2% in which the researcher focused on the parameters like angle proportion, conservativeness, linearity. The morphological component extraction strategy has been utilized numerous a times.

[15] Actualized a leaf acknowledgment framework utilizing leaf vein and shape. They proposed the calculation utilizing leaf shape extraction. Projection histogram was utilized to recognize the vein and after that the components were extricated. The exploratory result demonstrated an exactness of 97.19%.

Methodology:-

The remaining parts of plants like flowers are three dimensional in nature. So the process involved in image processing will be more complex as compared to two dimensional image processing. Since the leaf surface is flat it is a two dimensional structure. Image captured can be passed through two dimensional image processing algorithms. After image processing the resultant information characteristics are compared with e-botanists database and the traits will determine the leaf category. This will reduce the human effort by automating the entire process of leaf identification using dynamic programming techniques

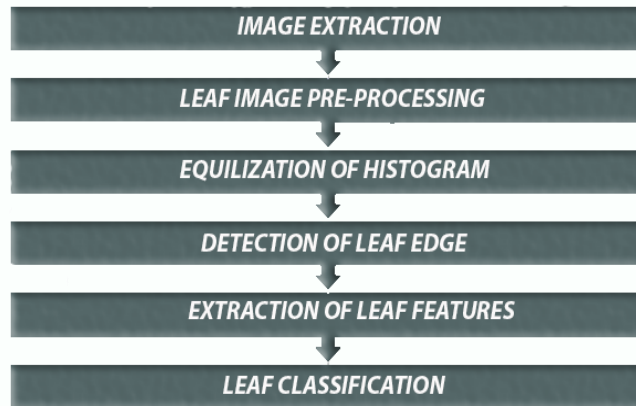


Figure 1:- Image based plant identification system

Image Processing:-

Leaf recognition process involves capturing of images. Image has to be captured using a 2D camera or a scanner. The equipment should be good enough to capture the external surface of the leaves crisp and clear. So that, veins or patterns on the leaves can be properly identified. High quality equipment is used for image capturing. Cameras capable of capturing high definition contents are used for this purpose. Once this process is over the captured image in digital format will be forwarded to the system for further processing. Here 2D cameras are used because the leaf surface is flat and at the same time the amount of details and algorithm complexity required for processing will be very less when compared to 3D image processing. Hence time complexity and space complexity can be reduced up to a great extent.

Image conversion involves the following steps:

- The captured image from the camera in RGB format.
- RGB format will be converted into Grayscale image.
- Grayscale image gets converted to Binary image.
- Next step is to smoothen the image.
- Final pre-processing step involves filtering of the obtained image after smoothening.

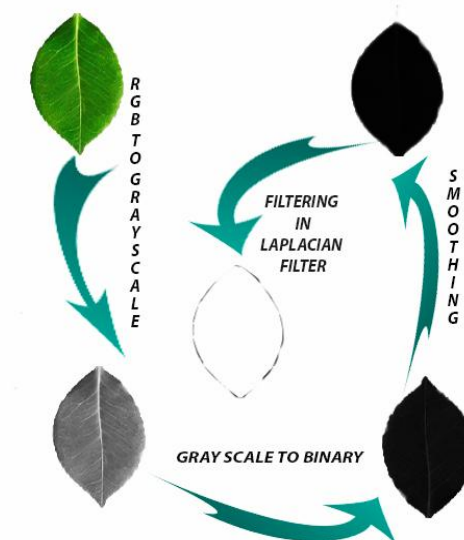


Figure 2:- Image pre-processing steps

The process of leaf recognition involves quite a few steps. The foremost step is to capture the leaf image, followed by two parallel processes. Leaf Contour extraction and Leaf Vein extraction. The following processes will determine contour-centroid distance and leaf direction determined using Projection histogram. The next process is distance sequence rearrange. There are subsequent processes for distance sequence rearrangement including FFT (it can be used for feature extraction), Feature extraction based on geometric and digital morphological features, and finally Feature extraction based on Convex Hull. Using FFT method, analysis is focusing more on frequency domain. Even though same plant can have different shapes for its leaves, the basic structure of the leaves will remain same. This makes the classification easier. After performing these complex techniques, the final result is obtained.

Conclusion:-

A number of methods were discussed in the above surveys. But very few works are available for the future researchers to work on and contribute to their research. As discussed earlier, databases for South Indian leaf species are few in numbers. So my work is to bring the attention of researchers all over the world towards the lack of availability of databases of the endangered leaf species of South India. So that they won't suffer extinction in the near future. There are wide varieties of medicinal herbs available in southern region of India which are facing extinction in the near future. So these species will also be preserved by the creation of South Indian leaf databases. With the creation of more precise methods of extraction of Indian based leaves more plant species can be protected.

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RESEARCH ARTICLE

INTRAORAL PHOTOGRAPHY IN ORAL MEDICINE –THE USE OF SMARTPHONE CAMERA.

Dr. Lakshmi. C and Dr. G. Maragathavalli

A study of Recent Advances in Technology.

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Abstract

The aim of this study was to report the use of smartphone cameras for intraoral photography in oral medicine. The smart phone photographs were evaluated to determine if they would be useful for diagnosis and other applications in oral medicine. In the absence of a standard intraoral camera system, it was found that smart phones could be useful in capturing and transmitting digital intraoral images that could assist in dental education, tele dentistry and other aspects.

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Introduction:-

Due to the technological revolution, the use of smart phone cameras substitutes in the absence of an intra oral camera or DSLR camera set up.

Smart phones are high end mobile phones with advanced computing ability and connectivity. Current models combine many functions and can be accessed via the internet. Smart phones have been sporadically mentioned in literature. The mobility, communication functions, relative to supporting treatment at distant locations, a process known as telemedicine.

The primary purpose of this study was to evaluate the feasibility of dental students acquiring and transmitting intraoral images to the dental specialist using a smart phone technology.

Material & Methods:-

The study was conducted in the Department of Oral Medicine & Radiology of our esteemed institute.

A smart phone with the following specifications were used.

Primary	16 MP, f/2.2, 31mm, OIS, autofocus, LED flash, check quality
Features	1/2.6" sensor size, 1.12 μ m pixel size, geo-tagging, touch focus, face/smile detection, panorama, HDR
Video	2160p@30fps, 1080p@60fps, dual-video rec., check quality

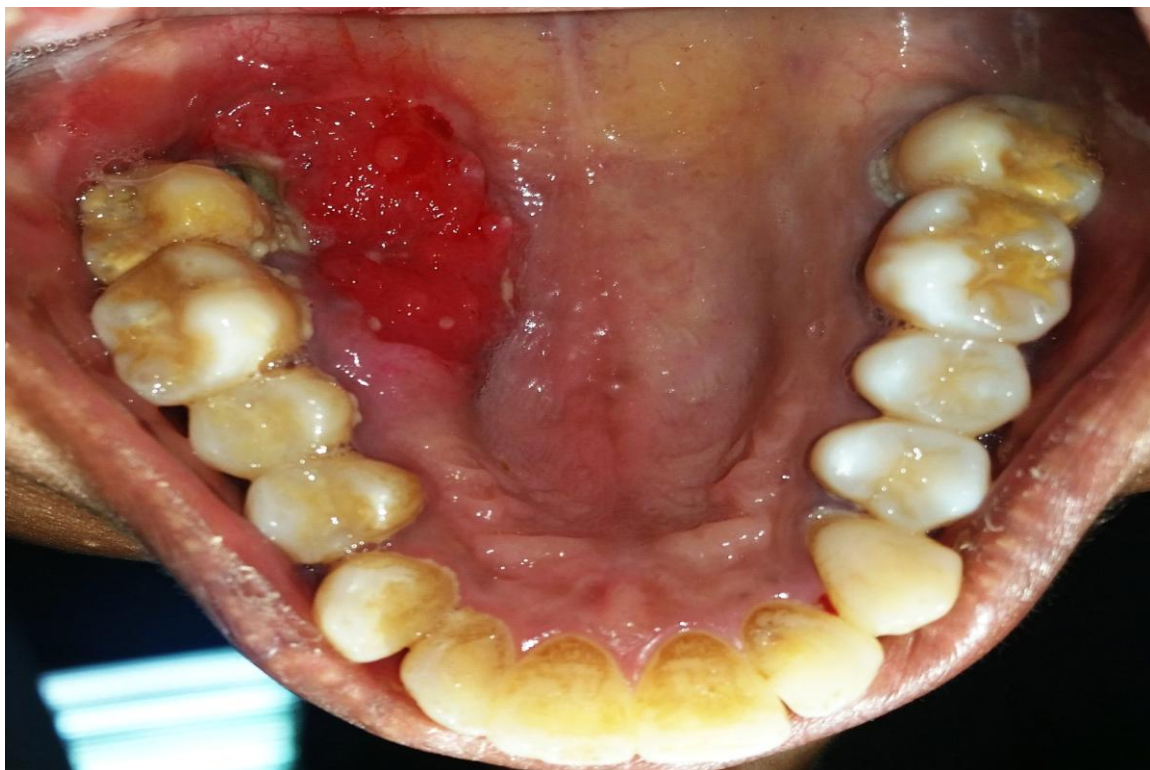
Intraoral photographs with no extra oral imaging were acquired from 4 different patients with four different lesions. Informed consent was obtained and patient's basic information recorded on a proforma. The smartphones were used by the postgraduate students. The brand of the smartphone would be kept hidden and will not be compared to another brand. The students were asked to take photographs in different angles and numbers.

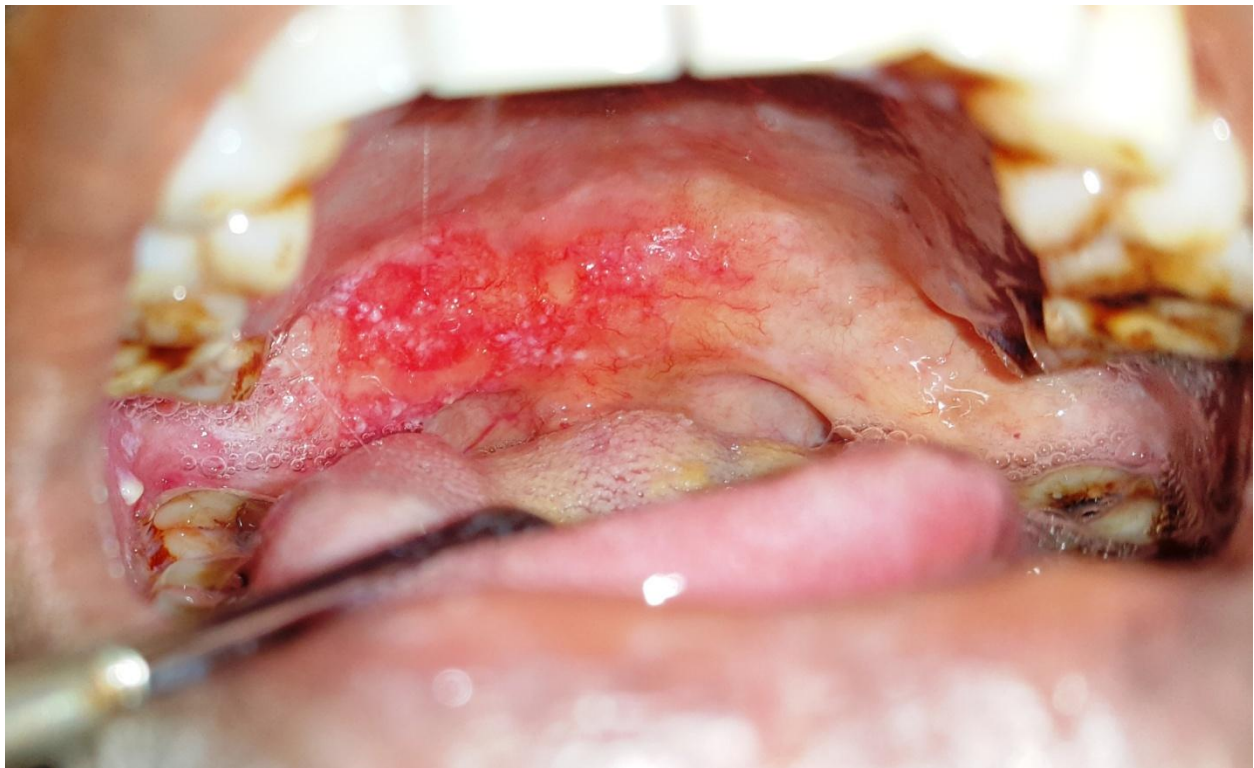
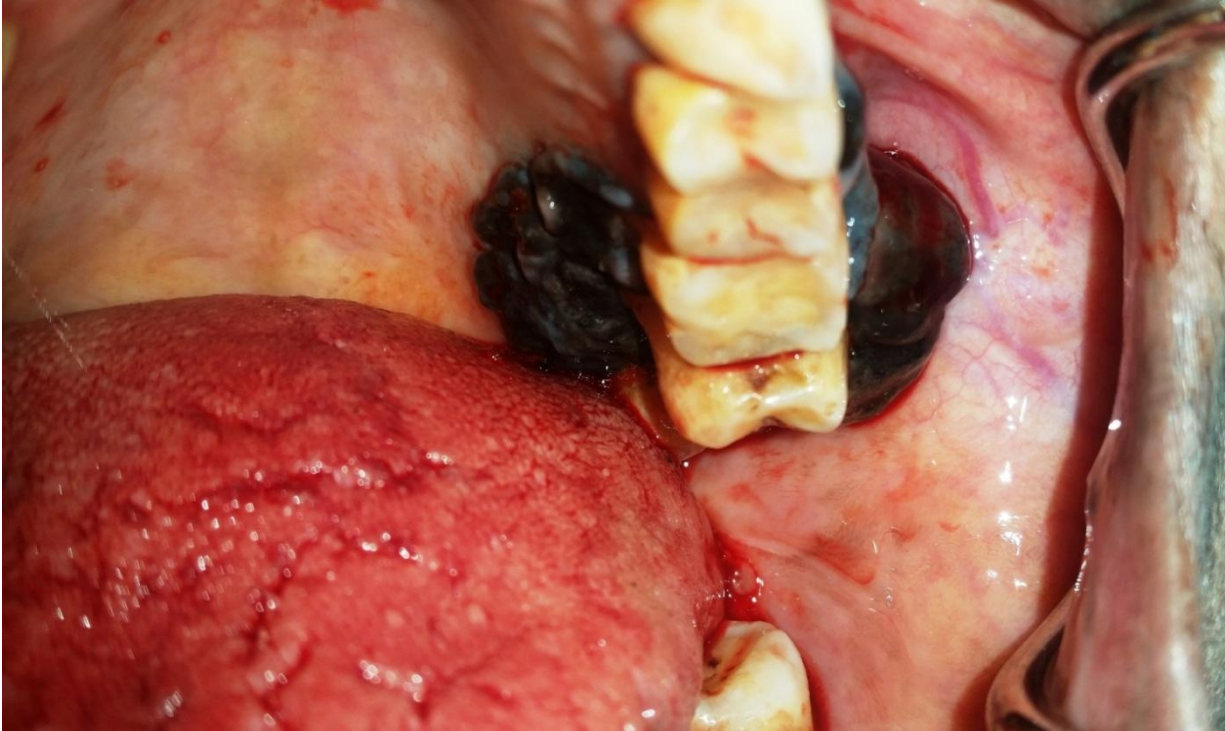
All intra-oral photographs were acquired with the smartphone cameras in a horizontal direction. Metal retractors and intraoral mirrors were used. No external light sources were used. The LED auto lighting of the camera flash was used.

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None of the images were enhanced or edited prior to transfer onto the computer system and were viewed by photography experts.



**Results:-**

The assessment of the photographs were done by the oral medicine specialist and a photography expert. They agreed that , given the characteristics of the intra oral photographs produced by the smart phone, they could use those photographs to assist in diagnosis, treatment planning.

According to the speciality of Oral Medicine, when coupled with the signs and symptoms, smart phone photographs: Could lead to a rank ordered clinical differential diagnosis provided to the practitioner via text messaging or email, with treatment recommendation. This would be beneficial for solo practitioners in locations without access to servers. Could assist in biopsy material submission. Intra operative surgical photos may provide valuable information regarding consistency and form of the lesions.

Discussion:-

The photographs could be taken without a hindrance with the use of metal cheek retractors and intraoral mirror. Tongue retraction however was difficult. If the smart phone device was held steadily, there were very low incidence of blurring of images.

Using smart phone cameras for intraoral photography is not seen as an alternate to the need of using specialized intraoral photographic equipment. As in all, the minimum requirements for the smart phone images to be useful include –

Proper focus on the intended area.

Sufficient lighting.

Consistency of colour of images.

Surprisingly, very few problems or no problems were faced with light source as the LED light being close to the lens in the smart phones.

The smart phones are compact to handle and due to pre specified settings, photographs could be taken more quickly. Intraoral video recording also was done successfully.

Conclusion:-

This project demonstrates that intraoral photographs can be efficiently taken using smart phone cameras and the images relatively quickly transferred or shared among inter disciplinary professionals for quick viewing, patient education and to store inter visit records,

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RESEARCH ARTICLE

THE STUDY OF VERMICOMPOSTING OPTIMIZATION OF ORGANIC WASTE.

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waste, vermicompost, earthworms

Abstract

The study of vermicomposting optimization of organic waste aimed to produce vermicompost from urban raw waste, and to select one of three kinds of worm which had significant effect in the production of vermicompost. The writers conducted a research during June to August 2012, by treating three varieties of worm named: G1 = *Lumbricus rubellus*; G2 = *Lumbricus terrestris*; G3 = *Eisenia foetida*, and used Fully Randomized Design (FRD) for each type of worms. The research variables were C/N ratio, C organic, N, P-2O₅, and K₂O. The writers analysed the vermicompost treatment results in the laboratory, followed by ANOVA. The researchers used gentong (barrel-shaped potteries) as a research tool; and obtained organic waste materials from the local landfill. The result showed that it was possible to produce organic fertilizer from urban waste materials with vermicomposting process using *Lumbricus rubellus*, *Lumbricus terrestris* and *Eisenia foetida*. Moreover, apparently, *Lumbricus rubellus* had the highest influence in the production of vermicompost.

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Introduction:-

Organic fertilizers are largely or entirely composed from organic materials which are derived from plants and animals that have been through a technical process. The fertilizers can be used to supply organic matter, and to improve the physical, chemical and biological soil [1]. Organic fertilizers can be made from waste, because it comprises of inorganic and organic materials. Organic waste consists of plant and animal materials which were taken from nature, or produced from agricultural activities, fisheries or other activities, these rubbish is easily described in the natural processes [2].

The use of waste as an organic fertilizer for direct application to soil generally experiences a variety of problems, for instance: (1) the waste contains inorganic material that is not biologically and easily weathered, such as: glass, plastic, metal, (2) trash has high level of C/N ratio, (3) the composition of organic waste varies too much, and sometimes there is a toxic compound [3]. Other problems are caused by sewage pollution of the environment by heavy metals and chemicals [4]. Toxic heavy metals such as As, Hg, Pb, and Cd can contaminate or damage the environment [5]. Heavy metals As, Cd, Pb, and Hg are very poisonous which can accumulate in crops such as rice, grass, vegetables, and other crops [6].

Vermitechnology is a system in which earthworms are utilized for the bio-conversion of organic waste into vermicompost. Vermicompost has wide applications in organic waste management and has been proven to be an efficient method to manage organic waste materials with diminutive complexity and economic feasibility [7]. Vermicompost is a process to convert organic waste into fertilizer [8]. Taking into account that the organic waste and compost application in agriculture has many positive effects [9], so that it is necessary to conduct research on organic fertilizer with vermicomposting method by utilizing worms performance to process raw materials from waste. Based on the above facts, therefore, the study of vermicomposting optimization of organic waste was conducted, which aims to (1) produce vermicompost from urban waste materials, (2) select one of the three types of worms that have the highest influence in the process of vermicomposting.

Waste:-

Waste is an invaluable material flawed or damaged goods in manufacturing or fabrication, or from excessive materials which are rejected or discarded. Waste is also from discarded materials from human activities and natural processes which do not have economic value. It is the leftover of human day-to-day activities and natural processes that is solid. Specific waste is a waste that needs a special treatment because of its nature, concentration, and volume. Organic waste is waste consisting of plant and animal materials which were taken naturally, or produced from agricultural activities, fisheries or other activities, this kind of waste is easily described in vermicomposting processes.

Vermicompost:-

Vermicompost is a method of waste disposal which is costly effective, fast and environmentally friendly [10]. Urban waste can be effectively degraded by earthworms in vermicompost process resulting in the production of pathogen-free fertilizer [11]. Organic waste can be more useful by converting them into fertilizer using vermicompost process. Although the nutrients contained in the fertilizer having no significant increase, the the ratio of C/N undergoes significant decrease making plants easily absorb the substance. Vermicompost plays an important role in protecting the environment because it uses waste as its raw materials, helps building soil fertility and improves soil health for sustainable agriculture [12].

Vermicomposting is in the same page with the principle of healthy environment because it has the value of resource conservation and sustainable practices, as a process for handling organic residue which is an alternative approach in waste management, which is not dumped or incinerated but recycled [13].

In the composting process, the C/N ratio is one of the important parameters that give an indication of the decomposition rate, and it is proven that vermicomposting decrease the ratio of C/N significantly [14].

Vermicompost can significantly improve the growth and yield of vegetables and fruits. The effect of vermicompost on plant is caused by the quality of the mineral provided. It is also to regulate the growth of other components such as plant growth hormones, and to improve soil quality by increasing microbial activity [15]. Giving efficient conversion of organic waste, stabilizing the soil conditions, helping in reducing the population of microbial pathogens and heavy metal toxicity, relatively easy and inexpensive economical technology, as well as its function as the most environmentally safe nutritional supplement for organic crop production is many advantages of using vermicompost [16].

Earthworms:-

Lumbricus has flattened body shape, and body segments around 90-195. Klitelum lies in the 27th-32th segment. *Lumbricus rubellus* has relatively small body size with a length of 8-14 cm; the body color, especially the back, is light beige to reddish purple. Most part of the belly is beige, and tail is yellowish; the shape of dorsal is rounded and vertically flat; the number of segment on the klitelum are around 6 - 7 segments; the male sex hole lies in the 13th segment; its movement is slow; and its body contains 70-78% of water [17]. Meanwhile, *Eisenia foetida* is 3 – 10 cm in body length, 0.4 – 0.6 g in body weight, 50 – 55 days of lifespan, cocoon production is 1 in every 3 days, cocoon incubation is 20 – 23 days [16].

Research Method:-

The study began in June to August 2012. The place of extraction of urban waste materials was in the landfill of Sekoto Village, Badas Sub-district, Kediri regency. The vermicomposting research process was conducted in Lirboyo Kediri, East Java, Indonesia.

The experiment was carried out using three types of worms G1 = *Lumbricus rubellus*; G2 = *Lumbricus terrestris*; G3 = *Eisenia foetida*, each worm was treated repeatedly for approximately three times using fully randomized design (CRD). The experimental variable is the content of: C/N ratio, C organic, N, P₂O₅, and K₂O, statistically analyzed by ANOVA. Laboratory analysis for C organic was measured using the Walkey-Black method, N using the method of Kjeldahl, P and K using calorimetric and photometric methods, C/N ratio was calculated from the measured values of C and N, the elements of As, Pb and Cd using AAS (Atomic Absorption Spectrophotometry), and Hg using Cold Atomic Absorption of Mercury Analyser.

Gentongs (barrel-shaped potteries) were used as the container during the experiment, and the experimental materials were organic wastes and three types of worms namely *Lumbricus rubellus* and *Eisenia foetida*, both worms were imported from wormbreeders in Bandung regency of West Java, while *Lumbricus terrestris* were taken from Kediri, East Java, Indonesia.

Research Implementation:-

The study began in June to August 2012. The place of extraction of urban waste materials was in the landfill of Sekoto Village, Badas Sub-district, Kediri regency. The vermicomposting research process was conducted in Lirboyo Kediri, East Java, Indonesia.

The research was started from the preparation of organic waste as the medium and worm food. Worm could not live in a pile of garbage in landfills, the waste needed to be sorted between organic and non-organic waste. Organic waste was composted using EM-4 and was left for 30 days, then it was used for the research materials. These materials were tested in the laboratory before being used; the result was used as a control in the statistical analysis. Vermikomposting has been proven to be better technology than the compost and more suitable for the management and recovery of nutrients from urban waste [18].

Nine barrel-shaped potteries were placed on the ground randomly. Sand was used to keep the moisture by watering them every day. Low temperature and bright light condition will disturb the earthworm; however, when the temperature is warm and humid, it will be favorable for the worm [19]. The barrel-shaped potteries were filled with organic waste, each as much as two pounds, then one kilogram of worms per barrel was put inside of them. Earthworms eat variety of organic wastes, the number of foods per day is equal to his weight [20]. One kilogram of earthworms can process one kilogram of waste per day, and produces 0.5 pounds of earthworms' waste [17].

In every three potteries, they were filled with the same type of worm. G1.1, G1.2, G1.3 = *Lumbricus rubellus*; G2.1, G2.2, G2.3 = *Lumbricus terrestris*; G3.1, G3.2, G3.3 = *Eisenia foetida*. Next, the barrels were closed using gauze so that the worms could not get out.

After two days, all the contents of the barrels were removed and separated between worms and vermicompost. It was estimated, that the organic waste would have been eaten by the earthworms in two days. Earthworms refuse to stay in their own excrement (vermicast) for a long time and die if there is no food available [19]. In the end, Vermicompost in each treatment was collected to be tested in the laboratory.

Result and Discussion:-

The vermicomposts were tested in the laboratory of soil science, Faculty of Agriculture, University of Brawijaya, Malang, Indonesia, to obtain the value of the variables. Further, they were statistically analysed using ANOVA, the result is as follows:

The analysis of the organic C content from the three types of worm treatment is presented in Table 1.

Table 1:- The Organic C content

Treatment	C organic %
Control	22,2 b
<i>Lumbricus rubellus</i>	6,01 a
<i>Lumbricus Terrestris</i>	5,84 a
<i>Eisenia foetida</i>	6,03 a
5% LSD	0,59

Description: The numbers followed by the same letter are not significantly different at 5% LSD.

The content of C-organic before worm treatments was 22.2%, after the worm treatments of *Lumbricus rubellus*, *Lumbricus terrestris* and *Eisenia foetida*, all organic C content decreased to 6.01%, 5.84% and 6.03%, significantly different from the previous treatment. The decomposition process is the release of carbon from complex into more simple bonds due to the use of C elements by the organism to get their life energy through respiration and biosynthetic processes, releasing CO₂ to organic material so that the organic materials that have undergone the decomposition process will have lower levels of C, compared to the C levels of raw materials [21].

The result of N content analysis from the three types of worm treatment is presented in Table 2.

Table 2:- Total N content

Treatment	N total %
Control	0,49 a
<i>Lumbricus rubellus</i>	0,63 b
<i>Lumbricus Terrestris</i>	0,69 b
<i>Eisenia foetida</i>	0,76 c
5% LSD	0,06

Description: The numbers followed by the same letter are not significantly different at 5% LSD.

The content of N-total before worm treatment was 0.49%, and after the worm treatment of *Lumbricus rubellus*, *Lumbricus terrestris* and *Eisenia foetida*, all content of N-total increased up to 0.63%, 0.69% and 0.76%, significantly different from the previous treatment. This is caused by the weathering process of mineralization of organic matter involving the performance of enzymes that hydrolyze the protein complex that will increase the nitrogen content in the vermicompost [22], on the other hand, there is a simultaneous addition of nitrogen by worms in the form of mucus and excretory material, this process occurs in a high intensity so that the content of nitrogen will increase [18].

The result of the C/N ratio content from the three types of worm treatment is presented in Table 3.

Table 3:- The C/N ratio content

Treatment	C/N %
Control	46,08 b
<i>Lumbricus rubellus</i>	9,54 a
<i>Lumbricus Terrestris</i>	8,46 a
<i>Eisenia foetida</i>	7,93 a
5% LSD	6,52

Description: The numbers followed by the same letter are not significantly different at 5% LSD.

The C/N ratio before worm treatment was 46.08, however, after the worm treatment of *Lumbricus rubellus*, *Lumbricus terrestris* and *Eisenia foetida*, the content of C/N ratio decrease up to 9.54, 8.46 and 7.93, all of them decreased significantly. The decline of C/N ratio was caused by the decreasing number of carbon as well as the increasing number of nitrogen.

The decomposition process is the release of carbon from complex into more simple bonds due to the use of C elements by the organism to get their life energy through respiration and biosynthetic processes, releasing CO₂ to organic material so that the organic materials that have undergone the decomposition process will have lower levels

of C, compared to the C levels of raw materials [21]. The decreasing amount of C/N ratio is also due to the increasing content as the result of mineralization, a process that changes the organic nitrogen into inorganic nitrogen through the weathering process which involves the work of enzymes that hydrolizes protein complexes, as well as the nitrogen that was produced from the worms' excrement [23].

P2O5 content analysis of the results of treatment of various worms are presented in Table 4.

Table 4:- The P2O5 content

Treatment	P2O5 %
Control	0,57 a
<i>Lumbricus rubellus</i>	0,97 d
<i>Lumbricus Terrestris</i>	0,78 c
<i>Eisenia foetida</i>	0,68 b
5% LSD	0,02

Description: The numbers followed by the same letter are not significantly different at 5% LSD.

The P2O5 content before worm treatment was 0.57%. Nevertheless, after the worm treatment of

Lumbricus rubellus, *Lumbricus terrestris* and *Eisenia foetida*, all contents of P2O5 increased up to 0.97%, 0.78% and 0.68%, significantly different from the treatment before. The total P value at the end of vermicomposting process will be higher than the initial value at the beginning of the process, this indicates that there is Phosphorus mineralization process [18]. In general, when the organic materials is passing through the worms' digestive organs, most of the phosphorus will be converted into the form of dissolved P by enzymes in the worm's digestive organs, namely acid phosphatase and alkaline phosphatase. Afterward, the P element will be released by microorganisms in the worm excrement [24].

K2O content analysis of the results of treatment of various worms are presented in Table 5.

Table 5:- The K2O content

Treatment	K2O %
Control	0,04 a
<i>Lumbricus rubellus</i>	0,07 b
<i>Lumbricus Terrestris</i>	0,10 c
<i>Eisenia foetida</i>	0,07 b
5% LSD	0,013

Description: The numbers followed by the same letter are not significantly different at 5% LSD. The content of K2O before worm treatment was 0.04%; however, after worm treatment of *Lumbricus rubellus*, *Lumbricus terrestris* and *Eisenia foetida*, all content of K2O raised up to 0.07%, 0.10% and 0.07%. It was significantly different compared to the initial treatment. Probably, this was caused by the fact that the remains of plants from organic waste was a source of potassium, and potassium is classified as mobile elements not only in the plant cell, but also in the plant tissue, as well as in the xylem and phloem. Potassium can also be found in the cytoplasm. The existing K element on the substrate is converted into a soluble form by the existing microorganisms in the gastrointestinal worms [18], which makes the element of potassium higher than the control which was treated without any worm treatment.

The content of heavy metal in organic waste before worm treatment, and the heavy metal content in the vermicompost, is descriptively presented in Table 6 as follows.

Table 6:- The Heavy metal content

Treatment	Pb	Hg	Cd	As
Organic wastes (pre-treatment)	21	0,7	5	22
EM-4 treatment	20	0,6	4	20
<i>Lumbricus rubellus</i>	tu	tu	tu	tu
<i>Lumbricus terrestris</i>	18	tu	tu	tu
<i>Eisenia foetida</i>	tu	tu	tu	tu

Description: tu = not measurable with AAS analytical methods and CA3M

The result from the laboratory said that the organic waste contained 21 ppm Pb, 0.7 ppm Hg, Cd 5 ppm and 22 ppm As. Next, the composting process using EM4 was done resulting in the decrease of Pb content to 20 ppm, Hg to 0.6

ppm, Cd to 4 ppm and As to 20 ppm. After the worm treatment with three types of worms, vermicompost contained no heavy metal such as Pb, Hg, Cd and As, but the vermicompost with the worm treatment of *Lumbricus terrestris* that contained Pb approximately 18 ppm. It is assumed that the process of vermicompost can reduce heavy metal toxicity [12] [16]. Worms can absorb high concentration heavy metals; the process is, worms absorb the heavy metal particles at first, and coat each particle with a specific protein which prevents these metals to interact with the inside of the body [25]. Worms can also absorb arsenic [26].

Next, regression test was conducted to test the effects of the worms in producing the vermicompost and to find out which one is the most influential worm in producing the vermicompost according to the standard of Minimum Technical Requirements of Organic Fertilizer by Permentan (The Regulation of Ministry of Agriculture) No. 28 / SR.130 / 5 / 2009. The regression analysis showed the value of F was 82.493 > F table 6.23. It could be concluded that the vermicompost from urban solid waste can be made using *Lumbricus rubellus*, *Lumbricus terrestris* and *Eisenia foetida* worm. Presumably, there was one of the most dominant worms to produce the vermicompost, it had been

proven using the regression analysis to select the highest value on the coefficient of partial determination or r^2 using SPSS 17.0 for Windows, and the results are presented in Table 7 below.

Table 7:- partial results coefficient of determination regression analysis.

Treatment	r	r^2
<i>Lumbricus rubellus</i>	0,954	0,910
<i>Lumbricus terrestris</i>	-0,887	not significantly
<i>Eisenia foetida</i>	-0,905	not significantly

The analysis showed that the effect of *Lumbricus rubellus* was 0.910 or 91%. This suggested that *Lumbricus rubellus* had contributed partially to the production of 91% vermicompost fertilizer. While the coefficients of partial determination for two other variables were negative, the negative values were less influential or not significantly meaningful so it could be ignored. From the results of this analysis, it could be concluded that the *Lumbricus rubellus* was a worm that had the highest influence in the production process of vermicompost. According to [27], *Lumbricus rubellus* is a major waste eater and it is also considered as the biodegrader earthworm species, they are used throughout the world for waste degradation and being the most important and successful thing in the ecology management of urban organic waste.

Conclusion:-

1). It is possible that urban waste could be made into vermicompost using the three types of worm (*Lumbricus rubellus*, *Lumbricus terrestris* and *Eisenia foetida*.) However, this vermicompost must still be in accordance with the standard of Minimum Technical Requirements of Organic Fertilizer by Permentan (The Regulation of Ministry of Agriculture) No.28/Permentan/SR.130/5/2009.

2). From the study, among the three worms, *Lumbricus rubellus* showed the most significant influence in producing many important contents in the process of vermicomposting.

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RESEARCH ARTICLE

Prevalence of Nutritional Anaemia among patients reporting to Saveetha Dental College: A SHORT STUDY.

Dr. Lakshmi. C and Dr. G. Maragathavalli.

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Abstract

Nutritional anaemia affects all the age groups and sex in developing countries like India. Iron deficiency is the most common micronutrient deficiency in the world affecting more than 2 billion people. In the present study, 50 patients were selected who were subjected to routine hemogram and were diagnosed anaemic. 2 ml of venous blood was drawn and analysed using autoanalyser method and were interpreted according to the WHO criteria. The study showed that 13 % of the patients were mildly anaemic, 34% were moderately anaemic and 03% were severely anaemic. Various factors like social, economic status, age, sex, social class, dietary habits are some of the major etiological factors for nutritional anaemia.

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Aims & Objectives:-

Aim:-

To study the prevalence of nutritional anaemia among the patients.

Objectives:-

Measuring the incidence of anaemia among the selected patients using hemoglobin percentage as cut off value provided by WHO.

To find out correlation between dietary habits and prevalence of anaemia.

Introduction:-

Nutritional anaemia is a worldwide problem with the highest prevalence in the developing countries. It frequently occurs due to inadequate iron intake, chronic blood loss or disease, malabsorption or a combination of these factors. Nutritional anaemia is prevalent all over the world with an estimated 2 billion people being iron deficient and is one of the most common nutritional disorders in the developing world. With an average prevalence of 40% among the general population that it affects nearly two third of pregnant and one half of non pregnant women which is three to four times higher than that in the developed countries, where prevalence of anaemia is between 4-12% among women of child bearing age (WHO/UNICEF 2001). Iron deficiency can arise either due to inadequate intake or poor bioavailability of dietary iron or due to excessive losses of iron from the body. Although most habitual diets contain adequate amounts of iron only a small amount (less than 5 %) is absorbed. This poor bioavailability is considered to be a major reason for the widespread iron deficiency. Women lose a considerable amount of iron especially during menstruation. Most commonly, people with anaemia report feelings of weakness, fatigue, general malaise and sometimes poor concentration and dysnoea on exertion.

In India, recent data from the District Nutrition Project (ICMR) in 16 districts and 11 states on prevalence of anaemia in non-pregnant adolescent girls (11-18 years) showed rates as high as 90.1% with severe anaemia ($Hb \leq 7 \text{ gm/dl}$) in 7.1% (Teoteja G.S, Singh P, 2002).

By far, the most frequent cause of nutritional anaemia is iron deficiency and less frequently folate or Vitamin B12 deficiency.

Materials & Methods:-

A cross sectional study was done among patients reporting to the institute. The age group of the patients were from 18- 65 years.

A total of 50 patients were taken. Study procedure was explained to the patient and a written informed consent was taken. All the patients' demographic details, habit history, dietary patterns etc., were collected in a structured questionnaire. A detailed clinical history taken from them for any present symptom regarding anaemia and physical examination was done to look for anaemic signs and symptoms and the same was noted in the proforma. The data and privacy of the patients were protected. 2 ml venous blood was obtained and the blood hemoglobin was estimated using the autoanalyser method and interpreted as per WHO criteria. Anaemia is established if the hemoglobin is below the cut off points as recommended by WHO.

INDICATOR	HB (gm/dl)
Non - anaemic	≥ 13 (males); ≥ 12 (females).
Grade 1 (mild) anaemia	10.0 -11.9
Grade 2 (moderate) anaemia	7.0-9.9
Grade 3 (severe) anaemia	≤ 7

Results:-

In the present study anaemia was observed in all the patients.

Figure 1:- Sex wise distribution of anaemia among patients.

MALES	FEMALES	TOTAL
03(6%)	47(94%)	50(100%)

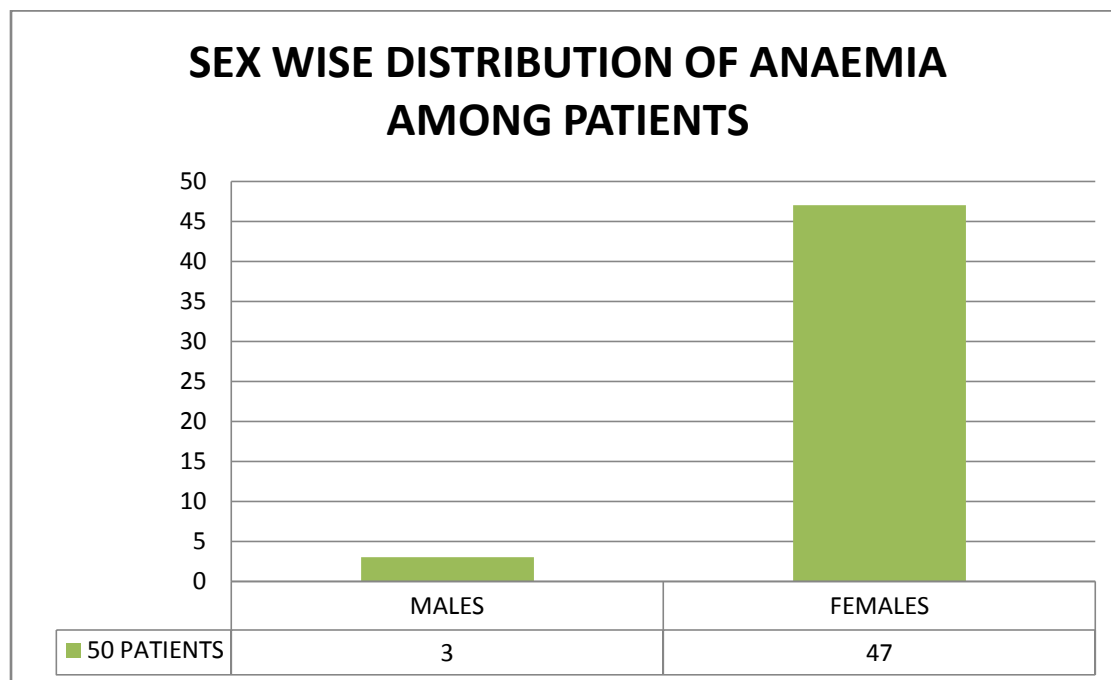


Figure 2:- Sex wise grading of severity of anaemia (WHO Criteria).

GRADING	MALES	FEMALES	TOTAL
MILD	1(1%)	12(12%)	13%
MODERATE	2(2%)	34(34%)	34%
SEVERE	0(0%)	03(3%)	03%

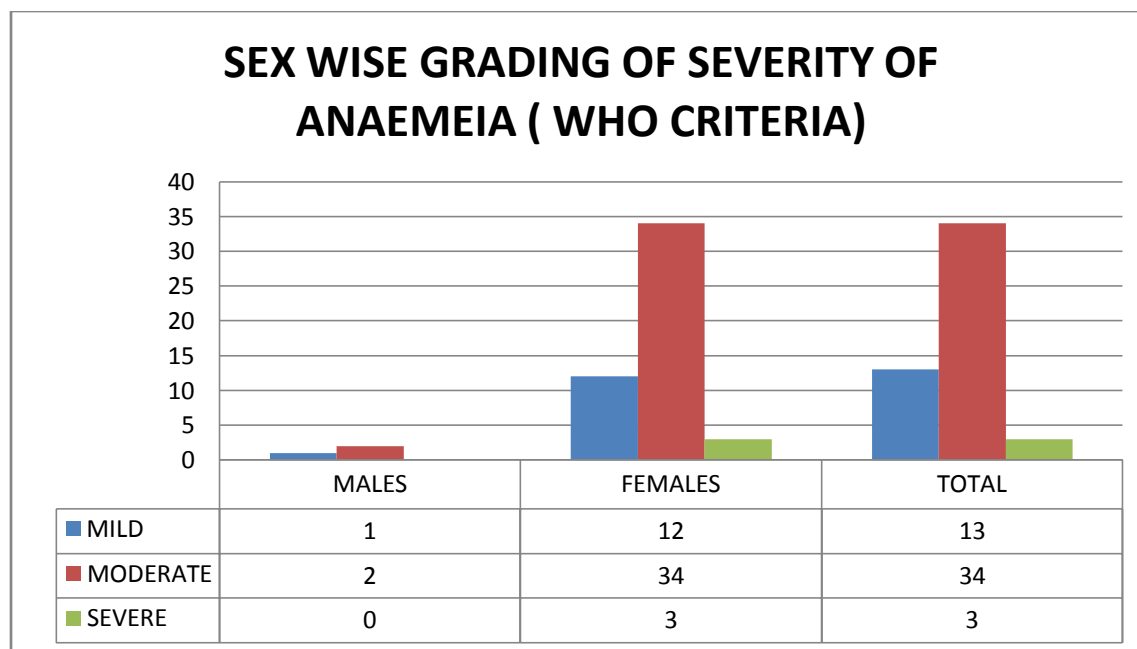
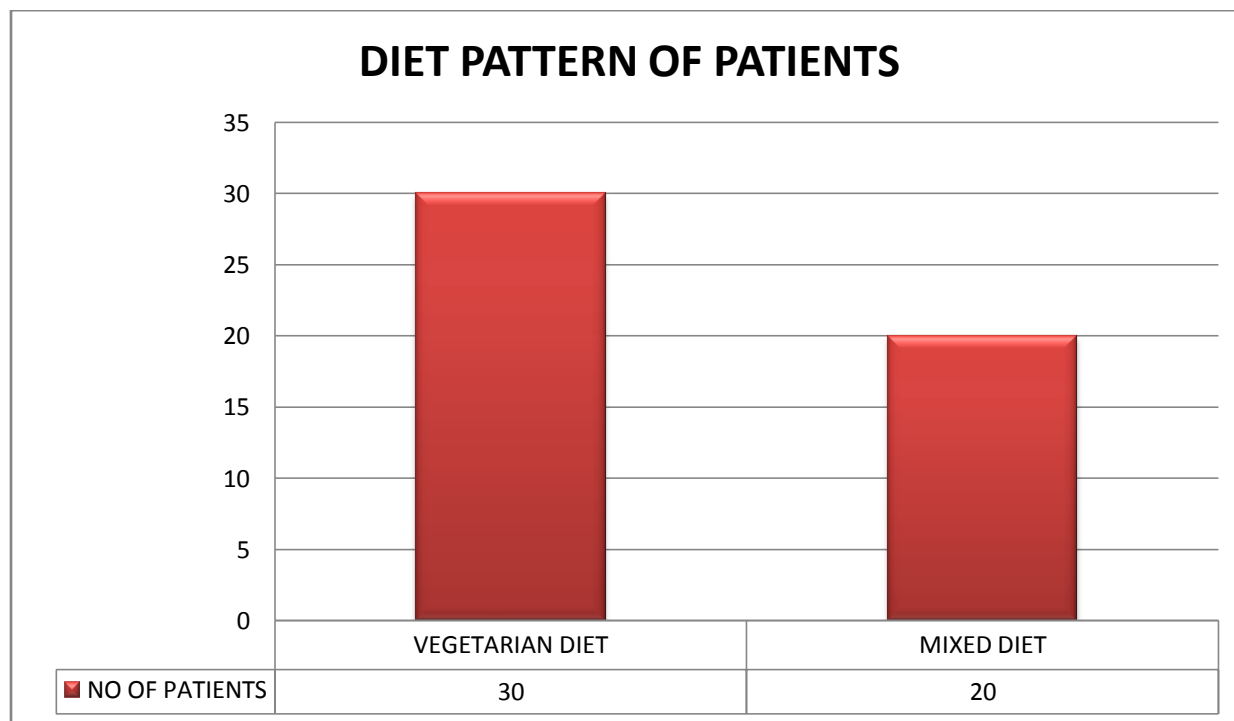


Figure 3: Dietary Pattern Of Patients :

VEGETARIAN DIET	30 (60%)
MIXED DIET	20 (40%)



Discussion:-

Nutritional anaemia is a major health threat to the developing countries. Interventions like iron and folic acid supplementation and other strategies such as diet modification, infection control, nutrition education must be undertaken.

Iron deficiency can be caused due to either inadequate intake, poor bioavailability of dietary iron or due to excessive loss of iron from the body.

Nutritional anaemia especially iron deficiency anaemia is more prevalent among women in the adolescent and reproductive age group due to menstrual blood loss, poor diet, bleeding during parturition etc., Patients must be monitored periodically, iron supplements and diet counseling should be given. In this study there is no significant correlation between the diet pattern of the patient and anaemia.

Conclusion:-

Nutritional anaemia especially iron deficiency anaemia is more prevalent among females especially adolescent girls due to causes like menstrual blood loss, poor diet and under nutrition as compared to males. Nutritional anaemia is easily preventable as well as treatable and the available measures are relatively affordable. Iron supplementation is required for this group of patients. Periodical screening, health check ups and routine hemograms must be performed. The patients should be motivated and educated about the importance of a balanced diet, nutritional supplements. If anaemia is severe, high doses of iron or blood transfusion may be necessary.

Further studies with large sample size are required to estimate the exact prevalence of nutritional anaemia in the target population so that appropriate preventive and treatment measures can be initiated.

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RESEARCH ARTICLE

ACUTE STROKE IN CHILDREN: AN OVERVIEW.

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Abd El-Azez and Ibrahim Elmenshawi.**

Manuscript Info

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Key words:-

stroke, childhood, etiology, clinical features, neuro-imaging.

Abstract

Objective: The aim of this study was to delineate the different characteristics of stroke in children and their outcome.

Method: Clinically diagnosed stroke in children patients underwent general, full neurological examination and radiological investigation with evaluation of severity by pediatric National institute of health scale of stroke (NIHSS) and outcome disability by using Modified Rankin Scale for children (mRS).

Results: The study was conducted on 60 patients with mean age \pm standard deviation was (5.98 ± 5.65) , median age 3.5 (1-18), 32 (53.3%) of them were male and 28 (46.7%) were female. Most of patients (83.3%) arrived to hospital after 12 hours from the stroke onset. Etiologically, 12 (20%) patients had congenital heart disease, 8 (13.3%) had hemorrhagic diseases, while 23 (38.3%) were cryptogenic. Ischemic stroke represent 83.3% of all patients. MRI brain imaging had better evaluation in patients with hemorrhagic stroke (2 of 6 patients (33.3%) revealed underlying pathology) rather than ischemic ones (only 2 of 44 ischemic stroke patients (4.5%) showed hemorrhagic transformation in one case and vascular anomaly in the other). Twenty (33.3%) patients had mild severity using pediatric NIHSS on admission, 35 (58.3%) were mild to moderate, 3 (5.0%) were severe and 2 (3.3%) were very severe. mRS for children after three months revealed 3 patients (5.0%) with complete recovery, 14 (23.3%) had no significant disabilities despite symptoms in clinical examination, 18 (30.0%) had slight disability, 10 (16.7%) with moderate disability, 2 (3.3%) had moderately severe disability while 13 (21.7%) patients died. There was significant weak positive correlation between severity of stroke at the onset and disability outcome three months later.

Conclusion: Etiological factors in childhood are quite different from those present in adulthood. Despite a careful investigation, some causes of stroke in children remain unidentified. MRI brain imaging is modality of choice in patients with hemorrhagic stroke.

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Introduction:-

The word stroke is used to refer to a rapidly developing focal or global neurological deficit lasting more than 24 hours if it has a vascular basis. The term "children" is between one month and 18 years of age (1), (2). In adults,

aetiology and outcome of stroke is well known, but it is quite few information about children especially in Egypt. The studies of stroke in children are few with variable results and under estimation of hemorrhagic stroke (3). The aim of this study was to delineate the different characteristics of stroke in children and their outcome.

Material and Methods:-

An informed consent was taken from at least one of each child parents. This study was conducted on sixty clinically diagnosed strokes in children patients obtained from Mansoura University Hospitals and other governmental hospitals in Mansoura from July 2011 till April 2016. With the following *Inclusion criteria*: stroke in children between one and 18 years of age, both hemorrhagic and ischemic strokes in children and both parenchymatous and non parenchymatous stroke in children. *Exclusion criteria*: includes stroke in the first year of life, in hospital stroke in children and hemorrhagic brain tumors in children. All patients were subjected to the following: Detailed history, including previous medical diseases and hospital delay duration in hours (<6 hours, 6-12 hours, 12-24 hours, 24-48 hours and >48 hours) according to Charlotte Martin et al. (4). General examination: with the following clinical items that should be considered in the diagnosis of stroke in children: Body mass index, measurement of head circumference, the skin should be examined for rash, signs of trauma, splinter haemorrhage or embolic skin rash and detailed cardiovascular examination including blood pressure. Neurological examination: Pediatric (NIHSS) at three points (on admission, on discharge and three months later) (5), fundoscopic examination for papilloedema or hemorrhage and (mRS) for children at three points (on admission, on discharge and three months later) (6). Laboratory investigations and radiological examination: Chest x-ray, electrocardiogram, echocardiogram, brain computed tomography, brain magnetic resonance imaging. The study was approved by the institutional board review of Mansoura University, faculty of medicine.

Statistical Analysis Of The Data:-

Data were fed to the computer and analyzed using IBM SPSS software package version 20.0. Qualitative data were described using number and percent. Quantitative data were described using (minimum and maximum), mean, standard deviation and median. Significance of the obtained results was judged at the 5% level. The used tests were Monte Carlo correction for categorical variables, to compare between different groups Correction for chi-square when more than 20% of the cells have expected count less than 5.

Results:-

The study was conducted between 2011 and 2016 on 60 patients with (mean age \pm standard deviation) (5.98 ± 5.65), median age 3.5 (1-18), 32 (53.3%) of them were male and 28 (46.7%) were female (Table.1). As regard distribution of studied patients according to their presenting complaints, lateralization was the most common presentation (in 44 (73.3%) patients), disturbed conscious level was found in 24 (40%) patients, convulsions in 21 (35%) patients, headache and/or vomiting in only 10 (16.7%) patients (Table.1). According to the hospital delay duration of studied patients, most of patients (56.7%) arrived to hospital within 12-48 hours, 16 (26.7%) patients were arrived after 48 hours while only 10 (16.7%) patients were arrived before 12 hours (Table.2). Twenty (33.3%) patients were mild severity using pediatric NIHSS on admission, 35 (58.3%) were mild to moderate, 3 (5.0%) were severe and 2 (3.3%) were very severe (Table.3). Cardiac evaluation of our cases revealed 12 (20%) patients with cardioembolic stroke (Table.4). Three cases had transposition of great arteries one of them associated also with patent ductus arteriosus (PDA) and atrial septal defect (ASD). Ventricular septal defect (VSD) proved in 2 patients. Infective endocarditis with VSD was seen in 2 cases. There was one case for each of the following: PDA, restrictive cardiomyopathy, mitral prolapse with mitral regurge, atrial myxoma with left atrial septal thrombosis and complex congenital heart disease (secondary to Down syndrome). Forty four (73.3%) patients were discovered ischemic stroke in CT brain imaging, however by MRI brain imaging, only 2 of them showed hemorrhagic transformation in one case and arteriovenous malformations (AVM) in the other, while 10 (6.7%) patients showed intracerebral hemorrhage (ICH) by CT brain imaging, 4 (6.7%) of them died before doing MRI brain imaging and 2 of the remaining 6 patients (33.3%) showed underlying pathology by MRI brain imaging (AVM and venous angioma) (Table.4).

The rest of the patients in our study were cryptogenic (38.3%), with 2 cases had AVM, 2 had moyamoya disease and one had venous angioma (Table.5).

Most of our cases (58.3%) showed no to mild disability after three months using mRS for children; three patients (5.0%) were completely recovered, 14 (23.3%) had no significant disabilities despite symptoms in clinical examination and 18 (30.0%) had slight disability. Moderate to severe disability was proved in (20%) of our patients;

10 (16.7%) patients showed moderate disability and 2 (3.3%) had moderate severe disability. The rest of patients (21.7%) died (mRS score 6) (Table.3). There was significant weak positive correlation between severity of stroke at the onset and disability outcome three months later (Table.3).

Table.1:- Age, gender and complaint

Age: Mean \pm Standard deviation	5.98 \pm 5.65
Gender: Number %	
• Male	32 (53.3%)
• Female	28 (46.7%)
Complaint: Number %	
• Lateralization	44 (73.3%)
• Disturbed conscious level	24 (40.0%)
• Headache/Vomiting	10 (16.7%)
• Convulsions	21 (35.0%)

Table.2:- Distribution of studied patients according to their hospital delay duration

	Studied patients n=60	
Hospital delay (hours)	N	%
<6	5	8.3%
6-12	5	8.3%
12-24	17	28.3%
24-48	17	28.3%
>48	16	26.7%

Table.3:- Distribution of studied patients according to NIH stroke scale on admission, on discharge and after 3 months

NIH	Studied patients n=60	
• On admission	N	%
Mild	20	33.3%
Mild to moderate	35	58.3%
Severe	3	5.0%
Very severe	2	3.3%
mRS	Studied patients n=60	
• After 3 months		
0	3	5.0%
1	14	23.3%
2	18	30.0%
3	10	16.7%
4	2	3.3%
5	0	0 %
6	13	21.7%
		mRS 3 months
NIHSS on admission	r	0.431
	p	0.001*

Table.4:- Distribution of studied patients according to the radiological investigations

Radiology	Studied patients n=60	
	N	%
ECHO		
• Negative	48	80.0%
• Positive	12	20.0%
CT		
• Ischemic	44	73.3%
• Hemorrhagic	10	16.7%
• Hemorrhagic infarction	6	10%
MRI		
• Died (hemorrhagic stroke)	4	6.7%
• Ischemic	42	70%
• Hemorrhagic	4	6.7%
• Hemorrhagic infarction	7	11.7%
• Space occupying lesions	3	5.0%

Table.5:- Distribution of studied patients according to their etiology

Etiology	Studied patients n=60	
	N	%
1. Cryptogenic	23	38.3%
2. Vasculitis	4	6.7%
3. Congenital Heart Disease, Rheumatic Heart Disease	12	20.0%
4. Mitochondrial cytopathy*	2	3.3%
5. Hemorrhagic diseases	8	13.3%
6. Sinus thrombosis	4	6.7%
7. Diabetes Mellitus	2	3.3%
8. Vascular anomaly	5	8.3%

*One case proved by biopsy and other proved by high lactate level with stroke, myopathy and diabetes Mellitus

Discussion:-

Estimates of the incidence of stroke in the general population of children vary widely, ranging from 2.5 to 13 cases per 100,000 children per year (7). It frequently has the sequelae of cognitive and motor impairment, as well as epilepsy (8), (9). In adults the frequency of cerebral stroke is more in males than females and this has been attributed to neuroprotective effects of estrogen in females (10). Our study revealed that the frequency of stroke in children is also more common in males than females (53.3% versus 46.7%). The same trend has been clarified in other worldwide childhood stroke studies (10), in spite of absence of estrogen predominance before puberty (11). Severity of stroke in most of our children (91.7%) measured by pediatric NIHSS was mild and mild to moderate with lateralization as the most common presenting complaint in (73.3%) of patients. In a study by Del Balzo about stroke in children: inherited and acquired factors and age-related variations in the presentation of 48 pediatric patients, lateralization was the most common presenting stroke manifestation (12). Signs of lateralization were early picked only by neurologist and this might explain the hospital delay of stroke in children as showed by Zacharias et al., (13). On the other hand disturbed conscious level proved in 24 (40%) patients and convulsions in 21 (35%) patients. These were agreed with Brian Chung et al., (14). Coma and convulsions were the higher warning signs for stroke in children diagnosis and helps to early hospitalization and rapid management (15). Hospital delay duration in our study was mostly (83.3%) after 12 hours. However in multiple researches most of patients about (35%) arrived early before 6 hours of the disease onset (16), (17). The reasons for delay of our studied cases were diverse but typically included underestimation of the disease severity by the parents or healthcare providers of first or second referral, especially less than 40% of our patients presented with the higher warning signs; coma and/or convulsions. These reasons were matched with results from similar societies in India and Taiwan (18), (19), which raise the attention to increase the public awareness of early stroke symptoms and signs. Heart disease and stroke statistics-2016 update

revealed that there is no major difference between ischemic strokes percentage in adults compared to children (about 85% of all strokes) (20). In our study, most of patients had ischemic insults (83.3%). MRI brain imaging is superior than CT brain imaging in diagnosis of hemorrhagic stroke secondary to vascular malformations (21). The use of MRI brain in our hemorrhagic stroke patients showed that 2 of 6 patients (33.3%) revealed underlying pathology while only 2 of 44 ischemic stroke patients (4.5%) showed hemorrhagic transformation in one case and vascular anomaly in the other. This highlights the importance of MRI brain in patients with hemorrhagic stroke. In adults, ischemic stroke is often associated with the atherosclerosis while major etiology in hemorrhagic stroke is hypertension (22), (23), but in childhood, ischemic stroke has various causes, such as blood diseases, sickle cell anemia and heart diseases while hemorrhagic stroke main etiology is coagulopathy (3), (24). Seven different causes of stroke in our series had been found. Cardiac diseases and hemorrhagic blood diseases were common in the studied patients. Congenital heart disease is one of the leading causes of stroke in children, accounting for 15-30% (25). Cardiac evaluation of our cases was not different from them, revealed 12 (20.0%) patients with cardioembolic stroke. Our study also showed 8 (13.3%) cases with hemorrhagic blood diseases. Another recognizable cause of stroke in children worldwide is sickle cell disease (SCD) (26), (27). Our study revealed no proved SCD cases using hemoglobin electrophoresis in anemic patients. This could be explained by genetic and racial differences as it is not frequent in Egypt along the Nile Valley (28), and HbS gene is almost nonexistent except in the Oases where the carrier rate varies from 9 to 22% (29). In pediatric stroke the cause is often unidentified. The clinical presentation is often subtle and early radiological investigation may reveal normal findings (30). While most of our patients were secondary to various causes, 38% were cryptogenic. In a systemic review in stroke in children: Lindsey Barst Gumer et al., the cryptogenic etiology were only 14% with no recognizable risk factors (23). The mismatching of our study could be explained by short follow up time in our research, because some etiologies needs longer period to diagnose like mitochondrial cytopathies and other metabolic inherited disorders (31). Also fund limitation restricted investigations needed to detect other rare causes incriminated in the pathogenesis of stroke in children. Assessment of the stroke survivors after three months in our case study showed significant improvement in severity and disability. This could be explained by the high neuroplasticity and regeneration in children brain cells (32), (33), (34). In conclusion, diagnosis of stroke in children proves challenging, because symptoms are often unspecific in nature which reflected on hospital delay time, etiology is diverse and different from those present in adulthood. MRI brain imaging proved more efficacious in patients with hemorrhagic stroke rather than ischemic ones. Our limitations were the unawareness of the disease and short follow up time. Our study was unfunded to clarify the various and rare genetic causes seen in stroke in children and highlight the need of specialized stroke management teams at pediatric hospitals, with rapid transport, specific neuroimaging protocols, early therapeutic strategies that have improved outcomes.

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RESEARCH ARTICLE

MUSCULOSKELETAL PAIN AMONG SAUDI MEDICAL STUDENTS IN MAKKAH.

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Abstract

Objective: To determine the prevalence and associated risk factors for musculoskeletal pain among medical students in Saudi Arabia.

Method: A cross-sectional analysis for one Saudi medical school in 2015-2016 academic year. The main outcomes were measured by The Nordic Musculoskeletal Questionnaire (NMQ) which was used to screen for pain in different anatomic joints during the past week and past year. Another questionnaire was developed to assess for contributing factors for the pain.

Result: 849 medical students were analyzed with a response rate (73.31 %). There were 57.85% of the students suffering of musculoskeletal pain in the past week, and 43.47% in past year. There is a higher prevalence among female medical students with p value <0.01. Previous trauma, family history of trauma, high computer hours use, being in clinical years, and high BMI were the main factors contributing to MSP in this study.

Conclusion: The study demonstrates a very high prevalence of musculoskeletal pain medical students which may affect the lifestyle and academic performance in medical schools.

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Background:-

Musculoskeletal pain (MSP) which includes commonly low back pain, shoulders pain and neck pain is a common health problem that affect both male and female. World-wide estimates of lifetime prevalence of low back pain (LBP) vary from 50 to 84 % and 20-30% at any given time [1]. Musculoskeletal pain affects people's mood leading to depression, anxiety, irritability, poor social interactions, and lower overall health status, and that can limit the individual productivity, induces stress and it is socially costing in terms of health care. Although the Musculoskeletal pain represented as a common occupational problem it may affects younger population like school and university students who have not yet entered their working life. Moreover, previous studies have demonstrated a higher prevalence of MSP among medical and dental students compared to other university students. Several studies investigated the prevalence and factors associated with MSP among medical students. However, there are no previous studies have been conducted among medical students in Saudi Arabia. The authors aim in this study to measure the prevalence of this health problem among medical students in Umm Al-Qura University, College of Medicine (UQU Med) in Saudi Arabia, compare it with other investigated risk factors in literatures, and look for other possible factors that may contribute to MSP.

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Method:-**Data Collection:-**

An observational quantitative cross-sectional study was conducted among medical students at UQUMed. A self-administered questionnaire was personally distributed to 520 medical students in each year. Questionnaires were distributed and collected over the second semester of the academic year 2016. The questionnaire included participants' demographic information (gender, academic year, height, and weight), social life characteristics (including time spent studying, using computer, watching television, exercise, coffee drinking, and smoking), co-existing medical illnesses, history of trauma, family history of MSP. The participants also were invited to Nordic questionnaire of musculoskeletal pain considering lower back, shoulders, hand, neck, knee, upper back elbow, hip and feet. In total, 849 out of 1160 medical students participated in the study (response rate: 73.31 %).

Data analysis:-

Data were obtained from the questionnaire forms and then entered into an SPSS spreadsheet (SPSS 21). Percentages, mean and standard deviation (SD) were used as descriptive statistics. Chi-square test was used to observe the association between the categorical and outcome variables. A p value of <0.05 was considered as statistically significant.

Result:-

Out of 1160 medical students at UQU, 849 medical students responded and returned the filled-up questionnaire forms with a response rate of 73.31%. Of these participants, 489 (57.6%) were female. The mean height and weight for this study were 163.63 cm (SD 11.63) and 64.74 kg (SD 16.68) respectively. History of previous trauma was positive in 28.64% of the participants and 35.93% had a positive family history of MSK pain. The majority spend between two to three hours in each of studying, using computer, and watching television every day. Table 1 shows the distribution of demographic and lifestyle variables of participants.

More than half of the respondents (n=596, 58%) were students in clinical years. The self-reported Nordic musculoskeletal pain level demonstrated prevalent of pain of low back among 291 (50.1%) medical students, followed by upper back pain in 249 (43%), and neck pain in 242 (40.8%) medical students Table 2. In the past year, the prevalence of MSK pain was significantly higher among the clinical year students compared to preclinical years (P .001). However, there is no statistically significant difference in MSK pain in past week among clinical and preclinical year students.

The gender of medical students is statistically significantly associated with history of MSK pain in past week and past year. Female medical students have higher MSK pain history than male medical students (P<.01). Moreover, those with history of prior trauma to neck, shoulder, or back has higher reported MSK pain than students with no prior history of trauma (P<.01). The medical students, who were studying in low tables, were suffering with MSK pain in the past week and past year, which is statistically significant Table3.

Discussion:-

The study displayed a high prevalence of MSK pain in different anatomic regions among medical students at Umm Al-Qura University. Similar to previously published researches, women population has a higher prevalence of MSK pain compared to men [1-3]. The most prevalent painful sites were lower back, upper back, and neck respectively. Similar to previous studies, lower back pain (LBP) was the most prevalent site where almost half of the students complained of lower back pain in the past week.

A cross-sectional study in an Indian medical college revealed a LBP prevalence of 48% in medical students which is very close to the 43% rate reported for medical students of the University of Colorado in the U.S. and a 53% rate reported for medical students at Paracelsus Medical University in Austria. Similar to previous studies, lower back pain (LBP) was the most prevalent site where almost half of the students complained of lower back pain in the past week [5].

Several studies show that there are other different factors which can affect the musculoskeletal pain among medical and dental students like age, gender, smoking, alcohol consumption, average weight and height and body mass index (BMI) [6]. A retrospective analysis of MSP among Chinese medical students shows more female than male in the MSP reporting group (55% vs. 45%). Similarly, this study show a higher significant prevalence of MSP among

female medical students compared with male medical students with p value >0.01 . There were 33.7% male medical students suffering from MSP in the past week compared to 66.3% female medical students in the past week. Also, MSP among the female group in the past year was higher with a p value >0.01 .

Previous history of trauma was significantly associated with a higher reports of MSP in the past week and year with P value 0.001, and >0.001 respectively. The MSP is also reported significantly with those with family history of MSP. This proves the multifactorial cofactors that may contribute to MSP which includes both acquired and inherited factors [1].

High Body Mass Index (BMI) was found to be a major factor of MSP. The risk of MSP is about two folds higher when BMI >24 [7]. Also, higher BMI is associated with a higher incidence of osteoarthritis which may affect and limit mobility in the future. In this study, higher BMI was also associated with a higher prevalence of MSP among medical students.

Limitation:-

This study is limited by the fact that it represents only one medical school in Saudi Arabia among 28 medical schools. Also, the ergonomic factors in medical school were not measured to look for possible other factors contributing to MSP. For these reasons, the results cannot be generalized to all medical schools in Saudi Arabia.

Table 1:- The distribution of demographic and lifestyle variables of participants

	Male n=360 (42.4%)	Female n=489 (57.6%)
Academic Year		
• Second	62 (17.2%)	111 (22.7%)
• Third	64 (17.8%)	16 (3.3%)
• Forth	91 (25.3%)	183 (37.4%)
• Fifth	54 (15.0%)	66 (13.5%)
• Sixth	89 (24.7%)	113 (23.1%)
History of trauma in the neck, shoulder, back, lower or upper limb		
• Yes	84 (23.3%)	105 (21.5%)
• No	276 (76.7%)	384 (78.5%)
Family history of MSP		
• Yes	116 (32.2%)	188 (38.4%)
• No	301 (61.6%)	545 (64.2%)
Exercise		
• Regularly	85 (35.6%)	87 (37.5%)
• Not regular	141 (59%)	141 (60.8%)
• No at all	13 (5.4%)	4 (1.7%)
Smoking		
• Yes	87 (24.2%)	69 (14.1%)
• No	273 (75.8%)	420 (85.9%)
Height	171.81 cm (9.02)	157.60 cm (9.44)
Weight	75.29 Kg (17.36)	56.97 Kg (10.92)
Hours of computer use/day	3.5hr (SD 2.4)	3.6hr (2.4hr)
Hours of study /day	3.05hr (2.03)	3.4hr (2.1hr)
Hours of TV watching/day	2.01hr (1.42)	2.3hr(1.98hr)

Conclusion:-

The study demonstrates a very high prevalence of musculoskeletal pain medical students which may affect the lifestyle and academic performance in medical schools. More interventions are needed to alleviate MSK pain among medical students and further improve the study environment. Exercises, appropriate study environment, and weight reduction can optimize the academic performance and wellbeing of medical students.

Table 2:- Prevalence of MSP during past week and past 12 months

Body site		Prevalence during past week		Prevalence during past Year	
		(n)	%	(n)	%
Neck pain	No	351	59.2	279	65.0
	Yes	242	40.8	150	35.0
Shoulders pain	No	424	73.7	315	80.8
	Yes	138	24.0	75	19.2
Elbows pain	No	525	90.5	328	90.4
	Yes	53	9.1	35	9.6
Wrists/hands pain	No	498	85.9	320	86.3
	Yes	75	12.9	50	13.5
Upper back pain	No	330	57.0	227	54.6
	Yes	249	43.0	189	45.4
low back pain	No	290	49.9	233	54.4
	Yes	291	50.1	194	45.3
One or both hips/ thighs pain	No	495	87.5	314	84.9
	Yes	71	12.5	56	15.1
one or both kness pain	No	418	71.9	267	69.4
	Yes	163	28.1	118	30.6
one or both ankles/feet pain	No	459	79.5	292	77.7
	Yes	118	20.5	84	22.3

Table3:- Prevalence of MSP in past week and year

		MSP in past week			MSP in past year		
		Yes	No	P	Yes	No	P
Gender	Male	118 (33.7%)	134 (52.5%)	P<.01	68 (27%)	184 (73%)	P<.01
	Female	232 (66.3%)	121 (47.5%)		195 (55.2%)	158 (44.8%)	
Academic year	Preclinical	61 (52.1%)	56 (47.9%)	.176	34 (29.1%)	83 (70.9%)	.001
	Clinical	289 (59.2%)	199(40.8%)		229 (46.9%)	259 (53.1%)	
History of trauma	Yes	107 (69.5%)	47 (30.5%)	.001	88 (57.1%)	66 (42.9%)	P<.01
	No	243 (53.9%)	208 (46.1%)		175 (38.8%)	276 (57.1%)	
Family history of MSP	Yes	148 (64.6%)	81 (35.4%)	.009	113 (49.3%)	116 (50.7%)	.023
	No	202 (53.7%)	174 (46.3%)		150 (39.9%)	226 (60.1%)	
Exercise	Regularly	74 (43.5%)	62 (40.3%)	.325	53 (39%)	83 (61%)	.033
	Not regularly	92 (54.1%)	84 (5.2%)		52 (29.5%)	124 (70.5%)	
	Not at all	4 (2.4%)	8 (5.2%)		2 (16.7%)	10 (83.3%)	
Level of study table	High	83 (54.4%)	102 (45.6%)	P <.01	57 (30.8%)	128 (69.2%)	P <.01
	Low	103 (66.5%)	52 (33.5%)		86 (55.5%)	69 (44.5%)	
		Mean (SD)			Mean (SD)		
Height		1.63m (SD 9.6)	1.66 m (SD 11.21)	P <.01	1.61m (SD 9)	1.64m (SD 12.54)	P<.01
Weight		63.66kg (SD 14.7)	69 kg (SD 19.27)	.002	62 kg (SD 13.6)	65.56 kg (17.8)	.005
BMI		23.76 (SD 4.1)	24.44 (SD 5.1)	P <.01	23.8 (3.6)	24.40 (SD 9.5)	P <.01

Hours of computer use/day		3.6 hr (SD 2.4)	3.11 hr (SD 1.9)	P < .01	3.3 hr (SD2.07)	3.75 hr (2.54)	.667
Hours of study/day		3 hr (SD 1.8)	3.4 hr (2.77)	.549	2.70 (SD1.44)	3.52 hr(SD 2.29)	. P<.01
Hours of watching TV/day		2.12 hr(1.2)	1.8 hr (SD 1.6)	P<.01	2.37 (SD 1.29)	2.11 hr (SD 2.03)	P<.01

Conflicts of interest:

The authors have no competing interests to declare.

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RESEARCH ARTICLE

INCIDENCE OF JAPANESE ENCEPHALITIS VIRUS INFECTION IN WEST BENGAL, INDIA – A TWO YEARS COMPREHENSIVE STUDY.

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Abstract

Japanese Encephalitis Virus (JEV), is a mosquito-borne pathogen, which causes Japanese Encephalitis (JE); a neurotropic disease with high mortality and can be accounted to be the main player in the increasing global trend of viral encephalitis with prime concern on general public health. JE was initially documented in the East Indian state of West Bengal in the year 1973. From that point forward it is being reported every year consistently from various regions of the state, although the vaccination programme has already being procured. In this way, it shows that there may be either an incomplete coverage of the vaccination approach or the gradual rise of transformed new strains of JEV. Considering this reality, to comprehend the frequency of JEV dissemination and its endemicity in the region, we apprehended a pilot scale epidemiological study on an aggregate of 159 positive patient samples gathered from the 533 clinically presumed patients with Acute Encephalitis Syndrome (AES), admitted in the different district health centres and hospitals of West Bengal, India, 2015-2016.

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Introduction:-

Japanese Encephalitis (JE) is a well-known mosquito borne viral disease. Currently it is one of the most dreaded and intriguing viral infections concerning the Indian health scenario. It is one of the leading varieties of viral encephalitis globally, mostly prevalent in south-eastern Asia, covering an area with a few billion population (1). Most JE infections are asymptomatic in nature, but if clinical symptom builds up rapidly it naturally effects with significant mortality and morbidity. Though not officially well documented but it can be estimated that, JEV causes approximately 50,000- 55,000 cases and 12,000-15, 000 deaths annually. Japanese Encephalitis is a disease with severe epidemic potential and high fatality rate and hence monitoring its prevalence status is considered to very important especially in countries like India. JEV mainly affects young adults and children areas with high endemicity. Japanese Encephalitis Virus (JEV) is a member of the Flaviviridae family and Flavivirus genus (2). The JEV virion is made up of three structural proteins - nucleocapsid, glycosylated envelope protein and non-glycosylated membrane protein as well as seven non-structural (NS) proteins. JEV exists as a zoonotic pathogen with mosquitoes and pigs as hosts (3). This deadly virus is accounted for severe neurotrophic disease with fever, acute flaccid paralysis, aseptic meningitis and meningo-myeloencephalitis in humans (4, 5). This virus has spread worldwide at an alarming rate and currently it has established itself as a major health concern in India.

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Approximately 30% of the infected patients die and about 50% of the surviving individuals suffer from severe neuropsychiatric sequelae (6, 7). In West Bengal, the first major outbreak of JEV infection took place in 1973 in the districts of Burdwan and Bankura where more than 700 cases and 300 deaths have been reported (8). Since then many outbreaks have been reported; every year sporadic cases are continuously being reported from different districts of West Bengal (9,10). This article presents a comprehensive analytical profile of JE cases from 2015-2016 and intends to specifically establish the current status of JEV endemicity in West Bengal.

Materials and methods:-

Sample collection:-

Different district hospitals, medical colleges and ID & BG Hospital in Kolkata, West Bengal, were selected for this study from 2015–2016. Patients admitted with AES, having high grade fever ($\geq 39^{\circ}\text{C}$) for 2–15 days with any two of the following symptoms: headache, vomiting, unconsciousness, convulsions, abnormal movements, stupor, delirium, altered sensorium, neck rigidity, presence of Kernig's sign, aphasia, drowsiness and rigor, were considered for this study. Details of the clinical event, investigations, treatment given and the prognosis of the patients during hospitalization were provided by the concerned clinicians. A short case history along with the results of CSF study of each case was recorded. Most of the patients had moderately high sugar levels (45–65 mg/dL), slightly higher protein levels, varying from 50–70 mg/dL, and a white blood cell (WBC) count of $\geq 6 \times 10^6/\text{L}$. Cerebral malaria and bacteriological etiology were ruled out by the hospitals concerned. Informed consent was obtained before the collection of samples. A total of 533 blood samples were collected from the clinically suspected AES cases in sterile gel-line vacutainers (Becton Dickinson, Franklin Lakes, NJ, USA) and also in sterile test tube (Gujarat Borosil Ltd., Bharuch, Gujarat, India) by vein and transported on dry ice to the Indian Council of Medical Research (ICMR) Virus Unit, Kolkata, where the serum was separated and both the serum and CSF were stored at -80°C till tested.

Cell and virus strain culture:-

JEV P20778 strain (GenBank Accession No. **AF080251**) was obtained from the National Institute of Virology (NIV), Pune, and was used as a positive control throughout the study. The virus was reconstituted and inoculated on *Aedes albopictus* C6/36 mosquito cell lines obtained from National Centre for Cell Science (NCCS), Pune in minimal essential medium (MEM; GIBCO BRL-Invitrogen, Grand Island, NY, USA) supplemented with 10% fetal bovine serum (FBS; GIBCO BRL-Invitrogen) and penicillin streptomycin antibiotics (PenStrep; GIBCO BRL-Invitrogen) in 75 cm² tissue culture flask (Nunc, Roskilde, Denmark). The flask was incubated at 28°C in an incubator under 5% carbon dioxide concentration. It was observed regularly for the appearance of cytopathic effect (CPE) up to 7–8 days. On appearance of the CPE, the tissue culture fluid was centrifuged at 1000 g for 5 minutes and the supernatant was used as positive control for RT-PCR or stored at -80°C .

Isolation of virus:-

Attempts were made to isolate the virus using C6/36 cell lines. For this purpose, only IgM negative to JEV samples with a history of ≤ 3 days fever along with any two of the symptoms stated earlier, were selected. 200 μl of selected serum samples were spread over the monolayer of C6/36 cell line and allowed to adsorb for 120 minutes in an incubator at 28°C under 5% CO₂ concentration. After adsorption, the excess sample materials were discarded and 1 ml MEM supplemented with 2% FBS and PenStrep were added in 24 well tissue culture plate (Tarsons Products Pvt. Ltd., Kolkata, West Bengal, India) and were incubated again at 28°C under 5% CO₂ concentration. It was observed regularly for the appearance of cytopathic effect (CPE) up to 7–8 days. For those samples which did not produce CPE, the tissue culture fluids were again passaged up to five times to facilitate the isolation of the virus, if any. After the appearance of CPE, the tissue culture fluid was collected by centrifugation at 1000 g for 5 minutes and the supernatants were kept in aliquots at -80°C till isolation of RNA, followed by RT-PCR test. Non-infected C6/36 cell culture was used as a negative control.

Serology:-

For the detection of JE IgM antibody in the collected samples, ELISA tests were performed with the kit, obtained from the NIV following the prescribed protocol.

RNA extraction:-

Those serum samples that produced prominent CPE and the JEV P20778 strain (used as the positive control throughout the study), were subjected for RNA isolation, using 140 μl of tissue culture fluid. The QIAamp® RNA viral kit (Qiagen, GmbH, Hilden, Germany) was used, following the manufacturer's protocol.

Reverse transcriptase PCR for envelope gene of Japanese encephalitis virus:-

For the detection of JEV by RT-PCR method, both serum and CSF samples with a history of ≤ 3 days fever were selected. Qiagen one step RT-PCR kit (Qiagen) was used according to the manufacturer's protocol, using RNA (50 pg to 1 g) and 0.6 M of primer pairs; forward primer: JEnvF (w) 942-ACCATCCTCCTGCTGTTGGTCGCT-965 and reverse primer: JEnvR (w) 2506-CTTGTGATGTCAATGGCACATCCAGTGTCA-2477 which anneal to the conserved region of structural envelop protein (E) specific for Japanese encephalitis virus.

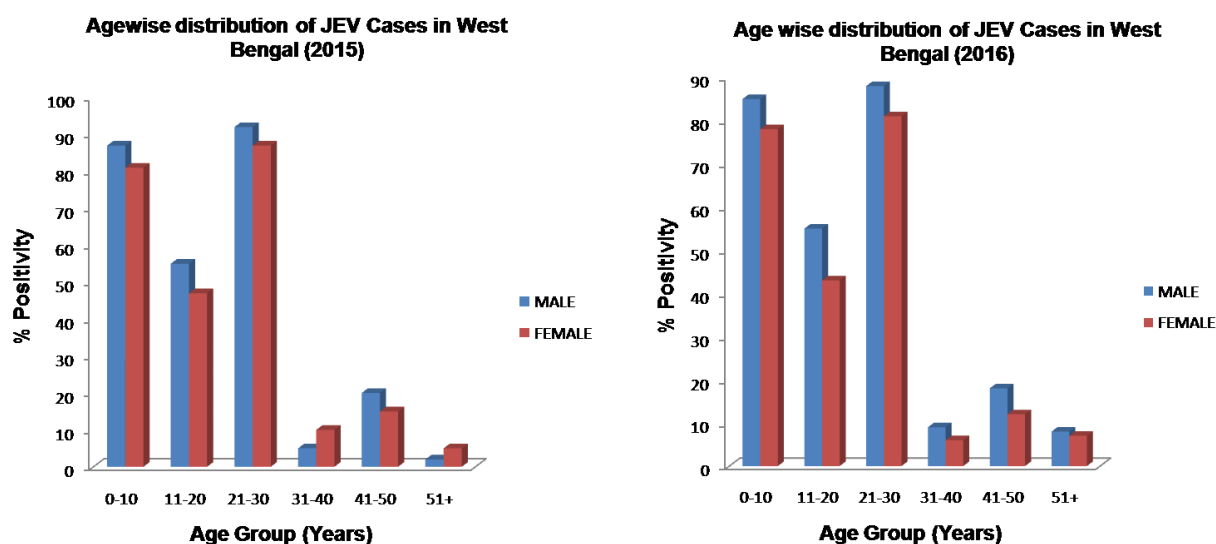
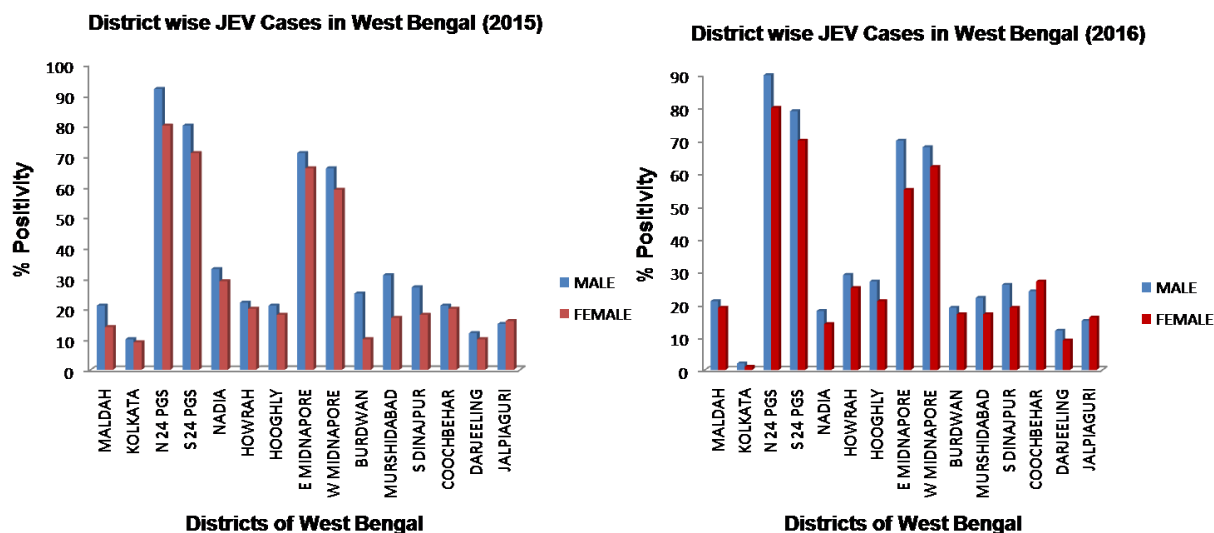
Results:-**Figure 1****Figure 2**

Figure 3



Discussion:-

As most JEV infections are subclinical in nature, a major portion of them remains undiagnosed (11,12). This study was particularly aimed to accurately identify the variable JE cases among the patients with Acute Encephalitis Syndrome (AES) in the East Indian state of West Bengal and to understand the current status of JEV in the state (13,14). JEV infection can be detected either directly or indirectly with the aid of several techniques. The conventional IgM specific capture ELISA is the approach for analysis of many infections including JEV as the primary screening test (15). The ELISA negative samples which had been amassed in the early clinical phase were additionally subjected to the reverse transcriptase (RT)-PCR for the identification of the virus (16). To fulfil this purpose, the collected clinical samples were preserved without disrupting the cold chain. In the present study, serum and CSF were chosen as the clinical specimens for serological and molecular diagnosis of JEV infection in the suspected cases. In India, JEV is a severe public health issue and mainly affects the young and adults up to the age of 30 years. In the present study, out of 533 samples tested in the two years, only 159 (29.8%) were positive to JEV. Percentage positivity of males during this time span was 31.68 % and females were 26.45%. All JEV positive patients had a history of illness for ≥ 10 days, indicative of active immune response at this stage of illness. This observation is proof of infection in the immediate past. Although, JE cases have been observed from all age groups, the highest numbers of positive cases have been recorded in the age group 21-30, followed by 0-10, in both male and female individuals almost identically (Figure 1) during 2015-2016, which almost corroborates with earlier studies. Only a few cases were detected in the age groups above 21-30 (elderly/higher age groups). The highest number of cases was observed in the 21-30 age groups of both male individuals and female individuals which are most likely because people from this age group actively take part in the cultivation of crop fields in rural areas. The vector usually breeds in the stagnant water in the cultivation fields and hence the majority from this age group are directly exposed to the vector. The next higher numbers were observed for the age group 0-10 mainly due to their underdeveloped immunity. The low number of JE cases in the higher age group is possibly due to the development of effective immunity, either by sub clinical infections or due to earlier vaccination. During both the years the maximum number of cases was reported during the months of July to September which corroborates with the warm, rainy season in West Bengal, an ideal environment for the growth of mosquitoes and disease spread (Figure 2). The major cases were observed from the districts of North 24 parganas, South 24 parganas, East Medinipur and West Medinipur (Figure 3). The reason may be due to the fact that these are sea connected and bordered districts with major chances of immigration carrying infected individuals through ports, as well as huge abundance of migratory birds which carry the virus with them. As males and females have been equally affected in almost all the cases it suggests that JEV does not have any sex preference.

Conflict of interest:-

There exists no conflict of interest among the authors.

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RESEARCH ARTICLE

DEGRADATION OF TEXTILE INDUSTRY'S EFFLUENT USING INTEGRATED CHEMICAL-BIOLOGICAL PROCESS.

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textile, effluent, coagulation, flocculation, activated sludge.

Abstract

Interest in ecologically friendly, wet-processing textile techniques has increased in recent years because of increased awareness of environmental issues throughout the world. With more stringent controls and more demand in environmental friendly product expected in the future, it is essential that control measures be implemented to minimize effluent problems. The textile dyeing industry consumes large quantities of water and produces large volumes of wastewater from different steps in the dyeing and finishing processes. Wastewater from printing and dyeing units is often rich in color, containing residues of reactive dyes and chemicals, and requires proper treatment before being released into the environment. The goal of this research was to study the performance of the integrated chemical (coagulation-flocculation)-biological (activated sludge) process and to develop a potential textile industry's wastewater treatment system for reuse. The results show that coagulation-flocculation process (optimum coagulant of 10 mg/l FeSO₄) was effective enough in removing color while activated sludge process effective in removing other organic materials contain in the effluent. Integrated chemical-biological process to treat textile effluent was able to remove organic with an efficiency of 71 - 91 %. Re-use of this kind of wastewater than to discharge it after treatment are preferably due to increase in the costs of chemicals, energy and water consumption.

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Background:-

The textile and clothing industry in Indonesia played a significant role in the national economy. In 2006, this industry contributes 11.7% to the total national exports, 20.2% of the national trade surplus, and 3.8% to the formation of Gross Domestic Product (GDP). This industry employs about 1.84 million workers and the lives of many Indonesian families are connected with the textile industry (agriculture, trade and services). Until year 2006, the number of Indonesian textile industry reached 2,699 companies, with a total investment of 135.7 trillion rupiah. This number is only slightly increases over the previous year, amounting to 2,656 companies. Textile industry is concentrated in West Java (57%), Central Java (14%), and Jakarta (17%). The rest are scattered in East Java, Bali, Sumatra and Yogyakarta [1]. The textile industry consumes large quantities of water and produces large volumes of

wastewater from different steps in the dyeing and finishing processes. These problems may occur during the production processes which usually require the input of a wide range of chemicals and dyestuffs, which generally are organic compounds of complex structure. Because all of them are not contained in the final product, they became waste and require proper treatment before being released into the environment. Major pollutants in textile wastewaters are high suspended solids, chemical oxygen demand, heat, color residues of reactive dyes and chemicals, acidity, and other soluble substances [2]. Effective and economic treatment of a diversity of textile effluents containing color has become a problem. No single treatment system is adequate for degrading the various dye structures. Currently, much research has been focused on chemically and physically degrading color dyes in wastewaters. These methods include chemical oxidation, which uses strong oxidizers such as hydrogen peroxide or chlorine dioxide. Chemical oxidation is sometimes coupled with UV light exposure to increase the color removal. Other techniques involve electrochemical or wet oxidation, activated carbon adsorption, reverse osmosis, or coagulation/flocculation. Many of these technologies are cost prohibitive, however, and therefore are not viable options for treating large waste streams [3]. To ensure the safety of effluents, proper technologies need to be used by treatment facilities when degrading fiber-reactive dyes. Many research efforts have focused on various biological, chemical, and physical techniques for treating dye wastes. There is evidence that all three areas have potential for remediating textile industry wastes. However, chemical treatment is often cost and application limited, while physical removal can lead to extra solid wastes and increased overhead. Biological treatment has been effective in reducing textile industry effluents, and when used properly has a lower operating cost than other remediation processes. Combinations of chemical and biological or physical and biological treatment have also proven to be effective. The primary objective of this study was to determine the best way to increase the color-reducing efficiency of the textile industry's effluent and to develop a potential textile industry's wastewater treatment system for reuse.

Materials and Methods:-

Wastewater used in this research was obtained from a synthetic textile dyeing factory located in the Nanjung industrial complex in Bandung, West Java. The characteristics of textile wastewater are showed in **Table 1**. Its average BOD, COD, pH, Ammonia, Oil&Grease and conductivity were 160 mg/l, 314 mg/l, 7.81, 4.46 mg/l, 16 mg/l and 5060 umho/cm, respectively. The combined process for textile wastewater treatment consists of chemical coagulation and flocculation pre-treatment followed by biological activated sludge treatment. Biological treatment was conducted in a 15 l reactor (10 l working volume) as described in **Figure 1** and 2000 mg/l of mixed liquor suspended solids (MLSS) were inoculated into the reactor. This reactor was equipped with a diffused aeration device at the bottom of the reactor. The wastewater was continuously fed to the reactor using peristaltic pumps with a desired flow rate. Chemical coagulation and flocculation were conducted in batch mode act as pre-treatment before biological treatment step. Jar-test at the laboratory scale was carried out in order to choose the adequate coagulant and the reaction condition (pH, dosage). $\text{Al}_2(\text{SO}_4)_3$, poly-aluminum chloride (PAC), FeSO_4 were tested as chemical coagulant candidates. One normal H_2SO_4 , NaOH solution was added to adjust pH of the solution to the desired value. Chemical coagulant was added and mixed for 2 min under rapid mixing condition (200 rpm) and the solution was mixed at slow flocculation (40 rpm) for 15 min after rapid mixing. And then, COD, SS, and pH of supernatant were measured after settling for 30 min. Standard methods [4] were used for the estimation of MLSS, COD, H_2S , Ammonia.

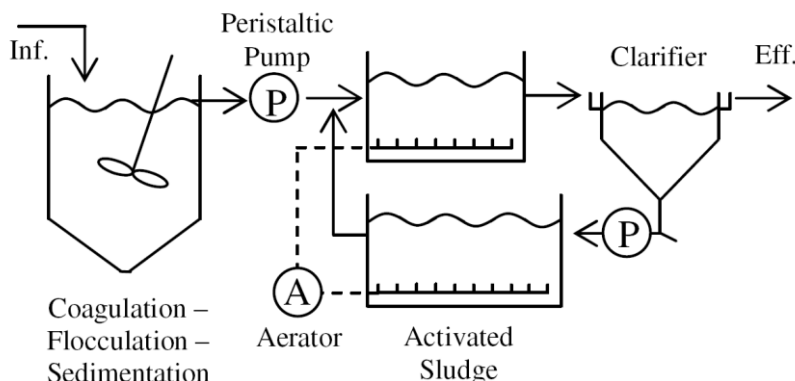


Figure 1 :- Reactor configuration schematic.

Results and Discussion:-

The raw textile wastewater samples were obtained from a large synthetic textile dyeing factory located in the Nanjung industrial complex in Bandung, West Java. The textile products processed in this plant were made primarily of cotton, linen and polyester fibers and small quantity of silk. According to the operational personnel, no less than 10 different kinds of dyes were used in the dyeing and finishing mill during the half-year period of this experimental study. Therefore tracking of the kind of dyes employed every day was virtually impossible because no record was kept at the mill. In general, the wastewaters had a BOD within the range between 125 and 172 mg/l, while COD within the range 281 and 315 mg/l. BOD:COD ratio of these wastewater within the range between 0.4-0.5 are quite biodegradable to biological process.

Table 1:- Characteristics of textile wastewater.

Parameters	Unit	Sample 1	Sample 2	Sample 3	Standard*
BOD	mg/l	160	125	172	60
COD	mg/l	314	281.6	315	150
TSS	mg/l	8	102	55	50
Phenol	mg/l	0.16	0.214	n.d	0.5
Cr Total	mg/l	0.008	0.088	n.d	1
Ammonia	mg/l	4.46	11.6	0.99	8
H ₂ S	mg/l	28	0.325	n.d	0.3
Oil & Grease	mg/l	16	12.4	4	3
Conductivity	us/cm	5060	4280	5300	-
pH	-	7.81	7.38	7.29	6-9

* Governor of West Java Decree No. 6, Year 1999; n.d.: no data

Conclusion:-

Studies on the effects of the optimum coagulant, dosage and pH are conducted in order to investigate the sorption capacity of coagulant in flocculation process. Since the Chemical Oxygen Demand (COD) level in wastewater from textile industry is considered as the most important parameter, so it has been used as the indicator on the sorption capacity of coagulant in these experiments by supporting with other parameter which is turbidity as total suspended solid/TSS. The jar test results showed that among others coagulant, FeCl₃ was the best coagulant for reducing organic in wastewater tested (performance data for Al₂(SO₄)₃ and PAC not shown in this report). FeCl₃ performance were presented in **Figure 2(a)** which showed the effects of coagulant (FeCl₃) dosage on COD level and the percentage of COD levels reduction. While **Figure 2(b)** showed the effects of coagulant dosage on turbidity levels and the percentage of turbidity levels reduction. From the jar test experiment, the curves for the both graphs were in the U-shape form for the condition of COD level and turbidity level versus coagulant dosage.

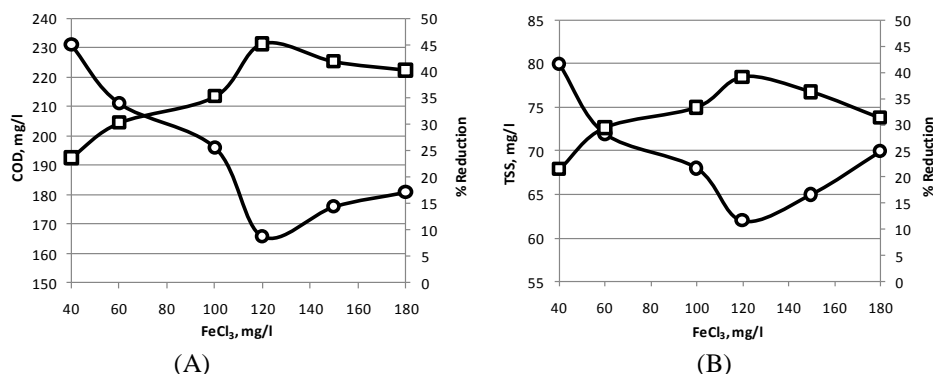


Figure 2 :- Effect of FeCl₃ dosage on (a) COD level (open circle) and the percentage (open square) of COD level reduction, (b) turbidity level as TSS (open circle) and the percentage (open square) of TSS level reduction

In wastewater treatment using metallic coagulants, pH plays a very important role in determining coagulation efficiency. Therefore, experiments were designed to determine the optimum pH for textile wastewater that allowed for maximum COD reduction and de-colorization. The effect of pH on the treatment efficiency was examined using optimum amount of coagulant at various pH conditions (4, 6, 8, 9, 10, 11, and 12). Lime and H₂SO₄ was used to adjust the desire pH. It is well known that pH affects the molecular structure of the dyes, which changes the absorbance of the solution. Optimum pH for COD reduction and de-colorization found within the range between 6 to 9. The optimum conditions for chemical coagulation flocculation process for pretreatment of textile wastewater are as follows: FeCl₃ coagulant with 120 mg/l dose with pH value 6-9. Pre-treated textile wastewater (with chemical coagulation process) then use as influent for biological (activated sludge) process for further treatment of wastewater (**Figure 1**). During operation, the DO concentration in the reactor was kept at 3 to 4 mg/l, MLSS was maintain at 2000-3000 mg/l and stable COD reduction were achieved after 2 d. In this case, about 72.9% of COD was removed. Other waste constituents like ammonia were reduced to 84%, phenol was reduced to 92% and oil&grease was reduced to 74% (**Table 2**). In addition, the result of COD removal capability in this study was compared with other similar combined processes for textile wastewater treatment as shown in **Table 3**. Two treatment operations comprised coagulation and activated sludge process, while the other comprised activated sludge followed with coagulation process.

Table 2:- Characteristics of textile wastewater after treatments

Parameters	Unit	Influent	Coagulation eff.	Activated sludge eff.	Removal efficiency
BOD	mg/l	152.3	70	36	76.3 %
COD	mg/l	303.5	165.9	82	72.9%
TSS	mg/l	55	32	23	58.2%
Phenol	mg/l	0.187	0.157	0.014	92%
Cr Total	mg/l	0.048	0.092	n.d	-
Ammonia	mg/l	5.68	10.2	0.909	84%
H ₂ S	mg/l	14.16	0.349	n.d	-
Oil & Grease	mg/l	10.8	8.8	2.81	74%
Conductivity	us/cm	4880	4310	n.d	-
pH	-	7.49	7.12	6.8	-

n.d.: no data

Table 3:- Comparison of the COD removal of textile wastewater treatment.

COD value after each individual process, mg/l				
Influent	Stage I	Stage II	Total reduction, %	Ref.
900	Chemical coagulation 600	Activated sludge 120	86.6	[5]
870	Activated sludge 272	Chemical coagulation 100	88.5	[6]
	Chemical coagulation	Activated sludge		[7]

694	391	236	65.9	
303	Chemical coagulation 165	Activated sludge 82	72.9	This study

The characteristic of wastewater discharged from textile industrial activities was strictly controlled by government. This was basically due to the wastewater from textile industry was contaminated with a complex set of oxygen demanding materials and poses a great problem to natural environment. As a result, the wastewater from textile industry was treated by using chemical via coagulation and flocculation processes followed with biological activated sludge for further treatment of textile wastewater. The goals of this study are multifold. It aims at enabling the textile industry to treat and reuse the wastewater from its operations using a simple and feasible technology (best available technology). It also aims at preventing the pollution that results from textile industries to the receiving water body and saving significant quantities of water for other domestic uses. In this point of view, the choice of studied technologies was kept to the simplest level to make it easy for the textile industry to accept and implement it.

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RESEARCH ARTICLE

IN VITRO ANTI INFLAMMATORY AND ANTI MICROBIAL PROPERTIES OF *ALLOPHYLUS SERRATUS* LEAF EXTRACTS.

Kero Jemal, B.V Sandeep and Sudhakar Pola.

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Anti microbial; Antibacterial; Anti-inflammatory; *Allophylus serratus*; leaf extract.

Abstract

Allophylus serratus is a local medicinal plant used traditionally for the treatment of diarrhea, inflammation, ulcer, elephantiasis and gastro intestinal disorders. The leaf extracts of this plant were assessed for *in vitro* anti-inflammatory and antibacterial activities. The *in vitro* anti-inflammatory activity assay of leaf extracts of *Allophylus serratus* were evaluated by Human Red Blood Cells membrane stabilization, inhibition of protein denaturation and protease inhibition methods. The anti microbial activity of leaf extracts were evaluated by agar well diffusion method against four bacterial and two fungi species. The membrane stabilization anti inflammatory test showed that, the methanolic leaf extracts exhibited highest membrane stabilizing activity of 77.64 ± 0.78 and 74.77 ± 0.66 , compared to that of standard Diclofenac sodium which exhibited 92.65 ± 0.56 % and 89.73 ± 1.18 at 1000 $\mu\text{g/ml}$ and 800 $\mu\text{g/ml}$ concentrations respectively. In inhibition of protein denaturation, 400 $\mu\text{g/ml}$ methanolic extract showed maximum protection 70.73% and standard drug provided 93.50 % protection. But at 1000 $\mu\text{g/ml}$ the percent inhibition of protein denaturation increased to 95.93%. More over protease inhibition test showed maximum inhibition (67.63%) at 100 $\mu\text{g/ml}$ concentrations by ethyl acetate extract. In anti inflammatory test, all extraction showed concentration dependent response. In anti microbial activity test the water, methanolic and ethyl acetate extracts were more effective against *Escherichia coli* and *Staphylococcus aureus*. Ethyl acetate extract also showed high activity against *Bacillus subtilis* ($20 \pm 0.00\text{mm}$). Chloroform and petroleum ether extracts showed high zone of inhibition (20 ± 0.17 and 22 ± 0.09 mm respectively) against *Pseudomonas aeruginosa* but no activity against the rest tested bacterial strains. All the extracts did not show any activity against tested fungal species. The results of this study therefore revealed that leaf extracts of *Allophylus serratus* possess anti inflammatory and anti bacterial activity which justify the use of this plant in the treatment of bacterial infections and inflammation.

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Introduction:-

Plants are the main sources of therapeutic agents for treatment of various ailments and diseases since ancient time. For example, in Africa, about 80% of the population depends on medicinal plants in managing various endemic

forms of infections (Agyare *et al.*, 2009). Over the past years, many medicinal plants have been proved to be a remarkable source of therapeutic agents and have therefore given attention in most research centers in the world (Iwu *et al.*, 1999; Agyare *et al.*, 2013). The advantage of using medicinal plants is that they are natural and have no side effects. According to World Health Organization, medicinal plants would be the best source of a variety of important drugs. Therefore, to better understand their efficacy, safety and properties, such plants should be properly investigated (Nascimento *et al.*, 2000). There are many reports on the use of medicinal plants as anti inflammatory and anti microbial agents. This is due to the presence of chemical compounds in plants that suppress inflammatory reactions and the reproduction and growth of micro organisms.

Inflammation and pain are our body's natural reaction to injury caused by infectious agents, burn, toxic or physical, chemical or traumatic damages (Battu *et al.*, 2011). The reaction comprises systemic and local responses (Mohamed *et al.*, 2011). One purpose of inflammation is to limit or eliminate the spread of injurious agent and protect the body site of an injury. There are different components to an inflammatory response such as a complex array of enzyme activation, mediator release, cell migration, tissue breakdown and repair. These responses are aimed at host defense and usually activated in most disease condition.

Modern drugs such as narcotics (e.g. opioids), non-narcotics (e.g. salicylates) and corticosteroids (e.g. hydrocortisone) which are in use presently for the management of inflammatory conditions are toxic and have well known side effects such as gastric ulcers and redistribution of body fat. In addition, these drugs are very expensive. On the other hand many medicinal plants had been used since long time as anti inflammatory without any side effects. At present, much attention has been given in the searching of medicinal plants with anti-inflammatory activity which is not only without side effects but also cheap. These medicinal plants used as anti inflammatory activity may lead to the discovery of new therapeutic agent that is not only used to suppress the inflammation but also used in diverse disease conditions where the inflammation response is amplifying the disease process.

Microorganisms such as bacteria and fungi cause various diseases in humans. To treat these microbial infections, various antimicrobial drugs have been discovered. Due to the indiscriminate use of these commercial antimicrobial drugs, resistance to these drugs by microorganisms has been developed and increased day by day (Davis, 1994; Ahamad, 1998; Karupiah and Mustaffa, 2013). The number of resistant microorganisms to these antimicrobial agents is increases exponentially. Therefore it is very important to understand the genetic mechanisms of resistance, to screen medicinal plants for the identification and development of newer and cost effective anti microbial agents to treat infections caused by resistant microorganisms. Medicinal plants have also been used as anti microbial agents. Many medicinal plants have been used to treat microbial infections because of their antimicrobial properties, which are due to compounds synthesized in the secondary metabolism of the plant. Therefore, such plants should be investigated to better understand their properties, safety and efficiency (Doughari *et al.* 2008).

Allophylus serratus (Roxb.) Kurz, (Synonym *Allophylus cobbe* Raeuschel; *Allophylus edulis* Radlk) (Dharmani and Palit, 2005), commonly known as Tippi in Hindi, belongs to the family Sapindaceae. It is a small tree or shrub found all over different parts of India. Traditionally this plant carries a strong ethno-pharmacological background and has been used as anti inflammatory, anti ulcer, to treat elephantiasis, oedma, and fracture of bones and gastrointestinal disorders such as diarrhea, anorexia and dyspepsia (Umashanker *et al.*, 2011; Gupta and Tandon, 2004). Dharmani *et al.*, (2005) and Kumar *et al.*, (2010), reported that the ethanolic extract of *Allophylus serratus* has potential anti ulcerogenic and anti osteoporotic activities respectively. The leaves are used to reduce fever, to relieve rashes, promote lactation, to treat colic to relieve stomach aches, as antiulcer and to reduce piles (Umashanker and Shruti, 2011; Devi *et al.*, 2013). The roots of this plant contain tannin and are considered astringent and used for treating nose bleeding, diarrhea and rheumatic pains (Umashanker and Shruti, 2011).

The presences of different phytochemicals such as steroids, glycosides, flavonoids, alkaloids and phenolics in this species have been reported (Sanmuga *et al.*, 2012). Phytochemical screening and Pharmacognostic studies of *Allophylus serratus* showed the presence of various chemical compounds in different parts of the plant. Leaves of the plant contain β -sitosterol. They also contain phenacetamide, a chemical known for its antiulcer activity (Rastogi and Mehrotra, 1995). The presence of Quercetin, Pinitol, Luteolin-7-O-B-D-glucopyranoside, rutin, apigenin-4-O-B-D-glucosid also reported by Kumar *et al.*, (2010). The aim of present study is to evaluate the *in vitro* anti-inflammatory activity and antimicrobial efficacy of leaves extracts of *Allophylus serratus*.

Materials and Methods:-

Plant material:-

Leaves of *Allophylus serratus* were collected between September and December 2015, from Andhra University, Visakhapatnam, India. The plant was identified and authenticated by Dr. Bodaih Padal, taxonomist, Department of botany, Andhra University, Visakhapatnam. The voucher specimens (21921) were deposited in the herbarium, College of Science and Technology, Department of Botany, Andhra University, India.

Preparation of plant leaves extracts:-

The leaves of *Allophylus serratus* were washed with distilled water and shade dried in dark for three weeks to a constant weight. The dried leaf samples were then ground into powder using a clean electric blender. 100 g of the powdered materials were extracted with water, methanol, ethyl acetate, chloroform and petroleum ether separately using soxhlet apparatus. The extracts were concentrated by evaporating the solvent in rotary evaporator. The yield was found to be 11.30 %, 15.40%, 9.80%, 6.90% and 6.20% w/w respectively with reference to the air dried plant.

Anti inflammatory activity:-

To test the anti inflammatory activity of leaf extracts of *Allophylus serratus* three *in vitro* methods (Human Red Blood Cells stabilization Method (HRBC), inhibition of protein denaturation and protease inhibition) were selected. There reason why these assay methods selected was due to ethical challenges and the nonexistence of rationale to use animals for pharmacological research of new chemical compounds (Bouhlali *et al.* 2016).

Human Red Blood Cells stabilization Method (HRBC):-

HRBC stabilization evaluation of the leaf extracts was carried out according to the Gandhisan *et al.*, (1991), Murugan and Parimelazhagan, (2014) and Bouhlali *et al.* (2016) methods with some modifications. Whole human blood was collected from volunteers (who had not taken any NSAIDS for 2 weeks prior to the experiment) and mixed with equal volume of Alsever's solution which was prepared by dissolving 2% dextrose, 0.80% sodium citrate, 0.05% citric acid and 0.42% sodium chloride in sterilized double distilled water. The collected blood was mixed with equal volume of sterilized Alsever's solution. The mixture was then centrifuged at 3000 rpm and washed with isosaline. The blood was reconstituted with isosaline and a suspension in 10% (V/V) was made. The assay reaction mixture which is 4.5mL consists of 1 mL phosphate buffer(0.15M, PH=7.4) , 2 mL hyposaline (0.36%), 1 mL of leaf extracts (1 mg/mL of respective organic solvents) and 0.5 mL RBC (10% v/v Red blood cells in isosaline) suspension (Sadique *et al.*, 1989 and Saket *et al.*, 2010). The reaction mixture without the extract was served as a control. The assay mixtures were incubated at 37°C for 30 min and centrifuged and the supernatant solution was estimated using UV analysis at 560 nm. Diclofenac sodium (100 and 200 µg/ml) was used as the reference drug. The control which was prepared by omitting the extract represents 100% HRBC lysis. Percent membrane stabilization activity was calculated by the formula (Sakat *et al.*, 2010; Reshma *et al.*, 2014)

$$\text{Percent protection} = 100 - \frac{Ab_1}{Ab_2} \times 100$$

Where Ab_1 is the absorbance of the sample, and Ab_2 is the absorbance of the control.

Inhibition of Protein denaturation:-

Inhibition of protein denaturation was evaluated according to the method of Mizushima and Kobayashi (1968), Sakat *et al.*, (2010), Rashma *et al.*, (2014) and Bouhlali *et al.* (2016) with some modifications. 0.5ml of bovine serum albumin (1% aqueous solution) was mixed with 0.1 mL of distilled water. pH was adjusted at 6.3 using 1 N HCl. 0.1 ml *Allophylus serratus* leaf extracts (mg/ml of respective organic solvents) was added to the reaction mixture. The mixture was then incubated at 37°C for 30 min and then heated at 51°C for 10 min. After cooling the samples to room temperature, 2.5 mL of phosphate buffer solution (PBS) was added. The absorbance was measured spectrophotometrically at 660 nm. For negative control 0.1 mL distilled water and 0.5 mL of bovine serum albumin were used. Acetyl salicylic acid was used as standard. The percentage inhibition of protein denaturation was calculated using the following formula.

$$\text{Percent inhibition} = \frac{Ab_1 - Ab_2}{Ab_1} \times 100$$

Where Ab_1 is the absorbance of the control, Ab_2 is the absorbance of the sample. The control represents 100% protein denaturation. The results were compared with acetyl salicylic acid (400 µg/ml) treated samples.

Protease inhibition Assay:-

The Inhibition of protease (trypsin) was evaluated by the previously reported method of Reshma *et al.*, (201 and Sakat *et al.*, (2010) with some modifications . To 100 ml of bovine serum albumin 100 µL of each leaf extract was added. The mixtures were incubated at room temperature for 10 minutes. 250 µL of trypsin was added to the mixtures to inhibit the reaction. Then the mixtures were centrifuged at 3000rpm for 5 minutes. The absorbance of the supernatant of each was measured at 210 nm. Acetyl salicylic acid was used as a positive control. Each experiment was done in triplicates and percent inhibition of protease was calculated by the following formula.

$$\text{Percent inhibition} = \frac{\text{Ab}_1 - \text{Ab}_2}{\text{Ab}_1} \times 100$$

Where Ab_1 is the absorbance of the control, Ab_2 is the absorbance of the sample.

Antimicrobial activity:-

The extracts were dissolved in DMSO to obtain different concentrations (10mg/ml, 50 mg/ml, 100mg/ml) The assay was performed in triplicate. DMSO (10% in water) was used as negative control. The bacterial strains used for the test were *Staphylococcus aureus* (*S. aureus*), *Bacillus subtilis* (*B. subtilis*), *Escherichia coli* (*E. coli*) and *Pseudomonas aeruginosa* (*P. aeruginosa*) and the fungal species used were *Aspergillus fumigatus* (*A. fumigatus*) and *Candida albicans* (*C. albicans*).

Four strains of bacteria were procured from microbial culture collection and loops full of all the bacterial cultures were inoculated in LB broth (Luria Bertani broth) (HiMedia) and were incubated at 37°C for 24 h. Fresh bacterial cultures were taken and spread on the LB agar (Luria Bertani agar) plates using glass rod to cultivate bacteria. Six millimeter diameter wells were made on agar plate with the help of gel puncture. Twenty micro liters of leaf extracts and DMSO (as control) were inoculated to the well, and then the plates were incubated in incubator at 37 °C for 24 h. The antimicrobial activity was measured based on the inhibition zone around the wells. The formation of clear inhibition zone of ≥ 7 mm diameters around the wells was regarded as significant susceptibility of the organisms to the extract (Okwori *et al.* 2007).

Statistical analyses:-

All the experiments were done in triplicates and the results were expressed as Mean \pm SD. The data were statistically analyzed using one way ANOVA followed by Duncan's test. Mean values were considered statistically significant when $p > 0.05$.

Results and discussion:-**Human Red Blood Cells stabilization Method (HRBC):-**

HRBC membrane stabilization method was used to investigate *in vitro* anti inflammatory activity of *Allophylus serratus* leaf extracts. The inhibition of hypotonicity induced HRBC membrane lysis (which is the response of inflammation) stabilization was taken as a measure of the anti inflammatory activity. HRBC membrane may be considered as a model of the lysosomal membrane which plays an important role in inflammation (Weissmann *et al.*, 1969). Lysosomal membrane stabilization is important in limiting the inflammatory responses. The mechanism is by the release of lysosomal constituents of activated neutrophil such as bactericidal enzymes and proteases which cause further tissue inflammation and damage upon extracellular release (Murugasan *et al.*, 1981). HRBC membrane stability test is based on the finding that non-steroidal anti-inflammatory agents inhibit heat induced lysis of erythrocytes, most likely by stabilizing the membrane of the cell.

Compounds in medicinal plants which prevent the lysis of HRBC membrane caused by the release of hydrolytic enzymes contained within the lysosomes may relieve some symptoms of inflammation (Hess and Milonig, 1972). When the HRBC is subjected to hypotonic stress, the release of hemoglobin from cells will be prevented by anti-inflammatory compounds in plant extracts due to membrane stabilization. This indicates that some compounds in medicinal plant extracts used as anti inflammatory drug, have the ability to exert anti inflammatory activity (Olajide *et al.*, 2000). The mode of action for membrane stabilization of the plant extracts, may be due to the binding of phytochemical compounds to the HRBC membranes and changing the charges of the surface of the cells (Oyedapo *et al.*, 2010) or according to Shinde *et al.*, (1999) through a deformation of cells by interacting with membrane proteins or other compound in the erythrocyte membranes. This interaction may later provoke alteration of the surface charges of the cells (Oyedapo *et al.*, 2004). Chopade *et al.*, (2012) reported that membrane protective effect of plant extract may be due to their ability to adjust the intracellular concentration of calcium into the erythrocytes.

For example tannins and saponins show membrane stabilizing effects by binding cations and other biomolecules (Oyedapo *et al.*, 2001).

The percentage of HRBC membrane stabilisation test for leaf extracts and positive control Diclofenac sodium were carried out at concentrations of 100, 200, 400, 800, 1000 $\mu\text{g/ml}$. The percent membrane stabilization and membrane protection of HRBC membrane by *Allophylus serratus* leaf extracts at different concentrations and positive control Diclofenac sodium are given in Figure 1 and 2. *Allophylus serratus* leaf extracts at different concentrations showed significant HRBC membrane stabilization. It was observed that methanolic extract of *Allophylus serratus* leaves exhibited highest membrane stabilizing activity of 77.64 ± 0.78 and 74.77 ± 0.66 compared to that of standard Diclofenac sodium which exhibited 92.65 ± 0.56 % and 89.73 ± 1.18 at 1000 $\mu\text{g/ml}$ and 800 $\mu\text{g/ml}$ concentrations respectively. The HRBC stabilizing activities of the extracts were concentration dependent, the membrane stabilizing activity increases with increasing concentration. This result is in line with previous report by Nagaharika *et al.* (2013) who revealed the anti inflammatory activity of leaves of *Jatropha gossypifolia* which increased with increasing concentration. Murugan and Parimelazhagan, (2014) also reported that methanol extracts of *O. parvifolia* at a concentration of 1 mg/ml significantly protected the lysis of erythrocyte membrane induced by hypotonic solution which is comparable to the standard Diclofenac sodium.

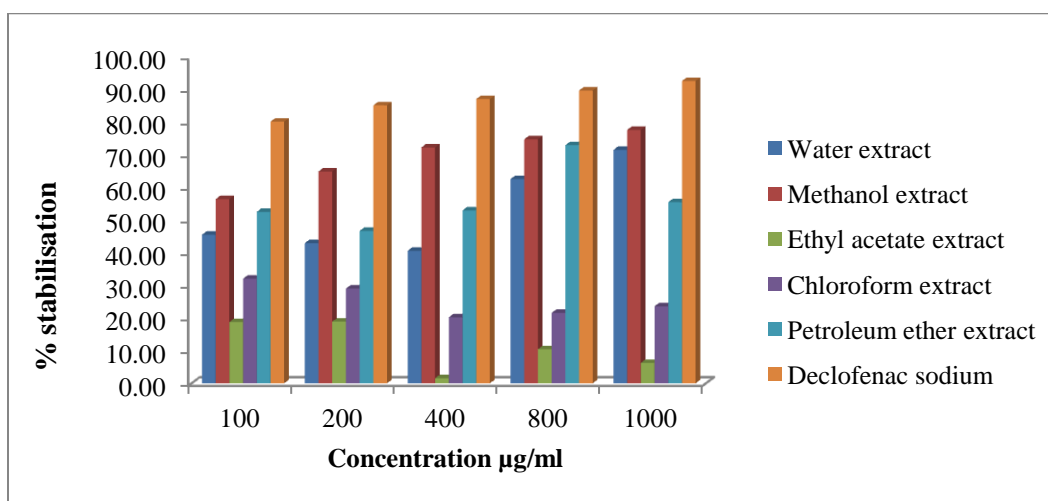


Figure 1:- Percentage of membrane stabilization of leaf extracts of *Allophylus serratus*.

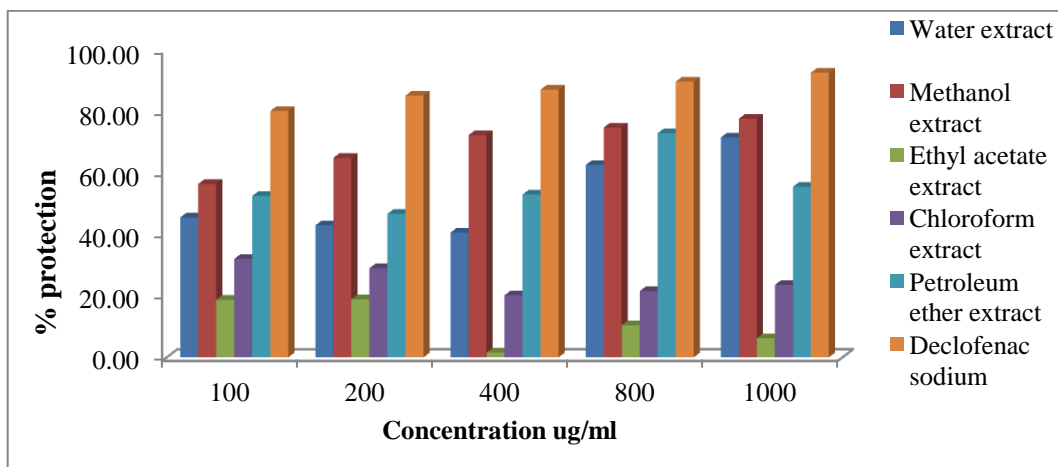


Figure 2:- Percentage of membrane protection of leaf extracts of *Allophylus serratus*.

Inhibition of Protein denaturation:-

The body response to infection, injury or destruction is what we call inflammation. Inflammation is characterized by pain, swelling, redness and disturbed physical functions. It is a normal body protective function against injury and

infection. One of the well documented causes of inflammation is protein denaturation. This is because, most biological proteins lose their function when denatured. The inhibition of protein denaturation by *Allophylus serratus* leaf extracts were studied by inhibition of protein denaturation method as anti inflammatory activity. The results of inhibitory effect of different extracts of *Allophylus serratus* leaf on protein denaturation are given in Fig 3 and Table 1. Among the different extracts, methanolic extract at concentrations of 200-1000 $\mu\text{g/ml}$ showed significant percent inhibition which was comparable to the standard Acetyl salicylic acid (93.50 %) at 400 $\mu\text{g/ml}$ (Fig 3). The methanolic extract showed the highest percent inhibition (70.73%) at 400 $\mu\text{g/ml}$ concentration. More over at 1000 $\mu\text{g/ml}$ the percent inhibition of protein denaturation increased to 95.93%. The results are in agreement with Murugan and Parimelazhagan, (2014) who reported that methanol extracts of *O. parvifolia* at a concentration of 1 mg/ml significantly protected the albumin from denaturation which is comparable to the standard Acetyl salicylic acid. The inhibitory effect was concentration dependent which increases with increasing the concentration of extract from 100 $\mu\text{g/ml}$ to 1000 $\mu\text{g/ml}$. Other studies also revealed that several anti inflammatory drugs also have shown dose dependent inhibition of protein denaturation (Grant *et al.*, 1970; Mizushima and Kobayashi, 1968). The ability of *Allophylus serratus* to inhibit protein denaturation is presumably due to its anti inflammatory activity.

Table 1:- Percent inhibition of protein denaturation by *Allophylus serratus* leaf extracts. Each value represents the mean \pm SD (n=3). ASA= Acetyl salicylic acid

Concentration ($\mu\text{g/ml}$)	% inhibition					
	Water extract	Methanol extract	Ethyl acetate extract	Chloroform extract	Petroleum ether extract	Acetyl salicylic acid
100	4.07	31.71	5.69	13.82	8.94	
200	8.13	41.46	10.57	16.26	18.70	
400	12.20	70.73	13.01	20.33	17.89	93.50
800	44.72	91.06	30.08	55.28	35.77	
1000	69.11	95.93	58.54	65.85	58.54	

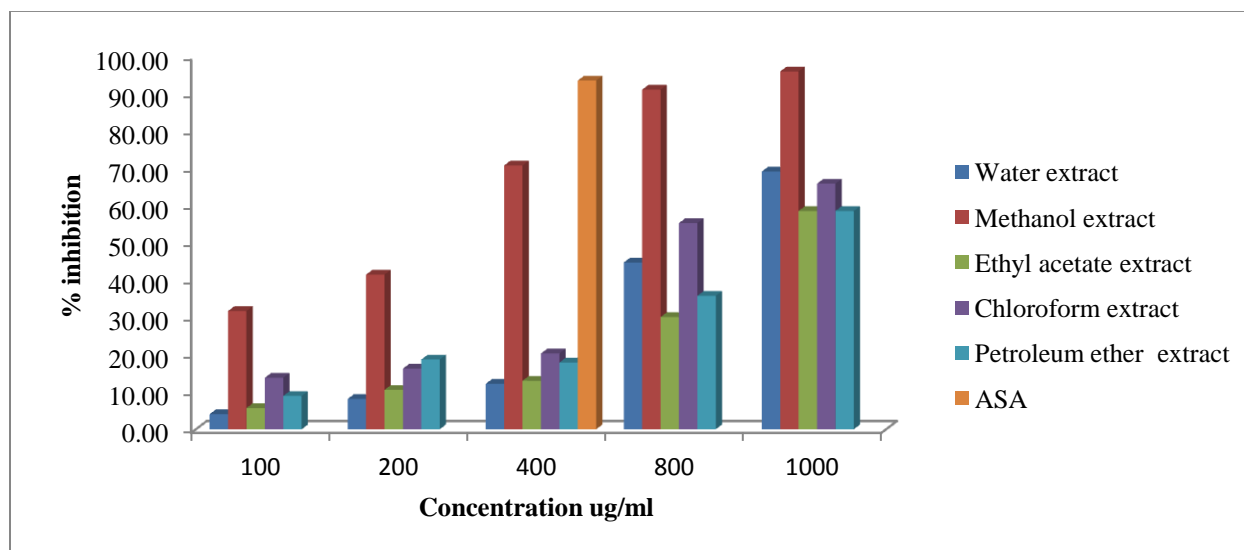


Figure.3:- Percent inhibition of protein denaturation by *Allophylus serratus* leaf extracts Each value represents the mean \pm SD (n=3). ASA= Acetyl salicylic acid

Protease inhibition Assay:-

During inflammation leukocytes protease enzymes play an important role in tissue damage. Protease inhibitors provide significant protection against this tissue damage developed by proteases during inflammatory reactions (Das and Chatterjee 1995). Phenolic compounds, tannins and flavonoids that are found in medicinal plants have been reported to have anti-inflammatory properties (Amarowicz, 2007). These compounds have the ability to inhibit protease and prevent tissue damage. In the present study *Allophylus serratus* leaf extracts were tested for their anti inflammatory activity by protease inhibition assay method. The result of this study showed that ethyl acetate extract has the highest protease inhibition activity compared to other extracts. The ethyl acetate extract produced the highest percentage inhibition (67.63%) at 100 $\mu\text{g/ml}$ concentrations. The protease inhibitory action was dose dependent with

the greater inhibitory effect of 81.02 % at 1000 µg/ml. Chloroform and methanol extracts at 1000 µg/ml also showed significant protease inhibitory activity 76.39 % and 75.42 % respectively Fig 4.

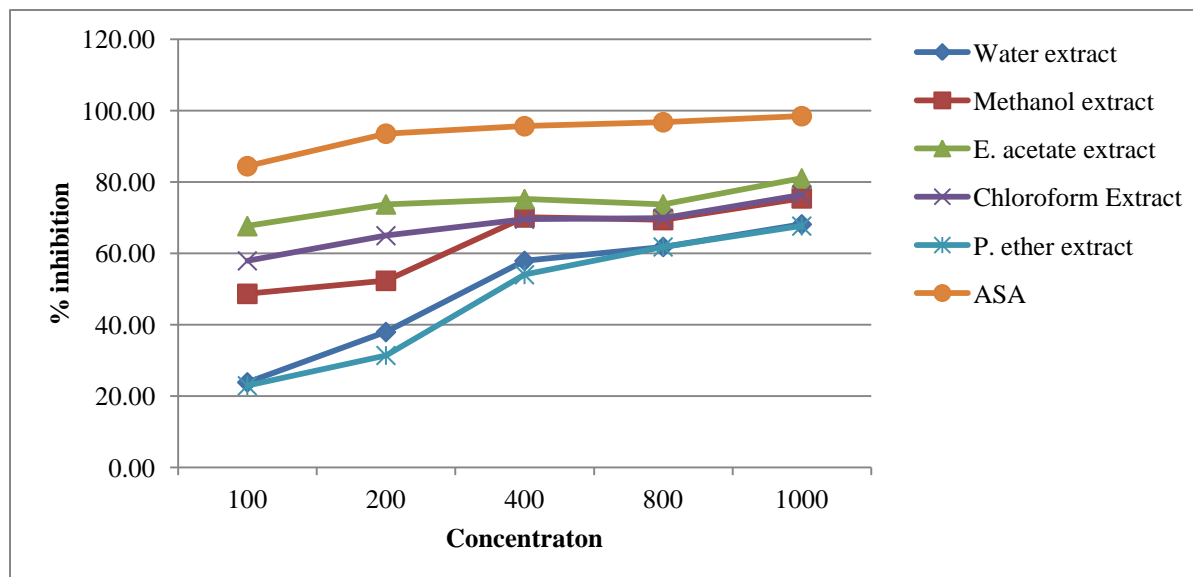


Figure 4:- Percent protease inhibition by *Allophylus serratus* leaf extracts Each value represents the mean \pm SD (n=3)

The anti inflammatory activity assay of different solvent extracts of *Allophylus serratus* leaf by HRBC membrane stabilization, inhibition of protein denaturation and protease inhibition produced promising results. The possible reason for the anti inflammatory activities of the plant is the presence of Phenolics, flavonoids, tannins and saponins which have anti inflammatory activity (Oyedapo, 2001). The in vitro anti-inflammatory effect of plant material may be due to synergistic effect rather than single constituent of their polyphenols contents (Bouhlali *et al.* 2016). This study also showed that the type of solvent used in extraction is important to find polyphenolic compounds from plant extracts. Comparison of these five solvents revealed that, they produce different anti inflammatory activities. Overall, methanolic extract, water extract and ethyl acetate extract were found to be good in obtaining anti-inflammatory substances from the extracts than the others.

Antimicrobial activity:-

An agar well diffusion assay method was used to determine the antimicrobial effects of the extracts of the leaves of *Allophylus serratus* against four strains of bacteria (*E. coli*, *B. subtilis*, *S. aureus* and *P. aeruginosa*) and two strains of fungi *A. fumigates* and *C. albicans*. The results of antimicrobial activity test are given in Table 2 and Fig 5. The results revealed variability in the inhibitory effect of each extracts against tested bacteria and fungi.

Table 2:- Antimicrobial activity of the different extracts of the leaves of *Allophylus serratus* on selected bacterial and fungal species

Microbial strain	Zone of Inhibition (mm)					
	Water extract	Methanol extract	Ethyl acetate extract	Chloroform extract	Petroleum ether extract	Control
<i>E. coli</i>	18 \pm 0.01	14 \pm 0.12	15 \pm 0.04	-	-	-
<i>S. aureus</i>	23 \pm 0.23	18 \pm 0.25	13 \pm 0.01	-	-	-
<i>B. subtilis</i>	-	-	20 \pm 0.00	-	-	-
<i>P. aeruginosa</i>	-	-	-	20 \pm 0.17	22 \pm 0.09	-
<i>A. fumigates</i>	-	-	-	-	-	-
<i>C. albicans</i>	-	-	-	-	-	-

The water, methanol and ethyl acetate extracts displayed low activity against the *B. subtilis* and *P. aeruginosa* except the ethyl acetate extract which showed high activity against *B. subtilis* (20 \pm 0.00mm). The water extract displayed high activity against *S. aureus* (23 \pm 0.23mm). The water extract, methanol extract and ethyl acetate extract were

slightly more active against *E. coli* (18 ± 0.25 , 14 1st and 15 mm respectively). Chloroform and petroleum ether extracts inhibited the growth of *P. aeruginosa* (20 ± 0.17 and 22 ± 0.09 mm respectively) but no activity against the rest tested bacterial strains (Table 2). Under the concentrations and conditions tested, Chloroform and petroleum ether extracts were active against Gram-negative bacterial strain (*P. aeruginosa*). On the other hand no any extract of *Allophylus serratus* showed anti fungal activity against tested strains of fungi (*A. fumigates* and *C. albicans*).

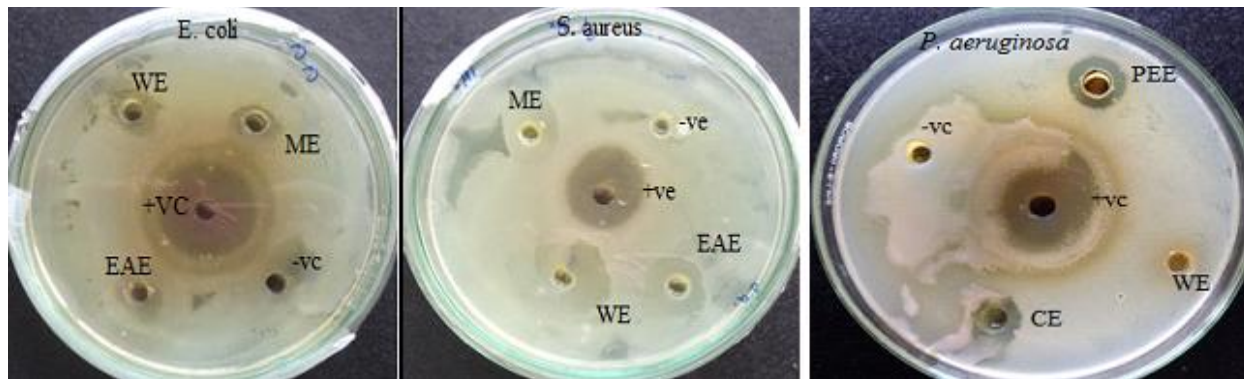


Figure 5:- Antimicrobial activity of *Allophylus serratus* leaf extracts. -vc= negative control, +vc= positive control, WE= water extract, ME= methanol extract, EAE= Ethyl acetate extract, CE= Chloroform extract, PEE= Petroleum ether extract.

The antibacterial activity of plant extracts as recorded in previous studies is attributed to the phytochemicals such as flavonoids, alkaloids and tannins present in plants. For example flavonoids in plants have antimicrobial (Maria Lysete *et al.*, 2009) and antiviral (Mehrangiz *et al.*, 2011) properties. Alkaloids isolated from plant are also found to have antimicrobial properties (Ahmed *et al.*, 2010). Tannins have antibacterial activity and kill bacteria by damaging its cell membrane (Mohamed Sham *et al.*, 2010). (Arokiyaraj *et al.*, (2009) reported that seeds extracts of *Vitex agnus-castus* has antimicrobial activity due to the presence of alkaloids, saponins, tannins, flavonoids, and glycosides contents.

Allophylus serratus as reported in previous studies also contain phytochemicals such as saponins, tannins, flavonoids and glycosides (Sanmuga *et al.*, 2012). There for the antibacterial activity of *Allophylus serratus* leaves extracts may be attributed to the presence of the phytochemicals reported to be present in it.

Conclusion:-

The anti inflammatory activity assay of *Allophylus serratus* leaves extracts exhibited membrane stabilization hypotonicity induced lysis of erythrocyte membrane, protein denaturation and protease inhibition effects. The plant extracts also showed significant anti bacterial activities against different bacterial strains. This implies that the extracts have significant anti inflammatory and anti bacterial activity compared to the standard medicines. The presence of various phytochemicals such as alkaloids, polyphenols tannins, saponins and flavonoids may be responsible of these activities. Therefore the results of this study provides scientific base for the traditional use of this plant as anti inflammatory and antibacterial agent. These results also showed that *Allophylus serratus* could be a potential natural source of compounds which have greater importance as therapeutic agent in inflammation related disorders and bacterial infections. Further studies are required to identify and isolate the active components of the extracts responsible for their anti-inflammatory properties and their mode of action.

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RESEARCH ARTICLE

INCIDENCE OF RH NEGATIVE BLOOD GROUP IN BLOOD DONORS FROM HIGH ALTITUDE AREAS OF JAMMU AND KASHMIR STATE, INDIA.

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Abstract

According to presence or absence of antigens, human blood can be divided into different blood groups. Rh blood group is one of the most complex blood groups in humans. Rh positive has D antigen while Rh negative lacks the D antigen. This one year study was done in which 600 blood donors were tested for Rh blood group in the blood-bank laboratory by test-tube method. 300 blood donors were from high altitude areas (above 5500ft the sea level), while the 300 blood donors were from plain areas, of Jammu And Kashmir State, India. In high altitude blood group donors 87% were Rh positive while 13% were Rh negative. In plain area blood donors 93% were Rh positive while 7% were Rh negative. Thus there is a high incidence (13%) of Rh negative donors in high altitude donors as compared to plain areas.

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Introduction:-

Humans contain a series of glycoprotein's and glycolipids on the surface of RBC's which constitute the blood group antigens. According to presence or absence of antigens human blood can be classified into different blood groups like ABO blood group, MNS blood group, Rh blood group Etc. The international society of blood transfusion definition of the blood group system is where one or more antigens are controlled at a single gene locus or by two or more very closely linked homologous genes with little or no observable recombination between them (1). The Rh blood group is one of the most complex blood groups known in humans. It was named after Rhesus monkey. With 49 antigens so far described, it is largest of 29 blood group systems. Rh antigens are located on two Rhesus proteins RHD and RHCE. Rhesus proteins are expressed only in membranes of red blood cells (2).

ABO blood group system were discovered by Austrian pathologist Karl Landsteiner (3).

Following discovery of ABO, blood group, Rh antigen was discovered by Levine and Stetson in 1939 (4).

Among the Rh antigens D,C,c,E and e are most important which are encoded by two adjacent gene loci, the RHD gene which encodes the RHD protein with the D antigen and variants(5).

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The RhCE gene which encodes RhCE protein with the C,E,c and e antigens and variants. Gene for Rh system is located on chromosome 1 (6).

Rh positive and Rh negative refer to D antigens only. Rh positive has D antigen and Rh negative does not have D antigen.

Incidence of Rh blood groups varies in different parts of world. Factors attributed to such difference include racial differences, geographical variation, external environment and genetic makeup (7,8,9).

The Caucasians of European descent have highest incidence of Rh negative phenotype (15-17%). It is less common in Africans, in Asians it is less than 1% (10).

Rh antigens, especially D are highly immunogenic. They have been implicated in hemolytic disease of newborn and hemolytic transfusion reactions. The knowledge of distribution of Rhesus blood group at local and regional levels is helpful in the effective management of blood banks and safe blood transfusion service (11).

Materials and methods:-

This prospective one year study was conducted from Nov 2015 to Nov 2016 in the department of blood bank of Sher-i-Kashmir Institute of Medical Sciences, Srinagar, India which is a deemed university, super specialty hospital of north India. This study was done to find out incidence of Rh negative blood group among donors belonging to high altitude areas (5500ft above the sea-level) of Jammu and Kashmir (sample size 300). This was compared with incidence of Rh negative blood group among donors belonging to plain areas (sample size 300). Both voluntary and replacement donors were included in the study. Donors were selected strictly as per AABB's (American association of blood bank) recommendation.

Procedure:-

Three methods are available for detecting Rh antigens, e.g. slide method, tube method and micro-plate or micro-well method. We selected test-tube method for our study, as it is simple, reliable, cost effective method. It allows for long incubation without any drying, unlike slide method which causes drying, of blood giving at times false impression of agglutination. The tubes can further be centrifuged to enhance antigen-antibody reaction.

Requirement & Material:-

- 75x 10mm tubes
- Test serum Igm monoclonal Anti D/ saline Anti D
- Test red cells
- Rh positive and Rh negative control cells (for monoclonal Anti D)

Methods:-

A detailed history and physical examination of donors was done. Their residential address, age and sex was noted. After taking informed consent of donors, blood samples were collected under all aseptic precautions. 300 blood samples were collected from high altitude areas donors (5500ft above the sea level). 300 blood samples were collected from non hilly areas or plain areas .

2-4% washed red cell suspension of test sample was prepared. To one drop of 2-4% red cell suspension 2 drops of anti D serum was added and was mixed by gently shaking the tube. The test tube was incubated at room temp for 60 minutes. Agglutination was seen against a well-lit background. Results, were recorded. All negative results were checked using a microscope. Relevant data was collected using a proper proforma form.

Result:-

1. A total of 600 donors , 300 each from high altitude areas (above 5500ft) and plain areas were screened for Rh typing.
2. Out of 300 donors of high altitude areas 291(97%) were males while as 9(3%) were females. In plain areas donors 288(96%) were males and 12(4%) were females.
3. In high altitude group donor , the age group of 18-30 years comprised 65% of the total donor. In plain area donors, this age group comprised 51% of total donors.

4. In high altitude group donors 261 donors (87%) were Rh positive while as 39(13%) were Rh negative. In plain area group donors, 279(93%) were Rh positive while as 21(7%) donors were Rh negative. Thus there is a high incidence of Rh negative group in high altitude area donors (13%), which is the salient feature of this study.
5. In high altitude area donors, of 291 males, 252(86.6%) were Rh positive and 39(13.4%) were Rh negative. Among females all 9 (100%) were Rh positive with no Rh negative females, were found.
6. In plain area donors out of 288 males, 267(92.7%) were Rh positive and 21(7.3%) were Rh negative. Among females all 21(100%) were Rh positive with no Rh negative females, were found.
7. In high altitude area donors the most common blood group was found to be B+ve (35%), while as in plain area donors two blood group were commonest. It was found to be B+ve (33%) and O+ve also (33%).
8. In high altitude area donors, the least common blood group was found to be AB-ve (1%), while in plain area donor group no AB-ve donor was found (0%).

Discussion:-

Blood groups are genetically determined. The incidence of ABO and Rh genes and their phenotypes vary widely across races and geographical boundaries. The resultant polymorphism remains important in population genetic studies, estimating the availability of compatible blood, evaluating the probability of hemolytic disease in new born, resolving disputes of paternity/ maternity and for forensic purposes.

In the present study among high altitude donors 87% were Rh positive and 13% were RhD negative. Among donors of plain areas 93% were RhD positive and 7% were RhD negative. Incidence of RhD antigen in most parts of India varies from 91-98% for RhD positive and 2-9% for Rh D negative. In Britain and U.S.A, the distribution of RhD positive is 83% and 85% respectively with RhD negative of 17% and 15% respectively (12).

It is postulated that ancestors of high altitude area donors, of Jammu and Kashmir state, India have migrated from central and south east Asia the erstwhile silk route which have a similar pattern of distribution.

In six muslim dominated districts of Aligarh, Uttarpradesh, what surfaced as a contrast was 15% incidence of Rh negative blood groups. It supports the ancestry background from middle east.

The comparison of frequency and distribution of Rh blood groups in blood donors of high altitude population and plain regions of Jammu and Kashmir, India with the studies carried out at national and international level is given in table 1 and table 2.

Table no 1:- Comparison of present study with Indian studies (in percentage).

RH D	Rh positive	Rh negative
Present study		
• High altitude areas	87%	13%
• Plain areas	93%	7%
Bangalore (13)	94.2%	5.79%
Chittoor (14)	90.6%	8.42%
Vellore (15)	94.5%	5.47%
Shimoga- Malnad(16)	94.9%	5%
Davangere(17)	94.8%	5.5%
Eastern ahmedabad (18)	94.2%	5.8%
Punjab (19)	97.3%	2.7%

Table no 2:- Comparison with studies outside India (in percentage).

Study	Rh D Positive	Rh D Negative
Present study		
• High altitude	87%	13%
• Plain area	93%	7%
Pakistan (20)	89.1%	10.9%
Nepal (21)	96.7%	3.3%
Britain (22)	83%	17%

USA (23)	85%	15%
Niger Delta(24)	93.9%	6.1%

The knowledge of distribution of Rh D negative blood groups is essential for the effective management of blood bank's inventory. The study data would help in establishment of more efficient blood transfusion service that would meet the increasing demand for Rh D negative blood groups. Voluntary blood donation camps can be organized in high altitude areas, so that resources for Rh negative bloodgroups, can be tapped. For this the general public of the area need to be motivated.

Conclusion:-

The study showed high incidence of Rh D negative blood group (13%) in population of high altitude areas (5500ft above sea level) , of Jammu and Kashmir state, India. This incidence was almost double (7%) as compared to the plain areas in studied area. It is hoped that the study data generated will help in establishment of more efficient functioning blood transfusion service especially the need for negative blood groups. It could also help formulate robust marrow registries in India a process in infancy yet.

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RESEARCH ARTICLE

SERUM TOTAL CALCIUM, MAGNESIUM, POTASSIUM AND PH IN SUDANESE PATIENTS WITH TYPE 2 DIABETES MELLITUS.

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Abstract

This study was carried out to measure serum levels of the calcium, magnesium, potassium and PH in patients with type 2 DM. Sixty samples were collected from patients in period between October to November 2016, chosen randomly from Military hospital and Sixty apparently, healthy individuals as controls, to assess the levels of calcium, magnesium, Potassium and pH in type2 D M.

Serum calcium and magnesium were measured by using automation, Mindray380, potassium by using easylyte and pH by using Cobas b 221, and results were analyzed using statistical of package social science (SPSS), computer program.

The study showed that, the serum levels of magnesium and potassium were significantly decreased, (p-value =0.00) in diabetic patients compared to control group. While the level of PH was significantly increased, (p-value= 0.000) and there was no significant difference between the mean of calcium level in patients and control group. Mean \pm SD cases versus control. (1.43 \pm 0.26versus2.10 \pm 0.29) For magnesium. (3.1 \pm 0.39versus4.10 \pm 0.48) for potassium. (7.99 \pm 0.06 versus 7.40 \pm 0.02) for PH.

(8.77 \pm 1.00 versus 8.79 \pm 0.80, p-value =0.997) for calcium.

There was insignificantly weak positive correlation between serum magnesium in patients used insulin and diabetic patients not used insulin (r= 0.1 / p= 0.627) , while there were no correlations between serum calcium, potassium and PH in diabetic patients used insulin and diabetic patients not used insulin.(r=0.073, p- value= 0.70),(r=0.027 , p- value= 0.888) ,(r=0.08, p-value=0.676) respectively.

It is concluded that; the levels of magnesium and potassium are significantly decreased in patients with type 2 DM, and the level of PH is significantly increased and there is no significant difference in the level of calcium in type 2 diabetic patients compared to control group.

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Introduction:-

Diabetes mellitus is characterized by hyperglycemia due to absolute or relative deficiency of insulin⁽¹⁾, leading to impaired metabolism of carbohydrates, proteins, fats, water and electrolytes. The persistence of these metabolic disturbances lead to permanent and irreversible functional and structural changes in the cells of the body which in

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turn lead to the development of “diabetic complications”, characteristically affecting, the cardiovascular system, eye, kidney and nervous system mainly⁽²⁾. A relationship between DM and minerals is frequently reported. Alteration in the metabolism of trace elements like calcium, magnesium is associated with DM⁽³⁾. Trace elements are accepted as essential for optimum health, because of their diverse metabolic characteristic and functions⁽⁴⁾. Trace elements participate in production of reactive oxygen species (ROS), which contribute to oxidative stress. Oxidative stress contributes to the pathogenesis of many diseases including DM. Chronic complications of diabetes mellitus can be divided into vascular and nonvascular complications. The vascular complications of DM are further subdivided into micro vascular (retinopathy, neuropathy, nephropathy) and macro vascular complications coronary artery disease, peripheral arterial disease, cerebrovascular disease⁽⁵⁾. Several of the complications of diabetes may be related to increased intracellular oxidants and free radicals⁽⁶⁾.

Magnesium, calcium and potassium are an essential element involved in glucose homeostasis. It is a cofactor for various enzymes in carbohydrate metabolism. It is also involved at multiple levels in insulin secretion, binding and activity. Reduced level of magnesium has been documented in type 2 DM^(3, 4,7 and 8). Hypomagnesaemia may have negative impact on glucose homeostasis and insulin sensitivity in type 2 DM patients⁽⁹⁾. Hypomagnesaemia may also have some effect in the development of diabetic complications with other risk factors⁽¹⁰⁾. The present study was designed to evaluate the serum levels of potassium; magnesium and calcium in type 2 DM.

Material and methods:-

Study Population The study was carried out at College of Medical laboratory Science and the subjects were recruited from military hospital in Khartoum from period between October to November 2016. A total of 60 type 2 DM and 60 apparently, healthy individuals as controls were enrolled in this study. The study was approved by hospital's ethics committee. Informed consent was obtained from patients before blood sampling.

Inclusion criteria:-

Sudanese patients with type 2 DM and apparently healthy volunteers (matches age and sex with the cases) were included.

Exclusion criteria: patients with hypertension, renal disease, heart disease and other disease related to pH imbalance were excluded.

Blood sample and analysis:

About 2ml of venous blood was collected from the antecubital vein by taking aseptic precautions. Care was taken to prevent venous stasis during the sample collection. The blood was allowed to clot and the serum was separated by centrifugation. The estimation of the parameters was carried out within 4-6 hrs. The samples were analyzed for serum calcium and magnesium by using automation, Mindray 380⁽¹¹⁾, potassium by Electrolyte analyzer (easlyte) and pH by using Cobas b 221⁽¹²⁾. The internal control sera of two different levels were used to calibrate the instruments.

Data was analyzed using SPSS computer program, the mean and standard deviation were obtained and the independent 't.test' used for comparison (p value of ≤ 0.05) was considered significant.

Results:-

In comparison with the controls, diabetic patients (type2) had significantly lower in means serum magnesium and potassium (1.43 ± 0.26 versus 2.10 ± 0.2951 mg/dL), (3.1 ± 0.39 versus 4.10 ± 0.48 mmol/L) p value < 0.05 respectively. There was significant increase in mean of PH in type 2DM compared to control group (7.99 ± 0.06 versus 7.40 ± 0.02) p value < 0.05 . There was insignificant difference in mean serum calcium compared to control group (8.77 ± 1.00 versus 8.79 ± 0.80 , p-value = 0.997) p-value ≥ 0.05 as in table 1.

Table (1):- means comparisons of serum calcium, magnesium, potassium and PH in type 2 diabetes mellitus compared to control group.

Variable	Patients (mean \pm SD) N=60	Control (mean \pm SD) N=60	P-value
Calcium (mg/dL)	8.77 \pm 1.00	8.79 \pm 0.80	0.997
Magnesium(mg/dL)	1.43 \pm 0.26	2.10 \pm 0.29	0.000
Potassium (mmol/L)	3.10 \pm 0.39	4.17 \pm 0.48	0.000
PH	7.99 \pm 0.06	7.40 \pm 0.02	0.000

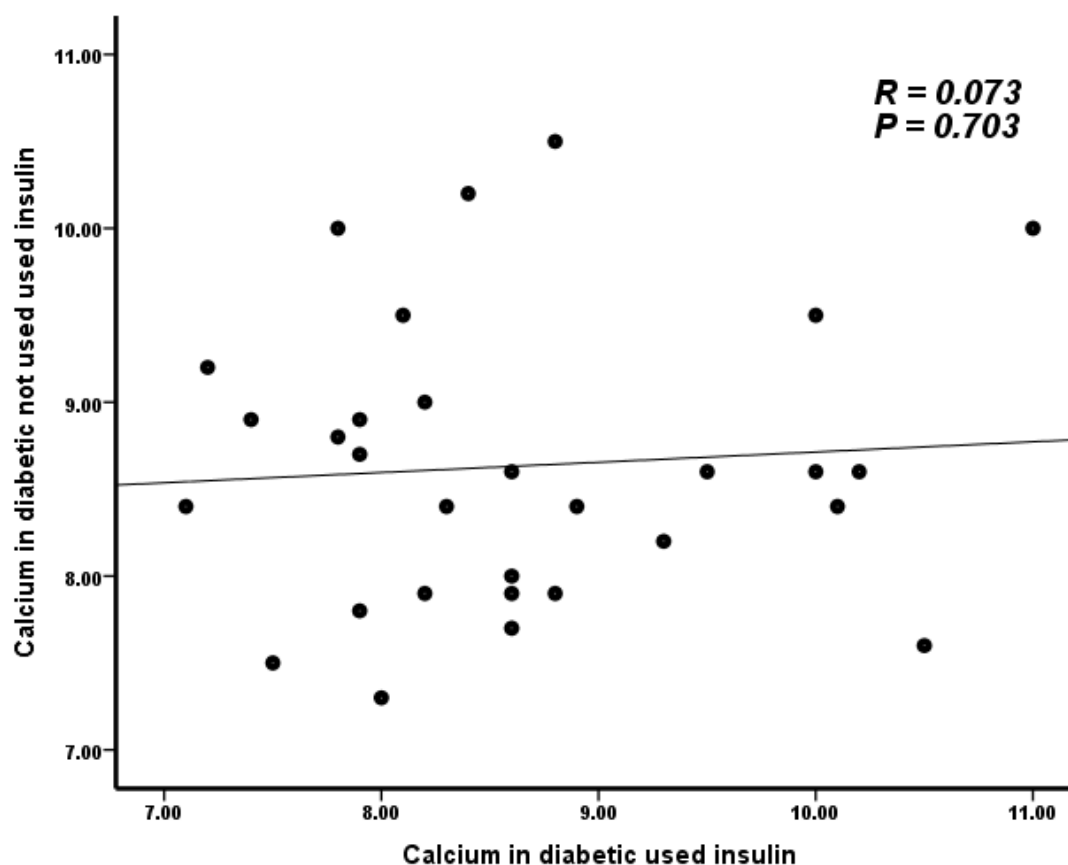


Figure (1):- A scatter plot shows the correlation between calcium level in diabetic patients used insulin and diabetic patients not used insulin ($r=0.073$, $p\text{-value}=0.703$).

There is no correlation between calcium level in diabetic patients used insulin and diabetic patients not used insulin.

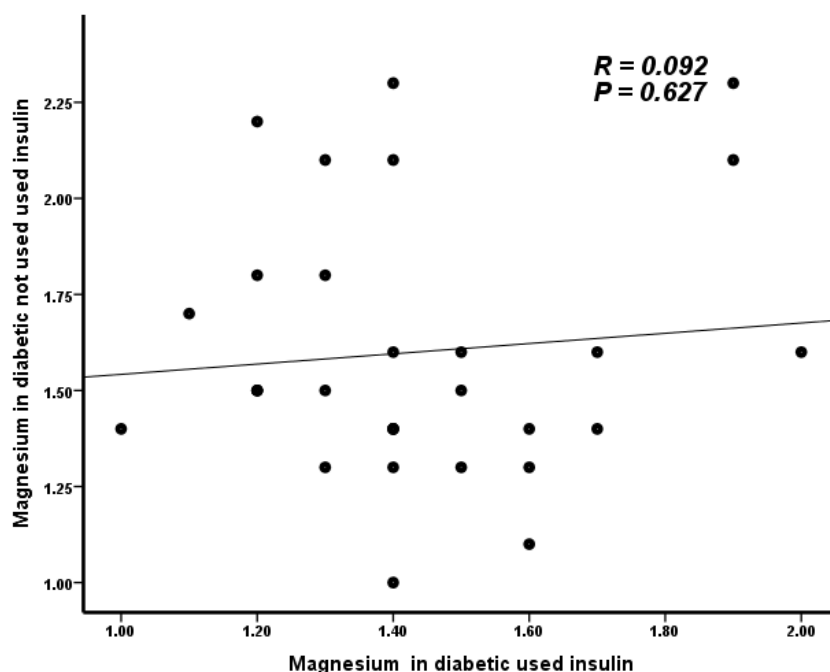


Figure (2):- A scatter plot shows the correlation between magnesium level in diabetic patients used insulin and diabetic patients not used insulin ($r=0.092$, $p\text{-value}=0.627$).

There is weak positive correlation between magnesium level in diabetic patients used insulin and diabetic patients not used insulin.

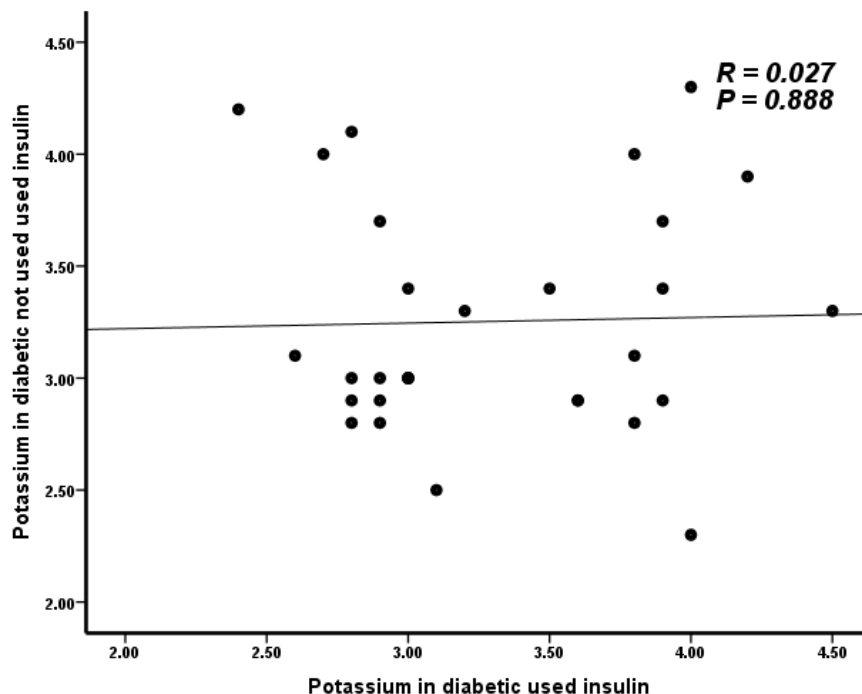


Figure (3):- A scatter plot shows the correlation between Potassium level in diabetic patients used insulin and diabetic patients not used insulin ($r=0.027$, $p\text{-value}=0.888$).

There is no correlation between potassium level in diabetic patients used insulin and diabetic patients not used insulin.

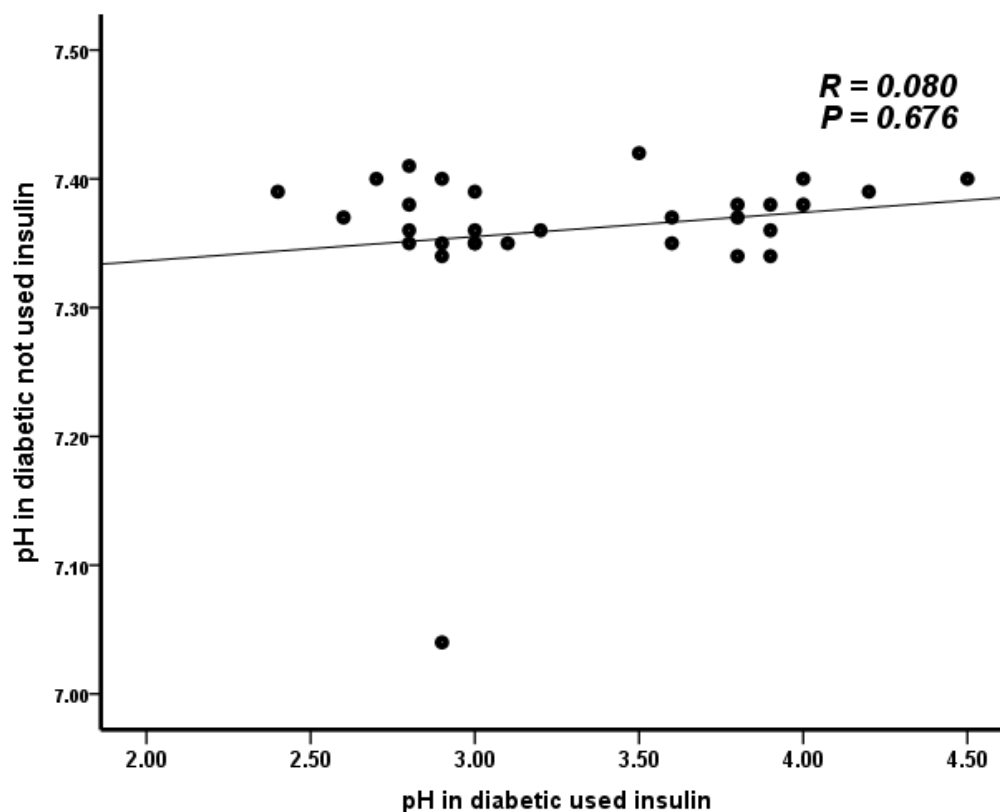


Figure (4):- A scatter plot shows the correlation between pH level in diabetic patients used insulin and diabetic patients not used insulin $r=0.080$, $p\text{-value}=0.676$).

There is no correlation between pH level in diabetic patients used insulin and diabetic patients not used insulin.

Discussion:-

A relationship between DM and minerals is frequently reported. Alteration in the metabolism of trace elements like calcium, magnesium potassium is associated with DM⁽³⁾. Trace elements are accepted as essential for optimum health, because of their diverse metabolic characteristic and functions⁽⁴⁾. Trace elements participate in production of reactive oxygen species (ROS), which contribute to oxidative stress. Oxidative stress contributes to the pathogenesis of many diseases including DM.

Results in this study found that, the serum levels of magnesium and potassium were significantly decreased in diabetic patients compared to control group. This results agree with another results carried by many authors^(13, 14), which finding confirmed that, there were significantly decreased in mean of serum magnesium and potassium in diabetic patients compared to control group, ($p\text{-value} = 0.00$). Also this study was in agreement with another study done by (Puri *et al.*, 2013), which showed hypomagnesaemia and hypokaleamia in diabetic patients and this may be a possible risk factor in development and progress of diabetic complications.

Also the results agree with another result which showed, Hypomagnesemia has been reported to occur at an increased frequency among patients with type 2 diabetes compared with their counterparts without diabetes⁽¹⁵⁾. Hypomagnesemia has been linked to poor glycemic control, coronary artery diseases, hypertension, diabetic retinopathy, nephropathy, neuropathy, and foot ulcerations. This study in agreement with another study, which finding confirm that, hypokalemia may reduce $\text{Na}^+\text{-K}^+\text{-2Cl}^-$ co-transport activity, the associated potassium extrusion through the potassium channel ROMK, and resultant diminution of the favorable trans membrane voltage that is required for paracellular Mg reabsorption. In addition, there is evidence to suggest that cellular potassium depletion may diminish Mg reabsorption at the DCT by yet unclear mechanisms.⁽¹⁶⁾

The result showed, the level of PH was significantly increased ($p\text{-value} < 0.05$) and there was no significant difference between the mean of calcium level in patients and control group ($p\text{-value} > 0.05$).

This result disagreed with another result, which showed Diabetes Mellitus is known to have its effect on almost all body system through alterations in structural and biochemical changes in tissues, alteration in calcium flux may adversely affect the insulin secretion as it is a calcium dependent process.⁽¹⁷⁾

There was insignificantly weak positive correlation between serum magnesium in diabetic patients used insulin and diabetic patients not used insulin ($r = 0.1$, $p = 0.627$). This result in agreement with result carried by researchers at the Institute of Internal Medicine, University of Palermo wrote, "Intracellular magnesium concentration has also been shown to be effective in modulating insulin action (mainly oxidative glucose metabolism). A poor intracellular Mg concentration, as found in noninsulin-dependent diabetes mellitus (NIDDM) and in hypertensive patients, may **result in a defective tyrosine-kinase activity at the insulin receptor level.**"⁽¹⁸⁾ There were no correlations between serum calcium, potassium and PH in diabetic patients used insulin and diabetic patients not used insulin. ($r = 0.073$, p -value = 0.70), ($r = 0.027$, p -value = 0.888), ($r = 0.08$, p -value = 0.676) respectively.

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RESEARCH ARTICLE

THE USEFULNESS & ACCURACY OF THE MODIFIED ALVARADO SCORE IN THE PREOPERATIVE DIAGNOSIS OF ACUTE APPENDICITIS.

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Abstract

This prospective study was conducted to evaluate the usefulness & accuracy of the modified Alvarado score in the preoperative diagnosis of acute appendicitis. The study included one hundred & fifty patients of different age groups & from both genders with provisional diagnosis of acute appendicitis presented to the emergency department of AL- Yarmouk Teaching Hospital over eighteen months period. All patients were evaluated on admission & given specific scores according to the variables of Modified Alvarado Score. In adult male patients acute appendicitis was confirmed in (40) of (42) patients giving sensitivity rate of (88.8%) & specificity rate of (91.6%), in adult female, it was confirmed in (25) of (28) patients with sensitivity rate of (80.6%) & specificity rate of (89.6%), while in children acute appendicitis was confirmed in (7) of (9) patients giving a sensitivity rate of (53.8%) & specificity rate of (75%). The overall sensitivity rate of Modified Alvarado Score was 80.9% & its specificity was 88.5%. Total negative appendectomy rate was 15.2%. The Modified Alvarado Score is simple, easy to apply & cheap complementary aid for supporting the diagnosis of acute appendicitis especially for junior surgeons.

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Introduction:-

The Alvarado Score was described in 1986.⁽¹⁾ & has been validated in adult surgical practice.⁽¹⁾ The Modified Alvarado Score is a simplified modification of the original Alvarado Score after the omission of one of its parameters by Kalan *et al.*⁽²⁾ Since 1886 when the classical signs & symptoms of acute appendicitis were first reported by Fitz.⁽¹⁾ Acute appendicitis has remained the most common surgical emergency.⁽³⁾ & surgery for acute appendicitis is the most frequent operation performed (10% of all emergency abdominal operations).^(5,6) Approximately 6- 7% of population will suffer from acute appendicitis during their life time .⁽⁷⁾ With early diagnosis & intervention the mortality rate from non complicated acute appendicitis is less than 0.1- 0.6% when there is gangrene, & 5% for perforated cases .⁽⁸⁾ The diagnosis of acute appendicitis is purely based on history, clinical examination & some simple laboratory investigations like white blood cells count. Recently the attempt to increase the diagnostic accuracy in acute appendicitis by imaging, laparoscopy & even radioactive isotope imaging have been included, however these techniques involve additional costs & are either complex or not easily available when they are most needed. A negative appendectomy rate of 20- 40% has been reported in literatures & many surgeons would accept rate of 30% as inevitable .⁽⁹⁾ The Modified Alvarado Score was designed to reduce the negative appendectomy rate without increasing the morbidity & mortality.⁽¹⁰⁾ At present many scoring systems for

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diagnosis of acute appendicitis are available & the Modified Alvarado Score is one of them, & is based on history, clinical examination & simple laboratory tests, & is very easy to apply as shown below.⁽¹¹⁾

	The	Modified	Alvarado
Score			
SYMPTOM			SCORE
A-Migratory right iliac fossa pain			1
B-Anorexia			1
C-Nausea & vomiting			1
SIGNS			
A-Tenderness in lower quadrant of abdomen			2
B-Rebound tenderness in right iliac fossa			1
C-Pyrexia equal or more than 37.5 c ⁰			1
3-Investigation			
A-Leucocytosis			2
TOTAL SCORE			9

The application of the score is based on the above variables (3 symptoms, 3 signs, 1 test). Patients with score 1- 4 are considered unlikely to have acute appendicitis, those with a score of 5- 6 have possible diagnosis of acute appendicitis not convincing enough to have urgent surgery, & those with score 7- 9 are regarded as probable acute appendicitis & underwent an appendicectomy.

Aim of the Study:-

1. To evaluate the usefulness & the validity of Modified Alvarado Score in the preoperative diagnosis of acute appendicitis.
2. To evaluate the accuracy of Modified Alvarado Score in relation to differences in age group & gender.
3. To compare the results of negative appendicectomy rate in those who were evaluated by Modified Alvarado Score with clinical literature figures.

Patients & Methods:-

This study was conducted on (150) patients admitted from the emergency department at Al- Yarmouk teaching Hospital with the clinical diagnosis of acute appendicitis during the period from June 2004 to December 2005. They were (79) males & (71) females with their age ranging from 8- 65 years, however, 21 of them were children (age < 12 years). All included patients were prospectively evaluated using the Modified Alvarado Score to determine whether they had acute appendicitis or not.

Following admission, initial assessment & baseline investigations, a specially designed forma was filled for each patient. This forma had general information about the patient plus seven variables based on the Modified Alvarado Score & information regarding the final outcome of each patient which was filled later on.

The admission of the suspected cases, evaluation & the decision to operate was made by the surgical, on call team, on the bases of one weekly duty for the eighteen months during which this study was conducted.

The sum of all variables were calculated for each patient & based on the results, patients were divided into:

Group one:- With score of > 7 (emergency surgery group).

They were (79) patients comprised of (42) male, (28) female & (9) children, these patients were prepared & underwent appendicectomy.

Group two. With score of (5- 6) (observational group). They were (71) patients comprised of (27) male, (32) female & 12 children. They were admitted & kept under observation with frequent assessments & re evaluations of the clinical data & their score, every 4- 6 hours. Abdominal ultrasonography was performed in 12 doubtful cases leading to the diagnosis of (2) cases of right ovarian cysts & (2) cases of right ureteric stones, while the condition of (41) patients were improved as shown by the decrease in the intensity of the symptoms & decrease in their score, & they were discharged with instructions that they should come back if their symptoms persist or increase in intensity. However, (26) patients of this group underwent appendicectomy for either persistence or increase in their symptoms, or increase score on subsequent reevaluation. Any patient presented with score of < (4) was not included in this

study as they were considered unlikely to have acute appendicitis & so they were discharged after initial symptomatic treatment with the instructions to come back if symptoms persist or increase in the intensity. Acute appendicitis was confirmed either peroperatively in those patients with macroscopical evidence of acute inflammation or by histopathological examination for those with macroscopically normal looking appendix

Results :- This study included (150) patients with clinical features suggestive of acute appendicitis. Among those patients (79) were male (52.7%), & (71) were female (47.3%). The age of the patients included in this study range from 8 year to 65 year, **The frequency distribution of the patients according to the results of Modified Alvarado Score demonstrate that the highest frequency was in those patients scored 5 (40 patients, 26.7%) & the lowest frequency in patients scored 9 (20 patients, 13.3),** However, from (150) patients (79) patients (52.6%) had score of (7) or more, comprised of (42) adult male, (28) adult female & (9) children, While (71) patients (47.4%) had a score of less than (7), (5- 6), comprised of (27) adult male, (32) adult female & (12) children. Another (41) patients with score of (5- 6) were discharged home as clinical improvement & decreased score was shown in subsequent reevaluation, & were instructed to come back if the symptoms persist or increase in intensity, while appendicectomy was performed in (26) patients with score of (5-6), they were (9) adult males, (10) adult females & (7) children.

Acute appendicitis was confirmed in (17) patients (false-ve) comprised of (5) adult male, (6) adult female & (6) children giving a specificity rate of (91.6%), (89.6%) (75%) respectively. **The overall sensitivity rate & specificity rate of Modified Alvarado Score were (80.9%) & (88.5%) respectively. The validity of Modified Alvarado Score for the (150) patients with provisional diagnosis of acute appendicitis.** All patients with score of (7) or more ((79) patients) underwent appendicectomy. Acute appendicitis was confirmed in (72) patients (true +ve), among them (40) patients were adult male out of (42) giving sensitivity rate of (88.8%), (25) adult female out of (28) giving sensitivity rate of (80.6%) & (7) children out of (9) giving sensitivity rate of (53.8). All patients with score of (5- 6) (71 patients) were admitted for observation & re evaluation. Abdominal ultrasonography was done for (12) patients leading to the diagnosis of (2) right ovarian cysts & (2) ureteric stones, & they were referred to the gynaecological & urological departments respectively following clinical improvement. Another (41) patients with score of (5- 6) were discharged home as clinical improvement & decreased score was shown in subsequent reevaluation, & were instructed to come back if the symptoms persist or increase in intensity, while appendicectomy was performed in (26) patients with score of (5-6), they were (9) adult males, (10) adult females & (7) children. Acute appendicitis was confirmed in (17) patients (false-ve) comprised of (5) adult male, (6) adult female & (6) children giving a specificity rate of (91.6%), (89.6%) (75%) respectively. Another (41) patients with score of (5- 6) were discharged home as clinical improvement & decreased score was shown in subsequent reevaluation, & were instructed to come back if the symptoms persist or increase in intensity, while appendicectomy was performed in (26) patients with score of (5-6), they were (9) adult males, (10) adult females & (7) children. Acute appendicitis was confirmed in (17) patients (false-ve) comprised of (5) adult male, (6) adult female & (6) children giving a specificity rate of (91.6%), (89.6%) (75%) respectively The overall sensitivity rate & specificity rate of Modified Alvarado Score were (80.9%) & (88.5%) respectively The total number of patients who underwent appendicectomy was (105) patients (70%) out of (150), among them acute appendicitis was confirmed in (89) patients (84.8) comprised of (72) patients with score of > 7 (true +ve) & (17) patients with score of < 7 (false -ve), among them (8) had perforated appendices, (10) patients had gangrenous appendices & (4) had appendicular mass, there was no operative findings of perforated, gangrenous appendices or appendicular mass in those patients with score <7. While normal appendix was found in (16) patients, (7) patients of them with score of > 7 (false +ve) comprised of (2) adult male, (3) adult female & (2) children. However, other surgical corrected pathology was found in (1) adult male with Meckel's diverticulitis & (2) adult female one with ruptured right ovarian cysts & the other with ruptured right tubal pregnancy. While in the remaining (4) patients no obvious other surgical pathology was identified. The other (9) patients with normal appendices were with score of < 7 (true -ve) comprised of (4) adult male, (4) adult female & (1) child, & other surgically corrected pathology was identified in (2) adult female only, one with ruptured right ovarian cyst & one with right ovarian endometriotic (chocolate) cyst, while in the remaining (7) patients no obvious surgically corrected pathology was identified. So the total (-ve) appendicectomy rate was (15.2%) & the adult male, adult female & children (-ve) appendicectomy rates were (11.7%), (18.4%) & (18.7%) respectively. The positive predictive value of the Modified Alvarado score was 91.1% & for adult males, adult females & children were 95.2%, 89.3% & 77.8% respectively.

Patients distribution with score > 7

Patients	Total no.	Score more 7	%
Adult male	69	42	53.2
Adult female	60	28	35.4
Children	21	9	11.4
Total	150	79	100

Specificity of Modified Alvarado Score

Patients	No.	Score	Operated	Appendicitis	Specificity
Male	69	27	9	5	91.6
Female	60	32	10	6	89.6
Children	21	12	7	6	75
Total	150	71	26	17	88.5

The sensitivity of Modified Alvarado Score

	No. of patients	Score	Appendicitis	Sensitivity
Male	69	42	40	88.8
Female	60	28	25	80.6
Children	21	9	7	53.8
Total	150	79	72	80.9

Specificity of Modified Alvarado Score

Patients	No.	Score	Operated	Appendicitis	Specificity
Male	69	27	9	5	91.6
Female	60	32	10	6	89.6
Children	21	12	7	6	75
Total	150	71	26	17	88.5

Results of application of Modified Alvarado Score

Item	Total No.	Score more 7	Score <7
Total patients	150	79	71
No surgery	45	0	45
Surgery	105	79	26
Acute appendicitis	89	72	17
Normal appendix	16	7	9

The results of operative & histopathological

Finding	No. of patients %	
Inflamed acute appendicitis		
Acute appendicitis	67	63.8
Perforated appendix	8	7.6
Gangrenous appendix	10	9.6
Appendicular mass	4	3.8
Normal appendix		
Right ovarian cyst	2	1.9
Meckel's diverticulitis	1	0.9
Ruptured tubal pregnancy	1	0.9
Endometriotic cyst	1	0.9
No pathology	11	10.6
Total operated on	105	100

Discussion:-

Acute appendicitis is undoubtedly, the most common surgical emergency, & the diagnosis of acute appendicitis is essentially & largely made on clinical grounds, but even then different possibilities are there. The prevalent age group in our study was from (11) to (30) years (71.4%) & the lowest rate were encountered in children & the elderly. These results are comparable with the results of a study done in Nineveh, Iraq, 1998.⁽¹²⁾ & also with a study done in Pakistan at 2003.⁽¹³⁾ The frequency distribution of the patients included in our study according to Modified Alvarado Score demonstrate that the highest frequency was encountered with those who scored (6) (40 patients, 26.7%) & the lowest frequency was in those scored (9) (20 patients, 13.3 %). However, (79) patients (52.6%) of the total (150) patients included in this study were with score of (7) or more & (71) patients (47.4%) were with score of less than (7) (5-6). These results are comparable to the results shown by a study done in Nineveh, Iraq, 1998.⁽¹²⁾ & also with the results of a study done in Pakistan at 2003.⁽¹³⁾

Initial assessment of the patients can be improved by clinical scoring system.^(14,15) & in the past few years various scores have been developed to aid the diagnosis of acute appendicitis.^(16,17,18) However, most of them are complex & difficult to implement in clinical situation.^(19,20,21) Our findings demonstrate that Modified Alvarado Score carries results which may vary according to the age group (adult versus children) & gender. For adult male patients with suspected acute appendicitis, the Modified Alvarado Score gives a sensitivity rate of (88.9%) while in adult female & especially those at child bearing age Modified Alvarado Score gives a sensitivity rate of (80.6 %) with a false (+ve) rate of (10.7). These findings are similar to the results of a study done in Nineveh, Iraq, 1998 & also similar to the conclusions of a study done by Owen *et al.*⁽¹²⁾ However, they are not similar to the results of a study done in Southern Region, Kingdom of Saudi Arabia (sensitivity male 56.4%, sensitivity female 48%). Our study also revealed that Modified Alvarado Score is more helpful in adult male than adult female patients by showing lower (-ve) appendectomy rate & higher (+ve) predictive value in adult male as compared to adult female patients, as the (-ve) appendectomy rate for adult male were (11.7%) while, in adult female it was (18.4%) & the (+ve) predictive value were (95.2%) & (89.3%) for adult male & female respectively. These results are comparable to a study done in Pakistan, 2003, also these results are supported by literature figures.^(23,24,25) Literatures show that if (-ve) appendectomy rate is less than (10- 15%), then the surgeon is operating too few patients thus increasing the risk of complications.^(26,27) In children with acute appendicitis the sensitivity rate of Modified Alvarado Score in our study was very low (53.8%) & its specificity rate was (75%) with a false (+ve) rate of more than (22%), & to be useful, a scoring system must be both sensitive & specific. These findings are comparable to the results of a study done in Nineveh, Iraq, 1998 & is also similar to the results of a study done by Macklin CP.⁽²⁰⁾ The (+ve) predictive value for the children included in our study was (77.8%). Studies evaluating the usefulness of Alvarado scoring system in paediatric age group show that it is accurate in children with (+ve) predictive value up to (85.7%).^(28,29,30) The overall sensitivity & specificity of our study were (80.9%) & (88.5%) respectively. These results are comparable to the results of a study done in Nineveh, Iraq, 1998 & is also comparable to the results of a study done by Fenyo G, but our results are not similar to the results of a study done in Southern Region, Kingdom Saudi Arabia. Our study revealed a total negative appendectomy rate of (15.2%) with a (-ve) appendectomy rate of the adult male, adult female & children of (11.7%), (18.4%), & (18.7%) respectively. These results are comparable to the results of a study done in Pakistan at 2003, & are also similar to the results of a study done by Fenyo.⁽³¹⁾ & is comparable to the figures shown in literature (15.62%).^(32,33) Our study showed a low incidence of perforation (7.6%) which can be explained by the fact that perforation in acute appendicitis is clearly more common in children & elderly people.^(34,35) The percentage of both of these groups is low in our study, as well as, the more accurate diagnosis depending on the application of Modified Alvarado Score. These results are comparable to the results of a study done in Nineveh, Iraq, 1998 & is also comparable to the results of a study done in Pakistan at 2003.

Conclusions & Recommendation:-

1. Modified Alvarado Score is a non invasive, safe diagnostic procedure which is simple, fast, reliable & repeatable tool to apply in emergency departments to role in acute appendicitis. It allows observation & critical reevaluation of the evolution of the clinical picture. We suggest its application in order to increase the diagnostic certainty of clinical examination in diagnosis of acute appendicitis especially by junior surgeons.
2. Modified Alvarado Score appeared to be effective in adult male patients, while in adult female patients specially those in child bearing age, additional investigations may be required, so we suggest the provision of (24) hours ultrasound facility to confirm or exclude the possibility of other pathologies in doubtful cases. In pediatric age group it was not successful enough to be useful complementary method in the diagnosis of suspected cases & because our study included only a small number of children we suggest a prospective study

to be conducted purely on patients within pediatric age group to assess precisely the validity of the Modified Alvarado Score in these cases.

3. Our study demonstrates comparable results of negative appendectomy rate with that of literature figures, which signifies that the application of Modified Alvarado Score improves the diagnostic accuracy & consequently reduces the negative exploration & complication rates.

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RESEARCH ARTICLE

EVALUATION OF T- REGULATORY CELLS IN BREAST CANCER.

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Abstract

Treg cells have an important role in cancer, enabling tumors to elude host anti-tumor immune response. CD4+CD25+ Treg cells constitute 5–10% of peripheral blood CD4+ lymphocytes. Also, CD8+CD25+ cells have been shown a suppressive effect and share phenotypic and functional features with CD4+CD25+ Treg cells.

Aim of the work: This work was done to evaluate the frequency of expression of different Treg cells in the peripheral blood of breast cancer patients and to elucidate their impact on clinical and laboratory parameters.

Patients and Methods: This study included forty-seven Egyptian breast cancer patients (30 de novo patients and 17 after surgery patients) and 13 healthy controls and 18 benign breast patients. They were tested by Flow Cytometry

Results: CD4+CD25+ Treg cells were significantly higher in the de novo compared to the healthy controls and in the after-surgery group compared to the healthy controls and benign breast groups ($P < 0.0001$), while CD4+CD25hi were significantly higher in both de novo and after surgery patients as compared to healthy controls ($P < 0.0001$).

Conclusion: some Treg cells as CD4+CD25+ cells are predominant in breast cancer patients.

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Introduction: -

Breast cancer is the most frequently diagnosed cancer and the leading cause of cancer deaths among females worldwide, accounting for 23% of the total new cancer cases and 14% of the total cancer deaths in 2008 (Jamal et al. 2011). Breast cancer patients have immune system functional deviations, that affect immune cells that infiltrate the tumor, but also cells from the blood compartment, even in the absence of a metastatic disease (Pinho et al., 2016).

Regulatory T-cells (Treg cells) are a variety of cells that display the capacity to suppress T-cell proliferation in vitro and prevent immune pathology in vivo. Treg cells have an important role in cancer, enabling tumors to elude host anti-tumor immune response (Maggiet al., 2005).

CD4+CD25+ Treg cells constitute 5–10% of peripheral blood CD4+ lymphocytes (Maggiet al., 2005). They express CD25 (the alpha chain of the IL-2 receptor), cytotoxic T lymphocyte-associated antigen-4 (CTLA-4), a glucocorticoid induced TNF receptor (GITR), and a novel member of the fork head transcription factors (Foxp3) (Zhang et al., 2007).

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CD8+CD25+ cells have been recently shown a suppressive effect. CD8+CD25+Treg cells share phenotypic and functional features with CD4+CD25+ Treg cells (Cosmi et al., 2003).

The aim of this study is to evaluate the frequency of expression of different Treg cells in the peripheral blood of Egyptian breast cancer patients and compare their expression with healthy controls and benign breast patients. In addition, to study the relationship between different Treg cells and clinical and laboratory findings and response to treatment in such patients.

Patients and Methods: -

Forty-seven breast cancer patients, 30 de novo patients (before surgery) and 17 after surgery patients, 13 benign breast disease patients, and 18 apparently healthy volunteering females as a control group were included in this study. The patients were presented to the outpatient's clinic at the National Cancer Institute (NCI), Cairo University over a period of consecutive 11 months. Malignancy was confirmed by histopathological examination. Written informed consent was obtained before enrollment into the study according to ethical committee. Investigations performed included routine hematological and biochemical investigations, and CEA and CA15.3 evaluations.

Methods: -

Flow cytometric analysis of Treg cells: -

Isolation of mononuclear cells and cell culture: -

Three to four ml of heparinized blood from each sample was added to 15 ml centrifuge tube and diluted 1:1 with sterile PBS without Magnesium or Calcium salts (*Biowest, France catalog no: L0615*) under sterile conditions. Ficoll-Hypaque media of density 1.077 gm/ml (*Lymphosep, Lymphocyte Separation Media- Biowest, France Catalog no: L0560*) was used to separate the PBMCs. The mononuclear cells were washed and re-suspended under sterile conditions in 3 ml RPMI 1640 with L-Glutamine medium (*Biowest, France catalog no: L050*) containing 10% Fetal Bovine Serum (*Biowest, South America catalog no: S1810*) and 2 % Antibiotic-Antimycotic 100x (*Biowest, France catalog no: L0010*) which contains Penicillin G, Streptomycin Sulfate & Amphotericin B. Two ng/ml IL2 (*Sigma, catalog no: 151342*) were added for T cell activation. The cells were incubated at 37°C, 5% CO₂ for 48 hours.

Monoclonal Antibodies: -

Fluorescent labeled mouse monoclonal antibodies anti-Human CD4-FITC (*Catalog no: F0766, Clone MT310*), CD8-PE (*Catalog no: R0806, Clone DK25*), CD25-FITC (*Catalog no: F0801, Clone ACT-1*), CD25-PE (*Catalog no: R0811, Clone ACT-1*) and CD3-RPE-Cy5 (*Catalog no: C7067, Clone UCHT1*) were obtained from DakoCytomation, Denmark.

Flowcytometric analysis: -

CD4+CD25+ Treg cells were studied with triple staining of anti-CD4 FITC, anti-CD25 PE and anti-CD3 RPE-Cy5, and CD8+CD25+ Treg cells were studied with anti-CD25 FITC, anti-CD8 PE and anti-CD3 RPE-Cy5 using COULTER® EPICS® XL™ Flow Cytometer instrument. To determine the percentage of CD4+Treg cells, lymphocytes were gated by plotting forward versus side scatter followed by gating on CD3+CD4+ T cells. These cells were then analyzed for CD25 expression; CD4+CD25hi were also identified as a tail from the major population of CD4+CD25 cells (Baumgartner and McCarter, 2009). CD8+ Treg cells were detected as percentage of CD8+CD25+ population gated from CD3+ lymphocytes (Fig 1).

Statistical analysis: -

Data management and analysis were performed using the Statistical Analysis System (SPSS) version 17 software. Comparison of groups with respect to numerical variables was done using the Mann-Whitney test. The Kruskal Wallis test was used to compare medians of 3 or more independent groups. Spearman's rank correlation was used to measure the strength of association between 2 numerical variables. All p-values were two sided, P-values ≤ 0.05 were considered significant.

Results: -

Characteristics of the de novo, after surgery & benign patients' groups are shown in (Table 1.a & 1.b).

CD4+ lymphocytes % were higher in normal controls and de novo patients compared to the after-surgery patients (P=0.04) (Table 2).

CD4+CD25+ Treg cells were significantly higher in the de novo compared to the healthy controls and in the after-surgery group compared to the healthy controls and benign breast groups ($P < 0.0001$), while CD4+CD25hi were significantly higher in both de novo and after surgery patients as compared to healthy controls ($P < 0.0001$) (Table 2).

A significant positive correlation was detected between CD4+CD25+ and CD4+CD25hi Treg cells in both de novo and benign patients' groups, while a significant negative correlation was detected between CD4+CD25 and CD8+CD25+ Treg cells in the after-surgery group (Table 3).

CD4+CD25+ Treg cells showed significant negative correlation with age in both de novo and after surgery groups ($P = 0.038$, $r = -0.388$ & $P = 0.012$, $r = -0.612$ respectively), and significant positive correlation with CEA in the benign group ($P = 0.005$, $r = 0.722$). CD8+CD25+ Treg cells showed significant positive correlation with age in the after-surgery group ($P = 0.048$, $r = 0.487$) (Table 4).

CD4+CD25+ and CD4+CD25hi Treg cells were higher in premenopausal compared to postmenopausal de novo breast cancer patients ($P = 0.017$ and 0.051 respectively). CD4+CD25hi Treg cells were significantly higher in the non-metastatic compared to the metastatic patients in the de novo group ($P = 0.041$), while CD4+CD25hi Treg cells were higher in grade III compared to grade II patients in the after-surgery group ($P = 0.011$) (Data not shown)

Discussion: -

In Egypt, breast cancer ranked the first, constituting 17.50% of total malignancies in females (Meloni et al., 2006).

Although T cells represent the most important immunological response in tumor growth, they become suppressive CD4⁺ and CD8⁺ Treg cells after chronic stimulation and interactions with tumor cells, thus promoting rather than inhibiting cancer development and progression (Mokhtar et al., 2007).

In this study, CD4+ lymphocytes were predominant in peripheral blood of de novo breast cancer patients and healthy controls groups compared to the after-surgery group ($P = 0.04$).

Mozaffari et al. (2007) reported the same predominance of absolute number of CD4+ lymphocytes although it was statistically non-significant. Leong et al. (2006) showed that CD4+ lymphocyte % was the 2nd predominant subset (after CD8+) in the tumor microenvironment. Different sample analyses and different sample sizes may be the contributing factors for this discrepancy.

Tumor cells commonly participate in the generation of Treg cells, which provides an explanation for the observation that elevated numbers of Treg cells have been found in many types of cancer. It appears that TGF- β , secreted by the tumor itself or tumor-stimulated myeloid cells, plays a central role in tumor-mediated development of Treg cells by converting naïve T cells into Treg cells (Perez et al., 2007).

As regards CD4+CD25+ and CD4+CD25hi, they were significantly higher in the de novo and the after-surgery cancer patients compared to the healthy controls and the benign patients' group.

Our result is consistent with Wang et al. (2011) who found the same result in their study on peripheral blood of de novo breast cancer patients.

In our study, CD4+CD25hi subset was higher in the after-surgery patients compared to the de novo patients ($P = 0.04$) and this was in accordance with the finding of Mozaffari et al (2007).

CD8+CD25+ Treg cells share phenotypic and functional features with CD4+CD25+ Treg cells (Cosmi et al., 2006). We detected that the benign patients group had the highest percentage of CD8+CD25+ Treg cells subset, followed by the after-surgery group. The de novo breast cancer patients and healthy controls group were almost the same, the differences were non-significant. Mozaffari et al. (2007) reported a significant increase in CD8+CD25hi Treg cells in breast cancer patients after receiving adjuvant therapy compared to the de novo patients. Consistently, Shao et al. (2017) reported that the proportion of regulatory T cells (Treg) after chemotherapy in the disease control patients decreased significantly compared with that of the progressive patients and this difference affected

significantly the overall survival. Pinho et al (2016) found that breast cancer patients have higher percentage of effector CD8+ T cells compared to healthy donors.

T-reg cells can offer a new insight into improving cancer therapies. Combined chemotherapeutic and immunomodulatory agents that showed their effect on T-reg can lead to marked suppression of tumor growth and prolongation of survival time (Xia et al., 2017 and Hekim et al., 2017).

A significant positive correlation was detected between the CD4+CD25+ % and the CD4+CD25hi % in the de novo breast cancer patients group ($P < 0.0001$, $r = 0.673$) and in the benign group ($P = 0.002$, $r = 0.79$), while there was a significant negative correlation between the CD4+CD25+ % and CD8+CD25+ % in the after-surgery patients group ($P = 0.018$, $r = -0.583$).

In agreement with our results, Zhu et al. (2015) and Leong et al. (2006) have demonstrated an insignificant negative correlation between the CD4+CD25+ cells and CD8+CD28+CD25+ obtained from TILs of breast cancer patients. They also found a significant positive correlation between CD4+CD25+ and CD8+CD28- subsets. In that study, CD8+CD28+CD25+ were defined as effector T cells and CD8+CD28- subset as Treg cells.

In our study, the observed negative relationship may be explained by the finding that not all CD8 + CD25+ population are Treg cells (Bisikirska et al., 2005).

Correlating CD4+CD25+ % and CD8+CD25+% with some of the prognostic factors of breast cancer revealed a significant negative correlation between the CD4+CD25+ % and the age in the de novo and after surgery groups. However, a significant positive correlation was found between CD8+CD25+% and age in the after-surgery group.

Similarly, Chui et al. (2004) reported that CD4+ Treg cells were higher in the blood of breast cancer patients < 65 years in age. Tsaknaridis et al. (2003) also demonstrated that the suppressive activity of human CD4+CD25+ T cells declines with age in his study done on normal subjects. In contrast, Leong et al. (2006) found a significant positive correlation with age (> 50 years old).

This diversity of results may be due to differences in sample size or ethnic diversity.

Regarding tumor markers, a significant positive correlation was detected in the benign patients between CD4+CD25+ % and CEA ($P = 0.005$, $r = 0.722$), while CA15.3 didn't show any significant results. In contrast, Abo-Elainen et al. (2008) reported a significant positive correlation between CD4+CD25+% in the peripheral blood of 30 female breast cancer patients and CA15.3 ($P = 0.880$, $r = 0.001$). This may be explained by the different stages of breast cancer patients enrolled in both studies.

CD4+CD25hi % was significantly higher in the non-metastatic group compared to the metastatic group in de novo patients ($P = 0.041$). Many studies have demonstrated that a higher Treg cells number within the tumor microenvironment or in peripheral blood would imply a worse prognosis and reduced survival (Wilke et al., 2010).

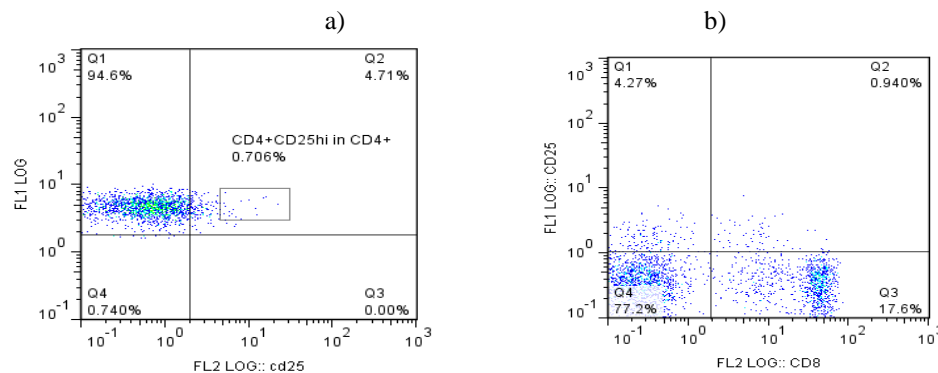
Significant higher percentage of CD4+CD25hi in the after-surgery group was detected in grade III compared to grade II ($P = 0.011$). This was inconsistent with Leong et al. (2006).

We didn't find any significant relationship between CD4+CD25+ % or CD4+CD25hi % and the tumor stage, ER, PR or Her2neu status in both the de novo and the after-surgery patients groups. Our results are consistent with Song et al (2015) but different from Perez et al. (2007). Also, Mahmoud et al. (2011) and Bates et al. (2006) found significant correlations between Treg cells infiltration and ER negativity in breast cancer patients. Liu et al (2014) found that CD4+CD25+FOXP3+ regulatory TILs are a poor prognostic indicator in ER+ breast cancer, but a favorable prognostic factor in the HER2+/ER-ve subtype. Leong et al. (2006) found a significant positive correlation between CD4+CD25- T subset of TILs in breast cancer patients and ER. Estrogen has been reported to play a role in regulating the activation of human T cells particularly CD4+ and CD8+ T cells. The importance of ER- α in regulating the immune system is further supported by an in vivo murine model, where a high frequency of immature double CD4+CD8+ thymocytes was found in ER- α -negative mice as compared to the ER- α positive mice (Erlandsson et al., 2001).

This discrepancy between results may be attributed to the predominance of ER positivity in our studied population, different ethnic groups or menopausal status of the studied population.

In conclusion, some Treg cells as CD4+CD25+ cells are predominant in breast cancer patients, and show negative correlation with age and with CD8+CD25+ lymphocytes, which are predominant in the benign patients and after surgery patients.

Treg cells may play a role in modulation of effector T cell responses against breast tumors. Some Treg cells such as CD4+CD25+ cells maybe increased in cases of malignancy, while others, as CD8+CD25+ lymphocytes seem to play a protective role against malignancy. Further studies and larger sample sizes are needed to clarify the diagnostic and prognostic role of Treg cells in breast cancer progression. Treg can act as an immune-modulator in the therapeutic treatment of breast cancer patients weather alone or in combination with therapeutic vaccines and cytokines.



(Fig 1) a):- Positive expression of CD4+CD25+ and CD4+CD25hi subsets gated from CD4+ lymphocytes in a de novo patient. b) CD8+CD25+ subsets histogram from CD3+ lymphocytes gate in an after-surgery patient.

Table (1.a):- Clinical data of the de novo, after surgery and benign patients.

		De novo patients		After-surgery patients		Benign patients		Total	
		n = 30	Percentage %	n= 17	Percentage %	n= 13	Percentage %	n= 60	Percentage %
Age	< 50 years	14	47	7	41	7	54	28	47
	≥ 50 years	16	53	10	59	6	46	32	53
Family history	Positive	4	13	0	0	0	0	4	7
	Negative	26	87	17	100	13	100	56	93
Marital status	Married	29	97	17	100	9	69	55	92
	Single	1	3	0	0	4	31	5	8
Menarche status	Pre-menopause	12	40	7	41	9	69	28	47
	Post-menopause	18	60	10	59	4	31	32	53

n: number of patients.

Table (1.b):- Clinical and pathological data of the de novo and after surgery patients.

		De novo patients		After surgery patients		Total	
		n = 30	Percentage	n= 17	Percentage	n= 47	Percentage
Tumor size	Tumor size \leq 5 cm	24	80	12	71	36	77
	Tumor size > 5 cm	6	20	5	29	11	23
Lymph node status	Positive	29	97	13	76	42	89
	Negative	1	3	4	24	5	11
Distant metastasis	Present	14	47	5	29	19	40
	Absent	16	53	12	71	28	60
Tumor stage *	Stage I	1	3	0	0	1	2
	Stage II	9	30	7	41	16	34
	Stage III	5	17	4	24	9	19
	Stage IV	15	50	6	35	21	45
Grade	II	26	87	12	71	38	81
	III	4	13	5	29	9	19
Tumor focality	Unifocal	26	87	14	82	40	85
	Multifocal	4	13	3	18	7	15
ER	Positive	24	80	10	59	34	72
	Negative	6	20	7	41	13	28
PR	Positive	11	37	14	82	25	53
	Negative	19	63	3	18	22	47
HER2/neu	Positive	4	13	7	41	11	23
	Negative	26	87	10	59	36	77

* Stage according to TNM classification for breast cancer (AJCC).

n: number of patients, **Grade:** Pathological grade, **ER:** Estrogen receptor, **PR:** Progesterone receptor, **HER2/neu:** Human Epidermal growth factor Receptor 2.

Table (2):- Comparison of different lymphocyte populations in the different studied groups (de novo, after surgery, benign patients and healthy controls) using Krusal-Wallis test:

	Healthy controls	De novo patients	After surgery patients	Benign patients	P-value
	Median & IR (Q1 – Q3)	Median & IR (Q1 – Q3)	Median & IR (Q1 – Q3)	Median & IR (Q1 – Q3)	
Total lymphocytes %	84.7 (82 – 87.8)	88.65 (86 – 92.3)	89.2 (81 – 91)	90 (88 – 91)	0.15
CD3+ lymphocytes %	76.95 (67.6 – 88.1)	69.8 (58.2 – 85.1)	67.15 (47.8 – 82.6)	77.8 (64.6 – 85.4)	0.27
CD4+ lymphocytes %	65.25 a (61.9 – 72.4)	65.6 b (57.5 – 75)	55.7 a,b (46.5 – 67)	52.7 (51 – 71.9)	0.04
CD8+ lymphocytes %	17 (12.1 – 20.5)	16.5 (13.5 – 20.9)	19.7 (12.7 – 33.7)	16.6 (9.4 – 20.9)	0.57
CD4+/CD8+ ratio	3.57 (3.4 – 5.9)	3.79 (3.3 – 4.6)	2.83 (1.45 – 4.1)	3.46 (3.2 – 5.1)	0.13
CD4+CD25+ %	2.1 c,d (1.7 – 2.5)	3.78 c (3.1 – 4.6)	4.46 d,e (3.95 – 4.9)	3.11 e (1.9 – 3.6)	<0.0001
CD4+CD25hi %	0.27 f,g (0.15 – 0.35)	0.7 f (0.47 – 0.85)	0.8 g (0.7 – 1.1)	0.43 (0.22 – 0.57)	<0.0001
CD8+CD25+ %	0.81 (0.54 – 0.85)	0.89 (0.5 – 1.12)	0.94 (0.7 – 1.2)	0.96 (0.56 – 1.7)	0.24

IR: Interquartile range

* Groups with the same letter show statistically significant difference

Table (3):- Correlation analysis between the different Treg cells subsets % in the different patients' groups

Table (b7) Correlation analysis between the different Treg cells subsets % in the different patients' groups				
			CD4+CD25hi %	CD8+CD25+ %
De novo patients	CD4+CD25+ %	r	0.673	-0.159
		P	< 0.0001**	0.438
	CD4+CD25hi %	r		-0.229
		P		0.261
After surgery patients	CD4+CD25+ %	r	0.33	-0.583
		P	0.212	0.018*
	CD4+CD25hi %	r		-0.007
		P		0.978
Benign patients	CD4+CD25+ %	r	0.79	-0.116
		P	0.002**	0.721
	CD4+CD25hi %	r		0.068
		P		0.842
* Significant.				
**Highly significant				

Table (4):- Correlation analysis between different Treg cells subsets % with the clinical, histopathological and laboratory data in the different patients' groups (de novo, after surgery and benign patients):

			CD4+CD25+ %	CD4+CD25hi %	CD8+CD25+ %
Age	De novo	r	-0.388	-0.214	0.119
		P	0.038*	0.266	0.554
	After surgery	r	-0.612	0.267	0.487
		P	0.012*	0.301	0.048*
	Benign	r	0.29	0.035	0.248
		P	0.336	0.914	0.437
Tumor size	De novo	r	-0.075	-0.201	0.21
		P	0.701	0.296	0.294
	After surgery	r	0.025	-0.051	0.467
		P	0.926	0.846	0.058
CEA	De novo	r	-0.279	0.034	-0.131
		P	0.143	0.859	0.515
	After surgery	r	0.148	0.221	0.108
		P	0.584	0.393	0.679
	Benign	r	0.722	0.493	0.155
		P	0.005**	0.103	0.631
CA 15-3	De novo	r	-0.025	-0.065	-0.002
		P	0.896	0.737	0.993
	After surgery	r	-0.091	-0.422	0.174
		P	0.737	0.092	0.504
	Benign	r	0.544	0.371	-0.315
		P	0.055	0.236	0.318

* Significant. ** Highly significant.

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RESEARCH ARTICLE

ISOLATION AND CHARACTERIZATION OF FUNGI FROM THE FRUIT OF ORANGE AND TOMATO IN JIMMA TOWN MARKET SELLERS, SOUTH WEST ETHIOPIA.

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C. sinensis, *L. esculentum*, *Penicillium* spp., *Aspergillus* spp., Spoilage.

Abstract

The study was carried out from February, 2013 to May, 2013 in Jimma to assess the common spoilage fungi from infected fruits of tomato (*Lycopersicon esculentum*) and orange (*Camellia sinensis*) in Jimma town (Agip, Kochi and Bishishe) market sellers. Mycological investigation on spoilage fungi from 40 (20 from each) apparently infected *C. sinensis* and *L. esculentum* was carried out. Potato Dextrose Agar was used for fungi isolation and purification following standard procedures. Pure cultures were identified morphologically to the genus level. A total of 56 spoilage moulds were isolated from the two different types of deteriorating fruit samples, from which 52 (92.85 %) isolates were identified and the remaining 4 (7.15 %) were unidentified. The fungal species *Aspergillus* spp., (13.46) *Penicillium* spp., (19.2) *Rhizopus* spp., (1.92) *Mucor* spp., (5.76) *Fusarium* spp., (3.84), *Byssoschlamy* spp. (3.84) and *Cladosporium* spp. (1.92) were found to be associated with deterioration of orange. The fungal isolates associated with the spoilage of tomato were *Aspergillus* spp., (7.69) *Penicillium* spp., (9.61) *Rhizopus* spp., (7.69) *Mucor* spp., (9.61) *Fusarium*, (9.61) and *Monilia* spp. (5.76). Generally, of all isolated moulds *Penicillium* spp. was the dominant isolate 15 (28.81%), followed by *Aspergillus* spp. 11 (21.15). *Byssoschlamy* spp. And *Cladosporium* spp. were the least encountered 2 (3.84%) and 1 (1.92%), respectively. The results of this study indicate that fruits sold at Agip, Kochi and Bishishe were massively infected with spoilage fungi due to lack of selling parameters of fruits found in Jimma town market sellers. So, appropriate measurement must be taken by responsible bodies to reduce the fungal load and to enhance the quality of fruits sold in Jimma town.

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Introduction:-

Fruits and vegetables are very important and have high dietary and nutritional quality. Fruits play a vital role in human nutrition by supplying the essential growth factors such as vitamins and essential minerals to the regular diet, which is necessary for the good and normal health. Consumption of fruit and vegetable products has dramatically increased by more than 30% during the past few decades (Barth et al., 2009). Fresh fruit and vegetable consumption increased by 25.8 and 32.6% respectively and far exceeded the increases observed for processed fruit and vegetable products. Fruits and vegetables are a major source of macronutrients such as fiber and micronutrients such as

minerals and vitamin C, thiamin, riboflavin, B-6 niacin, folate, A and E (Rickman et al., 2007). The fruits of tomato are popular throughout the world and are used in all kind of stews, soups and also eaten raw in salads. Ripe tomato fruits have high nutritive values, being a good source of vitamin A, B, C and minerals (Elsayed and Edrees, 2014). According to Wada and Ou (2002) fruits and vegetables are well known for their antioxidants compounds that protect against oxidative damage caused by free radicals, and they have been shown to be effective in helping to prevent retinal disease such as muscular degeneration. And also tomato plant contain antioxidant phytochemicals such as the carotene and lycopene (Wogu and Ofuase, 2014; Sravanthi and Gangadhar, 2015).

Traditional varieties of fruits like avocado, banana, orange and papaya are affected by a wide array of microorganisms. Spoilage refers to any change in the condition of food in which the food becomes less palatable, or even toxic: these changes may be accompanied by alteration in taste, smell, appearance or texture (Akinmusire, 2011). One of the limiting factors that influence health and economic values of the fruit is the relatively short lifespan. Fruits deteriorate rapidly after harvest and in some cases do not reach consumers at optimal quality after transport and marketing. The main causes of fruit deterioration are dehydration, weight loss, color change and microbial spoilage. However, deterioration rate is affected by different factors like temperature, relative Humidity and storage atmosphere condition (Sherratt et al., 2006).

Fungal fruits infection may occur during the growing season, harvesting, transport and post-harvest storage and marketing transport and post-harvest storage and marketing conditions, or after purchased by the consumer. The important post-harvest disease causing fungi include *penicillium*, *Aspergillus*, *Alteranaria species*, *Botrytis cinerea*, *Molniliinia lax* and *Rhizopus stolonifer* (Ogawa et al., 1995). Fruits particularly tomatoes and orange are eaten raw and tomatoes are also eaten cooked. Spoilage fungi that typically produce more diverse and greater amounts of extracellular depolymerases successfully attack both fruits (Barth et al., 2009). And also an abundance of extracellular pectinases and hemicelluloses produced by the fruits are important factors for fungal spoilage (Miedes and Lorences, 2004).

Although available literatures revealed that the importance of fruit is increasing daily, the incidence of microbial attack on this fruit demands attention. Janisiewicz and Korsten (2002) also remarked the exploration of plant extracts have shown to be great potential and may be used as alternative to synthetic fungicides. The purpose of the current investigation was to isolate and characterize some fruit spoilage fungi.

Materials and Methods:-

Description of study area:-

This research was conducted in Jimma town, which is the largest city in Southwestern Ethiopia. Jimma town is located 353km southwest of the capital city of Ethiopia, Addis Ababa. And the city is located between 1,500 - 2,400 m above sea level and the total population was about 120, 969 in 2007. It lies in a climatic zone locally known as Woyna degas which is ideal for agriculture as well as human settlement. The town is characterized by warm climate with a mean annual maximum temperature of 30°C and minimum of 14°C. The laboratory based study was conducted at microbiological Laboratory of Department of Biology, Jimma University during the period from February, 2013 to May, 2013.

Fruit source:-

Fruits of orange and tomato which were found with symptoms of fungal infection were purchased from Agip, Kochi and Bishishe market sellers in Jimma town.

Sample size determination:-

Purposive sampling technique was used to determine the sample size and assess the common fungal pathogens from infected orange and tomato, from Jimma town market sellers. A total of 40 samples comprising 20 samples from each fruit were collected randomly from Agip, Kochi and Bishishe markets. Ten of the samples (5 orange and 5 tomato) were collected from Agip market, the other 10 samples consisting 5 samples from each fruit were collected from Kochi market and the rest 20 samples comprising 10 samples from each fruit were collected from Bishishe market this is because Bishishe Market in Jimma town serves as the main depot where the major fruit dealers and suppliers sell to petty traders who in turn distribute to other local markets (Agip and Kochi) and cafeterias throughout the town. The fruits were sourced from areas like Mizan, Tepi, Addis Ababa and neighboring woredas within the Jimma zone itself. All of the 40 fruit samples were purchased from different vendors, so 40 fruit vendors were visited to collect all the samples.

Collection of infected fruit:-

Fruit types of orange and tomato with the common post-harvest disease symptoms were collected using sterile polyethylene bag from Agip, Kochi and Bishishemarkets in Jimma town. A total of 40 samples comprising 20 samples from each fruit were collected randomly from the venders and it was transported aseptically to the laboratory of microbiology, department of Biology, Jimma University for further analysis.

Isolation of fungi:-

Potato dextrose agar medium was prepared using distilled water and it was sterilized within an autoclave. The petridishes were also sterilized to kill any microbial contamination. In order to suppress bacterial growth, chloramphenicol of 35-100mg 100ml⁻¹ of PDA was poured on the already sterilized petridishes and it was left open for a few minute inside a laminar flow to be solidify, then the petridishes were closed to prevent other contamination. Fruits of orange and tomato were surface sterilized by exposing them in 90% ethyl alcohol for 1 min and 3 min to 1% sodium hypochlorite and then, it was rinsed three times in sterile distilled water. Segments (3-5cm), which cut with a sterilized scalpel was placed on previously prepared potato dextrose agar (PDA in case of both orange and tomato) in petridishes and they were incubated at 21°C for 5 to 7 days. After incubation, the colonies were observed. To get a pure culture, each of the emerging mycelium was transferred to fresh solid medium. The pure culture of each colony on different plates were obtained and they were prepared for characterization

Characterization of the isolates:-

The pure isolated fungi were identified using cultural and morphological features according to the most documented keys in fungal identification (Domsch *et al.*, 1993; Klich, 2002; Samson and Varga, 2007). The identification of fungal isolate was based on colony characteristics and conidial features of the isolates within genus level. The isolates were also identified by comparing their characteristics with those of known taxa, as described by Jolt *et al.* (1994) and Oyeleke and Manga (2008). To apply this, slide cultures were prepared for each of the isolates. The identified fungi were smoothly caught (2-3cm) with sterilized scalpel and they were placed on slide.

The technique of James and Natalie (2001) was applied for identification of the unknown isolated fungi using cotton blue in lacto phenol stain. The identification was achieved by placing a drop of the stain on clean slide with the air of mounting needle, where a small portion of mycelium from the fungal culture was removed and place on a drop of lacto phenol. The mycelium spread very well on the slide with the aid of the needle. A cover slip was gently applied with little pressure to eliminate air bubbles. The slides were mounted and observed under compound light microscope with magnification power of x10 and 40 objective lenses, respectively.

Results:-

Isolation of spoilage moulds from fruits:-

In this study, a total of 52 fruit spoilage moulds were isolated from 40 samples of deteriorating Orange (*Citrus sinensis*) and Tomato (*Lycopersicon esculentum*) fruits purchased from Kochi, Agip and Bishishe markets of Jimma town (Table 1). Of the total isolates, 26(50%) of the spoilage fruits were isolated from Orange (*Citrus sinensis*) and the remaining 26(50%) of the spoilage fruits were isolated from Tomato (*Lycopersicon esculentum*).

Table 1:- Total number of spoilage moulds isolated from deteriorating fruits sold at Kochi, Agip and Bishishe markets, Jimma town.

Fruits	No of samples	No of isolates	Isolates Proportion (%)
<i>C. sinensis</i>	20	26	50%
<i>L. esculentum</i>	20	26	50%
Total	40	52	100%

Characterization of the isolates

Physically observation of the diseased fruits revealed brownish, necrotic patches on the skin of the orange and tomato fruits. Table 2 shows that the colonial and morphological characteristic of the isolated spoilage moulds of fruits.

Table 2:- Colonial and morphological characteristics of moulds associated with the spoilage of fruits.

Isolates	Colonial characteristics	Microscopic morphology	Suggested Genus
JUO 01-02 JUT 03-07	Cottony, pink, purple, brown colonies.	Extensive septet mycelium and conidiophores simple or branched with ovoid to elongated conidia of variable size. Septate fusiform, slightly curved and pointed at both ends.	<i>Fusarium</i> spp.
JUO 08-17 JUT 18-22	Greenish or blue green colonies.	Conidia in long chains on repeatedly branched conidiophores resembling a brush like head (penicillus). Conidiophores smooth, relatively short. Penicillia mycelia arranged very irregular and asymmetrical with branches of various lengths.	<i>Penicillium</i> spp.
JUO 23-29 JUT 30-33	Colonies with loose white to yellow mycelium rapidly becoming dark brown to black on the development of conidia. Colonies light green-yellow.	Black, brownish black, purple brown Conidiophores and yellow to green conidia with dark sclerotia. Microscopically conidiophores arising from a foot-cell, catenate (basipetal) conidia on phialides (1or 2 series) on vesicle.	<i>Aspergillus</i> spp.
JUO 34 JUT 35-38	White to dark grey colonies, fast growing and filling the petri dish with dense cottony mycelium, producing mass of sporangia.	Nonseptate mycelium with root like rhizoids; black columellate, sporangiophores, in clusters and dark sporangia containing dark to pale spores.	<i>Rhizopus</i> spp.
JUO 39-41 JUT 42-46	Nonseptate hyphae, cottony colony, Smooth, non-striated Sporangiophores and Produce no rhizoids	Nonseptate or sparsely Septate, broad hyphae, sporangiophores, Sporangia and spores are visualized. Apophysis, rhizoid and stolon are absent.	<i>Mucor</i> spp.
JUO 00 JUT 47-49	Pink, gray, or tan conidia	Asterisk-like shape of the colony. Type "S" colonies grew slowly and stopped after days entirely.	<i>Monilia</i> spp.
JUO 50-51 JUT 00	Cottony colony Absence of ascocarps, asci in open clusters	Phialides with cylindrical bases that taper abruptly into long cylindrical necks and produce catenate conidia.	<i>Byssoschlamys</i> spp.
JUO 52 JUT 00	Thick, velvet colony, green, olive green, darkblue, black or brown colony and some lemon shaped conidia variously branched	Hyphae are Septate and brown in color. Conidiophores are brown and often Septate. Conidiophores are erect, straight or flexuous, unbranched or branched only in the apical region	<i>Cladosporium</i> spp.

Legend: O= Orange; T= Tomato; n= Code no of isolates

Occurrence of Fungi Isolates Associated with the Spoilage of Fruits:-

Of the total 56 spoilage moulds isolated from the two different types of deteriorating fruits (*C. sinensis* and *L. esculentum*), 52(92.85 %) isolates were characterized and identified as *Aspergillus*, *Fusarium*, *Penicillium* and *Rhizopus*, *Mucor*, *Monilia*, *Byssoschlamys* and *Cladosporium*spp. The remaining 4(7.15) were uncharacterized since

they were totally contaminated with bacterial growth. Generally, of all isolated moulds *Penicillium* sp. was the dominant isolate 15(28.81%), followed by *Aspergillus* sp. 11(21.15). *Byssoschlamys* and *Cladosporium* spp. were the least encountered 2(3.84%) and 1(1.92%), respectively (Table 3). In addition, most of the spoilage fungi were wide spread among all examined fruits. However, no *Monilia* was isolated from orange and no *Byssoschlamys* and *Cladosporium* spp. were isolated from tomato (Table 3).

From this outcome, it can be concluded that, *Penicillium* was the most spoilage fungi of orange and tomato than other fungal species. Next to *Penicillium*, *Aspergillus* was the dominant one for the spoilage of orange. And *Monilia*, *Byssoschlamys* and *Cladosporium* were found in small percentage compared to others.

Table 3:- Prevalence of spoilage moulds isolated from deteriorating fruits sold at Kochi, Agip and Bishishe, Jimma town, 2011/12.

	Orange (No of samples 20)	Tomato (No of samples 20)	
	No of isolates	No of isolates	Total
<i>Fusarium</i> sp.	2(3.84)	5(9.61)	7((13.45)
<i>Penicillium</i> sp.	10(19.2)	5(9.61)	15(28.81)
<i>Aspergillus</i> sp.	7(13.46)	4(7.69)	11(21.15)
<i>Rhizopus</i> sp.	1(1.92)	4(7.69)	5(9.61)
<i>Mucor</i> sp.	3(5.76)	5(9.61)	8(15.37)
<i>Monilia</i> sp.	-	3(5.76)	3(5.76)
<i>Byssoschlamys</i> sp.	2(3.84)	-	2(3.84)
<i>Cladosporium</i> sp.	1(1.92)	-	1(1.92)
al	26(50)	26(50)	52(100)

Note: Values in parenthesis are percentages of the isolated moulds.

Discussion:-

The findings of this study showed that *Aspergillus*, *Fusarium*, *Penicillium*, *Rhizopus*, *Mucor*, *Monilia*, *Byssoschlamys* and *Cladosporium* spp. were isolated from fruits sold in main markets of Jimma town (Kochi, Agip, and Bishishe). All the post-harvest spoilage moulds except *Monilia* spp. were found to be associated with spoilage or deterioration of orange (*Citrus sinensis*). Similarly, Al-Hindi et al. (2011) have reported that *A.niger*, *P. digitatum* and *R. stolonifer*, *Byssoschlamys* spp., *Cladosporium* spp. and *Mucor* were implicated in spoilage of *Citrus sinensis*. Tournas and Katsoudas (2005) also reported that *Fusarium* spp. were the most common fungi in citrus fruits. Bukar et al., (2009) also revealed that, the most predominant fungus isolated from the orange (*Citrus sinensis*) were, *Aspergillus* sp; others include *Mucor* sp, *Penicillium* sp, *Rhizopus* sp *Fusarium* sp, and *Alternaria* spp. A study conducted by Oviasogie et al., (2015) also confirmed that the fungal pathogens associated with the spoilage of orange (*Citrus sinensis*) were *Aspergillus* spp. *Penicillium* spp. *Mucor* spp. *Rhizopus* spp. *Candida tropicalis* *Saccharomyces cerevisiae* and *Alternaria* spp. in which *Aspergillus* spp. was the predominant fungal pathogen.

In the case of tomato fruits (*L. esculentum*), *Aspergillus*, *Penicillium*, *Rhizopus*, *Fusarium*, *Mucor* and *Monilia* spp. were implicated in its spoilage. In line with this, Mitra (1997) has discovered that the species of fungi associated with the spoilage of *Citrus sinensis*, *Asiminatriloba* and *L. esculentum* fruits include species of *Aspergillus*, *Fusarium*, Yeast, *Penicillium*, and *Rhizopus*. *A. flavus* and *A. Fumigatus* caused *L. esculentum* spoilage were also investigated by Al-Hindi et al. (2011). Seven fungal pathogens were associated and responsible for fruit rotting of *L. esculentum* caused by *Fusarium equiseti*, *A. flavus* and *A. niger*. Akinmusire (2011) also reported that fungi affecting *L. esculentum* includes *Fusarium oxysporum*, *Fusarium moniliforme*, *Aspergillus niger* and *Rhizopus stolonifer*. Ugwu et al. (2014) also discovered that the responsible fungal pathogens for the spoilage of tomato were *Candida tropicalis*, *Penicillium notatum*, *Aspergillus niger*, *Fusarium oxysporum*, *Absidia corymbifera*, *Rhizopus stolonifer*

In the current study *Penicillium* spp. was the most spoilage fungi of orange and tomato than other fungal species and this result is in agreement with the study conducted by Mbajiuka and Enya (2014) in which abundant presence of *Penicillium nalgiovense*, *Penicillium notatum* and *Penicillium expansum* were found among other fungi species involved in deterioration of tomatoes fruit.

The diseased fruits sampled from Kochi, Agip and Bishishe market were found to be massively infected with eight genera of fungi namely *Aspergillus*, *Fusarium*, *Penicillium* and *Rhizopus*, *Mucor*, *Monilia*, *Byssoschlamys* and *Cladosporium* spp. and four other uncharacterized spoilage moulds. The prevalence of fungi as the spoilage organism of fruits is due to a wide range of factors which are encountered at each stage of handling from pre-harvest to consumption and is related to the physiological and physical conditions of the produce as well as the extrinsic parameters to which they are subjected (Effiuvwevwe, 2000). Damage inflicted on produce at the time of harvest is a major cause of infection since most of the spoilage microorganisms invade the produce through such damage tissues; similarly, the extent of deterioration is influenced by the depth of the wound. Furthermore, the incidence of infection is worsened by poor sanitary practices such as cross-contamination, contact infection during the transportation of fruits (Effiuvwevwe, 2000).

Akinmusire (2011) also remarked that the contamination of fruits by fungi could also be as a result of poor handling practices in food supply chain, storage conditions, distribution, marketing practices and transportation. From personal observation at Agip, Kochi and Bishishe, the poor hygienic conditions of the vending house/store, the venders and the utensils used for keeping and weighing the fruits were responsible factors for mixing the deteriorating fruits with the healthier one and many other factors aggravate prevalence of fungi as the spoilage organism of fruits. In addition, the overall weather conditions of Jimma town, especially the high moisture content favours the growth of spoilage moulds on fruits and vegetables vended at Agip, Kochi and Bishishe. According to Alemu et al. (2011) from a climatic point of view, abundant rainfall makes this region one of the best watered of Ethiopian highland areas, conducive for the growth of post-harvest fruit spoilage moulds.

The overall analysis of the isolation and identification process implied various causes for the growth of the moulds on fruits. As clearly described by Korsten (2006), postharvest losses of fruits is a serious problem, because the values of fresh product significantly increase while passing from the farm to the consumers table and due to overpopulation the demand for fruits increases in the world. Fungal pathogens are mainly responsible for postharvest losses of fruits (Korsten 2006).

Conclusions:-

This study detected the profile of spoilage fungi involved in the deterioration of orange and tomato sold in main markets of Jimma town (Agip, Kochi and Bishishe). It showed that, fungi were involved in the spoilage of fruits. *Fungal flora of orange and tomato* samples found from February 2013 to May 2013 were dominated by moulds of *penicillium*, *Aspergillus*, *Mucor*, *Fusarium*, *Rhizopus*, *Monilia*, *Byssoschlamys* and *Cladosporium*. Generally, of all isolated moulds *Penicillium* spp. was the dominant isolate 15(28.81%), followed by *Aspergillus* spp. 11(21.15). *Byssoschlamys* and *Cladosporium* spp. were the least encountered 2(3.84%) and 1(1.92%), respectively.

The results of this study indicate that fruits sold at Agip, Kochi and Bishishe were massively infected with spoilage fungi due to several factors as poor hygienic conditions of the vending site/store, venders and vending utensils. In addition, the high humid content of the vending site also contributes for the spoilage. Mechanical injuries of fruits such as bruises or cuts was occur during harvesting or post harvesting, provide infection site for spoilage of fruits. The high moisture content of fruits and vending/storing site will be a serious limiting factor in their preservation. Since the fruits used in this study are produced in the neighboring woredas of Jimma town, thus, they are transported to the city in locally woven baskets and sacks under weather conditions that encourage the incubation of these contaminating fungi. Generally it can be concluded that, fruit spoilage fungi are caused due to the lack of occurrence of safety fruits within market sellers of Jimma town.

The high prevalence of the spoilage fungi demand that appropriate control measures against infection, should be employed if farmers expect good performance of their produce. Adequate mycological knowledge and handling practices of these produce would therefore help minimize wastes due to deterioration and unacceptability. It is therefore important that both the farmer who harvests the fruits into bags for transportation, the marketers and consumers take necessary precaution in preventing contamination and also try to create an environment that will not encourage the growth or multiplication of spoilage fungi. In addition, the hygienic conditions of vending/storing site, the venders and vending utensils should have to be improved in order to provide fresh and quality fruits for the consumers. This will help in providing fresh and quality fruits for the consumers as well as in preventing the consumption of contaminated fruits thereby reducing the risk of health problems which are produced by these fungi that have been isolated in this study.

This research pointed out the common fungi which spoil fruits of orange and tomato, which are sold in Jimma town market sellers. Thus proper handling of these fruits should be practiced to reduce the fungal genera. The majority of analyzed samples showed the presence of different fungal genera in fruits of orange and tomato. This indicates that the lack of selling parameters of fruits found in Jimma town market sellers. So, appropriate measurement must be taken by responsible bodies to reduce the fungal load and to enhance the quality of fruits sold in Jimma town.

In most unorganized markets, fruits were available in local retain shops without appropriate temperature control and unsuitable storage. This is purchased by households, this leads to the disturbance of the health of the people. So, inhibition of fungal growth by lowering storage temperature through storage under refrigeration and use of fungicides must be applied.

The hygienic and sanitary conditions observed in fruit sellers were not very satisfactory. Thus, they should improve the sanitary condition of fruit markets. In order to improve the quality of fruits, it is better to give awareness for consumers as well as sellers and to all others who have access these fruits must understand about the following critical control points;

Since molecular characterization of the isolates reveals the true diversity of spoilage fungi associated with fruits than the traditional morphological characterization, molecular technique is recommended. This would be the delimitation/drawback of this study.

Conflict of interests:-

The author(s) have not declared any conflict of interests.

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RESEARCH ARTICLE

FREQUENCY OF PERIPHERAL NEUROPATHY IN PATIENTS WITH CHRONIC KIDNEY DISEASE: A CLINICAL STUDY.

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Key words:-

CKD, Diabetes, ESRD, Hemodialysis,
Hypertension, Peripheral Neuropathy.

Abstract

Chronic kidney disease (CKD) is one of the major health problems in modern world. Peripheral neuropathy is considered to be an important complication in these patients. It can cause foot ulcers and may lead to circulatory problems. Regular and timely screening has the potential in reducing the incidence of complications due to peripheral neuropathy. The present study is conducted to measure the frequency of peripheral neuropathy among patients with CKD being treated with hemodialysis.

Objectives:- To measure the frequency of peripheral neuropathy among patients with chronic kidney disease.

Methods:- The current study was conducted in Al-Jabar Kidney Center under the Ministry of Health in Al-Ahsa region, Eastern province, Kingdom of Saudi Arabia. Peripheral neuropathy was evaluated clinically in patients included.

Results:- About 62.5 % patients of CKD were found to have peripheral neuropathy clinically. Out of these 78 % were diabetics while 94 % were hypertensive.

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Introduction:-

Chronic kidney disease (CKD) is a common disease that is characterized by pathological kidney abnormalities with persistent proteinuria. CKD is divided into 5 stages, based on glomerular filtration rate (GFR). In stage 1 and stage 2, GFR is near normal with functional and structural defects (e.g. hematuria and persistent proteinuria). Stage 3 has low risk of progression to kidney failure, the GFR being 30-59 mL/min. While in late stages, like 4 and 5, there is loss of significant renal function. The last stage (stage 5), called renal failure or end stage renal disease (ESRD), patients need dialysis or kidney transplant. End stage renal disease (ESRD) is the last stage of chronic kidney disease. It is irreversible condition of impairment of renal function when the kidney is unable to detect the balance of fluid and electrolytes, and unable to excrete waste product so, the patient develops uremia. The ESRD has more prevalence among diabetic and hypertensive patients, and is associated with various complications; like cardiac abnormality, neuropathy, hyperlipidemia, anemia and metabolic bone disease. Neurological complications are common in patient with ESRD, secondary to uremic state, affecting nervous system at any level; central and peripheral. So, patient may suffer from prolonged disability, weakness, loss of sensations, and alteration of mental state. Uremic peripheral neuropathy is symmetrical distal polyneuropathy with slowly progressive sensorimotor axonal neuropathy. The presentation of uremic peripheral neuropathy varies from patient to patient. Patients may present by sensory manifestations insidiously, which may consist of tingling and prickling sensation in the lower

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extremities, paresthesias, loss of touch, pain and temperature sensations. The rationale of this study is, if there is high frequency of involvement of peripheral neuropathy (loss of position, vibration, pain and touch) in ESRD patients on frequent hemodialysis; then screening for this should be among the routine clinical examination of patients with CKD.

Material and methods:-

The study aims to first determine the frequency of peripheral neuropathy in hemodialysis patient and plan for treatment and prevention if found. The setting was in hemodialysis department, the duration of the study was: 5 months. We used Cross-sectional Study design. Sample size of 80, of which 42 were females and 38 were males, the age group was from 30 to 60 years. Purposive non-probability sample technique. We selected all patients of 30 to 60 years of age, who had ESRD and were on hemodialysis. They varied from medical illness free to patient with hypertension or diabetes. We excluded patients who were previously diagnosed with Peripheral neuropathy.

Data collection procedure:-

After taking clinical history, 80 patients were included in the study with clinical features of renal failure; all patients were on hemodialysis, in department of dialysis, at Al-Jabar Nephrology center in Al-Ahasa. Informed consent was taken from each patient.

Detailed history was obtained. Frequency of neuropathy was detected by clinical examination of the sensations (touch, pain, position, vibration) supervised by consultant. Then according to data, we categorized patients according to age, gender, HTN, DM, the presence of peripheral neuropathy and the affected sensations. Data was entered using excel. Frequency and percentage of neuropathy was calculated with proportions of presence or absence of involvement of clinical symptoms.

Discussion:-

Neuropathy is the late complication occurring in the uremic nephropathy. It has also been resulted from nutritional deficiency, chronic kidney disease and diabetic nephropathy. It's outcomes generally not tolerated, and its main effect regarding sensations include pain, paraesthesias, amputations and disabilities (vagner, 2011). Hot feet sensation a less complications reported in 42% of cases. However, neuropathy is associated with Autonomic dysfunction. In patients with ESRD, sensory ataxia is an important complication. Physiotherapy is the main stay to prevent this ataxia. However, physiotherapy contribute to maintain independence as possible and insure easier mobility. Sensory neuropathy may cause an increased susceptibility to foot ulcer including infections, gangrene, amputations. The literature reveals many reported instances of amputations associated with uremic neuropathy, as well as report of amputations in CKD (PAUL, 1999). The national institute of neurological disorders (NINDS) recommends adapting self-care exercise. In some other studies, more complex treatments recommended with systemic diseases. Although our patients had renal failure, given the irreversible nature of disease with dialysis, uremia toxicity prevention recommended by American family physician (AFP) is kidney transplant. Reported that patients with renal failure who were complicated with neuropathy, had increase chance of morbidity. The median overall morbidity rate was 13% (hari, 2013). Our patients had significant loss of sensations clinically but further evaluation by nerve conduction studies, could not be performed. However, under clinical findings, neuropathy was confirmed by examination. In study we focused on peripheral neuropathy, but there are other studies (Doreen, 2011) on primarily axonal sensorimotor polyneuropathy and they noted that peripheral neuropathy occurs in 50–60% of patients who require hemodialysis. In our patients, we use clinical examination to detect peripheral neuropathy, and we found 50 of 80 patients were having peripheral neuropathy (62.5%), of which 39 were diabetics (78%) and 47 were hypertensive (94%), and we found similar results in other studies (Aggarwal, 2013)

Result:-

Peripheral neuropathy was diagnosed using the previously mentioned methods and the results came as the following:

Details	Number of all patients	Affected
Number of Male	38	21
	48%	42%
Number of Female	42	29
	53%	58%
Total	80	50
		62.50%
Details	Number of patients	

Number of all patients have DM (out of 80)	47
	58.75%
Number of affected patients have DM (out of 50)	39
	78%
Number of all patients have HTN (out of 80)	72
	90%
Number of affected patients have HTN (out of 50)	47
	94%

Type of lost sensation	Number of patients
vibration	19
position	20
pain	6
Touch	22
All	11

Out of 80 patients of renal failure, which were on hemodialysis, 50 patients were having peripheral neuropathy (62.5%) , of which 39 were diabetics (78%) and 47 were hypertensive (94%). Out of 42 female patients, 29 were affected (58%) and out of 38 male patients 21 were affected (42%). Out of the whole 50 affected patients, the fine touch sensation was affected in 22 of them (44%), position sensation was affected in 20 of them (40%) , and vibration was affected in 19 of them (38%), while pain was affected in only 6 of them (12%). We found total impairment of all sensations in 11 of them (22%) and all of these were diabetics for more than 10 years.

This reflects the affected tracts of sensory pathway, as the fine touch and proprioceptive sensation and vibration are the most affected sensations, this indicates that the neuropathy has more impact on the dorsal Column-Medial Leminiscal Pathway, and the least affect is on the lateral spinothalamic tract; which carries the pain sensation.

We also found that the duration of hemodialysis doesn't correlate with the progression of neuropathy, as many patients were affected in all of examined sensations while they just started hemodialysis, but it is correlated with other factors such as the control of the other co-morbidity in which here is diabetes, and the intensity of the hemodialysis and the progression of the underlying pathology of the kidneys.

Conclusion:-

Patients with end stage renal disease in Al-Ahsa have high frequency of peripheral neuropathy, which is more observed in patients with underlying diabetes. Control of diabetes and improved uremic control by frequent hemodialysis improve uremic neuropathy.

Limitations:-

- patients' education level sometimes affects the understanding and the perception of the clinical examination as the examination is subjective to what they feel or think.
- Patients' psychological status at time of examination affects the result either by exaggeration because of depression or denying because of fear.
- couldn't assess the nerve conduction velocity studies and get more accurate results because of lacking of facilities and access to the NCVS.
- no assessment of the crude touch and temperature sensations.

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RESEARCH ARTICLE

ASSESSMENT OF SPIROMETRY IN GASTROESOPHAGEAL REFLUX DISEASE (GERD) ASSOCIATED ASTHMA PATIENTS.

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Key words:-

Gastroesophageal Reflux Disease, chronic cough, asthma.

Abstract

Introduction: Gastroesophageal reflux disease (GERD) can cause several esophageal complications, as esophagitis, ulceration, stricture, hemorrhage, and Barrett's esophagus. Though, the associated problems with GERD have expanded to extra esophageal sites. Chronic cough and asthma are two clinical problems caused or triggered by GERD. Spirometric changes among GERD associated cough and asthma still a topic of ongoing research.

Aim of the study: to assess the spirometric parameters of GERD associated cough and asthma patients in relation to different clinical aspects of the disease including body mass index (BMI), disease duration, presence or absence of symptoms as well as endoscopic findings.

Patients and methods: 90 subjects of either sex with a mean age of 40.5±12 year are included in the study, 55 GERD associated cough and asthma patients served as test group while the remaining 35 subjects served as control group, each patient, each patient underwent spirometry and gastric endoscopy during the period from September 2015 to March 2016.

Results: Spirometric data of the control group were within the normal predicted range (80-120%) thus excluding the possibility of any asymptomatic obstructive airway disease. Whereas, the measured spirometric parameters (forced vital capacity (FVC), forced expiratory volume in 1 second ratio (FEV1%), peak expiratory flow (PEF), and forced mid expiratory flow (FMF) are significantly reduced in the test group with a clear restrictive pattern among the overweight subjects. Furthermore, neither the presence nor duration of symptoms affected the spirometric picture of the GERD associated cough and asthma subjects, unlike those with positive endoscopy findings who revealed significant obstructive pattern when compared to those with negative endoscopy findings.

Conclusions: GERD associated cough and asthma patients with positive endoscopy findings showed obstructive pattern of spirometry irrelevant to the presence or absence of symptoms or duration of the disease.

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Introduction:-

Gastroesophageal reflux disease (GERD) is a condition that develops when the reflux of stomach contents causes troublesome symptoms and/or complications (1). The true prevalence of GERD in the population is underestimated and the disease is often misdiagnosed. Approximately 20–40% of the adult populations in Western countries suffer from heartburn and/or regurgitation that have a negative impact on the patient's daily activities, sleep and working life, irrespective of the presence or absence of esophagitis (2, 3). According to the Montreal Definition of GERD, concomitant gastroesophageal reflux symptoms is “aggravating factor” to extraesophageal symptoms and lesions like reflux cough, reflux laryngitis and viewed as an reflux asthma (4). Gastroesophageal reflux is a potential trigger of asthma. Approximately 77% of asthmatics report heartburn (5). There is a cause effect relationship between asthma and gastroesophageal reflux which can turn into a vicious cycle (6).

The basic motivation behind conducting the present work was an attempt to answer the questions: Can GERD cause an abnormal spirometry test? An issue of ongoing research.

Patients and Methods:-

Ninety adult patients of either sex aged 40.5 ± 12 years (19– 65years) included in this study, from whom a written consent plus data sheet, including all necessary personal and health information were obtained.

They were classified as:-

(A) Control group: included 35 apparently healthy subjects recruited from patient companions, relatives, and friends in addition to hospital health staff workers after fulfilling the following criteria:

1. Never smokers.
2. No history of any significant pulmonary or cardiac diseases.
3. Clear chest on physical examination.
4. Absence of any musculoskeletal, neuromuscular disorders, or any other condition which could affect spirometry test.

(B) Test group: included 55 patients with gastro-esophageal reflux disease associated cough and/or asthma approved by history, clinical examination and other necessary investigations.

Patients were selected from outpatient subjects referred to the Endoscopy Unit and Pulmonary Function Units, during the period from September 2015 to March 2016.

Table 1, shows the distribution and clinical data of the test group.

Table 1:- Distribution and clinical data of the test group.

BMI		GERD Symptoms		GERD duration		Endoscopy	
Normal body wt BMI (19-25)	Over body wt BMI > 25	Present	Absent	< 3 months	> 6 months	Negative	Positive
N=14	N=41	N=40	N=15	N=21	N=34	N=18	N=37

Anthropometric measurements: Height, weight were measured using standard techniques as follows: height to within 0.5 Cm, without shoes; Weight to within 100g, without heavy clothing (table 2). Body mass index (BMI) was calculated by the following

Formula:-

$$\text{BMI} = \text{Weight (kg)} / [\text{Height (m)}]^2 \text{ (7)}.$$

Table 2:- Demographic and anthropometric data of the studied groups.

Parameter	Control group (N=35)			Test group (N=55)		
	Minimum	Maximum	Mean \pm SD	Minimum	Maximum	Mean \pm SD
Age (year)	19	58	39 ± 11	20	65	42 ± 13
Height (m)	1.6	1.86	1.68 ± 0.06	1.47	1.80	1.64 ± 0.08
Weight (Kg)	49.5	98.5	75.3 ± 13	52.8	117.5	87.7 ± 19.45
BMI	18.6	33	24.2 ± 5.27	17.9	42	28.2 ± 6.14

Spirometry: percent predicted forced vital capacity (FVC), forced expiratory volume in 1 second (FEV1 %) ratio, peak expiratory flow (PEF), and the forced mid expiratory flow (FMF) were measured using computer based spirometer. Standard procedure was adopted following the recommendations of the American Thoracic Society (ATS) (8).

The forced expiratory vital capacity procedure was described and demonstrated to the subjects before the test while encouraging breathing out as long and forcefully as possible. The best of three technically satisfactory maneuvers was recorded.

Statistical analysis: The data obtained in the study were analyzed using the SPSS statistics for data analysis. Standard statistical methods were used to determine the mean and standard deviation. Descriptive data analysis was used to describe different variables in addition to the Independent Sample T-test to compare different means.

Results:-

The percentage predicted spirometric values of the control and test groups shown in (table 3). The healthy control group is within the normal predicted range (80–120%) regarding all measured spirometric values, unlike the test group who showed highly significant reduction in the FEV1%, PEF and FMF ($p < 0.01$) revealing a clear obstructive airway pattern in addition to a significant reduction in FVC (restrictive index, $p < 0.05$).

Table (4) compares the percentage predicted spirometric parameters between normal body weight and overweight groups. The overweight group showed significant reduction in the FVC as compared to the normal weight group ($P < 0.05$) while other spirometric parameters (FEV1%, PEF and FMF) does not show significant difference.

Table (5) compares the percentage predicted spirometric parameters between asymptomatic and symptomatic GERD groups. All measured spirometric parameters (FVC, FEV1%, FPEF and FMF) were not found to be significantly different between the two groups.

Table (6) shows the percentage predicted spirometric parameters when compared between the test groups with short duration of GERD symptoms, less than 3 months and those with longer than 3 months duration of GERD symptoms. No significant differences were found between the two groups regarding again all measured spirometric parameters.

Table (7) shows the spirometric parameters within the test group with negative endoscopic findings as compared with those of positive endoscopic findings for GERD. No significant difference was observed regarding the FVC while there is a statistically significant reduction in the FEV1%, FMF ($p < 0.01$) and PEF ($p < 0.05$) among the group with positive endoscopic finding.

Table 3:- Comparison of the spirometric parameters between test and control groups.

Parameter (% predicted)	Mean \pm SD (minimum-maximum)		P value
	Control group (n = 35)	Test group (n = 55)	
FVC	99.76 \pm 9.35 (80.8-115.5)	93.66 \pm 15.44 (50.1-101.0)	<0.05
FEV1%	89.56 \pm 8.51 (82.3-116)	78.19 \pm 11.86 (49-84.9)	<0.01
PEF	96.69 \pm 11.84 (84.7-118.8)	82.22 \pm 17.41 (41.5-91.08)	<0.01
FMF	90.11 \pm 10.21 (82.8-119.9)	68.93 \pm 18.86 (41.2-83.02)	<0.01

Table 4:- Comparison of Spirometric parameters between normal body weight and overweight groups.

Parameter (% predicted)	Mean \pm SD		P value
	Normal body weight (BMI = 18.9-25) n=14	Over body weight (BMI > 25) n=41	
FVC	98.67 \pm (6.36)	90.38 \pm (17.11)	P < 0.05
FEV1%	71.10 \pm (10.18)	72.23 \pm (11.54)	NS
PEF	84.69 \pm (13.99)	81.64 \pm (18.01)	NS
FMF	58.82 \pm (11.58)	63.12 \pm (13.07)	NS

Table 5:- Comparison of Spirometric parameters between asymptomatic and symptomatic GERD groups.

Parameter (% predicted)	Mean \pm SD		P value
	Asymptomatic GERD n=15	Symptomatic GERD n=40	
FVC	89.23 \pm (14.53)	93.71 \pm (15.83)	NS
FEV1%	77.31 \pm (11.62)	75.93 \pm (9.02)	NS
PEF	82.93 \pm (15.7)	80.56 \pm (15.56)	NS
FMF	64.94 \pm (14.97)	63.93 \pm (13.27)	NS

Table 6:- Spirometric values of test group in respect to the duration of GERD symptoms.

Parameter (% predicted)	Mean \pm SD		P value
	GERD duration (< 3 months) n=21	GERD duration (> 3 months) n=34	
FVC	91.12 \pm (18.12)	93.34 \pm (12.51)	NS
FEV1%	74.96 \pm (11.62)	72.73 \pm (9.71)	NS
PEF	85.50 \pm (17.04)	78.55 \pm (14.05)	NS
FMF	65.00 \pm (12.05)	63.72 \pm (14.65)	NS

Table 7:- Spirometric values of test group in respect to endoscopic findings.

Parameter (% predicted)	Mean \pm SD		P value
	GERD (negative endoscopy) n=18	GERD (positive endoscopy) n=37	
FVC	94.03 \pm (18.06)	93.38 \pm (13.37)	NS
FEV1%	84.79 \pm (8.95)	73.07 \pm (11.39)	P < 0.01
PEF	87.25 \pm (18.01)	78.32 \pm (16.15)	P < 0.05
FMF	84.07 \pm (18.51)	57.21 \pm (7.23)	P < 0.01

Discussion:-

When spirometric measurements were compared between the age and height matched studied groups (table 2), the GERD associated cough and asthma group with an overweight BMI (table1) showed significant reduction in all measured spirometric variables, signifying the growing body of evidence relating overweight and obesity with a wide range of health conditions including chronic obstructive lung disease (COPD) and asthma (9,10,11). While the test group further subgrouped on the basis of BMI in an attempt to rationalize the restrictive pattern, the results revealed significant reduction in the percentage predicted FVC ($p < 0.05$) among the overweight group in respect to those with normal weight group (table 3); this finding has two implications, first, it could further confirm the fore mentioned relation between overweight and obstructive spirometric pattern, second, it can reinforce the role of gastroesophageal reflux in chronic cough and asthma(12).

Concerning the link between gastric acid reflux and respiratory symptoms (cough, dyspnea and wheezes), the test group was classified according to the presence of symptoms (symptomatic and asymptomatic) and to the duration of reflux symptoms into short duration (< 3 months) and long duration subgroups (> 3 months) (table 4 and 5). The obtained data (tables 4 and 5) of the nonsignificant differences in all measured spirometric values, revealing that neither the presence nor the duration of GERD symptoms could affect spirometry. These findings might be attributed to the control of acid reflux with medications for example, a proton pump inhibitor.

Consequently, the obstructive spirometric pattern (table 6) in the GERD subjects with positive endoscopic finding could be due to the effect of the acid reflux on the respiratory passageways. Though the cause-effect relationship between asthma and acid reflux isn't clear why, acid reflux can worsen asthma and asthma can worsen acid reflux (6). However, Chan WW and co-workers, 2011 claimed that gastroesophageal reflux disease occurs frequently among patients with asthma, despite that the therapy with proton pump inhibitors to improve asthma control remains controversial (13). Moreover, two proposed mechanisms involved in the pathogenesis of GERD associated cough: (1) acid in the distal esophagus stimulating an esophageal-tracheobronchial cough reflex, and (2) micro or macroaspiration of esophageal contents into the larynx and tracheobronchial tree. Dual-probe esophageal pH testing

with pH electrodes in the proximal and distal esophagus, esophageal acid infusion studies, and evaluation of agents inhibiting cough have added significantly to the understanding of GERD-associated cough (12).

Conclusion:-

The study concludes that Gastroesophageal reflux associated cough and asthma patients showed an obstructive spirometric pattern irrelevant to the presence or absence of symptoms or duration of the disease. On the other hand, the same obstructive spirometric pattern was reported in those with positive endoscopic findings when they were compared with subjects with negative findings.

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RESEARCH ARTICLE

SMOKING PREVALENCE AND ATTITUDE AMONG HEALTH SECTOR STUDENTS IN UMM AL-QURA UNIVERSITY, SAUDI ARABIA.

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Smoking, Dental Health Attitude, Prevalence.

Abstract

Background: Smoking is one of the major causes of death in the world. There is variability in the prevalence of smokers around the world.

Objectives: The aim of this study was to compare the prevalence and attitude of smoking among dental, pharmacy, and nursing students in Umm Al-Qura University.

Methods: The present study was a cross-sectional study, 850 students who are in the 2nd to the 6th year were participated in this study. Hard copy questionnaire were distributed among the students during lecture times. The questionnaire was composed of two sections. Section one included three demographic questions including age, college, and gender. Section two included 17 questions asked about (the reasons behind smoking habit, the role of the college to prevent smoking, the awareness of those smokers with the damaging effect of smoking, the reasons for failing to stop smoking and whether there are any members of his/her family smoke). The questionnaire take around 5 minutes to be answered.

Results: A total of 700 students answered the study questionnaires (82.35% of the distributed questionnaires). Smoking prevalence was 8%, the high percent of smokers was male students(73.3%) while female students were(26.7%). The prevalence of smokers among dental students was higher than pharmacy students (11% vs 9%) and found to be zero among nursing students. Most smokers exercised in order to avoid the pressure (44.6%) and get a relaxed feeling(60.8%) and the most smokers want to quit it(87.5%). A large percentage of students were dissatisfied with the role of their college awareness towards this habit (47.6%).

Conclusions: The prevalence of smoking was high among dental and pharmacy students, the health colleges should do more efforts to prevent smoking among their students.

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Introduction:-

According to the World Health Organization (WHO) Smoking is one of the major causes of death in the world. Each year more than a million people die due to smoking and each year this percentage is unfortunately increased. Smoking has been classified by the WHO as an epidemic because of its fast spread among peoples and also its dangerous effect on human (Tjandra, 2002 and WHO, 2003). More than 80% of those who practiced smoking are low-income peoples (WHO, 2013).

There is variability in the prevalence of smokers around the world. For instance, the prevalence of smoking in the Indonesia was 76.2% (the highest prevalence rate), 70.2 % in Jordan, 60% in Sierra Leone, 57.5% in Georgia, 48.8% in Bahrain, 32.4% in Germany, 20.4% in Sweden, 19.5% in United States of America and the lowest prevalence rate 13.1% in Ghana and Barbados (WHO, 2015).

In Saudi Arabia, for the general population many studies investigated the prevalence of smokers. These studies indicated that the prevalence of current smoking in Saudi Arabia ranged between (9-35%), and among school students the smoking prevalence was 16.5%, in university students 13.5%, among adults 22.6%, elderly peoples 25%, and males 26.5% while in females 9%. (WHO, 2015; Bassiony, 2009; Alnohair, 2011; and Hashim, 2000).

In fact, some studies investigated the prevalence of smokers among Saudi universities and colleges students in general, which found to be 14.5% (Mandil, 2011). Other studies focused on students who study in the health colleges (such as medicine and dentistry), and found the prevalence to be 16%, which considered to be higher than the previous percentage, and when it compared male students in different health colleges on a national scale in Saudi Arabia as (Central: King Saud University, King Fahad Medical City, Qassim university; North: Aljouf university, Hail university, Tabuk university; South: Najran university, Jazan university, King Khalid University; East: Dammam university; and West: King Abdulaziz University and Taibah university) and indicated that dental students smoke more than medicine students (Alrashedi et al, 2012). As the prevalence among dental students was 18% compared to medical students with a prevalence of 14%. However, this study did not include females, and did not consider Universities of Makkah city, despite the fact Makkah is considered one of the major cities nationally. Furthermore, a recent study was conducted in Najran university among health college students and found that the prevalence of smoking in male and female students (30.1 %, 0.5%) respectively, the students at the college of applied medical sciences had the highest prevalence of smoking 34% followed by the students at colleges of pharmacy (15%) and dentistry (9%), and the lowest prevalence (4%) was at the college of medicine. (Alqahtani, 2017).

There is a study to measure the prevalence of smokers in the Umm al-Qura in college of medicine among male students and found that the prevalence of smoking was 30% (Alqarni et al, 2015). According to our knowledge there was no previous studies carried out to investigate smoking among undergraduate students of dental, pharmacy and nursing colleges in Umm Al-Qura University. Thus, the aim of the present study is to compare the prevalence and attitude of smoking among dental, pharmacy, and nursing students in Umm Al-Qura University.

Aims:-

The aim of the present study is to compare the prevalence and attitude of smoking among dental, pharmacy, and nursing students in Umm Al-Qura University.

Methods:-

This study is a cross-sectional survey, investigated students in colleges of dentistry, nursing, and pharmacy, Umm Al-Qura University, Saudi Arabia. Only students who are in the 2nd to the 6th year were included in this study. Students in the 1st (preparatory year) and interns were excluded from this study. A hard copy questionnaire was distributed among 850 (316 male and 534 female) students. The questionnaire was disseminated to the students during lecture times by the assistance of the academic office in each college, and after the permission of lecturers, to be filled after the lecture time. The data collector was available to answer any question raised by students. After students fill the questionnaires, they were taken back.

The questionnaire was composed of two sections. Section one included three demographic questions including age, college, and gender. Section two included 17 questions asked about (number of smoking times per day, the reasons

behind smoking habit, the role of the college to prevent smoking, the awareness of those smokers with the damaging effect of smoking, the reasons for failing to stop smoking and are there any members of his/her family smoke).

The questionnaire was tested on a pilot group, and was adjusted before achieving the final version. This pilot study was conducted on 12 students from dentistry, nursing and pharmacy colleges. The participants included in the pilot study were excluded from the study. The questionnaire takes around 5 minutes to be answered. This study was approved by the institutional review board at the college of dentistry, Umm Al-Qura University.

Statistical analysis:-

Data was statistically analyzed using SPSS v.17. Descriptive statistics were generated using frequency tables. Chi-square test was used to compare the prevalence of smokers among different colleges and between male and female smokers. A p-value ≤ 0.05 was considered statistically significant.

Results:-

700 students out of 850 answered the study questionnaire, with a response rate of 82.35%. The mean age of the participants was 21.30 ± 1.40 years old. There were 268 male (38.3%) and 432 (61.7%) female students. There was a significant difference in the number of male smokers (41 smokers and 227 non-smokers) compared to female smokers (15 smokers and 417 non-smokers), with a $p < 0.001$ (Chi-square test). The prevalence of smokers among different health colleges is presented in table 1, the overall students experience to smoking is illustrated in table 2, the attitude and behavior among smokers only are presented in table 3 and the attitude of the health colleges in dealing with smokers is illustrated in table 4.

Table 1. The prevalence of smokers among different health colleges.

College	Smokers No (%)	Non smokers No (%)	Total No (%)	Significance
Dentistry	20 (3)	164 (23.3)	184 (26.3)	< 0.001
Pharmacy	36 (5)	348 (49.8)	384 (54.8)	
Nursing	0 (0)	132 (18.9)	132 (18.9)	
Total	56 (8)	644 (92)	700 (100)	

No= Number, comparison done using Chi square test

Table 2. Overall students' smoking experience.

	Number	%
Smoking		
Yes	56	8
No	644	92
Smoker in family		
Yes	420	60
No	280	40
I think smoking affect gum		
Yes	638	91.1
No	62	8.9
Efforts of my college against smoking		
Enough	248	35.4
Not enough	333	47.6
Never	119	17
My college give warning for smokers		
Yes	333	47.6
No	367	52.4

Table 3. The smoker students' attitude and behavior toward smoking.

	Number	%
Smoking type		
Cigarettes	33	58.9
Water pipes	23	41.1
Reasons for smoking		
Action against authority	3	3.7
Due to pressure	25	44.6
Draw attention	7	12.5
Due to friends	22	39.2
Smoking benefits from the smoker perspective		
Feel relaxed	34	60.8
Help to concentrate	12	21.4
Excuse for a break at work	10	17.8
Wish to stop smoking		
Yes	49	87.5
No	7	12.5
Stopped and returned to smoke before		
Yes	9	16.1
No	47	83.9
Visited specialty clinic for stopped smoking		
Yes	7	12.5
No	49	87.5

Table 4. The students' perception about health colleges attitude toward smoking.

	Dentistry	Pharmacy	Nursing	p-value
Faculty advise to quit smoking				
Enough	54	101	93	<0.001
Not sufficient	102	224	7	<0.001
Never	28	59	32	<0.001
Warning procedure for smokers				
Yes	92	130	111	<0.001
No	92	254	21	<0.001

Discussions:-

A series of studies set up to determine the prevalence of smokers among the population of the Kingdom of Saudi Arabia but there is no studies carried out on health colleges of Makkah city. The findings of the present study indicated that, the percentage of smokers among students of health colleges is only 8% (table 1). The low percentage of health care students who smoke may be due to the greater awareness of them with respect to tobacco, to being health care professionals and being able to see its pernicious effects directly in their daily clinical practice. These results were agreed with previous study which shown that the prevalence of smokers among dentists is considered a small percentage compared with the general public (Peidró et al 2006).

The proportion of smokers among male students is higher than female students. There were no smokers in the college of nursing (Table 1) and the main reason is the advices provided by the college for female students where there are a large percentage of them satisfied about their college advices compared to other colleges (Table 4).

The results of the present study showed that, the main reason for smoking is the pressure and simulation of their friends practiced smoking. Some practiced it for their sense of comfort and escape from the pressure, and a large percentage of them wish to quit smoking (Table 3).

Given the evidence of the action of health care students in tobacco use prevention and cessation, specific strategies should be developed to motivate and support oral health professionals in training for topics relative to tobacco use prevention and cessation in the health colleges as part of their routine clinical practice.

It is necessary to assess attitudes of health care students with regard to the regular establishment of advices on smoking prevention in their colleges in order to, if appropriate, prepare campaigns suited to these professionals ,both educational and with regard to the distribution of support materials for this activity.

Conclusions:-

The prevalence of smokers among dental students is higher than pharmacy students and found to be zero among nursing students. The smoking was more prevalent for male students than female. However, the prevalence of smoking among the health faculties in Saudi Arabia considered lower than the general population. Health colleges are recommended to do more efforts to coup with smoking among the students.

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RESEARCH ARTICLE

DENTIGEROUS CYST TRANSFORMING INTO MUCOEPIDERMOID CARCINOMA OF THE MAXILLARY SINUS IN A 10 YEAR OLD CHILD – A RARE CASE REPORT.

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Dentigerous cyst, mucoepidermoid carcinoma, impacted tooth.

Abstract

Mucoepidermoid carcinoma of the jawbones is an extremely rare malignant tumour involving maxillary sinus. We report a case of mucoepidermoid carcinoma of the maxillary sinus in a 10-year old male patient developing from a previously diagnosed dentigerous cyst. An excisional biopsy was performed and the histopathological features confirmed mucoepidermoid carcinoma. The origin of central mucoepidermoid carcinoma could be suggested to be from the epithelial lining of previously diagnosed dentigerous cyst. Thus, emphasizing the need for careful examination of the entire excision specimen to rule out such neoplastic transformation of epithelial lining of odontogenic cyst and provide appropriate and effective treatment.

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Introduction:-

Dentigerous cyst is a most common developmental odontogenic cyst that originates by separation of dental follicle around the crown of unerupted tooth. Usually asymptomatic and associated with dentigerous cysts unless there is secondary infection. Any tooth may get involved, but most commonly mandibular third molar is involved (Neville et al.2006). The age range varies widely, from 5 to 57 years (Shear 2007). The highest incidence of dentigerous cyst occurs during second and third decades of life and rarely seen during childhood. According to Browne et.al¹ the sex predilection of dentigerous cyst is 1.6:1(male: female). Sometimes, lining of dentigerous cysts may transform into malignancies like squamous cell carcinoma(SCC)^{2,3,4,5,6} (50%), mucoepidermoid carcinoma(MEC)^{7,8} (50%), ameloblastoma⁹ (14.6%) and adenomatoid odontogenic tumour(AOT)¹⁰ (73%).

Malignant tumors of the paranasal sinus are uncommon, constituting less than 1% of all malignancies¹¹ and 3% of all head and neck cancers and in that malignant transformation of odontogenic cysts into mucoepidermoid carcinoma is estimated to be between 0.13% and 2%, with most of the cases involving the mandible¹². Mucoepidermoid carcinoma accounts for 13% of all malignancies occurring in maxillary sinus¹³. 7 cases of mucoepidermoid carcinoma which was transformed from dentigerous cyst have been reported in children younger than age 16 of which 4 cases involved mandible and 3 cases involved maxilla^{14,15}.

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We report a case of mucoepidermoid carcinoma of maxillary sinus arising from previously diagnosed case of dentigerous cyst involving impacted permanent maxillary canine in a 10 year old male child.

Case Report:-

A 10 year old boy reported to GITAM Dental College and Hospital with a painful swelling in the right upper posterior region producing a feeling of dullness in the right maxillary sinus region. It has been present for about 3 months. Pain was continuous, radiating, aggravated on slight touch, during mouth opening and on lying down position and relieved by itself. There was no significant past medical history, family history or personal history. On general examination, patient was moderately built, nourished, co-operative and responsive. No rise in temperature and vitals were normal.

Extra-orally, on inspection, gross asymmetry on right side of the face was noticed (Fig.1). Swelling was irregular measuring about 2x3 cm, with obliteration of nasolabial fold. It extends from lateral wall of nose anteriorly to tragus of the ear posteriorly. It extends from 1cm above the ala-tragal line superiorly to 1cm below the ala-tragal line inferiorly. On palpation, swelling was very tender, firm, non-mobile, non-compressible, non-reducible, non-fluctuant, no sinus openings over the swelling.

Intra-orally, a swelling was seen on buccal and palatal portion of maxillary posterior region. Swelling was irregular extending from 53 to 16, completely obliterating the buccal vestibule (Fig.2).



Fig. 1:- Facial asymmetry



Fig. 2:- Swelling.

On palpation, swelling was very tender, firm, non-fluctuant, non-mobile, non-compressible, non-reducible. Teeth with tender on percussion were 53, 54, 55 and 16. The swelling was seen as an ill-defined bony mass, hard in consistency, erythematous surrounded by pinkish red area.

Panoramic radiograph revealed a massive radiolucent lesion from 53 to 16 involving maxillary sinus. The radiolucency was regular, well defined, trabecular pattern of alveolar bone was completely lost and lesion was invasive. Impacted canine (13) was displaced by the cyst to the roof of maxillary sinus. (Fig.3)



Fig. 3:- OPG: Unilocular well defined radiolucency.

Provisional diagnosis was dentigerous cyst and incisional biopsy was done. Histopathology revealed a 3-4 layered thick stratified squamous non-keratinized epithelium with fibrous connective tissue wall and few inflammatory cells suggestive of dentigerous cyst. A surgical approach was planned immediately after investigating blood reports. The standard treatment for dentigerous cyst of the maxilla involving the sinus is enucleation and extraction of the associated teeth via Caldwell-Luc procedure under general anaesthesia, which was followed for the present case (Fig.4). A crevicular incision was given from 21 to 16 region with an anterior releasing incision and the mucoperiosteal flap was reflected. The extraction of 53, 54, 55, 14, 15 and 16 followed by cystic enucleation and then by the extraction of 12 and 13 was done. There were no post-operative complications and the wound healed uneventfully. The excisional biopsy that was performed intraorally consisted of a bit of soft tissue specimen measuring about 2.5cm x 2cm x 1cm (Fig.5) which was grayish white in color and firm in consistency has been taken for processing. The H&E stained soft tissue section shows cystic lining epithelium with underlying connective tissue.



Fig. 4:- Surgical approach.

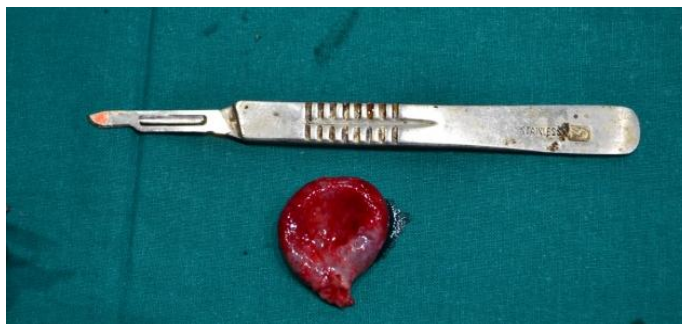


Fig 5:- Specimen.

Epithelial lining showed 3-4 layer reduced enamel epithelium. Cystic epithelium is transforming into hyper-proliferative epithelium with characteristic clear cells, intermediate cells and epidermoid cells arranged in nests pattern Fig 6 (a), (b). These cells are also present in connective tissue interspersed with chronic inflammatory cells. The findings were suggestive of Mucoepidermoid carcinoma arising from the lining of the dentigerous cyst. The radiographic evidence of lesion involvement of maxillary sinus along with histological features of clear cells, intermediate cells and epidermoid cells made us suggestive of “Dentigerous cyst transforming in to Mucoepidermoid carcinoma”. 2weeks post-operatively, a temporary obturator was given. Patient was on regular follow-up for the last 18 months without any evidence of recurrence.

Discussion:-

Dentigerous cyst can be defined as one which encloses the crown of an unerupted tooth and is attached at the cemento-enamel junction. It is a cyst arising by separation of follicle from the anatomical crown of an unerupted tooth within the jaws. The substantial majority of dentigerous cyst involves the mandibular third molars and the maxillary permanent canines, followed by mandibular premolars, maxillary third molars and rarely third molars¹⁶.

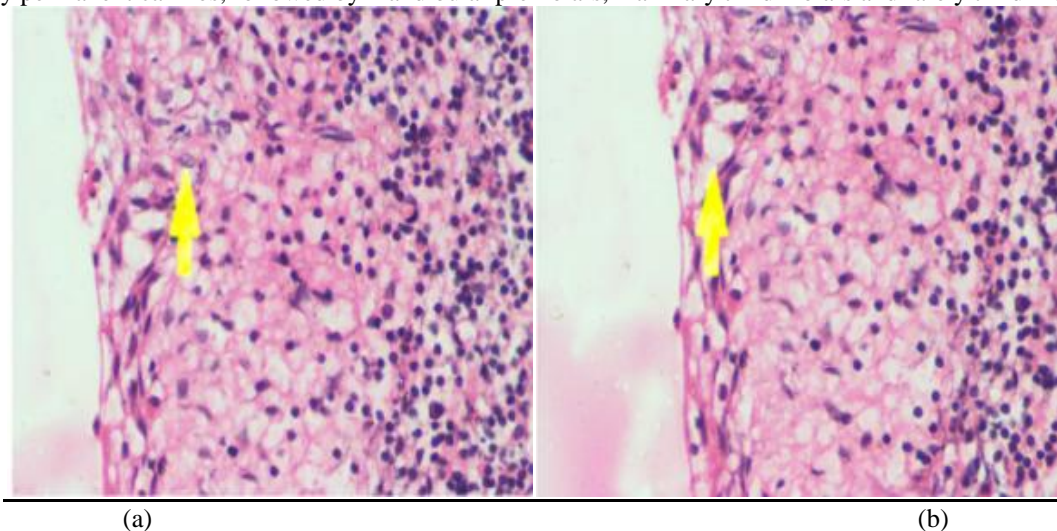


Fig. 6:- (a) H&E stained photomicrograph showing cystic spaces & mucous cells at 40x

(b) H&E stained photomicrograph showing cystic spaces, mucous cells & epitheloid cells at 40x

Unlike other odontogenic cysts, epithelial cells that line the lumen of dentigerous cyst arise from reduced enamel epithelium origin and possess an unusual ability to undergo metaplastic transition into other epithelial cell types such as SCC, MEC, AOT and ameloblastoma. Radiographically, the dentigerous cyst presents as a well-defined unilocular radiolucency, often with a sclerotic border. Since the epithelial lining is derived from the reduced enamel epithelium, this radiolucency typically surrounds the crown of the tooth.

Malignant tumors such as mucoepidermoid carcinoma (MEC) of the paranasal sinuses are uncommon, constituting less than 1% of all malignancies. MEC is a malignant epithelial neoplasm composed of both mucus secreting cells and epidermoid type cells in varying proportions. It was first studied and described as a separate entity by Stewart et.al in 1945 (Rajendran et al¹⁷). Central mucoepidermoid carcinoma is an uncommon lesion in children and only 7

cases in patients younger than 16 years were reported according to the literature revealed. After a systematic review of its histology and degree of differentiation, the WHO Classification in 1991 recommended that the term “mucoepidermoid tumour” be changed to ‘mucoepidermoid carcinoma’. It comprises 5-10% of all salivary gland neoplasms and accounts for 13% of all malignancies occurring in the maxillary sinus.

The criteria for diagnosis of intraosseous mucoepidermoid carcinoma are the following:

- (1) Presence of intact cortical plate
- (2) Radiographic evidence of bone destruction
- (3) Histologic confirmation
- (4) Positive mucin staining
- (5) Absence of a primary lesion in the salivary gland and
- (6) Exclusion of an odontogenic tumor.

The pathogenesis of this tumor is still unknown. Four possible origins have been considered:

- (1) Entrapment of retro-molar mucous gland within the mandible, which later undergoes neoplastic transformation;
- (2) embryonic remnants of the submandibular and sublingual glands trapped within the mandible during development;
- (3) neoplastic transformation and invasion from the lining of the maxillary sinus; or
- (4) neoplastic transformation of mucous-secreting cells from the epithelial lining of dentigerous cyst associated with impacted teeth^{16,17}.

MEC arising in the maxillofacial region can have its origin from maxillary sinus lining or central MEC arising from within the bone or from the minor salivary gland. In this case, histological picture is suggestive of MEC arising from the cystic lining of dentigerous cyst. Maxillary sinus malignancies are very difficult tumours to treat and traditionally have been associated with poor prognosis with the main reason being the close anatomic proximity of the nasal cavity and paranasal sinuses to the vital structures such as skull base, brain, orbit and carotid artery. This complex location makes complete surgical resection of sino-nasal tumours a challenging task.

Conclusion:-

The prognosis for most histo-pathologically diagnosed dentigerous cysts transforming into mucoepidermoid carcinoma is excellent as it is in early stage; also recurrence is a rare finding. It can be therefore suggested that after thorough enucleation of dentigerous cyst, microscopic examination of the entire specimen by the pathologists must be done to determine transformation into mucoepidermoid carcinoma though rare, still possible.

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RESEARCH ARTICLE

RIGHT OF CHILDREN TO MAINTENANCE.3

Dr. Mufti Imran Ul Haq Kilyanvi Bukhari.

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Abstract

The United Nations being a humanitarian organization demonstrates its love and care for children by observing Children's day in October each year. It is also the day of the declaration of the rights of children. Islam however, had drawn attention towards children's affairs over fourteen hundred years ago. Even before a child is born, its rights are given to it by Islam. At several places in the Quran and Sunnah, Allah and his Prophet (Peace Be Upon Him) have talked and acted lovingly towards the children, it thus leaves no possibility of Islam not paying particular attention to clearly charting out the rights of children. The following article brings out references from the Quran and Sunnah to highlight these very rights and collects them in one script of writing.

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Introduction:-

The United Nations demonstrates its care and love for children by observing Children's Day in October each year; it is also the day of the declaration of the rights of children.

On its part, Islam had aroused attention to children's affairs over fourteen hundred years ago. Even before the child is born, its right are given to it by Islam.

Islam draws the picture of childhood as a lovely world with happiness, beauty, dreams, love and amazing experiences. The verses of the Qur'an disclose Allah's love for children, for instance:

لَا أَقْسِمُ بِهَذَا الْبَلَدِ وَأَنْتَ حَلٌّ بِهَذَا الْبَلَدِ وَوَالِدٍ وَمَا وَلَدَ.

"I swear by this city. And you are a dweller in this city. And by the begetter and that which he begets" (90:1-3)
(The Urdu has..... by the parents and the child.) Children are mentioned as glad tidings.

يُزَكِّرُنَا إِنَّا نُبَشِّرُكَ بِغُلَامٍ اسْمُهُ يَحْيَى.

"O Zakariyya! We give you the glad tidings of a son. whose name is Yahya." (19:7)

They are described as joy to the eyes:

رَبَّنَا هَبْ لَنَا مِنْ أَزْوَاجِنَا وَذُرِّيَّتِنَا قُرَّةَ أَعْيُنٍ.

"Our Lord! Grant us coolness of eyes from our wives and our offspring". (25:74)

They are ornaments of the world's life

الْمَالُ وَالْبَنُونَ زِينَةُ الْحَيَاةِ الدُّنْيَا.

"Wealth and sons are an adornment of the life of this world". (18:46)

Allah's Messenger □ presented a child before us as if it was paradise. He said:

الاطفال دعاميص الجنة.

Children are butterflies of Paradise:-

The Prophet ﷺ also said "If infants were not fed mother's milk, old people did not bow down in worship and animals did not graze in the open fields then surely you would be seized with a severe chastisement."

He was delivering the sermon one Friday when Sayyidi-na Hasan and Husayn (may Allah be pleased with them), his two grandsons came running towards him — now stumbling, now checking them- self! — He paused and descended from the pulpit and lovingly picked them up. Returning to the pulpit, he continued his sermon, saying, "O people! Your wealth and your children are mean of your madness. I saw my two children fall and stumble as they scampered to me and could not restrain myself, so got down and carried them in my arms." (1)

He crawled on all fours once having placed Sayyidina Hasan (may Allah be pleased with him) on this back. Someone remarked, "Son, you are riding the best of transport." The Prophet ﷺ remarked, "The rider too is the best of riders!" (2)

If, during the salah he led, he heard a child weep, he would shorten the salah. He would remark, "I do not like to cause distress to its mother."

As he was passing by Sayyidah Fatimah's (may Allah be pleased with her) house one day, he heard Sayyidina Husayn (may Allah be pleased with him), crying. He was moved on hearing him cry, so went into the house and reprimanded Sayyidah Fatimah ﷺ: "Do you not know that I am pained when he weeps?"

He stressed the importance of teaching and training children, saying:-

لأن يؤدب الرجل ولده خير من أن يتصدق بصناع.

"A man's training his child is better than giving a sa' in charity." (3)

The Prophet & said:

ما نحل والد ولدا من نحل أفضل من ادب حسن.

"No father has given his children anything better than good manners" (4)

A child will be brought up according to the training the head of the family imports to him. Training a child is to groom him practically on how to live his life and to learn this in the household atmosphere.

Someone has said very rightly:

إذا كان رب البيت بالطبل ضاربا فلا تلم الا ولاد على الرقص.

"When the Lord of the house beats the drum then blame not the children if they dance." (5)

We must act as we want our children to do because their hearts are like mirrors that reflect our actions. (6)

ينسأه إذ قلبه كالجواهر الصافي
فسوف يأتي به من حفظه وافي

والطفل يحفظ ما يلقي اليه ولا
فانقش على قلبه ما شئت من خير

If a father does not train his children well then they cannot be trained all their lives. Sayyidina Umar Ibn Khattab (may Allah be pleased with him) said:

من لم يؤدونه الشرع الا ادبه الله.

"If anyone does not train the child according to Shari'ah then Allah also does not train him". (7)

Children's livelihood as outlined by the Qur'an:-

In pre-Islamic days, it was common to kill children. Poverty and want drove men to eliminate their children. It is more commonly believed that people killed their daughters, but the truth is that they killed all their children because they could not feed them. The Qur'an forbid people to do this repulsive thing. It said in very clear terms:

وَلَا تَقْتُلُوا أَوْلَادَكُمْ مِنْ إِمْلَاقٍ نَحْنُ نَرْزُقُكُمْ وَإِيَّاهُمْ.

"And slay not your offspring for (fear of) poverty. We provide sustenance for you and for them". (6:151)

وَلَا تَقْتُلُوا أَوْلَادَكُمْ خَشْيَةَ إِمْلَاقٍ نَحْنُ نَرْزُقُهُمْ وَإِيَّاكُمْ إِنَّ قَتْلَهُمْ كَانَ خِطْئًا كَبِيرًا.

"And slay not your children for fear of poverty we provide for them and for you. Surely the slaying of them is a great sin" (17:31)

The Qur'an instructs man not to appropriate to himself the duty of apportioning to sustenance. Instead, he must behave as Allah's slave. He had been like that child, helpless but the Provider of sustenance fed him he grew up to the age when he considers his children a burden. He still gets his provision. Then why should he forget the Provider and present himself as one.

This also rejects the ideas of family planning. The propagators are worried about population growth, who kill children of other people that their own bellies should be full. They collect large sums of money from western, irreligious forces on the pretext of promoting family planning. They inspire fear of having to spend on children but put down the drain millions of rupees on advertisement in newspapers and television. They could instead spend this money on the poor people's children.

Their falsification:-

The promoters of family planning have made bold to distort verses of the Qura'n in their book, Islam mein bachchon ki nighaydasht sponsored the ladarah Itfal Aqwaam Muttahidh, Pakistan. The writer has cited the verses 29 of an-Nisa; 151 of al-An'um, 32 of al-Ma'idah. He has presented only that piece of verse 151 of al-An'um as serves his purpose: **وَلَا تَقْتُلُوا أَوْلَادَكُمْ** (slay not your children). The words following it are a slap on the face of family planning **خَشْيَةَ إِمْلَاقٍ** (for — Fear of — porverty). The words following that are also dropped **نَحْنُ نَرْزُقُهُمْ وَإِيَّاكُمْ** (We provide sustenance for you and for them).

If they had not imitated the Jews and Christians in concealing the words of the Qur'an, they would have lost tens of thousands of dollars. Instead of a pat on the back, they would have faced the anger of their bosses. May Allah protect us! Aameen.

Children's live hood as outlined by the Hadith:-

Hadith is really a commentary and an explanation of the Qur'an. The Prophet ﷺ was asked once, "Which sin is the greatest?" He said: "That you ascribe partner to Allah although He has created you." The man asked, "What is the next great sin?" The Prophet ﷺ said:

ان تقتل ولدك خشية ان ياكل معك.

"That you kill your son for fear that he will share your food with you" (8)

So we see that right from the pre-Islamic days, people killed their children lest they become an economic burden. The Qur'an and Hadith state that people killed their children for food because the father is responsible for it and he shuns that responsibility and commits the crime of killing children.

Mother's love lauded:-

Allah has created love of children in His creatures so that they become active to raise them. In the field of maintenance, Allah has given excellence to offspring alike of men and animals that parents are made responsible for their feeding, and this carries on till they grow up. As they gain adulthood, parents let them earn their own livelihood; examples of this are seen in all creatures from birds to men. The Prophet is quoted in a Hadith:

**جعل الله الرحمة في مائة جزء فامسك عنده تسعة جزء، وانزل في الارض جزء واحد
فمن ذاك الجزء يتراحم الخلق حتى ترفع فرس حافرها عن ولدها خشية ان تصيبه.**

"Allah has divided mercy into a hundred parts, but retained ninety-nine of them with himself. He has sent one on earth because of which the creatures show mercy to each other so that even the mare lifts up her hoof away from her young colt lest she should trample on it" (9)

"The Prophet ﷺ demonstrated this mercy to his Sahabah. Sayyidina Umar (may Allah be pleased with him), reported that some captives were brought to the Prophet ﷺ, a woman among them. Her breasts were full with milk which overflowed. She ran till she found a child among the captives. She picked it up and suckled it. The Prophet ﷺ said, "Do you suppose that she would throw her child in fire?" They replied. "No. As long as she can, she will not throw her child in the fire." He said, "Allah is more Merciful to His slaves than this woman is to her son. (10)

These Aha'dith show us how a mother loves her children instinctly.

Equal division among offspring:-

The Prophet ﷺ also taught us that we should not discriminate among our children causing distress to the deprived. All our children have equal rights. If we prefer one over the others, they will suffer from inferiority complex. Sayyaidina Nu'man Ibn Baheer's (may Allah be pleased with him) father gave his son a slave as a gift and requested the Prophet ﷺ to bear witness for that. He asked him if he had given a similar gift to all his children to which he said that he had not. The Prophet ﷺ said to him, "Take back your gift." (11)

Some ulama' in their commentary on this Hadith have said that it is mustahab to show equality to all children. equality must be shown in the love bestowed on them, too. Sufyan Thawri رحمه الله عليه said that in bestowing them,

too Sufyan Thawri رحمه الله عليه said that in bestowing gifts, sons and daughters are equal. They are not treated as in inheritance but some maintain that, like inheritance, gifts will also be divided in the ratio 2:1 between sons and daughters, this is what imam Ahmad and Ishaq said.(12)

Initial upbringing:-

For the first two years a new-born is fed mother's milk. Allah has given this command and the duties of parents in the Qur'an. He says:

وَالْوَالِدَتُ يُرْضَعْنَ أَوْلَادَهُنَّ حَوْلَيْنِ كَامِلَيْنِ إِمَّا أَرَادَ أَنْ يُنْتَمِ الرِّضَاعَةُ ۖ وَ عَلَى الْمَوْلُودِ لَهُ رِزْقُهُنَّ وَ كِسْوَتُهُنَّ بِالْمَعْرُوفِ ۖ لَا تُكَلَّفُ نَفْسٌ إِلَّا وُسْعَهَا ۚ لَا تُضَارُّ وَالِدَةٌ بَوْلِدًا ۚ وَلَا مَوْلُودٌ لَهُ يَوْلَدُهُ ۖ * وَ عَلَى الْوَارِثِ مِثْلُ ذَلِكَ ۚ فَإِنْ أَرَادَا فِصَالًا عَنْ تَرَاضٍ مِنْهُمَا وَ تَشَاوُرٍ فَلَا جُنَاحَ عَلَيْهِمَا ۚ وَإِنْ أَرَدْتُمْ أَنْ تَسْتَرْضِعُوا أَوْلَادَكُمْ فَلَا جُنَاحَ عَلَيْكُمْ إِذَا سَلَّمْتُمْ مَا اتَّيْتُمْ بِالْمَعْرُوفِ ۚ وَ اتَّقُوا اللَّهَ وَ اعْلَمُوا أَنَّ اللَّهَ بِمَا تَعْمَلُونَ بَصِيرٌ

And mother shall suckle their children for two whole years, for one who desires that (he suckling be completed. It is upon the father to provide them their maintenance and clothing according to known fair manner. No one is burdened save to his capacity. A mother shall not be made to suffer on account of her child. Nor a father on account of his child. A similar duty to wean (the child) by mutual consent and consultation, there is no blame on them. And if you wish to have your children nursed, there is no blame on you provided you hand over according to known fair manner what you settled to offer (the fostering mother). And fear Allah, and know that Allah is seer of what you do.] (2:233)

The commands in the verse for the newborn are:-

1. Mother should suckle their babies for up to two years, parents can stop that before this period.
2. If mother's milk is not enough then the parents may feed the child other milk or nourishment. The father will bear the expenses.
3. It is a grave sin to stop feeding the child mother's milk without any reason or out of stubbornness (with-in the period).
4. It is wajib for a mother to suckle her child without remuneration as long as she is married to its father or, if he has divorced her, as long as she observes the iddah. It is unlawful to claim wages from her husband while she gets maintenance from him.
5. Maintenance of the wife, or expenses towards the iddah, is wajib on the husband. If she is divorced and the period of waiting (iddah) is over and the child's suckling period is not yet over then the mother can demand a reasonable remuneration for suckling the child. If she feeds milk to the child without charging wages then it is better and there is a reward in that for it because it is an obligation on her as its mother.
6. The maximum period of suckling according to Shari'ah is two years. It is a sin to exceed that period without reason.
7. When there is an excuse that is exempted.
8. In exceptional cases when a child does not take other nourishment or there is risk of his becoming seriously ill, there is permission to feed milk for two and a half years. This is one report from Imam Abu Hanifah.
9. It is allowed if father and mother agree to wean their child before two years are over provided that is beneficial to the child, or, at least, it is not harmful to it.
10. If the child would suffer if he is weaned before two years are over.
11. All other expenses, besides milk, like medical treatment, etc. are the father's responsibility.
12. When the child's mother is divorced, the father's responsibility for the child does not cease with wages for milk but he is responsibility for all other expenses of the child.
13. According to sharp ah, the mother has the right to raise up the child after she is divorced.
14. A boy of seven years or a girl of nine years must remain in their mother's care. If their mother marries a stranger and he is not willing to raise her children then the maternal grandmother or great grandmother has the right to raise them.
15. If they are not there then the right lies with the paternal grandmother, then paternal aunt and then maternal aunt. But, mother's right is foremost. "A mother shall not be made to suffer on account of her child, nor a father.(13)

The period of suckling:-

The authorities differ on the limit of suckling period. Most of them regard it as two years among them Abu' Hanifah رحمه الله عليه and Abu' Yusuf رحمه الله عليه. Imam Maalik رحمه الله عليه puts it at two years and two months, but many views are attributed to him including as much time after two years "as the child needs to accustom to another diet. (14)

Imam Abu Hanifah رحمه الله عليه puts it at two and a half years. Imam Jafar رحمه الله عليه holds that the total period of suckling is three years. (15)

Most go by the command of Allah:-

وَالْوَالِدَتُ يُرَضِّعْنَ أَوْلَادَهُنَّ حَوْلَيْنِ كَامِلَيْنِ (آيت: ٢: ٢٣٣)

Sayyidina Ibn Abbas (may Allah be pleased with him) reported:

قال رسول الله صلى الله عليه وسلم لارضاع الا ما كان فى الحولين. (16)

Imam Abu Hanifah رحمه الله عليه said about:-

وَالْوَالِدَتُ يُرَضِّعْنَ أَوْلَادَهُنَّ حَوْلَيْنِ كَامِلَيْنِ.

That it is not necessary that “حولين” stops suckling after that. He argues that “فان” in the following phrase فَاِنْ أَرَادَ فِصَالًا عَنْ تَرَاضٍ مِنْهُمَا وَتَشَاوُرٍ فَلَا جُنَاحَ عَلَيْهِمَا hints at the post-period of Hence, “حولين” this verse does not mean to limit the period of suckling but specifies that the maintenance of the woman who breast-feeds is wajib on the father for up to two years, not more than that.

Many deduce from the following verse too:-

وَحَمْلُهُ وَفِصَالُهُ ثَلَاثُونَ شَهْرًا.

“And the bearing of him and the weaning of him is thirty months” (45:15)

The minimum pregnancy period is six months, the remaining twenty-four months is of weaning. (17)

Imam Abu Hanifah رحمه الله عليه also bares his argument on this verse.

The outlier of Hidayah states that Allah has mentioned two things in this verse that pregnancy and weaning are over a period of thirty months.

كالاجل المضروب للدينين.

However, there is a drawback against pregnancy period. Sayyidah Ayshah (may Allah be pleased with her) said:-

لا يكون الحمل اكثر من سنتين قدر مايتحول ظل المعزل. (18)

This means that the maximum period of pregnancy is two years.

However, Mawlana Anwar Shah Kashmiri رحمه الله عليه said:

وما اجاب به صاحب الهداية ههنا فهو ركيك جداً.

The saying of Sayyidah Ayshah (may Allah be pleased with her) necessitates abrogation of the verse which is not correct. Hence, the correct answer is what Allamah Nusfa has suggested, "The meaning of حمله is to carry in the arms." Thus, the verse implies that the period of suckling is thirty months which is also the time during which the infant is carried in the arms. If an objection is vaied that the verse 46:15 ووضعتها كرها clearly refers to carrying the child in the womb, so حمله وفصاله would also refer to the same thing, then our contention is that the verse describes the different stages of hardship the mother undergoes for the sake of her child they are:

حملته امه كرهاى فى البطن (carrying it with hardship in her womb).

ووضعتها كرها (and brings it forth with hardship).

وحمله على الايدى (and carries it in the arms)

وفصاله (and weans it).

However, there is no doubt that the majority opinion is preferable.

Allamah Ibn Nujaym writes: (19)

ولا يخفى قودة دليلهما.

This is because the words:

وَالْوَالِدَتُ يُرَضِّعْنَ أَوْلَادَهُنَّ حَوْلَيْنِ كَامِلَيْنِ.

Continues to the verse:

لِمَنْ أَرَادَ أَنْ يُتِمَّ الرَّضَاعَةَ.

Which indicate that there is no sucking once (two years are over. However, someone might put forth the words:

فَاِنْ أَرَادَ فِصَالًا عَنْ تَرَاضٍ مِنْهُمَا وَتَشَاوُرٍ فَلَا جُنَاحَ عَلَيْهِمَا.

Indicate that weaning is subject to mutual constant after “حولين” so that if consent is not forthcoming suckling might carry on even after “حولين” (two years). The answer to this is that the consent is required within two years (to wean.). It is not needed thereafter when it is specified that breast-feeding will cease.(20)

Maintenance of child after two years:-

The father is responsible to arrange for the child's nourishment when he is able to subsist on that. Hence, he labours all day to earn that and at night lends a hand to the child's mother in looking after it and coaxing it to go to sleep. He wards off flies and mosquitoes from the child.

ويخشى عليه الثقل من وطاة الذر

and he even fears the ants on its account lest they bite the child. The father and mother say:

اكبادنا تمشي على الارض

وانما اولادنا بيننا

لامتنعت من الغمض

لو هبت الريح على بعضهم

(Our children. Our beloved walk on the earth among us. If the wind blows over any of them, my eye lashes will stop flickering.) (21)

This is a natural love that Allah has put into His creatures. The father is driven by those sentiments to see that his children have the best possible nutrition. However, that is not all. For, Islam has made it mandatory on the father to provide maintenance to his children who are minors. Allamah Ibn Nujaym رحمه الله عليه has written:

ولطفله اى تجب النفقة والسكنى والكسوة لولده الصغير الفقير.

“It is wajib on the father to provide maintenance to his minor and needy children” (22)

This maintenance is wajib for the father singly. No one else will share his responsibility for that. The Arabic word طفل (minor children) means a child from birth to end of adulthood. The qualification of فقير (poor) implies that if the child is rich then its maintenance is not wajib on its father, even if his property is a piece of land, sheets of cloth or drapery, etc. The father can sell those things and spend on the child. If both father and son are poor then according to Khassaf, the father will beg of people to meet the expenses of his child (ren). Or, the Bayt al-Maal is responsible for such people. (23)

It is stated in the marginal notes to al-Bahr-ur-Ra'iq by Allamah Ibn Aabidayn Shami رحمه الله عليه that the father will not compel his daughter to earn a livelihood. (24)

وليس له في الأنثى ذالك.

And Ramli has said that if the girl can sew or spin yarn and earn therefrom then it is not wajib for the father to provide them maintenance on condition that they earn according to their need. If their earnings fall short of their needs then it is wajib on the father to make good the short fall. (25)

Inheritance for children:-

Sometimes, a man has good intentions in protecting his legacy for his children through a waqf, or endowment. However, very soon the family grows, there are more children and relatives. Supervisors and guardians then quarrel with each other. This intensifies into rivalry and enmity. Many Muslim countries have therefore, abolished family endowments. Allah alone knows what His slaves require and what is good for them. The shares of inheritance are defined and Shari'ah has taken upon itself the responsibility of its apportioning because the rightful should get his right and people might not go to the courts and antagonize each other after death in their family member.

Allah, the Majestic, the Glorious has said:

يُوصِيكُمُ اللَّهُ فِي أَوْلَادِكُمْ لِلذَّكَرِ مِثْلُ حَظِّ الْأُنثِيَيْنِ ۚ فَإِن كُنَّ نِسَاءً فَوْقَ اثْنَتَيْنِ فَلَهُنَّ ثُلُثَا مَا تَرَكَ ۚ وَإِن كَانَتْ وَاحِدَةً فَلَهَا النِّصْفُ.

“Allah enjoins you concerning your children, for the male is the share equivalent of that of two females: but if there be more than two (or two) females only (and no male child) them for then are two thirds of what (the deceased) left, and if there be only one female child (and no male child), for her is the one half” (4:10)

Orphaned grandson's share:-

The family laws in our country have apportioned a share for an orphan from the legacy of his paternal grandfather. But, it is not reasonable that the right of other heirs is taken away to accommodate the orphan by force. This is altering the laws of Allah and His Prophet ﷺ. Rather, the options indicated by Allah and His Messenger ﷺ for the orphan must be observed. The laws of Shari'ah for maintenance must be enforced. To set aside a share for the orphan in the legacy of a paternal grandfather is not the solution of his problems because everyone does not leave behind a legacy when he dies. Also, it is not normally so abundant as to support an orphan. But, if the laws of maintenance are enforced then every helpless and poor orphan will have his difficulty removed. The Qur'an, the

Ahadith and the scholars have explained exhaustively the issues of maintenance. We present some here. A similar duty is upon the heir). (2:233)

وَعَلَى الْوَارِثِ مِثْلُ ذَلِكَ.

Thus, if a father dies then it is as wajib on the child's heirs to provide maintenance to his mother or wet-nurse during his suckling period. They are also responsible for the child's maintenance. (26) Imam Abu Bakr Jassas Razi (d. 370 A.M.) has written in explanation of this verse:

وعلى الوارث مثل ذلك: يعنى النفقة والكسوة وان لا يضاره اذا كانت المضارة قد تكون فى النفقة كما ذالك فى غير ما قل عطفاً على ذالك وعلى الوارث مثل ذالك كان ذالك موجباً على الوارث جميع المذكور وقد روى عن عمر وزيد بن ثابت والحسن وقبيصة بن ذؤيب وعطاء وقتادة فى قوله تعالى وعلى الوارث مثل ذالك، النفقة على الرجال والنساء على قدر مواريثهم وهو قول اصحابنا.

“Feeding and clothing are the responsibilities of heir. He should not cause hardship to women which is through maintenance or otherwise. Since Allah has placed it on the heir then it is necessary for him to observe all that Sayyidina Umar □, Zayd Ibn Thabit □, Hasan □, Qabeesah Ibn Zuwayab عليه رحمة الله and Qatadah عليه رحمة الله explain the verse as speaking of maintenance. Zayd Ibn Thabit □, said that maintenance is wajib on men and women according to inheritance. This is what our (Hanafi) ulama' say. (27)

Imam Fakhruddin Razi رحمه الله عليه has written in his Tafseer (exegetes):-

المراد وارث الاب يجب عليه بعد موت الاب كل ما كان واجباً على الاب ولهذا قول الحسن وقتادة وابى مسلم والقاضى.

“It means the heir of the orphan's father. All that was wajib on the father (in the favour of the orphan) will become wajib on him after the father's death. (28)

Allamah Mahmood Aaloosi رحمه الله عليه has written:-

والمراد بالوارث وارث الولد فانه يجب عليه مثل ماوجب على الاب من الرزق والكسوة بالمعروف ان لم يكن للولد مال وهو التفسير المأثور عن عمرو ابن عباس وقتادة ومجاهد وعطاء وابراهيم والشعبى وعبدالله بن عتبة وخلق كثير.

“The heir refers to the child's heir because all that is wajib on him which is wajib on father. It is feeding and clothing according to law provided the child does not have his own wealth. The exegesis of this verse is known from Sayyidina Umar □, Ibn Abbas □, Qatadah عليه رحمة الله, Mujahid عليه رحمة الله, Ata عليه رحمة الله, Utbah عليه رحمة الله, and many other authorities”

We learn of the following things from the verse and its exegesis by the scholars:-

1. After the death of the father, the heirs will shoulder every responsibility concerning the orphan.
2. The heirs are bound to give the orphan a proper education and training.
3. It is wajib on his paternal grandfather, paternal uncle and elder brothers to give him maintenance for all his needs.
4. His needs for food, clothing and other essentials will be determined according to dictates of Shari'ah in every circumstance. (30)

Maintenance of homeless children:-

Those children who are heirless will be provided maintenance from the Bayt al-Maal. This is what was done also in the initial days of Islam with a child without father or a child found on the thoroughfare. It was brought to Sayyidina Umar □ who would allocate for it a hundred dirham and whatever was needed for its sustenance and other essentials. The child's guardian would collect its stipend every month while the Ameer ul Mumineen would go every year and examine the progress of the child. He would also give instructions for kind treatment to it. (31)

The author ofai-Bahr has defined such a child thus:-

هو فى الشريعة اسم الحى مولود طرحه اهله خوفاً من العيلة او فرار من تهمة الزينة مضية آثم ومحزره غام.

“In Shari'ah, a laqeet is a living child whose people have abandoned it lest they have to raise it or from fear of being scandalized. Those who abandon it are sinners while those who protect it are sinners while those who protect it are worthy of reward” (32)

Such children are the responsibility of the state. This is also confirmed by Sayyidina Umar □ and Sayyidina Ali □ for the Laqeet is a Muslim who is unable to earn a livelihood and neither owns anything nor has relatives. His maintenance is from the state treasury.

ونفقته فى بيت المال، وهو المروى عن عمر وعلى ولاته مسلم عاجز عن الكسب، لا مال له ولا قرابة فاشبه المقعد الذى لا مال ولا قرابة. (33)

“Though it is the responsibility of the state to pick up abandoned children, if anyone does it in his individual capacity then he will earn reward.

ندب التقاطه لما فيه من احبائه وهو من افضل الاعمال.

“It is commendable to pick up such children because their life depends on it. It is the best of deeds (to let someone live).” (34)

In this connection, the very well-known social worker who has devoted himself to service to humanity, Abdus Sattar Edhi performs praiseworthy Tasks. At the Same time as doing other tasks for the common people, he has made reasonable arrangements to bring up heirless, homeless children and to give them maintenance. Innumerable children have been saved from destruction through his efforts.

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RESEARCH ARTICLE

PERCEPTION OF DISASTER PREPAREDNESS AMONG EMERGENCY PHYSICIAN IN INDIA.

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Abstract

INDIA is a country that's more prone to natural Calamities & disasters [UNESCO Bangkok, 2007.] .No city in the India can be said to be free from Risk and Hazard of facing such acts.

Disaster is a serious public health issue and one of man's oldest concerns, and we have no control over when and where they happen, but with proper preparedness and planning for all phases of disaster, we can minimize the consequences of any disaster.

In last few decades frequency and intensity of various disasters have been significantly increasing all over the world.

Emergency physicians are main players in disaster responses but their knowledge levels of disaster medicine and their psychosocial status are not readily available. This study aimed to evaluate psychosocial preparedness levels and training needs of disaster medicine among potential disaster responders and presented a necessity to popularize disaster medicine education

Methodology: This study is a survey based descriptive study, to fulfill this purpose a questionnaire was developed and a pilot study was done to validate the questionnaire. A web link for this survey sent through email, to Emergency department directors of various multispecialty hospitals with accredited emergency department, requesting them to circulate this link among their consultant / postgraduate trainees and residents.

Results: 124 participants from 16 states of India completed this survey over a period of 6 months. Majority of emergency physicians agreed hospitals in India are inadequately prepared to handle various kinds of disasters, whereas around 50% emergency physicians are not aware of either stockpiles of PPE of their hospitals or their adequacy. Where about 44% respondents did not know the access to resources in case of emergency events. Interestingly, in the study; around 83% of Emergency physicians were ready for being trained in the form of classroom teaching despite their busy schedules, which needs to be appreciated properly.

Conclusion : The study results show that there is enormous scope of further improvement in various aspects of disaster preparedness.

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Though in certain areas the result were satisfactory, majority of the results demand improvement. In the current scenario there is a need for an organized effort for the spread of disaster preparedness amongst the Emergency physician in India. As per the outcome of this study ,it can be concluded that perception of disaster preparedness among Emergency physician in India is still an iceberg phenomenon. Thereby emphasizing upon the fact that hospital administration authorities, government and nongovernmental agencies should take a serious note on it and take measures to inculcate awareness among EPs and should treat finding of this study with seriousness and treat it as a base for further studies for improvement in this field.

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Introduction:-

Disaster is a serious public issue and one of man's oldest concerns, and we have no control over when and where they happen, but with proper preparedness and planning for all phases of disaster, we can minimize the consequences of any disaster.

In last few decades frequency and intensity of various disasters have been significantly increasing all over the world.

Floods, drought, hurricanes and other disasters in last few decades have caused severe environment disruption as well as huge economic losses.

Emergency physicians are main players in disaster responses but their knowledge levels of disaster medicine and their psychosocial status are not readily available. This study aimed to evaluate psychosocial preparedness levels and training needs of disaster medicine among potential disaster responders and presented a necessity to popularize disaster medicine education.

The term DISASTER originated to the French word "Desastre" which is a Combination of two words 'des' meaning bad and 'aster' meaning star. Thus the term refers to 'Bad or Evil star'.¹

Disasters usually defined as "A serious disruption in the Functioning of the community or a society causing wide spread material, economic, social or environmental losses which exceed the ability of the affected society to cope using its own resources"^{1,2,3}

A disaster is a result from the combination of hazard, vulnerability and insufficient Capacity or measures to reduce the potential chances of risk.³

If we think back around 25-30 years, we rarely heard about a major disaster happening, now think in the present context, today it seems like every time we turn around there has been another major disaster.

As an aftermath of the 9/11 World Trade Center, USA event, Attack on Indian parliament on December 13, Indian Ocean tsunami on 24th December 2004 ,Mumbai attack 2009 & Recent Tsunami in Japan , and several incidence like that around the world and in India, has given a new management of disasters on a large scale, hence the importance of disaster management and disaster preparedness become quite evident even to the layperson.

India is one of the most disaster prone countries of the world^{4,5}.

It has had some of the world's most severe droughts, cyclones, chemical disasters, mid-air head-on air collisions, and earthquakes, rail and road accidents. India is also one of the most terrorist prone countries.

According to EM-DAT (2011) report Indian subcontinent acquired fourth position in term of reported disaster, after Philippines, USA, and China.

India is a huge country and it holds as many variations in religion, language, customs, art and cuisine as it does in topography and hence the disasters vary in their type and magnitude as well.

Disasters are typically viewed as low probability yet high impact events. Although various definitions have been used, a disaster is frequently viewed as a situation in which the number of patients presenting to the medical facility within a given time period exceed the ability of the hospital to provide care without external assistance. As such, the definition is institution specific, and therefore preparedness must be likewise institution specific.

The term “hospital preparedness” is a catch-all phrase, covering a multitude of inter-related areas of medical and non-medical disaster management. Although the Joint Commission on Accreditation of Healthcare Organizations (JCAHO) mandates specific standards for hospital preparedness, in many institutions these standards never extend beyond the written page. Hospital preparedness is focused on either natural or unintentional man-made mass casualty events such as accidents, earthquakes, tornadoes, commercial building collapse, and airline or school bus accidents.^{5,6}

Almost 85% of India’s area is vulnerable to one or multiple hazard. Of the 28 states and 7 union territories, 22 are disaster-prone. It is vulnerable to wind storms spawned in the Bay of Bengal and the Arabian Sea, earthquakes caused by active crustal movement in the Himalayan mountains, floods brought by monsoons, and droughts in the country's arid and semi-arid areas. India has also become much more vulnerable to tsunamis since the 2004 Indian Ocean tsunami.⁷

The size and location of our country gives us:

- i. 57% of landmass prone to earthquakes (high seismic zones III–V)
- ii. 68% of the total area are vulnerable to drought.
- iii. 12% to floods.
- iv. 8000 kilometers’ long coastline with an average of two cyclone seasons.

India is a country that’s more prone to natural Calamities & disasters [UNESCO Bangkok, 2007.] .

There is no city or district or village in India can be said to be free from Risk and Hazard of facing such events, hence the responsibility of the people functioning with government as well as private organizations increases manifold.

Beside as citizen of a country which has high risk of both natural and manmade disaster, we all have a certain amount of responsibility for facing and overcoming the socioeconomic as well as psychological impact of these incidents.

The medical impact of any major disaster is felt by the emergency physician and associated staff located within the geographical zone facing the event; they are first people to deal with victims of any disaster, especially Emergency Physician who has to lead a team in such kind of event and facing all challenges along with playing an important role in all phases of disaster, pre-disaster, in-disaster, and post-disaster.

Emergency medical services and emergencies physician should play a leadership role in preparedness activities such as training and education, development of performance metrics, establishment of memorandum of understanding (MOUs), and others planning.

So it is very important that most of the physicians have a good understanding about disaster preparedness. This study aimed to understand what is the current perception about disaster preparedness amongst the Emergency physicians of India.

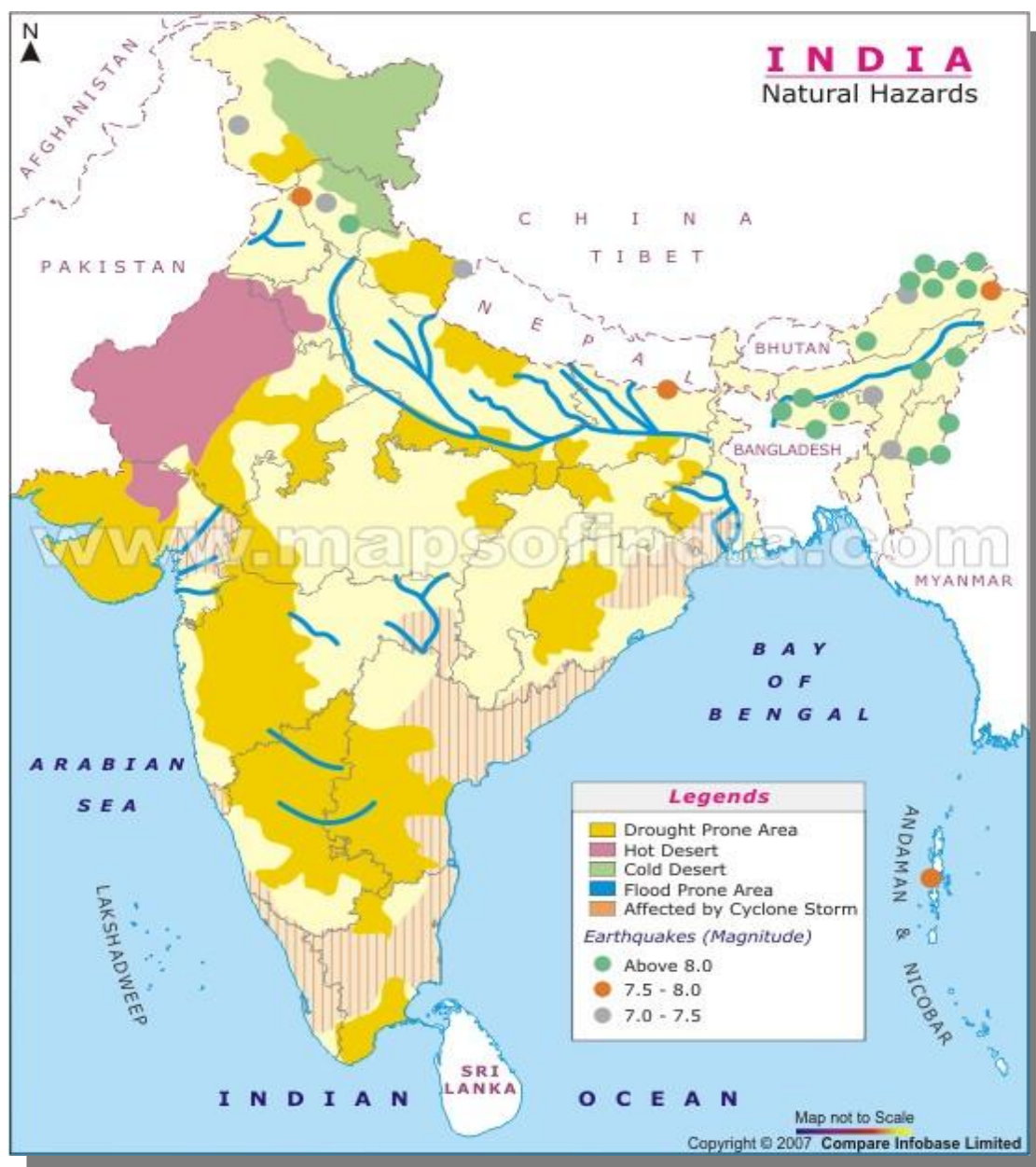


Fig 2.1:- showing Natural disaster prone zone in India.

Source: <http://www.mapsofindia.com>

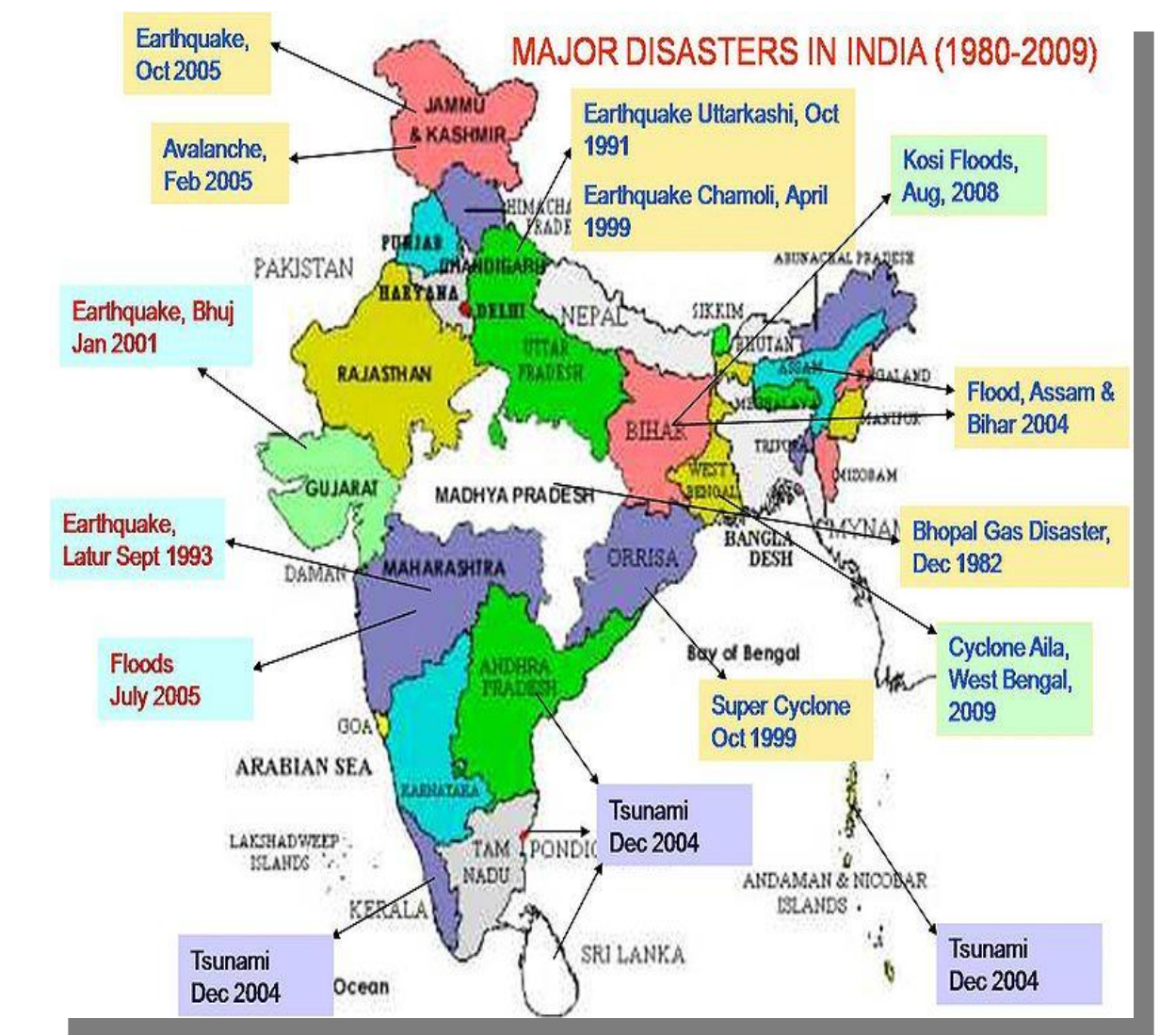


Fig.2.2:- major disasters in India in last 3 decades

Literature Review:-

Online databases were used to identify relevant studies for this Literature review,

The search was limited to human and English language. Search was restricted between the years 1995 to 2015 in order to provide only recent evidence. No previous study was identified on Emergency Physician's perception about disaster preparedness in India. Though no similar study was identified in the Indian context, the above search identified about 6 trials on nurse's perception, physician perception and preparedness done globally in last two decades, that was relevant to our topic. Also some textbooks and material from the internet were studied for the purpose.

A study was done to investigate Emergency Physicians' knowledge about the Superfund Amendments and Reauthorization Act (SARA) Title III legislation, passed by the United States Congress in 1986, and to determine the factors contributing to their level of preparedness in dealing with patients exposed to toxic chemicals. In this study, author found that out of 187 directors of EDs in New York, only 33% of the directors had heard about SARA. Despite the finding that some emergency physicians are involved in community preparations, two main problems persist in planning a medical response to environmental emergencies. First, the ED directors generally are unaware of the legislation that mandates these preparations and are not interacting with community planners. Second, there is not full involvement by the local EDs in the Local Emergency Planning Committee's planning efforts.⁸

A review of literature suggests that nurse's response to working during a bioterrorism event is not well described. The limited number of studies regarding nurses' concerns, fears, and anxieties is focused on nurses' experiences in natural disaster or war situations. Additional studies are needed to validate the appropriateness of applying findings from disaster response studies to bioterrorism events. During bioterrorism events, nurses will be expected to provide physical care and emotional and psychological support for victims and victims' families. Realistic bioterrorism plans should incorporate strategies to support nurses and address their physical, psychological, and emotional issues. Strategies to optimize safe working conditions and minimize psychological trauma such as technical training regarding bioterrorism agents and debriefing opportunities should be included.⁹

In another study author ranked nurses higher than physicians regarding their knowledge and skills in dealing with mass casualty events involving children, but the level of knowledge for mass casualty events involving children was low in both.¹⁰

Another study supported the above by concluding that considerably more training and information are needed to enhance preparedness for frontline healthcare workers (nurses) as important members of the disaster response community.¹¹

The importance of physician involvement and leadership in crisis preparedness is recognized by Uddin *et al* (2008). Their literature also suggests that few physicians are adequately trained to practice effectively in a large-scale crisis situation.¹²

In a recent pilot study on disaster training program to improve emergency physicians' preparedness. They concluded that Disaster preparedness is an essential area of clinical competence for EPs. Participation in a yearlong pilot training program demonstrated a statistically significant increase in cognitive competence among a pilot sample of Emergency Physicians.¹³

The above literature review clearly identifies that, very few physicians are adequately trained to practice effectively in a large-scale crisis situation.

Methodology:-

Participants:-

Participants for this study was consultants Emergency department, emergency department residents, and postgraduate trainees of emergency medicine from varies multispecialty, tertiary care level hospitals of India which have a fully functional Emergency department.

Those doctors who were working other than emergency department were excluded from this study.

Questionnaire:-

This study is a survey based descriptive study, to fulfill this purpose a 13 items questionnaire was developed including three demographic questions,(from question 1 to question 3) which included participant's gender, age, their professional role, residing state, contact number, and their emails.

Question 5 to question 10 had multiple sub-questions under their heading.

The questionnaire was divided in two part for understanding the following

1. Familiarity regarding specific term and activities in different emergency preparedness categories.
2. Participant's experience and training.

Survey:-

This survey was conducted through a web based survey site <https://www.surveymonkey.net>, a web link <https://www.surveymonkey.com/s/SK3BPRK> sent through email, to Emergency department directors of varies multi specialty hospitals with accredited emergency department, requesting them to circulate this link among their consultant / postgraduate trainees and residents. A multi stage sampling method was use to fulfill the objective of this survey. Apart from web based collection, some sampling was done through printed questionnaire, which was manually entered in website for proper analysis.

Questions Wise Result:-

124 participants from 16 states of India completed this survey over a period of 6 months.

99 respondents participated through an online survey tool and 25 participants were directly administered printed questionnaire.

An accurate estimate of the response rate is difficult to assess, as it was not possible to ascertain, what proportion of emergency physician have indeed been exposed to the email invitation to participate in this survey.

We excluded 10 responses from the final analysis as these were incomplete.

Key demographic characteristics (Q.No.1-3) of the respondents are detailed in following tables.

Table 5.1:- Representing gender distributions.

Gender			
Answer Options	Male	Female	Response Count
Numbers of respondents	86	28	114

Table 5.2:- Representing distributions of participant's age group

Age(in years)							
Answer Options	25-30	30-35	35-40	40-45	45-50	>50	Response Count
Number of participants	55	15	16	12	9	7	114

Table 5.3:- Showing distributions in work experience

Work Experience in Emergency department(in years)						
Answer Options	0-1	1-5	5-10	10-15	>15	Response Count
Number of respondents	31	46	25	12	0	114

Table 5.4:- Distributions in professional role

Professional role						
Answer Options	Consultant	Associate Consultant	Junior Consultant	PG Student	EMO	Response Count
Number of participant	24	8	9	69	3	113

Table 5.5:- Distribution of states

States	A.P	Asm	Bh	DL	chg	Jh.	Kar	krl	MP	MH	OR	PN	RJ	TL	UP	WB	RC
NOR	6	4	4	18	7	1	11	8	2	9	1	2	2	9	3	27	114

A.P-Andhrapradesh;Asm-Assam;Bh-Bihar;Chg.-Chandigarh;Del-Delhi;Jh.-Jharkhand;Kar-Karnatka;krl-kerala;MP-MadhyaPradesh;MH-Maharashtra;OR-Orisha;PN-Punjab;RJ-Rajsthan;TL-TamilNadu;UP-UttarPradesh;WB-WestBengal

RC-Response Count;NOR-number of respondents

Among the respondents 76.11% (86) were male and 23.89% (27) were female.

48.67% (56) were aged between 25-30 years,

5.31% (6) were aged more than 50 years, rest of the respondents were aged between 30 to 50 years.

Of the 114 respondents, majority 61.06% (69) were postgraduate emergency medicine residents, while 21.24 % (24) were working as consultant(s) in emergency department. and a small portion of respondents were working as junior consultants 7.96%(9), Associate consultant 7.08%(8) and 2.65%(3) working as emergency medical officer.

Majority of respondents 40.35% (46) had working experience (in the emergency department) between 1-5 years. 33.67%(33) had only 0-1 year working experience; while there is 13.27%(13) respondents had 5-10 years of experience, and only 4.27%(4) had more than 10 years of working experience in the emergency department .

Q.4: Familiarity with Emergency Preparedness Terms & Activities.

Table 5.6:- showing details percentage of familiarity with term and activities of emergency preparedness.

Answer Options	Not familiar	Somewhat familiar	Familiar	Very familiar	Rating Average	Response Count
Familiarity with triage in large scale emergency event	2	33	49	30	2.94	114
Assess and Respond to site safety issue for self, co-worker,& victim during a emergency event	10	26	49	27	2.83	112
Please provide an assessment of your overall familiarity with response activity/preparedness in the case of a large scale emergency event	7	36	60	11	2.66	114
Your hospital preparedness for responding to a large scale emergency event	10	40	50	14	2.60	114
Overall familiarity with sign/symptoms of exposure to different biological agent/contagious disease outbreak	18	38	47	8	2.41	111
Familiarity with emergency operations plan(EOP) in your hospital	19	56	32	6	2.22	113
Familiarity with incident command system(ICS)	27	26	36	24	2.50	113
Familiarity with rapid assessment of victim of emergency event	4	15	56	38	3.13	113
Familiarity with your facility's/community quarantine process	31	50	24	7	2.06	112
Use of all type of communication devices(fax/email/satellite ph.)	6	27	22	59	3.18	114
answered question						114
skipped question						0

The first question in this section was on *familiarity with triage in a large scale emergency event*. 42.98% (49) showed that they are familiar with triage, and 28.95% (33) respondents denoted that they are somewhat familiar ,26.32%(30)respondents replied that they are very familiar with triage in large emergency event while, only 1.75%(2) were not aware about triage in large emergency event.

Out of 114 respondents 43.75 %(49) denoted that they are familiar with *self safety and site safety issues*, 24.11%(27) think that they are very familiar with safety issues and a small percentage 8.93%(10) said that they are not familiar with site safety issue for self and co-workers during emergency events.

When asked about *familiarity with sign and symptoms of exposure to different biological agent/contagious diseases outbreak*, 42.34%(47) said that they are familiar, and 34.23% (38)replied “somewhat familiar”, while 16.22%

(18) were not familiar and only a small group of respondents 7.21% (8) were confident and very familiar with this issue.

Majority of respondents 49.56% (56) said that they are somewhat familiar with their hospitals emergency operation plan (EOP), 28.32% (32) respondents think that they are familiar, and 16.81% (19) respondents denoted that they are not familiar with their hospital's EOP. Only 5.31% (6) were confident in their knowledge regarding hospital EOP.

In assessment of another important component of emergency preparedness the *Incident Command System (ICS)* we looked at familiarity with this concept and found that 31.86% (36) respondents were familiar with ICS, and only 21.24% (24) of total participants said that they are very familiar with ICS, while a significant percentage of 23.89% (27) of the respondents denoted that they do not know about ICS. When asked about *familiarity with communication devices*, majority of 51.75% (59), shows that they are very familiar with use of the all kind of communication devices.

Q.5: Rate your responses to your hospitals preparedness.

Table 5.7:- details percentage of hospital preparedness to various kind of emergency events.

Answer Options	No Preparedness	Inadequately Prepared-ness	Adequately Prepared-ness	Do not know	Rating Average	Response Count
Do you think hospitals in India are prepared for disaster	17	87	6	4	1.97	114
Do you think your hospital is prepared for disaster.	11	67	29	6	2.27	113
What is level of preparedness of your hospital for bioterrorism?	72	20	6	16	1.70	114
What is level of preparedness of your hospital for radiologic/nuclear incident?	71	18	6	19	1.76	114
What is level of preparedness of your hospital for infectious disease outbreaks?	16	47	44	7	2.37	114
What is level of preparedness of your hospital for flood/cyclones?	16	37	51	10	2.48	114
What is level of preparedness of your hospital for earthquakes?	19	31	49	15	2.53	114
What is level of preparedness of your hospital for in hospital terrorist attack?	34	35	28	17	2.26	114
What is level of preparedness of your hospital in hospital hazmat accident?	25	44	31	14	2.31	114
answered question						114
skipped question						0

This section deals with hospital preparedness to various kind of emergency events.

Majority of respondents 76.32%(87) agreed that hospital in India are inadequately prepared to handle disasters; 14.91% (17) said that there is no preparedness amongst Indian's hospital to face large scale emergency events; while a small percentage 5.26%(6) of respondents think that hospital in India are adequately prepared for disasters.

When asked about participant's hospital disaster preparedness majority 59.29%(67) agreed that their hospital is inadequately prepared to face large scale emergency events, and 25.66%(29) of respondents think that their hospital is adequately prepared, while only 9.73% (11) think that their hospital is not prepared for facing disasters.

Majority of 63.16 % (72) and 62.28 % (71) emergency physician thinks that their hospital is not prepared for bioterrorism activities and radio/nuclear incident respectively, and only a small percentage 5.26 % (6) respondents said that they are adequately prepared for both events.

44.74 % (51) emergency physicians think that their hospitals are adequately prepared for flood/cyclone and 42.98%(49) replied that their level of preparedness is adequate for earthquakes.

Q. 6: Rate your responses to Accessing critical resources & Stockpiles.

Table 5.8:- Represents emergency physicians knowledge about availability of various stockpiles in their hospital

Answer Options	No	Yes	Don't know	Rating Average	Response Count
Do you know about your hospital's stockpiles of personal protective equipments?	58	35	21	1.68	113
Do you know about your hospital's stockpiles of personal protective emergency medication/antidotes?	54	42	18	1.69	113
Do you know about your hospital's stockpiles of oxygen/electricity backup?	39	47	27	1.89	113
Do you think your hospital has adequate stockpiles?	33	25	56	2.21	113
Do you know that, during an event, where to quickly access up-to-date resources about specific (Radiological, Chemical, Nuclear, Biological, and Explosive) agents	51	32	31	1.83	113
answered question					114
skipped question					0

Large number of emergency physician 50.44%(58) and 46.90% (54) are not aware about stockpiles of personal protective equipments (PPE) and emergency medication/antidotes respectively in their hospital, while 30.97% (35) and 37.17%(42) said they know about stockpiles of PPE and medication of their hospital.

When asked whether available stockpiles is adequate or not most emergency physician 49.56%(56) has no idea about this as they replied they do not know, and 28.32% (33) responded that the available stockpiles is not adequate and 22.12%(25) emergency physician thinks that their hospital has adequate stockpiles of PPE and emergency medications. Out of 114 respondents 44.25% (51) replied that they don't know, from where to quickly access up-to-date resources about specific agent (radiological/chemical/biological /explosive) and 28.32 % (32) people know about this, while 27.43 % (31) of emergency physician has no idea about this as their answer was "do not know".

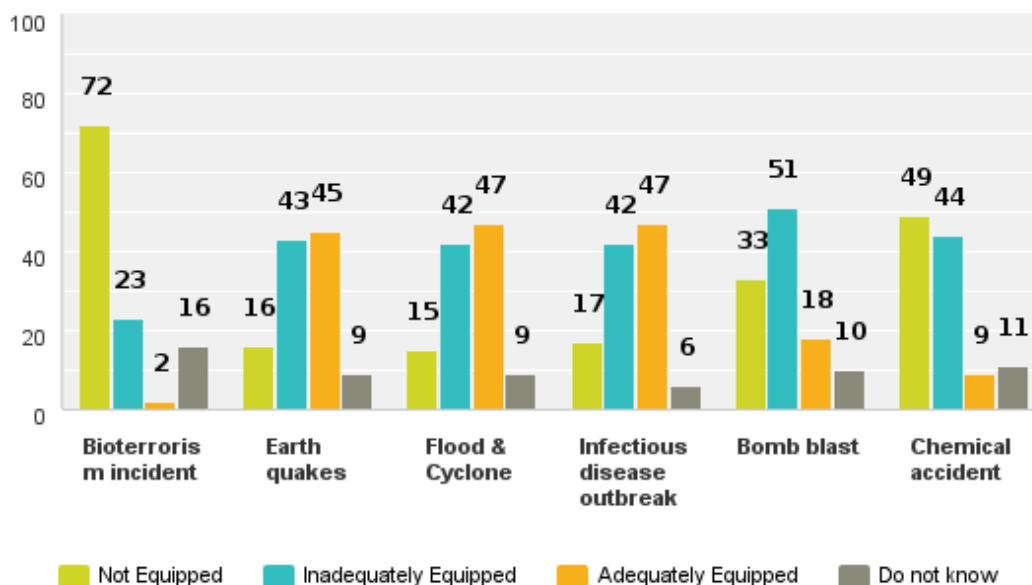
Q. 7: Do you feel adequately equipped to work during various kinds of disaster?

Fig 5.1:- percentage of emergency physician's perception about how well they are equipped, working in different type of disaster.

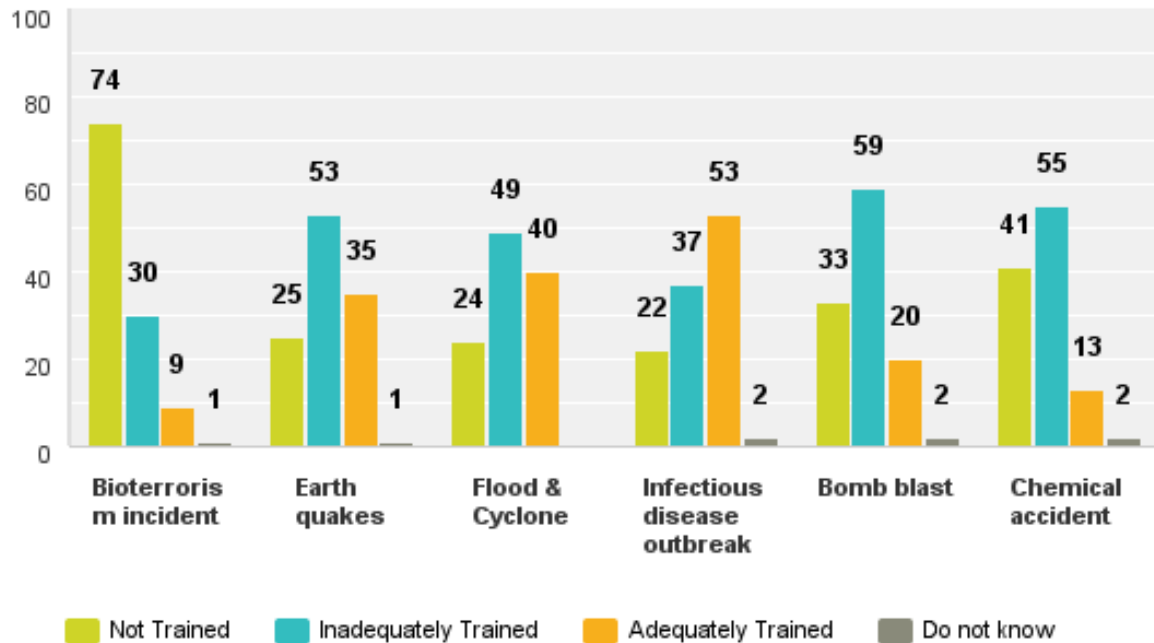
In this section we tried to access that how well Emergency physicians are equipped working in different type of disaster.

A majority of 63.72% (72) thought they are not equipped to work in case of bioterrorism event, while 20.35% (23) thinks inadequately equipped and only 1.77% (2) respondents replied that they feel adequately equipped.

39.82% (45) thought that they are adequately equipped to work in a earthquake event, and 41.59% (47) replied that they are adequately equipped for flood/cyclone.

In case of bomb blast injuries 45.54% (51) and 29.46% (33) of the participants replied that their hospital(s) were 'inadequately equipped' and 'not equipped' respectively, while 16.07% (18) replied that they were adequately equipped. Only 7.96% (9) respondents think that they are adequately equipped to face any bomb blast event.

When asked about an infectious outbreak 41.96% (47) emergency physicians said that they are adequately equipped for working in an infectious outbreak, and 37.50% (42) replied that they are inadequately equipped. A major number of respondents 43.36% (49) replied that they are not equipped for working in a chemical accident, while 38.94% (44) said they are inadequately equipped.

Q.8 : Do you feel adequately trained to work during various kinds of disaster?**Fig5.2:-** shows emergency physician's formal training on different kind of Disasters

Majority of the responding emergency physicians 64.91%(74) and 36.94%(41) have stated that they are not trained for working during bioterrorism and chemical accident respectively. While 26.32% (30) and 49.55%(55) denoted that they are inadequately trained for working in event of bioterrorism and chemical accident.

A small percentage of 7.89 % (9) and 11.71% (13) said they are adequately trained for working during such events. Majority of emergency physician 51.75% (59) said they are inadequately trained for working in a case of bomb blast event, and 28.95% (33) confirmed that they are not trained for working in bomb blast event(s), while 17.54% (20) were confident about the adequacy of their training for working on an event of bomb blast. Regarding training for working during earth quakes and flood/cyclone 46.49%(53) and 43.36%(49) respectively stated that they were not adequately trained.

35.40% (40) emergency physician stated that they are adequately trained for working during flood/cyclone, and 30.70% (35) confirmed their adequate training for working during an earth quake.

46.49 % (53) respondents were confident about the adequacy in their training for working during an infectious disease outbreak.

A significant number of emergency physician 32.46% (37) denoted their training was inadequate for working during an infectious outbreak; while 19.30 % (22) stated that they are not trained for working in such a scenario.

Q. 9 : Rate your responses to your experience & training.**Table 5.9:-** details of emergency physician's work experience in disastrous events and hospital's disaster plan and policies

Answer Options	No	Yes	Rating Average	Response Count
Have you had any training in disaster medical response?	39	75	1.66	114
Have you had any experience in working during mass casualty event?	38	76	1.67	114
Have you had any experience in working during mass infectious disease outbreaks?	75	39	1.34	114
Do you know in case of a disaster who commands the disaster response in your hospital?	42	72	1.63	114
Does your hospital have a plan and policies to respond to a large scale mass causality incident?	35	77	1.69	112
Did your hospital have a fire/disaster drill?	15	97	1.87	112
Have you had any experience in working during mass casualty event?	40	71	1.64	111
Have you been trained on the disaster plan?	47	64	1.58	111
As an emergency physician do you think that you will be legally liable by the Indian legal system for practicing altered level of care because of limited supply of resources, excessive loads of patients, as usually happens during disasters?	50	63	1.56	113
answered question				114
skipped question				0

This section deals with Emergency physician work experience in disastrous events and hospital's disaster plan and policies.

Majority of the respondents 65.79%(75) stated that they have received formal training for working during an external disaster; while 34.21% (39) were untrained.

66.67% (76) emergency physicians had experience working in a mass causality event.

When asked about their experience of working during infectious disease outbreak, majority of 65.79% (75) of the emergency physician stated that they have never worked in an infectious outbreaks.

when inquire about their hospital's plan and policies of mass causality events and incident commander of their hospital 68.75% (77) and 63.16% (72), respectively said that they know about these while 31.25%(35) and 36.84% (42) emergency physicians stated that they are unaware about their hospital's disaster plan and policies and incident commander of their hospital.

A large number of emergency physician 86.61%(97) agreed that their hospital conducts simulation/disaster/fire drill, and 13.39%(15) replied 'NO'. to this question.

Majority of respondent 57.66% (64) had training on hospital disaster plan in their hospital and but a significant percentage 42.34% (47) of emergency physician did not receive any formal training on disaster management plan.

55.75% (63) emergency physician thinks that they are legally liable by "Indian legal system" for practicing altered level of care in emergency events because of limited supply and excessive loads of victims. While 44.25% (50) said they are NOT legally liable.

Q.10 : How far would you be willing to travel if a disaster occurred today?

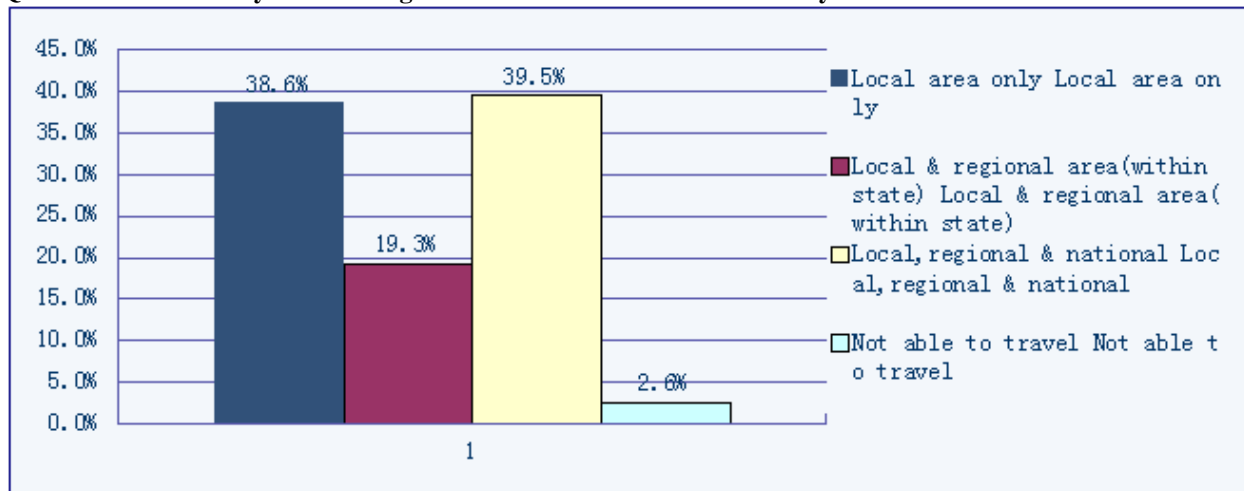


Fig 5.3:- -explaining about willingness of travel to work in case of disaster.

Only 39.50% (45) emergency physician agreed to travel nationwide to work in disastrous event, and 38.60% (44) said they can travel only locally. While a small number of 2.63% (3) respondents were not keen to travel to work in disaster.

Q.11 : How many times in a year your hospital/organization conducts fire drill/simulation drill ?

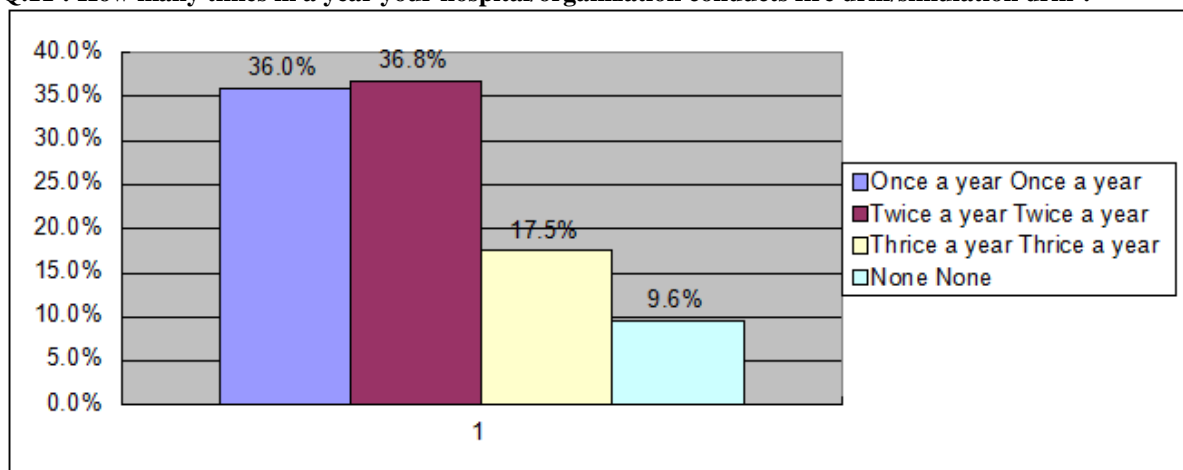
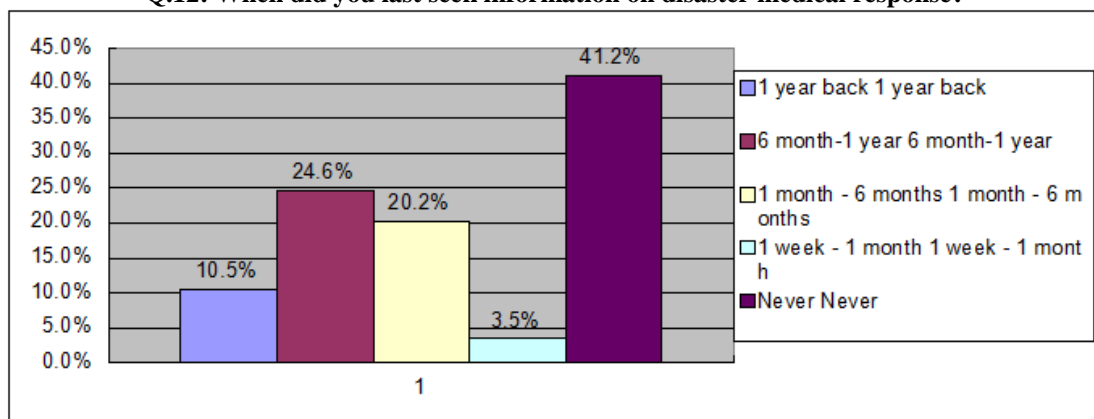
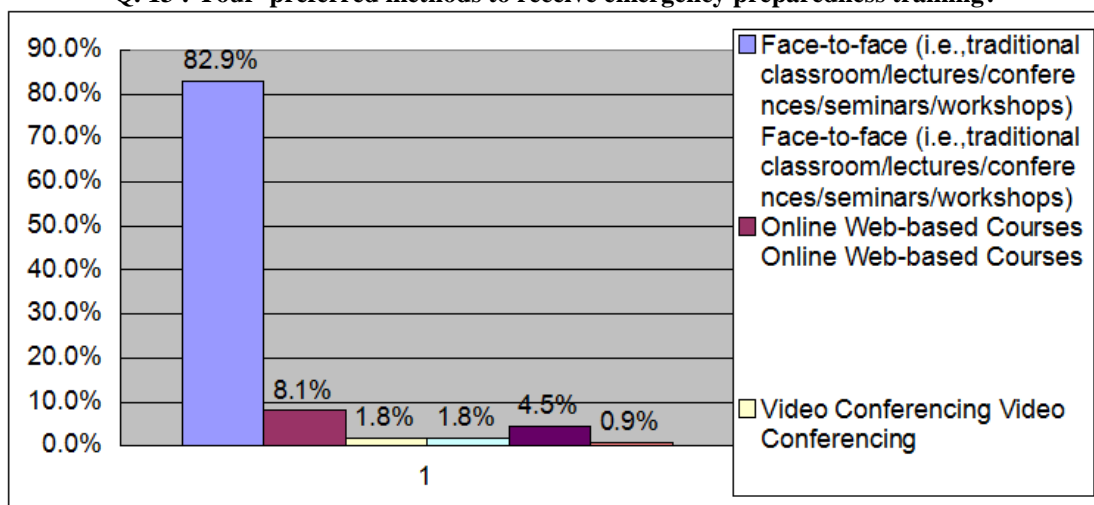


Fig 5.4:- Explaining about simulation drills at participant's hospitals.

Q.12: When did you last seek information on disaster medical response?**Fig.5.5:-** Participant's last gathered information on disaster medical response.**Q. 13 : Your preferred methods to receive emergency preparedness training?****Fig 5.6:-** Emergency physician's preferred method of training

A significant number of 41.23%(47) participants agreed that they never sought information regarding medical disaster response.(fig 5.5)

82.90% emergency physicians are ready to take training on disaster response by traditional face to face method, while others are interested in others methods like web based courses, CV/DVD, or pocket reference books.(fig.5.6)

Discussion:-

Emergency medicine is a broad specialty comprising of disaster medicine as one of its vital branches.

The knowledge of disaster medicine is a paramount importance among the emergency physicians playing the responsibilities of dealing with different kinds of calamities occurring in society.

Natural and human-induced disasters have significantly increased in last few decades which inturn called attention to the fact that emergency physicians play pivotal role in disaster planning.

Though emergency physician are involved in planning, mitigation, response, and recovery aspects of disasters as well as actively participating in actual disaster events, mock drills and further educational opportunities, specific to disaster preparedness; data regarding their knowledge level are not readily available.

This has led to the essence of a pioneer research work to assess **“Perception of disaster preparedness among emergency physician in India”**

This unique analysis of perception of disaster preparedness amongst the working emergency physician was conducted on pan India basis among 16 states over a period of 6 months, with a total 124 participants. The study questionnaires were divided in to several subsections which covered various aspects and facts of disaster preparedness.

The result has revealed several interesting findings. The salient features are as follows.. 26% of emergency physicians were found to be familiar with triage in large emergency event and a significant percentage of them,(around 24%) did not know about ICS ,an important aspect of disaster preparedness. This is the grey zone with enormous scope of improvement.

Majority of emergency physicians (around 76%) agreed that hospitals in India are inadequately prepared to handle various kinds of disasters, whereas around 50% emergency physicians are not aware of either stockpiles of PPE of their hospitals or their adequacy. Where about 44% respondents did not know the access to resources in case of emergency events. Thereby emphasizing upon the fact that hospital administration authorities should take a serious note on it and take measures to inculcate awareness among EPs.

On the other hand, Only 5.26% emergency physicians thought that their hospitals were adequately prepared to handle a bioterrorism activity as well as radio/nuclear incidents, while around 63% had an opposite view on the same.

When we tried to gather information that whether they are adequately equipped or not to work in various disastrous events, majority of emergency physicians (around 64%) stated that their hospitals were not equipped to work in case of bioterrorism event, while around 40% and 42% thought that they are adequately equipped to work in earthquake and flood/cyclone respectively. Only a small percentage ,around 16% emergency physicians thought that their hospitals were adequately equipped to work in case of bomb blast injuries. On the contrary, other participants replied that either they are inadequately equipped or not equipped. The reason for such differences in views should be sorted out in a systematic way and should be addressed in a proper way accordingly. Thus opening an opportunity for further scientific study on the topic.

When enquired about their formal training dealing with various kinds of disasters, only around 8% and 12% were adequately trained in bioterrorism and chemical accident management respectively, where about only 18% were confident about adequacy of their training for working in bomb blast scenarios. Most of these participants were either working as consultant or having 5-10 years of working experiences in Emergency Medicine.

In due course, around 42% of Emergency physicians revealed that they did not receive any formal training on the hospital disaster management plan.

When asked, around 40% Emergency physicians were prepared to travel nationwide to contribute in the management of disastrous events.

Interestingly, in the study; around 83% of Emergency physicians were ready for being trained in the form of classroom teaching despite their busy schedules, which needs to be appreciated properly.

Limitations:-

This study is limited to those who have access to internet because this is a questionnaire based survey .

This survey was limited to major cities of India. There are emergency physicians working peripherally in the interiors of India ,who are possibly working in more ouster conditions and facing natural calamities more closely .It would be really interesting to know their perception on disaster preparedness.

Conclusion:-

Emergency physicians are forefront for dealing with disasters its consequences; they are important cadre of any hospital and society, as they play important role in all phases of disaster.

The study results show that there is enormous scope of further improvement in various aspects of disaster preparedness. Though in certain areas the result were satisfactory ,majority of the results demand improvement. In

the current scenario there is a need for an organized effort for the spread of disaster preparedness amongst the Emergency physician in India. The administrative authorities should treat these results with seriousness and treat it as a base for further studies for improvement in this field.

As per the outcome of the study, it can be concluded that the perception of disaster preparedness in Emergency Physicians of India is still showing an iceberg phenomenon. The level of understanding is still at primordial level, which needs proper standardization to meet the requirements of international standards. This is only feasible by following a proper scientific method to raise awareness among the Emergency Physicians of India & organizing regular mock drills as well as workshops for the expansion of the horizon of their perception about importance of disaster preparedness in India.

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RESEARCH ARTICLE

LA GOUVERNANCE DES DESTINATIONS TOURISTIQUES DANS LA NOUVELLE POLITIQUE TOURISTIQUE MAROCAINE: ENTRE CONSTRUCTION INSTITUTIONNELLE ET REALITE OPERATIONNELLE.

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 2020.

Abstract

Promoted since the early sixties as a priority sector in the strategy of socio - economic development, the Moroccan tourism sector, in view of the achievements and the great potential of tourism of the country and of the important growth of world tourism Will undergo a radical change after the Year 2000 in its development policy by moving from a tourism policy mainly promotional by state to the era of strategy in the framework of ambitious, voluntarist and integrated development visions based on contractualization and public-private partnership. These new visions placing tourism as a driving force in the development of the country have focused on strengthened governance to ensure the steering of the strategy and ensure the effective implementation of strategies and action plans.

The purpose of this article is to present an analytical reading of the new model of tourism governance in Morocco in order to characterize it and to analyze its effectiveness and the main limitations to its operational implementation. On the methodological level, we have mobilized the theories and models of strategic management and the governance of tourist destinations and a documentary analysis backed up by a qualitative study by semi-directive interviews with the main actors of the sector.

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Introduction:-

Depuis que le tourisme s'est confirmé comme un secteur très important pour le développement socio-économique des pays et au vue des perspectives prometteuses de son développement futur les questions du management, de la compétitivité des destinations touristiques a retenue l'attention tant des chercheurs, des professionnels, des gouvernements que des organismes internationaux. L'environnement turbulent (Graulier2000) et concurrentiel (Chazaud, 1999) mais aussi indifférencié (Rauche, 2000) dans le quel s'inscrivent les destinations touristiques ont remis en cause les politiques touristiques classiques basées sur des logiques de promotion et de commercialisation tout azimut et ont imposé aux acteurs et destinations touristiques de réfléchir et de mettre en place de nouvelles stratégies tenant compte de leurs attributs attractifs et des lourdes mutations qui s'opèrent sur le marché. De même, les destinations touristiques étant un complexe d'acteurs autonomes et indépendants du point de vue organisationnel mais en principe fonctionnellement interdépendants dans la chaîne de valeur de production touristique, leur management repose sur une nécessaire mobilisation et coordination d'acteurs à logiques et intérêts divergents. Cette particularité conduit ce management à s'étendre pour s'insérer dans un contexte de gouvernance de la destination (Zhang et Zhu, 2014). Le concept de gouvernance des destinations touristiques malgré sa jeunesse a donné lieu à

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une importante littérature visant à explorer son champ, ses logiques et dimensions (Gerbaux, Boudières et Marcelpoil, 2004; Gerbaux et George-Marcelpoil, 2004; Gerbaux et Moreau, 1996; Gerbaux et Paillet, 1999, François et Marcelpoil, 2006; Marcelpoil, Marcelpoil et Langlois, 2006 ; Bensahel et Vles, 2007). De même, qu'il a donné lieu à des modèles théoriques cherchant à proposer des schémas d'organisation et du management de la gouvernance des destinations touristiques.

Au Maroc le tourisme représenté un secteur fondamental dans la dynamique de développement socioéconomique du pays. Depuis l'année 2000 ce secteur va être érigé en locomotive de développement et une ambitieuse stratégie de développement va lui être consacrée rompant avec les politiques basées sur la seule promotion étatique engagées depuis le milieu des années 60. Cette nouvelle stratégie baptisée l'ère des visions (Vision 2010 et Vision 2020) va miser sur la gouvernance comme pilier du pilotage stratégique et de la mise en œuvre des stratégies et des plans d'action territoriaux qui en découlent.

Notre propos dans cet article est de mener une recherche exploratoire pour tenter d'une part, d'analyser et de caractériser le nouveau modèle de gouvernance des destinations touristiques au Maroc et d'autre part, de jauger de son opérationnalisation et de ses limites à quatre années de l'horizon stratégique retenu. Pour ce faire nous avons d'une part mobilisé les principales théories et les principaux modèles du management stratégique et de gouvernance des destinations touristiques et sur le plan empirique nous avons allié pour la collecte des données l'exploitation de nombreux documents officiels de ce nouveau modèle de gouvernance et les données publiées sur sa mise en œuvre (statistiques, CPN, CPR, PDRT, études, articles de presse et publications dans les sites web des organismes touristiques nationaux et régionaux et bibliographie), une étude qualitative par entretiens semi-directifs, en face à face, auprès d'acteurs du secteur, les rapports des ateliers que nous avons animés au profit de l'AIH et enfin la participation active à diverses réunions, séminaires et manifestations publiques ou privées organisées par les acteurs au niveau national et régional.

La politique touristique au Maroc : des programmes aux Visions:-

Evolution de la politique touristique au Maroc:-

L'analyse du système de management et de gouvernance du tourisme dans la destination Maroc nous oblige à revenir sur l'évolution de la politique touristique dans le pays et sa caractérisation. La Politique touristique est une expression générique qui donne lieu à diverses interprétations. Avec l'évolution du tourisme cette expression a connu aussi une nette évolution dans son contenu. Bécherel (2001) distingue quatre étapes différentes dans la formulation de la politique touristique. Au cours de la première étape, la politique touristique était orientée vers la «promotion» à travers la mise en place d'attractions publicitaires par les autorités touristiques. Avec l'affirmation du secteur touristique comme secteur économique significatif et porteur, la politique touristique s'est orientée plus vers l'optimisation du potentiel de production en multipliant les investissements dans l'offre d'hébergement et dans les infrastructures de bases et les actions de marketing et de promotion. Ces actions étaient généralement portées par les autorités nationales, régionales et locales faute d'un secteur privé fort ou à cause de son désintéressement. Dans cette deuxième étape, la politique touristique s'est orientée ainsi plus vers «la promotion et le produit». L'intensification de la concurrence sur le marché touristique mondial a amené les concepteurs de la politique touristique se concentrer sur la recherche de l'amélioration de la compétitivité du secteur en créant un cadre statutaire destiné à gérer, contrôler et améliorer la qualité et l'efficacité du secteur et à protéger les ressources. Actuellement, dans un marché touristique hyperconcurrentiel (Chazaud, 1999) et très évolutif tant de point de vue de l'offre que de la demande, la seule politique touristique basée sur la paire «promotion et produit» n'est plus suffisante pour assurer la compétitivité touristique. La stratégie et la planification stratégique deviennent des éléments clés de la politique touristique. Dans cette quatrième étape, les destinations doivent réfléchir en termes de stratégie, la politique doit être conçue de manière à attirer les marchés identifiés, à créer de la valeur à travers l'innovation, à encourager les partenariats entre les acteurs et à coordonner les actions et les initiatives. En outre, de nos jours, les parties intéressées et impliquées dans la gestion du secteur sont nombreuses et le sujet est bien plus vaste. «Compétitivité», «Viabilité» et «Durabilité» sont devenues les mots-clés de la vision moderne de développement du tourisme. Dans ce sens, La politique touristique a un double objectif : apporter un maximum de bénéfices aux intéressés d'une ville, d'une région ou d'un pays, tout en minimisant les impacts négatifs. Dans ce registre, Goeldner, Ritchie et McIntosh (2000) nous proposent une définition satisfaisante de l'expression «politique touristique»: «un ensemble de réglementations, règles, directives, objectifs de développement/de promotion et stratégies permettant de définir le cadre dans lequel sont prises les décisions collectives et individuelles qui affectent directement le développement du tourisme et les activités quotidiennes pour une destination donnée ». Ainsi, pour ces auteurs, le rôle le plus important de la politique touristique est de s'assurer qu'une destination a une idée claire de ce vers quoi elle se dirige ou de ce

qu'elle cherche à devenir à long terme. En parallèle, elle doit avoir l'ambition de créer un climat favorable à la collaboration entre les nombreux acteurs du secteur touristique. Cela sous entend qu'une politique touristique efficace se doit de viser à offrir aux visiteurs des expériences de qualité, profitables aux parties intéressées de la destination tout en évitant de compromettre son intégrité sociale, culturelle et environnementale. Bécherel, sur la base de ces nouveaux enjeux a déterminé cinq domaines propres à la politique touristique de ce «nouvel âge» du tourisme : La politique de communication, la politique de produit, la politique environnementale, la compétitivité : qualité et efficacité, la stratégie: positionnement et ciblage.

La politique touristique marocaine n'a pas échappé aux mêmes règles d'évolution développées ci-dessus. En effet, l'activité touristique au Maroc a débuté certes sous le protectorat français mais c'est après l'indépendance du royaume que le secteur, sous l'impulsion des organismes internationaux (BM, ONU), va être retenu comme priorité de développement socioéconomique du pays. La politique touristique marocaine va connaître deux principales phases dans son développement : la phase dites des programmes, allant de 1965 à la fin des années 90 caractérisée par au début un engagement total de l'Etat assurant le leadership du secteur et ensuite par son désengagement au profit du secteur privé tout en maintenant le rôle d'orientation. Et la phase dite des «visions» à partir de l'année 2001. En effet, l'attrait touristique du Maroc et ses caractéristiques géographiques, historiques et culturelles ainsi que le succès du tourisme international dans certains pays du pourtour de la Méditerranée vont être à la base de l'importance accordée au tourisme dans la stratégie de développement du pays au milieu des années soixante. C'est ainsi que le secteur va être érigé en priorité national dans les plans de développement successifs allant de 1965 à la fin des années 90. Cette période a été marquée par une politique touristique active, où le secteur public jouait un rôle dynamique en investissant massivement dans le secteur et en le réglementant. L'effort d'investissement et d'aménagement va se concentrer en premier lieu sur des zones prioritaires choisies en fonction de leur forte attraction touristique (Tanger, Al Hoceima, Restinga, Smir et Agadir) pour lesquelles des sociétés d'aménagement spécialisées vont être créées, puis sur les villes impériales disposant d'un patrimoine culturel ancestral (Marrakech, Rabat, Fès et Mekhnès). Cet effort va s'accompagner aussi par une action massive en matière de promotion. Les pouvoirs publics vont aussi par le biais d'organismes publics et semi-publics, tels que l'Office National Marocain du Tourisme (ONMT) et la Caisse de dépôt et de gestion (CDG), procéder à la création des premières unités d'hébergement dans les régions sous-équipées ou déshéritées. Un effort considérable a été aussi accordé à la variable ressource humaine avec la création des établissements de formation professionnelle dédiés au secteur. Les années 70 ont encore marqué cette orientation en instituant un code d'investissement spécifique au secteur dans lequel l'Etat a mis en place un dispositif de mesure incitant le privé à investir dans le secteur (prise en charge et garanti sous forme d'exonérations fiscales et de facilités de financement par le Crédit Immobilier et Hôtelier (CIH) jusqu'à près de 75 % du coût des investissements des opérations à caractère touristique). La décennie 80 va connaître, dans un contexte d'application du programme d'ajustement structurel, une orientation vers le développement des équipements hôteliers de catégorie moyenne de façon à répondre à la diffusion du tourisme de masse et une recherche de la diversification de l'offre et des produits par l'intégration de certains type de tourisme jusqu'alors marginalisés, tels que le tourisme familial, le tourisme de montagne et de sports d'hiver, le tourisme rural, le tourisme de nature, etc...

Le début des années 90 a été marqué par une crise d'essoufflement du tourisme (crise du golf, attentas terroriste de Marrakech). Dans ce contexte, une importante étude portant sur l'analyse du secteur et la mise en place d'une stratégie nationale d'aménagement touristique a été réalisée par le ministère. Cette étude, constatant le caractère vétuste du produit Maroc et la faiblesse de la qualité de service dans la grande majorité des établissements d'hébergement et son incapacité à rivaliser avec les nouvelles destinations touristiques va tracer les grandes lignes d'une stratégie de développement du tourisme au Maroc pour les années 1995-2010 et suggérer un programme d'action qui devraient l'accompagner. La mise en application des premières mesures du plan d'action de l'étude de la stratégie nationale d'aménagement touristique va rapidement donner ses fruits et l'année 2000 s'est soldé par des résultats forts encourageants (près de 4 300 000 non résidents dont 2 275 000 touristes étrangers de séjour, sans compter les croisiéristes, 22,3 millions de nuitées dont 50 % dans les hôtels classés). En 2001, la Fédération du tourisme, membre de la Confédération générale des entreprises du Maroc (CGEM), et le gouvernement adoptent une stratégie de développement touristique sous la forme d'un contrat-programme 2001-2010, « le tourisme : une vision, un défi, une volonté », qui actualise et développe la stratégie de développement 1995-2010. Cette nouvelle stratégie va rompre avec la politique touristique jusqu'alors poursuivie et va faire entrer le pays dans une nouvelle ère de stratégie de développement touristique baptisée l'ère des visions.

Les visions « 2010 » et « 2020 » de développement touristique : une lecture croisée:-

Au vue de l'importante croissance que connaissait le tourisme mondial et possédant des atouts naturels importants et un patrimoine culturel riche et diversifié et conscient du rôle déterminant du secteur dans le développement économique du pays, le secteur touristique au Maroc va, à partir de l'année 2001, connaître un grand changement dans sa politique de développement et de promotion à travers le délaissement des politiques promotionnelles ponctuelles en faveur de la mise en place d'une vraie stratégie de développement touristique volontariste susceptible de déclencher une dynamique de développement durable et intégrée. Cette nouvelle ère baptisée l'ère des « Visions » va marquer clairement le point de rupture avec le passé en matière de politique touristique marocaine en érigeant le tourisme en priorité économique nationale et en dotant le secteur d'une stratégie prospective avec une vision à long terme confortée par des contrats programmes chiffrés et détaillés. La première vision dite « Vision 2010 », lancée lors des premières assises du tourisme tenues à Marrakech le 10 janvier 2001 et marquée par la signature de l'Accord Cadre entre le Gouvernement et la Confédération Générale des Entreprises Marocaines (CGEM) et par la suite de la signature de son Accord d'Application, contractualisant ainsi l'engagement des deux parties (publique et privée) de mettre en œuvre le dispositif stratégique de la nouvelle politique touristique, a défini des objectifs très ambitieux aussi bien en termes quantitatifs que qualitatifs.

Tableau n°1:- Les objectifs de la vision 2010

- | |
|---|
| <ul style="list-style-type: none"> - Entrées aux frontières : 10 millions - Capacité hôtelière additionnelle : 160 000 lits - Nuitées touristiques dans les hôtels classés : 50 millions - Taux d'occupation : 70 % - Emploi additionnel : 600 000 emplois - Contribution du tourisme au PIB: elle devrait progresser en moyenne annuelle de 8.5%, ce qui la porterait à près de 20% à l'horizon 2010 |
|---|

Source : Vision 2010, Ministère du tourisme marocain.

Cette vision a permis une grande avancée dans le secteur touristique national et a pu insuffler une grande dynamique surtout en matière d'investissement et de réalisation de stations touristiques. Toutefois les objectifs en matière d'arrivées touristiques et de taux d'occupation n'ont pu être pleinement atteints. Cela est dû à la conjugaison de plusieurs facteurs d'ordre conjoncturel et structurel (MEF-DÉPF, 2011, p:12). Sur le plan conjoncturel, le secteur touristique national a subi de plein fouet les effets négatifs de la crise économique qui a affecté les principaux pays émetteurs depuis 2008. Sur le plan structurel, malgré les réalisations physiques en matière d'investissement touristique, l'offre touristique marocaine reste peu diversifiée et spatialement concentrée dans quatre grandes villes (Marrakech, Agadir, Casablanca et Tanger) qui concentrent à elles seules plus de 65% de la capacité totale d'hébergement. De plus, l'offre balnéaire demeure prépondérante dans la palette touristique globale avec une part de 23%, ce qui place le Maroc en concurrence directe avec les pays compétitifs du pourtour méditerranéen.

A la vue des résultats optimums de la Vision 2010, le pays a décidé de reconduire sa stratégie touristique à travers la « Vision 2020 », qui mise sur un développement touristique plus équilibré et diversifié, capable de renforcer l'offre de chaque région et qui serve de mécanisme de croissance pour ces dernières. En effet, en construisant sur les acquis et les enseignements de la Vision 2010, ainsi que sur les perspectives d'évolution du marché touristique mondial de la prochaine décennie, le but central de la Vision est d'assurer le rôle du tourisme comme moteur et levier principal du développement du pays. Pour cela la vision a défini comme ambitions de hisser la destination Maroc en 2020 parmi les 20 premières destinations touristiques mondiales tout en la positionnant comme une destination de référence en matière de développement durable sur le pourtour méditerranéen.

Tableau n°2:- Les objectifs de la vision 2020

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| <ul style="list-style-type: none"> - Doubler les capacités d'hébergement, avec la construction de 200.000 nouveaux lits. - Atteindre la barre des 20 millions de touristes issus de différents marchés émergents - accroître les recettes touristiques pour atteindre environ 140 milliards de dirhams en 2020 - accroître de deux points la part du PIB touristique dans le PIB national, - Porter la part du tourisme interne à 40% des nuitées. - Créer 470000 nouveaux emplois - et monter à la 20ème destination touristique mondiale. |
|--|

Source: Vision stratégique de développement touristique « VISION2020 », Contrat- programme 2011-2020, 30 Novembre 2010.

Comme feuille de route pour la réalisation de cette ambition et de ce positionnement, la vision cherche à offrir aux touristes des expériences variées, authentiques, d'un niveau de qualité répondant aux meilleurs standards internationaux, qui valorisent les ressources naturelles et socioculturelles du pays à leur juste potentiel, qui permettent aux populations locales de participer pleinement à l'activité touristique et qui soient respectueuses de leur environnement et de leur qualité de vie. Pour ce faire, aux deux principaux pôles d'activité que sont Marrakech et Agadir, sont venus s'adjoindre plusieurs nouveaux pôles touristiques, dont la montée en charge devrait permettre d'intéresser de nouveaux segments de touristes, de rééquilibrer l'activité touristique et d'en diffuser les bénéfices économiques sur l'ensemble du territoire. La Vision 2020 s'est appuyée ainsi sur une démarche volontariste et intégrée d'aménagement territorial visant d'une part à permettre la mise en valeur des potentialités de chaque région et d'autre part à exploiter toutes les synergies possibles entre les régions, afin que dans leur ensemble, les régions du pays soient plus compétitives et plus visibles sur les marchés internationaux. Cette démarche devrait être rendu possible grâce au concept de territoire touristique (la vision a retenu huit territoires touristiques) défini comme une zone géographique présentant une offre touristique complète, une forte cohérence et identité propres qui en font son originalité, et une masse critique en termes de capacités d'hébergement, de ressources touristiques et de desserte internationale (CPN- Vision 2020, P:11). Pour opérationnaliser la mise en œuvre de cette stratégie, la vision a prévue de manière détaillée et chiffrée un plan de mise en marche de six programmes d'actions **structurants pour un portefeuille diversifié de produits adossé à un dispositif intégré de développement d'un tourisme durable, une promotion et une desserte aérienne en adéquation avec l'ambition des territoires, des mesures de soutien pour une dynamique d'investissement soutenue et durable, un plan de valorisation du capital humain et de formation d'excellence, une stratégie pour la compétitivité du secteur et la professionnalisation des acteurs de la chaîne de valeur touristique et un système de gouvernance claire et adéquat.**

La dynamique créée par ces deux visions a pu rapidement donner ces fruits en permettant des performances remarquables. En effet, les données actuelles montrent une nette progression et amélioration de tous les indicateurs du secteur en perspective de la réalisation des objectifs retenus malgré un environnement géopolitique mondial et régional marqué par les risques et l'incertitude. Ainsi, grâce aux efforts déployés dans les deux visions, le Maroc est classé comme la 26ème meilleure destination mondiale et l'objectif retenu pour 2010 a été atteint à hauteur de 93% (9.3 millions en 2010 de touristes internationaux contre 4.4 millions en 2001) avec une croissance annuelle moyenne de 8.7%, très supérieure à la moyenne mondiale. Cette croissance a continué pour atteindre 10,282 944 millions de touristes internationaux à fin 2015 avec une progression de +2,4% par rapport à 2014. Le secteur a aussi été d'une contribution décisive aux grands équilibres macro-économiques. En effet, entre 2001 et 2010, les recettes touristiques (hors transfert des Marocains Résidents à l'Etranger) sont passées de 29 à plus de 56 milliards de dirhams, soit un taux de croissance de 7.5% par an. Les revenus issus du tourisme sont ainsi devenus la première source de devises du pays, devant les transferts de MRE, et loin devant les autres secteurs de l'économie (phosphate, textile, ...). En 2015, ces recettes en devises générées par les non-résidents ayant séjourné au Maroc (hors transport international) se sont élevées à près de 57,2 milliards de dirhams représentant près de 29% des exportations des biens et services et le solde de la balance des voyages a couvert 24% du déficit de la balance commerciale en 2015. Le secteur représente aussi actuellement le deuxième contributeur au PIB national. En effet, le Produit Intérieur Brut du secteur du tourisme a connu entre 2001 et 2010 une croissance annuelle moyenne de 7.6%, supérieure à celle de l'économie dans son ensemble et a pu gagner 2 précieux points dans le PIB national (8% contre 6% au début de la décennie). En 2015, cette croissance s'est encore confirmée et la part du secteur est passée à 12% du PIB confirmant ainsi l'importance du secteur. Cette dynamique qu'a et que connaît le secteur a eu un impact socio-économique important, puisque le secteur représente désormais près de 450.000 emplois directs, avec une hausse de 40% du nombre d'emplois durant la décennie. Tendance qui s'est confirmée à fin 2015 puisque le nombre d'emplois a dépassé la barre de 500000 emplois directs soit près de 5% de l'emploi dans l'ensemble de l'économie faisant du secteur le deuxième contributeur d'emplois dans le royaume.

Dans le registre de l'hébergement et suite aux efforts entrepris et aux réalisations des deux visions 2010 et 2020, la capacité hôtelière va connaître une nette évolution depuis l'année 2000. Ainsi d'une capacité de près de 93000 lits à la fin des années 90, la capacité litère classée a atteint plus de 230 628 lits à fin 2015.

Tableau n°3:- Chiffres clés du secteur touristique marocain (2015).

Part dans le PIB	Nb. d'emplois	VA	Taux de croissance	Capacité d'hébergement	Croissance des recettes de voyage
12%	1200000 (500000 emplois directs soit près de 5% de l'emploi dans l'ensemble de l'économie.	8%	4%	230 628 lits	5,2%

Source : Ministère du tourisme marocain, 2015.

Somme toute, le tourisme occupe une place de choix dans la structure économique et financière du pays et représente un levier considérable pour l'accélération de la croissance socio-économique. Véritable moteur de croissance, le tourisme impacte pratiquement tous les domaines de l'activité économique du Maroc et exerce ainsi une grande influence sur les autres secteurs de l'économie. Toutefois, malgré ces réalisations, la grille de lecture du secteur touristique marocain manifeste encore quelques retards par rapport aux objectifs et ambitions annoncés et notamment un flagrant manque dans la capacité d'hébergement comparativement aux principaux pays et destinations concurrentes.

Cadre théorique et conceptuel:-

Du management à la Gouvernance des destinations touristiques:-

Comme le rappelle Lozato-Giotard (2003), la filière tourisme est constituée de la production de divers secteurs d'activité constituant une chaîne de fonctions, base de la chaîne de valeur associée au produit touristique. Une destination touristique ne constitue pas, ainsi, une organisation intégrée, mais le regroupement d'un ensemble de professionnels, simultanément autonomes et indépendants. Le management étant défini comme: «la manière de conduire, diriger, structurer et développer une organisation... l'animation d'un groupe d'hommes et de femmes qui doivent travailler ensemble dans le but d'une action collective finalisée » (Thietard et al. 1999), celui de la destination touristique relève des mêmes considérations et suggère ainsi l'analogie entre une destination et une entreprise. Toutefois, compte tenu des caractéristiques des destinations touristiques, ce management présente certaines particularités. La première particularité du MDT provient même du fait de l'existence d'un grand nombre d'acteurs autonomes, privés et publics aux poids, objectifs et intérêts divergents. La deuxième principale particularité réfère à la nécessité de prendre en compte de nombreux autres acteurs animant le territoire de la destination et qui agissent de manière intentionnelle ou non sur de multiples composantes constituant des ressources pour le tourisme et l'attractivité de la destination et donc une grande part du service touristique global lui-même, ou du "bien support" du service touristique, au sens de la théorie des services (Eiglier et Langeard, 1987). Ces acteurs doivent ainsi être sensibilisés et mobilisés pour la réussite de ce management. Ainsi, le MDT exige la prise en compte de la façon la plus opérante de nombreux acteurs fonctionnellement interdépendant mais souvent indépendants du point de vue organisationnel, avec des logiques propres et d'autres parties prenantes souvent non intéressés par l'activité touristique et ainsi dépasser la simple gestion d'une activité économique dans ses fonctions les plus génériques pour embrasser d'autres fonctions liées à la mobilisation, la coordination et l'implication. De ce fait, compte tenu de la grande multiplicité et l'importance des différentes parties prenantes présente sur le territoire de la destination et influençant son développement, le management de la destination n'impliquant que les acteurs touristiques, doit intégrer la dimension coordination entre tous les acteurs et ainsi ce management doit s'étendre pour s'insérer dans un contexte de gouvernance de la destination; celle-ci impliquant tous les acteurs au sein de la destination (Zhang et Zhu, 2014).

De la gouvernance des destinations touristiques:-

Concept ancien (Stoker, 1998), la notion de gouvernance, issue de l'économie institutionnelle, dans les années 1930 où elle désignait le comportement de l'organisation face à un environnement complexe, elle est devenue, depuis les années 1980, une expression qui s'insère dans les sociétés et les économies d'une façon controversée: à la fois une notion instable et en évolution, elle est aussi une pratique managériale (Paquet, 2008), un outil mobilisateur des acteurs (Letourneau, 2009) pour les territoires et les organisations confrontés aujourd'hui à une féroce compétition internationale. Repris d'abord par les politologues le concept de gouvernance exprime l'idée d'une transformation de l'action publique faisant intervenir de nouveaux acteurs et recourant à de nouvelles modalités de coordination (Borraz et Le Galès, 2001), il va connaître une grande évolution en termes des disciplines qui l'étudiaient, telles que

les sciences politiques, le management des organisations (Ruhanen et al., 2010) et la géographie (Leloup et al., 2005) et un foisonnement d'études et de recherches vont lui être consacrées pour en cerner le sens, les caractéristiques et les dimensions. Parmi les nombreuses définitions données à ce concept, celles les plus citées nous retenons la définition fournie par P. Le Gales (1995) qui considère la gouvernance comme « l'ensemble des arrangements et relations, formelles et informelles entre intérêts publics et intérêts privés, à partir desquels sont prises et mises en œuvre des décisions. » et celle de Benko (1992) pour qui « la gouvernance renvoie à des formes intermédiaires de régulation, ni marchandes, ni étatiques, qui articulent intérêts privés et publics, aspects sociaux et économiques. ». En sciences de gestion, la gouvernance a été définie comme « l'ensemble des mécanismes qui ont pour effet de délimiter les pouvoirs et d'influencer les décisions des dirigeants, autrement dit, qui 'gouvernent' leur conduite et définissent leur espace discrétionnaire » (Charreaux, 1997 in Charreaux et Wirtz, 2006), ce que Pérez (2003) résume par « le management du management ». Les systèmes de gouvernance sont alors supposés influencer le processus de création de valeur par le biais, notamment des décisions d'investissement et de répartition de la richesse créée. Dans ce domaine, le cadre théorique dominant la recherche en gouvernance est fortement inspiré des théories contractuelles de la firme, en particulier la théorie de l'agence, la théorie des coûts de transaction et théorie des parties prenantes.

Le tourisme approché comme une filière liant une grande multitude d'acteurs indépendants mais aussi très interdépendants constitue un contexte intéressant dans l'étude de la gouvernance de la destination (Ruhanen et al. 2010). Toutefois, malgré la multiplication d'approches n'a pas pu jusqu'alors aboutir à une définition universelle de la gouvernance des destinations et à un corpus théorique propre bien établi. Cela est dû en grande partie à la diversité des champs de recherche des chercheurs et du caractère des études consistant en grande partie en des études de cas qui ne tiennent compte que des dimensions appropriées au contexte de l'étude en question (Beritelli et al. (2007). De nombreux autres auteurs se sont intéressés à l'étude de la gouvernance des destinations touristiques (Beritelli et al, 2007, Ruhanen et al, 2010, Laws et al. 2011, Zhang et Zhu, 2014). En se basant sur les meilleures micro-théories de la gouvernance (théorie des coûts de transaction, théorie des droits de propriété, théorie d'agence, théorie des réseaux), Beritelli et al. (2007) ont pu identifier six dimensions principales de la gouvernance de la destination: « coût de transaction », « asymétrie du pouvoir », « interdépendance », « confiance/contrôle », « connaissance » et « relations personnelles informelles ». Pour mieux opérationnaliser leur analyse, ces auteurs ont retenu d'autres dimensions non moins importantes, notamment : l'histoire de la destination et son développement, la taille de la destination, les performances de la destination, l'existence d'un groupe privé dominant, le rôle de la commune et de l'organisme du management de la destination (DMO), les facteurs d'attractivité, le comportement de coopération, la confiance mutuelle et le climat interne.

De leur côté et sur la base d'une recherche menée sur 53 articles traitant de la gouvernance dans deux disciplines majeures à savoir les sciences politiques et les sciences de management Ruhanen et al. (2010) ont cherché à relever les dimensions de la gouvernance de la destination les plus citées et utilisées. Ainsi, sur une base de 40 dimensions soulevées et en ciblant celles qui étaient citées au moins une dizaine de fois dans ces articles, ces auteurs ont pu retenir les principales dimensions suivantes : imputabilité, transparence, implication, structure, efficacité et pouvoir. En ne contentant uniquement d'énumérer ces dimensions sans pour autant montrer leur importance eu égard à la gouvernance de la destination, ces auteurs concluent que malgré l'existence d'un cadre théorique relativement construit, le concept de la gouvernance de destination reste multidimensionnel. Cette multi-dimensionnalité nous pousse à considérer la destination comme un système constitué par plusieurs acteurs et dans lequel chaque acteur exerce une certaine influence et est influencé par ledit système. D'où la nécessité de faire appel à l'approche systémique pour analyser la gouvernance de la destination (Laws et al. 2011:10). Cette conclusion est confortée par celle de Zhang et Zhu (2014 :125-126) qui affirment que la gouvernance de la destination se caractérise par la complexité due à l'interaction entre les secteurs public et privé, lesquels sont interdépendants en termes de ressources. Pour ces auteurs, complexité, interdépendance sur les ressources et relations entre le public et le privé sont les trois principaux aspects qui caractérisent la gouvernance de la destination.

Somme toute, les différents travaux ayant touché à la gouvernance des destinations attestent de sa grande complexité et de la difficulté de saisir toutes ses dimensions voir encore de juger de son efficacité. Sur cette base plusieurs modèles du management et de la gouvernance des destinations touristiques ont été développés et discutés sur la base d'expériences en vogue dans les pays du nord notamment aux Etats Unis et en Europe. Dans le point suivant nous exposerons les modèles les plus importants et nous discuterons de leur pertinence et efficacité.

Les modèles de Management et de la gouvernance des destinations touristiques:-

Avec l'importance de plus en plus grandissante du tourisme dans les économies et dans les stratégies de développement des pays ces trois dernières décennies, la question du management et de la gouvernance des destinations touristiques a retenue l'attention tant des chercheurs, des professionnels, des gouvernementaux et des organismes internationaux (OMT, OCDE). Dans ce registre plusieurs modèles de Management de destination touristique ont été proposés. L'un de ces modèles qui reste le plus répandu et le plus cité est celui développé par les deux fameux auteurs américains Crouch et Richie (Crouch et Ritchie, 1999; Ritchie et Crouch, 2000). En prenant comme référence les travaux développés par M. Porter sur la compétitivité, ces deux auteurs ont élaboré un modèle présentant les éléments constituant la destination touristique et les principales fonctions que le ou les organes de gestion de la destination (DMO) doivent accomplir pour lui assurer réussite et compétitivité. Pour ces auteurs, les organes responsables de la gestion de la destination touristique ont à leur charge deux types de fonctions : celles dites du «management simple» (gestion des ressources, marketing, organisation...) et celles dites «politiques» ou de «management stratégique» (délimitation du système, élaboration d'une vision partagée, définition d'un positionnement, définition des choix stratégiques de développement et des choix de coopération-compétition, définition procédés de diagnostic, et de contrôle-évaluation). En empruntant la voie à ces deux auteurs, Flagestad et Hope (2001) vont développer un autre modèle aussi important et aussi répandu que celui de Crouch et Richie. Considérant la nature et les spécificités des destinations touristiques caractérisées par leur complexité et l'existence de nombreux intervenants et acteurs à intérêts divergents référant à un jeu entre des forces ayant pour objectifs prioritaire la satisfaction des résidents (collectivités publiques) et celles d'autres orientées vers la satisfaction d'objectifs économiques (les entreprises), ces deux auteurs ont tenté d'articuler, malgré leur rivalité, les grandes approches de la stratégie que sont celle du positionnement, celle des ressources et compétences, et sur d'autres plans, celle de la chaîne de valeur de Porter pour mettre en exergue la difficulté d'aboutir à un système de management concerté et coopératif entre les acteurs de la destination. Sur ce, les deux auteurs vont dans le sens de proposer un système de management de la destination hybride (dit aussi d'organisation en réseau) se situant entre celui hiérarchique (dominance d'un acteur fort) et celui du marché. Leur modèle va retenir l'attention de nombreux autres auteurs qui l'ont enrichi et élargi via l'intégration de la théorie des parties prenantes en proposant la prise en compte des autres acteurs opérants dans le territoire de la destination et leur nécessaire mise en réseau dans une vision politique du système (Gundolf, Jaouen et Loup, 2006, Dredge, 2006).

Les deux modèles proposés par Crouch et Richie et Flagestad et Hope ont été à la base d'une florissante littérature sur les modes d'organisation, du management et de gouvernance des destinations touristiques qui va permettre de générer et de développer, sur fond de deux théories qui se présentent rivales dans la littérature mais d'apparence complémentaires dans la réalité : les théories contractuelles et la théorie des parties prenantes, deux principaux corpus théoriques présentant modèles extrêmes du management et de la gouvernance des destinations à savoir le Corporate model et le Community model, entre les quels plusieurs situations peuvent se présenter.

Le **corporate model** (ou modèle entrepreneurial ou modèle d'entreprise) est celui le plus fréquent dans l'Amérique du Nord et repose sur les principes et les postulats des théories contractuelles. Ce type de modèle épouse le schéma d'une «business corporation» selon Flagestad (2001) et considère la destination touristique comme une entreprise dominée par une grande firme ou un groupe privé qui cherche à atteindre des objectifs d'efficience et de profit. Agissant au nom de la communauté locale et détenant le pouvoir politique et de décision sur les modalités de développement et d'exploitation de la destination, la firme dominante applique une gestion de type intégrée pour la destination sous fond d'une logique de profit avec une stratégie orientée marché et l'intégration des entreprises de services soit par incorporation dans le groupe soit par contractualisation. Plusieurs avantages sont associés à ce type de modèle. D'une part, la recherche de la maximisation du profit étant l'objectif principal, malgré la présence d'autres petites et moyennes entreprises agissant indépendamment de la grande entreprise, c'est cette dernière qui détermine l'orientation stratégique ainsi que les objectifs principaux de la destination et son pouvoir et sa capacité de mobiliser les ressources nécessaires et de solidifier le réseau d'entreprises incorporées ou liées permet de minimiser et de maîtriser les coûts et de réaliser des performances importantes pour toute la destination et d'assurer ainsi le rôle de leader pour les autres acteurs de la destination. (Flagstad et Hope 2001, p. 451-453) D'autre part, en rappelant l'hypothèse émise par Powell (1990) selon laquelle les entreprises hiérarchiquement organisées sont plus susceptibles d'entreprendre des actions (R&D, Investissement,...) impliquant une incertitude quant à leur résultat et qui nécessitent un investissement substantiel de ressources (argent, temps et énergie), ce type d'organisation permet le développement de l'apprentissage organisationnel nécessaire au renforcement de la position de l'entité et plus important encore pour le processus d'innovation dans la destination étant donné que les investissements dans les innovations de produits, de marchés et de procédés sont un aspect essentiel pour maintenir la compétitivité d'une

destination. Un autre avantage certain de ce type de modèle, mentionné par Powell (1990), est celui relatif à la question de l'opportunisme. Cet auteur note, en effet, que l'opportunisme des acteurs est atténué par le pouvoir d'autorité exercé par la grande firme. Il avance aussi que les différentes parties ont tendance à développer une identité plus forte quand elles sont regroupées ou liées au sein d'une même société. Ainsi, pour le cas des destinations touristiques gérées dans le cadre du Corporate Model cela se traduit par une diminution du risque d'être affaibli par les comportements opportunistes et déloyaux des acteurs et prestataires et une plus grande identification des acteurs individuels avec une entreprise fortes renforce l'identité de toute la destination. Et cette forte identité permet à tous les acteurs de développer des comportements cohérents qui auront un impact positif sur l'expérience du visiteur et par conséquent sur la compétitivité de la destination.

Le principal inconvénient associé à ce modèle réside dans le risque que la société dominante elle-même tend à agir de manière opportuniste en tirant parti du déséquilibre de pouvoir existant entre elle-même et les petites entreprises indépendantes et les autres acteurs dans la destination. En effet, la recherche de la maximisation du profit pourrait pousser cette société ou groupe dominant à exploiter son pouvoir et son influence pour ne servir que ses propres intérêts ou ne pas remplir ses obligations envers la communauté.

A l'opposé le modèle dit communautaire (**community model**) très spécifique à l'Europe repose sur la théorie des parties prenantes. Ce modèle considère que la destination touristique est très fragmentée et est constituée d'un grand nombre d'unités de productions de tailles et d'activités différentes, indépendantes, opérant dans une démarche décentralisée, sans pouvoir de domination mais aussi d'autres parties prenantes dont les acteurs publics, les collectivités territoriales et d'autres groupes d'intérêts distincts opérant dans le territoire. Ce modèle, se caractérise donc par la présence de nombreuses parties prenantes qui doivent pour une meilleure gestion parvenir à de fréquents compromis en raison de leurs divergences d'intérêts. Contrairement au premier modèle, le modèle communautaire ne reconnaît pas la dominance d'une unité ou d'un acteur sur les autres et le management stratégique des destinations ayant ces caractéristiques posent la problématique de la gouvernance de ces différentes parties prenantes. En général, au vue de la grande multitude des intervenants, la réglementation de ce modèle s'effectue de manière décentralisée, en vertu de laquelle l'organisation locale de gestion des destinations est responsable de la coordination des membres individuels, du marketing et de la formulation de l'orientation stratégique. Cependant, l'influence de cette organisation sur les mesures réelles dans la destination et son pouvoir décisionnel sont limitées. La caractéristique d'une destination communautaire est le fait que ni une entreprise dominante ni l'organisation de gestion ne contrôlent les unités d'affaires de manière hiérarchique.

Comme le font remarquer Flagestad et Hope (Flagestad et Hope 2001, p. 452) dans un tel système les forces politiques sont plus fortes que la vision économique. Et l'organisation du management de la destination dépend plus du comportement coopératif des entreprises indépendantes et des acteurs de gouvernance locale.

Caractérisant deux systèmes différents, ces deux modèles donnent l'apparence qu'ils sont exclusifs. Certes, chacun des deux modèles apporte des éléments d'analyse importants : le corporate modèle mis plus l'accent sur l'importance de l'existence d'un groupe privé fort tant sur le plan financier que sur le plan de la culture d'entreprise adoptant une logique de gestion d'entreprise comme moyen pour assurer la performance et la compétitivité de la destination alors que le modèle communautaire introduit le rôle fondamental des collectivités locales à coté des autres prestataires de services touristiques et la nécessité d'un bon système de gouvernance pour assurer la compétitivité de la destination. Cependant à la question centrale « quel modèle est le plus pertinent en termes de stratégie et de compétitivité ? », la réponse ne peut en aucun cas être à l'absolu. Et pour cause, en retenant la conclusion fondamentale d'Arvid Flagestad (2001) qui statue sur l'existence d'un continuum de situations entre le corporate model et le community model et qu'il ne s'agit pas de se placer dans l'un ou l'autre de ces deux modèles. Dans les destinations caractérisées par la présence de nombreuses entreprises à finalité économique et commerciale et sans réelle domination et de plusieurs acteurs territoriaux gérant les biens publics et garants de l'intérêt général, aucun des deux modèles ne peut être appliqué totalement. Ainsi, dans ces types de destinations, le système d'organisation et de management est à bâtir dans une logique de coopération et de partenariat entre les unités économiques et le pouvoir politique local. En effet, les premiers étant incapables d'assurer le leadership total dans la destination et les second aussi à eux seuls ne peuvent pas non plus assurer la dynamique économique de la destination, une nécessaire coordination et collaboration doit être développée et mise en œuvre entre ces deux sphères publique et privée pour pouvoir mettre en place un système de management et de gouvernance adapté et efficace. Ainsi, compte tenu de ces considérations, il est peu probable que les systèmes d'organisation et de management des destinations observables dans la réalité soient une forme extrême. De ce fait, entre ces deux

modèles tout un ensemble de situations hybrides est possible et chaque destination doit définir son modèle en fonction de ses spécificités et de la complexité ou non de son contexte et de sa situation.

Analyse du modèle de gouvernance des destinations touristiques au Maroc:-

L'analyse du système de gouvernance touristique des destinations touristiques au Maroc dans le cadre de la nouvelle stratégie de développement touristique sera menée en deux temps. Dans un premier temps nous tenterons de caractériser le modèle promu à travers sa présentation selon le CPN. Dans un deuxième temps nous procéderons, à une lecture de son opérationnalisation à travers l'analyse de son processus de mise en œuvre et son état actuel à quatre années de l'horizon 2020.

Présentation et caractérisation du nouveau modèle de gouvernance des destinations touristiques au Maroc:-

Jusqu'à l'avènement de la nouvelle stratégie touristique, que nous avons qualifié de l'ère des visions, le secteur touristique était géré en grande partie par les pouvoirs publics dans le cadre de plan de développement dont l'Etat définissait la politique et les plans d'action et en assurait la mise en œuvre, le suivi et la régulation sur le plan national et régional. C'est avec la vision 2010 que les premiers jalons de la gouvernance touristique vont être posés à travers une démarche de conception et de mise en œuvre basée sur la concertation et le partenariat comme cadre et principes fondateurs. Cette volonté d'asseoir une meilleure gouvernance s'est traduite aussi dans le cadre de cette vision par la modernisation de certaines structures publiques et la création d'autres. Ainsi, dans le cadre de cette vision, l'Etat a procédé à la modernisation de l'Office National Marocain du Tourisme (ONMT) par son recentrage sur son métier de promotion nationale et du marketing et en le dotant de moyens conséquents, la création par le Ministère du Tourisme de la Société Marocaine d'Ingénierie Touristique (SMIT), en remplacement de la Direction des Aménagements et des Investissements (DAI) pour soutenir et orienter le développement de l'offre, assurer également le suivi des grands projets de la Vision 2010, la création des Conseils Régionaux du Tourisme (CRT), instances regroupant les professionnels privés, les élus et les autorités locales, pour poser les premières bases d'une gouvernance locale, la création de l'Observatoire du Tourisme, pour professionnaliser la publication de statistiques et d'études et la tenue annuelle des Assises du Tourisme pour effectuer le bilan des réalisations et débattre des priorités à venir. Dans cette foulée, l'Etat a aussi procédé, à partir de l'année 2002, à la création des Centres Régionaux d'Investissement (CRI) dont la mission est principalement de centraliser et de simplifier les démarches administratives permettant la création des entreprises et de soutenir et d'accompagner les investisseurs au niveau régional. Concernant le privé, la FNT, acteur majeur représentant le secteur privé, visait d'une part le regroupement en son sein de l'ensemble des professions touristiques et d'autre part le renforcement des structures des différentes professions pour plus d'efficacité et de participation aussi bien au niveau régional que national (FNIH, FNAVM, FNR, STT, ALASCAM et FNAGAM). Malgré cela nous pouvons souligner que la gouvernance est restée le parent pauvre de cette vision. Et pour cause les efforts étaient plus concentrés sur l'accentuation de la réalisation des investissements programmés dans le cadre du Plan Azur (programme de réalisation des stations balnéaires) et sur le développement de l'offre culturelle. Ainsi, les efforts étaient plus centrés sur le développement de l'offre en matière de produits (balnéaire et culturel) et sur ce plan l'Etat a pu en grande partie gagner le pari au vu des réalisations physiques enregistrées et a pu ainsi mobiliser les investissements et les acteurs et créer une dynamique positive dans le secteur. Dans son rapport publié en mai 2008, intitulé « Bilan stratégique de la vision 2010 » la FNT, atteste que globalement le chantier "organisation institutionnelle" a accusé beaucoup de retard avec un déficit de gouvernance et faiblesse des organes de pilotage ainsi qu'une faiblesse des capacités de gestion des associations du secteur (CRT, Fédérations, Associations professionnelles,...).

C'est avec la vision 2020 que l'on peut parler d'un véritable système de gouvernance dans le tourisme marocain. En effet, reconnaissant la nature transversale du tourisme et son importance à l'échelle nationale les concepteurs de la vision ont cherché à asseoir un fort leadership et une gouvernance renforcée afin d'une part de renforcer la légitimité institutionnelles du secteur (vue la multiplicité des acteurs) et de faciliter les prises de décision au niveau national et régional à travers un pilotage national stratégique de la vision et la garantie de la bonne exécution des stratégies et plans d'actions territoriaux (CPN, p :30). En sous-jacent à cette logique, l'objectif est de mettre en place des mécanismes et des moyens de pilotage adaptés, mieux partagés et plus synthétiques pour éviter un nombre d'acteurs trop importants (FNT, 2008, p28). Pour ce faire, de nouvelles structures programmées dans le cadre de cette vision se sont venues s'ajouter aux autres structures nationales, régionales et locales déjà en place. Il s'agit principalement de la Haute Autorité du Tourisme (HAT) (dénommée initialement : Conseil National du Tourisme) et des Agences de Développement Touristique (ADT). D'après la Mesure 37 du CPN (CPN-Vision 2020 ; p : 30), la HAT est une instance nationale de pilotage, réunissant dans un partenariat renouvelé l'Etat, les régions et le secteur privé qui aura pour principales missions de s'assurer de la cohérence de la vision à long terme et du suivi de son exécution, de

suivre et évaluer l'action des différents acteurs clés publics et privés de la gouvernance du tourisme ainsi que de suivre et évaluer l'exécution des feuilles de routes régionales en veillant en particulier à un pilotage coordonné de tous les paramètres clés de l'équation touristique, à la régulation dynamique des capacités hôtelières et de formation des ressources humaines; d'assurer le suivi des efforts des autorités concernées pour une adéquation entre les capacités aériennes et les capacités littorales et de proposer les ajustements nécessaires à la mise en œuvre de la Vision et le cas échéant à la révision de ses paramètres stratégiques. Véritable organe de pilotage et de gouvernance du secteur au niveau national la HAT, dont la mise en place était prévue pour au plus tard à la fin de l'année 2011, est composée, sous la tutelle du Ministère du Tourisme, qui en assure le Secrétariat Général, une dizaine d'autres ministères, la CGEM et 5 représentants du secteur privé (représentants des fédérations professionnelles). Pour accompagner et réussir la régionalisation touristique retenue dans la vision 2020 et de s'assurer de la mise en œuvre des stratégies territoriales (CPR), la HAT sera reliée sur le plan régional par les ADT qui devraient être progressivement créées dans les 8 territoires touristiques retenus (4 agences à fin 2011 et 4 agences à fin 2012). Ces agences, d'après la mesure 38 du CPN (CPN-vision 2020 ; p : 31), devraient avoir pour principales missions de garantir la bonne exécution des Contrats Programmes Régionaux (CPR) de leur territoire respectif pour en favoriser un développement touristique harmonieux, rapide, pérenne et rentable et ce à travers, outre la déclinaison des missions de la HAT au niveau régional, de développer l'attractivité globale des territoires, en direction des investisseurs et grands acteurs du secteur, de développer la compétitivité des territoires via la coordination, d'assurer la promotion, la commercialisation et l'accompagnement du développement de la desserte aérienne de leur territoire respectif auprès des marchés ciblés, d'assurer le contrôle de la qualité sur l'ensemble de la chaîne de valeur et au sein de l'environnement touristique, d'assurer le suivi et l'orientation des projets stratégiques et d'orienter les professionnels locaux du secteur en fonction des tendances du marché mondial ainsi qu'une diffusion des meilleures pratiques et des démarches novatrices.

Ainsi, dans cette nouvelle configuration, les ADT assureront le rôle du principal interlocuteur des acteurs publics et privés pour toutes les questions liées au tourisme, et constitueront une plateforme d'échange et de concertation au niveau de leur territoire. Dans ce schéma les structures actuelles, notamment la Délégation Régionale du Tourisme, les Conseils Régionaux du Tourisme (CRT) et les Conseils Provinciaux du Tourisme (CPT) seront remplacés progressivement par les ADT des territoires concernés. En matière de gestion et de fonctionnement, ces ADT, étant des établissements publics sous la tutelle du Ministère du Tourisme, seront pilotés par un Conseil d'administration, composé de représentants des secteurs public et privé (MT, les représentants des Ministères, les élus et les représentants des associations professionnelles régionales relevant du territoire) et opéreront par des «contrats de performance» pluriannuels validés et suivis par la HAT. Dans leurs activités opérationnelles les ADT auront la tâche d'une part, en collaboreront avec les centres régionaux d'investissement (CRI), de développer de l'offre produit local en cohérence avec la stratégie nationale, d'orienter et d'accompagner les investisseurs spontanés et de piloter et d'assurer le suivi des projets planifiés. Et d'autre part d'élaborer et de mettre en œuvre une Stratégie Intégrée de promotion et de Marketing Territorial en parfaite cohérence avec la stratégie de marque nationale.

En référence aux modèles théoriques présentés auparavant, et en plaçant la gouvernance basée sur le partenariat public-privé comme pilier fondamental de la mise en œuvre et de la réussite de la nouvelle politique touristique, le système de management et de gouvernance touristique marocain s'apparente plus à un modèle hybride entre le corporate modèle et le community modèle avec un rapprochement de ce dernier. En effet, en misant sur les ADT comme organe de management et de territorialisation de la mise en œuvre de la stratégie de développement touristique le modèle marocain va dans le sens de l'émergence d'un acteurs collectif au sens de Gerbaux (Gerbaux 2003) ou d'acteur stratégique (Gerbaux et Marcelpoil, 2003) composé de différentes parties prenantes, publiques et privées et qui est propres aux destinations organisées de façon communautaires. Toutefois dans un environnement rapidement évolutif caractérisé par le déplacement des barrières entre acteurs publics et privés ce modèle de gouvernance ne pourrait réussir que si trois principaux éléments sont réunis : la capacité d'agir, l'existence d'acteurs et d'intérêts organisés et des relations entre acteurs afin d'agir ensemble (Le Galès, 1995, pp. 81-82). Pour cet auteur, l'existence d'une réelle coalition d'acteurs publics et privés est la condition nécessaire pour définir et mettre en place des politiques efficaces. Cette avancée est toutefois minimisée par Lorrain (2000) pour qui l'existence d'une coalition n'est pas suffisante pour faire émerger et réussir un acteur stratégique, du fait que gouverner un territoire suppose, selon lui, de parvenir à des compromis entre des intérêts potentiellement divergents, sur un horizon temporel lointains dont les résultats sont aléatoires. Pour cet auteur, ni les institutions politiques en raison des contraintes électorales ni les acteurs privés évoluant dans un environnement de plus en plus concurrentiel et mutant ne sont disposés à s'engager sur ces horizons lointains aux résultats incertains. Ces contraintes risquant de compromettre la mission de l'acteur stratégique l'auteur privilégie la thèse de l'acteur réactif à celle de l'acteur

stratégique. C. Van Der Yeught (2009) de son côté avance que l'émergence d'un acteur stratégique collectif exige la réunion de trois conditions fondamentales relevant de la gouvernance des destinations à savoir l'hybridation des modes de coordination déployés, la relative indépendance politique de l'équipe dirigeante et sa forte légitimité.

L'hybridation des modes de coordination consiste en le déploiement d'un dispositif de gouvernance permettant d'assurer la coordination des nombreuses parties prenantes impliquées dans le projet productif, de doter le groupe d'une intention stratégique et de maintenir le cap fixé à long terme, indépendamment des jeux de pouvoir et des aléas du calendrier électoral. L'indépendance politique de l'équipe dirigeante est la seconde condition favorisant l'émergence d'un acteur stratégique. Cette indépendance est nécessaire pour ne pas compromettre et entraver le déroulement des projets à long terme. Quand à la légitimité de l'équipe dirigeante elle repose soit sur l'existence d'une réglementation claire ou sur une volonté collective des parties prenantes basée sur des valeurs partagées et une confiance mutuelle. Cette analyse montre que l'émergence d'un acteur stratégique collectif au sein d'une DT ne va pas de soi, mais dépend plutôt de la structure de gouvernance déployée. Qu'en est-il dans le cas du modèle marocain ? Les ADT prévues dans le cadre du CPN seront des établissements publics sous la tutelle du MT, constituées par la fusion des Délégations Régionales du Tourisme, des CRT et des CPT. Or la délégation est une administration alors que les CRT et les CPT sont des associations professionnelles avec l'autonomie budgétaire. Cette nouvelle structure abritera en son sein ainsi des fonctionnaires et des professionnels et sera régie par les règles du droit administratif et des finances publiques. Au regard des trois exigences d'une gouvernance efficace énoncées ci-dessus il paraît très difficile que les ADT deviennent l'acteur stratégique collectif au sens de Van Der Yeught. Une faible légitimité étant donné la faible représentation de la sphère privée pierre angulaire de la mise en œuvre de la vision, une faible indépendance de l'équipe dirigeante à cause de prédominance de la sphère publique et politique dans la gestion et le financement et un problème de gestion étant donné les divergences des visions et des compétences en matière de gestion et de prise de décision entre les fonctionnaires et les gestionnaires issus des structures fusionnées et par conséquent un risque majeur d'émergence de problèmes de coordination et d'adhésion de tous les acteurs aux intérêts divergents.

Après avoir présenté et caractérisé le modèle de gouvernance des destinations touristiques marocaines dans le cadre de la nouvelle stratégie de développement nous allons dans le point suivant comment cette nouvelle gouvernance s'est traduite dans la pratique après pratiquement 6 années de son adoption et à 4 années de l'horizon des objectifs tracés.

La gouvernance des destinations touristiques marocaines à l'épreuve de la mise en œuvre et de l'opérationnalisation:-

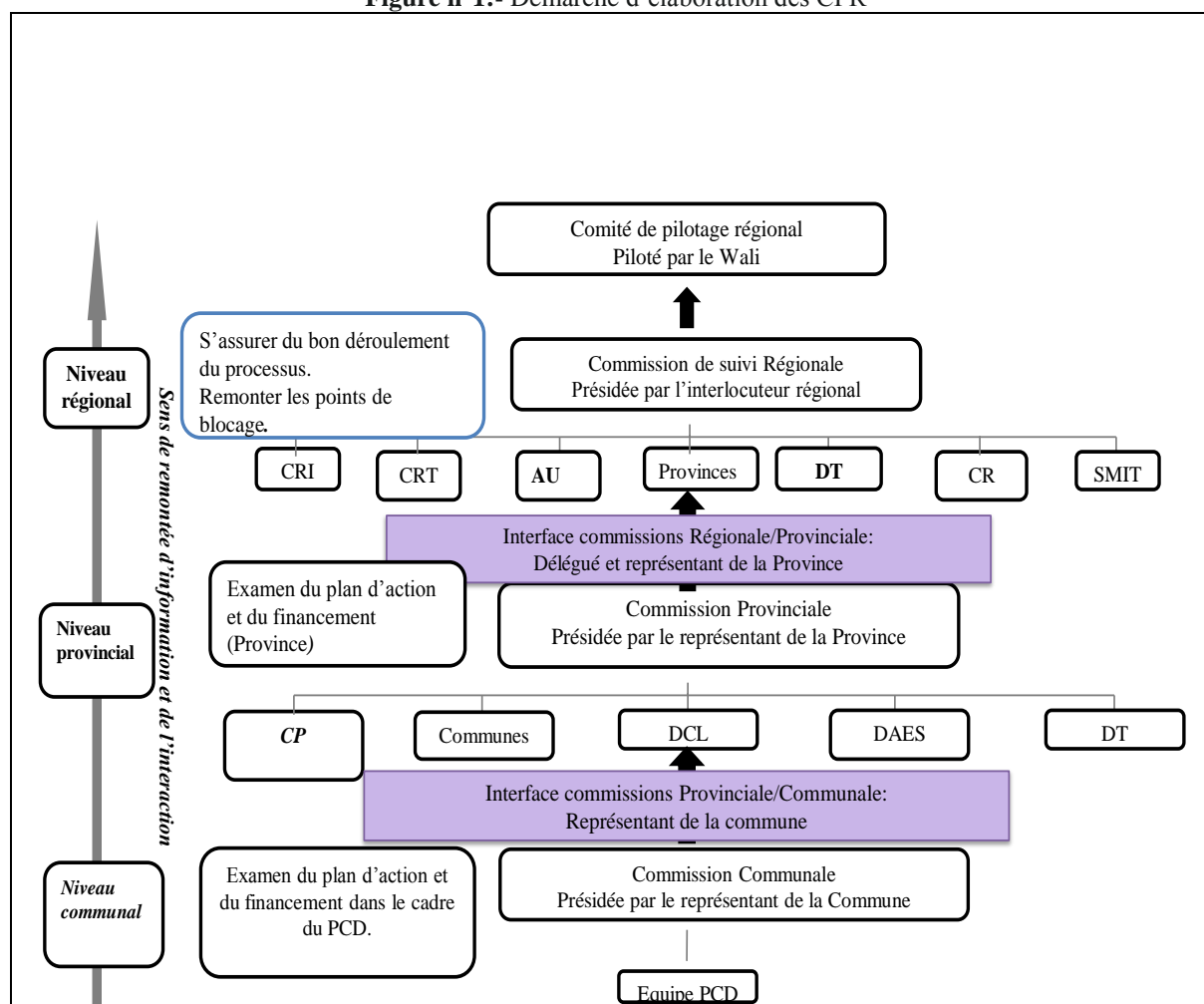
Notre étude de la gouvernance des destinations touristiques marocaines dans le nouveau modèle de développement touristique a été opérée en deux principaux temps. L'analyse de la gouvernance dans le processus de mise en œuvre de la nouvelle stratégie touristique à travers une lecture documentaire et son opérationnalisation effective sur la base des résultats de notre étude terrain.

En retenant qu'un processus de gouvernance est un «ensemble défini d'activités qui sont effectuées dans le cadre de la gouvernance pour permettre la définition cohérente, l'application et, au besoin, la modification des règles qui organisent et régulent les activités des participants pour mettre en œuvre les politiques énoncées» nous chercherons à caractériser la gouvernance dans le processus de mise en œuvre de la gouvernance dans la nouvelle politique touristique marocaine. Ce processus dans le cas du Maroc à connu trois principales phases. La première phase caractérisée par une implication et participation limitée des acteurs dans le cadre de l'élaboration du CPN et une deuxième phase marquée par une forte implication et participation des acteurs selon une approche up-top dans le cadre de l'élaboration des CPR. La troisième étape, compte tenu du retard accusé dans la mise en place des structures prévues (ADT), est caractérisée par un scénario secours donnant le pilotage régional des contrats.

D'après les données disponibles, c'est en 2009, au vu du bilan provisoire de la vision 2010, que les acteurs concernés (MT et FNT) avec l'aide de bureaux d'études étrangers ont entamé les réflexions et les travaux pour l'élaboration des orientations stratégiques qui devraient aboutir à la vision 2020. Ces travaux qui vont durer près de trois ans n'ont pas été marqués par une implication et participation de tous les acteurs du secteur. L'urgence de présenter la nouvelle stratégie dans les assises du tourisme de 2010 a fait que l'implication et la concertation ce sont plus limitées à l'information des acteurs. Pour nombre de professionnels cette approche n'a pas respecté les principes d'une bonne gouvernance et s'apparente plus à une approche top-down masquée par une tentative d'implication ad hoc des acteurs.

La vraie démarche de concertation et d'implication de tous les acteurs et donc de gouvernance n'a pu se concrétiser que dans le cadre de l'élaboration des CPR. En effet, si l'élaboration du CPN n'a pas connu, à l'avis de tous et plus particulièrement des professionnels, une réelle concertation et participation, c'est l'élaboration des CPR qui a connu la collaboration et la coordination des différents acteurs impliqués sur les plans local, régional et national selon une approche bottom-up. En effet, le CPN stipule (Mesure 1, CPN p :13), comme l'une de ses recommandations principales pour la mise en œuvre de la vision 2020, que chacun des 8 territoires (ou destinations touristiques) identifiés par la Vision 2020 élabore son Contrat Programme avec des objectifs fixés à partir des potentialités naturelles de chaque région, des pistes de financement pour la construction d'infrastructures et d'une offre touristique spécifique pour accueillir les touristes dont le nombre potentiel est aussi déterminé pour chaque destination. Ces CPR constituent, ainsi, des feuilles de routes précises qui doivent s'élaborer en étroite concertation avec l'ensemble des acteurs locaux: élus, professionnels, société civile et acteurs publics de chaque territoire. La démarche générale d'élaboration des CPR est présentée dans le schéma suivant :

Figure n°1:- Démarche d'élaboration des CPR



Source : élaboré par les auteurs sur la base du processus décrit dans le CPN.

Suivant une démarche interactive l'élaboration des CPR a débuté par la consultation du niveau local (communes) à travers le listage des projets touristiques retenus dans les plans communaux du développement (PCD) puis leur filtrage au niveau de chaque province en présence du représentant du Ministère (Délégué) et leur validation au niveau régional pour leur mise en œuvre sous forme du programme. le programme validé sur le plan régional est enfin remonté au niveau national pour un dernier filtrage visant à vérifier la concordance et la convergence des projets avec le positionnement du territoire d'implantation tel que prévu dans la vision 2020.

Si la démarche voulue a été globalement respectée plusieurs limites sont à soulever et qui entravent le processus de gouvernance mais aussi la réalisation des objectifs de la vision 2020. En premier, il ya lieu de noter la grande problématique de non concordance entre les découpages administratif et touristique et leurs mutations dans la période d'élaboration et de mise en œuvre des CPR. En effet, en cherchant à développer une expérience touristique beaucoup plus diverse, tant en termes de nombre de destinations que de variété de l'offre au sein de chacune de celles-ci, la vision 2020 s'est construite sur un découpage, qui transcende les barrières administratives à fin de permettre à chaque région de tirer profit de l'ensemble de ses potentialités et ainsi huit territoires touristiques en été retenus. Toutefois, le découpage administratif de l'époque était composé de 16 régions. Ainsi, 16 CPR étaient prévus pour huit territoires touristiques. D'autant plus que depuis 2011 l'organisation administrative du pays a connu un changement notable en limitant le nombre des régions administratives à seulement 12 régions (régionalisation avancée dont le projet de décret a été adopté par le conseil de gouvernement en février 2015). Dans ce contexte de mutation et du retard de mise en œuvre de la régionalisation avancée et des structures de gouvernance touristique notamment les ADT, chargées de la mise en œuvre territoriale de la vision, confrontés à l'horizon de réalisation des objectifs de la vision, l'élaboration des CPR a été l'œuvre de sous-commissions créées aux différents échelles régionales. Ainsi, encore une fois c'est le découpage administratif et non touristique qui a prévalu étant donné que pour outrepasser les problèmes et les tensions entre territoires et entre acteurs en jeu, c'est à l'échelle des provinces que les projets ont été filtrés pour leur simple validation au niveau de la région. Dans ce processus qui a pris au moins trois années pour l'élaboration des CPR pour toutes les régions nous pouvons ainsi relever la prédominance du politique sur les autres acteurs étant donné que le comité régional est présidé par le wali de la région. Cette situation montre que la régionalisation touristique n'a pas été respectée, ni dans sa dimension territoriale ni dans sa dimension gouvernance, et les CPR, s'apparentent plus à une collection de contrats-programmes provinciaux loin de la logique d'une réelle régionalisation touristique supposant une vraie appropriation du territoire par tous les acteurs comme espace permettant la résolution de leurs problèmes identifiés comme communs. En effet, les retards accusés dans l'opérationnalisation de la régionalisation avancée et de la régionalisation des structures de gouvernance touristique ont fait que les différents acteurs ont adopté la voie de la collaboration et de la coordination en se référant au territoire administratif et non au territoire touristique.

En second, un autre aspect important de la défaillance de la gouvernance touristique lors du processus d'élaboration des CPR réside dans la manière avec laquelle les projets ont été proposés et adoptés. En effet, alors que la démarche prévoyait donner la priorité aux acteurs locaux pour la proposition des projets pour faciliter leur territorialisation et leur appropriation il n'en été rien dans la réalité, lors des concertations, les représentants régionaux du ministère du tourisme se sont déplacés aux communes avec une banque de projets conçus à l'avance et concordants avec la vision 2020 pour chaque territoire. Cette procédure à faciliter l'alignement de ces communes qui se sont appropriées ces projets et les ont intégré dans leur propres programmes communaux de développement au lieu de l'inverse. Ainsi, la remontée de l'information, le filtrage et la validation des projets et du CPR se sont trouvés faciliter sans réelle concertation et implication de tous les acteurs territoriaux et sans réelle vraie territorialisation des projets.

En troisième, la question de la gouvernance dans le contexte actuel est vécue différemment par les deux sphères publique et privée et les départage. Du côté de la sphère publique la course vers la réalisation des objectifs de la vision 2020 est vécue comme une contrainte majeure et conduit à une sorte de gouvernance de la crise. En effet, après plus de trois ans de l'adoption des CPR et le retard très important accusé dans la concrétisation de la grande majorité des projets d'investissement retenus, une simple analyse de l'agenda des comités du pilotage régionaux, montre la grande priorité consacré au suivi de l'état d'avancement des projets et à la recherche de l'élimination des facteurs de blocage à leur réalisation et à la recherche d'investisseurs preneurs et de financement et les questions du management et de la gouvernance des destinations dans leur aspect promotionnel et marketing sont reléguées au second ordre. Du côté de la sphère privée, la situation est différente. En effet, les professionnels du secteur, acteur majeur dans la mise en œuvre de la vision et des CPR, touchés pour leur grande majorité par la conjoncture difficile que travers le tourisme national, sont plus préoccupés par la situation économique de leurs unités en grande majorité en difficulté. Malgré, qu'ils avancent adhérer sans faille aux politiques et décisions prises (une sorte d'adhésion de façade ou sous silence), ils sont plus demandeurs de mesures de soutien et d'actions orientées vers la promotion de la destination et de la recherche de marchés et marginalisent les préoccupations majeurs des acteurs publics plus orientés vers l'accélération de la concrétisation des programmes retenus dans les CPR. Au sein même de cette sphère privée les avis sont très disparates. Dans presque toutes les destinations du pays compte tenu de la conjoncture difficile que travers le tourisme, nous assistons à une structure à double palier. Alors que les grandes structures hôtelières et plus particulièrement celles appartenant à des chaînes internationales se comportent bien la grande majorité des structures indépendantes sont en difficultés et sont plus préoccupées par la recherche de

solutions commerciales ou économiques à leur problèmes. Dans ce contexte, pour ces acteurs, parler de gouvernance, de collaboration et d'adhésion relève presque de l'impossible.

Somme toute, à quatre années de l'horizon 2020, en mettant de côté les trois seuls points vraiment lumineux dans cette nouvelle politique consistants en le partage de l'ambition par tous et la validité du modèle de gouvernance promu et une forte tendance tant de la sphère publique que celle privée vers une vraie régionalisation de la gestion et de la gouvernance des programmes et des projets touristiques, nous sommes en droit, au vue des grands retards accusés tant dans la mise en place de la régionalisation avancée, des structures de gouvernance touristiques régionales et des projets et programmes retenus, de se poser des questions sur la validité des objectifs retenus et aussi de se poser la question légitime de savoir s'il s'agissait d'une véritable stratégie pour laquelle on s'est donné réellement et concrètement les moyens de sa mise en œuvre et réussite ou simplement d'une période transitoire à l'apprentissage du management et de la gouvernance du tourisme dont il faudra tirer les enseignements nécessaires et enclencher les mesures et actions efficaces pour bâtir le vrai cadre d'un management stratégique et de gouvernance efficace de nos destinations touristiques.

Conclusions:-

En posant la problématique de la gouvernance des destinations touristiques marocaine nous avons cherché à placer le modèle de gouvernance promu dans le cadre de la nouvelle stratégie de développement touristique du pays dans le référentiel théorique mais aussi à le questionner dans le processus de sa mise en œuvre et opérationnalisation. En plaçant la gouvernance basée sur le partenariat public-privé comme pilier fondamental de la mise en œuvre et de la réussite de la nouvelle politique touristique, le système de management touristique marocain s'apparente plus à un modèle hybride entre le corporate modèle et le community modèle. Toutefois, si le schéma institutionnel de la gouvernance tel que décrit dans le CPN semble respecter les principes d'une bonne gouvernance des destinations touristiques, sa traduction dans le terrain a connu des limites. Les réformes territoriales et organisationnelles n'étant pas encore achevées conjuguées à la conjoncture difficile que traverse le tourisme national ne doivent pas pour autant entrainer les acteurs dans un scénario de gestion de la crise mais plutôt d'appréhender et de voir en cette situation une occasion d'apprentissage pouvant engendrer une dynamique positive susceptible d'entrainer des améliorations dans leur manière de percevoir les logiques du secteur et son organisation pour un meilleur management et gouvernance. Chaque destination étant spécifique et ayant son contexte territorial et d'acteurs particulier, seule une gouvernance basée sur le partage d'objectifs communs, la coordination et la concertation de tous les acteurs dans un climat de participation, de cohérence, de confiance, d'ouverture, de responsabilité et d'efficacité peut assurer les bases d'une meilleure compétitivité des destinations touristiques marocaines. Le tourisme est et restera encore et pour toujours un pilier de l'économie marocaine, ré-ouvrir, ainsi, le chantier de la gouvernance, le rediscuter et le réussir à l'échelle territoriale pertinente semble être la voie prometteuse pour asseoir un management stratégique performant pour asseoir la compétitivité et la durabilité des destinations touristiques marocaines.

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RESEARCH ARTICLE

SURVEY PAPER ON TEXT RETRIEVAL AND TRANSLATION FROM SIGN BOARD IMAGES.

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Abstract

The system assists people specially travelers to find their paths while travelling in English speaking countries without language barrier. Sign Board helps travelers to complete their journey with ease. But, understanding sign boards in English will be difficult for travelers who don't know English. To solve this, we are using text retrieval and translation from English to Marathi from sign boards. Text retrieval can be performed by using methods like Optical Character Recognition, Maximally Stable Extremal Region etc. Text translation can be performed by using various free API services available from organizations like Google, Microsoft, Yandex etc. For text retrieval, we found Optical Character Recognition as the best technique and for translation API services available from Google are the best.

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Introduction:-

Sign boards provide route guidance using short and precise information. It conveys information that helps common people to take decisions while travelling. The language barrier among different English speaking countries is one of the major difficulties faced while travelling. The reason being tourists are unable to understand sign boards in the native language of the region. They may suffer consequent loss of important information which may lead to misguidance. The sign boards may also convey some warnings or hazards. The gaining momentum of internet and portable mobile phones makes it easy to drift from traditional dictionary translation to a better web app which is user friendly. The world is moving towards image records and retrieval of information from the same. In this sense, it is expected that a high percentage of the world population will own a smart phone with an embedded camera and an internet connection, which is our system's requirement.

Various techniques are used to retrieve text from an image. One of them is Optical Character Recognition (OCR)[1]. OCR is the process of converting a scanned document image into machine encoded format. The scanned document image may include photograph of a document, image of sign boards and billboard from a landscape photograph. OCR finds its applications in the fields of pattern recognition, artificial intelligence, handwriting recognition, etc. Although OCR works efficiently with digital images, it doesn't work well with natural scene images [2].

Another image processing technique is Maximum Stable Extremal Region (MSER). It is a technique used to detect blobs in images. Blobs are regions which differ in properties like brightness or color. MSER is useful while detecting text from a natural scene image. It also provides great results for images which are captured blur or of varying region density and size.

For taking pictures of the sign boards, we are using a mobile phone camera of a good quality so that we can get clear and stable pictures of the sign boards. Quality of a camera is going to affect the quality of the image of a sign board.

So, we should take care of that while selecting the camera. Each of the methods of a text retrieval has some advantages and also some drawbacks. The next section i.e. Section 2 gives us the detailed idea about these methodologies used to retrieve the text and translate the text from sign board images. Section 3 gives us comparison between these methodologies. Section 4 gives us the conclusion of this survey.

Related Work:-

The system of text retrieval and translation from sign board images works in two phases i.e. text retrieval and text translation.

Text retrieval:-

It is the process of detecting & retrieving of the text from the images. There are two popular methods of text retrieval.

OCR- optical character recognition:-

The Optical Character Recognition(OCR) is considered as machine replication of human reading. Today's most common way of inputting the data into computer is by using the keyboard but, it is time consuming and it requires human efforts [3]. OCR provides a desirable way to extract the text from the images. So, that we can efficiently input the data to the computer. OCR is considered as mechanical or electronic conversion of scanned image where image can contain handwritten or typewritten text. OCR is the process of digitizing the printed text so that it can be used electronically in various machine processes. OCR is the process of converting a scanned document image into machine encoded format. The scanned document image may include photograph of a document, image of sign boards and billboard from a landscape photograph. OCR finds its applications in the fields of pattern recognition, artificial intelligence, handwriting recognition, etc.

Nowadays, many institutions and organizations are moving towards OCR to eliminate the human interference and increase the efficiency [5]. OCR is of two types and they are offline recognition and online recognition. In offline recognition either an image or scanned form of image is considered. In online recognition, successive points are chosen as function of time and order of strokes are also available. The OCR methodology has the following features:

- ❖ No more retyping
- ❖ Edit text.
- ❖ Save space.
- ❖ Quick digital searches

Steps of a OCR are as follows:-**Pre-processing:-**

OCR software often "pre-processes" images to improve the chances of successful recognition. Here input image is taken and it is converted to the gray scale image. The gray scale image is finally converted into binary image. This process is known as binarization of the image. Scanned image from the scanner may contain some noise due to the unnecessary details present in the image. This noise should be removed from the image for the proper application of OCR. Some algorithms are used to denoise the image and this denoised image should be referred for the further use in the system. Other operations that are performed in the pre-processing of the image are: Deskew, Despeckle, Fragmentation, Line removal, layout analysis.

Character Extraction:-

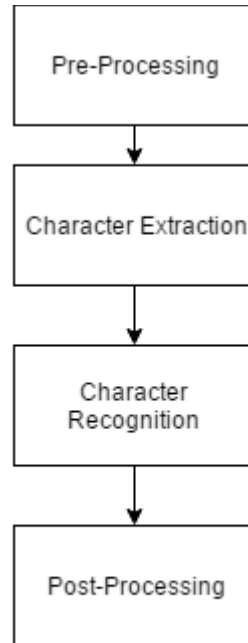
Preprocessed image is provided as input to this step to extract each and every character from text which is obtained from the given image.

Character Recognition:-

The image from the extraction stage is correlated with the templates which are preloaded into the system. Once the correlation is done, the template with maximum correlated value is declared as the character present in that image. There are two basic types of core OCR algorithm, which may produce a ranked list of candidate characters. Matrix matching involves comparing an image to a stored glyph on a pixel-by-pixel basis; it is also known as "pattern matching" or "pattern recognition". This relies on the input glyph being correctly isolated from the rest of the image, and on the stored glyph being in a similar font and at the same scale. This technique works best with typewritten text and does not work well when new fonts are encountered. This is the technique the early physical photocell-based OCR implemented, rather directly.

Post Processing:-

After the character recognition, if there are still unrecognized characters in the image then that characters are given their meaning in post processing step. Some extra templates can be added for providing wide range of compatibility checking in the system database. The Levenshtein Distance algorithm has also been used in OCR post-processing to further optimize results from an OCR API.



Block Diagram of OCR

We can face some challenges while implementing the traditional OCR on mobile devices. These challenges are-

- 1) Non-contact nature of digital cameras in mobiles.
 - 2) Poor quality of images.
 - 3) Processing speed and memory are not that sufficient to run the desktop OCR algorithms on mobile devices.
 - 4) Mobile devices don't have Floating Point Unit(FPU) which is required for floating point arithmetic operations.
- Hence, we are using computationally efficient and light weight OCR in our system which will run smoothly on the mobile devices.

MSER- Maximally Stable Extremal Regions:-

MSER denotes a set of distinguished regions, which are defined by extremal property of its intensity function in the region and on its outer boundary [10]. In computer science, Maximally Stable Extremal Regions(MSER) are used for blob detection in scanned images. This technique was suggested by Matas et al. to find correspondences between image elements from two images with different perspectives. This method of extracting a ample number of analogous image elements contributes to the wide-baseline matching, and it has led to better stereo matching and object recognition algorithms. Extremal regions in this context have two important properties, they are as follows:

- 1) Continuous transformation of image coordinates. This means it is affine invariant and it doesn't matter if the image is warped or skewed.
- 2) Monotonic transformation of image intensities. The approach is of course sensitive to natural lighting effects as change of day light or moving shadows.

Advantages of MSER are-

- 1) Invariance to affine transformation of image intensities.
- 2) Stability: only regions whose support is nearly the same over a range of thresholds is selected.
- 3) The set of all extremal regions can be computed in worst-case $O(n)$, where n is the number of pixels in the image.

For region detection invariance transformations that should be considered are illumination changes, translation, rotation, scale and full affine transform (i.e. region should correspond to same preimage for different viewpoints. View point changes can be locally approximated by affine transform if assuming locally planar object and orthographic camera, that is perspective effects ignored). Region detection should be repeatable, stable and capable of discriminating between regions.

MSER is the method of blob detection in images. The MSER algorithm extracts from an image a number of co-variant regions called MSER. MSER is a stable connected component of gray level set of images. MSER is based on the idea of taking regions which stay nearly the same through a wide range of thresholds. All the pixels below a threshold are white pixels and all those above or equal are black pixels. If we are shown a sequence of threshold images I_t with frame t corresponding to threshold t , we would see first a *black* image, then *white* spots corresponding to local intensity will appear then grow larger. These *white* spots will eventually merge, until the whole image is *white*. The set of all related components in the order is the set of all extremal regions. Elliptical frames are attached to the MSERs by fitting ellipses to the regions. Those regions descriptors are saved as features. The word *extremal* refers to the property that all pixels inside the MSER have either higher (bright extremal region) or lower (dark extremal region) intensity than all the pixels on its outer boundary.

The operation can be accomplished by sorting all pixels by gray value and incrementally adding pixels to each connected component as the threshold is changed. The area is monitored. Regions such that their variation w.r.t the threshold is minimal are defined maximally stable.:

Let's make all the pixels below a threshold, white and all others, black. Considering a sequence of threshold images with increasing thresholds sweeping from black to white we pass from a black image to images where white blobs appear and grow larger by integration, up to the ultimate image. Over a enormous range of thresholds the native binarization is steady and shows some invariance to affine transformation of image intensities and scaling.

Text Translation:-

Our system is mainly divided into two parts, first one is text detection phase and second one is text translation phase. In text detection phase, we are using OCR and MSER together. In text translation phase, we are using text translation API. There are various free APIs available in market for the translation purpose like Microsoft Translator API, Google Translator API, Yandex Translator API, Frengley Translator API etc. Out of these APIs, some APIs are free and some APIs are paid. We have used the free API for the translation purpose in our System.

Microsoft Translator API:-

Microsoft Translator Text API [13], part of the Microsoft Cognitive Services API collection, is a cloud-based machine translation service supporting various multiple languages. This Translator can be used to build various applications, websites, tools, or any solution requiring multilanguage support.

It is Built for business. Microsoft Translator is a proven, customizable, and scalable technology for machine translation. Microsoft Translator technology powers translation features across Microsoft products, including Office, SharePoint, Yammer, Visual Studio, Bing, and Skype. By simply integrating translation into web, desktop, or mobile applications, the Translator API provides a rich functionality set for any developer. The features of Microsoft Translator API are- it is built for enterprise, customizable, effectively scalable.

Yandex Translator API:-

The Yandex Translator API [12] gives access to the Yandex online machine translation service. It provides support for more than 70 languages and can translate separate words or complete texts. The API makes it possible to use Yandex Translate in a mobile apps or web services for end users Or to translate large quantities of text, such as technical documentation. We can use any of the following interfaces for accessing the Yandex Translate API over HTTPS:

- ❖ XML interface (The response is an XML document).
- ❖ JSON interface (The response is JavaScript objects with the same names and semantics as the XML elements).
- ❖ JSONP interface (The response as JavaScript objects wrapped in a callback function with the specified name).

All the interfaces have the same functionality and use the same set of input parameters.

Yandex Translate is a statistical machine translation system. This system translates words, complete texts, and web pages into the desired language. It is available as a web service and mobile application, and is also used in other

products, such as translating web pages in Browser. Yandex Translate has an automated dictionary that sets it apart from the limited number of similar existing services. The technology, developed by a Yandex team of linguists and programmers, combines current statistical machine translation approaches with traditional linguistic tools.

Py-translate:-

Py-translate is a translation tool for humans. Its end goal is to translate text from one language to another. Translation is done in terminal using command line arguments. It is so good that it can translate a book in mere 5 seconds. It was made for python 3 but still it works on python2. It is fast & easy to install. It is easy to use & handle. It supports translation from any language. It is having highly composite interface, the power of Unix pipes & filters. It is having a simple API.

Comparison:-

Comparison of OCR and MSER

	OCR	MSER
Font Size	Fixed	Variable
Robustness	Less	More
Usability	Easy	Hard
Reliability	More	Less
Flexibility	Less	More
Time Complexity	Fast	Slow
Space Complexity	More	More

Comparison of Google API, Microsoft API and Yandex API

	Google Translate API	Microsoft Translator API	Yandex Translate API
Supported Request Format	Atom, GData	SOAP, XML, URI Query String/CRUD, JavaScript, HTTP POST, GET	XML, JSON, JSONP
Supported Response Format	GData, Atom, XML	XML, JSON, SOAP	XML, JSON
Is this Hypermedia API?	No	Yes	No
Languages Supported	100+	60	46
Functionality	Cloud Based API	Cloud Based API	Statistical Machine Translation

Conclusion:-

This paper focuses on the various methodologies that are used for text retrieval and translation from the images of the sign boards. By referring to the above sections, we can conclude that for the text retrieval, Optical Character Recognition(OCR) is better than any other methods for the digital images & Maximally Stable Extremal Regions(MSER) is best for natural scene images. As they produce the desired output with high accuracy. For the text translation, API services which are available from the Google are the best.

Future work aims at the development of a such system that performs text retrieval by using above methods and translation of that text from one language to another for the sign board images.

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RESEARCH ARTICLE

PREVALENCE OF DENTINAL HYPERSENSITIVITY IN DAKSHINA KANNADA POPULATION

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Key words:-

Dentine hypersensitivity, Dakshina
Kannada.

Abstract

The aim of the survey was to study the prevalence, common causes, aggravating factors and awareness about dentine hypersensitivity in urban and rural patients of Dakshina Kannada population. A total of 1000 patients were randomly selected, who reported at the A. B. Shetty Memorial Institute of Dental Sciences, Nitte University and Satellite Rural Centres. Dentine hypersensitivity was evaluated by questionnaire, oral and clinical examination. On analysis of data, it was found that the prevalence of dentine hypersensitivity was 19.2%. Cold was the most common (68.2%) aggravating factor and the most common cause was gingival recession (47.9%), followed by caries (24%). Most common age group affected by dentine hypersensitivity was between 21-30. It was also noted that population had limited awareness about the causes, symptoms and treatment modalities of dentine hypersensitivity.

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Introduction:-

Dentine hypersensitivity (DH) has been defined as a short, sharp pain arising from exposed dentine as a response to stimuli typically thermal, evaporative, tactile, osmotic or chemical and which cannot be ascribed to any other form of dental defect, disease or pathology [1, 2]. DH is a very common condition that is frequently encountered in clinical dental practice. It is a clinical condition in permanent teeth caused by dentin exposure to the oral environment as a consequence of loss of enamel and/ or cementum [3]. Several theories have been proposed to explain the mechanism of dentine sensitivity. Of these the most widely accepted theory is called hydrodynamic theory of sensitivity. This theory postulates that rapid shift in either direction of the fluid within the dentinal tubules following stimulus application, results in activation of sensory nerves in the pulp/inner dentine region of the tooth [4, 5]. Several studies indicate that DH was found in adult populations, with prevalence of more than 40% worldwide, 4 to 74% in India and 26% in southern India [6]. The rural population may have a lot of barriers preventing them to attend the dental clinics/institutes which are mainly situated in urban settings. A number of people with mild tooth sensitivity do not visit the dentist. Therefore, it may be difficult to get the actual prevalence of dentine hypersensitivity in the population. Patients usually do not report this painful condition to their dental practitioners and when they do, they report experiencing sharp pain after a number of stimuli's [7]. DH may occur also as a result of history of any dental treatment as well. DH may even result in emotional changes that alter

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behaviour, in extreme cases [8]. The purpose of this cross sectional study was to evaluate the prevalence, common causes, aggravating factors, association with previous dental treatment and patients' awareness about dentine hypersensitivity among the urban and rural patients of Dakshina Kannada population.

Materials and Methodology:-

The study was conducted on a total of 1000 patients over a period of 3 months from June to August 2016, after obtaining written consent, out of which 500 were examined from the Out-Patient section of Department of Conservative Dentistry and Endodontics and rest 500 were examined in Satellite Rural Centres of A. B. Shetty Memorial Institute of Dental Sciences, Nitte University, Deralakatte, Mangaluru.

Inclusion criteria:-

- Patients between the age of 15-70 years
- History of dentine hypersensitivity

Exclusion criteria:-

- Teeth with prosthodontic crowns
- Patients consuming analgesic drugs, anti-psychotic drugs or mood altering medications

Data obtained was entered in M S Excel spreadsheet and statistical analysis was done using the Statistical Package for the Social Sciences (SPSS 22). Difference between variables were analyzed using Pearson Chi-Square test.

Results:-

Out of 1000 patients, 192 patients showed a positive response to dentine hypersensitivity (prevalence is 19.2%). The prevalence was highest in the age group of 21-30 years (32.3%) [Fig.2]. Males (59.9%) were more affected than females (40.1%) [Fig.1]. Most common aggravating factors for DH is cold (68.2%), followed by sweet (27.6%) and hot (4.2%) [Table.1]. Most common causes found were gingival recession (47.9%), caries (24.0%), and tooth abrasion (19.8%) [Fig.3]. Out of the 192 cases that presented hypersensitivity, 75 cases (39.1%) had history of regular soft drinks consumption ($p=0.013$). Out of 1000 patients, only 38 cases reported of any recent history of dental treatment and 39.4% of them had undergone oral prophylaxis/root planing, 36.8% underwent orthodontic treatment including proximal stripping.

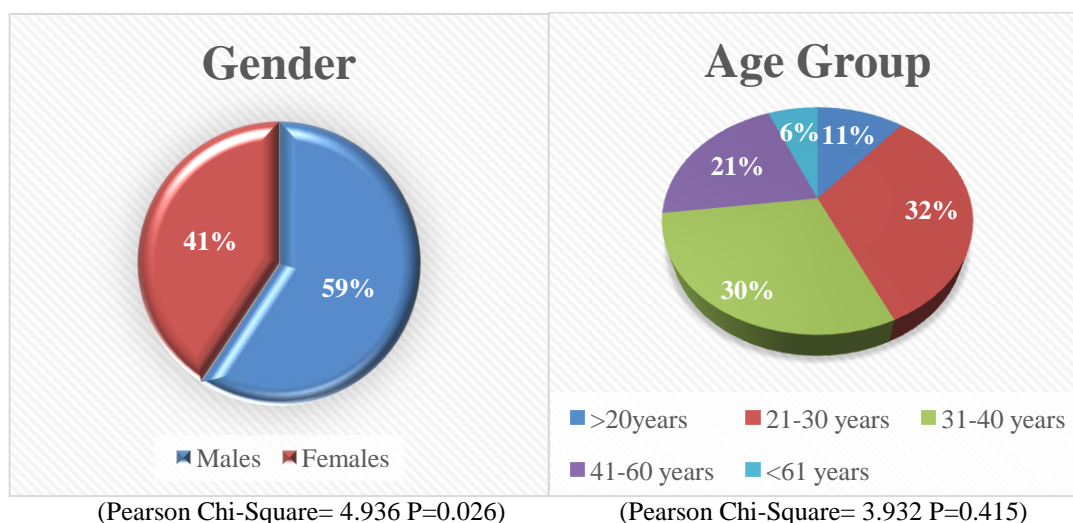


Figure 1:-
Gender distribution of hypersensitivity

Figure 2:-
Association of DH with age

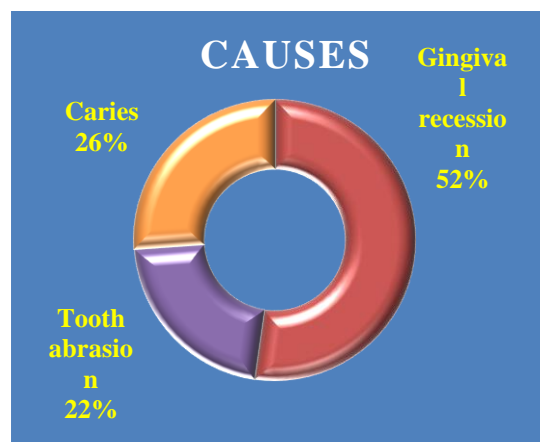


Figure 3:- Causes of DH.

Table 1:- Aggravating factors of DH.

Factors	Frequency	Percent
Hot	8	4.1
Cold	131	68.2
Sweet	53	27.6
Total	192	100.0

Discussion:-

According to the results of the questionnaire and clinical examination, the present study showed that the prevalence of dentine hypersensitivity was overall 19.2%, which is similar to the previous study done by the Hegde et al 22.5% [9] and Dhaliwal et al 25% [10]. Some studies that have been carried out with questionnaires had reported values of more than 50% or less than 20% [11, 16]. There are studies from dental practices which have reported prevalence values from 4-15% [12, 19]. The differences may be due to the type of sample and methods used in the studies. It could also be due to the variation in the geographical locations with different dietary habits and oral hygiene practices.

In the present study, dentine hypersensitivity is more prevalent in males than females as against studies done by Rees JS, Addy M [13], Hsin Cheng Liu [14] and N A Chrysanthakopoulos [15] which may be due to their overall healthcare and better oral hygiene awareness while the study done by Chabanski M B [17] is in accordance with this study.

In the present study dentine hypersensitivity is more prevalent in age group of 21-30 years and least above the age of 60. This particular age distribution may be because of more frequent and aggressive oral hygiene regimen adopted. A reduction of DH in seniors could be plausible due to the development of secondary or reparative dentine and subsequent sclerosis of the dentinal tubules accompanied by reduced sensitivity. Maxillary teeth seem to be more affected than mandibular teeth with 122 of the 192 patients affected by hypersensitivity complaining of the same in maxillary teeth compared to the 70 others who had hypersensitivity in the mandibular teeth. This is in accordance with study done by Addy M et al [18].

Several possible etiologic and predisposing factors for dentine hypersensitivity have been proposed [19]. In this study, gingival recession was seen to be the leading cause of dentinal hypersensitivity (52%). Dentine tubules may become exposed as a result of enamel loss from attrition, abrasion, erosion (acid dissolution) or abfraction (cervical stress lesion) [20], but dentin exposure often may be a result of gingival recession and cementum loss from root surfaces, most frequently in canines and premolars [21]. Aggressive or frequent tooth brushing [22] and periodontal treatment (such as scaling and root planning [23]) could also result in elevating DH.

Soft drinks had a direct association with dentine hypersensitivity in the present study, with 39.1% of the positive respondents to DH having a habit of regular consumption of soft drinks. The study had similar findings with Colak et al study among students in Turkey [8]. Soft drinks are carbonated that leads to tooth wear by erosion of enamel

surface and then dentine, leading to DH. According to studies done by S. Wongkhantee [24], ME Barbour [25] and Vanuspong W [26], acidic beverages and cola drinks depending on pH, temperature and frequency of exposure are responsible for softening of dental hard tissues and restorative materials that are exposed.

Cold was the most common factor which initiated the pain as experienced by most of the patients (68.2%). This result was similar to the studies done by Rees JS et al & Tan CS et al [27]. Sweet stimulus was the second most common aggravating factor (27.6%), which was in accordance with study done by J.S Dhaliwal et al [10] but not in agreement with studies done by Fischer et al [3], Rees JS et al [13], Flynn J et al [28] where heat was found to be the second most common aggravating factor for DH as pain from heat takes longer to develop than pain from cold, because heat causes inward movement of tubular fluid while the outward movement caused by cold develops more rapidly.

Conclusion:-

The prevalence of dentinal hypersensitivity among Dakshina Kannada population is 19.2%. Dentine hypersensitivity is a clinically relevant and worldwide problem that may affect about a quarter of the adult population. Even though dentine hypersensitivity may not be considered a fatal or severe problem, it affects the quality of life of patients and, therefore, it should be properly addressed in research, dental education, prevention, and treatment.

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RESEARCH ARTICLE

SYSTEMATIC VEHICLE TRACE WITH RELAY CONTROL AND PULSE SURVEILLANCE.

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Abstract

Modern technological development in automobile industry has drastically improved people's way of living; therefore there is a need for monitoring of vehicles to heighten its longevity. Mortality rate is also on the rise due to the lack of timely and proper medical attention. Continuous health monitoring of patients using wearable and implantable body sensor networks will increase detection of emergency conditions during health crisis. By integrating vehicle tracking and continuous wireless patient monitoring system; we can enhance safety during travel. This system is user friendly, easily installable, easily accessible and economically viable. The conventional system of tracking using battery operated radio frequency identification (RFID) is only suitable for small range. To overcome this, GPS and GSM technologies are used which will receive the location coordinates and provides real-time information which can be observed from any other remote location. The established health monitoring system is not compact and understanding the output analog signals is a tedious task. Hence, in the designed health monitoring system patient's physiological signals are acquired by the sensors attached on the patient body; if there are any abnormalities then a notification is sent and the vehicle is turned OFF using relay.

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Introduction:-

Advanced ingenuity has proved that we are living in the golden era of science where for any anomaly some kind of solution exists. Transportation is a vital activity in everyone's life; we can't avoid it just because of the fact that daily 400 accidents occur. However, the issue that's not getting any attention is the growing number of people having heart attacks while driving. According to a Times of India article published in the year 2016 India is witnessing two million heart attacks in a year, one person for every 33sec dies of cardiac arrest and majority of them are youngsters. An ambulance must be called within five minutes after the patient collapses as part of an emergency treatment. Although it seems like an easy thing to do many obstacles are there the main being lack of surveillance of

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the patient's heartbeat (pulse rate). Health monitoring technology has been flourishing proficiently in recent years, due to its ubiquity, accessibility and ease to use. However, quality and affordability of the health care systems are a major cause of concern around the globe. A large number of people with low income facing health issues with the high cost of healthcare system. Between 2004 and 2014 medical expenditure per hospitalization for urban patients increased by about 176%. Early detection can always prevent long term hospitalization in order to this we need to use a device such as cell phone that has become a part of everyone's identity so that it is affordable for everyone be it people in rural or urban area. That is why in our proposed system mobile healthcare monitoring system was developed.

Wireless Technologies is all the hype due to the invention of low-power electronics, internet of things (IOT) and wearable sensor systems. A heartbeat sensor is a great tool giving you clear indication and evaluation of the condition of your cardiovascular system. There are many devices used by doctors to monitor patient's vital parameters such as electrocardiography, Holter Monitor, long term ambulatory monitors and pulse rate oximeters. Though they are proficient they can't be put to daily use since it restricts mobility of person and can't be possibly bought by an average person. This is the main reason why we use heart beat sensors because they are efficient, cost-effective and doesn't affect movement.

In the last decade, we observe the drivers fatigue driving and vehicle theft activity has exponentially increased which causes social real time problem, accidents and many more hazards conditions. These incidents are widely prevalent and have affected our day to day activities causing security concerns. So there is a need for monitoring and tracking the vehicle and also storing and updating database with relevant information which can have future impact. In our proposed system, there is prevention of occurrence of accident due to sudden health crisis and to develop a remote control mechanism that will avoid loss of control of vehicle when the driver collapses. A driver of any kind of vehicle is responsible to not lose control of his vehicle and cause harm to innocent passersby. However, what if a driver causes personal injuries to others because of unconsciousness due to cardiac arrest that is why a relay control is used which automatically turns down the engine when a turn OFF command comes from the command centre as soon as the drivers pulse rate deteriorates.

Problem Statement:-

In conventional system, Radio Frequency Identification (RFID) was predominantly used. It is a small electronic device that consists of a small chip and an antenna. This device serves the same purpose as a code on the back of a credit card or ATM card; it provides a unique identifier for the object. And, just as a code must be scanned to get the information, the device must be scanned to retrieve the identifying information and used in short range and less effective. Since this system make use of the electromagnetic spectrum, they are relatively easy to jam using energy at the particular frequency. Reader collision occurs when the signals from two or more readers overlap. Tag collision occurs when many tags are present in a small area; since the tags can be read without being swiped or obviously scanned like a code, anyone with this tag reader can read the tags embedded in your clothes and other consumer products without your knowledge. For various reasons, the reader/tag systems are designed so that distance between the tag and the reader is kept to a minimum to prevent tag collision. However, a high-gain antenna can be used to read the tags from much far away, leading to privacy problems.

The conventional health monitoring requires large equipment which are costly and causes loses of mobility of patients. Monitoring of vital parameters needs various devices and can include measuring blood pressure and heart rate, and preferably also pulse oximetry and respiratory rate. Multimodal monitors that simultaneously measure and display the relevant vital parameters are commonly integrated into the bedside monitors in critical care units, in rooms. So, this leads to high expenditure of money and extreme consumption of space. Patients are not satisfied with the treatment which doctors normally use for finding the heartbeat count. There are instruments available in the market to track internal body changes, but there are some limitations due to their heavy cost. Conventional monitoring of cardiac activity is performed in a clinical setting in real-time during a visit, by recording electrocardiograph (ECG) signals. Monitoring the heart activity with ECG signals is a very common technique, performed by placing at least three electrodes on the skin to measure the electrical activity of heart. Traditionally, Holter monitors were used for ambulatory monitoring during the recovery period after cardiac surgeries Although Holter monitors are capable of providing continuous monitoring, the central unit of the monitors is bulky and each electrode is connected to the central unit with wires. Therefore, the use of the these monitor interrupts the daily routine of the patient and is not feasible for continuous monitoring. Over the few years, with the advancement in wireless technologies, these monitors have been miniaturized and evolved into complete wire-free monitoring

devices. Although ambulatory wire-free devices look for promising continuous monitoring, there is a still need for development of such devices.

Usage of other controller will lead to difficulty in manipulation which is not user friendly. ARM chip is RISC processors that were created by Acorn and the design now owned by ARM Limited. They license the design to manufactures to add on to their own chips to create “systems on a chip” that contain many features on one chip, and with the ARM design which is able to include the CPU as well. The ARM is very prevalent in low power embedded applications. They are designed for low power consumption, a laptop using an ARM processor which is going to possibly have better battery life than an x86 based laptop.

The disadvantage of the ARM processor is that it is not binary compatible with x86. For the little cost, the Raspberry Pi is powerful, but it has some limitations. Although you probably use it as a desktop computer, its power is closer to a mobile device (like a tablet) than a modern desktop PC. Even though Raspberry Pi can perform different tasks, there are some limitations due to the hardware. Because of its processor, it cannot run X86 operating systems. Some common area like Windows and Linux is not compatible. In addition, some applications which require a high demand on CPU processing is off-limits. The memory of the Raspberry Pi is more limited than the usual one, with just 512MB or 256MB available. You can't expand that with extra memory as the desktop PC.

Mechanism Used:-

Vehicle Tracking using GPS&GSM:-

In **GPS tracking system** the location of vehicle is sent to remote place and it is indicated by GSM modem. Global Positioning System (GPS) modem requires minimum 3 satellites to calculate the exact location. This modem communicates only in one way with 8051 microcontroller. This means that it can only transmit data to microcontroller. GPS Modem will not receive any data from microcontroller. At the same time GPS modem does not send any data to Satellite, it only receives signal from satellites. GSM modem is used to send this information via SMS. SMS will be sent to the owner of the vehicle.

It would be time consuming to track location on maps. But now various websites and apps are available on internet which shows online map. Google maps are one of the main and website. You can use any one of these websites to track the location of vehicle. You can track the location using Longitude and Latitude received in SMS. Using these maps you can track vehicle and get the exact location as well as directions. All you need to do is copy the contents of Text SMS received from SMS based Vehicle tracking system using GPS into the Google map.

GPS based Vehicle tracking system is required in many arena, like in case of car theft detection. Car tracking using GPS will be useful when our car is stolen. Also if somebody wants to track school bus of their children, it will be helpful to find out the location of their kids. One more situation is when some company wants to track the location of the cab or transport bus of employee then in that case, this vehicle tracking system will be very useful. This is a cheaper solution than a two-way GPS communication system where the communication is done in both ways with GPS satellites, GSM modem with a SIM card used here implements the same communication technique as in a regular cell phone.

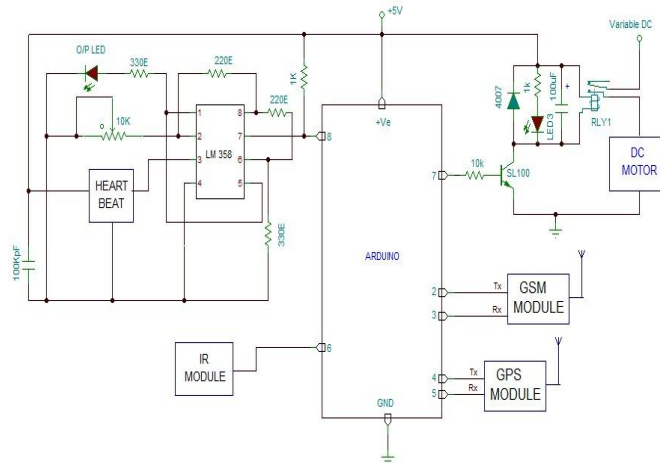


Fig 1:- Circuit diagram of System.

+

Measurement of vital parameters using heartbeat sensors for health monitoring:-

The adoption of mobile health-care technology is a promising one to enhance the quality of life of patients and the elderly. Furthermore, it offers the potential to alter the current health-care system by enabling out-patient care and preventing hospitalisations. Designing a telemetry system for health monitoring is very cumbersome task. There are many key issues to be addressed, including: designing sensors; ensuring the transmission of vital sign data; providing privacy and security of an individuals. The system Heart Attack Detection by Heart Rate Monitoring helps to inform if a person is about to have a heart attack. This system is done by detecting the heart beat level and informs as soon as the heart beat level fall within the permissible limit. Thus this system can be used to save the life of many people as it alerts the doctor about the patient's heart beat level. For this the system has two circuits. One is the transmitting circuit which is the patient's one and the other is the receiver circuit which is being supervised in the control room.

The heartbeat sensor is based on the principle of photo phlethysmography. It measures the changes in volume of blood through any organ of the body which causes a change in the light intensity in that particular organ. In case of applications where heart pulse rate is to be monitored, the timing of the pulses is important. The flow of blood volume is decided by the rate of heart pulses and as the light is absorbed by blood, the signal pulses are equivalent to the heart beat pulses. The basic heartbeat sensor consists of a light emitting diode and a detector. The heart beat pulses causes a variation in the flow of blood to different parts of the body. When a tissue is illuminated with the light source, the light emitted by the led, it either reflects or transmits the light. Some of the light is absorbed by the blood and one which is transmitted or the reflected light is received by the light detector. The amount of light absorbed depends upon the blood volume in that tissue. The detector output is of the form of electrical signal and is proportional to the heart beat rate. The signal is actually a DC signal relating to the tissues and the blood volume and the AC component is in phase with the heart beat. Thus the major requirement is to isolate the AC component.

Control of vehicle using relay driving circuit:-

One easy way to increase system reliability and performance is by using relays to switching devices (lights, fuel pumps, fans, etc.). A relay is an electro-mechanical switch. An electro-magnet is used to pull a set of contacts, or pins. The relay uses a tiny amount of current to trigger the larger switch in the relay, which is far efficient and safer. A perfectly wired relay will provide the shortest electrical path between the battery and the device which is controlled by the relay. Combined with the proper gauge wire, this will minimize the voltage drop between the battery and the device, allowing it to function at a peak performance levels. Relays, allow you to control a number of devices with a single switch (a master ignition switch on a race car). Having only one switch to turn off is safer in an emergency and it's more convenient as well.

System Specification:-

Relay:-

A relay coil is an electromagnet and is also an inductor. When power is applied to the coil the current in the coil builds up and levels at its rated current (depends on the DC coil resistance, $I = V/R$). Some of the remaining energy is now stored in the coil's magnetic field ($E = 0.5LI^2$). When the current flowing in the coil is turned off this stored energy needs a way out. The voltage across the coil quickly increases to keep the current in the coil flowing in the same direction ($V = Ldi/dt$). This voltage spike can reach high range of volts and can damage electronic parts.

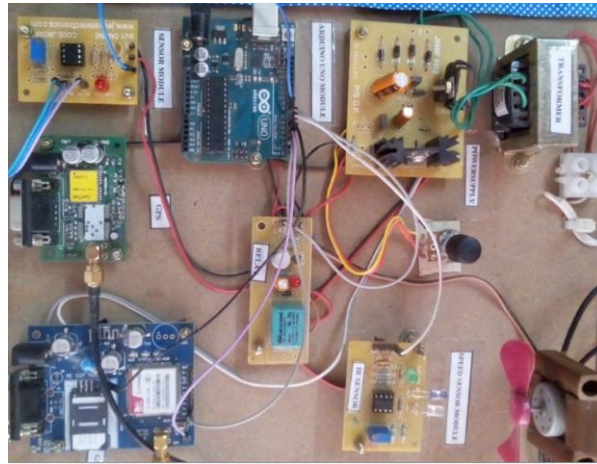


Fig 2:- Experimental setup of Proposed System.

By the addition of a fly back diode the current has a path to flow through coil until the stored energy is used up. The voltage across the coil is clamped to about 0.7V protecting the electronics. The stored energy dissipates at a fast rate in the diode ($E = V \cdot I \cdot t$). The current flow is interrupted and the relay is turned off. The diode should be able to deal the coil current for a short time and switch relatively fast. A resistor or zener diode is connected in series with the diode to dissipate the stored energy quicker. This result in the increase of amplitude of the voltage spike above 0.7V but the energy is used up quicker (i.e. the voltage spike is short lived).

Mc78xx/Lm78xx/Mc78xxa:-

This type employs internal current limiting, thermal shut down and safe operating area protection, making it durable. If tolerable heat sinking is provided, 1A output current can be obtained. Although initially it was designed to be as fixed voltage regulators, these devices can be used with external components to obtain adjustable voltages and currents.

GPS:-

GPS is the space-based satellite navigation system which provides location and time information in all weather conditions, anywhere on the Earth. GPS receivers are popularly used for navigation, positioning, and other research purposes. The GPS consists of satellites which orbits the earth. These satellites are geosynchronous with an orbital period that is the same as the Earth's rotation period. So they maintain exactly the same position with respect to the earth. All the GPS satellite transmits radio signals, which are then captured by the GPS receiver and used to calculate its geographical position. The minimum number of four satellites may be required to compute the four dimensions of X, Y, Z (latitude, longitude and elevation) and time. GPS receiver converts the received signals into position and estimates time and some information depending on the application and requirements. GPS determines the distance between a GPS satellite and the GPS receiver by measuring the amount of time taken by a radio signal to travel from the satellite to the receiver. To obtain accurate information, the satellites and the receiver uses very accurate clocks, which are synchronised so that they generate the same code at exactly the same time

GSM:-

The overall system definition for GSM describes not only the air interface but also the network. By adopting this approach it is possible to define the operation of the whole network.

GSM cellular technology has 200 kHz RF channels. These are time division multiplexed to enable up to eight users to access each carrier.

- It has good subjective speech quality
- It should have a low or terminal cost
- Terminals should be able to be handheld
- The system is to support international roaming
- It should offer good spectral efficiency
- The system should offer ISDN compatibility

The base transceiver station is organized into small groups, controlled by a base station controller which is typically co-located with one of the BTS. The BSC with its associated BTSs is termed the base station subsystem.

Further into the core network is the main switching area. This is known as the mobile switching centre. Associated with it is are the location registers, namely the home location register and the visitor location register which track the location of mobiles and enable calls to be routed to them. Additionally there is the Authentication Centre, and the Equipment Identify Register that is used in authenticating the mobile before it is allowed to the network and for billing.

Often termed the mobile equipment, this is the one which end user sees. One important feature that was first implemented on GSM was the use of a Subscriber Identity Module. This card carries with it the user's identity and other information to allow the user to upgrade a phone, while retaining the same identity on the network. It was also used to store other information such as "phone book" and other items. This item allows people to change phones very easily, and this has fuelled the phone manufacturing industry and enabled new phones with additional features to be launched.

Speech or voice calls are obviously the primary function for the GSM cellular system. To achieve this speech is digitally encoded and later decoded. A varieties of coders are available for use, being aimed at different sectors.

In addition to the voice services, GSM cellular technology supports a variety of other data services. A variety of data services were supported with user data rated up to 9.6 kbps. Services including Group 3 facsimile, videotext is also been supported.

One service that has grown at a rapid rate is the short message service. Developed as part of the GSM specification, it has been added into other cellular technologies. This service has become particularly popular.

Arduino Uno Controller:-

Arduino Uno is a microcontroller board based on the ATmega328. It has 14 digital input/output pins, 6 analog inputs, a 16 MHz quartz crystal, a USB connection, a power jack, an ICSP header and a reset button. It contains everything needed to support the microcontroller; simply connect it to a computer with a USB cable or t with a AC-to-DC adapter or battery to get started. The Uno board and version 1.0 of Arduino Software (IDE) were the reference versions of Arduino, now evolved to newer releases. The Uno board is the first in a series of USB Arduino boards, and the reference model for the Arduino platform for an extensive list of current, past or outdated boards see the Arduino index of board.

Working Principle:-

This project has a different approach. Arduino is used for controlling whole the process with a **GPS Receiver and GSM module**. GPS Receiver is used for detecting coordinates of the vehicle, GSM module is used for sending the coordinates to user through SMS. And an optional 16x2 LCD is also used for displaying status messages or coordinates. We have used GPS Module SKG13BL and GSM Module SIM900A. When hardware is ready then after programming, we can install it in our vehicle and power it up. This system has Global Positioning System (GPS) which will receive the latitude and longitude coordinates from the satellites among other c information. These co-ordinates are sent to the Arduino controller.

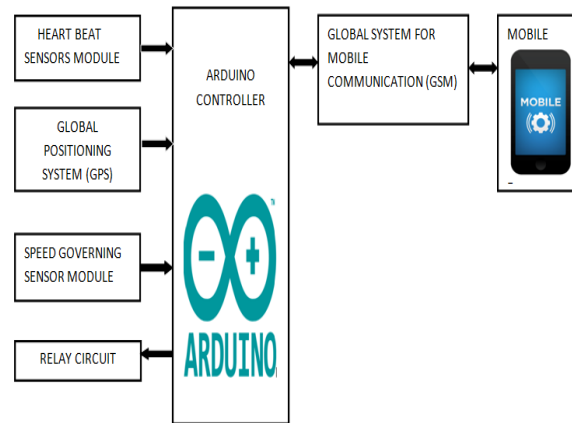
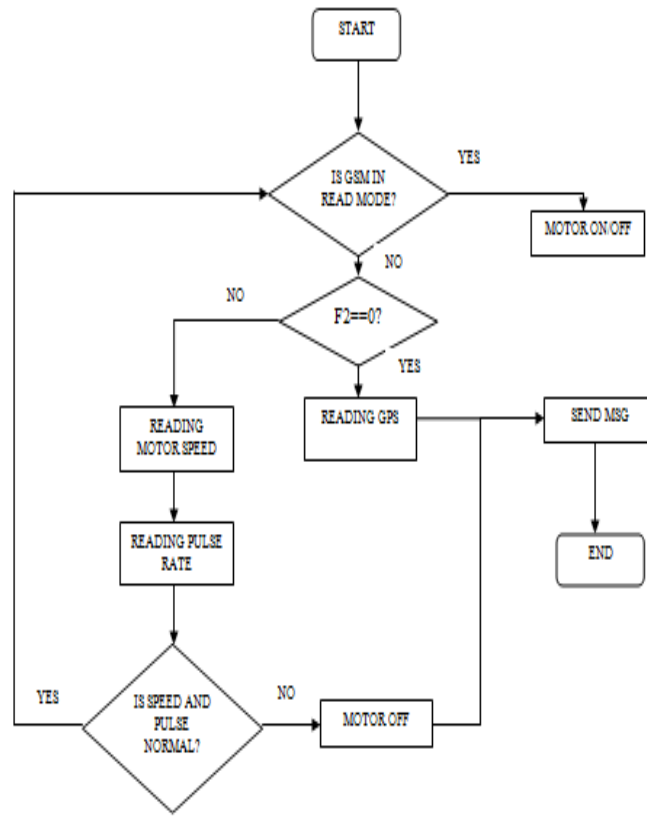


Fig 3:- Block Diagram Of Proposed Model.

This proposed model consists of two sensor modules namely, heartbeat sensor and speed governing sensors. The driver is provided with the wearable device which is used to monitor the driver's health conditions which consists of a heartbeat monitor sensors which can be used to measure the driver's pulse rate and blood pressure level during various conditions. So in order to detect the pulse we will pass LED from one side of the finger and measure the intensity of light received on the other side using a Light Dependant Resistor. As a result the value of resistance of the LDR increases. This variation in resistance is converted into voltage using a signal conditioning circuit usually an OP-AMP. On the other hand, the speed governing sensor module senses the speed of the vehicle and transmits it to the controller using the IR sensor. IR Sensors work by using a specific light sensor to select light wavelength in the Infra-Red spectrum. By using an LED which produces light at the same wavelength as the sensor is, you can look at the intensity of the received light. When an object is close to the sensor, the light from the LED bounces off the object back into the light sensor. This results in a large jump in the intensity that can be detected using a threshold. An infrared sensor is an electronic device that emits in order to sense some aspects around the surroundings. An IR sensor can measure the heat of an object as well as detects the motion. Usually in the infrared spectrum, all the objects radiate some form of thermal radiations. The emitter is simply an IR and the detector is simply an IR photodiode which is sensitive to IR light of the same wavelength as that emitted by the IR LED. Whenever the IR light falls on the photodiode, the resistances and these output voltages, change in proportion in accordance to the magnitude of the IR light received.

Arduino's Analog to Digital Converter converts them to the digital form. This digital data is transmitted to the cellular device through the Global System for Mobile Communication (GSM). The cellular device gets a Short Messaging Service (SMS) which comprises of position, speed of vehicle and driver's psychological data like pulse rate and blood pressure level. By monitoring this data, we can ensure the safety of the vehicle and the passengers. In case of abnormal pulse rate or blood pressure the control centre is given a alert message and the vehicle can be brought to halt using a relay connected to the engine. Relays are switches that open and close circuits electromechanically or electronically. Relays control one electrical circuit by opening and closing contacts in another circuit. A relays useful life depends upon its contacts. Once contacts burn out, the relays contacts or the entire relay has to be replaced. Thus by implementing this system we can tremendously reduce the human loss and material loss

Flowchart:-**Result:-**

```

File Edit Sketch Tools Help
sketch_feb09a $
delay(500);
if(a==5)
{
  gsm.println("AT+CMGR=1");
  while(gsm.read()!='+');
  while(gsm.read()!=' ');
  while(gsm.read()!=' ');
  while(gsm.read()!=' ');
  while(gsm.read()!='\r');
  delay(200);
  Y[1]=gsm.read();
  delay(200);
  Y[2]=gsm.read();
  delay(200);
  Y[3]=gsm.read();
  delay(1000);
  if(Y[2]=='0' && Y[3]=='N')
  {digitalWrite(rxly, HIGH);}
  if(Y[2]=='0' && Y[3]=='F')
  {digitalWrite(rxly, LOW);}
}

```

Fig 4:- Arduino Coding.

An emergency notification system refers to a collection of methods that facilitate the one-way dissemination or broadcast of messages to one or many groups of people with the details of an occurring or pending emergency situation.

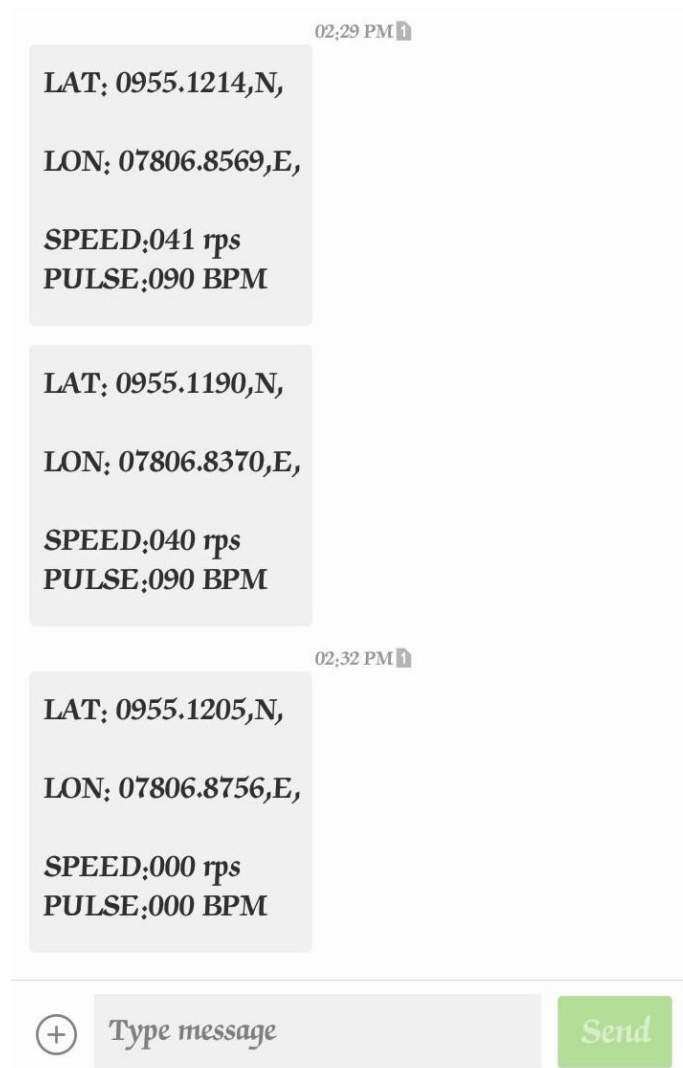


Fig 5:- Screenshot of the Message Received.

Conclusion:-

Our proposed system has the following features:-

- You can locate your stolen vehicle easily using your mobile without any extra cost.
- It can be used for vehicles carrying valuable goods, to keep track of the status of delivery and location
- You can also use it to keep tab on your driver. It reduces vehicle abuse and ultimately results in significant cost-savings for individuals, fleet owners and the like.
- Inform the location and route travelled by a vehicle using Global Positioning System (GPS) and Global System for Mobile Communication (GSM).
- Remotely monitor various vital parameters such as heartbeat rate and blood pressure level using sensors.
- Prevents occurrence of accident due to sudden health crisis.
- A remote control mechanism that will avoid loss of control of vehicle when the driver collapses.
- It is user friendly as it is easily available in human platform for anytime usage.
- GPS and GSM technology is suitable for long range communication. Using cellular device for transmitting purpose neglects the usage of RFID tags

Later Innovation:-

- Use of the advanced technology can be further enhanced by the use of camera and by developing a mobile based application.
- For diabetic patients, an artificial pancreas which is a device designed automatically adjusts blood sugar level.

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RESEARCH ARTICLE

EVALUATION OF AGD IN DIGITAL BREAST TOMOSYNTHESIS RELATIVE TO THOSE IN TWO-VIEW-FULL-FIELD DIGITAL MAMMOGRAPHY.

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Abstract

Digital Breast Tomosynthesis (DBT) is performing in high resolution limited angle tomography at radiographic dose levels. Advanced system facilitates the DBT along with 2 view Full Field Digital mammography (FFDM). The objectives of this study were to compare the Average Glandular Dose (AGD) of DBT and FFDM and to calculate the percentage of radiation dose reduction when using DBT with compare to FFDM. The study was carried out using data base of DBT system in a private hospital. In the investigation we analyzed dose of 251 patients who underwent mammographic examinations of both FFDM and DBT. All data analyses were done using IBM SPSS statistical software version 20.0. P-value 0.05 was considered as statistically significant. The explanatory variables were age, breast thickness, kVp, mAs, target/filter combination, and AGD values of DBT and FFDM. Mean values for the patient age and compressed breast thickness were 50 years and 49 mm (± 11.9 SD) respectively. The majority of the images were acquired using W/Rh target/filter combination and 51% patients came for the diagnostic mammograms and 49% for screening mammograms. A wide kVp range was observed for DBT than FFDM while mAs range was lower in DBT. According to the results total average glandular dose (TAGD) from FFDM and DBT for diagnostic was 4.21mGy (± 1.46 SD), for screening 4.04 (± 1.31 SD) and there was a statistically significant difference between mean values of TAGD. Mean AGD for Mediolateral Oblique (MLO) view in DBT was 2.05mGy (± 0.60 SD), in FFDM 2.73mGy (± 1.02 SD). AGD for Craniocaudal (CC) view in DBT was 1.63mGy (± 0.36 SD) and for FFDM it was 1.83mGy (± 0.66 SD). AGD to the breast from DBT was significantly lower than that for FFDM while range was lower in FFDM than DBT. There was a significant difference between mean values of CC and MLO views in DBT and FFDM ($P < 0.05$). It was evident that AGD from DBT was lower than that for FFDM further AGD was reduced by 55.3% by using DBT with compared to TAGD,

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using both FFDM and DBT together for same patient and AGD was reduced 19.19% by using DBT with compared to AGD from FFDM.

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Introduction:-

Mammography is considered to be the most effective method for the early detection of breast cancer^[1]. Although mammographic process is using low energy x-rays to examine the human breast the use of ionizing radiation implies the risk of inducing fatal breast cancer^[2]. Minimizing radiation risk is important in general as manifested by the as low as reasonably achievable (ALARA) principle.^{[3][4]}

As glandular tissue is radiosensitive organ, the average glandular dose (AGD) to the breast is the most important quantity to estimate the risk of radiation-induced carcinogenesis from mammography. AGD is currently accepted as an estimation of the patient dose in mammography.^[4]

Digital Breast Tomosynthesis (DBT) is a FDA approved radiological imaging method for breast imaging^[1]. It performs high resolution limited angle tomography at radiographic dose levels^{[5][6]}. DBT allows the detection of a greater number of expansion lesions and a better morphological analysis of masses and architectural distortions^[7]. Advanced system facilitates the DBT along with two view Full Field Digital mammography (FFDM)^{[8][9]}. Therefore the purpose of the study was to compare the Average Glandular Dose (AGD) of DBT and FFDM and to calculate the percentage of radiation dose reduction when using DBT with compare to FFDM.

Methodology:-

The study was carried out using data base of Digital Breast Tomosynthesis system at a private hospital, Colombo. This was a quantitative, retrospective research project. The study sample was included 251 patients exposure parameters. Data was collected through machine storage in the period of 01.01.2016 to 01.04.2016. KV, mAs, thickness, age, force, view, AGD, ESD, Exposure Index (EI) was recorded for each individual subject in to excel sheet. The data was analyzed by SPSS 20.0 software with $P < 0.05$ indicating level of significance and using descriptive statistics.

Results:-

Below table, contain the variable measurements of Full field digital mammography (FFDM/2D imaging) and Digital breast tomosynthesis (DBT/3D imaging).

	Variables	Mean	Standard deviation	Minimum	Maximum	Range
FFDM/2D imaging	Age	50.492	9.720	32.000	87.000	55.000
	HVL	1.083	1.690	0.486	559.000	558.514
	kVp	28.695	5.342	21.000	521.00	283.00
	mAs	171.30	64.71	58.00	415.00	357.00
	Thickness(mm)	309.28	11.999	14.000	93.000	79.000
	Force(lb)	25.180	60.66	238.00	51.900	41.400
	AGD(mGy)	2.2834	0.9675	0.3400	6.5200	6.1800
	ESD(mGy)	8.340	4.811	1.800	30.430	28.630
DBT/3D Imaging	EI(Exposure index)	309.28	60.66	238.00	4449.0	283.00
	Kv	31.927	2.518	27.000	48.000	21.000
	mAs	102.65	18.88	60.60	249.00	188.40
	AGD	1440.5	0.5416	1.1500	9.2100	8.0600
	ESD	5.8203	474.6	814.0	14.8800	13.8000
	EI	31.927	2.0968	1.0800	521.00	3635.0

Total average glandular dose (TAGD) from FFDM and DBT for diagnostic was 4.21mGy (± 1.46 SD), for screening 4.04(± 1.31 SD) and there was a statistically significant difference between mean values of AGD. Mean AGD for

Mediolateral Oblique (MLO) view in DBT was 2.05mGy (± 0.60 SD), in FFDM 2.73mGy (± 1.02 SD). AGD for Craniocaudal (CC) view in DBT was 1.63mGy (± 0.36 SD) and for FFDM it was 1.83mGy (± 0.66 SD). AGD to the breast from DBT was significantly lower than that for FFDM while range was lower in FFDM than DBT. There was a significant difference between mean values of CC and MLO views in DBT and FFDM ($P < 0.05$).

Relationship of average glandular dose with breast thickness for FFDM and DBT

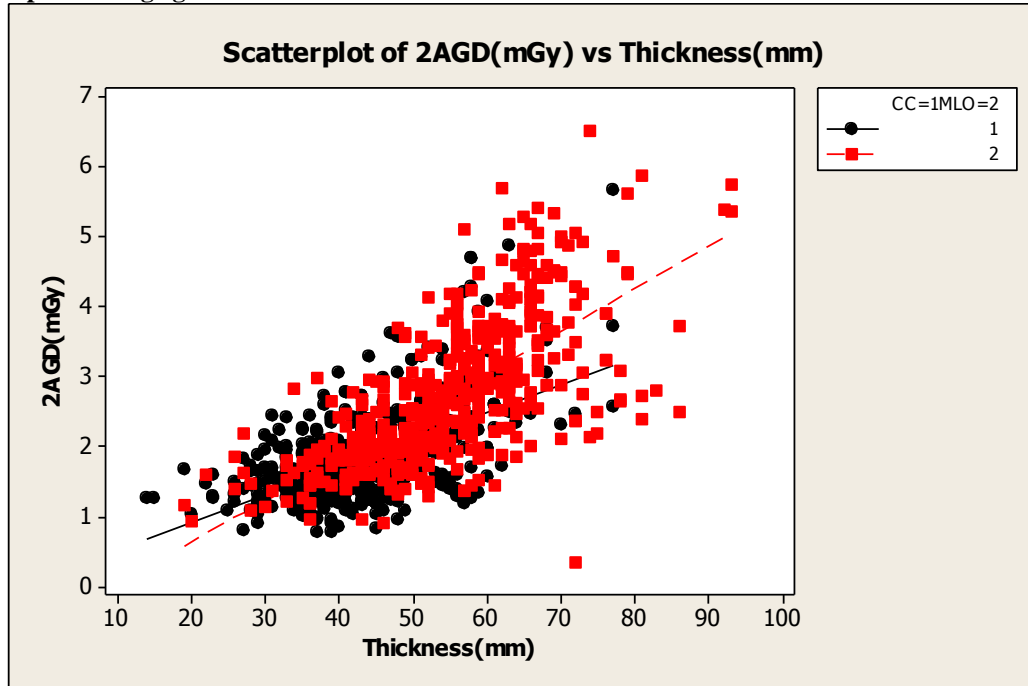


Figure 1:- AGD Vs Breast thickness in FFDM(2D imaging).

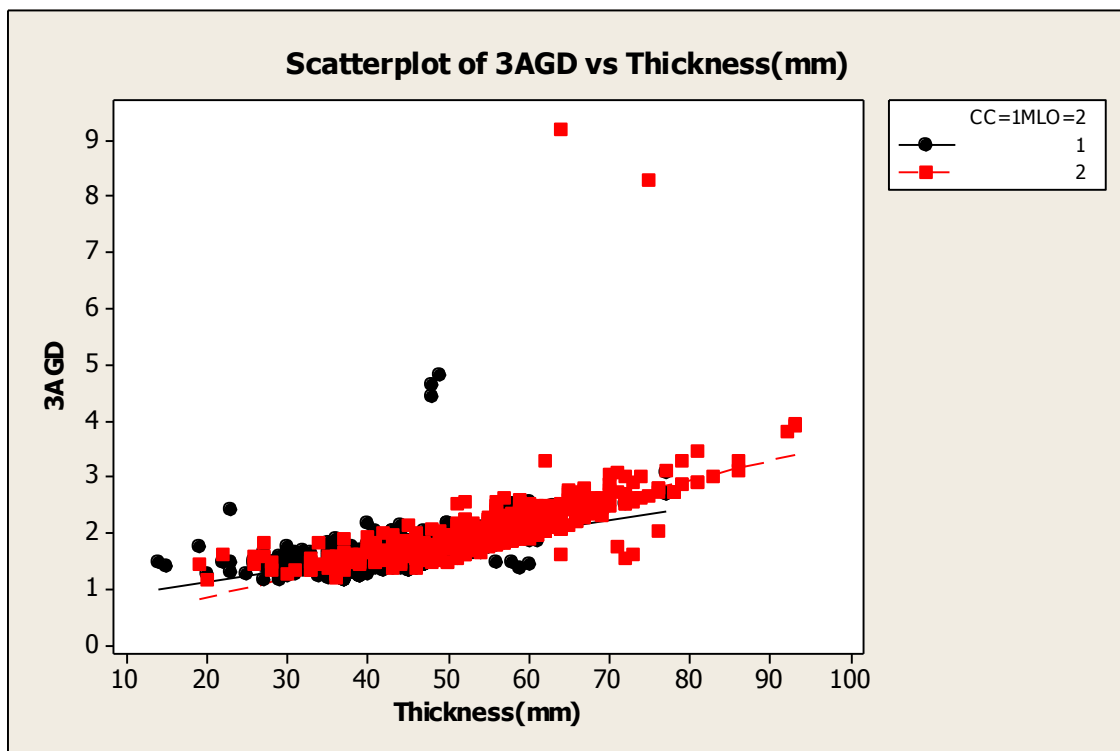


Figure 2:- AGD Vs Breast thickness in DBT(3D Imaging)

The above graphs show positive linear regression between AGD and thickness for DBT and FFDM in both views

Relationship of average glandular dose with age for FFDM and DBT

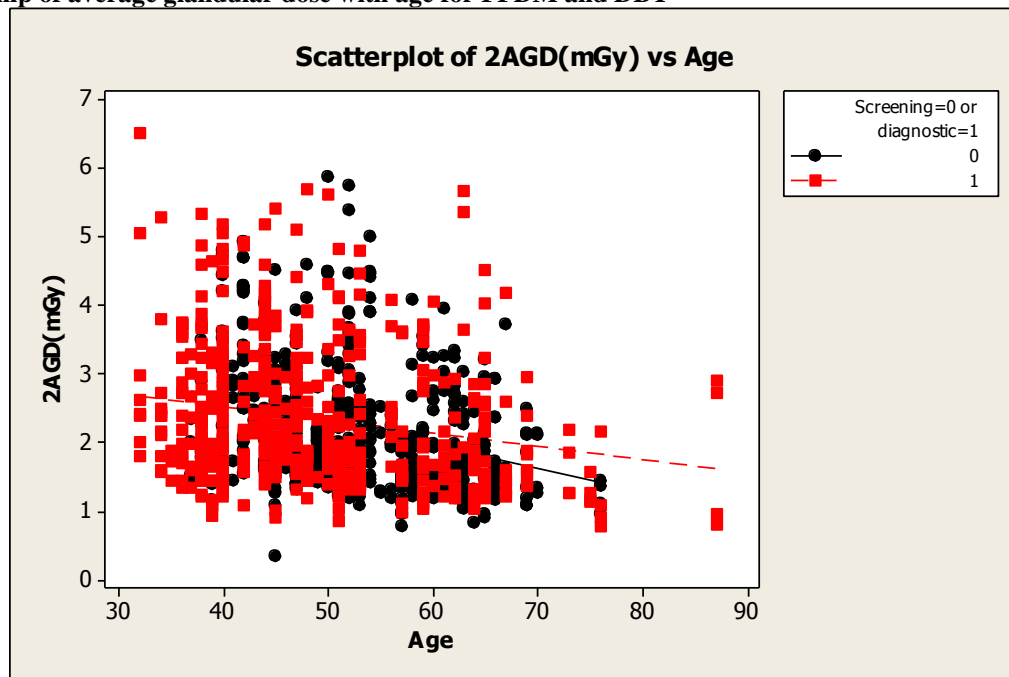


Figure 3:- AGD Vs age in FFDM (2D imaging).

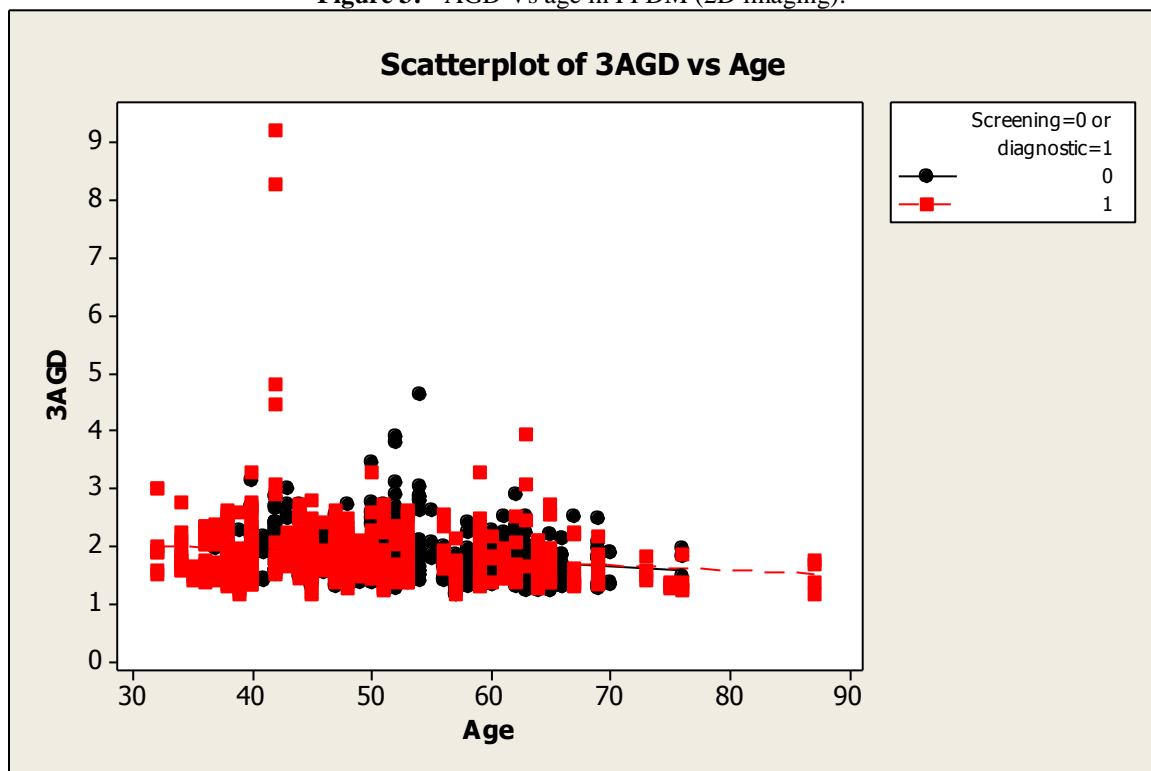


Figure 4:- AGD Vs age in DBT (3D Imaging).

The above graphs show negative linear regression between AGD and age for both DBT and FFDM in both views.

Discussion:-

Ionizing radiation such as x-rays widely used in radiological examination. Hence there should be a special concern on radiosensitive organs which are exposed to direct x-ray beam. In mammography average glandular dose should be kept minimum due to high radiosensitivity of mammary glands. Chevalier M et al states that TAGD was 3.8mGy and it slightly lower than TAGD from current study. Further mean age and thickness from both study were similar.^[9] In both study AGD from CC view was lower than MLO due to thickness of the CC view lower than MLO.

Total average glandular dose (TAGD) from FFDM and DBT for diagnostic was significantly higher than that for screening owing to dose is directly proportional to mAs and screening category has much lower mAs value than diagnostic^[1]. And it was ensure that negative linear regression between AGD and age for both DBT and FFDM of screening and diagnostic categories due to the fatty tissue of the breasts may become more prominent than the glandular tissue as women ages.

According to the results mean average glandular dose to the breast from DBT was significantly lower than that for FFDM. It was evident that AGD from DBT was lower than that for FFDM. Which is similar to the study of Svahn TM et al but controversial to the study of Olgar T et al^[3]. Further AGD was reduced by 55.3% by using DBT with compared to TAGD, using both FFDM and DBT together for same patient and AGD was reduced 19.19% by using DBT with compared to AGD from FFDM.

Conclusion:-

The dose values delivered in breast DBT(3D imaging) were lower than in FFDM (2D imaging) mode. Hence DBT (3D imaging) has superior AGD reduction to that of FFDM(2D imaging) in the diagnostic and screening settings. DBT (3D imaging) only can use for screening programme in order to minimize the AGD.

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RESEARCH ARTICLE

EFFICIENCY OF SYSTEMIC VERSUS INTRALESIONAL INJECTION OF BONE MARROW CELLS ON BUCCAL WOUND HEALING IN DIABETIC RATS.

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Diabetic wound, PCNA, Bone marrow cells.

Abstract

The multipotent capability of bone marrow cells gave an impelling reason to study the effect of bone marrow cells in diabetic wound healing. Diabetes is characterized by delayed and poor wound healing. The goal of this study was to evaluate the efficiency of systemic versus intralesional injection of bone marrow cells on buccal wound healing in diabetic rats. Sixty four adult male albino rats were used in the study and divided randomly into four groups. All groups were subjected to buccal wound induction. Group I; non-diabetic group (negative control) and group II; diabetic group (positive control) and did not receive any treatment. The groups III and IV diabetic groups received intralesional and intravenous injection of whole bone marrow cells respectively two days following wound induction. Specimens were obtained after 3, 7 and 12 days following wound induction and were processed to be immunostained with mouse monoclonal antibody PCNA. Immunohistochemical results revealed marked expression of PCNA in treated groups III and IV at day 3 compared to non-treated groups I and II, although there was no significant difference between the treated groups. Our results reported that intralesional injection of whole bone marrow cells showed faster healing than intravenous injection and display a more mature histology. In conclusion, Injection of the whole BMCs intralesional is more efficient compared to intravenous route in accelerating the diabetic oral wound closure and enhancing the wound healing quality. The use of whole bone marrow cells appears to be beneficial in the treatment of diabetic oral wounds.

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Introduction:-

Diabetes is the disease that compromises the body's ability to heal wounds and injuries. Wound healing in diabetic patients is a complex and multifactorial process due to dysregulated molecular interactions and signal transductions that delay wound healing (Fekrazadet al., 2015). Diabetic patients may have problem in the process of wound healing because they suffer from impaired vascularization, anti-inflammatory and antioxidant response, damage from reactive oxygen species and advanced glycation end products. Moreover, growth factor production and

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collagen synthesis are impaired in diabetes (**Brem and Tomic-Canic, 2007; Enoch et al., 2008; Yadollah-Damavandi et al., 2015**). In dental sciences, the issue of delayed healing of oral wounds and periodontal lesions in diabetic patients has raised great controversy. The delayed healing may cause pain, infection, discomfort and even affect the prognosis of the patient, leading to the failure of the treatment and the need for repeated therapies (**Fekrazadet et al., 2015**). Optimum healing of oral wound requires a well-orchestrated integration of the complex biological and molecular events of cell migration and proliferation.

Extending the hypothesis that cell therapy may be required to recondition chronic wounds and accelerate their healing leads to the conclusion that bone marrow cells may offer even greater advantages. Previous studies demonstrated that re-epithelialized mucosa of grossly “regeneration” experimental ulcers has prominent histological and ultrastructural abnormalities: Reduced height, increased connective tissue and a disorganized microvascular network. These prominent abnormalities may interfere with the mucosal defense and cause ulcer recurrence when ulcerogenic factors are present. Therefore, the quality of mucosal structural restoration may be the most important factor in determining the future ulcer recurrence (**Tarnawski et al., 1990; Tarnawski et al., 1991**). In some situations, such as for chronic non-healing wounds, the objective of cellular therapy is to reverse those cellular and vascular events that compromise repair (**McFarlin et al., 2006; Dash et al., 2009**).

Whole bone marrow cells (BMCs) participate in tissue repair processes and may have a role in wound healing. One role based on the capacity of bone marrow cells to secrete a variety of growth factors and cytokines that are required to orchestrate tissue repair and regeneration. This mechanism would regulate cellular processes such as chemotaxis, cell proliferation, angiogenesis, extracellular matrix production and remodeling. Another role for bone marrow derived cells is to differentiate into multiple cell types including fibroblast and myofibroblasts after migrating to the injury site (**Yamaguchi et al., 2005**). A recent study has shown that whole bone marrow is more efficient compared to bone marrow cultured cell and mesenchymal stem cell in reducing wound size in a murine model of delayed wound healing. They demonstrate that mixed bone marrow cell preparations may be superior to more purified stem cell formulation in stimulating wound healing (**Rodriguez-Menocal et al., 2015**).

Currently, there are two basic delivery methods: Systemic infusion of cells into the vascular circulation and direct application of therapeutic cells to wound sites (**Sorrell and Caplan, 2010**). Therefore the purpose of this study was directed toward the comparison of the efficacy of systemic versus intralesional injection of whole BMCs in regeneration of diabetic oral wound.

Material and methods:-

Sixty-four adult male Albino rats weighing (200-250g) were used in this study; they were housed in the laboratory of Nile Center for Experimental Researches under controlled condition, with constant temperature and humidity. Animals were divided randomly into four groups (n=16).

Group I (negative control): subjected to buccal wound followed by injection of Hank's balanced salt solution (HBBS)/bovin serum albumin after two days. Group II (positive control): induced for experimental diabetes, subjected to buccal wound after one week of diabetic induction followed by injection of (HBBS)/bovin serum albumin after two days. Group III: subjected to experimental diabetes and buccal wound followed by intralesional injection of 1×10^6 suspended in HBBS/bovine serum albumin of bone marrow cells after two days. Group IV: subjected to same procedure in group III and received Intravenous injection of 1×10^6 of bone marrow cells suspended in HBBS/bovine serum albumin of bone marrow cells after two days.

Streptozocin- induced diabetes:-

Diabetes was successfully induced in rats of groups II, III, IV, via single intraperitoneal injection of freshly prepared streptozotocin (STZ) (Sigma, St. Louis, MO, USA) dissolved in 0.1ml citrate buffer (pH 4.5) at a dose of 45 mg/kg as described previously (**Rajasekar et al., 2014**). Diabetes was assessed by measuring the blood glucose level in rat's tail blood using a glucometer (B. Braun, Seoul, Korea) 7 days after injection of STZ. Rats whose blood glucose level exceeded 300mg/dl were considered diabetic and were included in this study (**Hie et al., 2007**). Serum blood glucose was checked daily using ACCU-check advantage strips and 1 unit of Insulin therapy was administered if the animal had high glucose readings. High glucose reading were indicated on the glucometer as high (**O'Loughlin et al., 2013**).

Wound induction:-

All rats were anesthetized with 30 mg/kg ketamine and 10 mg/kg xylazine intraperitoneally (Fekrazad et al., 2015). Wound measuring 4 mm in diameter was made at buccal mucosa opposite to molar teeth midway between mandibular and maxillary teeth using a standard punch biopsy technique. The day of wound induction was considered as day zero.

Isolation of whole BMCs:-

Bone marrow was harvested from albino rats weighing (100-140g). In order to prevent diabetic cellular dysfunction (because of glycosylation and other mechanisms) from adversely affecting wound healing, we isolated the BMCs from non-diabetic albino rats (Lin et al., 2008). Briefly, the rat BMCs were flushed from tibias and femurs with cold HBSS and 0.5% bovine serum albumin. The suspension was then strained through a 70- μ m nylon filter and washed twice with HBSS/bovine serum albumin. The concentration of the cells was adjusted to 10^5 viable cells/ μ l. The viability of BMCs was checked using Trypan Blue with an average of 95% viability before injection. Cell-free extract was prepared by subjecting the BMCs to three freeze-thaw cycles using an ethanol/dry ice bath, followed by microcentrifugation at 14,000 rpm to remove insoluble material (Yeghiazarians et al., 2009).

Bone marrow cells transplantation:-

Two days after wound creation, all rats were anesthetized and treated as follows: rats of Group III (n=16) were treated once by intralesional injection (at two points around the wound) of 1×10^6 BMCs suspended in 10 μ l HBSS/bovine serum albumin; rats of group IV (n=16) were treated once by intravenous injection via tail vein with BMCs at a dose of 1×10^6 /10 μ l HBSS/bovine serum albumin (Yeghiazarians et al., 2009), while rats of groups I (n=16) and group II (n=16) were received 10 μ l HBSS/bovine serum albumin only.

Tissue preparation:-

Following sacrifices of rats at 3, 7 and 12 days post wounding, tissue specimen containing the wound was dissected out from each rat, fixed in a 10% buffered formalin solution and paraffin embedded, sectioned into 4 μ m-thick slice and prepared for routine hematoxylin and Eosin staining and immunohistochemical staining.

Immunohistochemical analysis:-

Immunohistochemistry was performed using mouse monoclonal antibody PCNA as a cell proliferation marker (Thermo Scientific, Fremont, USA). Each slide was screened microscopically and the area with the most intense PCNA staining were selected for quantification. Slides were photographed using Olympus[®] digital camera installed on Olympus[®] microscope with 1/2 X photo adaptor, using 40 X objective. The resulted images were analyzed on Intel[®] Core I3[®] based computer using Video Test Morphology[®] software (Russia) with a specific built-in routine for immunohistostain analysis and stain quantification. Five images for each slide were taken for analysis.

Statistical analysis:-

Data analyses were performed using IBM Statistical Package for Social Sciences (SPSS) software package version 20.0. The description of data presented in the form means (+/-) standard deviation (SD) for quantitative data. The statistical comparison between the different groups and the significance of difference was tested using one way ANOVA and repeated measure ANOVA tests followed by Post Hoc test for pairwise comparisons.

Results:-**Histological analysis:-**

Examination of H&E stained sections of Group I showed wound area filled with granulation tissue, intense inflammatory cells infiltration and disorganization of the epithelium with mitotic activity at the wound edges at day 3 (Fig.1A) while in group II showed less mitotic figures than group I (Fig.1B). In comparison to group III and group IV, the epithelia showed increase in mitotic activity (Fig.1C, D). At day 7; group I revealed moderate inflammatory cells infiltration in the granulation tissue and more mitotic activity in the epithelial edges compared to day 3 (Fig.1E) while Group II showed intense inflammatory cells infiltration in the granulation tissue with less mitotic activity within the epithelium (Fig.1F). Group III showed advanced healing process. The wound area was covered with epithelia with little irregularity in the basement membrane and the lamina propria revealed well-formed new blood vessels (Fig.1G). Group IV revealed epithelial migration into the wound bed and the epithelial cells showed mitotic activity (Fig.1H). At day 12; the wound area in group I was covered by thin epithelium with irregular basement membrane and shallow rete ridges. Lamina propria showed newly formed blood vessels with few inflammatory cells infiltration (Fig.1I) while in Group II, the granulation tissues showed intense inflammatory cells infiltration and the

Epithelia at the wound edges showed increase in the mitotic activity (Fig.1J). The epithelium showed marked re-epithelization and complete healing with obvious mitotic figure in group III (Fig.1K). However, the wound area covered with thinner epithelium in group IV (Fig.1L).

Immunohistochemical observations:-

The expression of PCNA was detected within the epithelial cells;

Group I revealed moderate expression (3.901 ± 0.001) at day 3, intense expression (6.5 ± 0.45) at day 7 and mild expression (2.4 ± 0.84) at day 12. Group II revealed moderate expression (3.8 ± 0.82) at day 3 which increased slightly at day 7 (4.2 ± 1.03) while showed mild expression at day 12 (2.8 ± 0.68). Group III showed intense expression (7.2 ± 1.61) at day 3, moderate expression (3.4 ± 0.47) at day 7 and less expression of PCNA at day 12 (2.2 ± 0.59). Group IV showed intense expression (6.068 ± 0.824) at day 3 which began to decrease at day 7 (4.2 ± 1.06) and became less at day 12 (3.4 ± 0.51). (Fig. 2)

Statistical analysis results:-

Descriptive statistics of the PCNA expression were represented by the mean \pm SD for all groups. The highest mean value was in group III at day 3. One way ANOVA followed by Post Hoc test revealed statistical significant difference between the different groups represented in table 1 and Figure 3.

Repeated measures ANOVA revealed significant difference among different days in group II, III and IV ($p < 0.05$) while a highly significant difference in group I ($p = 0.005$). (Fig.4)

Discussion:-

Cell therapy has come into attention in all medical fields as a new paradigm for future medicine. Whole bone marrow cells participate in tissue repair processes and may have a role in wound healing. Wound healing is a complex process that is regulated by cytokines and growth factors and modulated by systemic conditions such as diabetes (Rai et al., 2005). A serious complication of diabetes is impaired healing. Optimum healing of oral wound requires a well-orchestrated integration of the complex biological and molecular events of cell migration and proliferation. When the normal biological process fails for any reason, this healing process can stall resulting in chronic wounds (Giles et al., 2015).

Siqueira et al., 2010 stated that a critical component of a vigorous healing response is a generation of a sufficient number of cells to participate in repair. Recently, Garget et al., 2014 has been reported that BMCs could home to site of injury or inflammation, play a direct role in vasculogenesis and modulate epithelial regeneration. A current problem is the development of strategies that ensure that these cells reach wound beds in a timely fashion and in sufficient numbers to maximize their therapeutic benefits. The aim of our study was directed toward the comparison of the efficacy of systemic versus intralesional BMCs in regeneration of diabetic oral wound.

In the present study, STZ induced diabetic rats were used as a model to study the healing efficiency of bone marrow cells. The significance of diabetes is that delayed wound healing and thus, acceleration of healing is crucial in oral and maxillofacial surgery. Rees & Alcolado, 2005 stated that STZ induced diabetes in rodents is considered to be a model of insulin-dependent diabetes mellitus and is widely used in the study of insulinopenia and hyperglycemia. Also, Papaccio et al., 2000 reported that STZ selectively destroys beta cells, inhibits the synthesis and release of insulin and causes the onset of diabetes mellitus.

A study done by Roriguez-Menocal et al., 2015 demonstrated that the whole BMCs appeared to stimulate a greater healing response than bone marrow mesenchymal stem cells in a radiation induced delayed wound healing animal model. The whole BMCs have a greater mixed population of cell types thus they were capable of producing a more wide-ranging cytokine profile that more closely resembled that seen in healing wounds. The results of the previous study support the use of a mixture of cell types in designing clinical approaches to wound healing. Based on the previous statement, the whole BMCs were used in the present study.

In hematoxylin and eosin results the wound area in group I (negative group), at day 7 showed slight proliferation of covering epithelia with moderate inflammatory cells infiltration within granulation tissue while after 12 days the wound area was covered by thin epithelium which showed irregular basement membrane and shallow rete ridges. These results are consistent with Karavana et al., 2011 who reported that oral mucosal ulceration in animal model showed complete healing on the 12th day post ulceration.

In the present study, the results in group II (positive group) at day 3 showed prolonged excessive inflammatory cells infiltration, loss of attachment and decreased mitotic activity within epithelia and connective tissue. This is in accordance with **You & Han, 2014** who reported that one of main contributing factors for the non- or delayed healing wound is decreased function (mitotic activity, extracellular matrix and growth factors synthesis) of the key cells for wound healing including fibroblast and keratinocytes. Therefore, the wound remains open for long period of time which leads to infection.

The wound area in Group III (intralesional group) at day 3, showed granulation tissue formation with dense inflammatory cells infiltration, increased dilated blood vessel and mitotic activity increased at the epithelial edges of the wound, while at day 7, the wound area was covered with epithelia. These results are in many respects to those of **Lin et al., 2008** who demonstrated that topical subcutaneous transferring bone marrow progenitor cells into full thickness excisional dorsal cutaneous wound in diabetic mice significantly improves diabetic wound healing. The authors hypothesized that these progenitor cells will differentiate into endothelial cells, increase wound vascularity and improve wound healing.

The wound area in group IV (intravenous group), at day 7 was partially covered by epithelium, while at day 12 healing process was evident and the wound area covered with thin epithelia. This is in agreement with **Roríguez-Menocal et al., 2015** who explained that whole BMCs were able to achieve higher new blood vessel formation because it stimulates endothelial cells and angiogenesis in tissue repair.

Detection of proliferating cells in tissue sections can be achieved by a number of methods, **Hall & Woods, 1990** and **Muskhelishvili et al., 2003** reported that immunohistochemical methods of assessing cell proliferation have particular advantages over the other techniques because of maintenance of cellular and tissue architecture, the relative simplicity of methodology and rapidity of results. In order to identify the efficiency of intralesional and intravenous injection of BMCs on cellular proliferation, PCNA was used as a cell proliferation marker.

In the current study, PCNA expression appeared obvious mainly in basal cell layer of all groups. This result could be explained by **Ishii et al., 2014** who mentioned that the cells proliferation occurs principally in the basal layer of the epithelium in which keratinocytes undergo terminal differentiation as they migrate to the surface. Also, **Yue et al., 2005** stated that the basal layer of the stratified squamous epithelium consists of a heterogenous population of proliferative and differentiating cells.

In addition, PCNA expression was obvious in the marginal epithelia in group III and group IV, at day 3 specifically, (7.2 ± 1.61 and 6.068 ± 0.824) respectively. These findings imply that the wound healing is provoked not only by the proliferation and differentiation of basal cells but also by supplying cells from the wound margin to the epithelial outgrowth as mentioned by **Takaichi et al., 2014**.

In group II, there was moderate expression of PCNA at day 3 and 7 days (3.8 ± 0.82 and 4.2 ± 1.03) respectively while mild expression at day 12 (2.8 ± 0.68). This is consistent with **McLaughlin et al., 2013** who reported that initial delays in cell proliferation following wounding in diabetic rats were related to changes in DNA synthesis rates in basal epithelial cell layers.

In the present study, immunohistochemical results showed marked expression of PCNA in groups III and IV (treated groups) after 3 days (7.2 ± 1.61 and 6.068 ± 0.824) respectively compared to groups I and II (non-treated groups) (3.901 ± 0.001 and 3.8 ± 0.82), although there was no significant difference between treated groups. These findings is in agreement with **Isakson et al., 2015** who mentioned that both topical and systemic delivery methods have been shown to be effective in chronic wound healing. Moreover, these results are in analogues in many respects to those of **Borue et al., 2004** who found that transplantation of whole bone marrow cells in wound environment, allow the progenitor cells enter the wound area and fuse with the damaged epithelial cells, thereby taking on keratinocytes properties.

Also, these results are supported by the findings of **Paradells et al., 2015** who mentioned that BMCs could help to enhance local endogenous proliferation and probably stimulate proliferation and subsequent migration from surrounding zones. Moreover, **Rodríguez-Menocal et al., 2015** reported that the whole bone marrow cells were able to achieve higher new blood vessel formation compared with mesenchymal stem cells. This implies a potential significant advantage for whole bone marrow to stimulate endothelial cells and angiogenesis in tissue repair.

To date, experimental studies have not presented definite answers regarding the role of BMCs and related mechanisms. However, several hypotheses have been proposed. First, **Liu et al., 2006** and **M. Isakson et al., 2015** stated that BMCs have the capacity to secrete a variety of growth factors and cytokines. The production of growth factors by BMCs potentially plays a role in coordinating and regulating the cellular processes, such as chemotaxis, inflammation, cell proliferation, tissue granulation, and wound remodeling which are critical for the efficient wound healing. In addition to the cells themselves participating in the process of wound healing, ultimately differentiating into the cell types required for closure of the wound. Second possibility relates to the tremendous plasticity and the ability of BMCs to differentiate into a variety of cell types and integrate into tissues. Therefore, BMCs may facilitate wound healing as mentioned by **Liu et al., 2006**. Third, **Vaananen, 2005; Aly et al., 2014** mentioned that BMCs release paracrine factors and thereby induce the proliferation of specific cells in the damaged tissue or contribute to making the microenvironment more appropriate for the repair of injury by maximally inhibiting the tissue injury. In the present study, group III (Intralesional injection) showed faster healing and more organized re-epithelization than group IV (intravenous injection) although there was no significant difference between the two groups in PCNA expression. These results are consistent with **Kwon et al., 2008** who attributed that the differences in efficacy of systemic versus local BMCs therapy in wound healing, to the decrease in number of BMCs migrating to the wound after systemic administration. Also, he thought that the decrease in migration of BMCs to the wound treated systematically may be due to arterial-venous shunts in diabetic skin. In addition, **Karp & Leng, 2009** stated that during vascular transit, BMCs risk being taken out of circulation, and going to other organs such as the lungs, spleen and liver. This may reduce the numbers of cells that finally appear at a target site.

Another study done by **Sorrell & Caplan, 2010** revealed that the topical delivery of cells allows for concentrated doses of cells to be delivered to a wound and not become trapped in other sites in the body. Also, it was hypothesized by **Chung et al., 2011** that applying cells topically may promote neovascularization from inside the wound not only from the wound periphery.

All of these findings demonstrated that BMCs can contribute to wound repair in diabetic rats and may provide the cell source for regenerative therapy. Despite the current limitations to widespread clinical use, BMCs are highly promising cell sources for the treatment of diabetic wounds either via intravenous or intralesional route. However, further studies are necessary to study the critical factors that make up the microenvironment that supports the survival and differentiation of these cells.

Conclusion:-

Our findings proposed the intralesional injection of BMCs directly to improve the healing of oral diabetic wound, in an attempt to overcome both decrease in number of BMCs and impaired homing to the site of wound via intravenous route.

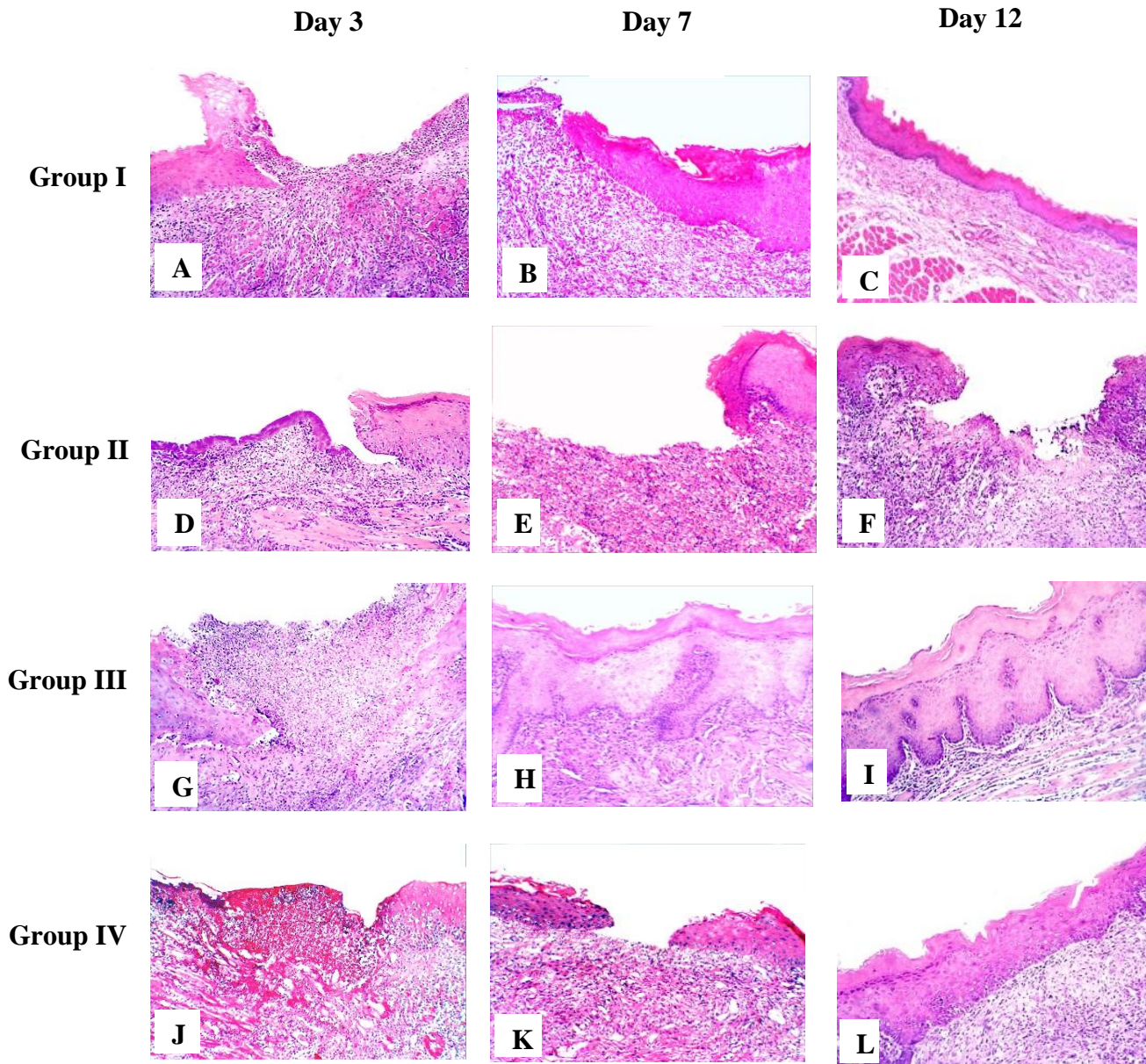


Figure 1. Histological observations of oral mucosa sections in different studied groups (I, II, III and IV) at day3, 7 and 12 (H&EX100). **At day 3** shows the wound area filled with granulation tissue with intense inflammatory cells infiltration (A, G and J), moderate inflammatory cells in (D). **At day 7** shows slight proliferation of covering epithelium (B), intense inflammatory cells infiltration within the granulation tissue (E), epithelium covering wound area with irregular basement membrane and fibrous lamina propria (H), epithelial migration of wound edges and granulation tissue with moderate inflammatory cells infiltration (K). **At day 12** shows thin epithelium with irregular basement membrane covering the wound area over fibrous lamina propria (C), discontinuity of the epithelium with intense infiltration of inflammatory cells (F), marked epithelization with deep rete ridges and initial organization of collagen fibers in the underlying connective tissue (I), thin epithelium covering wound area with short rete ridges (L).

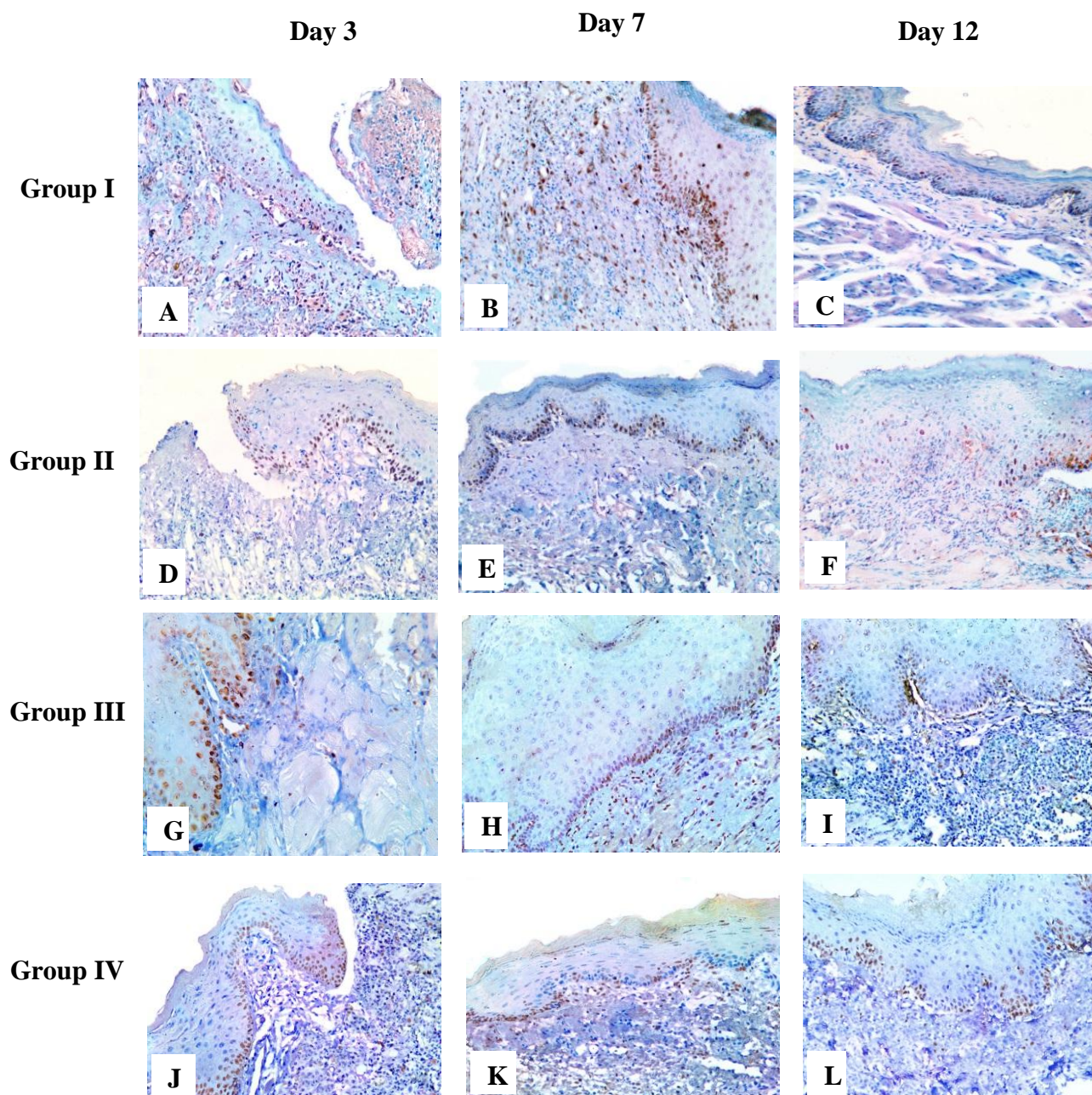


Figure 2.Immunohistochemical expression of PCNA during wound healing in different studied groups (I,II,III and IV) at day 3, 7 and 12 (IHC staining, PCNA X200).At day 3 shows moderate expression in group I and II (A, D) and intense expression in group III and IV (G, J).At day 7shows moderate expression in group II, III and IV (E, H, K) and intense expression in group I (B).At day 12shows mild expression in all studies groups (C, F, I, L)

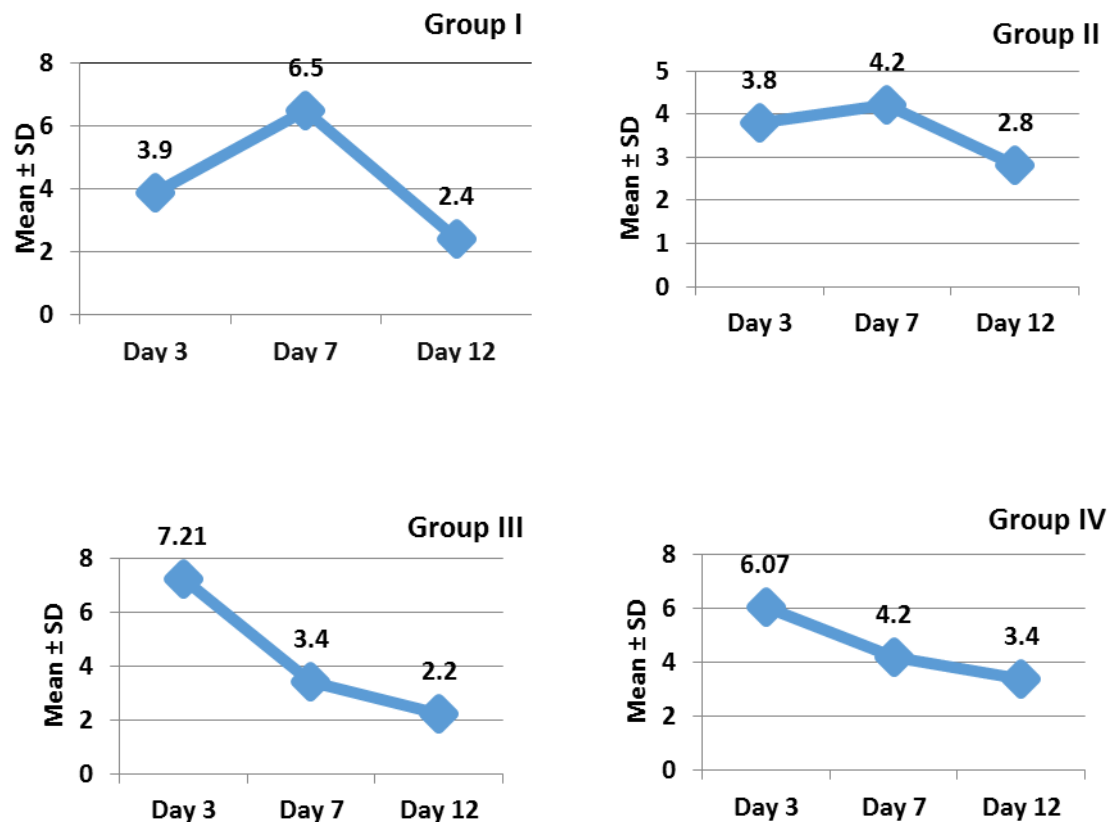


Figure 4:- Line charts shows mean \pm SD of PCNA expression in different studied groups at different sacrifice dates.

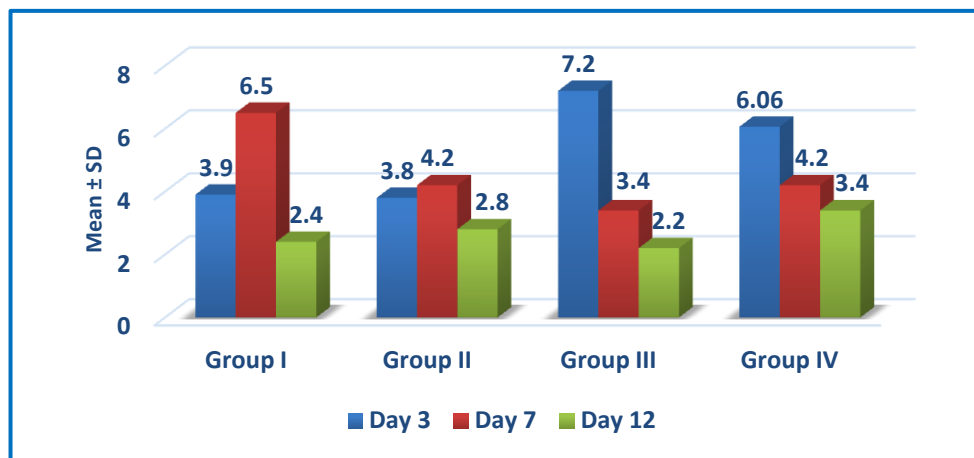


Figure 3:- Bar chart shows mean \pm SD of PCNA expression in the studied groups.

Table 1: Shows mean \pm SD for proliferating cell nuclear antigen (PCNA) at days 3, 7 and 12 among the studied groups

	Group I	Group II	Group III	Group IV	Significance
Day 3	3.901 \pm 0.001 ^{ab}	3.8 \pm 0.82 ^{cd}	7.2 \pm 1.61 ^{ac}	6.068 \pm 0.824 ^{bd}	F=12.098 P=0.001**
Day 7	6.5 \pm 0.45 ^{abc}	4.2 \pm 1.03 ^a	3.4 \pm 0.47 ^b	4.2 \pm 1.06 ^c	F=11.023 P=0.001**
Day 12	2.4 \pm 0.84	2.8 \pm 0.68	2.2 \pm 0.59 ^a	3.4 \pm 0.51 ^a	F=2.418 P=0.117

F=One Way ANOVA

** High statistically significant

abcde Similar letters denote significant difference between groups using Post Hoc LSD test

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RESEARCH ARTICLE

TRIPLE TEST SCORE AS AN EVALUATING FACTOR FOR PALPABLE BREAST LUMP.

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Abstract

Background: Breast lump is a very sensitive issue and cause of great worry and anxiety to the patient, so a reliable, preferably non-invasive and prompt diagnosis is required. Breast lump should be managed effectively and confidently with a proper protocol plan, ensuring early and best possible treatment for every patient. Triple test assessment was a major breakthrough in this direction, which streamlined the management of palpable breast lump. When all the components of triple test assessment which are Clinical Examination, FNAC, Mammography point to one possibility (are concordant) then the diagnosis is almost certain and management can be confidently planned in such a situation. But if there is discordance among the components of triple test, then what should be the next step in the management plan is the question to be answered. This is where triple test score shows us the path.

Objective of the Study:

- To perform Triple Test Score in patients with breast lump.
- To perform Histopathological Examination (HPE) of the breast lump resected.
- To evaluate the efficacy of TTS in comparison with HPE.
- To develop a standard protocol for management of breast lump especially when discordant results are obtained from triple assessment.

Materials and Methods: Study was conducted on 200 patients presenting with breast lump to the department of General Surgery at Tertiary Care Teaching Hospital, during the period from January 2012 to Dec 2016.

It was a prospective study. Women presenting for evaluation of palpable breast lump underwent assessment by clinical examination, mammography and FNAC and got the Triple Test Scoring done.

All patients who underwent a complete TTS at our institution were entered into the study.

All patients were subjected to necessary surgery, post TTS and followed up with Histopathology of the specimen.

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A structured proforma was used to collect relevant information from each patient selected.

Results: In our study the mean age of the patients was found to be 46.12 ± 1.48 years, most of the patients were in the age group of 35-45 years (60%). Positive family history was found in 17%. Patients on an average took 6 months to seek medical help after recognition of the breast lump. Most common location of breast lump was upper outer quadrant. 38 cases were malignant and 148 were benign 14 were intermediate group commonest lump was fibroadenoma, Histopathology was 100% specific in both the groups of study conclusions were made based on clinical radiological and pathological assessment.

Conclusion: The study clearly demonstrates the superiority of TTS over other components of triple assessment or all of them put together. A ITS of ≤ 4 is consistent with a benign lesion; a TTS of ≥ 6 indicates malignancy. Only in patients in whom TTS score is 5, biopsy is recommended to obtain a definitive diagnosis. Thus a standard protocol can be developed, for the management of breast lump even with discordant results obtained via triple test assessment, which can be followed universally, thus empowering surgeon to go ahead in managing breast lump effectively and confidently.

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..... Introduction:-

"He will manage to cure best who has foreseen what is to happen from the present state & matters"

Hippocrates:-

The frequency of breast diseases, their recognition and the attempts at primeval cures by various cultures and societies historically antedate the therapy of diseases of other solid organs. Diseases of breast, with their uncertain causes and confusion of treatments, have intrigued physicians and medical historians throughout the ages. Despite centuries of theoretical meanderings and scientific enquiry, cancer of the breast remains one of the most dreaded of human ills. Although primarily thought of as a disease of women, it may occasionally afflict men with results just as lethal. The breast as a paired organ further increases its exposure to disease. As appendage of the skin it usually reveals its disorders to touch or sight.

A breast mass can be a difficulty at times. It may be difficult for the patient because of the anxiety associated with her underlying fear of a breast malignancy and it may be difficult for the physician to feel confident that what he or she is palpating is truly a mass rather than a variation of normal breast parenchyma. Breast symptoms and signs are common problems in clinical practice. Majority of breast symptoms or lesions will prove to be of a benign etiology. Physical, psychological and financial costs of investigating benign breast disease, primarily to exclude malignancy are substantial. Much concern is given to malignant lesions of the breast because breast cancer is the most common malignancy in women; however, benign lesions of the breast are far more frequent than malignant.

Because the majority of benign lesions are not associated with an increased risk for subsequent breast cancer, unnecessary surgical procedures should be avoided. It is important for pathologists, radiologists, and oncologists to recognize benign lesions, both to distinguish them from in-situ and invasive breast cancer and to assess patient's risk of developing breast cancer, so that the most appropriate treatment modality for each case can be established.

The first step in evaluation of breast lump is the clinical assessment. Although many a times clinician can confidently make the diagnosis of benign or malignant lesion, the possibility of mistake is always there even in experienced hand. This is where triple assessment has played a significant role in breast lump management.

The triple assessment for breast diseases involves,

1. Clinical assessment
2. Imaging modality — Mammography
3. Fine needle aspiration cytology

Clinical diagnosis of breast cancer is of higher sensitivity than specificity and has high diagnostic error. Mammography and FNAC respectively have lower sensitivity than specificity but have high positive predictive values.

When combined in the triple assessment, a definitive diagnosis can be made when the diagnoses concur, suggesting that the triple assessment has a high sensitivity, specificity, positive predictive value and negative predictive value with minimal error and excellent Kappa statistic. The output of the triple assessment is reproducible, making it a valid and reliable diagnostic approach to diagnosis of breast cancer.

With increasing prominence and greater visibility in country specific health profiles around the world, breast cancer and its prevention, detection and treatment will continue to emerge as a major priority and challenge, for the health system in the near future.

Need For The Study:-

Breast lump is the clinical presentation of numerous breast diseases ranging from innocent benign cysts to malignant lesions. Distinction of benign from malignant is of paramount importance for patient care and proper management. Breast cancer is the most common site specific cancer in women and is the leading cause of death from cancer for women of age 40 to 44 year¹. It accounts for 33% of all female cancers and is responsible for 20% of the cancer related deaths in women².

Presently a wide range of diagnostic modalities are available for the evaluation of breast lump. Conventional open biopsy, considered to be the gold standard for confirming diagnosis, has significant morbidity, is costly and time consuming. All of these cause significant trauma to the patient and are not patient friendly.

Mis-diagnosed breast cancer accounts for the greatest number of malpractice claims for errors in diagnosis. Litigation often involves younger women whose physical examination and mammography may be misleading². Two techniques that are currently available with excellent patient tolerability are mammography and fine needle aspiration cytology. However if employed alone the reliability of mammography and FNAC is only around 82% and 78% respectively³.

There are numerous reports that if the results of clinical assessment, mammography and FNAC are all combined, the accuracy of diagnosis reaches 100%⁴. Furthermore these techniques provide information on tumor size, number, extent and grade pre-operatively⁵.

There is a direct need for evolving a method for establishing the diagnosis pre-operatively, which is cost effective, least invasive and least disturbing to the patient, with accuracy comparable to open biopsy. An efficient evaluation and prompt diagnosis is necessary to maximize cancer detection and minimize unnecessary testing and procedures. A thorough Clinical Breast Examination (CBE), Imaging (Mammography), and Tissue sampling (FNAC) are needed for definitive diagnosis².

The triple test assessment is the combination of results from CBE, imaging, and tissue sampling. When the three assessments produce concordant results (point to the same possibility), the triple test diagnostic accuracy approaches 100 percent⁶⁻⁸. But, in discordant results, there is no clear cut protocol to follow and it is in such situations that triple test scoring will be the next step forward. Discordant results are seen in 40% patients, who are subjected for open biopsy for the confirmation of diagnosis⁹.

The Triple Test Score (TTS) was developed to help clinicians interpret discordant triple test results.⁶⁻¹⁰ A three-point scale is used to score each component of the triple test (1 = benign, 2 = suspicious, 3 = malignant). A TTS of ≤ 4 is consistent with a lesion; a TTS of 5 indicates malignancy. Only in patients in whom TTS score is 5, biopsy is recommended to obtain a definitive diagnosis. Thus a standard protocol can be developed, for the management of discordant results in triple test assessment, which can be followed universally.

The scope of improvement in arriving at correct and confident diagnosis of the breast lump is still enormous when we keep cent percent perfect diagnosis as our goal. This study is an attempt to travel part of that journey towards the goal.

Materials and Methods:-**Source Of Data:-**

Study conducted on 200 patients presenting with breast lump to the department of surgery in Tertiary Craae Teaching Hospital during the period from January 2012 to December 2016.

Inclusion Criteria: Patient aged ≥ 35 years, presenting with palpable breast lump.

Exclusion Criteria: Obvious malignant lesions (fungation, ulceration).

Method of collection of data:-

It being prospective study, women presenting for evaluation of palpable breast lump to the department of surgery at Tertiary Care Teaching Hospital underwent assessment by clinical examination, mammography and FNAC and Triple Test Scoring was done.

All patients who underwent a complete TTS(Triple Test Scoring) at our institution were entered into the study.

All patients were subjected to necessary surgery, post TTS and followed up with histopathology of the specimen.

A structured proforma was used to collect relevant information from each patient selected. Each component of the triple assessment was compared with the gold standard histopathology, so also TTS was compared with histopathology and findings were analyzed.

All of patient details and relevant information was entered into the proforma.

Analysis:-

All the three components of triple test i.e., physical examination, mammography and FNAC findings were categorized as benign, suspicious and malignant. The Triple test (TT) was considered concordant if all the elements indicated a malignant condition or all indicated a benign condition, otherwise TT was considered non-concordant.

Sensitivity is defined as percentage of cases in which biopsy proven cancer was correctly diagnosed by the test.

Specificity is defined as percentage of cases in which biopsy proven benign lesion was correctly diagnosed by the test.

The values were determined by the following formula:

Sensitivity = $TP / TP + FN$; Specificity = $TN / TN + FP$

TP - true positive,

TN - true negative,

FP - false positive

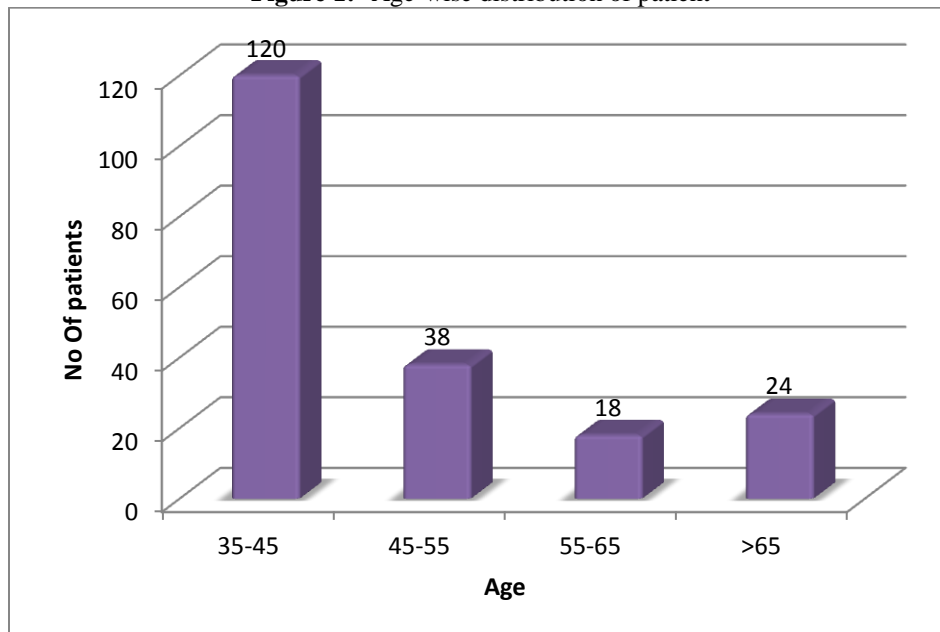
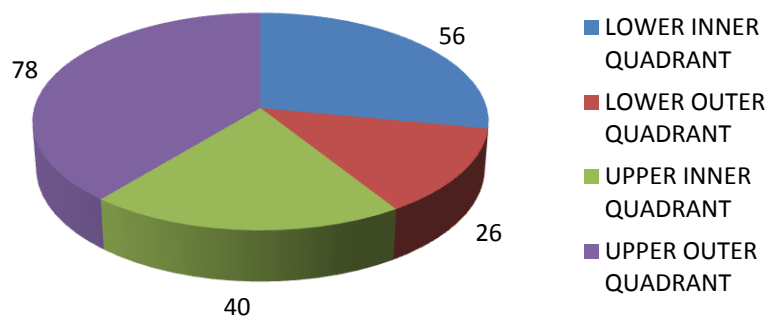
FN - false negative

Further, triple test score (TTS) was given; physical examination, mammogram and FNA were each assigned a score of 1, 2 or 3 for benign, suspicious or malignant results; TTS is the sum of these scores. TTS has a minimum score of 3 (concordant benign) and a maximum score of 9 (concordant malignant).

Results:-

A total of 200 patients who satisfied the inclusion criteria were enrolled into the study, all the patients were subjected to clinical examination followed by mammography and FNAC, individual scores were given and the triple test score was calculated. All the patients were subjected to the appropriate surgery and the specimen sent for histopathology.

In our study the mean age of the patients was found to be 46.12 ± 1.48 years, most of the patients were in the age group of 35-45 years (60%). Positive family history was found in 17%. Patients on an average took 6 months to seek medical help after recognition of the breast lump.

Figure 1:- Age-wise distribution of patient**Figure 2:-** Location of breast lump

Most common location of breast lump was upper outer quadrant (Figure).

Among the 200 cases which had histopathologic correlation 156 were benign disease and 44 malignant.

Table 1:- Comparison Of Clinical Assessment With Histopathology

CLINICAL ASSESSMENT	HISTOPATHOLOGY		Total
	Benign	Malignant	
Benign	146	2	148
(%)	91.1%	4.5%	53.0%
Suspicious	10	4	14
(%)	8.9%	9.1%	9.0%
Malignant	0	38	38
(%)	0.0%	86.4%	38.0%
Total	156	44	200
(%)	100%	100%	100%

Specificity: 100 %

Positive Predictive value: 100 %

Negative Predictive Value: 96.23 %

Accuracy: 97.80%

The scoring for clinical examination revealed a score of 1, 2 and 3 in 53%, 9% and 38% respectively. The clinical diagnosis of benign and malignant was comparable with HPE.

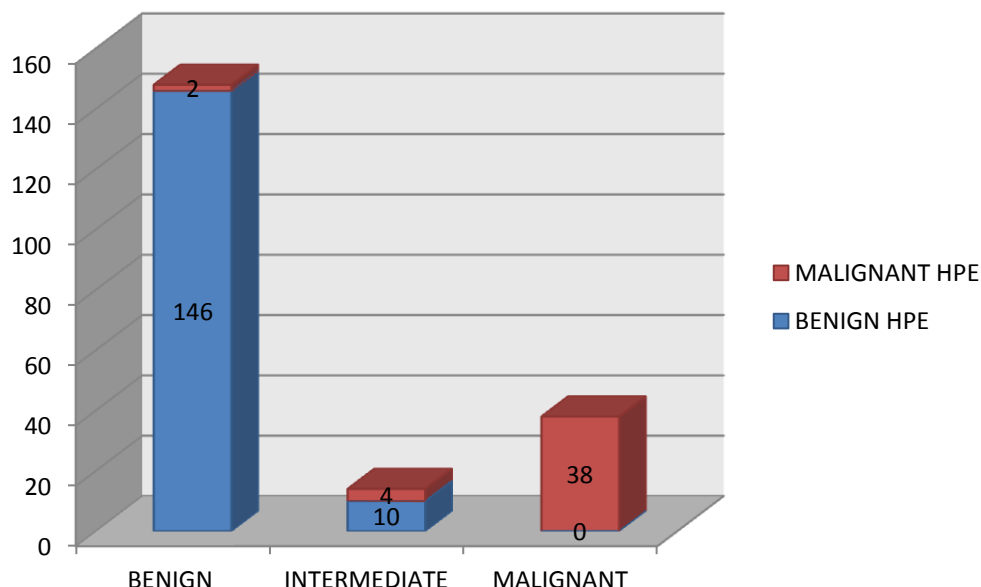


Figure 3:- Comparison of clinical assessment with histopathology

Out of 14 cases with suspicious interpretation in clinical diagnosis: 10 were diagnosed to be benign and 4 were diagnosed as malignant. 4 cases which were diagnosed clinically as benign turned out to be malignant on HPE.

Table 2:- Comparison Of Fnac With Histopathology

FNAC SCORE	HISTOPATHOLOGY		Total
	Benign	Malignant	
Benign	152	1	153
(%)	96.4%	2.3%	55.0%
Suspicious	4	3	7
(%)	3.6%	6.8%	5.0%
Malignant	0	40	40
(%)	0.0%	90.9%	40.0%
Total	156	44	200
(%)	100%	100%	100%

The scoring for FNAC revealed a score of 1, 2 and 3 in 55%, 5% and 40% respectively. The clinical diagnosis of benign and malignant was comparable with HPE.

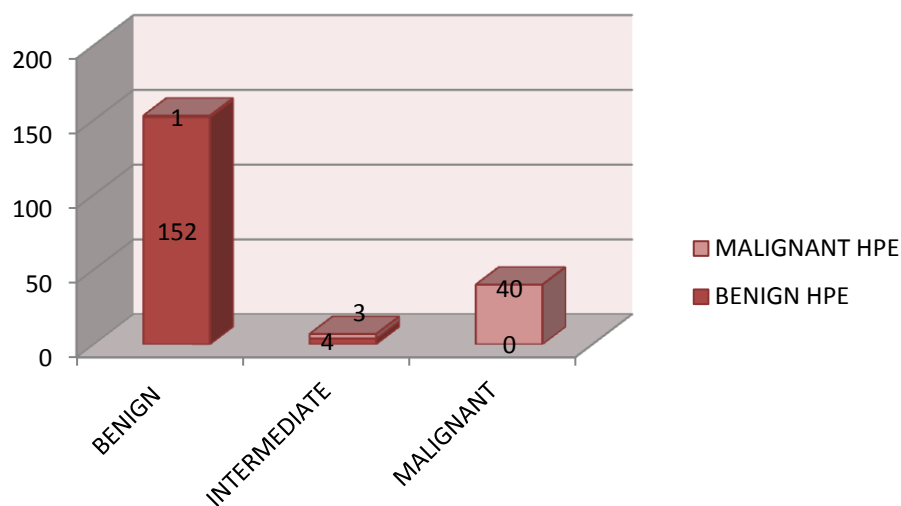
Sensitivity: 97.56 %

Specificity: 100 %

Positive Predictive value: 100 %

Negative Predictive Value: 98.18 %

Accuracy: 98.94%

Figure 4:- Comparison of FNAC with histopathology.

Out of 7 cases with suspicious interpretation in FNAC: 4 were diagnosed to be benign and 3 were diagnosed as malignant. One case which was diagnosed as benign turned out to be malignant on HPE.

Table 3:- Comparison Of Mammography With Histopathology

MAMMOGRAPHY	HISTOPATHOLOGY		Total
	Benign	Malignant	
Benign	156	3	159
(%)	100%	6.8%	59.0%
Suspicious	0	6	6
(%)	0.0%	13.6%	6.0%
Malignant	0	35	35
(%)	0.0%	79.5%	35.0%
Total	156	44	200
(%)	100%	100%	100%

The scoring for mammography revealed a score of 1, 2 and 3 in 59%, 6% and respectively. The mammography diagnosis of benign and malignant was comparable with HPE.

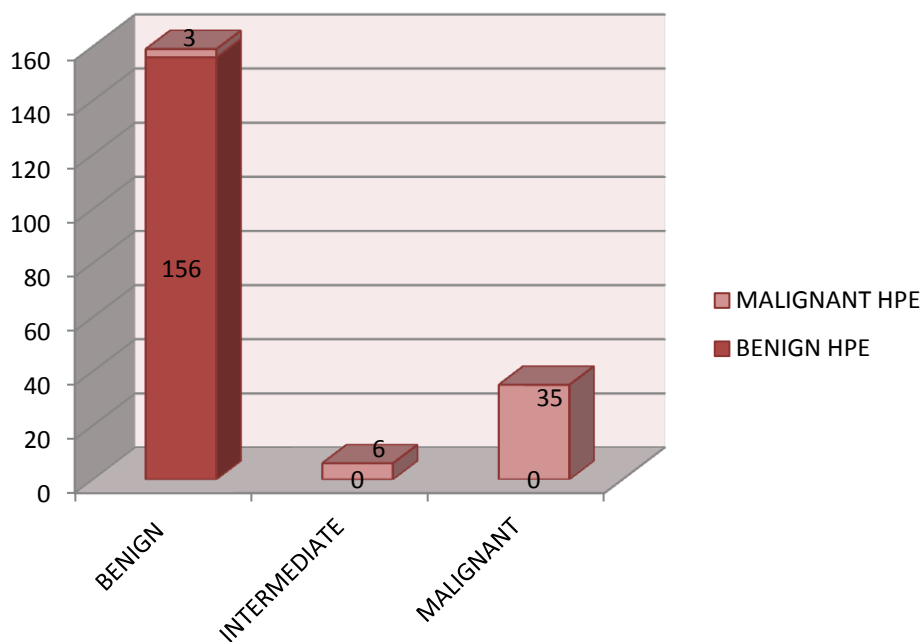
Sensitivity: 92.11 %

Specificity: 100 %

positive Predictive value: 100 %

Negative Predictive Value: 94.92 %

Accuracy: 96.80%

Figure 5:- Comparison of Mammography with histopathology

Out of 6 cases with suspicious interpretation in mammography all were diagnosed as malignant. Three cases which were diagnosed as benign turned out to be malignant on HPE.

Table 4:- Comparison Of Triple Test Score With Histopathology

TRIPLE TEST SCORE	HISTOPATHOLOGY		Total
	Benign	Malignant	
Benign	155	0	155
(%)	100%	0.0%	52.0%
Suspicious	1	0	1
(%)	1.8%	0.0%	4.0%
Malignant	0	44	44
(%)	0.0%	100.0%	44.0%
Total	156	44	200
(%)	100%	100.0%	100%

All the cases diagnosed as malignant with TTS were proved malignant by HPE, all cases diagnosed as benign were proved benign on HPE, one case with TTS of 5 required a further test in form of biopsy for confirmation, it turned out to be benign,

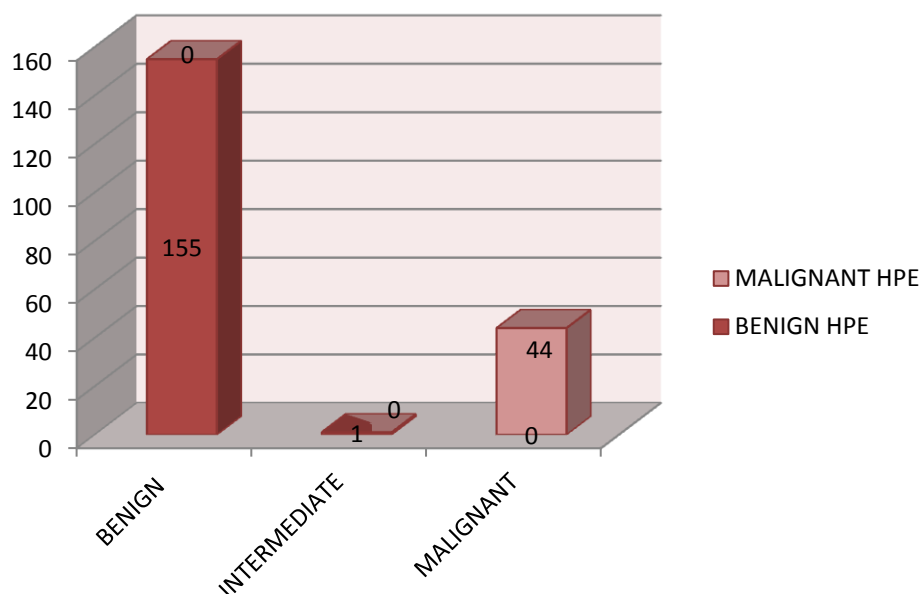
Sensitivity: 100 %

Specificity: 100 %

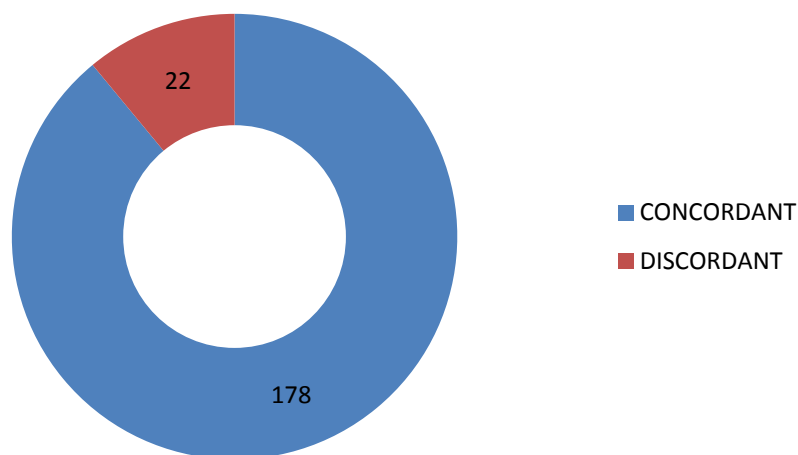
Positive Predictive value: 100%

Negative Predictive Value: 100 %

Accuracy: 100 %

Figure 6:- Comparison of TTS with histopathology**Table 5:-** concordant v/s discordant results in triple assessment.

CONCORDANT	DISCORDANT
178	22

Figure 7:- Depicting the ratio between concordant and discordant results

Among the 22 discordant results, score of 7,6 and 5 were seen in 14,6 and 2 patients respectively.

Table 6:- Results Derived From Various Modalities Used In Breast Lump Analysis

Diagnosis	Clinical	FNAC	Mammography	Triple test	HPE
Benign	148	153	159	155	156
Intermediate	14	7	6	1	0
Malignant	38	40	35	44	44
Total	200	200	200	200	200

Table 7: Comparison Of All The Components Used In Breast Lump Analysis

Investigation	Sensitivity (%)	Specificity (%)	Positive Predictive Value (%)	Negative Predictive Value (%)	Accuracy (%)
Clinical examination	95	100	100	96.23	97.80
FNAC	97.56	100	100	98.18	98.94
Mammography	92.11	100	100	94.92	96.80
TTS	100	100	100	100	100

Table 8: Kappa Agreement Between Biopsy And Other Diagnostic Modalities

Diagnostic Modality	Kappa value	P value
Clinical Examination	0.795	<0.01
FNAC	0.884	<0.01
Mammography	0.758	<0.01
Triple test	0.903	<0.01

Discussion:-

Prospective analysis of TTS on 200 patients and confirming the results with histopathological finding showed to be highly sensitive and specific. In the present study 100 patients with age ranging from 35 years to 90 years with a mean age of 46.12 \pm 11.48 years who presented with complaint of breast lump were evaluated. The mean age here was considerably less than that seen in the western population (57 years) ⁹ and comparable to study done at Nepal (48 years). 60% of the patients belonged to age group between 35-45yrs. Benign diseases were more common than malignant. Fibroadenoma being the most common benign lesion and Infiltrating ductal carcinoma being the most common malignant lesion. Most of the patients aged above 55 years with breast lump were diagnosed with a malignant lesion reinforcing the fact that age is an important risk factor in carcinoma breast. The lesion was found to be present commonly in upper outer quadrant (39%). Women on an average sought medical help with a delay of 6 months after realizing the presence of breast lump, thus delaying the treatment which in cases of malignancy carry bad prognosis, thus emphasizing the need of better education of the mass at large.

In our study, clinical diagnosis (physical examination) showed a sensitivity of 95%, a specificity of 100% and positive predictive value of 100%, negative predictive value of 96.23 with an overall accuracy of 97.80% . Other studies showed that clinical examination could diagnose accurately only 70% of cases of carcinoma. Egan recorded an accuracy of 65% detection by physical examination ¹¹. Our study showed an accuracy of 97.80% by clinical examination. This relatively high accuracy in detecting malignancy by clinical examination is due to the fact that our patients rarely present early in the course of the disease. Breast lump in our patients on an average was about 4x3 cm on presentation. Out of 9 cases with suspicious interpretation, in clinical 5 were diagnosed to be benign and 4 were diagnosed malignant. Two cases which were diagnosed clinically as benign turned out to be malignant on HPE .

In examining the triple test elements individually, we noted that FNAC is typically more accurate than physical examination or mammography . This agrees with the study of Morris et.al. and Vetto et. Al ¹²⁻¹⁸. In our study, the sensitivity of FNAC was 97.56%, the positive predictive value was 100%, specificity was 100%, and the negative predictive value was 98.18% with no false positives, but 1 false negative . These results are in accordance with those of Morris et.al. Vetto et. Al ¹²⁻¹⁸. reported a sensitivity of 96% for FNAC, with a specificity of 100%, and a positive predictive value of 100%. Rubin and Joy concluded that FNAC is the first reliable diagnostic step in detection of breast carcinoma. They reported a positive predictive value of 100%, a specificity of 100%, a sensitivity of 87%, and a negative predictive value of 89%.

The widespread use of mammography has helped in better management of breast lump. In our study, the accuracy of mammography was 96.80%, the sensitivity 92.11%, the specificity 100%. The positive predictive value was found to be 100%. There were 3 false positives and 6 cases were inconclusive . In a Dutch study of breast cancer screening, Romback found that if mammography alone has been used the sensitivity of breast cancer diagnosis would have been 95%. Rodes et.al. reported that mammography was the sole detection modality in 56% of cases. When combined with Physical examination, an additional 30% were detected, while physical examination alone detected 14% of cases.

In our study, the best results was got by TTS, it showed sensitivity of 100%, the positive predictive value was 100%, specificity was 100%, and the negative predictive value was 100% almost in perfect alignment with that of histopathology. In one case where the TTS was 5 an additional test in the form of biopsy was required.

In our study, when all three components showed malignancy, the sensitivity and specificity were 100%. Kaufman et al described a sensitivity of 100% and a specificity of 57% for the triple test and a negative predictive value of 100% in concordant cases. Clinical examination remains indispensable for detection of different breast lesions. Mammography remains the method of choice in radiology of the breast. FNAC has proved to be a very effective diagnostic aid. It is an easy technique, safe and very acceptable to patients. TTS outweighs all of these components and also helps us proceed further even in difficult scenarios of discordant results with triple assessment, thus reducing the fall back on the option of open biopsy which carries with it a number of disadvantages.

The use of the triple test score (with highest kappa agreement-0.903) has proved itself to be a reliable tool for the accurate diagnosis of palpable breast lump. Triple test score when implemented streamlines the management of breast lump, more so when triple assessment can't come to a definitive diagnosis and thus biopsy which usually is resorted to in such a scenario can be avoided, saving the patient from anxiety, repeated operative procedure, financial burden, undue delay in treatment and also providing the surgeon a platform to base his further management.

Summary:-

This prospective study was done to evaluate the efficacy of triple test score in management of palpable breast lump and to develop a standard protocol for management of breast lump especially when discordant results are obtained from triple assessment.

A total of 200 women with complain of breast lump presenting to the department of surgery at Tertiary Care Teaching Hospital, underwent assessment by physical examination, followed by mammography and FNAC. Triple Test Scoring was done for each component and the total score calculated. Patients were subjected to appropriate surgery and the specimen sent for histopathology. Each component was analyzed and compared with histopathology, the parameters sensitivity, specificity, positive predictive value, negative predictive value, accuracy were calculated and it was found that triple test score was best in predicting the nature of the breast lesion compared with any of the individual components of triple assessment or all of them put together.

The real value of triple test scoring was better appreciated when there was discordant results among the components of triple assessment. As no proper guidelines exist in managing such cases, it was here that triple test score can provide us something concrete on which we can base our definitive management effectively and confidently. Being a non-invasive, cost effective method it gives us the result without any undue delay and helps to manage patient in a better than managing such cases as per individual surgeon's choice.

Triple test score when implemented streamlines the management of breast lump, more so when triple assessment can't come to a definitive diagnosis and thus biopsy which usually is resorted to in such a scenario can be avoided, saving the patient from anxiety, double operative procedure, financial burden, undue delay in treatment and also providing the surgeon a platform to base his further management.

Conclusion:-

Triple test score can be safely used as an accurate and least invasive diagnostic test and based on its interpretation, definitive treatment can be initiated which would reduce the need for unnecessary biopsies. The strength of TTS seems to lie in its ability to reliably predict benign lumps and thus avoid major surgeries. Given the increased incidence of malignant lumps in elderly females and the tendency to hide asymptomatic lumps, we need more awareness programs targeting this age group.

When patient presents to us with breast lump, it has been the usual practice to do a thorough clinical assessment, reaching a provisional diagnosis, which is then confirmed by using FNAC. With triple assessment gaining popularity mammography too was included into the scheme of breast lump evaluation for more apt diagnosis thus leading to better management of the patient.

When all the components of triple assessment are concordant, that is agree on common grounds the diagnosis is easily reached and patient is managed accordingly. When the components are discordant, that is differ in their interpretation of the breast lump, what would be the next step forward is the area which needs more light to be shed upon. It is precisely in this area where triple test score can be the answer to this dilemma. TTS being non-invasive and economical, with certain diagnosis in most of the cases (except in score of 5) can be relied upon as an effective test for further management of the patient.

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RESEARCH ARTICLE

Assessment of Physico chemical changes in Nigeen lake.

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Abstract

A study of the water quality changes of NigeenLake was conducted for 6 months, which began in August 2010 to February 2011. Three sampling stations were selected representing the different areas in the lake. A total of 11 water quality parameters were measured. The physical and chemical variables were temperature, dissolved oxygen, conductivity, pH, total dissolved solid, turbidity, ammonia-nitrogen, nitrate-nitrogen and ortho- phosphate

. Due to siltation, encroachment and pollution the lake has become shallower and the lake shore has turned into marsh at various places. The higher ranges for Sp. Conductance ($192-560 \mu\text{Scm}^{-1}$ @ 25°C), Total-Alkalinity ($106-435 \text{ mgI}^{-1}$), Calcium ($29.6-44 \text{ mgI}^{-1}$), Nitrate-Nitrogen ($275-931 \mu\text{gI}^{-1}$), Ammonical-Nitrogen ($38-360 \mu\text{gI}^{-1}$), Total-Phosphorus ($122-722 \mu\text{gI}^{-1}$) which exceed the permissible limits is a cause of concern. Hence, the water quality is found to be poor due to increased anthropogenic pressures from the catchment area.

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Introduction:-

Dal lake ($34^{\circ}-06'N$, $74^{\circ}-45'E$; altitude 1583 m) is a post glacial lake with a shallow depth, bounded on the south-west by the capital city Srinagar and encompassed on the other sides by terraced gentle slopes at the base of the precipitous mountains. The Dal lake and its adjunct Nigeen has remained at the focal point of Kashmir's illustrious history, culture and tourism. Situated against the backdrop of Zabarwan, Shankaracharya and KohiMaran, the serene and placid waters have bestowed unparalleled beauty and magnificence, attracting the tourists from all over the world. The Nigeenlake offers a novel attraction to tourist because of its calm, placid and serene waters, water sports like skiing, swimming and staying in houseboats. The lake ecosystem of varied cultural ethos offers recreation to tourists who want to fish in its waters or indulge in bird watching. The sunset in Nigeenlake when the entire body of the Nigeen looks like a sparkling jewel to which the word 'Nigeen' is related.

Although extensive research has been conducted on Dal lake but the aim of present study is to provide the status of the Nigeen lake (which is an important basin of Dal lake) on the basis of physico-chemical analysis of water.

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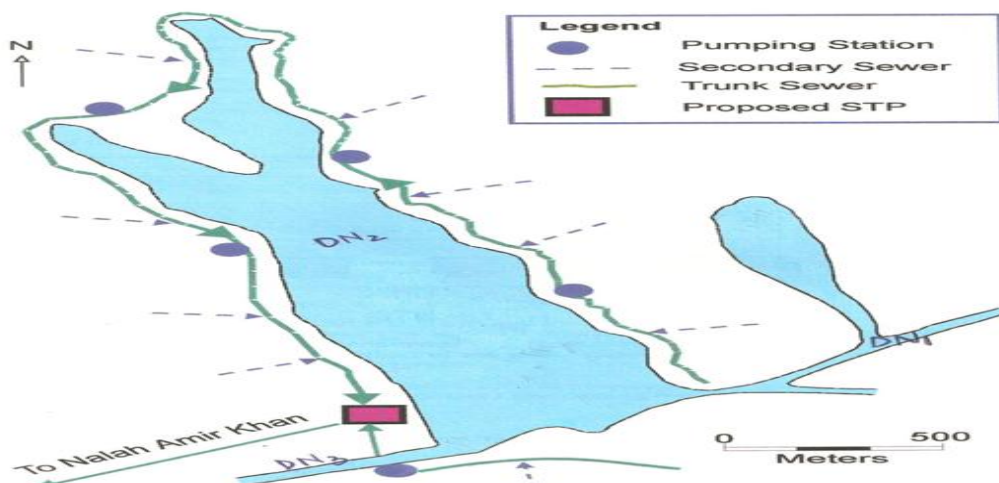



Materials and Method:-**Study site:-****Fig.1:-** Outline map Nigeen lake showing sampling sites**Table 1:-** Description of sampling sites.

Table 1. Description of sampling sites.																
S	i	t	e	C	o	d	e	S	i	t	e	i	m	a	g	e
D	N	1	Entry of water into Nigeen via Dal lake													
D	N	2	Nigeen Centre													
D	N	3	Exit of water from Nigeen to Anchar lake via Nallah Amir Khan													

Methodology:-

The water sampling at the desired sites and depths were taken by Ruttner's sampler. The sampling was done usually between 10:00 to 11:00 hours. The samples were immediately transported to the lab. for detailed analysis. The water analysis was carried out using the methods outlined in APHA, AAWA, WPEF (1997), Golterman and Clymo (1969) and Mackereth (1963), P. K. Gupta (2004).

S . N o	P a r a m e t e r	M e t h o d / E q u i p m e n t
1	Water Temperature ($^{\circ}\text{C}$)	D i g i t a l T h e r m o m e t e r
2	Water Depth (cm)	L e a d W e i g h t
3	Water Transparency (cm)	S e c c h i D i s c (2 0 c m d i a)
4	W a t e r p H	D i g i t a l p H M e t e r M K V I
5	Specific Conductivity (μS)	Digital Conductivity meter (Systronics)
6	Dissolved Oxygen (mg/l)	Winkler's Method -Azide Modification (APHA, 1998)
7	C h l o r i d e (m g / l)	Argentometric Method (APHA, 1998)
8	A l k a l i n i t y (m g / l)	Titrimetric Method (APHA, 1998)
9	A m m o n i a ($\mu\text{g} / \text{l}$)	P h e n a t e M e t h o d (A P H A , 1 9 9 8)
1 0	N i t r a t e ($\mu\text{g} / \text{l}$)	Salicylate Method (CSIR, 1974)
1 1	Ortho-Phosphorus ($\mu\text{g/l}$)	Stannous Chloride Method (APHA, 1998)

Results and Discussion:-

The monthly air temperature (fig 2) ranged between 6.5°C (Jan, 2011) to 30°C (Aug, 2010) at DN1 & DN3) with an overall average of 16.5°C during the investigating period.

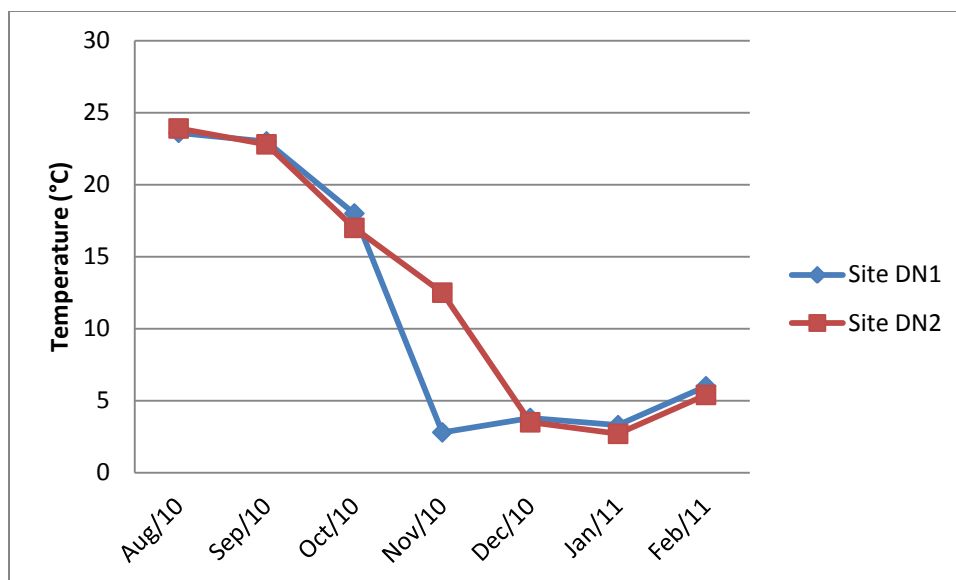


Fig. 2:- Monthly variation in Air Temperature (°C) at various sites of Nigeen Lake.

The monthly water temperature (Fig.3) ranged from 2.8 °C (Nov, 2010 at DN1) to 25.0 °C (Aug, 2010 at DN3) with an overall average of 11.9 °C during the study period.

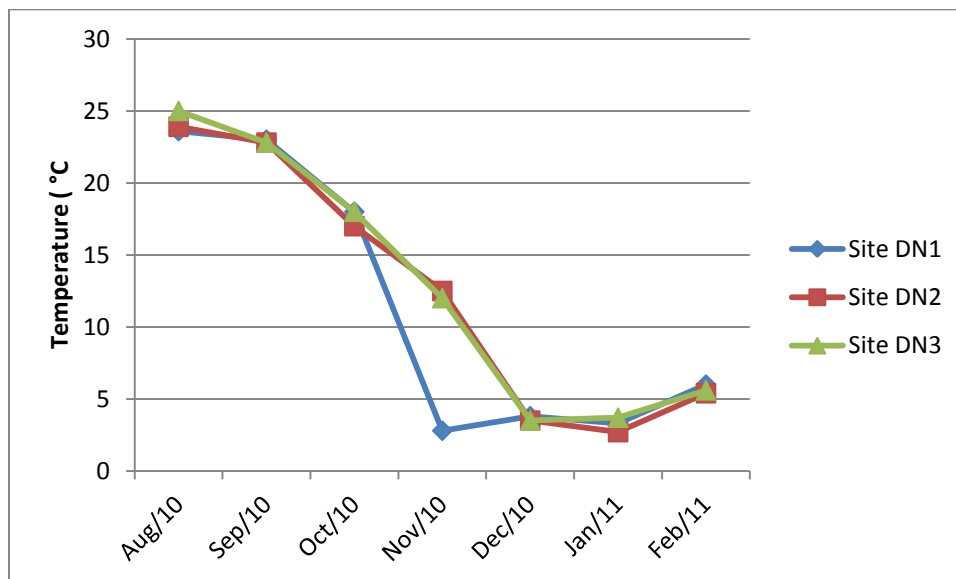


Fig. 3:- Monthly variation in Water Temperature (°C) at various sites of Nigeen Lake.

The monthly transparency measured at the study sites of Nigeen Lake is given in Fig 4. The value for transparency ranged from 0.6m (Sept,Oct, 2010 at DN1) to 2.7m (Jan, 2011 at DN2'S') with an overall average of 1.3m during study period.

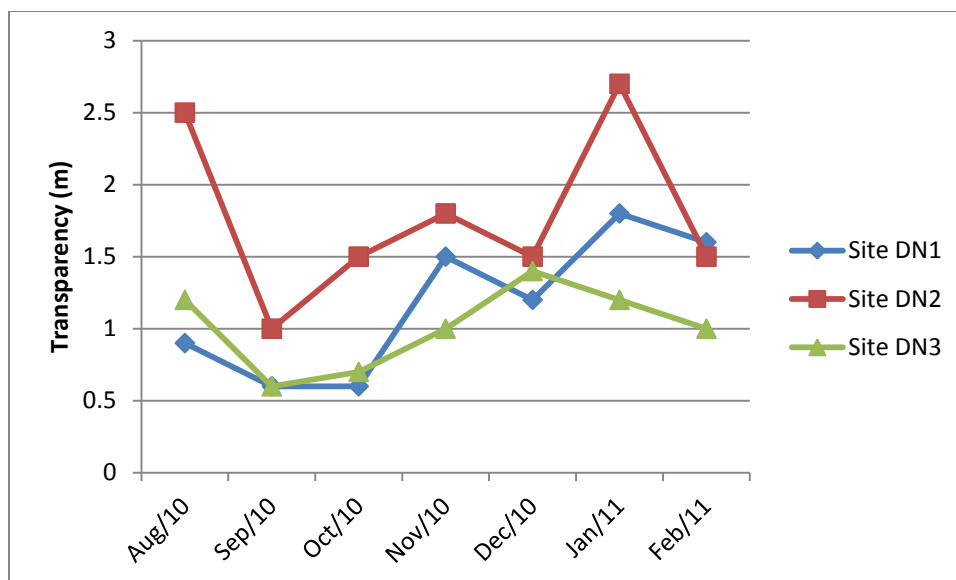


Fig 4:- Monthly variation in Transparency(m) at various sites of Nigeen Lake.

The monthly pH measured at the monitoring sites of Nigeen Lake is given in table (05). The value for pH ranged from 7.6 (Jan, 2011 at DN1) to 8.5 (Aug, 2010 in surface water at DN2) with an overall average of 7.8 during the study period.

The pH of water ranged between 7.5 to 8.5 during the study period and depicted alkaline nature of the water. No major shift has been recorded in pH concentration when compared with earlier records (table 1). The waters of Nigeen lake are thus well buffered which is in conformity with the findings of Edmodson and Hutchinson (1939), Zutshi (1968) and Kundangar and Adnan (2006).

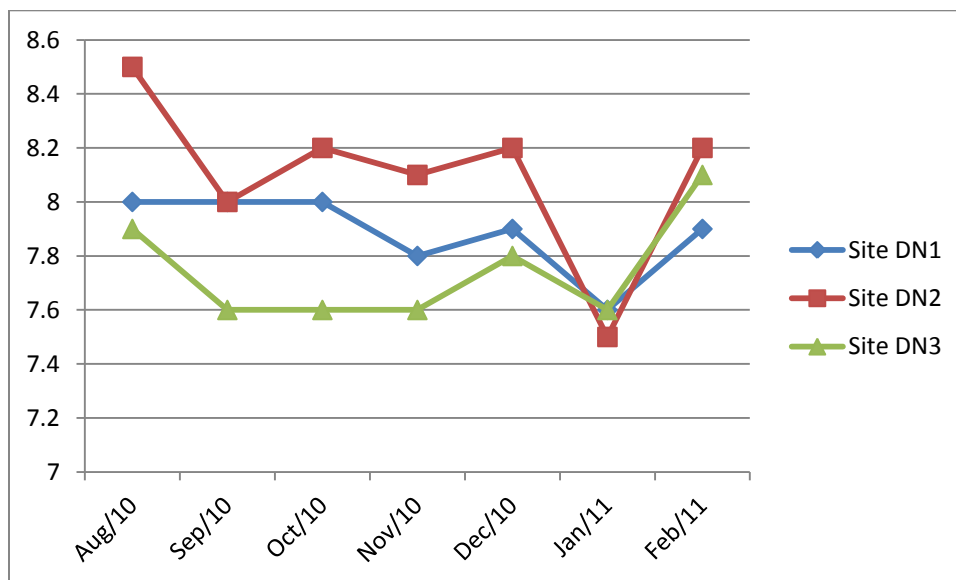


Fig 5:- Monthly variation in pH values at various sites of Nigeen Lake.

The monthly specific conductivity measured at the study sites of Nigeen Lake is given in Fig (06). It ranged between 192 μS (Aug, 2010 in surface water at DN2 & DN3 respectively) to 560 μS (Aug, 2010 in bottom water at DN2) with an overall average of 302.25 (μS at 25°C) during the study period.

The specific conductance of Nigeen water ranged from $192\mu\text{Sm}$ at 25°C to $560\mu\text{Sm}$ at 25°C which is higher than earlier records. Zutshi (1987) observed moderate conductivity value in Dal lake during 1980-1982 while as Kundangaret al.(1992-93) and Kundangar and Adnan(2006) reported progressive increase in conductivity values of Nigeen waters. Mathew and Vasudevan(2000) attributed the higher values of sp. conductivity to the mineral inputs while Agarwal and Kanan (1996) hold the concentration of particular ions, TDS and Electrical conductivity when keep rising, water quality may deteriorate. Nararian et al(1995) Jain et al.(1998) are of the view that rising electric conductance will deteriorate the physico-chemical and bacteriological status of the waters to a greater extent.

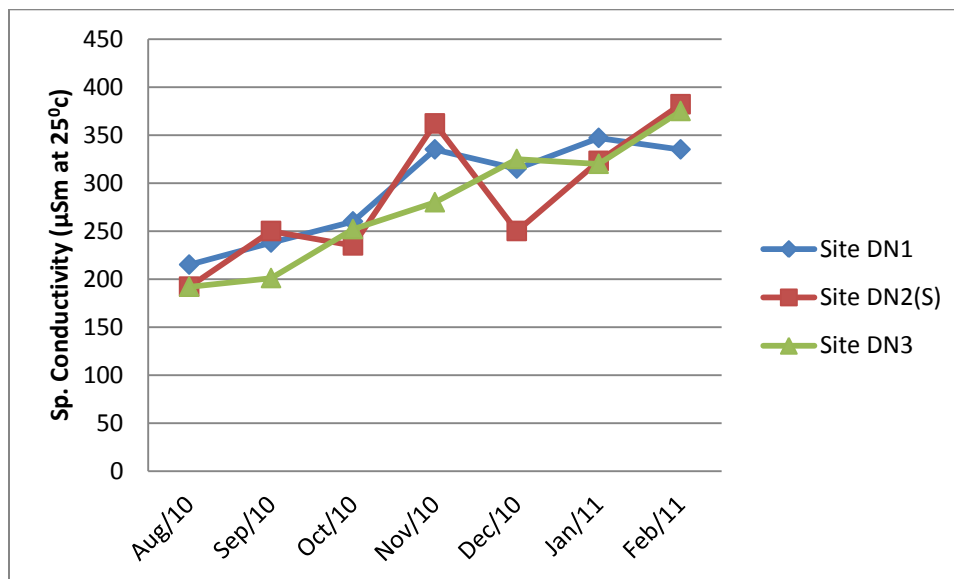


Fig 6:- Monthly variation in Sp. Conductivity (μSm at 25°C) at various sites of Nigeen Lake.

The monthly dissolved oxygen (mg l^{-1}) measured at the monitoring sites of Nigeen lake is given in Fig. 7. The value for dissolved oxygen ranged between 5.5mg l^{-1} (Oct, 2010 at DN1) to 12.0mg l^{-1} (Sept, 2010 at DN3) with an overall average of 6.4mg l^{-1} during the investigating period. The oxygen content in Nigeen waters ranged from 4.0 to 12mg l^{-1} and no major shift was observed than the earlier records where the oxygen content was recorded between 5.9 to 13mg l^{-1} Kundangaret al. (1992-93). However the oxygen depletion was observed in the bottom waters which can be attributed to summer stratification. These findings are in agreement with those of Zutshi(1987).

Das and Pande(1982) attributed the depletion of oxygen content in the bottom waters to anaerobic bacteria who take over the process of decomposition of biological organic matter releasing foul smelling hydrogen, methane and ammonia(aerobic bacteria evolve only CO_2 when organic matter is decomposed in presence of oxygen) these gases not only deplete oxygen in water but are toxic, killing most of the plankton, algae and zooplankton in the lake. A difference of 8mg l^{-1} of oxygen content was recorded by Kundangaret al. during (1992-93). According to Vass (1980) Nigeen shows a positive heterograde along with clinograde type of oxygen curve, which according to the author was biogenic. Kundangar and Adnan(2006) observed the depletion of oxygen content in the bottom waters of Nigeen throughout the year which according to authors depicted the shift in oxygen regime.

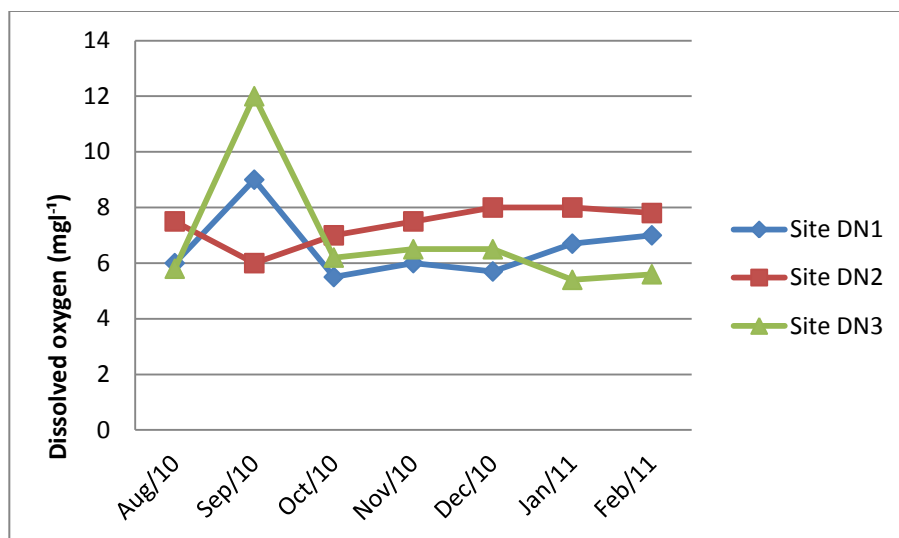


Fig. 7:- Monthly variation in Dissolved oxygen (mg l⁻¹) at various sites of Nigeen Lake.

The monthly total alkalinity measured at the study sites of Nigeen Lake is given in Figure 8. The value for total alkalinity ranged between 106 mg l⁻¹ (Aug, 2010 in surface water at DN2) to 435 mg l⁻¹ (Jan, 2011 in surface water at DN2) with an overall average of 226 mg l⁻¹ during the investigating period.

The total Alkalinity of the waters in Nigeen ranged between 106 to 435 mg l⁻¹ which was comparatively much higher than the earlier records of Kundangaret *et al.* (1992-93). The alkalinity was of bicarbonate type which is in conformity with Freiser and Ferriande (1996) who stated that when total-alkalinity is higher, the carbonate system persists and p.H usually remains alkaline.

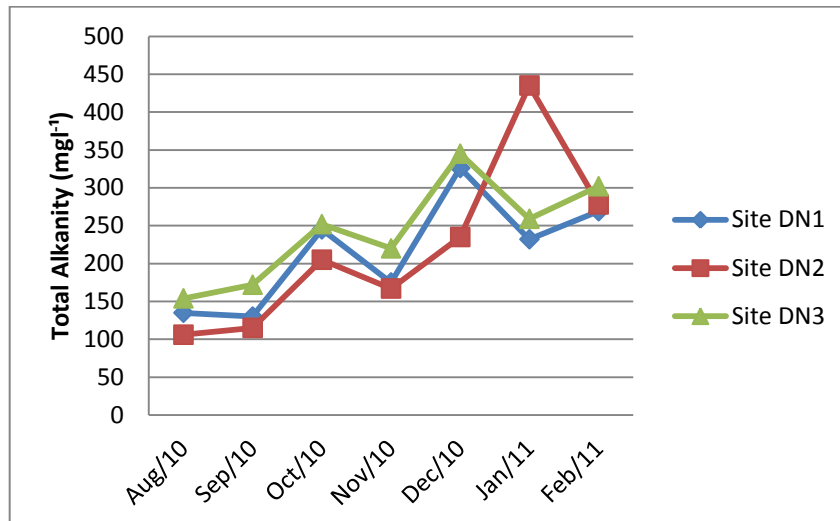


Fig 8:- Monthly variation in Total Alkalinity (mg l⁻¹) at various sites of Nigeen Lake.

The monthly calcium measured at the investigated sites of Nigeen Lake is given in Figure (9). The value for calcium ranged from 29.6 mg l⁻¹ (Nov, 2010 in surface water at DN2) to 44.0 mg l⁻¹ (Aug, 2010 at DN3) with an overall average of 36.3 mg l⁻¹ during the investigating period.

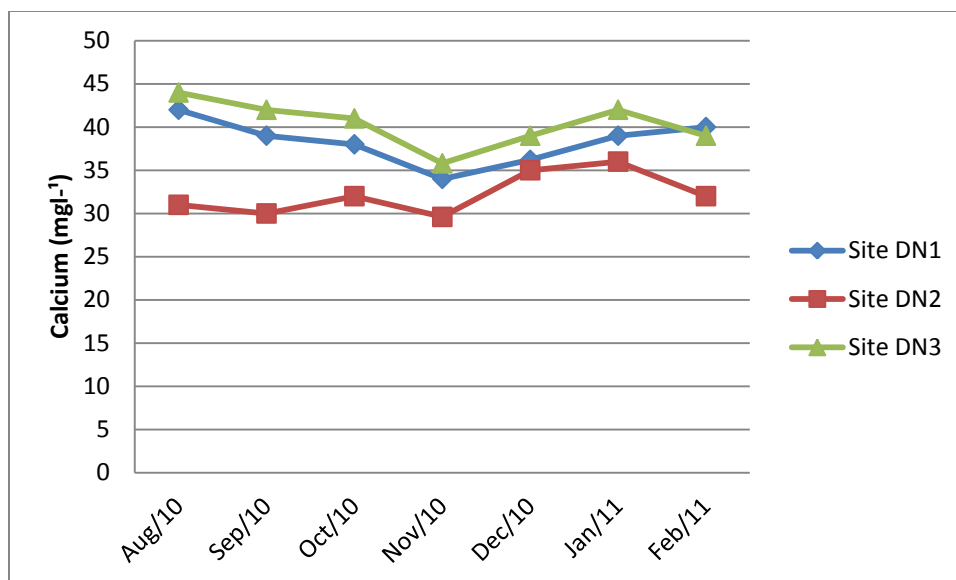


Fig 9:-Monthly variation in Calcium (mg l⁻¹) at various sites of Nigeen Lake.

The monthly magnesium measured at the monitoring sites of Nigeen Lake is given in Figure 10. The value for magnesium ranged from 0.9 mg l⁻¹ (Feb, 2011 at DN1) to 6.0 mg l⁻¹ (Aug, 2010 at DN3) with an overall average of 4.0 mg l⁻¹ during the study period.

The calcium content during the study period was recorded between 29.6 – 44 mg l⁻¹ which shows the lake water is basically a marl lake and the water is rich in calcium, similar observation were made by Kundangar *et al.* (1992-93) and Kundangar and Adnan *et al.* (2006). The values of calcium are much higher than those of Zutshi and Khan (1988). According to Kundangar and Adnan (2006) the high levels of calcium in water and presence of marl on the leaves and stems of the macrophytes in all the basins of Dal lake may adsorb liable organic substance and thereby limits the plankton growth.

The direct relationship between bicarbonates and calcium has been observed by Pearsall (1923) and Zaffar (1964), whereas the inverse relationship between carbonates and bicarbonates has been noted by Ganapati (1940). Average Magnesium content in the lake water with high dissolved organic content are alkaline in nature and magnesium salts are more soluble in them which is in conformity with the finding of Adnan and Kundangar (2008) while studying the ecological status of some flood plains lakes with Jehlum river basin of Kashmir.

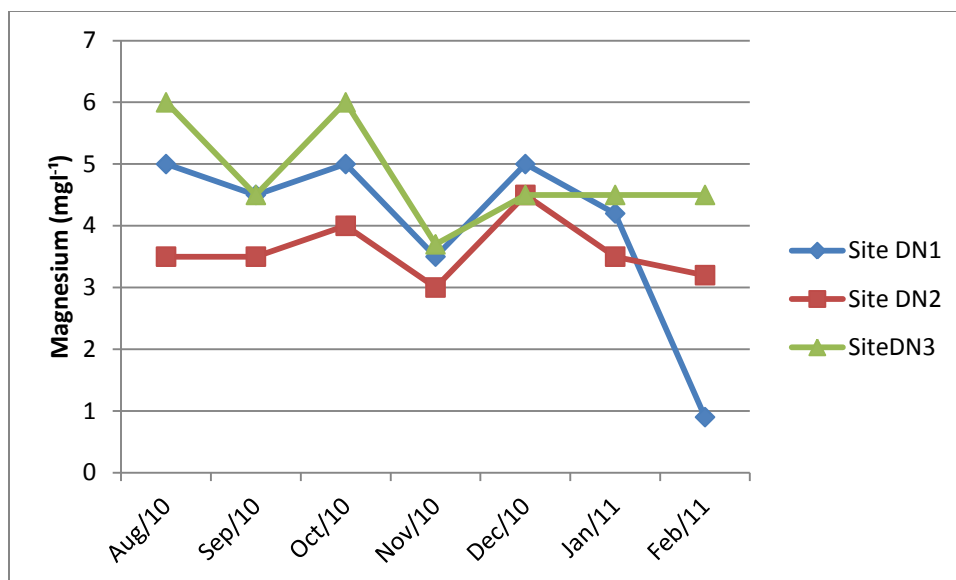


Fig 10:- Monthly variation in Magnesium (mg l⁻¹) at various sites of Nigeen Lake.

The monthly nitrate-nitrogen measured at the investigated sites of Nigeen Lake is given in table (13). The value for nitrate-nitrogen ranged from 275 $\mu\text{g l}^{-1}$ (Dec, 2010 in surface water at DN2) to 931 $\mu\text{g l}^{-1}$ with an overall average of 483.42 $\mu\text{g l}^{-1}$ during the investigating period.

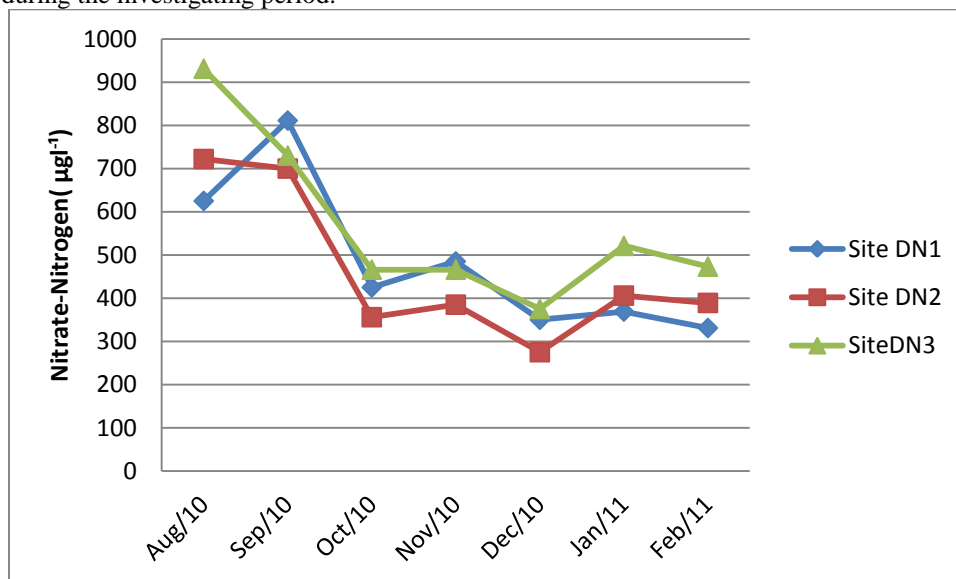


Fig 10:- Monthly variation in Nitrate-Nitrogen ($\mu\text{g l}^{-1}$) at various sites of Nigeen Lake.

The nitrate-nitrogen and Ammonical-nitrogen content in the Nigeen waters ranged from 275-931 $\mu\text{g l}^{-1}$ and 36-360 $\mu\text{g l}^{-1}$ respectively which is comparatively much higher than the earlier records of Vass(1980), Zutshi and Khan(1988), Kundangar *et al.* (1992-93) and Kundangar and Adnan(2006). Besle and Suckling(1944) have attributed the nitrogen richness of a fresh water body to the pollution of animal origin.

The monthly Ammonical nitrogen, measured at the monitoring sites of Nigeen Lake is given in figure 11. The value for Ammonical nitrogen ranged from 38 $\mu\text{g l}^{-1}$ (Aug, 2010 in surface water at DN2) to 360 $\mu\text{g l}^{-1}$ (Jan, 2011 in bottom water at DN2) with an overall average of 133.57 $\mu\text{g l}^{-1}$.

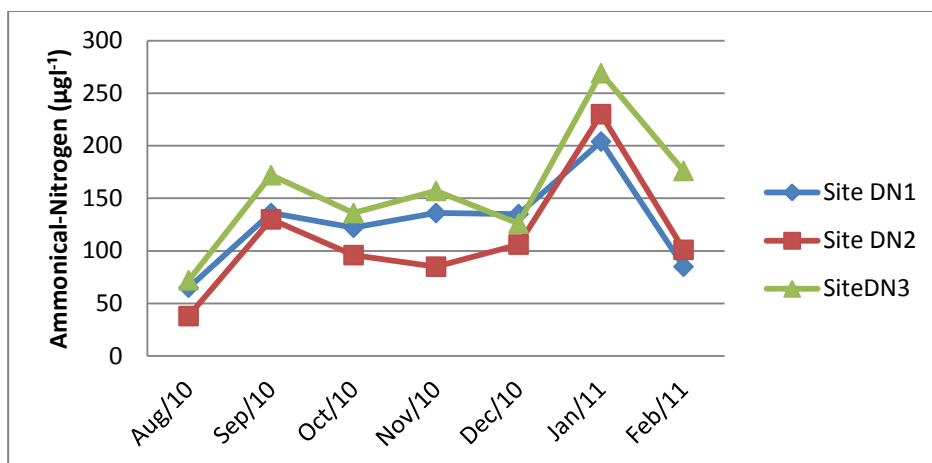


Fig 11:- Monthly variation in Ammonical-Nitrogen ($\mu\text{g l}^{-1}$) at various sites of Nigeen Lake.

The monthly ortho- phosphorus measured at the study sites of Nigeen Lake is given in fig. 12. The value for ortho-phosphorus ranged from $34 \mu\text{g l}^{-1}$ (Nov, 2010 at DN1) to $432 \mu\text{g l}^{-1}$ (Nov, 2010 in bottom water at DN2) with an overall average of $132.13 \mu\text{g l}^{-1}$ during the study period.



Fig 12:- Monthly variation in Ortho- Phosphorus ($\mu\text{g l}^{-1}$) at various sites of Nigeen Lake.

The Phosphorus content in the Nigeen waters ranged between $122\text{--}722 \mu\text{g l}^{-1}$ the values are much higher than those of earlier records. The higher Phosphorus content in the lake water can be attributed to the influx of raw sewage entering the lake through surface drains. Hutchinson (1957) also reported the increase of Phosphorus as a result of sewage contamination. Schindler *et al.* (1971) singled out phosphorus for attention because it is believed to be nutrient frequently controlling Eutrophication.

Lund (1965) and Einsele (1936) support the view that phosphorus play a major role in eutrophication as well as in production. Wetzel (1975) holds in the view that number of bacteria increase with increasing productivity and concentration of organic and inorganic compounds in lake.

In case of Nigeen waters Nitrogen and Phosphorus can be considered as two major elements responsible for pollution and limiting primary production. In this lake phosphorus is expected to be released in water from bottom mud during the oxygen deficiency in the bottom waters during the summer months. The water quality is found to be poor due to increased anthropogenic pressures from the catchment area.

Conclusion:-

The observation recorded during the study period reveal that the Nigeenlake has undergone marked change in morphometry including the depth. Due to siltation the lake has become shallower and the lake shore has turned into marsh at various places. The excessive inflow of nutrient rich effluents from the immediate catchment and also due to sedimentation the lake bed has not only become shallower but has resulted in serious weed infestation.

The unabated encroachments by way of extension of floating gardens beyond legal titles coupled with fertilizing method of using lake mud and weeds which eventually reaches to the bottom of the lake transforms floating gardens into permanent dry lands. Over a period of time such practices have continued and encroaching of open water body has become an unending problem.

The drainage, natural sedimentation and continued reclamation of the lake basin to provide vegetable growing land besides enormous increase in the area of floating garden have combined with natural process to reduce the area of open water within the basin. On assessment of physico chemical parameters, The lower values are recorded in dissolved oxygen content reaching to an anoxic condition at certain times which lead to algal blooms and fish kills. The pH has remained throughout alkaline indicating buffered nature of lake waters. The population explosion in the immediate catchment of Nigeen and increasing sewage loads has aggravated the ecological problems of the Nigeenlake. The changes in the water quality over a period of time must have brought insignificant changes in the biodiversity of the lake.

The laying of garland sewer for pollution abatement though initiated has not yet been completed, the construction of sewage treatment plants and their doubtful working have jeopardized the entire sewage scheme. The sewage treatment and solid waste management of the houseboats has remained an unaccomplished job till date and has become a matter of grave concern. The program of lake conservation for last three decades has been very slow and more of engineering nature rather than scientific.

The higher ranges for Sp. Conductance ($192-560 \mu\text{Scm}^{-1}$ @ 25°C), Total-Alkalinity ($106-435 \text{ mgI}^{-1}$), Calcium ($29.6-44 \text{ mgI}^{-1}$), Nitrate-Nitrogen ($275-931 \mu\text{gI}^{-1}$), Ammonical-Nitrogen ($38-360 \mu\text{gI}^{-1}$), Total-Phosphorus ($122-722 \mu\text{gI}^{-1}$) which exceed the permissible limits are worrisome as the lake waters are used for portable purpose by the PHE Deptt. Since the conventional treatment plants cannot remove the Nitrate-Nitrogen and Total Phosphorus content of the water, thus putting the health of thousands of consumers at stake.

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RESEARCH ARTICLE

STRATEGIC RESOURCES FOR SUSTAINABLE COMPETITIVE ADVANTAGE.

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SCA (sustainable competitive advantage), RBV (resources based view), Valuable, Rare, Not Imitable, Not Substitute, firm, EXSTRIM

Abstract

Companies are trying to position ourselves and try to have a sustainable competitive advantage, by leveraging the strength to exploit opportunities and neutralize threats, and can avoid or fix vulnerabilities that exist in the company. The gain the competitive advantage is the influenced by two factors, there are Internal environment and the external environment. Internal environment are important elements in the company. The Analysis of internal environment beneficial for the company, especially the resource analysis. This analysis is done for internal resource can be strength of the company, but also can be a source of weaknesses. Terms of resources can be a force if these resources are managed and empowered well, but on the contrary, if internal resources are not managed and controlled properly it will be the weaknesses of the company. Companies can achieve competitive advantage through resource, resources the company has four criterion the namely of Valuable, Rare, Low Inimitable Non- substitute (VRIN). However, based on this study Authors to identified resource from VRIN into EXSTRIM. The resources EXSTRIM that means of: Excellent, Not Substitute, Not Trade, Rare, Not Imitable, Mover & Modification can be the strategic resource configuration in the company to creating a sustainable competitive advantage (SCA).

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Introduction:-

Strategies management become indispensable in managing fluctuating environmental conditions, the environment can affect to the performance and company's goals. The company's strategy is formulated at three levels: operational level, functional level and strategic level, in the context of industry competition, the main concern is the strategic level. Each level of the strategy in general in order to create a competitive advantage that includes; cost leadership, focus and strategy that differentiate it from other companies Porter (1991). A business manager should be able to analyze the environment to understand opportunities and threats facing. Organizations need to build strength and improve weaknesses; this process is not just a single step, but a process that includes various steps. One of them is the scanning, the business situation, which, according to Albright (2004) scanning is an internal communication to get information relevant external information on the Issues of potential influence on corporate decision-making process in order to face the competition. The company may face competition when companies that have a sustainable competitive advantage by harnessing the power companies to exploit opportunities and neutralize the threat while avoiding or fixing a weakness or so-called analysis 'SWOT'. Porter's Five Force Model, Porter conducted an analysis of the competitiveness of the industry, which is affected by the five forces that emphasizes analysis of existing industrial structures. The Five forces such as; bargaining power of suppliers, bargaining power with customers, the strength of the threat of new entrants, as well as the strength of the threat from competitors as

well as existing industry. While Porter framework has provided many useful insights both practitioners and researchers to concentrate on external analysis 'OT' nevertheless still experiencing some significant problems. The same thing Porter (1991), Valentin (2001) the SWOT analysis is an analytical tool for the environment that identify strengths weakness opportunities and threat for the company. The business strategy is a way to master the wide expertise and managing a company's profit with its surroundings. To realize that all companies should determine the factors which might be in the face of competition once expected to be used to win the competition? One factor that can be used to win the competition is a strategic resource company. Determine the combination of the company's strategic resources is not easy, but if it is done well and seriously then the combination can be achieved with either. The company's strategic resource that can be used to win the competition must be resources that have a competitive advantage compared to the resources owned by other companies.

Understanding Resources:-

Many definitions of resources raised by the experts which is according to Barney (1991) Barney define resource company covers all assets, capabilities, organizational processes, firm attributes, information, knowledge, which is controlled by the company. According to Anderson (2007) the resource can be divided into internal and external resources, external resources consist of relations with actors outside the company; customers and suppliers as well as its reputation. Javad & Carlos (2010) introduced the idea that the company should be analyzed in terms of resources, not only in terms of its products alone. According to resources based view (RBV), resources are defined as: anything that can be regarded as a strength or weakness of a particular company. Resource companies as a factor in the company and are owned or controlled by the company. The resource is generally defined as "all assets, capabilities, organizational processes, firm attributes, information, and other knowledge that can be controlled by the company, resource as an asset or an input to production tangible or intangible owned by an organization and can be controlled, or in access to a semi-permanent.

Resources Advantage:-

According to Anderson (2007) special ability to create such varied resources management, entrepreneurial and dynamic capability has been used to describe the ability to generate strategic resources. Furrer at.al (2008) indicates that the resource-based grouping took the example of a bank that explains differences in corporate performance better than the market grouping based products. However, market-based group still provides explanations of the significance of the difference in performance in two of the three performance measures used. Meanwhile, according to Grant (1991) by using the difference between the resources and capabilities, resources are tangible or intangible assets, and capabilities are the ability generated from these assets. Anderson (2007) the resources can lead management capabilities such as the ability of the market, production capabilities, and the ability of innovation. To obtain optimal composition, the researchers suggest that the fit between the available resources of the company and the requirements of the product-market activity in terms of concepts such as generic strategies should be carried out it is appropriate revelation (Furrer at.al 2008). Resources Strategic Change, and Organizational Performance The concept of strategic Resources by Kraatz & Zajac (2001) the resource that has an advantage can influence strategic changes to the company, the company can emphasize how resources can hinder or may lead to organizational learning in order to adapt. According to the opinion Banrey (1991) the latest research is a strategic resource is a resource company that is difficult to replicate. Meanwhile, according to Chaharbaghi & Lynch; (1999) indicates that strategic resources are not the only applications that efficiently and effectively to provide the level of high competitiveness and ability to generate profits, but more important than that is how to make resources the mutual synergy so really widened superior resources and strategies for the company in realizing a sustainable competitive advantage. One way to the competitive advantage if the company has the dynamic strategic resource. The dynamic the resources within the company the combination of resources within the organization based on the time dimension will always be changing its composition, therefore, according to the authors' opinion that in order to realize the resource made resources that strategic managers must be able to combine existing resources in the company. According to Großler (2007) that resources are located will affect the performance capability of the organization / company (if not, they will not be located) there is a reciprocal process: the company's performance also affects resource and capability. Großler (2007) exemplifies the relationship between the ability to produce products with high quality and sales revenue as a performance indicator. Producing high quality (among other factors) affects the sales, which determines the sales revenue. Indicator of the understanding of strategic resources and its relation with the ability to improve the performance of the company. As Großler (2007) Barney (1991) found a strategic resource must have at least four major characteristics, namely: Durable, has a positive long-term effect on the company's strategic position; Non-tradable, it is impossible for competitors to easily afford; Non-applicable, it is impossible for

competitors to easily duplicate or copy them; Non substitutable, it is impossible for competitors to easily find a replacement.

Meanwhile Barney (1991) that a strategic resource that can create competitive advantage and sustainability performance is a resource that has the following criteria: Valuable (V): Resource-value, the resource company that has a value that is able to set up the company in creating market opportunities and can reduce the challenges that exist in the market. Rare (R): The resources are scarce (unique), whose existence is difficult to find the resources and these resources are unique and not shared with other companies. Imperfect imitation (I): The resource that is difficult to imitate, this resource is difficult to imitate by other companies. Non-substitutability (N): The resources irreplaceable, resources in the other companies are difficult replacement to match.

Based on several definitions of strategic resources there is a difference between an expert opinions with another expert opinion, the difference is due to the perspective and different educational backgrounds. Therefore, the author tries to back describe strategic resources to get a thread or a new definition of the strategic resources. But before describing the author or authors redefine about strategic Resources will provide an overview of some of the definitions neighbor Strategic resources being addressed by some experts that will be displayed in the following table.

Table :- Definition of Strategic Resources

No	Author	Definition
1	Banrey (1991)	Business resources and difficult to imitate
2	Black and Boal (1994)	Identify how the performance of the company and its sustainability depends on the uniqueness, rarity.
3	Chaharbaghi & Lynch (1999)	Strategic resource not only the application of efficient and effective to provide the level of high competitiveness and ability to generate profits, but more important than that is how to make resources the mutual synergy that really widened resource excellence and strategic for the company in realizing SCA
4	Kraatz & Zajac (2001)	Resources that have the advantage may affect strategic change in the company, the company can emphasize how resources can hinder or may lead to organizational learning in order to adaptation
5	Anderson (2007)	Special ability to create such varied resources management, entrepreneurial and dynamic capability has been used to describe the ability to generate strategic resources.

Based on Banrey (1991) the identification of the resources are: V: Valuable, R: Rare, I: Not Inimitable N: Not Substitute, Black and Boal (1994) identification of the resources are VRIO that mean are V: Valuable, R: Rare, I: Not Inimitable, O; Organization Orientation, they are differences existing definition. So the Author can the conclusion of the strategic resources is "The unique resource obtained through a process that is adaptive and proactive to spearhead (guns) as a means to win the competition and excellence continuous competitiveness. This resource should have the first criteria; excellent namely resource superior or resources are: Excellent, Second: Not Substitute are resources that not be replaced, Third: Not Trade resources that are not easily assessed by monetary or not be traded, Fourth: rare is a scarce resource that is difficult to obtain, Fifth: Not Inimitable that the resources that are difficult to imitate by other companies, Sixth: Mover & Modification is easy to be adapted to the dynamically changing environment, means that in the past, now becoming a strategic resource, but not necessarily in the future come can be a strategic resource back. Therefore, if the company to gain a sustainable competitive advantage the company must make resource adaptive, proactive and reactive to the environment. Based on the above, the Authors refer to as strategic resources are: EXSTRIM or the means of: Excellent, Not Substitute, Not Trade, Rare, Not Inimitable, Mover & Modification.

Anderson (2007) The ways to realize the strategic resource contribution-based view of research taking the difference in the configuration of resources between companies, if there is a description of how these strategic resources are developed, the discussion usually focuses on direct investments as a way to obtain strategic resources. However, differences in the configuration of resources between companies can also be explained by factors other than the difference in making investment decisions. Usually the activity that takes place within the company in terms of

organizational processes and activities carried launching products in the market and can also develop resources in order to obtain strategic resources. Thus, a strategic resource can be obtained in three ways:

1. Through direct investment;
2. Through the process of organization, and
3. Through the activities undertaken in the product markets.

Barney (1991) identifies the barriers that used to barrier process of imitation, the things that will be used when analyzing barriers to imitation are:

1. The social complexity;
2. Ambiguity causal,
3. Conditions unique history

Social complexity refers to the intangible factors such as social and cultural relations, while the causal ambiguity exists when the relationship between resources and competitive advantage is unknown. Conditions unique history confirms that each company is a unique entity and that the possibility of acquiring the resources depending on the time point, where and when to get those resources.

Strategic Concept advantage:-

According to Barney (2002) the company will gain a competitive advantage if the company's strategy and values which not be done by the company's competitors. Strategic advantage, on the other hand, covers a broader perspective in which this matter, so Chaharbaghi & Lynch (1999) include the following:

1. Provide the organization with a unique ability to develop strategic directions that create new opportunities and future competitive environment.
2. Is a source of leadership resources in one or more of the products on the market.

Sustainable competitive advantage (SCA) is a process that needs to compete today without compromising the ability of organizations to meet the requirements to compete in the future. Chaharbaghi & Lynch (1999) the process of developing such an organization as a dynamic concept, which covers the basics of the following:

1. Sustainable Competitive Advantage is a conservation-oriented as the organization does not have unlimited resources. Given these limitations, conscious management of existing resources is needed.
2. Sustainable Competitive Advantage is based on the needs of the organization in which economic activities should be concerned with the needs of the market for goods and services resulting from economic activity.
3. Sustainable Competitive Advantage is future-oriented, placing itself outside day-to-day management of long-term development resources to bring strategic advantages
4. A Strategic Advantage is the standard for corporate renewal, it is very dependent and influenced by, the development and timely introduction to strategic resources.

Based on the results from the description above one of the factors that can be used to create a sustainable competitive advantage SCA is a resources firm. Configuration was good between resources is expected to be decisive to the sustainable competitive advantage.

Conclusion:-

Based on the discussions that have been submitted at the beginning to the end that was found disagreement among experts on the categorizing of corporate resources is generally grouped into three categories, namely: physical, tangible resources, intangible resources and human resources. A good combination between the three elements contained in these resources is expected to establish a sustainable competitive advantage SCA. For the competitive advantage for the company to be created in earnest with how to configure the use of the existing resources within the company to the optimum. Appropriate combination or selection of the resources for the company is the main capital in determining the level of a sustainable competitive advantage. The resources based view RBV has contributed much to the formation of a strategic resource in promoting a sustainable competitive advantage. Resources that have a sustainable competitive advantage if they meet the criterion VRIN namely; Valuable, Rare, Non Imitable and Non Substitute. Meanwhile, according to the Authors that strategic Resources should have the criterion: first Excellent is the resource superior or resources excellent, Second: not Substitute is the resources that not be replaced, Third: Not trade is the resources that are not easily assessed by monetary or not be traded, Fourth: Rare is the resource that is difficult to obtain, Fifth: Not Imitable is the resources that is difficult to imitate by other companies, Sixth: Mover & Modification is easy to be adapted to the dynamically changing environment, means

that in the past, is now becoming a strategic resource, but not necessarily in the past that will come can be a strategic resource back. Based on the above, the Authors refer to as a resource to resource strategic are the EXSTRIM resources criterion.

Recommendation:-

The first performed by the expert company strategy is the identification of all the factors that may affect its business. In this process, that important to know and to do is the related to internal activities involved in the business, this includes all the systems of internal structure, strategies used, and the culture of the organization. The company's activities will interact with the environment components outside the company such as customers, competitors, and supplier; in addition it includes all parties (stakeholders) such as trade unions, media, and government. Competition has entered a new era with the constant changes. The infrastructure problems that disrupt the organization can't be solved effectively without resource-based strategy is therefore necessary to incorporate the following principles:

1. Strategic resources must be regenerated more quickly than those used by other companies
2. The resources must be used more quickly excel
3. Basis of resources should be considered to be invested faster than the rate of acceptance of the environment (adaptive) and proactive to realize resource from VRIN criterion to E X S T R I M to targeting in the sustainable competitive advantage (SCA).

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RESEARCH ARTICLE

Digital Wiring Harness Tester.

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Abstract

This paper consists of a distributed cable harness tester. It has a few functions such as connection and detection of a wire, proper connections of wire etc. In this project, application is designed in order to improve the tester's performance, and the software design of upper computer and the framework of hardware of tester nodes are introduced in details. The tester is designed to ensure quality and reliability of cable harness. In detection process early failure products such as breakage circuit, short circuit can be rejected. We can also print the data regarding the Harness. It improves the efficiency of system, decreases the length of cable, the cubage and weight of the equipment and makes the test and maintenance convenient and fast.

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Introduction:-

A cable harness, also known as a wire harness, cable assembly, wiring assembly or wiring 100m. It is an assembly of cables or wires which transmit signals or electrical power. A wiring harness can be very small in size such as a small group of wires found in your computer or car. A wiring harness can also be very huge such as some of the ones that Interconnect manufactures for aircraft. These huge wiring harnesses may have thousands of wires in them and hundreds of connectors. Wiring harnesses have many different names including: electrical wiring harnesses, looms, cable assemblies, coax cables, RF cables, injection molded cable assemblies, fiber optic cables, and many times are simply called harnesses. It is much easier and faster to route a group of wires already bundled together than routing them individually.

A cable harness tester it has a few functions such as connection detection of a wire, diode orientation testing and resistor's etc. The application is designed in order to improve the tester's performance, and the software design of upper computer and the framework of hardware of tester nodes are introduced in details. The tester is designed to ensure quality and reliability of cable harness. In detection process early failure products such as breakage circuit, short circuit and wrong conductor arrangement can be rejected. It improves the efficiency of system, decreases the length of cable, the cubage and weight of the equipment and makes the test and maintenance convenient and fast.

Faulty End Recognition tool identifies wiring faults and their physical location of the product. This tester is fast, accurate and easy to use and saves a lot of time involved in manual checking. Harness testers have a unique, patented graphic display to pin-point problems when wiring errors are detected, and offer one second pass/fail testing for production environments. Quickly locate intermittent-connection and identify their position in the cable.

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Harness Testers are commonly used in automobiles, as well as construction machinery, cable harnesses provide several advantages over loose wires and cables. For example, many aircraft, automobiles and spacecraft contain many masses of wires which would stretch over several kilometers if fully extended. By binding the many wires and cables into a cable harness, the wires and cables can be better secured against the adverse effects of vibrations, abrasions, and moisture. By constricting the wires into a non-flexing bundle, usage of space is optimized, and the risk of a short is decreased. Since the installer has only one harness to install (as opposed to multiple wires), installation time is decreased and the process can be easily standardized. Binding the wires into a flame-retardant sleeve also lowers the risk of electrical fires. Perform high speed electrical interconnect testing on cables and harnesses for wiring correctness and insulation quality.

Block diagram:-

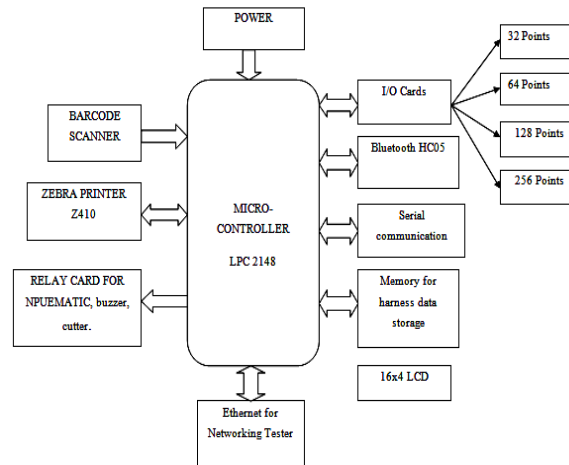


Figure 1: Block diagram of Harness Tester

Working: The wires are first cut to the desired length, usually using a special wire-cutting machine. The wires may also be printed on by a special machine during the cutting process or on a separate machine. After this, the ends of the wires are stripped to expose the metal (or core) of the wires, which are fitted with any required terminals or connector housings. The cables are assembled and clamped together on a special workbench, or onto a pin board (assembly board), according to the design specification, to form the cable harness.

LPC2148 Controller has limited i/o pins, but we needed large no. of i/o pins, so we can use the i/o cards. Which is extended the i/o pins. That is depends on the how many inputs or cables can be checked. i/o cards are use for expanding the input output connections.

These are four points for checking the wires/cables like A, B, C, D. In first case: If A point connected to C point and B point connected to D point then the message displayed is "harness is OK" on LCD. In second case: If A point connected to D point and B point connected to C point then the message displayed is "harness is EXCHANGED" on LCD. In third case: If A point connected to C point & B point is not connected to D point then the message displayed is "harness is OPEN" or no connections between B & D.

In this project we can use barcode scanner, which is used for scanning the barcode of product and they display the specifications or any other information of that product.

Ethernet is used in LAN connection, it means as earlier one man use to supervise going to every machine, now it can be supervised on one PC connected to other PC's on other machines. Printer is used in order to print any data of the product needed on papers. Thus we are reducing man power and making task earlier by having many functions on one device.



Figure 2: Digital Harness Tester

Results:-

There are many results we can display on LCD as follows:

1. Firstly the options are displayed on LCD that are, TEST WIRE, SET RTC, TIME/DATE, SEND HRN(Harness)DATA.



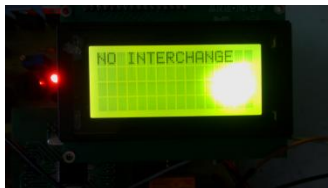
2. In second case we can check the points. Suppose two points are open then it will display that as OPEN POINT. In below picture point 1 13 are opened.



3. In third case we can check the faulty points. Suppose some faults occur in harness then it will display that as FAULTY POINTS. In below picture point 4 16 are faulty.



4. In fourth case we can check the interchange connections. Suppose point 1 is connected to point 15 and point 2 connected to point 16 then there is interchange between that points. But in below picture all points are connected to correct points so there is NO INTERCHANGE.



5. In fifth case suppose some connections are required between points then it will display REQUIRED POINTS. Shown as below.



6. In sixth case we can check the harness. If data is not passed through harness then it will display HARNESS FAIL. Shown as below



7. In seventh case we can check the harness and if harness is ok or good then it will display HARNESS PASS. As shown below.



Advantages:-

1. All functions are available in one device.
2. Less time consumption.
3. Reduced man power.
4. Accuracy.

Applications:-

1. Complex wiring testing (open circuit, wrong connection, wire interchange)
2. Electronic devices. (oven, fridge, washing machines etc.)
3. Automobile Industries. (vehicles etc.)
4. Aircrafts.

Conclusion:-

Hence in this project we solved the problems of time consumption, man power etc. which occurred in earlier projects of Harness tester. Harness Tester is designed to test a harness by Checking each point with each other point. IT learns the harness and Compare with the already stored harness information and locates open points, extra points, group short. Cable Harness Tester facilitates two modes of testing (auto / manual) harnesses. Testing speed of harness is high .It also facilitates indirect saving of space by accommodating different types of harnesses on rig.

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RESEARCH ARTICLE

FREQUENCY DOMAIN ANALYSIS OF HEART RATE VARIABILITY BETWEEN PREMENOPAUSAL AND POSTMENOPAUSAL KNOWN DIABETICS - A COMPARATIVE STUDY.

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Key words:-

Diabetes Mellitus, Heart Rate Variability, Premenopausal and postmenopausal ageing, estrogen.

Abstract

Back Ground: Diabetes mellitus mainly type II is mainly due to insulin resistance. The hyperglycemia caused by diabetes lead to micro and macrovascular complication that endangers with life. Premenopausal women with diabetes had the risk similar to that of men with diabetes. Postmenopausal diabetic women had additional risk due to estrogen hormone deficiency.

Aim And Objective: To compare the Frequency Domain Analysis of Heart Rate Variability between premenopausal and postmenopausal known diabetic females.

Materials& Methods: 100 Type II diabetic females around the age of 40-65yrs (both pre and postmenopausal) from diabetic OPD were recruited from Stanley Medical College Hospital. Institutional Ethical committee approval was obtained. After obtaining written and informed consent from the subjects. ECG(LEADII) was recorded for five minutes in supine position using RMS Digital Polyrite. HRV analysis was done using Frequency domain methods using RMS Digital Polyrite software version 2.1.

Results: Our study states that there is a lower HRV in postmenopausal known diabetic females when compared to that of premenopausal known diabetics. Further, decline in estrogen level and diabetes gives a additional risk of increased sympathovagal balance in postmenopausal diabetic women.

Conclusion: Type II postmenopausal diabetic females have increased level of autonomic dysfunction. Hence they require hormonal replacement therapy, regular periodic evaluation of cardiac autonomic status in order to prevent future cardiovascular morbidity and mortality.

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Introduction:-

Diabetes Mellitus is a group of common metabolic disorders that share the phenotype of uncontrolled blood sugar levels (hyperglycemia). The metabolic abnormalities associated with diabetes mellitus causes secondary pathological changes in multiple organ system that impose a tremendous burden on the individual with diabetes and on the health care system¹. With evolving trend across worldwide, Diabetes Mellitus will be a leading cause of mortality and morbidity. The worldwide prevalence of Diabetes Mellitus has risen drastically over past two decades, from an

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estimated 30 million cases in 1985 to 285 million in 2010. In individuals aged more than 65 years, the prevalence was 26.9%. Worldwide estimates project that in 2030 shows that the greatest number of individuals with Diabetes will be aged 45-64 years¹. Diabetic Autonomic Neuropathy (DAN) is among the most recognized and silent complications of diabetes, in the face of its significant harmful impact on survival and quality of life in people with diabetes². DAN may be either clinically evident or subclinical³. Reduced heart rate variability is the earliest indicator of CAN⁴.

Materials & Methods:-

100 Type II Diabetic females around the age of 40-65 yrs (both pre and postmenopausal) from diabetic OPD with duration of diabetes of 5-15 yrs with Random Blood Sugar ≥ 200 mg/dl or Fasting Blood Sugar ≥ 126 mg/dl were recruited from Stanley Medical College Hospital. Institutional Ethical committee approval was obtained. After obtaining written and informed consent from the subjects ECG (LEAD II) was recorded for five minutes in supine position using RMS Digital Polyrite. HRV analysis was done using Frequency Domain methods using RMS Digital Polyrite software version 2.1.

Exculsion Criteria:-

Subjects with a history of Asthma, Hypertension, Cardiovascular Disease and those on Chronic Medication.

Experimental Protocol:-

The short term Heart rate variability recording is usually performed for research, clinical investigations and followed the procedure given in the Task-Force report on Heart Rate Variability. Subjects were instructed to avoid heavy physical activity and also instructed to refrain from all caffeinated beverages for 12 hours prior to research activity. All the study subjects and controls have a prestructured proforma completed. Subjects were screened after measuring height, weight, blood pressure. The basal recording of blood pressure was done using sphygmomanometer by standard Riva Rocci method. Ask the subjects to lie down comfortably in the supine position in the Neurophysiology lab, Department of Physiology, Stanley Medical College. (5 mins rest). Placed the ECG electrodes on the limbs of the subjects and connect the leads to the machine for lead II ECG recording. Transfer the data from RMS Polyrite to window based PC loaded with software for Heart rate variability. Removed ectopics and artifacts from the recorded ECG. Extracted the R-R tachogram from the edited 256-second ECG using the R wave detector in the Acq Knowledge software and saved it in the ASCII format which is later used offline for short-term HRV analysis. Performed HRV analysis using the HRV analysis software version 2.1 (Biosignal Analysis group, Finland). Mean R-R is measured in second(s). Variance, defined as power in a portion of the total spectrum of frequencies, is measured in milliseconds squared (ms²). Mean R-R is measured in seconds.

Parameters Studied:-

Spectral indices (LF ms², HF ms², LF/HF ratio) are calculated.

Statistics:-

Data are expressed as mean \pm SD. Data between the study groups were compared using unpaired Student t-test. Differences were considered statistically significant at ($P < 0.05$). The collected data was analysed with SPSS 16.0 version. Data were normally distributed based on the Kolmogorov-Smirnov Z test for normality. To describe about the data mean and S.D was used. To find the significant difference between the Patients and controls Independent t-test was used.

Results:-

Table 1:- Subjects Characteristics, Anthropometric Measures.

PARAMETER		MEAN	SD	t-value	p-value
BMI	pre	173	35	1.02	0.315
	post	29.8	150		
SBP(mmHg)	pre	130.2	6.6	7.333	0.000
	post	141.4	6.2		
DBP(mmHg)	pre	34	6.0	7.666	0.000
	post	42.3	36		

No significant difference between pre and postmenopausal study subjects

Table:- Frequency Domain Analysis Between Pre And Postmenopausal Known Diabetic Individuals

		MEAN	SD	T-VALUE	p-value
Pm2LF	PRE	1109.6	160.2	2.999	0.001
	POST	3304.1	499.9		
Pm2HF	PRE	501.1	222.1	10.989	0.000
	POST	190.9	180.1		
LF/HF	PRE	3.8	1.2	18.989	0.000
	POST	19.9	5.9		

Compared to premenopausal diabetics, postmenopausal diabetics had lower HF, Higher LF and high LF/HF ratio.

Discussion:-

Diabetes Mellitus is characterized by hyperglycemia mainly in Type II diabetes due to reduced action of insulin (Insulin resistance)¹. It is the major cause for cardiovascular morbidity and mortality. The main advantage of using frequency domain analysis of Heart rate variability is that one can study the signal's frequency-specific oscillations. Thus both the amount of variability and the oscillation frequency (number of heart rate fluctuations per second) can be obtained. Spectral analysis involves decomposing the series of sequential R-R intervals into a sum of sinusoidal functions of different amplitudes and frequencies by the FFT algorithm. The LF fluctuations are predominantly under sympathetic control with vagal modulation, whereas the HF fluctuations are under parasympathetic control². Three main spectral components are distinguished in a spectrum calculated from short-term recordings of 2 to 5 minutes^{6,7,8,9,10}: VLF, LF, and HF components. Frequency domain analyses contributed to the understanding of autonomic background of RR interval fluctuations in the heart rate record.^{11,12} Silent ischemic heart disease or cardiac arrhythmias have both been invoked as contributors to sudden death. In Asymptomatic Diabetics (DIAD) study of 1123 patients with type 2 diabetes, cardiac autonomic dysfunction was a strong predictor of ischemia. Results from the European Diabetes Insulin-Dependent Diabetes Mellitus (IDDM) Complications Study showed that patients with impaired HRV had a higher corrected QT prolongation than without this complication. Cardiac autonomic neuropathy (CAN), which can be documented by abnormal heart rate variability (HRV), occurs commonly in patients with diabetes and is associated with silent myocardial ischemia⁶ and increased mortality⁷. In a recent large meta-analysis, Maser et al. reported that the presence of cardiac autonomic neuropathy was associated with a greater than threefold increase in mortality and sudden death⁷. Autonomic imbalance between the sympathetic and parasympathetic nervous systems regulation of cardiovascular function contributes to metabolic abnormalities and significant morbidity and mortality for individuals with diabetes.⁸⁻¹⁰ The presence of CAN was associated with a greater than threefold increase in mortality and sudden death. Silent ischemic heart disease or cardiac arrhythmias have both been invoked as contributors to sudden death. Meta-analyses of published data demonstrate that reduced cardiovascular autonomic function as measured by heart rate variability (HRV) is strongly associated with an increased risk of silent myocardial ischemia.^{9,15} Regular HRV testing provides early detection and thereby promotes timely diagnostic and therapeutic interventions. HRV was found to be an independent predictor of all-cause mortality during a period of 9 years, in a population-based study using Cox proportional hazard models. Moreover, the Hoorn study by Gerritsen et al demonstrated that impaired autonomic function is associated with increased all-cause and cardiovascular mortality and that CAN in patients already at risk (diabetes, hypertension, or history of CVD) may be especially hazardous. Clinical manifestations of cardiovascular autonomic dysfunction (e.g., exercise intolerance, intraoperative cardiovascular liability, orthostatic tachycardia and bradycardia syndromes, silent myocardial ischemia) can result in life-threatening outcomes¹⁰⁻¹⁴.

Results:-

Compared to premenopausal diabetics, postmenopausal diabetics had lower estrogen level, lower HF and high LF/HF ratio. Further, decline in estrogen level and diabetes gives an additional risk of increased sympathovagal imbalance in postmenopausal diabetic women.

Conclusion:-

The postmenopausal women had a significantly reduced overall fluctuation in autonomic input demonstrated by lower HF, increased LF, HF ratio in postmenopausal diabetic suggests that more sympathetic dominance. Therefore my study suggests that decline in levels of estrogen from pre to postmenopausal makes shift of autonomic balance towards the sympathetic dominance. Type II postmenopausal diabetic females have increased level of autonomic dysfunction. Hence they require hormonal replacement therapy, regular periodic evaluation of cardiac autonomic status in order to prevent future cardiovascular morbidity and mortality.

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RESEARCH ARTICLE

DENGUE FEVER IN CHILDREN: ROLE OF ULTRASONOGRAPHY.

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 Hepatomegaly, Splenomegaly,
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Abstract

Introduction: Dengue fever is one of the most important emerging vector-borne viral diseases. There are four serotypes of dengue viruses, each of which is capable of causing self-limited dengue fever or even life-threatening dengue haemorrhagic fever and dengue shock syndrome. The aim of this study was to evaluate the role of sonographic findings as early diagnostic modality in children with dengue fever.

Materials & Methods: This cross-sectional observational study was carried out in the department of pediatrics of a tertiary care hospital. A total of 88 patients who were clinically and serologically (NS1 antigen/ IgM/ IgG antibody) diagnosed as dengue positive were included in this study. These patients also underwent USG abdomen and chest within the first week of the illness.

Results: Out of the 88 confirmed dengue cases, 56 (63.6%) cases were males. All the cases had fever (100%). Other clinical symptoms were vomiting in 64.8%, abdominal pain in 62.5%, bodyache 53.4%, Shock in 28.4%, headache in 48.9% and rashes in 23.9%. The USG findings were hepatomegaly (39.8%), gall bladder thickening (53.4%), splenomegaly (20.5%), pleural effusion (38.6%) and ascites (30.7%). Overall 65 (73.8%) out of 88 cases had one of the positive finding in ultrasonography.

Conclusion: Ultrasonography can be used as a first-line imaging modality in all patients with suspected dengue fever to detect early signs suggestive of the disease.

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Introduction:-

Dengue fever is an acute febrile infectious disease in subtropical and tropical areas that is progressively making its way from being "one of the great neglected diseases of mankind" [1] towards being acknowledged as one of the world's major infectious diseases [2]. In the last 50 years, incidence of dengue has increased 30-fold with increasing geographic expansion to new countries and from urban to rural settings. An estimated 50 million dengue infections occur annually and approximately 2.5 billion people live in dengue endemic countries [3].

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The etiologic agents include all four serotypes which belong to the genus *Flavivirus* in the family *Flaviviridae*. The principle vector is mosquito, *Aedes Aegypti*, which breeds largely indoors in clean waters mainly in artificial water containers, and feeds on humans in daytime [4].

The clinical manifestations of dengue vary with the age and immunity of the patient. It can present as 1) In apparent infection 2) non-specific febrile illness, 3) classic dengue fever, 4) dengue hemorrhagic fever, 5) Dengue shock syndrome (DSS) and 6) encephalopathy and fulminant liver failure [5,6].

Guzman et al. studied to show that most cases present as classic dengue fever (DF) with high fever, retro-orbital pain, severe myalgia/arthralgia, and rash. However, in some cases, illness progresses to life-threatening dengue hemorrhagic fever/dengue shock syndrome (DHF/DSS), characterized by vascular leakage leading to hypovolemic shock and a case fatality rate up to 5% [7,8].

Hammond et al. [9] studied to show that preschool children and infants have rather more often nonspecific febrile illnesses while preadolescent children often develop fever and moreover younger children with DHF are known to experience more severe clinical outcome (e.g. higher case fatality ratio) than adults.

Early identification of patients at risk of developing severe dengue is critical to provide timely supportive care, which can reduce the risk of mortality to < 1% [10]. Distinguishing dengue from other febrile illnesses (OFIs) early in illness is challenging, since symptoms are non-specific and common to other febrile illnesses such as malaria, leptospirosis, rickettsiosis, and typhoid fever in dengue-endemic countries [11].

Positive serology (anti dengue antibody) is the mainstay in the diagnosis of DF. But serology takes approximately 7 days to give a positive result [12,13]. The diagnosis of DF is often delayed owing to time taken for availability of serology test results [14]. Ultrasonography (USG) is a cheap, rapid and widely available non-invasive imaging method [15,16]. Some authors concluded findings of hepatomegaly, pericholecystic edema, thickened gall bladder, ascites, pleural effusion (right sided/ both) and splenomegaly are early and significant markers of dengue, which can be confirmed by serology later on [17,18]. Several studies concluded that ultrasonography of the chest and abdomen can be an important adjunct to clinical profile in diagnosis DF and diagnosis can be made early in the course of disease compared with other modes of diagnosis [14]. It can be used as a first-line imaging modality in patients with suspected dengue fever to detect early signs suggestive of the disease prior to obtaining serologic confirmation test results, especially in a dengue fever epidemic area [17].

These findings may also occur in other viral infections and enteric fever but in other viral infections the historical profile, symptom complex evolution and physical findings do not mimic those of dengue fever [14]. Thus, the aim of present study was to evaluate the sonographic findings of dengue fever which may be useful as an early diagnostic tool.

Materials and Methods:-

Study Type & Area:-

A prospective observational study was conducted at Department of Paediatrics of a tertiary care centre during July 2014 to August 2016.

Inclusion Criteria:-

1. Patient between the age group of 0 – 18 years.
2. Patients with confirmed diagnosis of dengue fever (positive test for NS1 antigen/IgM dengue/IgG dengue).

Exclusion Criteria:-

1. Patient with serological test negative for dengue.
2. Patients having fever of more than 2 weeks duration.

Sample Size:-

Consecutive type of non-probability sampling was used for the selection of study subjects. A total of 88 diagnosed pediatric patients of Dengue fever admitted in our hospital were taken for study after informed consent from parents.

Study Methodology:-

All patients were admitted and treated as indoor patients. On admission, detailed history and complete physical examination findings were recorded. The vitals (temperature, pulse, respiration, blood pressure) of the patients were recorded on admission and thereafter till discharge. In all patients, complete blood count and serological tests for dengue (NS1 antigen/ IgM dengue/ IgG dengue) were carried out depending on duration of fever on presentation.

Following laboratory investigations were done in all patients on admission:-

1. Dengue NS1 antigen test/ IgM dengue antibody/ IgG dengue antibody- depending on duration of fever on presentation. All the three tests will be done by the ELISA (enzyme linked immunosorbant assay) technique.
2. Complete blood count (CBC) including platelet count.
3. Coagulation Profile
4. USG abdomen

Other investigations like liver function test, renal function test, S. electrolyte, X-ray chest/abdomen, blood culture, peripheral smear for malarial parasite were performed based on the clinical presentation.

Based on the investigation results, patients were classified into 3 categories as per WHO guidelines: Dengue fever, Dengue Hemorrhagic fever and Dengue shock syndrome. All patients will be treated as per WHO guidelines- Dengue guidelines for diagnosis, treatment, prevention and control [19].

Results:-

Out of the 88 confirmed dengue cases, 56 (63.6%) were males while 32 (36.4%) were females. Fever was present in all the cases, while other features were: vomiting (64.8%), abdominal pain (62.5%), bodyache (53.4%), shock (28.4%), headache (48.9%) and rashes (23.9%) (Table 1). Investigations revealed thrombocytopenia in 78.4%, hemoconcentration in 48.9%, raised liver enzymes in 68.2% while raised creatinine levels in 2.3% patients (Table 2). The ultrasonographic findings showed hepatomegaly in 35 (39.8%) cases, gall bladder thickening in 47 (53.4%) cases, splenomegaly in 18 (20.5%) cases, pleural effusion in 34 (38.6%) cases while ascites was seen in 27 (30.7%) cases (Table 3). Overall 65 (73.8%) out of 88 cases had one of the positive finding in ultrasonography. Final Diagnosis of Dengue was given in 15.9% cases while diagnosis of dengue with warning signs and severe dengue was given in 55.7% and 28.4% cases respectively (Table 4). Out of total 88 patients, 2 (2.27%) patients expired who were suffering from severe dengue.

Table 1:- Distribution of patients according to presenting complaints.

Presenting Complaints	N	%
Fever	88	100%
Headache	43	48.9%
Bodyache	47	53.4%
Rash	21	23.9%
Vomiting	57	64.8%
Abdominal Pain	55	62.5%
Shock	25	28.4%

Table 2:- Distribution of patients according to laboratory findings.

Lab Findings	N	%
Thrombocytopenia	69	78.4%
Hemoconcentration	43	48.9%
Raised Liver Enzymes	60	68.2%
S. Creatinine (> 1.2 mg%)	2	2.3%

Table 3:- Distribution of patients according to sonographic findings.

USG Findings	N	%
Hepatomegaly	35	39.8%
GB wall Thickening	47	53.4%
Splenomegaly	18	20.5%
Pleural Effusion	34	38.6%
Ascites	27	30.7%

Table 4:- Distribution of patients as per Final Diagnosis.

Diagnosis	N	%
Dengue	14	15.9%
Dengue with warning signs	49	55.7%
Severe Dengue	25	28.4%
Total	88	100.0%

Discussion:-

Dengue has become a major international public health concern in recent years [20]. It is emerging as one of the most important mosquito borne diseases in India. *Aedes aegypti* mosquitoes that transmit the disease breed in man-made containers such as tanks, pitchers, discarded containers etc. in which water has stagnated for over a week. Thus, the success of control measures have become a reflection of sanitation and hygienic practices achieved. The cases of dengue peak in the monsoon season in most parts of the country but have become perennial in the southern states and Gujarat [21,22].

Dengue has myriad clinical manifestations with unpredictable evolution and outcome. The disease typically begins with an acute febrile phase lasting 2-7 days and is accompanied by flushing, generalized body ache, myalgia, arthralgia and headache. Increased capillary permeability reflected by progressive increase in hematocrit heralds the beginning of critical phase at around 3-7 days of illness. Severe hemorrhagic manifestations and shock secondary to plasma leakage may occur at this stage. Leukopenia and declining platelet counts are also seen preceding this stage [23]. Serological diagnosis is confirmatory of dengue and includes direct methods such as virus isolation and NS1 antigen detection and indirect methods such as IgM and IgG antibody detection [24]. As serological diagnosis is not available in most of the places in a resource scarce country like India, sonography has an increasingly important role. Sonography is a readily available and cost-effective method for the diagnosis, which impacts the management of patients of dengue. The aim of present study was thus to evaluate the role of sonographic findings as early diagnostic modality in children with dengue fever.

In present study, male predominance was seen with 63.6% male cases to 36.4% females with male to female ratio of 1.74:1. In a study by Hema M et al. [25], the sex ratio of male to female was 1.32: 1, while in that of Srinivasa et al. [26], it was 1.13:1. The sex distribution is also consistent with previous study findings that dengue fever occurs more in male sex [22,24,27].

In present study, fever was present in all the cases, while other features were: vomiting (64.8%), abdominal pain (62.5%), bodyache(53.4%), shock (28.4%), headache (48.9%) and rashes (23.9%). In the study by Srinivas et al. [26], fever and vomiting were the most frequent symptoms. Similar observation was also seen in earlier studies [14,28,29]. Srinivasa also observed rashes in 10.5% cases while Ahmed FU et al [30] noted rashes in 12% of children.

We also observe thrombocytopenia in 78.4% cases while hemoconcentration and raised liver enzymes were observed in 48.9% and 68.2% cases respectively. In the study by Mittal Hema et al. [25], thrombocytopenia and hepatic enzyme derangement was present in 92.6% and 60% of the cases respectively. In the study by Narayanan M. et al. [31], serum SGPT levels were elevated in 59.7% of the total patients in whom the test was performed. Serum SGPT levels were elevated in 65% of DF, 63.6% of DHF and 66% of DSS patients. In the study by Aggarwal Anju et al. [32], serum SGPT levels were elevated in 57% of the patients in whom the test was performed.

The ultrasonographic findings showed hepatomegaly in 35(39.8%) cases, gall bladder thickening in 47(53.4%) cases, splenomegaly in 18 (20.5%) cases, pleural effusion in 34(38.6%) cases while ascites was seen in 27(30.7%) cases. Overall 65 (73.8%) out of 88 cases had one of the positive finding in ultrasonography. This clearly shows that Ultrasound can be used as an early, non-invasive and economical diagnostic tool.

In the study by srinivasa et al. [26], 72% of the cases had ultrasound findings in the form of either hepatomegaly or gall bladder wall thickening. The ultrasound findings showed gall bladder wall thickening in 30.5%. In a study by Quiroz-Moreno et al. [33] gallbladder thickening was seen in 86% of the patients, Sai et al. [14] observed in 56% patients while Thulkar et al. in 35.1% [34]. Setiawan et al. attempted to investigate whether gall bladder wall thickening measured by ultrasonography can be used to predict the onset of severe dengue fever. It is found that a

thickness of 5mm is useful as a criterion for identifying DHF patients at high risk of developing hypovolemic shock [35].

In our study pleural effusion or ascites was apparent on clinical examination in some of the cases. But sonography helped in diagnosing all of them. Similarly 15 cases of hepatomegaly and 11 cases of splenomegaly could not be diagnosed by clinical examination but were diagnosed correctly by ultrasonography. So this study clearly demonstrates the importance of ultrasonography in the accurate and complete clinical evaluation of dengue fever. All the cases which went into hypotension and shock showed gall bladder wall thickening, with moderate to severe pleural effusion and ascites.

Conclusion:-

Abdominal USG should be used as a first-line imaging modality in all patients with suspected dengue fever to detect early signs suggestive of the disease along with obtaining serologic confirmation tests.

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RESEARCH ARTICLE

EVALUATION OF STRAWBERRY CULTIVARS FOR GROWTH AND YIELD CHARACTERISTICS IN SUB TROPICAL REGION OF PUNJAB.

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Key words:-

Strawberry, Chandler, Selva, yield,
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Abstract

A research trial was carried out in the experimental field of Department of Horticulture, Khalsa College, Amritsar during the year 2015-2016 to evaluate some strawberry cultivars in sub tropical region of Punjab. The runners of five strawberry cultivars i.e.. Chandler, Tioga, Fern, Selva and Blackmore were planted at 30x40 cm apart on the raised beds at three planting times of mid October, end October and mid of November respectively. The design of experiment was Randomised block design (RBD) with factorial arrangement. The results of the study indicated that out of the cultivars tried, the strawberry cv. Chandler proved to be the best in producing maximum number of shoots, leaves, runners and flowers along with yield parameters while strawberry cv. Selva was the least among all these parameters. Out of the times of planting, middle of October proved to be the best in all the parameters.

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Introduction:-

The modern cultivated strawberry (*Fragaria x ananassa* Duch.) is a hybrid of two largely dioecious, octoploid species, *Fragaria chelonensis* Duch and *Fragaria virginiana* Duch. Basically, it is herbaceous perennial and short day plant grows predominantly in the temperate climate. Strawberry is known for its pleasant aroma. It is amongst the few crops, which gives quick and very high returns per unit area on the capital investment, as the crop is ready for harvesting within six months of planting (Bakshi *et al.* 2014). It is a crop cherished by the growers of sub-tropical and temperate regions of the world due to its low cost of growing and high returns per unit area. It is widely, adapted in geographically diverse areas in world (Biswas *et al.* 2009). It can also be cultivated successfully under subtropical climate of Punjab (Dhillon, 2005). It is cherished in gardens and commercial fields with glamorous red, delicious with a unique shape, highly perishable fruit with a pleasant flavor. It is refreshing, nutritious soft fruit having a distinct tantalizing aroma (Sharma and Yamdagni, 2000). It is highly nutritious having a rich source of vitamins A, B, C and niacin, minerals like phosphorus, potassium, calcium and iron (Karkara and Dwivedi, 2002). It is utilized for the production of purees, juice concentrate, jams, preserves and rose red wine. Medicinally, strawberries have been known to kill certain viruses like polio, herpes and these may block the formation of nitrosamines, which can cause cancer, furthermore these contain relatively high quantities of ellagic acid, which has a wide range of biological activity only recently discovered (Rieger, 2006). The wide diversity of strawberry plant gives an idea of its potential for selection work. Choice of cultivars is of paramount importance for successful strawberry cultivation (Asrey and Singh, 2004; Ahsan *et al.* 2014). Selection of new stable strawberry cultivars for field condition can ensure better yield and quality. Considering above facts the present study work was undertaken with a view to determine the performance of different strawberry cultivars.

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Materials and Methods:-

The present experiment was conducted in the nursery of Horticulture Department, Khalsa College, Amritsar during 2014-15. The runners of strawberry cv. Chandler, Tioga, fern, selva and Blackmore were planted. A unit of 10 runners comprising a treatment was planted in each plot, 30 cm apart on the raised ridges at three times of planting i.e. mid of October, end of October and mid of November taking care that the crown of the runners lies just at the surface of soil. The experiment was laid out according to randomized block design with factorial arrangement with three replications. The observations were recorded on vegetative, floral characteristics and yield parameters. The pooled data of both the years were analysed statistically.

Results and Discussion:-

Maximum shoot number plant⁻¹ (7.94 during first and 8.34 during second season) was counted in strawberry planted on end of Oct (T₂) during both the cropping seasons, it was followed by mid of Nov (T₃) and mid of Oct (T₁) in the first season which recorded 7.51 and 5.67 shoot number respectively and mid Oct (T₁) and (T₃) times in the second cropping season with 7.87 and 5.56 shoots respectively. The differences were found to be statistically significant. Maximum shoot number plant⁻¹ (8.05) was observed in cv. Chandler during first cropping season, it was followed by cvs. Tioga (7.95) and Blackmore (7.52) while the cvs. Fern and Selva recorded 5.95 and 5.73 shoot number respectively. In the second cropping season of study maximum shoot number plant⁻¹ (10.35 each) was counted in cvs. Chandler. It was followed by cvs Tioga, Blackmore and Fern with 8.10, 7.77 and 6.58 shoot number plant⁻¹ which was found to be at par with the former two cultivars and minimum shoot number per plant to the tune of 6.01 was recorded in cv. Selva. The differences were found to be statistically significant. The interaction between the time of planting and type of cultivars were also found to be statistically significant during both the years of study. In the first year maximum shoot number plant⁻¹ (9.11) was counted in cv. Blackmore planted during end of Oct (T₂) in the cropping season it was 10.00 in cv. Chandler planted on mid of Oct (T₁) and minimum (5.09) in cv. Fern planted on mid Nov (T₃). The varying shoot number may be due to the temperature variation, changing weather conditions which affects growth of plants. The variation in shoot number in different varieties may be due to the particular varietal character and their growth habits.

The time of planting and type of cultivars exerted significant effect with respect to the number of leaves per plant. Maximum leaf number plant⁻¹ (19.71 during first and 21.02 during second cropping season) was recorded in planting time of mid Oct (T₁), it was followed by end Oct (T₂) and mid Nov (T₃) planting time which recorded 19.46 and 18.69 leaf number during first season, 19.52 and 18.63 leaf number per plant respectively during the second cropping season of study. In the first year the differences during all the three times were found to be statistically at par with each other while in the second year of study the time T₃ showed statistically non significant differences. Maximum number of leaves (23.78 in first and 24.15 in second cropping season) were counted in cv. Chandler followed by cv Tioga (23.62 in first season and 23.84 in second cropping season) and cv. Blackmore (23.01 in first and 23.38 in second cropping season), while minimum leaf number (10.11 in first and 10.45 in second cropping season) was recorded in cv. Fern during both the cropping seasons. The differences were found to be statistically significant. The interaction between cultivars and the planting time were found to be statistically significant with regard to the leaf number plant⁻¹ during both the cropping season of investigation. Maximum leaf number plant⁻¹ (24.75 during first and 25.92 during second cropping season) was recorded in cv. Chandler planted on mid Oct (T₁) as against minimum (9.00 during first and 8.20 during second cropping season) in cv. Fern planted on end Oct (T₂) during both the cropping seasons.

The maximum leaf area 94.17 cm² during first cropping season was measured in runners planted on end Oct T₂ but during second cropping season it was 93.51 cm² in T₁. Leaf area produced by T₂ planting time remained at par with its following time of planting during both the cropping seasons and the differences were found to be statistically significant. Different cultivars also found effective in altering leaf area in both the cropping seasons of study, cv. Blackmore produced maximum leaf area (110.97 cm² during first and 110.35 cm² during second cropping season), it was followed by cv. Chandler with 107.86 cm² and 107.38 cm² leaf area in the first and second cropping seasons. Minimum leaf area was obtained in cv. Selva during both the years of study. The differences were found to be statistically significant during both the cropping seasons of study. The interactions between planting time and strawberry cultivars exhibited significant effects in both the years of investigation where the maximum leaf area 112.25 cm² and 110.38 cm² were produced by cv. Blackmore in end Oct (T₂) planting time. Variation in leaf area among different strawberry cultivars could be attributed to the fact that different progenies may have reacted differently to photoperiod, light, temperature and status of media (Darrow, 1966 and Tanaka and

Mizuta,1974).Hence, it seems that a negative correlation between these two character variable between these two variables coexists. According to Dhillon (2005) plant condition or environment or both affect these two variables differentially or independently.

In the first cropping season runners of T_1 planting took 177.0 days for runner formation which is significantly higher than the T_2 and T_3 plantings which took 164.0 and 155.0 days for runner formation. During the second cropping season significantly more (176.0) days were taken by runners of T_1 planting than T_2 and T_3 plantings which registered 164.0 and 156.0 days for runner formation. During both the cropping seasons cv. Chandler took minimum days (154.0 days during first and 152.0 days during second cropping season) for runner formation followed in line by cvs. Blackmore, Fern, Selva and Tioga which registered 162.0, 165.67, 170.0 and 175.0 days during first cropping season and 159.0, 164.4, 172.0 and 180.0 days for runner formation respectively. The interaction between time and cvs. were found to be non- significant statistically. The vast differences in the time of runner formation, runners plant⁻¹ in the various cultivars may be ascribed either a varietal character or genetic make up of the plants or both which affect the plants to react differently under the same agro-climatic conditions. The temperature of the site also affected the runner formation. These results are in line with the findings of Dhillon 2005 respectively.

Time of planting influenced number of runners emerged per plant significantly. Maximum number of runners (4.92) were counted in T_1 planting followed by T_2 and T_3 plantings which gave 4.51 and 3.33 runners plant⁻¹ respectively in the first cropping season and in the consecutive season, T_1 was also able to produce maximum (5.36) runners followed by T_2 and T_3 timings with 4.72 and 3.52 number of runners plant⁻¹. The difference during both the cropping seasons were found to be statistically significant. All the cultivars showed a great variation with respect to number of runners plant⁻¹ among each other. Maximum number of runners plant⁻¹ (6.25) were obtained in cv. Chandler, it was closely followed by the cvs. Blackmore and Fern during the first cropping season which gave 6.03 and 4.26 number of runners plant⁻¹ while cv. Tioga registered minimum (1.30) runners plant⁻¹. During the second season cv. Chandler produced 6.80 runners plant⁻¹ followed by cvs. Blackmore, Fern and Selva with 6.22, 4.42 and 3.73 runners plant⁻¹ while cv. Tioga registered minimum runners. Similar results have been reported by Athwal (1976), Singh (1979) and Singh (1982) under Punjab conditions which further confirms the results of the present study. The interaction of planting time and different cultivars were found to be statistically non- significant .

The data with regard to the number of days taken for flower initiation depicted that during first cropping season minimum average days taken for flower initiation were in the planting of T_3 (121.54 days) while the planting of T_2 and T_1 times taken little more time i.e. 122.14 and 122.61 days for flower initiation, but the differences were found to be statistically non-significant. During the second cropping seasons, time of planting T_3 also recorded minimum 121.73 days taken for flower initiation followed by T_2 and T_1 planting times which took 122.55 and 122.83 days. During both cropping season the difference were found to be statistically significant. It is obvious from the data that different cvs. showed appreciable differences in respect of number of days taken for flower initiation and the differences were found to be statistically significant during both the cropping season of study. In the first cropping season cv. Fern took minimum (120.53) number of days for flower initiation followed in ascending order by cvs. Blackmore, Tioga, Chandler and Selva with 121.34, 121.55, 121.79 and 125.31 days respectively. In the second cropping season again cv. Fern took minimum (120.68) days for flower initiation followed in ascending order in cvs. Tioga, Chandler, Blackmore and Selva with 121.41, 122.02, 122.15 and 125.61 days respectively. It was noted that during both the cropping seasons cv. Selva took maximum number of days flower initiation. The interaction between time of planting and cultivars was found to be non-significant statistically during both the cropping seasons.

The saplings planted during mid Oct (T_1) time took maximum days for flowering (118.88 days during first and 115.81 days during second cropping season) followed by end Oct times of planting with 97.16 days during first, 98.51 days during second cropping season while the time of planting T_3 registered 82.19 days during first, 87.22 days during second cropping season. The differences were found to be statistically significant during both the cropping seasons. In the first cropping season cv. Selva remained in flowering for 105.75 days followed by cv. Fern and Blackmore with 103.29 and 100.54 days of flower duration respectively and these three remained at par with each other while the cv. Tioga and Chandler recorded minimum 93.93 and 93.55 days duration of flowering. In the second year of study cv. Selva remained successive with having 103.81 days of flower duration and it was followed by cv. Fern and Blackmore with 102.51 and 102.41 days duration in flowering and likewise the former season of study these were also remained at par with each other and minimum duration of flowering was noted in cvs. Tioga and Chandler with 100.26 and 93.57 days respectively. The results of the present study are collaborated by the findings of Dhiman (2003). However, Dhaliwal and Singh (1983) while working on strawberry in Ludhiana

conditions, Beniwal *et al.* (1989) and Kidmos *et al.* (1996) in Denmark conditions recorded similar type of observations which confirms the findings of present study. The interaction between planting time and cultivars were found to be non-significant statistically during both the years of study.

The maximum number of flowers were registered during the mid Oct (T_1) planting time which was followed by end Oct (T_2) and mid Nov (T_3) during both the years. During the first cropping seasons planting time T_1 , T_2 and T_3 recorded 13.37, 13.27 and 13.15 number of flowers plant⁻¹ respectively and during the second cropping season the times of planting T_1 , T_2 and T_3 registered 13.41, 13.25 and 13.22 flower plant⁻¹ respectively, but the differences only in the first cropping season were found to be statistically non-significant. The kind of cultivar showed statistically significant differences with regard to number of flowers plant⁻¹ during both the cropping seasons. The cvs. Chandler, Blackmore, Tioga, Fern and Selva recorded 15.22, 15.14, 13.68, 11.18 and 11.08 number of flowers plant⁻¹ during the first and during the second cropping season, these were 15.35, 15.05, 13.73, 11.16 and 11.17 respectively, same pattern in the orderliness was noted with regard to the variation in the cultivars during both the cropping seasons. The interactions between time of planting and type of cultivars were found to be non-significant statistically during both the cropping seasons.

During both the seasons T_1 time of planting registered maximum number of fruit set plant⁻¹ (11.21 during first and 11.18 during second cropping season) followed by T_2 (10.79 during first and 10.83) during second cropping season and T_3 (10.53 during first and 10.56 during second cropping season). Significant differences were observed on number of fruit set plant⁻¹ due to the type of cultivars. During the cropping season number of fruit set plant⁻¹ were maximum in cv. Chandler followed by cvs. Black more and Tioga with 13.41, 12.43 and 10.31 number of fruit set plant⁻¹ while the minimum (8.73) number was recorded in cv. Selva, Same pattern of number of fruit set plant⁻¹ was observed during the second cropping season where the cvs. Chandler, Blackmore, Tioga, Fern and Selva registered 13.47, 12.40, 10.35, 9.31 and 8.74 of fruit set plant⁻¹. It was noted that the fruit set per cent was reduced with the planting time as the early planted saplings received the favourable climatic conditions but the saplings planted in the mid Nov faced the low chilling temperature rising temperature at the fruit set time which resulted in the complete necrosis of the developed fruit or the flower. Per cent fruit set of strawberry depends upon the agro-climatic field conditions of the place of there cultivation. Different research workers reported variability in the per cent fruit set of various varieties depending upon agro-climatic conditions of the area of cultivation. Singh (1982) reported maximum per cent fruit set of Pusa Early Dwarf followed by cv. Tioga under Ludhiana agro climatic conditions of Punjab. The variability in the per cent fruit set under Amritsar's prevailing agro-climatic conditions may be due to the effect of temperature and humidity of the area. The interaction between time of planting and type of cultivars was found to be statistically non significant.

The early plantation resulted in maximum yield then it showed descending trend with the late plantations during both the cropping seasons. In the first cropping season planting times T_1 , T_2 and T_3 registered 115.71, 104.32 and 83.34 g yield plant⁻¹ respectively and in the second cropping season it was 117.98, 103.35 and 81.05 g plant⁻¹ respectively and differences were found to be significant statistically during both the cropping seasons. It is also evident from the data that statistically significant differences were observed in the yield plant⁻¹ of different cultivar. The maximum yield plant⁻¹ to the tune of 137.15 g on average basis was obtained in cv. Chandler, cv. Blackmore with a yield of 116.47 g plant⁻¹ stood runner up, followed in line were cvs. Tioga, Fern and Selva with 109.18, 86.91 and 55.91 g plant⁻¹ yield respectively while during the first cropping season. During the second cropping season cv. Chandler registered 134.94 g plant⁻¹ yield followed by cvs. Blackmore, Tioga, Fern and Selva with 117.00, 106.25, 85.65 and 60.22 g plant⁻¹ fruit yield. Sharma and Yamdagni (2000) also reported cv. Chandler as high yielding cultivar amongst other cultivars and mentioned variability in the yield and marketable yield of strawberry fruits of different cultivars. The yield plant⁻¹ also decreased with the planting time. Similar observations on yield plant⁻¹ have been reported in different cvs. of strawberry by Singh (1982) and Sharma and Badiyala (1980). These results are in accordance with the findings of several research workers (Bedarad *et al.*, 1971; Lal and Seth, 1980; Hancock *et al.*, 1983). They found a strong correlation with fruit number per plant and weight with total fruit yield but in contrast Dhaliwal and Singh (1983) reported that higher yield of strawberry cultivars was due to number of fruits per plant which may be smaller and lighter in size. The interaction between cvs. and the planting time were found to be non significant during both the seasons of study.

The yield attributes of marketable yield were influenced significantly with the advancement in time of plantation during both the cropping seasons. The time of planting T_1 , T_2 and T_3 yielded 114.00, 103.18 and 81.26 g plant⁻¹ marketable yield respectively in the first cropping season and 117.33, 102.69 and 80.72 g plant⁻¹ marketable yield

during the second cropping season and differences were found to be statistically significant during both the seasons of cropping. The type of cultivars also showed their effect on the marketable yield of strawberry fruits during both the cropping seasons. The cv. Chandler yielded maximum marketable fruits to the tune of 135.71g plant⁻¹ and 133.31g plant⁻¹ during first and second cropping seasons. The cvs. Blackmore, Tioga, Fern and Selva followed the cv. Chandler by registering 114.65, 106.26, 83.46 and 57.31 g plant⁻¹ (minimum) marketable yield respectively during the first cropping season and in the second season these were 116.47, 106.68, 85.59 and 59.19 g plant⁻¹ (minimum) marketable yield respectively and the differences were found to be statistically significant. The interaction between the time of planting and different type of cultivars were found to be statistically non-significant during both the cropping seasons.

Table 1: Effect of planting time and cultivars on shoot number plant⁻¹ of strawberry.

Cultivar	2014-15				2015-16			
	T ₁	T ₂	T ₃	Mean	T ₁	T ₂	T ₃	Mean
Chandler	7.03	8.10	9.02	8.05	10.00	12.50	8.55	10.35
Tioga	6.75	9.00	8.12	7.95	8.03	8.07	8.21	8.10
Fern	5.48	6.33	6.05	5.95	7.22	7.44	5.09	6.58
Selva	4.00	7.19	6.00	5.73	5.11	5.59	7.34	6.01
Blackmore	5.12	9.11	8.34	7.52	9.00	8.11	6.22	7.77
Mean	5.67	7.94	7.51		7.87	8.34	5.56	

LSD (P=0.05)

Time	1.05	1.92
Cvs.	1.35	1.48
Time x Cvs.	1.12	0.23

Table 2: Effect of planting time and cultivars on leaf number plant⁻¹ of strawberry.

Cultivar	2014-15				2015-16			
	T ₁	T ₂	T ₃	Mean	T ₁	T ₂	T ₃	Mean
Chandler	24.75	24.10	22.50	23.78	25.92	23.47	23.07	24.15
Tioga	24.28	24.25	22.34	23.62	25.35	25.17	21.01	23.84
Fern	10.36	9.00	10.96	10.11	12.34	8.20	10.82	10.45
Selva	15.10	17.13	19.35	17.25	16.29	17.08	17.02	16.80
Blackmore	24.06	24.10	20.88	23.01	25.20	23.70	21.25	23.38
Mean	19.71	19.46	18.69		21.02	19.52	18.63	

LSD (P=0.05)

Time	1.72	1.48
Cvs.	1.85	1.91
Time x Cvs.	0.56	0.12

Table 3: Effect of planting time and cultivars on leaf area (cm)² of strawberry.

Cultivar	2014-15				2015-16			
	T ₁	T ₂	T ₃	Mean	T ₁	T ₂	T ₃	Mean
Chandler	107.25	109.00	107.34	107.86	108.34	106.45	107.36	107.38
Tioga	80.98	83.16	80.26	81.46	80.36	82.94	80.97	81.42
Fern	86.27	86.23	86.03	86.17	86.97	86.00	86.14	86.37
Selva	80.96	80.22	81.30	80.83	81.75	80.10	81.35	81.06
Blackmore	110.12	112.25	110.54	110.97	110.11	110.57	110.38	110.35
Mean	93.12	94.17	93.09		93.51	93.21	93.24	

LSD (P=0.05)

Time	1.67	1.45
Cvs.	1.32	1.87
Time x Cvs.	0.58	0.21

Table 4: Effect of planting time and cultivars on time taken for runner formation (days) of strawberry

Cultivar	2014-15				2015-16			
	T ₁	T ₂	T ₃	Mean	T ₁	T ₂	T ₃	Mean
Chandler	166.00	155.00	140.00	154.00	160.00	153.00	142.00	152.00
Tioga	185.00	173.00	166.00	175.00	190.00	179.00	170.00	180.00
Fern	178.00	160.00	159.00	165.67	175.00	158.00	157.00	164.40
Selva	183.00	170.00	158.00	170.00	184.00	172.00	160.00	172.00
Blackmore	173.00	160.00	152.00	162.00	170.00	157.00	149.00	159.00
Mean	177.00	164.00	155.00		176.00	164.00	156.00	

LSD (P=0.05)

Time 10.2 9.56

Cvs. 12.4 10.3

Time x Cvs. NS

NS

Table 5: Effect of planting time and cultivars on number of runners plant⁻¹ of strawberry

Cultivar	2014-15				2015-16			
	T ₁	T ₂	T ₃	Mean	T ₁	T ₂	T ₃	Mean
Chandler	6.81	6.35	5.59	6.25	7.55	6.95	5.90	6.80
Tioga	1.35	1.35	0.25	1.30	2.55	1.25	0.75	1.52
Fern	4.35	4.35	3.25	4.26	5.35	4.45	3.45	4.42
Selva	3.60	3.60	2.95	2.42	4.20	4.00	3.00	3.73
Blackmore	6.90	6.90	4.25	6.03	7.20	6.95	4.50	6.22
Mean	4.51	4.51	3.33		5.36	4.72	3.52	

LSD (P=0.05)

Time 0.68 0.64

Cvs. 0.34 0.78

Time x Cvs. NS NS

Table 6: Effect of planting time and cultivars on days taken for flower initiation of strawberry

Cultivar	2014-15				2015-16			
	T ₁	T ₂	T ₃	Mean	T ₁	T ₂	T ₃	Mean
Chandler	123.64	121.91	119.81	121.79	122.95	122.16	120.95	122.02
Tioga	120.91	122.22	121.53	121.55	121.17	122.63	120.42	121.41
Fern	121.24	120.10	120.25	120.53	121.58	120.25	120.21	120.68
Selva	125.76	125.21	124.97	125.31	126.17	125.76	124.89	125.61
Blackmore	121.52	121.24	121.22	121.34	122.32	121.94	122.19	122.15
Mean	122.61	122.14	121.54		122.83	122.55	121.73	

Table 7: Effect of planting time and cultivars on duration of flowering (days) of strawberry

Cultivar	2014-15				2015-16			
	T ₁	T ₂	T ₃	Mean	T ₁	T ₂	T ₃	Mean
Chandler	113.11	87.49	80.04	93.55	109.61	91.11	80.00	93.57
Tioga	113.12	95.66	73.00	93.93	114.35	103.22	83.23	100.26
Fern	120.23	105.21	84.44	103.29	114.09	101.05	92.38	102.51
Selva	134.42	101.28	81.56	105.75	114.33	101.06	96.05	103.81
Blackmore	113.56	96.16	91.89	100.54	126.67	96.10	84.46	102.41
Mean	118.88	97.16	82.19		115.81	98.51	87.22	

LSD (P=0.05)

Time 8.20 7.71

Cvs. 11.2 7.95

Time x Cvs. NS

NS

Table 8: Effect of planting time and cultivars on number of flowers plant⁻¹ of strawberry

Cultivar	2014-15				2015-16			
	T ₁	T ₂	T ₃	Mean	T ₁	T ₂	T ₃	Mean
Chandler	15.32	15.25	15.10	15.22	15.56	15.31	15.19	15.35
Tioga	13.86	13.75	13.44	13.68	13.89	13.68	13.61	13.73
Fern	11.27	11.16	11.12	11.18	11.21	11.19	11.09	11.16
Selva	11.18	11.07	11.00	11.08	11.27	11.000	11.25	11.17
Blackmore	15.22	15.12	15.10	15.14	15.11	15.05	14.98	15.05
Mean	13.37	13.27	13.15		13.41	13.25	13.22	

LSD (P=0.05)

Time NS NS
 Cvs. 1.45 0.95
 Time x Cvs. NS NS

Table 9: Effect of planting time and cultivars on number of fruit set of strawberry

Cultivar	2014-15				2015-16			
	T ₁	T ₂	T ₃	Mean	T ₁	T ₂	T ₃	Mean
Chandler	13.89	13.23	13.12	13.41	13.92	13.34	13.15	13.47
Tioga	10.56	10.22	10.16	10.31	10.58	10.25	10.22	10.35
Fern	9.67	9.22	9.11	9.33	9.56	9.28	9.10	9.31
Selva	9.02	8.95	8.23	8.73	9.12	8.82	8.28	8.74
Blackmore	12.93	12.34	12.01	12.43	12.72	12.45	12.04	12.40
Mean	11.21	10.79	10.53		11.18	10.83	10.56	

LSD (P=0.05)

Time NS NS
 Cvs. 0.87 1.05
 NS NS
 Time x Cvs. NS Cvs.

Table 10: Effect of planting time and cultivars on fruit yield plant⁻¹ (g) of strawberry

Cultivar	2014-15				2015-16			
	T ₁	T ₂	T ₃	Mean	T ₁	T ₂	T ₃	Mean
Chandler	141.19	140.52	129.73	137.15	143.35	137.92	123.56	134.94
Tioga	120.47	116.31	90.76	109.18	118.54	114.23	86.00	106.25
Fern	109.26	90.70	60.78	86.91	112.63	85.97	58.35	85.65
Selva	77.74	50.54	39.44	55.91	84.32	53.34	43.00	60.22
Blackmore	129.89	123.52	96.00	116.47	131.10	125.32	94.34	117.00
Mean	115.71	104.32	83.34		117.98	103.35	81.05	

LSD (P=0.05)

Time 11.7 15.1
 Cvs. 15.2 15.7
 Time x Cvs. NS NS

Table 11: Effect of planting time and cultivars on marketable yield plant⁻¹ (g) of strawberry

Cultivar	2014-15				2015-16			
	T ₁	T ₂	T ₃	Mean	T ₁	T ₂	T ₃	Mean
Chandler	140.00	138.21	128.93	135.71	141.47	136.93	121.54	133.31
Tioga	119.45	114.11	85.22	106.26	118.08	114.00	87.96	106.68
Fern	108.36	85.82	56.21	83.46	113.59	85.21	57.97	85.59
Selva	75.98	53.00	42.95	57.31	83.16	53.22	41.18	59.19
Blackmore	126.24	124.76	92.97	114.65	130.34	124.13	94.94	116.47
Mean	114.00	103.18	81.26		117.33	102.69	80.72	

LSD (P=0.05)

Time 12.4 12.1

Cvs.	16.0	15.7
Time x Cvs.	NS	NS

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RESEARCH ARTICLE

CONGENITAL ANOMALIES OF THE KIDNEY AND URINARY TRACT NEOPLASMS AND IN THE ELDERLY.

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Key words:-

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Abstract

Introduction: Congenital abnormalities of the kidneys and urinary tract represented a spectrum of malformations often grouped under the acronym for CAKUT. Normal development of the kidneys and urinary tract is the result of the precise interaction, spatially and temporally coordinated, between two structures, the metanephric mesenchyme (which originates from the nephrogenic cord) and the ureteric bud (which originates from nephric duct or Wolff). Any alteration, it's genetic, environmental, or stochastic, which influence this process can result in kidney malformations and streets urinariedel present study is to evaluate in relation to clinical attention the incidence of risk in neoplastic 'association cancer and malformation. in adults

Materials end Methods: Male patient of 75 suffering from gross hematuria and hernia inquinal sn accompanied by dysuria symptoms, in heavy smoker instrumental ultrasound examination of the urinary tract showed a solid lesion in the posterolateral wall sn aggettante in the bladder lumen The uro-CT examination showed a " abnormality of the urinary tract development with ectopia crusade and merger of the two kidneys located alongside sn (fot, 3.4) of the renal pelvis are addressed before the district complete with double ureteral sn .In at the urethral meatus will highlight a focal thickening protruding into the lumen wall to finely irregular margins, with enhancement after contrast medium. (photo 5.6) the CT examination conducted in the chest (photo 7) confirmed the marked signs of emphysema, the anterior segment of the left upper lobe, in close relationship with bronchus and vessels segmented solid training in margins lobed (29x20 mm axial diameter approximately). At PET-CT accumulation of the tracer in the upper field of the left lung in the paramedian attributable to non-specific reactive process .Fig (8.9), then he performed genetic test that confirmed mutations in the PAX2

Results: The UroCT is the imaging examination with greater sensitivity and specificity. The limit lies in the poor ability to detect flat lesions non-invasive muscle. The urography has been almost entirely supplanted by UroTC. The current biomarkers do not have a sensitivity and specificity greater than urinary cytology, and The aromatic amines, NAT2 and NAT1 of tobacco smoking products, are considered the

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main cause of the vesicula flexible cystoscopy cancer (Narrow Band Imaging) is being promoted as first-invasive test to be practiced in the patient suspected of having a bladder tumor. Uro-RM, similarly to what happens with the urography and CT, it is possible the study of the urinary tract, in order to seek sincrone. l'associazione urothelial lesions and tumors in malformations has prompted our research towards an evaluation genetics through genetic testing the result of which was the presence of a mutation in the PAX2,

Discussion: Renal abnormalities, according to the classification of Ridson, are divided into: number of anomalies, seat of anomalies, anomalies of form, abnormalities of differentiation, The presence of a double district pyelo-ureter. Observed in the patient was incomplete, with the two ureters who join before leading into the bladder through a common orifice, Figure 3 also the duplicity era asymptomatic, and was accompanied by a fusion anomaly: a horseshoe kidney. It was also present malrotation renaleuna genetic evaluation by performing genetic tests given to the search for possible correlation between cancer and malformation with oncological risk assessment identifies a heterogeneous set of tests designed to identify changes in the DNA sequences of the germline or of the products that are derived directly from the modification of transmissible genomic sequences .. the geographical correlation allows altresì to compare health risk levels allows you to answer the question whether there is a relationship between risk of death or birth defects as evidenced by numerous studies there is a risk increased cancer and malformations in the presence of high socioeconomic deprivation With increasing environmental pressure has been a steady trend in the health risk

Conclusions: Genetic testing can predict with great accuracy the future development of the health of a single individual in the presence of defects is necessary to provide genetic testing for the purpose of assessing the health of the individual. You can assume the major socio-ethical-legal implications for presymptomatic testing, the only genetic tests that can predict with certainty and in the future occurrence of a disease tests for clinical purposes provide information on the health status of subject. Those carried out for research purposes usually generate information that is not indicative of health status or genetic risks of the subject, but it is useful for the purposes of statistical or scientific research:

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..... Introduction:-

Congenital abnormalities of the kidneys and urinary tract represented a spectrum of malformations often grouped under the acronym for CAKUT (Congenital Anomalies of the Kidney and Urinary Tract). These malformations can affect one or more urinary structures (kidney, ureter, bladder) and can occur in isolation or in various combinations in the same family or even in the same individual, the CAKUT includes alterations of the renal parenchyma, such as agenesis kidney or hypodysplasia (RHD), and ureteriche abnormalities, such as obstructive stenosis of the joint ureteropelvic junction (UPJO), duplication of the excretory system (DCS), the megaureter congenital stenosis of the joint uretero vaginal bladder (UVJO) and reflux ureteral vesicles (VUR) (1, 2,3,4). This anatomical classification is often not informative .The malformations of the urogenital tract are present in more than 20% of birth defects [5] and can be observed in greater than 1% of live births (6,7). The application of routine ultrasonography during pregnancy has led to an increase of CAKUT diagnosis in pre-natal period. when it occurs at birth or within the first year of life [8]. Children who survive have a mortality rate 30 times higher than children of the same age. Normal development of the kidneys and urinary tract is the result of the precise interaction, spatially and temporally coordinated, between two structures, the metanephric mesenchyme (which originates from the nephrogenic cord) and the ureteric bud (which originates from nefrico duct or Wolff). . Any alteration, it's genetic, environmental, or stochastic, which influence this process can result in kidney and urinary tract malformations. In family forms of

CAKUT, accounting for about 20-30% of cases, the disease segregates in an autosomal dominant form in this case the genetic model that agrees more with the epidemiology is attributable to mutations that give loss-of-function or isomorphic to alleles that produce moderate malformations without significant an impact reproductive fitness. In point monogenic forms an important role in the pathogenesis mainly due to malformations of the kidneys and secondary urianarie ways to mutations in the PAX2, TCF2, SALL1, WT1, SIX1, EYA1 and other genes [9]. These diseases are frequently diagnosed by extra-renal manifestations, which led to the re-evaluation of extra-renal disease of patients with these mutations, resulting in an improvement of the clinical treatment. The degree of clinical variability can in fact be partially explained by the interaction at the expense of three or more of the BBS loci. In fact, it is well recognized that individuals carrying the PAX2 gene mutations may have different phenotypes. This phenotypic variability is due to the fact that all of the urinary components derived from two embryological tissue, the metanephric mesenchyme and the ureteric bud, As a consequence, individual defects in each of these compartments may have a pleiotropic effect on the development of the entire section urinary. The determination of PAX2 [10]. it is also very important for the high risk of the association observed in 20% of patients with congenital anomalies of the kidney and urinary tract diseases and the neurocognitive development, recognition in utero of congenital malformations of the kidneys and urinary tract, give kidney and urinary tract the sentinel role for an early diagnosis can optimize and customize treatment strategies in children with risk of development of other disorders also involving the central nervous system. Finally. Other abnormalities include unilateral or bilateral renal malposition, duplication of the pelvis or ureters (double kidney district) and bladder cancers aim of this study is to evaluate in relation to clinical attention the incidence of risk in neoplastic 'association cancer and malformation . in adult.

Materials and Methods:-

December 2013 to January 2017 at the II Clinical Surgery and digestive surgery department of general surgery and specialized company Polyclinic II University of Catania presents to our patient observation CS male of 75 suffering from gross hematuria and hernia inquinaal sn accompanied by symptoms dysuria, in heavy smoker .The patient was subjected to study and instrumental diagnostic ultrasound of the urinary tract showed a solid lesion in the posterolateral wall sn contained in the bladder lumen size of about 19mm. (Fig 1 2)

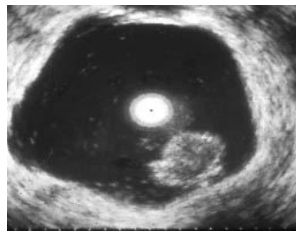


Fig 1:- bladder neoplasm



Fig 2:- side elevation

The uro-CT scan showed an "abnormality of the urinary tract development with ectopia crusade and merger of the two kidneys located alongside sn (fot, 3.4) of the renal pelvis are addressed before the district complete with double ureteral



Figure 3:- CT double urinary District



Fig 4:- CT fusion localized kidney to sn

sn .In at the urethral meatus will highlight a focal thickening of the wall jutting into the lumen a finely irregular margins, with enhancement after contrast medium. (Photo 5.6)

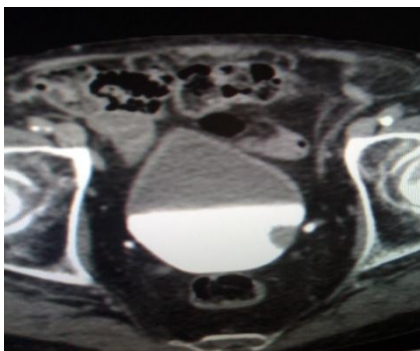


Fig 5:- bladder cancer CT

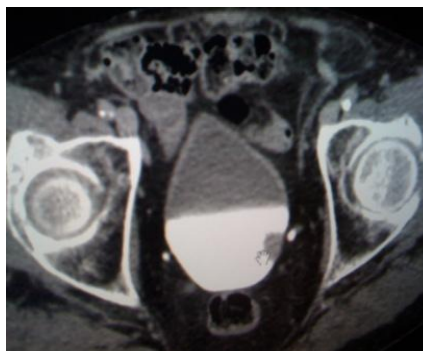


Fig 6 :- CT side wall neoplasia sn

The CT examination conducted in the chest (photo 7) confirmed the marked signs of emphysema, the anterior segment of the left upper lobe, in close relationship with the bronchus and vessels segmented solid training in margins lobed (axial diameters 29x20 mm approx).



Figure 7:- CT lung neoplasm lobar sup sn nonspecific

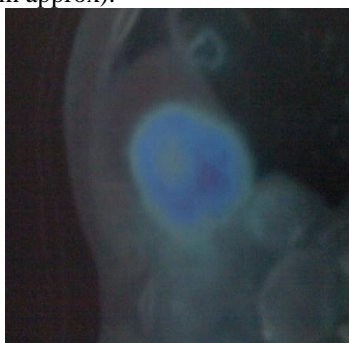
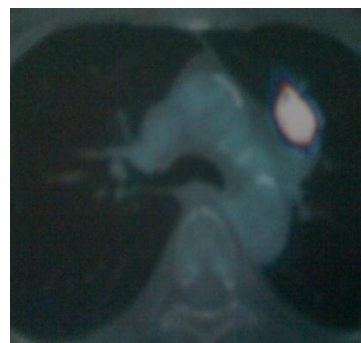


Fig.8:- Fig.9:- PET PET accumulation outline reactive process



At PET-CT accumulation of the tracer in the upper field of the left lung in the paramedian attributable to non-specific reactive process .Fig (8.9), then he performed genetic test that confirmed mutations in the PAX2,

Results:-

Although there is no evidence to recommend a screening of the first level in the event of gross hematuria, during the consensus conference, the prevailing view (91%) is in favor of 'ultrasound. The sensitivity and specificity may reach 95% without exposing the patient to the risk of ionizing radiation, however, in the absence of scientific evidence about it, the prevailing view (94%) is that the outpatient preoperative cystoscopy in case of positive ultrasound can be omitted . The UroCT is the imaging examination with greater sensitivity and specificity. The limit lies in the poor ability to detect flat lesions non-invasive muscle. The urography has been almost entirely supplanted by UroTC. The current biomarkers do not have a sensitivity and specificity greater than urinary cytology, and The aromatic amines, NAT2 and NAT1 of tobacco smoking products, are considered the main cause of bladder cancer. THE observed patient is a heavy smoker with two neoplastic lesions in different venue. The recent introduction of multidetector CT equipment, has made possible one urinary multiphasic study, executable with higher resolutions both spatial and contrast, allowing the simultaneous evaluation of the renal parenchyma, vascular structures and the urinary tract, as well as all remaining surrounding abdominal structures. In fact it can be a method of imaging "one-stop-shop" that allows you to diagnose the possible cause of hematuria and, in the context of a bladder tumor, and perform an accurate local and remote staging. A methodology of rigorous study of the urinary tract includes at least four phases of acquisition (in basal conditions and in the arterial phase, venous and urographic, after intravenous injection of at least 120-150 ml of iodinated contrast material, with a flow rate of about 2 , from 5 to 3.5 ml / sec), a thin layer (layer thickness of 1-2.5 mm, with reconstruction interval of at least 0.6-2 mm) Fundamental, is also the integration of axial images with reconstructions two- and three-dimensional images, which represent the real added value of multidetector CT, allowing to overcome the inherent limitations axial imaging. (11,12) It is still in the process of validating the utility of virtual cystoscopy in the diagnosis of bladder cancer, which, however attractive method,

does not have to date a real added value in diagnostic terms, except in specific conditions (13,14), it could in fact be performed even in cases where the traditional cystoscopy is contraindicated or in the case of disease localized in a diverticulum. With Uro-MRI techniques, similarly to what happens with the urography and with CT, it is possible study of the urinary tract, in order to seek synchronous urothelial lesions. In this regard, the first developed technique is called "pyelography RM", in which the high weighing sequences in T2 provide imaging of a static fluid, which urine excretory renal axis, without the use of means of intravenous contrast. With these sequences, obtained both in inspiratory apnea with breath synchronization, the urine signal is enhanced by providing an image cast of the lumen of the urinary tract, while the consensual reduction until complete cancellation, the signal from the parenchyma and from the surrounding tissues, it helps to further maximize the signal. It is the ideal method for obstructive uropathy study with varying degrees of dilatation of the urinary tract. Subsequently, there was also the so-called "excretory urography RM", in which the visualization of the renal excretory tract is made possible thanks to the renal excretion of the paramagnetic contrast agent, in a manner entirely analogous to what occurs in conventional urography and in uroTC. As for the CT urography, the two- and three-dimensional reconstructions are performed for complete visualization of the urinary tract. It is a method to be considered as a valid alternative conventional or all uroTC urography for the evaluation of the urinary tract, even in the absence of dilation of the same and for the possibility to study the lesions even without use of contrast medium and in the absence of exposure to ionizing radiation the spread of flexible cystoscopy (Narrow Band Imaging) is being promoted as a first-invasive test to be practiced in a patient suspected of having bladder cancer. In the most recent literature is applied a new technique to Flexible cystoscopy: the NBI. The narrow band of light (narrow-band) is strongly absorbed by hemoglobin and penetrates only on the surface of the fabric, increasing the visibility of capillaries and other delicate surface structures through an auto resection of the bladder tumor, which was followed by a typing of lung lesion with a needle biopsy CT guided whose outcome he called a borderline malignancy (cancer) which resulted in correlation to patient age implementing a radiation therapy.

Discussion:-

renal abnormalities, according to the classification of Ridson, are divided into: number of anomalies, seat of anomalies, anomalies of form, abnormalities of differentiation, The presence of a double district pyelo-ureter. Observed in the patient was incomplete, with the two ureters who join before leading into the bladder through a common orifice, Figure 3 also the duplicity era asymptomatic, and was accompanied by a fusion anomaly: a horseshoe kidney. It was also present malrotation renale. fig 4 lower poles of the two kidneys are fused, before the aorta, to left over an isthmus formed by fibrous or kidney parenchyma tissue. The etiology is probably naturally from the union of the two early research nephrogenic blastema during migration. Kidney fused masses showed two excretory systems and two ureters, therefore the malformation was totally asymptomatic, occasionally diagnosed in the presence of haematuria. Capricious. combine is also the seat of anomalies such as renal malrotation a malformation asymptomatic, relative-frequent mind, caused by a partial rotation of the kidney during the medialisation process. The kidney took the form of an ellipse with goblets oriented medially. finally to complete the clinical picture was present also the ectopic kidneys, relatively frequent anomaly. asymptomatic and is manifested only if the renal parenchyma undergoes infections or compressing an abnormal blood vessel dilates the urinary tract. In this uniqueness it is associated the presence of a dual solid education on both the superior lobe closely related bronchus and vessels Fig 7,8,9 and another in the bladder trigonal fig 1,2,5,6. In presence of this association we have performed a genetic evaluation through the execution of genetic tests aimed to research the possible link between cancer and malformation with the cancer risk assessment. With genetic testing expression identifies a test diverse set designed to identify changes in the DNA sequences of the germ line or products which are derived directly from the modification of transmissible genomic sequences. often sick children have inherited the syndrome from parents who, being affected in less severe form, I'm not aware of. Genetic diseases are localized Sui chromosomes, or inside the cell nucleus, and are lined up and organized the genes. (15,16,17,18) A small proportion of genes is extranuclear, as present in the mitochondria (cellular organelles that are inherited from the mother). (19,20,21,22) The alterations of mitochondrial and nuclear genes and, often, the interaction with environmental factors, contribute to identify five main groups of hereditary diseases. a) Chromosomal. They originate from a change in the number or structure of chromosomes. b) Genomic. They are due to the loss or acquisition of a number of genes (often whole segments of DNA, DeoxyriboNucleic Acid, containing tens or hundreds of genes) from a specific chromosomal region. c) Single gene or Mendelian. Due to the mutation of a single gene. They are classified into autosomal diseases, when the gene is not located on the sex chromosomes, and X-linked, in the case in which the disease gene is localized in the female sex chromosome. (23,24,25,26) They are also divided into dominant and recessive diseases, depending on which is sufficient only one copy or two copies of the mutated gene to determine the disease. d) multifactorial or complex. They are due to the additive effect of genes and environment. e) Mitochondrial. They originate from the small

circular chromosome mutation present in multiple copies in the mitochondria. It is a relatively small number of diseases, mainly of neuromuscular interest. In asymptomatic family members, taking into account the psychological and legal implications, as well as medical, accompanying the presymptomatic diagnosis of hereditary diseases, it is necessary that the adoption of a special protocol, which provides for the implementation of an interview in the presence of the geneticist and psychologist.(27,28,29,30) of meetings with the patient and with the help of other specialists reflected on the benefits and risks arising outcome survey genetics, in particular effects on the reproductive and career choices. In order to ensure the autonomy of individual choice, in carrying out predictive testing, it is useful to program only in people older. During the counseling is important that we provide the most accurate information on the natural history of the disease and about the advantages and disadvantages of the tools available for the risk reduction, in order to help the patient to make the most appropriate choices in his case. (31,32,33,34) On the basis of personal and family history, or the finding of a mutation prognosis. Special attention is paid to predictive testing, targeted at asymptomatic relatives of patients with an inherited malformation. The first mutational event (M1), which predisposes to the development of the tumor and / or malformation, is due in most cases to truncating mutations: nonsense mutations or deletions / insertions frameshift, located in exons of the gene. genotype-phenotype correlation studies have shown that usually individuals with large deletions of the gene develop a milder phenotype, compared to individuals with other types of mutation with total loss of protein function.(35,36,37,38) The second mutational event (M2) is represented by mitotic recombination (50%) 16, non-disjunctions, with or without duplication (about 40%), microscopic and submicroscopic deletions, point mutations, and inactivation of the gene (10%). The second mutation occurs more frequently than the first 94, and is more sensitive to environmental factors, especially those that determine chromosomal rearrangements, such as ionizing radiation. correlation between the clinical features and the parental origin of the deletion Genetic counseling as part of hereditary diseases or defects, is set up as a communication process, which aims to bring home to patients and their families, the information relating to the characteristics of disease, methods of transmission, the risk of recurrence and possible therapies, including reproductive options, which are relevant to their condition.(39,40,41,42) Key Features of genetic counseling is to not be directed to the whole family, and not having in any way influence the decisions of family members. (43,44,45,46) The identification of the mutation allows you to extend the analysis to other family members and therefore identify, in a short time, those who are at risk of developing the disease and pass it on to their offspring. In cases where the molecular analysis does not identify a constitutive mutation in the gene it should emphasize the need for close monitoring. The geographical correlation allows also to compare health risk. It allows to answer the question levels if there is a relationship between risk of death or birth defects as evidenced by numerous studies, there is an increased risk of cancer and malformations in the presence of high socioeconomic deprivation. With increasing environmental pressure has been a steady trend in the health risk. The risk of congenital malformations, respectively increases, by 54%, and 83% significantly higher. The correlation study also shows that the total mortality grows on average about 2%, in both sexes, from a more environmental pressure category to the next in higher pressure with a statistically significant trend showing an increase of excesses of congenital malformations with increasing hazard (47,48,49,50)) the clinical diagnosis therefore can be confirmed by molecular examination in 75% of patients. They are reported genotype-phenotype correlations in the field of clinical expression of patients with mutations in specific genes. Some genetic disorders can have partially overlapping clinical manifestations and can then ask for differential diagnosis in the diagnostic process underway. Many human hereditary characters are distributed in families proportionally different from those expected by Mendelian characters.(51) As the patient examined the interaction between genes and the interaction between genes and environment have changed the expression of a phenotype. (Heavy smoker, birth defects, presence of mutation, economic deprivation) Some human diseases have a polygenic inheritance and / or multifactorial. In these cases the hereditary transmission is not easily recognizable family tree. Many birth defects and many common adult diseases are inherited as discontinuous multifactorial traits. The susceptibility factors to a discontinuous multifactorial phenotype in the population are distributed according to a normal curve, which predicts that only a few subjects have a small number or, respectively, high susceptibility factors, while the majority has an average number of factors. The discontinuous phenotype occurs only when the number of susceptibility factors exceeds a threshold value empirically defined. At the manifestation of the phenotype contribute genetic and environmental factors. The relatives, who share with people with a number of genetic susceptibility factors is proportional to the degree of consanguinity, have a relatively higher risk of developing the same phenotype. The assessment of the risk of recurrence is more complex than the illnesses.

Conclusions:-

Genetic testing can predict with great accuracy the future development of the health of an individual. They concern both the subjects in good health as those with symptoms of a disease, and may have profound repercussions for relatives of the person who is subjected to a test. Therefore, it is important to provide an adequate level of support for the patient and his family before proposing a genetic test and to communicate the results. With the progress of scientific knowledge on the complex genetic traits and their influence on the phenotype, genetic tests are undergoing an evolution that tends to shift their scope from traditional medicine to the molecular. They always cover the most complex traits rather simple, and their sphere of relevance will move from the family to the individual; for diagnostic and preventive services that will be in addition to those of the risk determination and the predictive; the methodology will shift from the analysis of chromosomes and genes to the genome; the genetic research clinical protocols will be oriented more clearly towards complex diseases. In the presence of defects it is necessary to provide genetic testing for the purpose of assessing the health of the individual. You can assume the major socio-ethical-legal implications for presymptomatic testing, the only genetic tests that can predict with certainty and in the future occurrence of a disease tests for clinical purposes provide information on the health status of subject. Those carried out for research purposes usually generate information that is not indicative of health status or genetic risks of the subject, but it is useful for the purposes of statistical or scientific research. Understanding the reasons for the functioning of our body with the understanding of the mechanisms of disease does not lead directly to a cure, but produces useful materials in studies focused on therapeutic perspectives. To understand well the role of the environment, complementary to that of the genes

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RESEARCH ARTICLE

QUANTITATIVE ESTIMATION OF HYDROQUINONE IN FOUR COSMETICS SAMPLES FROM LOCAL MARKET OF SANGAMNER TEHSIL, DISTRICT AHMEDNAGAR (M.S.), INDIA.

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Cosmetics, Hydroquinone, Oxidizing agent etc.

Abstract

Hydroquinone has been used for decades as a skin lightening agent. However the use of excessive concentration of hydroquinone in cosmetics poses skin problems hence was banned. Four cosmetics samples were sampled from the local market of Sangamner Tehsil, District Ahmednagar (M.S.), India. The labels on the packages did not indicate the presence of hydroquinone.

Hydroquinone can be quantitatively oxidised using mild solution of H_2O_2 in presence of catalytic amount of Fe^{+3} and the quinone is quantitatively estimated using Systronics Visible Double Beam Spectrophotometer Model-1203 at 363 nm. The samples are found to contain 0.08641% to 0.11666% w/w hydroquinone and are said to be safe.

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Introduction:-

Tyrosine is the precursor for the synthesis of melanin. Tyrosinase is the key and rate-limiting enzyme responsible for the conversion of tyrosine into melanin by melanocytes in human skin^{1,2}. Many compounds that bind to the tyrosinase active site and inhibit melanin synthesis have been developed as agents to lighten skin color, including hydroquinone³. Hydroquinone is indicated clinically as a 2-5% ointment for the gradual bleaching of hyper-pigmented skin in conditions such as melasma, freckles and senile lentigines as well as chloasma⁴. Hydroquinone based products could be potential carcinogens as most of the benzene metabolites and derivatives are health hazards⁵. The U.S. Environmental Protection Agency has not established a reference dose (RFD) for hydroquinone. However EPA has calculated a provisional RFD of 0.04mg/Kg/d, hence there is no occurrence of chronic, non cancer effects of this dose but as the amount and frequency of exposure exceeding the RFD increases, the probability of adverse health effects also increase⁶. So it has been recommended to ban in cosmetics. Several analytical methods for the determination of hydroquinone in skin preparations (cosmetics) are described, including High Performance Liquid Chromatography⁷⁻¹⁰, Capillary Electrochromatography¹¹ and other Spectrophotometric techniques¹²⁻¹⁶.

The aim of the present study is to quantify the hydroquinone in some popular kinds of whitening creams in local market of Sangamner tehsil, District Ahmednagar, (M.S.) India.

Materials and Methods:-

Instrument:-

The absorbances were measured using Systronics Visible Double Beam Spectrophotometer Model No.1203 with 1cm rectangular quartz cells.

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Chemicals and Reagents:-

All the chemicals used were of analytical reagent grade. Distilled water was used for the preparation of all solution. 100 ppm Standard solution of hydroquinone was prepared in water. 10 ppm aq. Fe^{+3} in the form of FeCl_3 were used as a catalyst and 0.6 % solution of H_2O_2 was used as mild oxidising agent.

Procedure for Spectrophotometric Determination of Hydroquinone:-

2, 4, 6 & 8 ml of 100 ppm hydroquinone were added into a series of labelled test tubes. To each tubes, 5 drops of Fe^{+3} and mild solution of 2 ml H_2O_2 were added. The contents were mixed well and heated in water bath at 100°C for 20 minutes. The solutions were cooled at room temperature and diluted to 25 ml using distilled water. The absorbances were measured within the stability period of 3 hrs at 363 nm against reagent blank (Table 1). A calibration plot was obtained by plotting Absorbances versus ppm of Hydroquinone.

Estimation of Hydroquinone in Cosmetics Creams:-

2 g. of each sample was boiled with distilled water and contents were filtered through Whatmann No.42. This filtrate was diluted to 25 ml with distilled water. Into a series of labelled test tubes; 2, 4, 6 & 8 ml of diluted sample solution was added. To each tube, 5 drops of Fe^{+3} and mild solution of 2 ml H_2O_2 were added. The contents were mixed well and heated in water bath at 100°C for 20 minutes. After heating, the solutions were cooled at room temperature and diluted to 25 ml using distilled water. The absorbances were measured within the stability period of 3 hrs at 363 nm against reagent blank (Table 2).

Results:-

The hydroquinone content of each sample was deduced by extrapolation at corresponding Absorbance from Standard Curve (Table 3). The results of spectrophotometric analysis confirmed the presence of hydroquinone in four samples having varying levels. All the four samples contained less than 2% hydroquinone which is permissible limit given by WHO.

Table 1:- Absorbances for oxidation product at different concentration of HQ (Standard solution)

Sr.No.	mL of 100 ppm HQ	Drops of Fe^{+3}	mL of H_2O_2	Dilution (mL)	Absorbance
1	2	5 drops	2	25	0.044
2	4	5 drops	2	25	0.166
3	6	5 drops	2	25	0.296
4	8	5 drops	2	25	0.311

Table 2:- Absorbances for oxidation product at different concentration of HQ (Sample solutions)

mL of sample	S_1	S_2	S_3	S_4
2	0.115	0.075	0.066	0.077
4	0.158	0.206	0.102	0.135
6	0.162	0.249	0.175	0.327
8	0.219	0.293	0.212	0.407

Table 3:- HQ content of samples deduced from Standard Curve

Sample	HQ in Working solution (ppm)	HQ in Sample (% w/w)
1	72.72	0.0909
2	93.33	0.1166
3	69.13	0.0864
4	86.15	0.1076

Conclusions:-

1. In the present study, we developed a new method for quantitative estimation of hydroquinone in cosmetics samples. Mild solution of H_2O_2 was used for oxidation of hydroquinone to quinone in presence of catalytic Fe^{+3} .
2. Although there is no adequate warning on product labels about the levels of hydroquinone present, this work revealed that the four collected samples of beauty creams from the study area contain 0.08641 % to 0.1166 % w/w of hydroquinone and hence are said to be safe.

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RESEARCH ARTICLE

Changing trend in choice of schooling in selected areas of rural Almora, Uttarakhand: Some case studies.

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Parental choice, quality education, discipline, safety, public and private school

Abstract

In recent times the number of public and private schools has flourished in all the states of the country, including Uttarakhand and thus the idea of school choice between government schools vis-a-vis public/private schooling has received considerable attention in the minds of the parents. Consequently the trend to send children- son and daughter towards English speaking private schools has grown significantly amongst middle and lower-middle income group families. This has mainly arisen due to the growing awareness on the quality aspect of education. Since private schools are expensive, therefore, cost of schooling does influence girl's schooling in India. The gendered nature of parental choice in the context of schooling is known to play some role; this aspect has not been considered in any of the studies undertaken in this region of the country. Hence a field study/survey has been undertaken in rural areas of Almora district in Uttarakhand comprising, four selected blocks, focusing on some case studies, with a view to understand the attitudinal change of parents on the choice of schooling for their daughters irrespective of their family status like identity and financial conditions, parental education, caste and gendered nature particularly in reference to their daughters.

This study reports a current growing concern not only among urban parents but also amongst parents in rural areas who realise the importance of quality education for their children. Appropriately, a growing trend has emerged to send their children, including daughters to private schools. A close analysis of interviews with students, parents and teachers reveals that educational choice exercised in these middle or lower middle income group are connected with issues of marriage, self dependent (job), or gendered norm of conduct (like discipline, honesty). Apart from considerations of costs, gendered ideas of school proximity, quality education, security and disciplines are of prime significance in their choice of high school for children and particularly for daughters.

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Introduction:-

'Choice' is a relative term and at times be deceptive. Choice is generally exercised keeping in mind several thoughts and/or constraints in actual situations. Hence it is used as a tool to examine the complex dynamics of social life (Goswami, 2015). The idea of school choice between government schools vis-a-vis public/private schooling is changing in the minds of the parents. The conception of the social space in which parental choice of school is generally exercised has been questioned. At the same time framing of issues in individualistic terms of consumptions at the expense of social process of choice can be debated. On the other hand some scholars have pointed out how the capacity of, and the inclination towards, making choice vary with class based and gendered practices of parenting (Bowe et al. 1994; Reay, 1996, 1998; Ball and Vincent, 1998; Sarangapani and Winch, 2010).

It must be mentioned that relatively few studies have been conducted in India on the class and gendered nature of parental choice in the context of school market expansion (Goswami, 2015). It has been reported that a larger proportion of boys as compared to girls attend private schools, since private schooling involves higher investment (De et al., 2011). Although the cost of schooling do influence girl's schooling in India, nevertheless, it does not give a full account of the multifarious ways in which class, gender and community identity is reflected in specific situations (Goswami, 2015). Some studies have, however, examined family strategies of schooling across class and presented situations where girls, instead of boys, are sent to high cost private schools (Chopra, 2005; Nambissan, 2010).

The concept of choice needs to be examined in a social context which necessarily is interaction of class, caste and gender (Goswami, 2015). Debates on school choice have gained attention in the South Asian context in recent years, especially in reference to the category of the urban poor (Alderman et al., 2001; Tooley, 2009). Expansion of private school in urban India has occurred in recent times and some scholars have presented evidences that indicate acts of resistance by the poor against the state controlled bureaucratic education system that generally fails to deliver quality education (Tooley et al., 2007; Tooley, 2009). In this context, some studies have highlighted the need to understand the social context of schooling in South Asia and have found flaws with the quality of low-cost private schools (Sarangapani, 2009; Sarangapani and Winch, 2010). Further, some scholars have also focused on the class strategies made by relatively well off group of people to convert their economic capital into cultural capital. Although Nambissan (2010) has reviewed the reproduction of educational advantages produced by middle classes in India, however, while highlighting the class aspect of family strategies, many of these have tended to over look the fact that the schooling process itself is shaped by and is constitutive of gender relations in a family setting (Goswami, 2015).

Low-cost private schools appear to provide the necessary numbers, quality and security to parents concerned about girls' education (Tooley, 2009). While providing an account of private schools for the poor in Kenya, China and India (Hyderabad), he documents narratives of parents concerned about quality education and safety of their daughters, and mentioned that parents relied more on low-cost private school for these reasons. These narratives of *parental* choice for girls' education show them simply as urban poor devoid of any history and social identity (Tooley, 2009). Another aspect which requires attention when analysing the parental preference is to examine the dynamic of class, caste ethnicity and gender in a particular social setting. While examining the girls' education in a village in Rajasthan (Hill et al., 2011) the issue of parental choice beyond that of an expanded school market was envisaged through a study of class and caste relations in a village. However, in this study by Hill et al. (2011) the gender issue was examined based on only cost of school and attempt to address the variation in gender norms by class and caste groups was not made. Further, only limited studies have attempted to examine the schooling patterns in light of the advances made in feminist scholarship in family and kinship studies (Chanana, 1993). Gender concerns regarding schooling in India are often expressed through macro indicators of gender gaps in dropouts and enrolments of students from various categories (Govinda and Bandopadhyay, 2012). However, as mentioned by Goswami (2015), nuanced investigation of gendered process implicated in the schooling process and situated in family context is rare.

Family aspects also play an important role in schooling. Thus at the family level, decisions regarding the future roles of children are deeply influenced by hierarchies of sex and age, and ideologies of conduct which shape gendered identities of young men and women. These identities are reflected differently in a plural society where different communities have their own ways of preparing young generation for adult roles (Goswami, 2015). In South Asian societies, the upbringing of children is marked by gender ideologies of seclusion and segregation (Dube, 1997). In the Indian context, Chopra (2005) provided the linkage of educational choices with particular acts of feminine

identity constructions to gain access to globalised marriage market among land owning caste families in agrarian Punjab. Further, linkage between family structures, gendered norms of marriage and girls' schooling have been reported by several workers (Chanana, 1993; Seymour, 1999; Manjarekar, 2003; Hasan and Menon, 2005). However, the implications of linkages between ideologies of sexual segregation and educational choice have not been examined in the context of the expansion of school market in recent years (Goswami, 2015).

In this article a survey has been undertaken to understand the attitudinal change of parents for the choice of schooling for their daughters irrespective of their family status like income, parental education, caste and gendered nature. The first part of the article reviews/discusses on preference of schools, i.e. school choice; the second part describes the study/field site and the methodological approach adopted for the empirical study involving a set of students and their parents located in the rural area in Almora. The third part dwells with the various cases studies, focusing on the gendered nature of choices and constraints, regarding schooling options negotiated by parents and children at different stages during schooling.

Methodology and Field area:-

Over the past few decades the number of low cost private schools in the country has markedly increased. Studies have indicated that even lower income group families are moving away from government schools to private schools in rural areas (Hill et al., 2011). But one must acknowledge the fact that there is a wide category of private schools which widely vary in their human and physical resources and in their degree of independence from the state (Goswami, 2015). In the hilly areas of this country the number of private schools could be noticeable right from the time of post-independent days but these were for mostly upper class families. But in recent times large number of low costs private schools have flourished. In rural areas of Uttarakhand too, many private schools both at the primary and secondary levels, are growing very quickly in number. Consequently the trend to send children- son and daughter towards English speaking private schools has grown significantly amongst middle and lower-middle income group families.

The study was conducted in the state of Uttarakhand where the literacy rate increased from 71.62% in 2001 to 78.82% in 2011, with 87.40% for male and 70.00% for female. The overall literacy rate of 78.82% is higher than the national rate of 74.04% (in 2011). The field survey was conducted during September 2013 to May 2014 in Almora district, Uttarakhand which comprises of 11 blocks (Bhasyia-chhana, Bhikyasen, Chaukhutiya, Dhaula Devi, Dwarahat, Hawalbagh, Lamgarah, Saalt, Syalde, Takula and Tarikhet) and the corresponding forest area. Based on the female literacy rate, the blocks were arranged into two categories, i.e. high and low (source: Vikas Bhawan, Almora, Govt. of Uttarakhand; based on Census of India, 2011). Since the female literacy rate in rural areas of Almora district is 67.88%, blocks with literacy rate up to 67.88% were categorized under 'low level' while the blocks with literacy rate of above 67.88% were placed under 'high level' of literacy. Thus from each of these categories, two blocks each, i.e. with lowest and highest literacy rates, comprising a total four blocks, namely, Dhaula Devi (60.67% literacy rate; 35 km from Almora), Lamgarah (64.09%; 33 km from Almora), Hawalbagh (73.69%; 14 km from Almora), and Tarikhet (74.04%; 56 km from Almora) were chosen. The location of these sites (in map) and their geographical position, area, population, literacy rate, etc. in these four different blocks selected for the study have been detailed earlier (see Nandi and Joshi, 2015).

The principal occupation in the rural areas of Almora district is cultivation, cattle rearing, poultry farming, small scale business, and wage labour. More recently there has been an expansion of economic opportunity in the informal service sector like semi-skilled labour or driving. Most of the families are nuclear but vary in size according to number of children. In some of the families, the mother also worked to supplement the family income. Most families relied on wage work combined with seasonal cultivation and cattle farming to supplement their income. Majority of the parents had basic literacy education through primary or secondary schooling. In a few cases (particularly fathers) were found to possess higher secondary and above, while in some mothers never attended any school.

Information relating to geographical position, area, population, literacy rate and number of school/college of under different blocks/sites in Almora district selected for the study has been provided earlier (Nandi and Joshi, 2015). A brief description of the four blocks is given below:

Lamgarah Block:-

Lamgarah block head quarter is located about 33 km from Almora town. Most of the area is situated in a hilly mountainous area (Figure 1) and the overall economic condition of the people residing is quite poor with a number

of people living below poverty line. Majority of the people living in this block, work as daily wage labourers. Lamgarah block is an under developed area; when the male and female literacy rate is compared, female literacy rate is much low (64.09%).

Hawalbagh Block:-

Hawalbagh, the block headquarters, nearest to the Almora town among all of the selected blocks, situated about 14 km from Almora town, is a relatively developed block with a population of 67,447. River Kosi, the main source of water for Almora town runs just below the area; the forest area is relatively less and with small scattered habitation. The people are mainly engaged as daily wage labour or carry out small business. It is well connected and walking distance from the Almora-Kausani and Almora-Ranikhet highway. The overall literacy rate of this block is good and the female literacy rate (73.69%) is well above the district's female literacy rate (69.93%).

Dhaura Devi Block:-

Dhaura Devi block is situated about 35 km from Almora town on way towards Jageshwar temple, historically an ancient temple of Lord Shiva; also called one of the twelve Jyotirlinga temples. The block has a population of 60,620 with a total literacy rate of 73.01%, and a female literacy rate of 60.67% which is lowest in rural Almora. Thick forest with small habitation is scattered in the block (Figure 2). The block has the largest area of 324 sq km among the four selected blocks but is most back ward as far as the female literacy rate is concerned.

Tarikhet Block:-

Tarikhet, a well developed block as far as literacy rate is concerned, is situated about 56 km away from Almora town. It is well connected to most of the important places of the state but is far away from Almora town (Figure 3). It has highest female literacy rate of 74.03% among the selected four blocks in rural areas. The well known Dwarahat Engineering College (now Bipin Tripathi Kumaun Institute of Technology) is located about 43 km away. The Army Cantonment area, based in Ranikhet town, is only 14 km away. The remarkable literacy rate of 88.75% can be attributed to the presence of the institution (Army base) and several schools in the area.

The criteria for selection of respondents was only the girl children aged between 6 to 18 years (excluding married, widow and divorce) with their parents (without any caste, race or creed) in order to get a complete range of information. The sample is representative of various variables like age, standard or class, family, financial condition, parental education, and parental attitude of the respondents. The sample consisted of only school going students of various types of school (Government, Private and Under schemes).

Information was gathered through in-depth interview and conversation with students, some parents and teachers. It comprised of questions on socio-educational and demographic characteristics as well as on various aspects related to educational choices in these families. For parents, apart from expenditure, considerations of proximity, security, discipline and quality education (mostly English medium) are of prime significance while discussing their choice of school for children.

All the interviews were basically conversation of educational patterns and their future plans of their children. In order to gain rapport with the respondents I gave a brief introduction of myself and explained the actual purpose of the study. I also assured them about the importance of their contribution and promised them to maintain confidentiality. In order to make the interviews more efficient they were conducted in a lonely place where no other person was present. Initially interviews started with common general talk and when the respondents were comfortable then proceeded to the actual questioning. Despite the initial inhibition, the girls gradually adjusted to the interview situation and appeared to enjoy talking and sharing their experiences.

The interviews were conducted in Hindi depending on understanding of the respondents (all dwellers of the block) and on weekdays in the afternoon, except interview with the male teacher and office staff of G.I.C in Tarikhet blocks. Later I translated the interviews in English. This study draws primarily on data collected from a set of students and their parents; it also relies on the secondary data from state government office and case studies undertaken in four blocks.

Field experience and case studies

In order to understand the reasons why the parents exercise a choice of schooling in different blocks undertaken in the study, several criteria emerged from the discussion. Three themes, as observed by Goswami (2015) in a recent

study in Assam, also emerged from the interviews of this investigation; these are costs, security and discipline. Therefore the case studies are broadly grouped under these 3 groups.

1. Cost of schooling:-

In the given familial context, strategy about school may be conceived of as consisting of a range of actions regarding beginning continuing switching and discontinuing school attendance. Some of these identified limited resources in terms of net income and provision of low-cost schooling. It is important to note that in many families, expansion of educational provision through the government seems to have made the choice of schooling much easier for the present generation of children as compared to their parents. The majority of the parents said they themselves did not have as many facilities such as free meals, uniform, and books; moreover even the school infrastructure was reported to have been very poor, and they had to sit on a mat and the roof was not 'pucca'. The costs of the schooling of children continued to be a common concern within family. The costs included not only school fees, but many other expenses like transportation, stationery requirements, uniform other items (Goswami, 2015). However, the time and situation has gradually changed in the last two decades. The parents are quite concerned about not only education, but also the quality of education for their children, equally for both sons and daughters. When it comes to schooling decisions within a family, costs are not merely calculated in monetary terms, and even when they are, the calculations are not same for every child in the family. The small family with stable income are more interested about quality higher education for their children. Besides the formal school education they want their children be smart, disciplined, honest, and able to speak English. Therefore, they are extremely selective about the choice of schooling for their children, not only for boys but equally for their daughters. They have felt that they would not be able to fulfill their desire through government schools. These experiences made them attracted towards private or public schools. The following cases from the different categories of family with contrasting experiences somewhat reveal these complexities.

Case 1 (Lamgarah block):-

I interviewed Sri Madan Singh Bisht, a resident of Lamgarah. A home maker wife with two sons and three daughters, Sri Bisht is an owner of small grocery and stationery shop cum tea stall. With a monthly income ranging between Rs. 5000-7500 (Lower middle income group) he takes sufficient care of his children in education. Due to lack of degree colleges in the area both the first and second daughter are enrolled as private students studying Bachelor of Arts (1st year and 3rd year, respectively). Although a new degree college has started recently but due to insufficient number of teachers, the student enrolment is quite less. The third daughter (a student of class 10) attends a nearby Government Inter college. In contrast, his two sons have been admitted in a nearby private school (Upper primary). During survey it was noted that there are a few other private schools in Lamgarah block. He said that due to financial constraint he cannot afford to support the third daughter to private school due to higher school fees, nevertheless, he has arranged private tuition for her. He highlighted that his wife completes domestic work on her own and occasionally, if required absolutely, calls her daughters in helping her. Sri Bisht and her wife want that all their daughters continue studies up to graduate level and above, and dreams that after completion of daughter's education they will take up jobs and enjoy their independence.

While discussing about the quality of education, he indicated that although there are a number of Government Junior schools in the block but student enrolment is very less compared to private schools. The number of student is around 50 in a government school while it is 300-400 in private schools. He also mentioned that presently there has been a growing trend among parents to send their children to private schools where children are taken care and given more attention. In general, it was understood that parents do send their children (of both government & private schools) weak in studies, for private tuition to local teachers who charges fees between Rs 200-250 per month.

Thus it is clear that there has been a gradual change in the attitude of parents who pay sincere effort to educate their children thus fostering educational development.

Case 2 (Hawalbagh Block):-

The survey revealed that people of this block are quite serious about education and ensure that every child goes to school. During interview in one of the villages of the block, i.e. in village Mahetgaon I met Mrs Seema Negi (B.A.), a primary school teacher of a private school located in Gwalakot village (Figure 4). The school, run by an NGO, is Upper Primary (up to 8th standard) and 10 km away. Mrs Negi is mother of two children- a son (3 years old) and a daughter (6 years old) who are students of LKG and class 1, respectively in the same school. She and her children have to put in some effort since they live at the top of the hill, about 2 km from the main road. Both the children

accompany her in a school bus owned by the same NGO. She appreciated the bus facility and said that without transport it would have been very difficult for her going to school with two little kids. The bus service provided by the school makes easy for her and many students of that area. She is a caring mother with no mental bias between her son and daughter, and therefore has admitted both of them to the same private school. Education, according to her is the right of a child and duty of parents is to provide good education to their children; whatever demands the children put forth, efforts must be made to fulfill it. Mrs Negi lives in a nuclear family and is of the opinion that one can take care of the family in better manner than a large family. She said that as proper upbringing of children is extremely important, therefore, the number of children should be restricted. Another girl student (of class 2) of the same village is being escorted by Mrs Negi in the same school bus (Figure 4).

Case 3 (Hawalbagh Block):-

While discussing with Mrs Negi it was noticed that a person was listening to our conversation very carefully. On interacting with him, Sri Surinder Singh Mehta, owner of small road side tea shop has a different story to tell. He informed that with a monthly income of little more than Rs 5000 (Lower middle income group) he is making sincere efforts to make a decent living. His two children, a son (15 years old & student of class 9) and one daughter (17 years old & student of class 11), attends Government Inter College located in Chaura, about 3 km walking distance from his home. He said that his daughter wants to join Polytechnic after completion of her intermediate studies.

According to him expenditure in a private school is much higher than in government school. In general most villagers want to send their children to Private school since personal attention to students is lot better and facility like school bus is also available. He mentioned that student enrolment in private schools has increased considerably. However, he admitted that with little income he can't afford to send his children to a private school. Since the government school has no bus facility, his children has to walk a distance of 3 km which results in daily loss of 2-3 hours, without proper reason, and he felt sorry for this wastage of time.

Education, according to him, is very crucial in this modern age and education of woman is even more important because it can be used as a tool at the time of crisis in terms of widowhood, desertion and also for economic upliftment. Sri Mehta also said that he cannot afford to give any dowry for his daughter's marriage but is of the opinion that if he educates his daughter who then gets a job, it will be a gift more than a dowry. Besides he said that only educated woman can bring up a generation of enlightened citizens. On the number of children in a family, he mentioned that "Poor people like us cannot afford to have too many children in today's expensive era". According to him, more teachers need to be appointed for improving the quality of education in government schools. He categorically mentioned that Government should pay more attention and the various educational schemes that are launched must be monitored and assessed for their success/failures. Moreover, more High schools and Degree colleges need be established in nearby areas so that poor students can study and fulfill their dreams.

2. Quality and strict schools around:-

In recent times preference for a good quality education and strict school selected by parents for their children, not just for son but also for their daughters, is a rising demand and plays an important role in working middle class family's decisions about school choice. Studies by Reay and Ball (1998) on parents from varying class position suggested that 'for many working class families, locality and community represent security belonging and connection; places and spaces to feel (relatively) safe and comfortable in' (Gewirtz et al., 1995 as cited by Reay and Ball, 1998). Thus in ethnically diverse setting the conception of safe and comfortable spaces for working class parents will actually vary. Moreover, the sense of varied types of threat may arise quite often during conversation with mothers mainly with reference to their daughter's safety. However, such consciousness does not surface in the case of each and every school, and parents generally negotiate their community identity in their concerns about school quality (Goswami, 2015).

In this study, experiences and views of parents and children of desirable schools from a given range of options available to them were obtained. The choice expressed was mainly based on quality of education and discipline in the schools compared to other government schools on various attributes. Many parents ignored the distance of school and admitted their daughters in private schools only on the basis of quality education. Such parents also dropped their children (son and daughter) to school bearing own responsibility, ensuring maximum security for their children, particularly daughters. It emerges on various occasions that the proximity of school location is expressed not simply in terms of physical but also in terms of status. Further, a striking pattern among the majority dwellers in the rural area has been in their preference for the English medium private schools for their children, both son and

daughter. In families with more than one child, the parents were inclined to send the younger sibling to same school as the older child.

During discussion with Principal of a popular English medium private school in Hawalbagh block several facts emerged as to why parents send their children (son and daughter) to private school even if nearby government school are available. The Principal indicated that currently there is a growing trend amongst parents to send their children (both son and daughter) to private school, even if the school is situated far away from their locality as the parents can be assured of their children's safety in this school compound. The children cannot bunk school and roam in school uniform; if someone is identified outside the school campus, the matter is reported to the Principal and the students is likely to be thrown out of school. But in government school the situation is different. The first and foremost point the parents prefer is safety of their daughters and thus choice of a private school over a government school can be seen. Moreover, quality of education, discipline and attention to every individual student is given; Parent-Teacher meetings are regularly conducted, and the performance of the students are reflected in the Report card handed over to students in the presence of parents. Thus such methods not only help in maintaining discipline but also improve the performance and quality of students. Moreover, parents can be made aware of the performance (good or bad) of their wards. In this way the parents, teacher and Principal are fully engaged in maintaining the well being of the students. The Principal also mentioned that at times parents complain about the excessive home work and indicate that their daughters would fall sick completing the burden of assignments. The Principal made it clear that all students compulsorily need to complete their home work and there is no excuse; however, in case due to some reason a student cannot complete, she/he may be allowed to complete it in school, prior to the morning assembly/prayer time, with the permission of class teacher. The Principal stressed that only through hard work and dedication can students achieve the target result. Thus the resultant effort of hard work of teachers and students this school is well known for good performance in High school results.

It is important to indicate that the above said school was founded in 2006 with an aim to start an English medium school in rural area of Hawalbagh block. The number of students getting admitted has grown rapidly each year, particularly in Nursery and K.G. classes. Currently student enrolment is more than 350, indicating a growing demand of this Private school by parents of younger generation. The school, situated near Kosi market and on the Kosi-Almora main road has about 16 teachers along with 10 support staff. While the school is well known for maintaining a strict discipline citing a particular case (below) is worth mentioning.

On a Monday morning when I started the field activity at 9.00 O'clock, I observed that a group of four students (in school uniform) of the above mentioned private school are standing in front of a 'Barber shop'. This group included three boys and one girl. Quite surprised, I enquired the reason for their waiting near the shop? One of them replied that every Monday is a 'checking day' by the class teacher; short hair for boys, white hair ribbon for girls, nails be cut, clean uniform, shoes polished, etc must be maintained by all students, otherwise they will be punished. So the boys were having a haircut in the shop. The girl replied that she has forgotten to bring her class test copy and hence requires purchasing a new one. They also added that for every subject class tests are held weekly and the class test copy should be brought; the marks of these class tests are important as they are included in the 'Annual Report card'. The combination of careful attentive teachers and disciplined environment is what convinces the parents to send their children to a quality Private school with confidence.

Case 4 (Hawalbagh Block):-

Amrita Pathak and Deepika Mehta, two students of class 10 attending the above said school in Hawalbagh block were also interviewed. Both of students are very intelligent compared to other students of the class and compete with each other during exams to attain position in the class. They indicated that they loved their school extremely and attend classes every day without being absent. Both the girl's parents belong to Upper income group (above Rs 10000).

Amrita's father is a rich business man, educated up to 12th standard, while her mother, class 10 pass, is a homemaker. She has a brother who studies in 9th standard in the same school. They live in Shitlakhet and travel to school every day by their own car covering a distance of 22 km (one way) spending about 2-3 hours every day. After reaching home after school every day she feels very tired but when she thinks about fulfilling her dreams then she recharges herself. She is quite good in English and has a passion for the subject, thus aims to be a journalist cum English writer. She not only wants to be independent but also wishes to make an identity of herself. According to her, a girl should acquire as much education as she can. The main objective of education according to her is to gain

knowledge but she also mentioned about the other benefits of education as well. Besides gaining knowledge, her formal education can contribute in various ways in her future life, i.e. to fetch a job, a good match, keeps her tuned to the new developments that are unfolding virtually every day, she can enjoy high status, economic independence and respect in society. She said that in order to fulfill her dreams she gets every possible support from her parents who are more serious about her future than her. Since she belongs to a joint family she noticed that it was very difficult for her mother to take care of her every essential need, but she (her mother) always tried to solve her problems related to studies or personal matters. In order to assist her in studies, her parents also appointed a private lady teacher. When asked about the age of marriage, she mentioned that between 25 to 30 years is perfect age. Since a person needs time to settle them, Amrita was of the view that now a day not only men but also women should first take time to settle down and then marry. Whenever she gets spare time she loves to watch TV programmes because she thinks that through TV she can obtain quick information on what's happening in the modern world.

Like Amrita, Deepika Mehta is also a student of class 10 of the same school. Her father is a government employee while mother (educated up to class 8) a homemaker. She has a younger brother who is also a student of class 5 in the same school. They live in Mahetgaon which is about 3 km (one way); they travel to school using public transport. She is a very intelligent girl and her dream is to become an Environmentalist. According to Deepika education is a basic necessity of life in today's modern era and without education it's not possible to keep pace with the fast moving world. She lives in a joint family and she always noticed that her mother has no role in decision making; on the other hand seniors in the family, particularly her grandparents take all the decisions. There is no gender biasness in her house. Her family give equal facilities both to her and her brother. Deepika rarely enjoyed any leisure time because of burden of school home work, and likes to help her mother whenever she finds time, particularly in light domestic work like chopping vegetables, washing lentil, etc.

Both the students agreed that the atmosphere of the school is extremely suitable and strict discipline maintained helps to generate quality students. The attitude of most of the teachers towards students are very caring, friendly and helpful, nevertheless, are very strict. If any student is not able to complete home work the teacher ensures to get it completed within the school working hour. All class work is finished otherwise teachers help to complete their work after the school hour. They had a high praise for their school.

Case 5 (Hawalbagh Block):-

One fine sunny day I reached Katarmal village, close to the famous Sun temple. I met the Gram-Pradhan of this village. She has been living in this village since the 8-10 years along with her husband, who works in a nearby government office. She holds a Post Graduate degree and a mother of two kids, a son (8 years) and a daughter (2 years). The son is a student of class 3 in a nearby private school. When asked why they did not admit their son in a nearby Government Primary School (Junior Basic; up to class 5) situated close to home, she mentioned that she was not very satisfied with the quality of education in this school, and because this school has around 25-30 students of varying classes with only one teacher. It is difficult for the teacher to teach properly and at the same time take care of the students alone; in addition she has to perform other school duties including assigned Government work. The Gram-Pradhan also added that after lots of effort, another teacher was appointed. In addition, there is a Government Senior Basic School (up to class 8) where the quality of education is relatively better than in the Government Primary school, and sufficient number of teachers is present. When enquired about the quality of education, she wasn't satisfied with the standard and quality in the Government Primary School in the village; improvement is needed. She mentioned that at present there is a trend amongst all parents (belongings to all categories) to send their children to private school where teachers pay more attention and quality of teaching is also better. Being not fully satisfied with the educational standard of girls in her own village she narrated that they have to put in lot of hard work to compete with their counter parts of Almora town. She also added that extra educational schemes are necessary for promoting girls' education in this village.

I had an opportunity to conduct a *Group discussion* with villagers and parents of village Katarmal, to learn about the educational situation of their daughters. There are two Government schools (one up to class 5 and other up to 8) in the village where free education, mid-day meal and dress are provided. This has been very encouraging and excellent facility, particularly for the poor villagers, who mainly work as daily wage labourers. It was found out that presently there are no drop-out girls in their village; all girls are enrolled. Some parents expressed the desire to send their children to English medium private school but due to financial constraint unable to do so. They certainly felt that some expenditure has to be incurred to get quality education that can be obtained in Private school. Nevertheless, some parents of the village do send their children to a nearby low-cost Private school located 2 km

away in GBPIHED Campus (up to class 5) where the quality is better. In general, all the villagers expressed that the quality and standard of the teaching in government primary school is comparatively poor than in the private school.

Case 6 (Dhaura Devi Block):-

A survey and interaction with several girl students and their senior English teacher of a co-educational Intermediate College (semi-Govt) in Panuanaula was of the opinion that due to limited interest and financial problems literacy rate in that area is low but currently parents, students and also teachers are trying to overcome this situation. The student enrolment in this school is nearly seven hundred with about 70% girl students. He indicated that in the year 2015, there was no drop out case in the school. In general, after completing intermediate studies the students take admission in Kumaun University situated in Almora town. The attendance of girl students is better than other schools and they are relatively serious in studies; moreover the performance of girls is better than boys. Most students walk to school daily covering about 4-5 km (one way); while some live on top of the hill others stay down the road side. On the quality of education, he responded that in general it is good but some important aspects were missing out, i.e. out-knowledge, communication with modern world and exposure; these need to be improved for their betterment. He said that the people of the area, particularly the teen agers are very simple, unlike city dwellers they do not know what is happening around them. So he mentioned the urgent requirement of counseling and guiding the students of his school by some educated personalities (professors, scientists, doctors, and others). Various extracurricular activities (games, dance, drama, arts & NCC) are also taken up by the students; it was observed during the interview that both boys and girls were undertaking NCC training. He also said that NCC camps are being organized every year where both girls and boys participate. At present most parents are serious and particular about their children's education, providing equal treatment to their sons and as well as daughters.

During conversation we met three girl students of the above said Intermediate College. They live in a nearby village called Gunja. They narrated a similar tale, living a distance of about 4 km away (one way) which takes more than 2-3 hours every day. Daily they attend school, and try not to be absent. One student, named Kiran Negi (a student of class 12) said that "My father does nothing, every day he drinks and beat my mother, brother and me; it's a daily drama". Her mother works as domestic help in a nearby house, earning some money to support family, including their education. Thus despite many obstacles at home Kiran attends school daily with lot of hope that those good days will arrive one day when she and her brother will obtain a job after completing education. Her brother is also a student of class 9 in this same school. Kiran wants to study up to Bachelor's level and then would try getting a job as she desires to help her mother in sharing house hold expenses. She also added that since her father has no job and daily drinks alcohol, hence once she begins to earn she will take up the responsibility and then his father's mental situation will change. When asked about the age of her marriage, she quickly replied that for her it is a sort of problem, and has no time to think of marriage at present. Hence, she would complete her study first and then she would try to solve family's financial problem. She said at the end "my first duty is my home".

Case 7 (Tariket Block):-

Tarikhhet block has several schools within close proximity to the block headquarters. The Jawahar Navodaya Vidyalaya (Boys only) an important government school, and a girls' college, i.e. G.G.I.C. are located within 100 metres of the block headquarters. Two students of class 8 studying in G.G.I.C. were asked about their opinion for improvement in their life, they expressed that they would first like to complete schooling and subsequently higher studies (Graduate, PG, etc). They attended school daily and were not absent; but the most serious problem they faced every day was shortage of public transport (bus, small van, jeep, etc). The government schools have no facility of transport and some of them stayed in remote rural area about 5-6 km (one way) from their school. Moreover, according to the students after reaching home nearly most of them performed some domestic work. Following that they have to also complete school assignments. Thus they argued that it was difficult for them to perform well and attain good result like the students of English medium private schools as they reach home quickly because of school bus service and after schooling they can also take tuition. These students of private schools also take part in extracurricular activities. On the other hand the girl students of G.G.I.C. waste their precious time waiting for public transport. At times due to shortage of public transport they do not find seat in public transport (bus, small van, jeep, etc.) and have to travel in a pick-up van. During survey such a situation was observed and was disappointing for me to watch. Thus they strongly suggested that all government schools should try to provide vehicle facilities for children who live in distant locations like them.

Further, during this survey exercise I visited G.G.I.C. and discussed with two school officials about their school. Currently the total student enrolment is 315 (from class 1 to 12). It can be appreciated that 75 students (from class 1

to 8 class) are being provided with nutritious free lunch daily under the 'Mid-day meal' scheme. On enquiring about the performance of the students, they mentioned that they did extremely well. However, there is a gradual decrease in the number of girl students in their school and other government schools due to increasing trend among most parents to admit their children (son or daughter) in English medium private schools. It was observed that there are several private schools (mostly English medium) in that area.

3. Disciplining daughters through school:-

Parental perceptions of discipline and strictness of a school emerges as an overarching theme cutting across community affiliations, especially in reference to the daughters' education. A common yardstick of discipline in schools is to keep daughter safely in check within the school premises. A lack of supervision of students and/or lack of exposure, however, may be enough to earn the school a poor reputation, as observed in case of students of a semi-Government schools, that during interview they were not confident, initially getting quite nervous and were hesitating to give answer.

The parents wanted their daughters must go to school but should also be safe throughout the total travelling period for school. Thus it can be observed that in Case no. 4, the parents drop their daughter with their own responsibility. The anxieties and tensions about women's sexuality are ever strongly expressed with a consciousness of changing times, when the material cultural symbols are rapidly changing across class. Therefore, in an age when the daughters may have access to mobile phones, which not only opens up channels of communications but may also raise awareness about ones sexuality, the selection of a strict school is particularly significant (Goswami, 2015).

These anecdotal references to the disciplinarian measures of the school, gathered from girls and their family, added to the schools reliability in taking custody of girls in their growing age. The concern for safety and security of their girls cut across community line led to the selection of either a private school in the neighbourhood or a strict school at a distant location

Case 8 (Hawalbagh block):-

I met Meena Bisht, a 17 year-old class 11 student of a English medium private school; she has a younger brother Vijay of 13 years, a student of class 8 studying in the same school. The school is situated around 3 km away and so her father drops and brings both of them in his motor bike. Her father is a rich business man (studied up to 10th standard) and owner of famous sweet shop. Her mother is a homemaker who studied up to 8th class. While talking it was mentioned that she is an adopted daughter, they are not biological parents; she was only three years when they adopted her and brother Vijay was born the following year. Her parents are extremely nice and not biased at all; they love her more than Vijay and always fulfill her necessary desires as per her demands. They want that Meena must study up to the level she wishes. After finishing her studies in school, Meena wants to join Hotel Management course in Almora town. Her main intention is to enlarge her father's business and open a modern hotel. According to her these days tourism sector is much popular in the Hills; however, number of hotels with modern facilities is few, particularly in rural areas.

Case 9 (Tarikhet block):-

Ananya Negi, a student of class 6 (12 years old) and her 7 year-old younger brother (reads in class 2) are students of a local Government High School. On enquiring about her family, it was found that her father is an employee in a Government office and is in a transferable job. Earlier they lived in Rudrapur but recently (six months ago) have come here. In Rudrapur she studied in an English medium school but in her village there is no such good English medium school, thus they were forced to take admission in this school. Since the school is close to their house her mother drops them to school regularly. Ananya said swinging her head and turning her eyes that "We are very small, so our mother comes with us". She also said that her father promised them that in the following year they will be surely admitted to a good English medium school which is a bit far from their village. Ananya is pretty and sincere girl; she added that she loves to study and at the same time much fond of dancing. Dance is her passion. In her previous school in Rudrapur, she performed a dance programme in the Annual Day when all teachers, parents and students appreciated. She said that on completion of her intermediate study she would take a course in Dance and like to become a traditional dancer. Her mother said that she will give support in her capacity.

Case 10 (Tarikhet block):-

After completing the day's interviewing schedule while standing near a bus stop, I observed a Maruti van stop and a 16 years old girl student, wearing a clean school uniform and proper school shoes come out. She then went straight

in to a stationery/book shop and purchased two note books and a bundle of large white glazed paper. I introduced myself and wanted to know about her. She also gave her introduction; her name is Sunita Salal (studies in class 10). She is the only daughter with two brothers; one is elder, a student of class 12 and the younger, a student of class 6. They all study in the same English medium private school (co-educational) which is situated around 12 km from their home. Since her father is a retired army personnel, he drops them daily in their own vehicle. Some children of same school and of their own village, also join them in their vehicle. She said her father is very serious about her study and gives all support whatsoever she demands. She showed me that she purchased the glazed sheets for preparing a Science Project which needs to be submitted within two days.

She admired her father and told me that her father stopped the van to purchase stationery items although they have been quite late and have to travel another 10 km from here; they will reach home late when the sun has set. When asked why they did not take admission in a nearby school then she replied “My father will never compromise with quality of studies and discipline in school; it is his children’s future and hence admitted us in a good school. My father is so concerned about my studies that he participates regularly in Parent-Teacher meetings. He discusses regularly with our Principal on minor issues related to my study”.

She also indicated that after completing her intermediate she will write the entrance test for joining an Engineering College; she is interested to join the Engineering College in Dwarahat and become a Civil Engineer. According to her these days there is frequent occurrence of disasters and extreme events, like earthquakes, flash floods, cloud bursts, heavy rainfall, hailstorms, etc. She is of the opinion that by obtaining a degree in Civil Engineering she would be able to assist in solving these problem with her department. She would also help in construction of earthquake proof buildings and dams, good roads and bridges. This would help in reducing disaster and improving safety, and consequently better the living conditions of the hill people of Uttarakhand.



Figure 1: A panoramic view of a village and hilly terrain in Lamgarah block.



Figure 2: A panoramic view of some villages close to village Panuanaula, Dhaula Devi block; nicely terraced land for cultivation is seen in the foreground.



Figure 3: A panoramic view of some villages in Tarikhet block.



Figure 4: Students and a mother (teacher) returning home after attending a private school located in Gwalakot village, Hawalbagh block.

Conclusions:-

This study mainly focused on the objective of school choice among a set of different income-group families from four development blocks in rural areas of Almora district; while discussing the process of school choice one must take into account social and economical aspect and cultural discourses which in all manner of ways structures experiences of people located in a particular setting. In all these accounts it is the overarching concerns of costs, discipline and quality through which a deeply gendered choice is exercised by these parents. In this study, costs of schooling did differentiate on the decisions but it varied within a family and between son and daughters (Lamgarah block). Moreover, in some case there was no difference made between son and daughter (Hawalbagh block).

During interviews a significant and quick change in the people's attitude (including parents, girls, villagers, teachers etc.) was observed in the rural area in Almora, and this could be noticeable throughout the whole interviewed sequence. The socio-educational profile of school going girl children indicates that a remarkable changing scenario is going through education and now the confident girl children in different age groups with different economical category are the product of different educational effort.

The present study also attempts to profile the educational and sociological status of the girls in rural Almora District. Relevant information has been gathered from the respondents, particularly on their educational condition, choice of schooling, cause of choice schooling, and from other stakeholders like parents, Gram-Pradhans, teachers and school officials. The data for the study was collected through in-depth interview schedule. It was collection of questions on socio-educational and demographic issues. The empirical data gathered demonstrates that education has contributed significantly in raising the status of girl children in rural Almora. All the girls aged between 6 to 18 years were found to be enrolled in schools. The number of drop out students is negligible, indicating that at present the parents are aware about the importance of education. It can be also noted from the educational status of the parents that new generation of parents had greater levels of education than the older ones and was found to influence respondents' education, i.e., higher the education of the parents, higher was the educational level of the girl children. The parents formed the source of encouragement for higher education of their daughters. The nature of encouragement from parents is indicative of the high practical value they had for higher education which contrast with a number of stereo-typed notions about girls' education in rural areas. Although girl students studying in higher classes (10 and 12) have moderate percentage of illiterate parents, nevertheless, due to changing trend and importance of education such parents do encourage their daughters to get educated. Better income of parents was likely to place the children in a better educational position. Parents whose children were sent to a private school for acquiring quality education believed that their children would obtain quality education and give good performance, did not really consider the

school fees or the distance of the school (Hawalbagh & Tarikhet block). It must be mentioned that the parents with poor financial background were also found to influence the daughter's accessibility to education.

The present study also suggests that now a day there has been a trend among parents to send their children to private school. It also reveals that, according to parents, the quality of education and caring for students in private school are better than in government schools. Moreover, vehicle facilities are available in most private schools (in Hawalbagh and Tarikhet block). The expansive growth of private schools in the country due to fact that government schooling has not been able to provide quality education has also been reported in earlier studies (Nambissan, 2003; Balasundran, 2005). It was observed that majority of the girl children goes to school with neat and clean uniform, proper school bag, and maximum students carried drinking water. In addition, a number of the parents themselves escorted their children to school and brought them back to home (Figure 4).

In general, the parents were of the opinion that the quality of education in government schools is just satisfactory, and not comparable with the private schools, and this is reflected in unsatisfactory level of education standard of girls. The girl students in rural areas are quite simple, and unlike their city counterparts, some important facets were missing in them, particularly out-knowledge, exposure and communication with the modern world (in Dhaula Devi block). Further, more attention in studies need to be given by the girl children at home.

India has the second largest education system (World Bank, 2003) and largest number of children out of school in the age group 6-10 years (Huebler, 2007). But over the last few decades there has been considerable change in status of girl's education in hilly areas of Uttarakhand. The situation has improved considerably following the formation of Uttarakhand state since currently the drop out girl students of 6-17 years age form only a very small proportion, i.e. 2.9% in rural Almora and 0.5% in urban Almora (Annual Health Survey 2011-12. Fact sheet- Uttarakhand. <http://www.censusindia.gov.in>. Retrieved on 10th May, 2014). Hence, it can be concluded that a gradual change is occurring in rural areas and going to change further; the rural parents now want to give an opportunity to their daughters to live a hazard-free, open and enjoyable life.

The present study has depicted the 'role of education for all' changing the status of girl children in rural Almora. The status of girl children in rural areas needs to be examined further. Moreover, several challenges would come their way but the girls have to overcome them. They need to study seriously, improve their skills, and establish themselves in the competitive world. They need to first love, care and value themselves, increase their self esteem, then only can they understand the value of others; they should become good citizens and good human beings. Their aim must be not only to become a working and independent woman but also a caring and loving mother in their future.

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RESEARCH ARTICLE

DEVELOPMENT AND STANDARDIZATION OF MENTAL MATURITY SCALE FOR PRE-SERVICE STUDENTS.

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Key words:-

Tool construction, Standardization, mental maturity, pre-service teachers.

Abstract

The present study is designed to construct the mental maturity scale for pre-service teachers. The Mental maturity scale was developed and standardized by administering it on 55 randomly selected pre-service teachers of Madurai district and conducting item analysis to eliminate the inconsistent items in a tool. The final form of mental maturity scale consists of 42 items classified into 7 sections namely mental health, self-awareness, problem solving, cognitive self management, multiple intelligence, social attitude and emotional maturity. All the items of mental maturity scale are Likert type with five point rating.

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Introduction:-

Maturity implies putting away of childish things and regarding oneself as an adult ready to shoulder responsibilities that develop upon one in general in worldly affairs. It is related to the kind of personality one has developed. It is also concerned with the extent to which one is able to think act for oneself. A mature person is highly socialized and independent. He has built up a set of values for himself which guide his actions and behavior consistently. Emotional stability, intensity, social adjustment, professional interests and life aspirations go on increasing with the process of attaining maturity. A mature person is expected to understand things of importance and be aware of his rights and duties. A mature person does not act impulsively and does not waste time in the world of his own imagination.

Mental maturity is generally believed that a person with average intelligence acquires mental maturity between 13 and 15 years of age and a brilliant one gets it within 16 and 25 years of age. By the term mental maturity we mean mature ideas. The criteria of mental maturity are:

- The power of independent judgement without being guided by one's caste, class, traditions, or some influential person.
- Readiness to fulfill one's responsibility and duty.
- To analyse a situation objectively without being influenced by one's sentiments.
- To reach a right decision when confronted with complex situations.
- To decide issues with a spirit of give and take and not to overlook future consequences when overpowered by emotions.

The ultimate aim of teacher education is to prepare effective teachers who are capable of bringing about the desired behavioural changes in pupils. The quality of teaching is determined by the quality of teaching learning process

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made in the classroom situations. So the pre-service teachers must improve mental maturity. Then only they can improve the quality of school education. So the investigator decided to construct and standardize the mental maturity scale for pre-service teachers.

Objectives:-

- To construct the mental maturity scale for Pre-service teachers.
- To standardize the mental maturity scale for Pre-service teachers.

The Mental maturity scale was constructed to measure the skill of mental maturity of Pre-service teachers. The steps followed for its construction and standardization are as follows:

1. Planning
2. Preparation of Preliminary form
3. Pre-tryout
4. Editing
5. Pilot study
6. Item Analysis
7. Preparation of final form

Planning:-

During planning it was decided to prepare the statements with reference to mental health, self-awareness, problem solving, cognitive self management, multiple intelligence, social attitude and emotional maturity.

Preparation of Preliminary form:-

The investigator developed the preliminary form of mental maturity scale for pre-service teachers with 70 simple, clear and concise statements for better understanding. Care was taken to avoid ambiguity and repetition in the statements. At the end of each statement, five graded options were given namely. "Strongly agree", "Agree", "Undecided", "Disagree" and "Strongly disagree" having scores 5,4,3,2 and 1 for positive statements and 1,2,3,4 and 5 for negative statements. The investigator classified the statements of preliminary form of mental maturity scale under different sections namely mental health, self-awareness, problem solving, cognitive self management, multiple intelligence, social attitude and emotional maturity.

Pre-tryout:-

The Preliminary form of mental maturity scale for pre-service teachers was given to 10 teacher educators for their observation and criticism regarding the clarity of statements, appropriateness of the language and the pattern against each statement. Based on their suggestions, the developed research tools were further edited.

Editing:-

Soliciting the responses of the teacher educators approached at the pre-tryout stage, the editing of the developed research tool was completed. On the basis of criticisms and suggestions of the experts involved, 5 statements were rejected and 65 statements were retained.

Pilot study:-

For the standardization of the constructed tool, the investigator conducted a pilot study. The constructed tool was administered on 55 randomly selected pre-service teachers studying in DIET, Madurai district. The pre-service teachers were instructed to mark their responses for all the statements of mental maturity scale. Further, they were given assurances that their responses would be used only for research purpose. They were also convinced that their responses would be kept confidential. There was no time limit, but the pre-service teachers took 40 to 50 minutes for giving responses to all the statements of the tool. The responses of pre-service teachers were scored according to the positive or negative nature of statements.

Item Analysis:-

The investigator used item whole correlation to find out 'r' values. The item having 'r' values between 0.35 and 0.65 were retained and the other items were rejected. In the final mental maturity scale, 42 items were retained with positive (32) and negative (10) items.

Table 1:- Item whole correlation for mental maturity scale.

Item. No.	r' Value	Remarks	Item number in mental maturity scale
1	0.514170691	Retained	1
2	0.314755875	Rejected	
3	0.286770536	Rejected	
4	0.050745996	Rejected	
5	0.470296511	Retained	2
6	0.425550036	Retained	3
7	0.031395406	Rejected	
8	0.651537975	Retained	4
9	0.407366878	Retained	5
10	0.534555892	Retained	6
11	0.29546716	Rejected	
12	0.230880957	Rejected	
13	0.403585573	Retained	7
14	0.593752854	Retained	8
15	0.374328076	Retained	9
16	0.057879103	Rejected	
17	0.071226087	Rejected	
18	0.565367422	Retained	10
19	0.427614185	Retained	11
20	0.565141707	Retained	12
21	0.448991621	Retained	13
22	0.405815439	Retained	14
23	0.398733473	Retained	15
24	0.405408639	Retained	16
25	0.486430903	Retained	17
26	0.465951917	Retained	18
27	0.466950226	Retained	19
28	0.526462766	Retained	20
29	0.205382954	Rejected	
30	0.592337104	Retained	21
31	0.227275458	Rejected	
32	0.327493852	Rejected	
33	0.345958714	Rejected	
34	0.405605684	Retained	22
35	0.389239412	Retained	23
36	0.326807346	Rejected	
37	0.402965297	Retained	24
38	0.41060083	Retained	25
39	0.490937768	Retained	26
40	0.402597389	Retained	27
41	0.38383348	Retained	28
42	0.453662757	Retained	29
43	0.632777954	Retained	30
44	0.44091366	Retained	31
45	0.630455047	Retained	32
46	0.522314852	Retained	33
47	0.232215771	Rejected	
48	0.110913755	Rejected	
49	0.245486829	Rejected	
50	0.553845456	Retained	34
51	-0.077709067	Rejected	
52	0.149146801	Rejected	

53	-0.181720204	Rejected	
54	0.357098688	Retained	35
55	0.627565969	Retained	36
56	0.595588646	Retained	37
57	-0.00274471	Rejected	
58	0.179960345	Rejected	
59	0.41938927	Retained	38
60	0.52725274	Retained	39
61	-0.079612746	Rejected	
62	0.389781483	Retained	40
63	0.381927493	Retained	41
64	0.285229562	Rejected	
65	0.397709655	Retained	42

Table 2:- Section-wise Distribution of Items of Mental maturity Scale.

Sl. No.	Dimensions	Item number in mental maturity scale	Positive items	Negative items	Total number of items
1.	Mental health	1-8	1,5,6	8	4
2.	Self-awareness	9-18	9,10,13,14,15	18	6
3	Problem solving	19-23	19,20,21,22	23	5
4.	Cognitive self-management	24-29	24,25,27,28	26	5
5.	Multiple intelligence	30-38	30,34,37,38	35	5
6.	Social attitude	39-54	39,40,41,42,43,44,45,	46,50,54	10
7.	Emotional maturity	55-65	55,56,59,60, 65	62,63	7

Preparation of the final form:-

After item analysis the final form of mental maturity scale for pre-service teachers was prepared with 42 retained items.

Reliability and validity of the tool:-

Split-half reliability coefficient of mental maturity scale was determined on the basis of scores of 55 randomly selected pre-service teachers. In the split-half method, the tool was divided into two equivalent halves and for these two halves half-test reliability coefficient was found using Karl Pearson's coefficient correlation formula. From the half-test reliability coefficient, whole-test reliability coefficient was estimated by using Spearman Brown Prophecy formula.

Whole-test reliability coefficient, Split half-test and Guttman Split-Half Coefficient of mental maturity scale for pre-service teachers were 0.797, 0.754 and 0.744 respectively. Hence the developed tool was reliable.

On the basis of opinions of experts the items of mental maturity scale were structured. Hence the developed tool has face validity and content validity.

Conclusion:-

The education of the students largely depends on their teachers. It is important that pre-service teachers who are the future teachers of the country must possess the skill of mental maturity. This will inculcate the pre-service teachers to be efficient. The great teacher should be necessarily equipped with knowledge, skills and competencies. The teacher should be properly enlightened, educated through proper training and develop sense of responsibility and love towards his profession and with rapid expansion of primary education both in number and extent, the teacher must come out of teacher educational institutions with great perfection in teaching competencies and allied aspects.

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RESEARCH ARTICLE

ASSESSMENT OF PUBLIC SATISFACTION AND TREATMENT TOWARD PHC AND PUBLIC PRIVATE HOSPITAL IN EASTERN PROVINCE, KSA.

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Abstract

Objectives:-

1. To determine the compliance and satisfaction to doctor's prescription among general population in Eastern Province of Saudi Arabia by type of health facility used, age, gender, and education.
2. To determine the reasons for non-compliance, and opinions on how to improve compliance.

Materials and Methods:-

Cross-sectional study conducted for public in eastern province. Data Collected using web-based self-administered questionnaire to fill online through newspaper, and social media like Facebook, Twitter, etc. for 6 month and periodically repeated throughout the study period, SPSS 17.0 used for data analysis. Chi-square or Fisher's Exact test used to test for statistical significance by gender, age, and education.

Background:- Compliance in healthcare is defined as the extent to which a patient's behavior coincides with the healthcare provider's recommendations for health and medical advice, we conduct unique study to involve the public instead of narrow spectrum of patient and diseases.

Results:- Total of 587 participants 62.9% were Satisfied during last visit to their physician. 32.2% stop taking medication if they feel better, 31.7% always committed to treatment. The reasons of non-compliance with medication mainly 35.6% due to large number of drugs, 28.3% due to Inappropriateness of drug, 20.8% do not Know the function of drug. the more acuteness of illness more noncompliance, avoiding reading the patient file and lack of knowledge. More than half of participant said doctor misbehavior affect their trust about prescription. Private hospital showed to have more regular visit and satisfaction and governmental hospital showed less satisfaction. 58.8% said that nearby health centers would not affect their compliance. 78% of the Health Centers lack communication with patient outside health facility. The best option to overcome non- compliance showed to be follow-up by social media.

Conclusion:- Males and people at age of 30 are more compliance to drugs and more satisfied. Side effect and accumulated drugs are common cause of non-compliance, best option to overcome non-compliance are follow up by social media and explain side effects of drugs , people less satisfied with governmental hospital and more compliance to PHC so high qualified health system, patient -centered relationship, health education are needed.

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..... **Introduction:-**

Compliance in healthcare is defined as the extent to which a patient's behavior (in terms of taking medication, executing the lifestyle changes, undergoing medical tests or keeping appointments with the physicians) coincides with the healthcare provider's recommendations for health and medical advice. [1] Non-compliant patients are those whose health-seeking or maintenance behaviors lack congruence with the recommendations prescribed by a healthcare provider. [2]

Patient non-compliance is a serious healthcare concern that poses a great challenge to the successful delivery of healthcare. This is widespread and has been reported from all over the world.

Previous research of specific communication variables indicates that patient-centered encounters result in: (1) the duration of the office visit remaining the same [3], [4] (2) better patient satisfaction, [5] (3) higher physician satisfaction, and (4) fewer malpractice complaints.

Medication management outcomes can be improved by adopting more client-centered approaches. To examine the implications of a client-centered relationship this reviews regarding client involvement in: (1) identifying treatment goals; (2) choosing from regimen options; (3) monitoring symptoms and evaluating regimens; and (4) self care with nonprescription pharmaceutical products. , A compliance study conducted in Saudi Arabia for those on short-term medication found 67.8% compliance.

However, compliance of patients tends to decrease with time being lower in patients on long-term medication than in those on short-term medication. [6] Another study done in Saudi Arabia found an overall 65.8% non-compliance in patients suffering from hypertension. [7] The noncompliance to long-term therapy severely compromises the effectiveness of treatment and adversely affects the patient's condition. [8]

Materials and Methods:-

Study population and area:- Population of Eastern Province.

Study design:- Cross-sectional study

Study period:- Six months until 12-1- 1438 H

Data Collection:- Data Collected using web-based self-administered questionnaire. The information collected included demographic data, type of health facility visited, satisfaction with last visit to physician, compliance to doctor's prescription, use of medicine without doctor's prescription, questions related to reasons for non-compliance, and suggestions to improve compliance to doctor's prescription.

The public in the study area was informed about this survey (along with the web link) and requested to fill the online questionnaire through newspaper, and social media like Facebook, WhatsApp, Twitter, etc. This was periodically repeated throughout the study period

Data Analysis:- SPSS 17.0 used for data analysis. Descriptive statistics included frequency and percentages for categorical variables. Chi-square or Fisher's Exact test, as appropriate, used to test for statistical significance by gender, age, and education. The sample size using Epi info version 3 with confident level 97% is 471. The results were expressed as mean values \pm SD. A P value of < 0.05 was considered significant.

Results:-

Total of 587 participated in this study, 79.5% of the participants were from the Alhasa and Dammam and Qatif. Majority of participants their age Found to be between 21-30 years (SD=1.11) and about two third of them were females (74.7%). All the participants were educated (100%, n = 587).

The demographic characteristics of the participants are summarized in Table 1 and Geographic distribution in figure 1

Table 1:- Demographic characteristics of the study population.

Characteristic	Number	Percentage
Gender		
Male	148	25.2
Female	439	74.8
Age (Years)		
≤ 20	126	21.5
21-30	259	44.2
31-40	103	17.6
> 40	99	16.9
Education		
High school or less	126	21.4
College or higher	461	78.5
Nationality		
Saudi	574	97.8
Non-Saudi	13	2.2

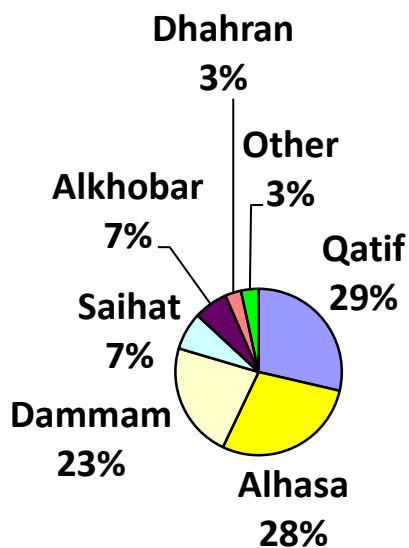
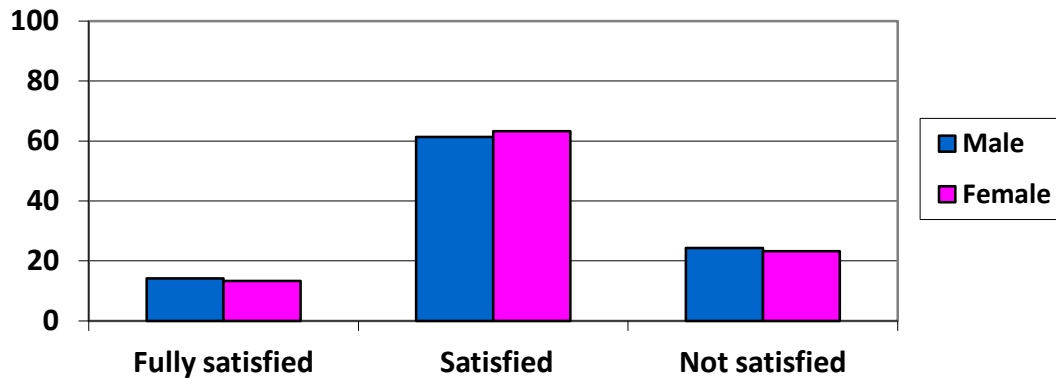


Figure 1 :Geographic distribution

According to geographic distribution as shown in figure 1, 29% of participants from Qatif , 28% from Alhasa , 23% from Dammam , 7% from Saihat , 7% from Alkhobar , 3% from Dhahran , 3% others.

Figure 2:Satisfaction during last visit to the doctor by gender



According to satisfaction during last visit to doctor by gender as shown in figure 2, 14.18% of male and 13.34% of female are fully satisfied while 61.4 of male and 63.3 of female are satisfied and 24.32% of male and 23.23% of female are not satisfied.

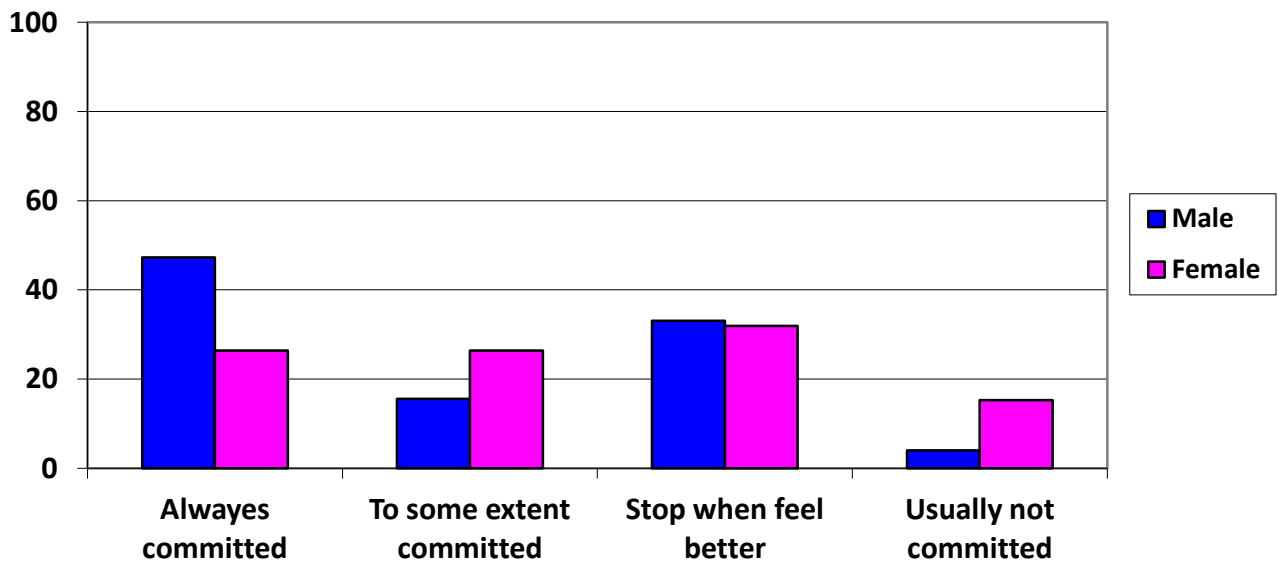


Figure 3:- percentages of compliance to drugs the doctor prescribed by gender.

According to compliance to drugs the doctor prescribed by gender as shown in figure 3, males more committed 47.29% than females 26.4% and 15.54% of males and 26.4% of females said they committed to some extent and 33.1% of males and 31.89% of females said they stop taking medication when they feel better and females more not committed 15.26% compared to males 4%, this comparison chart are statistically significant ($p < .001$).

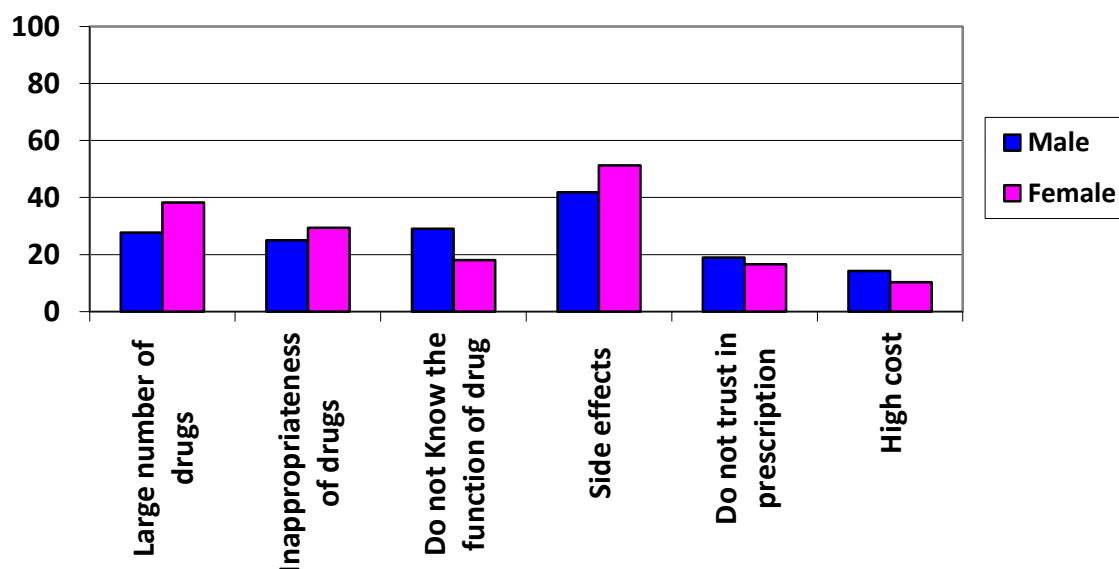
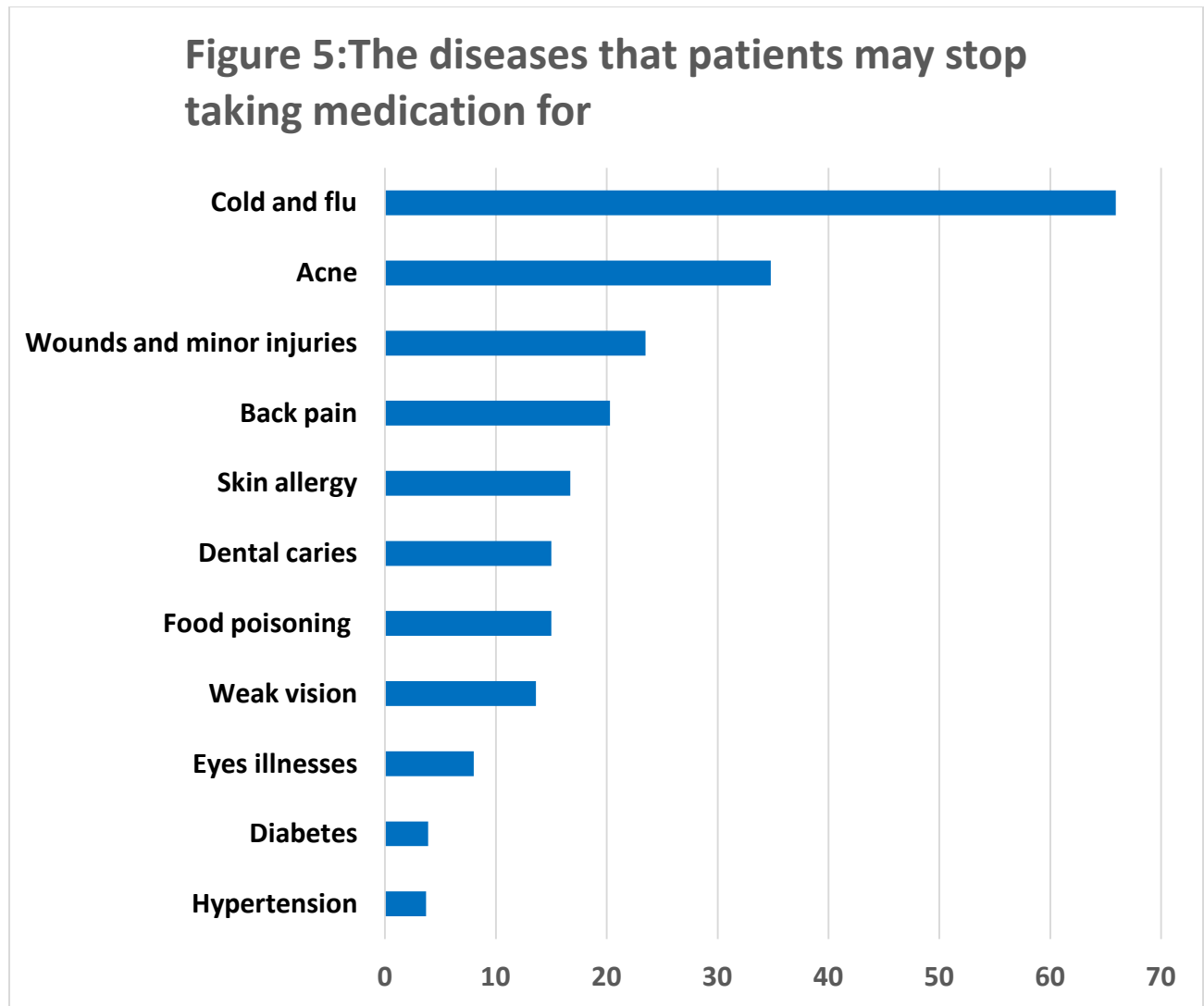


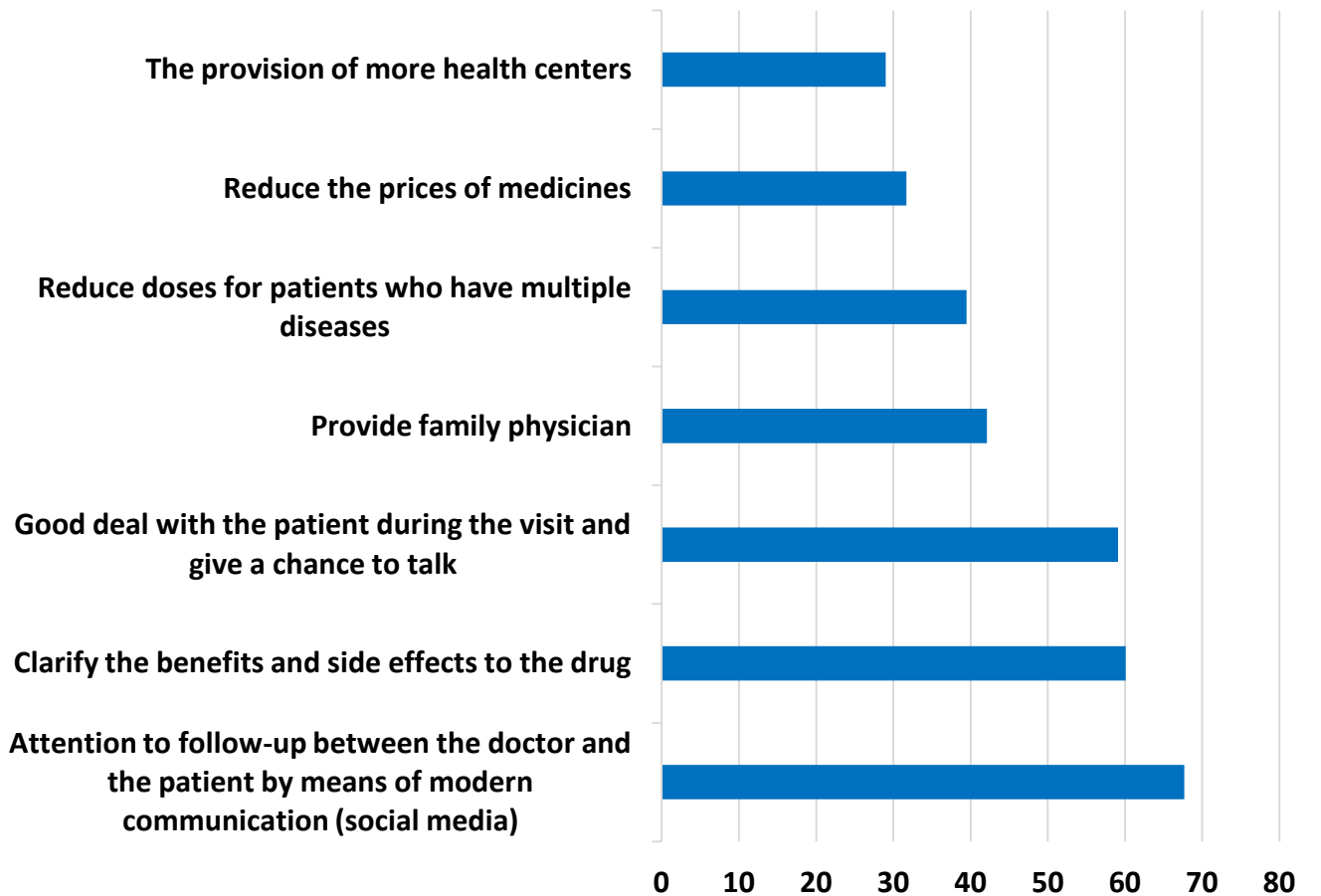
Figure 4:- The opinion of participants of what are the reasons which may lead to failure in compliance with medication by gender.

According to opinion of participant of what are the reasons which may lead to failure in compliance with medication showed that females are more non-compliance to treatment due to large number of drugs 38.3% compare to males 27.7% ($p < .012$) also due to inappropriateness of drugs 29.4% compared to males 25% , 29.1 % of males said that lack of knowledge about function of drugs contribute to their non-compliance to drugs compared to 18% of females ($p < .04$), side effects contribute to non-compliance in 41.9% of males compared to 51% of females ($p < .03$), distrust of doctor prescription contribute to non-compliance in 18.9% of males compared to 16.6% of females , high cost of drugs contribute to non-compliance in 14.2% of males and 10.3% in females.

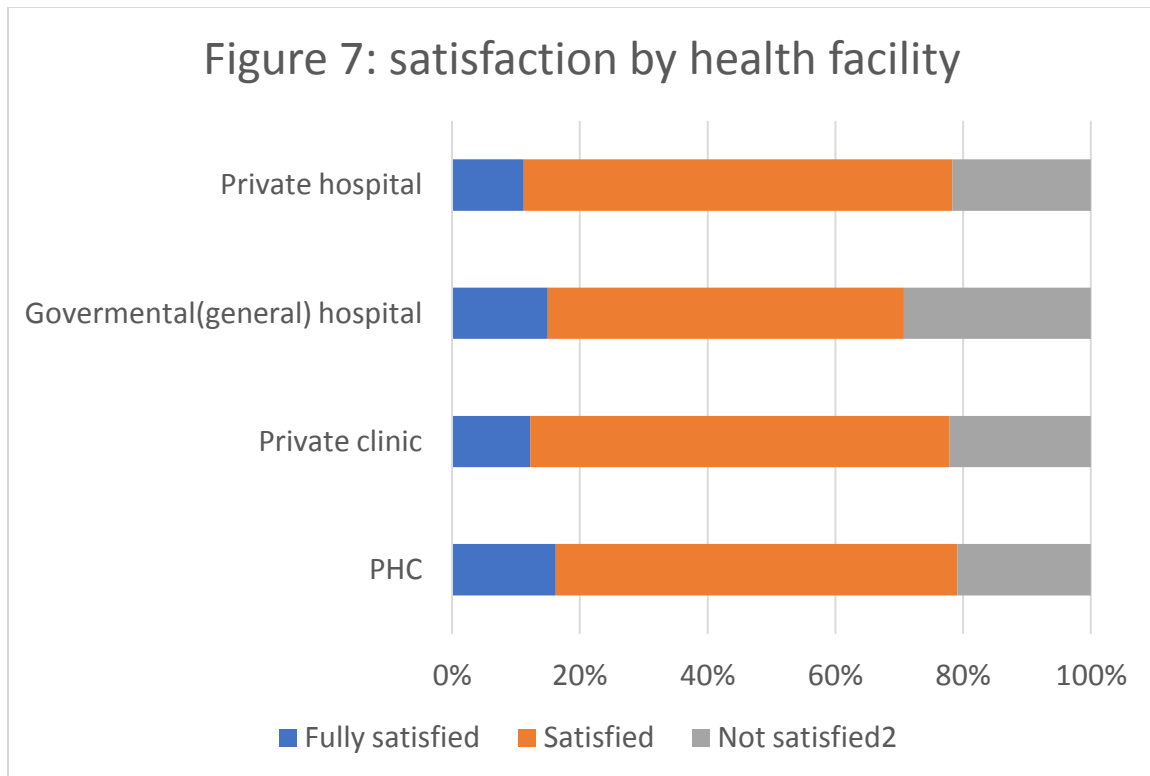


According to above figure 5 showed that 65.9% of participants said that cold and flu in the top of diseases that they left treatment for followed by Acne 34.8 % ($p < .002$), wounds and minor injuries 23.5%, back pain 20.3%, skin allergy 16.7%, dental caries 15%, food poisoning 15%, weak vision 13.6% ($p < .029$), eye illnesses 8%, diabetes 3.9%, hypertension 3.7%..

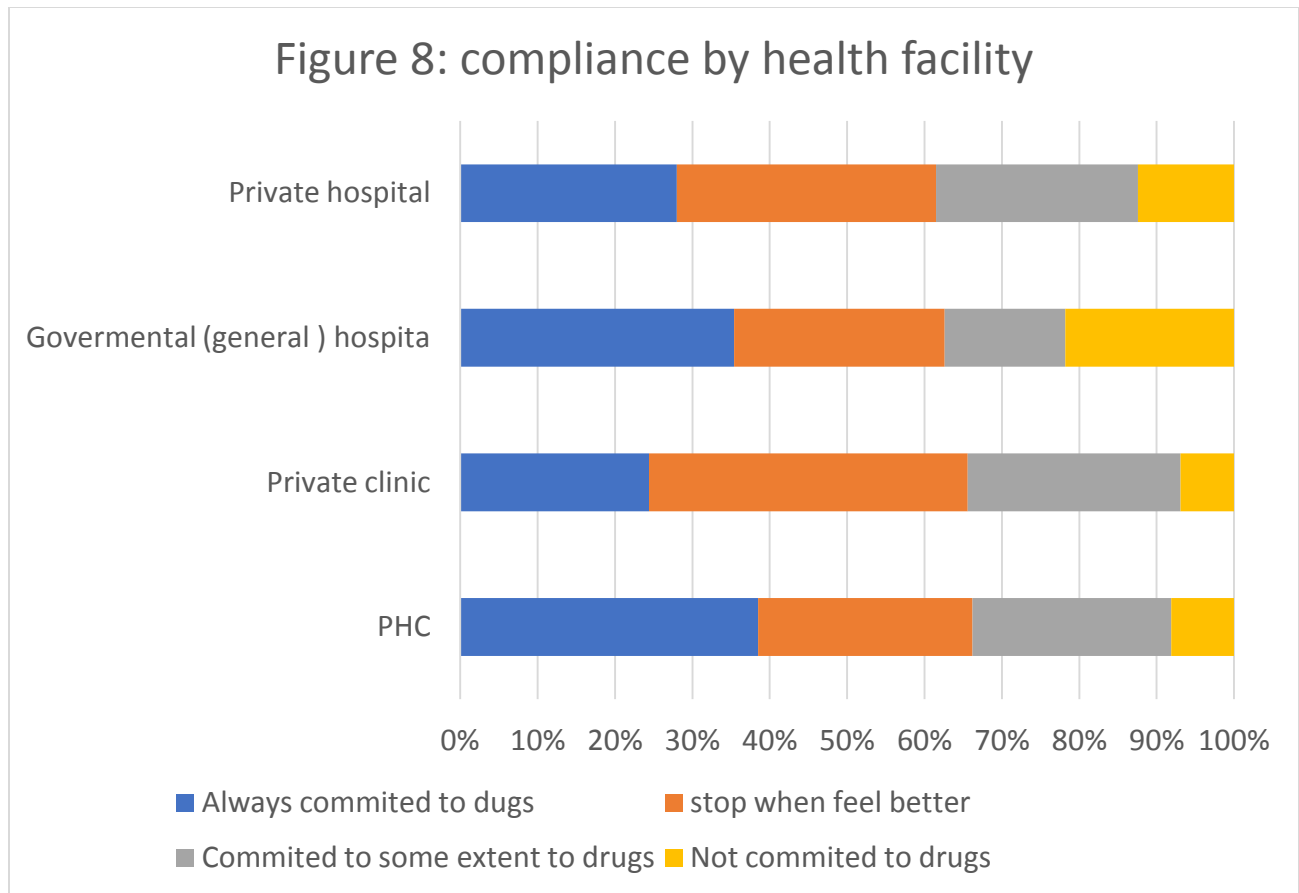
Figure 6:patients' opinion regarding the possible solutions to overcome the non-compliance with medication



According to patients' opinion regarding the possible solutions to overcome the non-compliance with medication in figure 6 showed to be follow up by social media 67.6% followed by clarification of side effects 60.1%, give chance to talk 59.1%($p<.01$, more in males), provide more family physician 42.1% , reduce the number of doses who have multiple diseases 39.5%($p<.001$, more in females) , reduce the prices of drugs 31.7%($p<.04$, more in females), provide more health centers 29%.



According to satisfaction by health facility in figure 7 showed that the most fully satisfied health facility is PHC (primary health care center) 16.2% followed by governmental hospital 15% ,private clinic 12.2% , private hospital 11.2% and the most satisfied health facility is private hospital 67.1% followed by private clinic 65.6%, PHC 62.8% , governmental hospital 55.8%, the most not satisfied health facility is governmental hospital followed by private clinic 22.1%,private hospital 21.7%, PHC 20.9%.



According to compliance by health facility as shown in figure 8, participants are more compliance to PHC 38.5% followed by governmental hospital 35.4%, private hospital 28%, private clinic 24.4 and participants stop taking medication prescribed by doctor more in private clinic 41.2% followed by private hospital 33.5%, PHC 27.7%, governmental hospital 27.5% and participants committed to some extent to drugs more in PHC 25.7% followed by private clinic 27.5%, private hospital 26.1%. governmental hospital 15.6% and the health facility that have high rate of noncompliance to drugs found to be governmental hospital 21.8% followed by private hospital 12.4%, PHC 8.1%, private clinic 6.9%, this comparison chart are statistically significant ($p < .001$).

According to compliance by age we found that people more than 30 years old are the age group with more compliance rate compared to other age groups ($p < .001$)

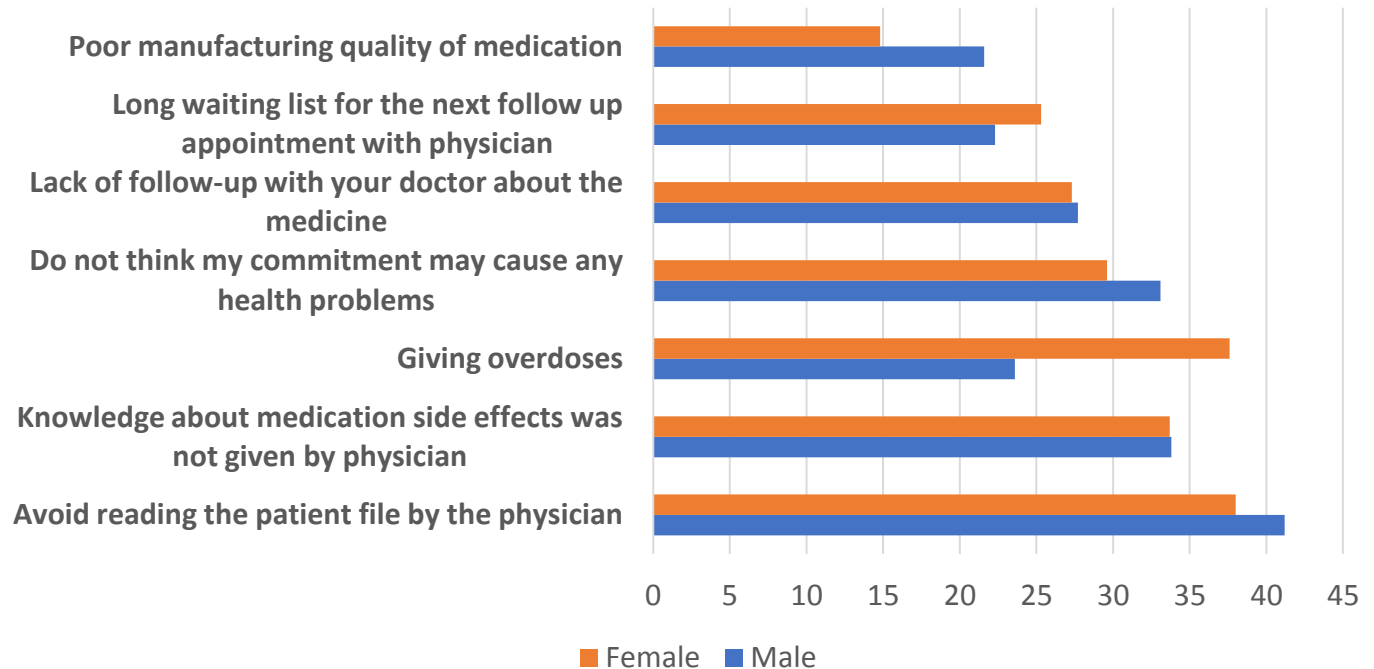
Also that particular age group had the higher rate of satisfaction compared to other age groups ($p < .002$)

Table 2:- Participants perception toward compliance to medication.

	Number	Percentage	P value
1- Do you think that the mishandling of the doctor may be a reason not to trust the physician medical advice and leave taking medication?			
Yes	302	51.4%	<.01*
No	55	11.2%	
To some extent	219	37.3%	
2- Which of the following age groups noticed his lack of medication compliance or rejected?			
Children	268	45.7%	<.1
Teens	258	44%	
Adults	221	37.6%	
Elderly	235	40%	
3- lack of medication compliance may worsen my health status?			
Yes	245	41.7%	<.02*
No	62	10.6%	
To some extent	280	47.7%	
4- I think that my commitment to the drug may bring me and other health problems?			
Yes	212	36.1%	<.3
No	375	63.9%	
5- Which of the following health centers commitment to taking the medication out more?			
PHC	78	13.3%	<.09
Government	198	33.7%	
Special	115	19.6%	
Teaching	36	6.1%	
Private Clinic	160	27.3%	
6- Which health facility you visit usually?			
PHC	148	25.2%	<.01* Male more in PHC
Private Clinic	131	22.3%	
Government	147	25%	
Special	161	27.4%	
7- What types of follow-up used by the health facility that you visit to complete your follow-up?			
Telephone	101	17.2%	<.001*
Email	11	1.9%	<.1
Doctor visit me	30	5.1%	<.3
Internet & New social media	52	8.9%	<.001*
No communication outside health facility	458	78%	<.001*

*: statistically significant

Figure 9 :The reasons that brings health problems despite adherence to medical prescription by gender



According to the reasons that brings health problems despite adherence to medical prescription as figure 9 showed, we emphasized significant value which is giving overdoses ($p < .002$, more in females) and poor manufacturing quality of medication ($p < .03$, more in males)

Table 3: Other related questions for drug compliance .

	Number	Percentage	P value
1- Do you buy drugs from pharmacy without doctor prescription?			
Yes	472	80.4%	<.005*
No	115	19.6%	
2 -The presence of health centers near the residential neighbourhood may reduce non-compliance to drugs?			
yes	99	16.9%	<.01*
no	345	58.8%	
to some extent	143	24.4%	
3- Lack of a commitment with medication are widespread in the city?			
Yes	245	41.7%	<.1
No	62	10.6%	
To some extent	280	47.7%	
*: statistically significant			

Discussion:-

Effects of Compliance: adherence is a very important issue in medical field for many causes so as we know that non-compliance to treatment imposes a considerable financial burden upon health care systems.

Doctor – patient Relationship:-

A paternalistic approach to be avoided because the doctor patient relationship, And communication and shared decision-making are important factors affecting compliance as shown in our survey that %51.4(n=302) will not committed to medication if they face misbehavior from their physicians that will affect the relationship between them also Knowing each patient's health beliefs and physician plan are the key feature of the new doctor patient encounter.

Doctor-patient decision Making:-

We have to discuss the treatment plan where both patient and doctor can adhere, as we know a lot of people in KSA use new social media in communication recently so as we found according to survey that the best option to resolve a medical non- compliance regarding participant opinions showed Good following up between doctor and patient by mean of modern social media 67.6% (n= 397) followed by Explain the benefits and the side effects of medications to patients 60.1% (n= 353), Good behavior between doctor and patient and give the patient chance to talk 59.1% (n= 347) and other causes as mentioned in results, so as we can see the great role of patient in making decision about their treatment the thing that will decrease patient non-compliance , we need to review our style of communication and provide special office only for patient relationship and communication outside health facility because as we see in the result that more than 70% of health facilities lack these services .

Satisfaction:-

We need to study "seeing the same physician" as in case of family physician and its influence on patient satisfaction, that reflect the need of family physician in our health system as shown in our study that one of the most important thing to overcome noncompliance is Provide more family physicians 42.1% where the patients can follow again and again the same physician to complete their health issues and get maximum benefits.

Role of neighbor PHC:-

According to survey showed that %58.8 said that availability of neighbor primary health center have no effectiveness in decrease non- compliance , and we conduct study to show which health care facility have more rate of compliance and the result showed that %33.7 which mean more than 1/3 of participants are more compliant toward governmental hospital followed by private clinic(%27.3) , private hospital (%19.6), PHC(%13.3) , teaching hospital , as we can see governmental hospital goes with highest rate of compliance but we recommended other study to be conducted to see what factors affect compliance in these health facilities.

History of PHC:-

There was study that have explanation on why PHC it occupies middle place in patient adherence, By the year 1987 the Ministry of Health in Saudi Arabia had established 1477 Primary Health Care centers all over the kingdom, Saudi study done 3, June 1993 [9] was to assess the satisfaction of patients with different aspects of Primary Health Care services in Riyadh, the results showed that the patients are generally moderately satisfied with the services. They are most satisfied with the effectiveness and humaneness aspects of care.

Compliance of general population:-

Regarding compliance to medication of general population , many research done in KSA but did not mentioned the compliance of general population and most of their research confined to certain group of patient with certain disease , so in our study we have conducted a study to involve the public and we found that most of responses 32.2% said they stop taking medication if they felt better , while 31.7% said they always committed to the drugs that prescribed by the doctor, 23.7% said Sometimes , 12.4% will never committed to the drugs that prescribed by the doctor, so here we can see that more than 1/3 of participants stop taking medication if they feel better that indicate poor knowledge about side effect of some drugs like antibiotics and the risk of development of bacterial resistance as result of that ,as we consider our participants highly educated despite we found higher rate of non-compliance to treatment the thing that imposes serious attention. According to our study %65.5 stop taking medication in flu and cold (that sometimes may need antibiotic prescribed by GP).

Relationship between education level and compliance:-

As we mentioned before that higher educational levels of participants were found to be %32.2 stop when they feel better, Several studies have found association between higher education level and compliance,[10] while some studies have found no such association, A study conducted in the UK has shown that patients with a lower level of education have better compliance.[11,12] It may be presumed that patients with a lower educational level may have more trust in the physician's advice. However, these results show that education may not be a good predictor of therapeutic compliance.

Causes of non-compliance among general population of eastern province:-

Our study and other studies shown many causes of non-compliance, in our study we found many causes e.g., the large number of drugs that must be taken, Inappropriateness of medication, lack of knowledge about the function of drugs, Side effects, lack of trust in doctor prescription, high cost of medications and other studies conducted in KSA shown other causes [13], Those patients who did not get adequate information on what to do in the event of their missing a dose, or if they experienced any side effects of the medicine, were more non-compliant.

Numerous researches involving:-

Various diseases have evaluated the effect of the patient- physician relationship on patients' compliance, and has found it to be another strong factor in favor of patient compliance. [14-16], Compliance to treatment advice was good when the physicians were supportive, supplied vital information, and listened patiently to patients [17].

Suggested solutions to overcome non-compliance among general population of eastern province:

The best option to overcome medical non-compliance regarding participants opinions showed to be following up between doctor and patient by modern social media 67.6% (n= 397), Explain the benefits and the side effects of medications 60.1% (n= 353), Good behavior between doctor and patient and give the patient chance to talk 59.1% (n= 347), Provide more family physicians 42.1% (n= 247), Reduce the doses of patients who have a lot of diseases 39.5% (n= 232), Reduce the prices 31.7% (n= 186), provide more Health Centers 29% (n= 170).

Conclusion:-

The findings show that males and people at age of 30 more compliance to drugs and more satisfied, side effect and accumulated drugs are the common cause of non-compliance, best option to overcome non-compliance found to be follow up by social media and explain side effects of drugs, people less satisfied with governmental hospital and more compliance to PHC so there are a definite need for Hight qualified health system, patient-centered relationship, health education and to improve patient-doctor communication and health services.

Acknowledgments:-

Department of family and community medicine at abdurahman bin faisal university .

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RESEARCH ARTICLE

ROLE OF COMPUTED TOMOGRAPHY IN ACETABULAR FRACTURES.

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Abstract

In the 21st century, use of automobiles have not only revolutionized transport system across the globe, but have changed the lifestyle of human beings. The chief drawback of this gift is increase in cases of accidents. Most of accidents associated with polytrauma involve pelvis. In pelvic injuries, acetabular fractures and fracture dislocations of hip joint are common. The radiological evaluation of hip and acetabulum has been revolutionized by development of computed tomography because of its fine definition of sectional anatomy and accurate evaluation of pelvic trauma. The study included 30 polytrauma cases, attending the Radiodiagnosis Department of Rajindra Hospital Patiala.

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Introduction:-

Acetabular fractures constitute between 20% and 25% of all pelvic fractures in adults. CT is the modality of choice for evaluating these injuries. Axial images should be performed with contiguous slices no thicker than 3mm. Sagittal and coronal reconstructions and three dimensional images are often helpful to conceptualize the fracture pattern before surgery (2). CT is helpful in delineating the extent and configuration of fractures of the acetabulum as the acetabular fractures and fracture dislocations of the hip joint are frequently complex and routine radiographs do not easily demonstrate the precise pathological anatomy. Intraarticular bony fragments, joint space incongruity and femoral head fractures may also be revealed by CT examination (3). Unconventional radiography, in addition to standard AP view of pelvis, 45 degree oblique view, iliac oblique view, inlet and outlet views are taken which provide only means whereby spatial relationship of bones of pelvic ring can be assessed (5). There is no significant difference between plain radiography and computed tomography in detection of fractures of the iliac wing, anterior pelvic column, posterior pelvic column and pubic rami. However computed tomography is more sensitive than plain radiography in detecting pelvic fractures involving sacrum, quadrilateral surface, acetabular roof, posterior acetabular lip and primary loose bone fragments (6). In present study, the significance of computed tomography in diagnostic evaluation and management of the pelvic trauma patients was assessed. The computed tomographic findings in diagnosis and management of acetabular fractures were also compared.

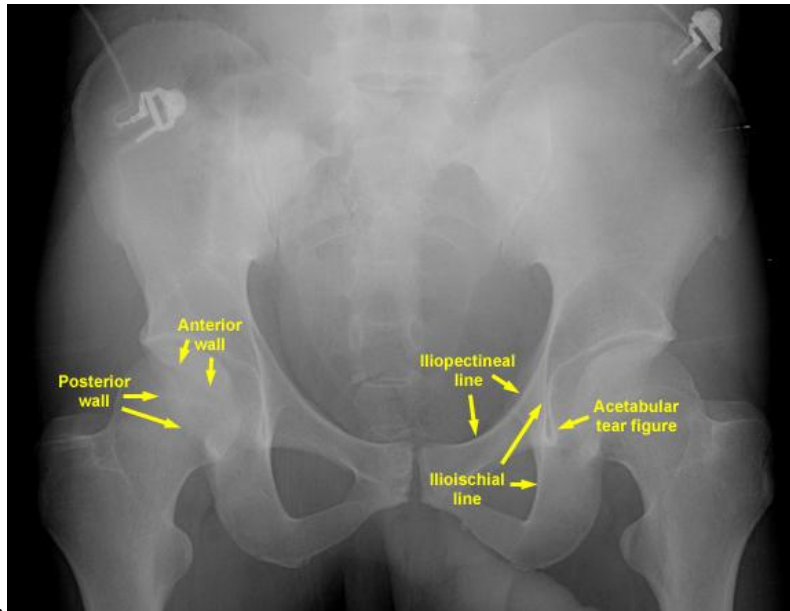


Figure 1:- Anteroposterior (AP) radiograph of the pelvis. The iliopectineal (or iliopectic) and ilioischial lines serve as landmarks for the anterior and posterior columns, respectively. The larger and more lateral posterior wall is visualized more easily than is the smaller, more medial anterior wall. The acetabular tear figure is a composite shadow of the inferomedial structures that compose the acetabulum. The ilioischial line should pass through the teardrop on a true AP view of the pelvis.

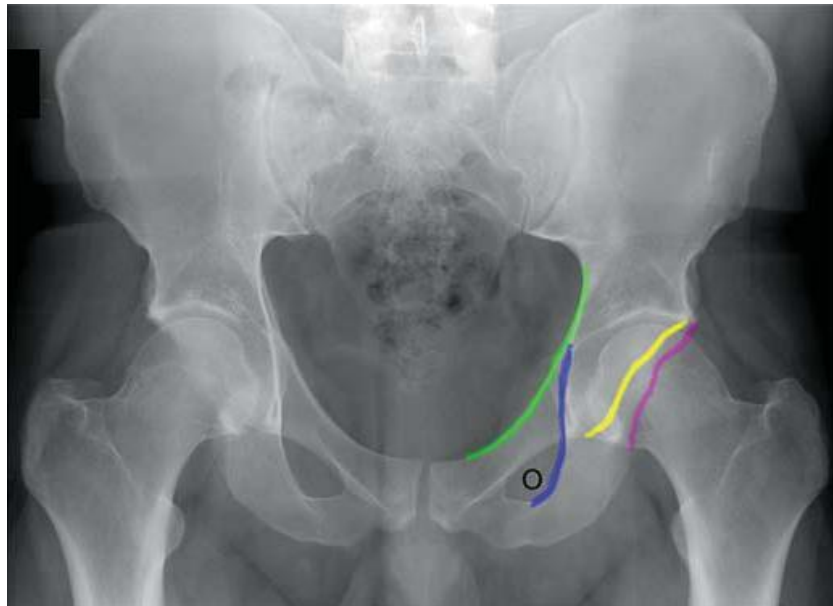


Figure 2:- Anteroposterior radiograph shows iliopectineal line (green), ilioischial line (blue), anterior acetabular wall (yellow), posterior acetabular wall (pink), and obturator foramen (O).

Classification Of Acetabular Fractures:-

The classification of acetabular fractures described by Letournel and Judet is most widely accepted (1,18). They divided acetabular fractures into two basic groups: elementary fractures and associated fractures.

Elementary Fractures comprise fractures in which a part or all of one column of acetabulum has been detached. There are five elementary forms:-

1. Fractures of posterior wall of acetabulum
2. Fractures of posterior column of acetabulum
3. Fractures of anterior wall of acetabulum
4. Fractures of anterior column of acetabulum
5. Transverse Fracture

Posterior column and posterior wall fractures account for nearly 30% of all acetabular fractures, most common and are often as a result of posterior hip dislocation.

Anterior column and anterior wall fractures account for 6 to 7% of acetabular fractures, result in a separation of the articular surface together with the corresponding segment of iliopectineal line and mostly a fragment of anterior column is separated from rest of the innominate bone.

Transverse Fractures:-

Account for 8% to 10% of acetabular fractures, are axially oriented, separate the innominate bone into superior and inferior fragments usually at the level of acetabular roof. They are often associated with central fracture dislocation.

Associated Fractures:-

Include at least two of the elementary fracture forms from above. There are five principal associations.

1. T-shaped fractures.
2. Fractures of the posterior column and posterior wall.
3. Transverse and posterior wall fractures.
4. Fractures of anterior column or anterior wall associated with a hemi transverse fracture posteriorly.
5. Fracture of both the columns.

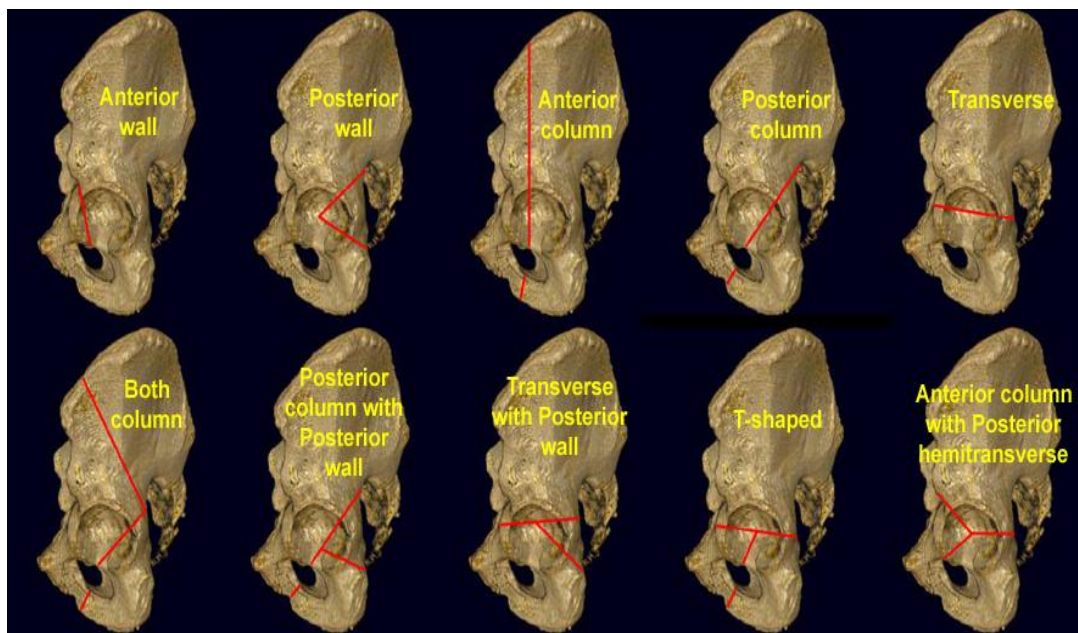


Figure 3:- Acetabular fracture classification system. Judet and colleagues (1964) described the classification scheme that is most commonly used today. Of the 10 types, 5 are elementary fractures (top row), and 5 are associated fractures (bottom row). Elementary types involve 1 primary fracture plane. Associated types involve more than 1 fracture plane

Materials and Methods:-

Study Design:-

Hospital based prospective correlative and comparative study. Source of data: 30 patients with clinically suspected or known hip fractures referred to department of Radiodiagnosis, Rajindra Hospital Patiala.

Protocol:-

A detailed history regarding mode of trauma and other sites of injuries besides pelvis were noted. Basic haematological investigations- Hb, TLC, DLC and other biochemical investigations like renal function tests, liver function tests were done if indicated. Conventional radiographic evaluation was done which included Antero-posterior views of pelvis including the area extending from iliac crest to the lesser trochanter of femur including both hip joints and sacroiliac joints. Oblique –lateral views of injured hip were done specially in cases in which dislocation of femoral head was suspected or evident on AP view. The plain radiographs were studied in detail and fracture lines were traced. Observations were recorded in the performa in all the cases. CT examination was done after initial radiographic

CT protocol:-

Patients were scanned with High Resolution Siemens Somatom Emotion, in supine position with 6mm to 8mm sections. 3D reconstructions were done from axial images using surface imaging technique in cases asked by the Orthopaedician, especially in patients with comminuted fractures. Clinical and imaging findings were recorded as per performa.

Radiographic Anatomy Of Acetabulum:-

Computed tomography has revolutionized diagnostic imaging of acetabular trauma as the three dimensional pictures of the fracture are vital for the diagnosis and treatment of these injuries. Crucial to interpretation of the axial computed tomography is an understanding of the normal cross sectional anatomy of acetabulum. The acetabulum can be described as an incomplete hemispherical socket with an inverted horseshoe shaped articular surface surrounding the non articular cotyloid fossa. The acetabulum is formed by anterior and posterior columns of bone which join in superior acetabular region (1,8,9). The anterior and posterior walls extend from each respective column and connect to axial Skeleton through a strut of bone called sciatic buttress. The anterior column represents larger portion which extends superiorly from superior pubic ramus into iliac wing. The posterior column extends superiorly from ischiopubic rami as ischium towards ilium. The anterior and posterior column of bone will unite to support the acetabulum. In turn sciatic buttress extends posteriorly from anterior and posterior column to become articular surface of sacroiliac joint which attaches column to axial skeleton. On radiograph, iliopectineal line represents border of anterior column and ilioischial line represents posterior column. The fractures which traverse the anterior column disrupt the iliopectineal line, whereas fractures which traverse the posterior column disrupt ilioischial line. The column concept is used in classification of these fractures and is central to the discussion of fracture patterns, operative approaches and internal fixation.

Observations:-

Distribution of patients according to age:-

The maximum number of patients were in the age group of 20-40 yrs. Only three patients included in the study were above 60 years of age

Distribution of patients according to sex:-

Males were seen to be frequently involved in pelvic trauma as compared to females. Males were 83.3%. Only five females (16.7%) were seen in the study.

Mode of injury:-

The patients who sustained pelvic trauma due to roadside accident were 28 patients (93.3%). Only 2 patients (6.7%) had a positive history of fall from the height leading to pelvic injury.

Acetabular fractures detected on pelvic skiagram:-

In 26 patients (86.7%), definite acetabular fractures were seen, however in 13.3% of cases, no obvious fracture of acetabulum was detected on pelvic skiagrams.

Pattern of acetabular fractures detected on pelvic skiagramsview:-AP

Out of total 30 cases, comminuted fracture of acetabulum was seen in 16 patients (53%), whereas linear fracture was noted in 10 patients (34%). In 04 patients, definite fracture line was not seen on X rays, however there was high index of suspicion on account of associated fractures of pelvic bones.

Dislocation of femoral head detected on pelvic skiagram AP view:-

Dislocation of femoral head was seen in 14 cases (46.6%), whereas in the remaining (53.3%) patients, no associated dislocation of femoral head was seen.

Disribution of acetabular fractures on pelvic skiagrams:-

Most common acetabular fractures in our study were those in the roof, anterior and posterior column seen in 08 patients (31%). Posterior wall fracture was seen in 08 patients. Fracture of quadrilateral plate was seen in 03 patients.

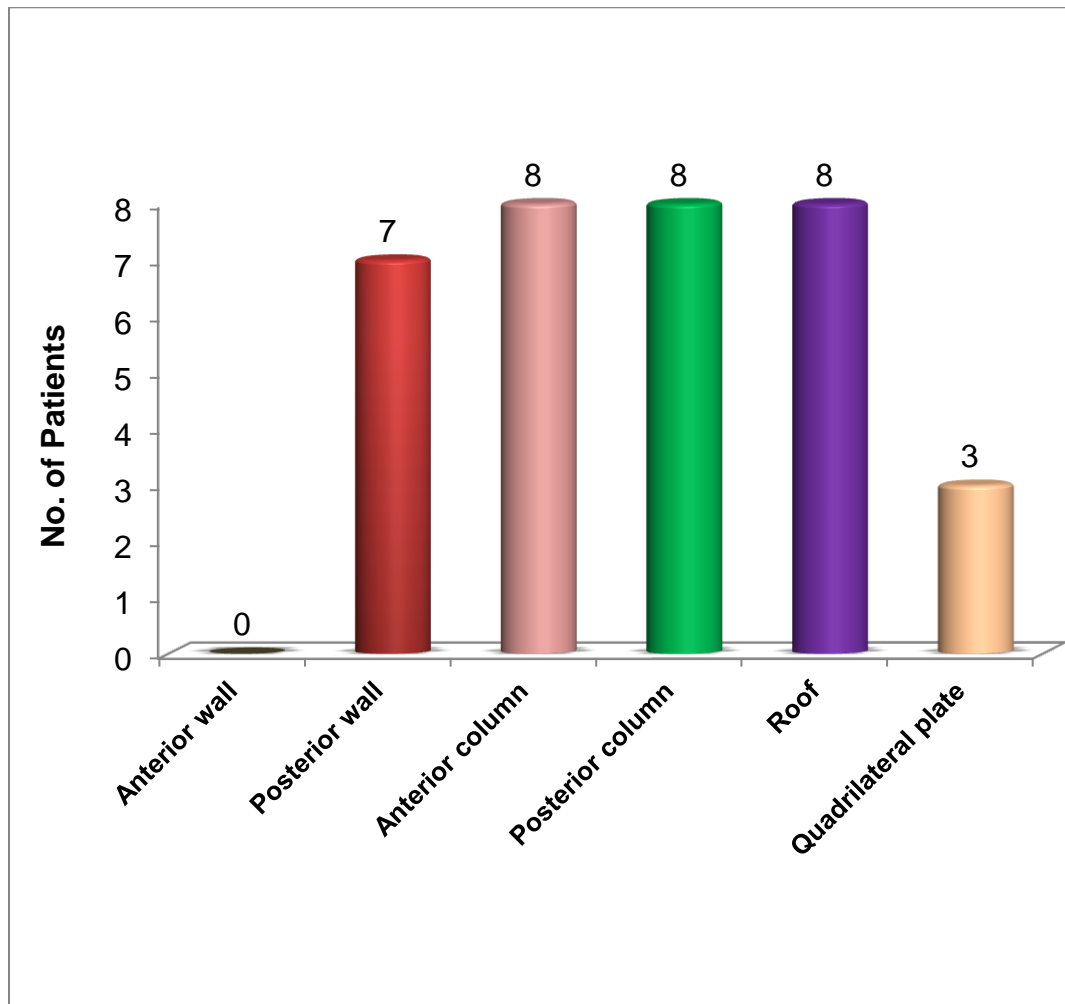


Figure 4:- Distribution Of Acetabulum Fractures On Pelvic Skiagram.s

Unilateral or bilateral acetabular fractures seen on computed tomography:-

Unilateral acetabular fracture was seen in 26 patients and bilateral acetabular fracture was seen in three patients on CT while on conventional radiographs it was seen in only one case.

Pattern of acetabular fractures as detected on computed tomography:-

Acetabular fractures were seen in 29 patients on CT. Out of these 29 patients, pattern of comminuted fracture was seen in 18 patients and 11 patients had linear pattern. In one patient, no acetabular fracture was detected on CT.

Dislocation of femoral head as detected on computed tomography:-

Out of 29 positive cases of acetabular fractures, associated dislocation of the femoral head was seen in only six cases. Most of the patients with dislocation underwent reduction of the dislocated hip prior to CT examination.

Type of dislocation of femoral head detected on computed tomography:-

Acetabular fractures were associated with dislocation of the femoral head in six cases. Out of these, posterior dislocation of the femoral head was seen in four cases, central dislocation in two cases whereas none of the cases had anterior dislocation.

Distribution of acetabular fractures on computed tomography:-

Most commonly involved segment of the acetabulum on CT was roof seen in 18 cases (62%) and quadrilateral plate seen in 12 cases (41.4%). Involvement of the posterior wall, anterior wall, anterior and posterior column was seen in 9 cases (31%). Fracture involving the anterior wall was seen in 3 patients (10.3%).

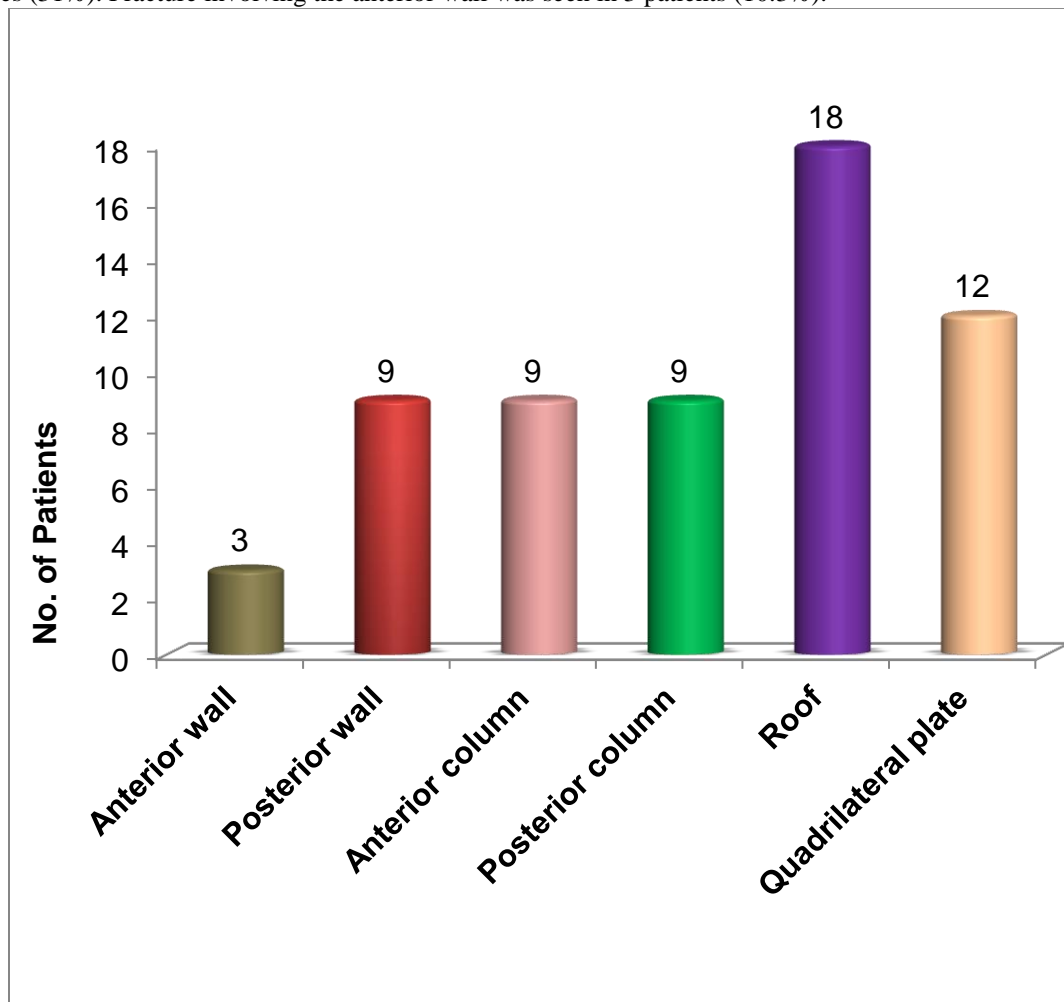


Figure 5:- Distribution Of Acetabular Fractures On Computed Tomography.

Associated fractures of pelvic bones seen on computed tomography:-

Along with acetabular fractures, the fractures of the pelvic bones were detected. Fractures involving the pubic bone were seen in 14 cases (27.5%), fracture of sacrum was seen in 5 cases (17.2%) and fracture of femoral head was seen in 3 cases (10.3%).

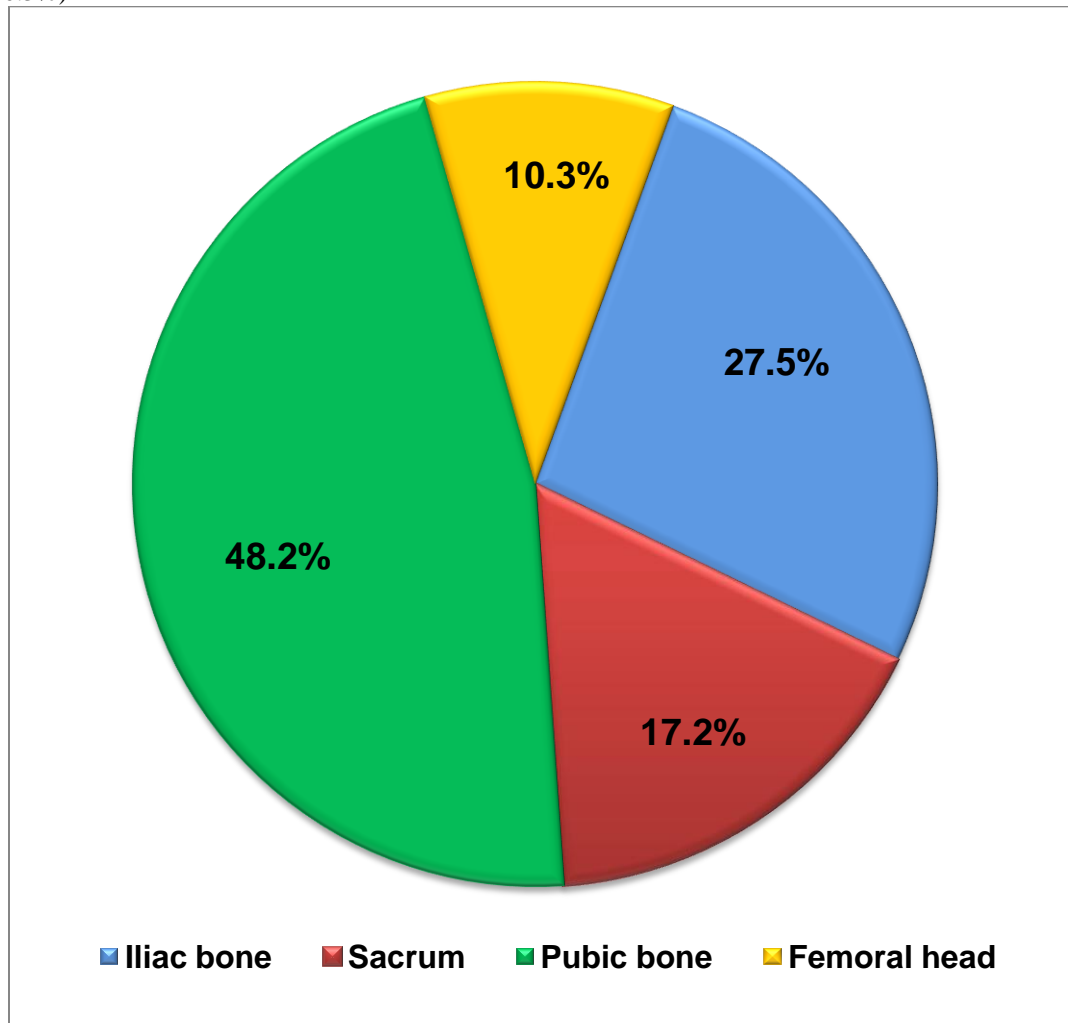


Figure 6:- Associated Fractures Of Pelvic Bones Seen On Computed Tomography (N=29)

Combination of pelvic fractures as detected on computed tomography:-

In some patients, more than one segment of acetabulum was fractured. The most common combination was seen in fracture of roof and quadrilateral plate of acetabulum seen in 7 cases (24%), next common combination was fracture involving both columns (13.7%).

Number of intra-articular bony fragments detected on computed tomography:-

Intra-articular bony fragments were seen in 7 (24%) of patients on CT. Single bony fragment was seen in 4 cases (57%). Multiple bony fragments were seen in 3 cases (43%).

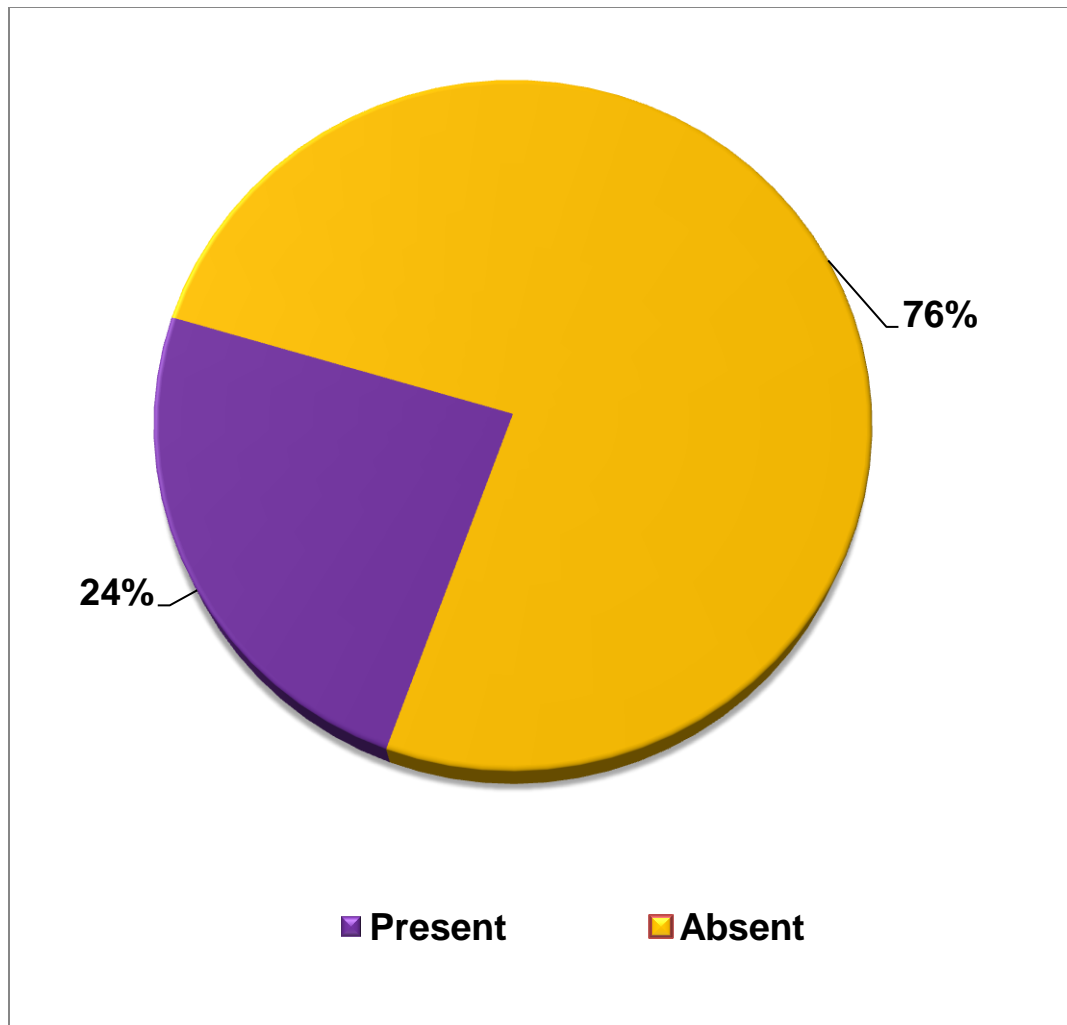


Figure 7:- Intra-Articular Bony Fragments Detected On Computed Tomography.

Discussion:-

In our study, the maximum number of patients were in the age group of 20-40 years (60%) with mean age of 36.26 yrs and median age of 30. This reflects the most active age in life, especially with regard to travelling. Our results were comparable with those of Adam et al (4). In their study of thirty patients, the age of patients ranged from 17-73 yrs with mean age of 34.5 yrs.

Roadside injury due to motor vehicle injury was the leading cause of pelvic trauma in 28 patients (93.3%), whereas fall from height accounted for 2 patients (6.6%). High incidence of pelvic trauma (93.3%) caused by motor vehicle accidents is comparable to the study of Snohiadski et al (13) who reported traffic accidents as a cause of acetabular fractures in 84% of patients.

CT is superior to plain radiography in not only confirming or excluding the fractures but also clearly demonstrating whether the fractures are unilateral or bilateral. Bilateral involvement of acetabulum on conventional radiography was noted only in one case while on CT, bilateral involvement was seen in 3 patients. There were two cases where bilateral involvement was missed on X-ray.

Hip dislocation was detected in 14 patients (46.6%) on radiography whereas it was seen in only 6 cases (20%) on CT examination. This discrepancy could be attributed to the fact that most of these patients were subjected to CT examination after post reduction of the dislocated hip. The incidence of dislocation of femoral head associated with acetabular fracture in our study was comparable to that of Sauser et al (11). In their study 53% of patients had

associated dislocation of femoral head as compared to 46.6% in our study. High incidence of posterior dislocation (78.5%) in the present study was comparable to that of Brown et al (12) who also reported 14 patients of posterior dislocation out of 17 patients included in their study.

According to Harley et al (6), the sensitivity of CT is more in detection of fractures of quadrilateral plate and roof as transverse axis of the CT images allows the precise characterization of the abnormality of this structure, which is not possible with conventional radiography. In their study of 26 patients of pelvic trauma, the authors reported 100% sensitivity of CT in detecting fractures of roof and quadrilateral plate compared to 73% for conventional radiography. The overall sensitivity of X-ray in detection of acetabular fractures was 26/29 (89%) and overall sensitivity of CT was 100%. Authors also observed that there was no significant difference between plain radiography and CT in detecting fractures of anterior column, posterior column and posterior wall. This can be attributed to the fact that these structures are so oriented that are quite well seen on conventional radiography. We also found almost equal sensitivity of CT and conventional radiography in detecting fractures of anterior column (89%), posterior column (89%) and posterior wall (78%) on conventional radiography and 100% on CT for all the segments of acetabulum. Martinez et al (1) also illustrated the above fact in their study.

Sensitivity of CT in detecting intra-articular fragments in present study compares well with the other authors. The intra-articular fragments were detected on CT in 7 patients, while on conventional radiography of pelvis, intra-articular fragments were not seen in even a single patient. Harley et al (6) detected intra-articular bony fragments in 18 out of 26 patients with acetabular fractures, whereas plain radiography showed no intra-articular bony fragment. Sauser et al(11) did not detect any bony fragment on plain skiagrams as compared to CT in which bony fragments were seen

In 3 patients. Adab et al (4) also emphasized superiority of CT in detection of any intra-articular bony fragment as compared to conventional radiography as they detected one or more loose bodies in 33% of patients included in their study whereas only one loose body was apparent on conventional radiography.

The use of 3D surface reformations of complex anatomical structures from sets of contiguous axial CT sections had been discussed by Guy et al (7), Magid et al (17), Fishman et al(15) and Rommense et al (16). Guy et al(7) observed that fracture lines demonstrated on plain radiographs and axial scans are not always apparent on 3D reconstructions, hence according to authors, 3D is complementary and not a substitute to good quality plain radiographs and axial computed tomographic sections. In our study, all the axial sections of CT scan were superior to the reconstructed images in the detection of acetabular fractures. 3DCT did not alter the initial radiological diagnosis but it did provide the best and most easy interpretation of overall assessment of the fractures.

The comparison between CT and plain radiography shows that CT is more sensitive than conventional radiography in detecting linear, undisplaced fractures of acetabulum, detecting fractures involving quadrilateral plate and acetabular roof. In addition, abnormalities of the hip joint space like loose bony fragments were detected more often on CT. Hence CT has been advocated as an adjuvant to conventional radiography in the evaluation of acetabular fractures of the pelvic bones.

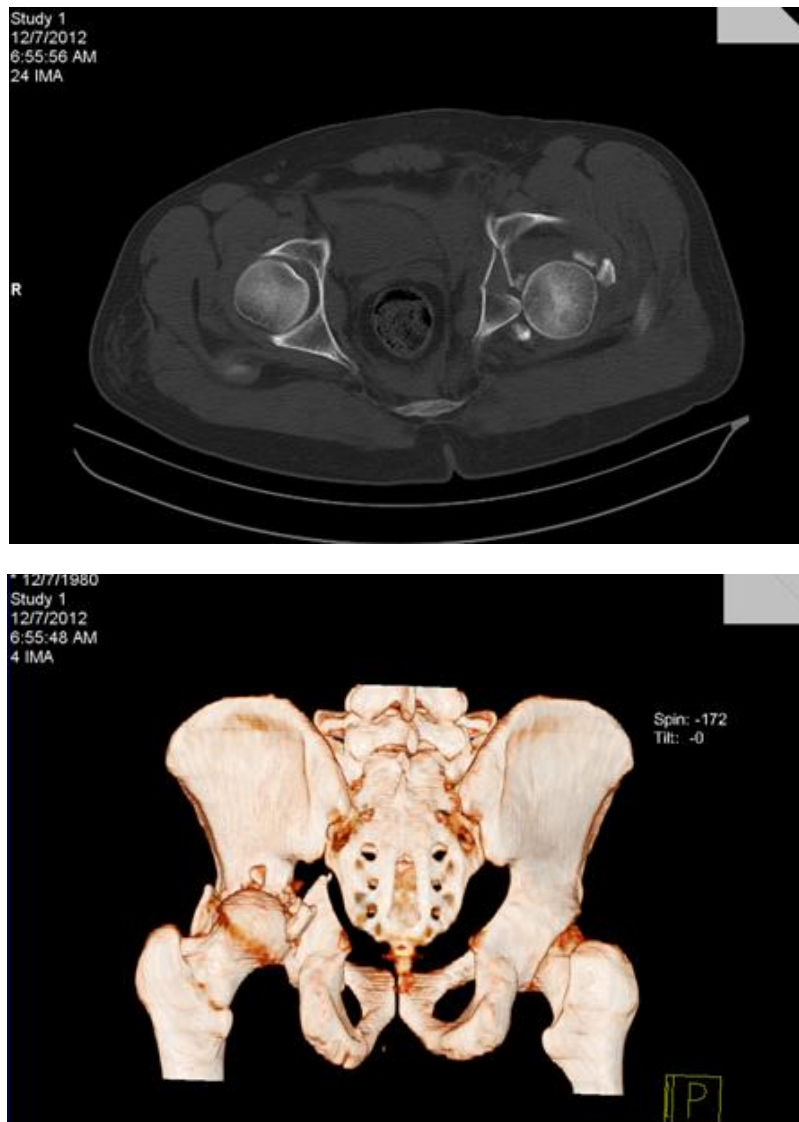


Figure 8:- Axial and 3D Reformatted CT images showing T shaped fracture of Left Acetabulum with Posterior Dislocation of Left Femur.



Figure 9:- Coronal and axial CT images showing Comminuted fracture of both the columns extending superiorly to involve iliac bone on left side.



Figure 10:- Axial CT images showing Fractures of Posterior wall and Posterior column of Left Acetabulum.



Figure 11:- Coronal and 3D CT images showing comminuted fracture of both columns of right acetabulum with extension into iliac bone.



Figure 12:- X ray showing disruption of iliopectineal line and fracture of both superior and inferior pubic rami.

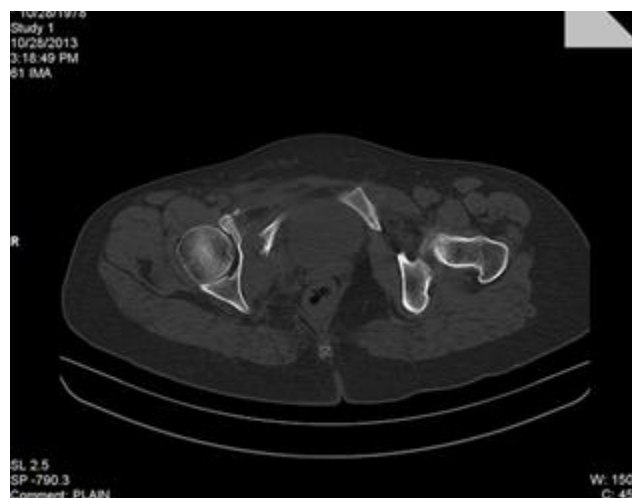




Figure 13:- Axial and 3D Reformatted CT images showing Fracture of Anterior wall and Anterior Column of Right Acetabulum.

Summary and Conclusion:-

The radiological evaluation of hip and acetabulum has been revolutionized by development of computed tomography because of its fine definition of sectional anatomy and accurate assessment of pelvic trauma. CT is superior to plain radiography in not only confirming or excluding the fractures but also clearly demonstrate whether the fractures are unilateral or bilateral. The overall sensitivity of X-ray in detection of acetabular fractures is 86% (26/29) and overall sensitivity of CT is 100%. From the present study, we conclude that computed tomography is superior to conventional radiography in detecting acetabular fractures, involvement of specific segment of acetabulum, associated fractures of pelvic bones and in detection of intra-articular fragments. CT displays the better anatomy of acetabulum in axial plane, thereby providing the missing pieces of information and greatly facilitating the classification of acetabular injury.

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RESEARCH ARTICLE

TOBACCO CESSATION COUNSELING – THE ROLES AND RESPONSIBILITIES OF A DENTIST.

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Abstract

India is the fourth-largest consumer of tobacco in the world and the third-largest producer of tobacco after China and Brazil. There are about 250 million tobacco users in India who account for about 19% of the world's total 1.3 billion tobacco users. India's tobacco problem is made more complex by the fact that tobacco is used in various forms in different parts of the country. Where research once concentrated on tobacco-related diseases and their treatments, emphasis has been shifting toward examining underlying behaviors. Understanding risk profiles helps determine where and how treatment should be focused and which therapies are most effective. It is for this reason that oral health team members are in an ideal position to give their patients specific, authoritative information concerning the adverse oral effects of tobacco use and the means to tackle it. Optimizing the implementation of routine tobacco cessation intervention in general dental practice remains a key aim in the delivery of dental care.

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Introduction:-

Tobacco use can lead to death, as it is one of the causes for chronic diseases. Such deaths are preventable. In 2005, tobacco use was responsible for more than 5 million deaths around the world and this number is expected to rise to 10 million deaths annually by 2020. [1] India is the fourth-largest consumer of tobacco in the world and the third-largest producer of tobacco after China and Brazil. There are about 250 million tobacco users in India who account for about 19% of the world's total 1.3 billion tobacco users. [2]

India's tobacco problem is made more complex by the fact that tobacco is used in various forms in different parts of the country. The prevalence of all types of tobacco use among men is about 47% (11%–79% in different states) and, among women, smokeless tobacco use varies between 0.2% in Punjab and 61% in Mizoram (Where the prevalence of women's smoking reaches a high of 22%). Among school-going children in grades 8 through 10, current tobacco use varies from 2.7% in Himachal Pradesh to 63% in Nagaland. [3]

In India, at least 800,000 deaths every year are related to tobacco use (700,000 of them due to smoking). Conservative costs of treating selected tobacco-related cancers, heart and lung disease amounted to Rs 30,833 crores (US\$ 7.2 billion) in 2002–2003. [2]

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Over the past few years, India has demonstrated a resolve to deal with the issue of tobacco use. In 2003, the Indian Parliament passed the Tobacco Control Act and in 2004 it ratified an international treaty—the World Health Organization's Framework Convention on Tobacco Control (WHO-FCTC). Efforts are urgently needed at the implementation level to reduce the current and projected harm caused by tobacco. [4]

Tobacco And Oral Health:-

Since the early 1970s, dental professionals have become increasingly aware of the damage that smoked and smokeless tobacco causes to tissues in and around the oral cavity.[5] Doubts about tobacco's adverse effects on oral health are evaporating in the face of evidence from a large and growing body of scientific literature that establishes it as an underlying cause of numerous diseases.

Ranging from mild to life-threatening, the following tobacco-related oral conditions may develop: halitosis, hairy tongue, dental calculus, periodontal disease, acute necrotizing ulcerative gingivitis, abrasion, discoloration of teeth and restorative materials, miscellaneous tissue changes, delayed wound healing, sinusitis, leukoplakia, and oral cancer. [6] Additionally, tobacco use increases and complicates treatment risks by compromising the prognosis for periodontal and other oral diseases and increasing the likelihood of the occurrence and reoccurrence of mouth cancers.[7]

Indeed, in light of current knowledge, mechanical and pharmaceutical treatment of oral diseases without addressing patient smoking is akin to medicating and binding a sliver in the hand but not removing it. The source of the problem must be managed. For e.g., treating tobacco use and dependence is as important to success in caring for periodontal disease as are restorative measures for dental caries, and preventing initiation of tobacco use by youths is as fundamental to the prevention of periodontal disease as providing fluoride and sealants are to caries prevention.[8] Where research once concentrated on tobacco-related diseases and their treatments, emphasis has been shifting toward examining underlying behaviors. Understanding risk profiles helps determine where and how treatment should be focused and which therapies are most effective. [9]

It is for this reason that oral health team members are in an ideal position to give their patients specific, authoritative information concerning the adverse oral effects of tobacco use and the means to tackle it. [10] Moreover, dentists who implement an effective smoking cessation program in their practices can expect to achieve quit rates up to 10-15 percent each year among their patients who smoke or use smokeless tobacco.[11]

These rates will rise further if additional help is harnessed for smoking cessation counseling by referral to smoker's clinics and by appropriate use of pharmacotherapy. Thus, optimizing the implementation of routine smoking cessation intervention in general dental practice remains a key aim in the delivery of dental care. [12]In this regard, the 1996 clinical practice guideline Smoking Cessation recommended methods based on more than 3,000 clinical studies in several countries that had been published in peer-reviewed journals over a fifteen- year period. [13]

The guideline became the most successful of a long series produced by the Agency for Health Care Policy and Research (now the Agency for Healthcare Research and Quality), and in four years, more than 1 million copies were distributed worldwide. The 1996 guideline focused primarily on individual patient encounters. With rapid advances in the science of tobacco treatment, the 3,000 studies initially available had expanded to more than 6,000 by 1999.[14]

The 2000 guideline, *Treating Tobacco Use and Dependence*, placed an emphasis on regarding tobacco use as a chronic disease. Repeated care is needed to manage the long-term effects and the slow, partial recovery process. Relapse is almost inevitable during recovery, so clinician monitoring and reinforcement are needed just as is done when treating periodontal diseases, diabetes, hypertension, and other conditions requiring sustained management.[15]

The new guideline addresses issues commonly confronted, such as triage, care of special patients, and the special conditions, policies, and procedures needed to nurture effective patient cessation and maintenance services. Key recommendations in the current guideline are as follows:

1. Tobacco dependence is a chronic condition that often requires repeated intervention. However, effective treatments exist that can produce long-term or even permanent abstinence.
2. Because effective tobacco dependence treatments are available, every patient who uses tobacco should be offered at least one of these treatments:
 - a. Patients willing to try to quit tobacco use should be provided with treatments identified as effective in the guideline.
 - b. Patients unwilling to try to quit tobacco use should be provided with brief interventions designed to increase their motivation to quit.
3. It is essential that clinicians and health care delivery systems (including administrators, insurers, and purchasers) institutionalize the consistent identification, documentation, and treatment of every tobacco user seen in a health care setting.
4. Brief tobacco dependence treatment is effective, and every patient who uses tobacco should be offered at least brief treatment.
5. There is a strong dose-response relationship between the intensity of tobacco dependence counseling and its effectiveness. Treatments involving person-to-person contact (via individual, group, or proactive telephone counseling) are consistently effective, and their effectiveness increases with treatment intensity (i.e., minutes of contact).
6. The following three types of counseling and behavioral therapies were found to be especially effective and should be used with all patients attempting tobacco cessation:
 - a. Provision of practical counseling (problem solving/skills training);
 - b. Provision of social support as part of treatment (intra-treatment support);
 - c. Help in securing social support outside of treatment (extra-treatment social support).
7. Numerous effective pharmacotherapies for smoking cessation now exist. Except in the presence of contraindications, these should be used with all patients attempting to quit smoking.
 - a. Five first-line pharmacotherapies were identified that reliably increase long-term smoking abstinence rates: bupropion SR, nicotine gum, nicotine inhaler, nicotine nasal spray, and nicotine patch.
 - b. Two second-line pharmacotherapies were identified as efficacious and may be considered by clinicians if first-line pharmacotherapies are not effective: clonidine and nortriptyline.
 - c. Over-the-counter nicotine patches are effective relative to placebo, and their use should be encouraged.
8. Tobacco dependence treatments are both clinically effective and cost-effective relative to other medical and disease prevention interventions. As such, insurers and purchasers should make sure that:
 - a. All insurance plans include as a reimbursed benefit the counseling and pharmacotherapeutic treatments identified as effective in the guideline; and
 - b. Clinicians are reimbursed for providing tobacco dependence treatment just as they are for treating other chronic conditions.

Role Of The Dentist ^[16]

In the clinic, dentists have an important role in helping patients quit tobacco and, at the community and national levels, to promote tobacco prevention and control strategies.

Dentists in the clinics:-

Can check for the harmful effects of tobacco in the oral cavity, they are in an ideal position to counsel patients, can target young children and youth as patients and can influence them to adopt a tobacco-free lifestyle, Can treat women of childbearing age and can inform them of the dangers of tobacco use during pregnancy. They spend more time with patients than other clinicians and use this time to counsel tobacco users to quit and reinforce messages given to patients by physicians and other caregivers about the dangers of tobacco use and the need to quit.

Dentists in community and nation:-

They can act as role models by not using tobacco or by quitting successfully. Tobacco use by dentists is a significant barrier to tobacco cessation counseling. They are also in a position to speak with authority in the community about the dangers of tobacco use; for example, the need to curb tobacco use in public and educate children about the dangers of tobacco use. Thus, they can be effective advocates for tobacco control in the community.

Behavioral Counseling:-

Counseling is a collaborative process in which the counselor assists the client in facilitating behavior change, enhancing coping skills, promoting decision making and improving relationships. [17] Tobacco use decreases when

patients receive counseling from a health care provider. Physicians and other primary health care personnel are valuable and effective in the management of individuals with tobacco cessation. The physician has the influence, as a credible expert in a position of authority, to suggest and advise patients to quit tobacco use. [18]

Patients, who are willing to quit tobacco, should be provided with treatments identified as effective. However, for those who are unwilling to try and quit tobacco, interventions should be designed to increase their motivation to quit. [19] Under these circumstances, patients undergo a series of stages of willingness and preparedness to quit. At each stage a person is thinking and feeling differently about the tobacco use habit. These 5 stages are described below which can be categorized as: [20]

Stage-1) Pre-contemplation

Here a person is not thinking about quitting, they are not interested in change. This is because:

- i) They do not see their smoking habit as a problem.
- ii) They think that Tobacco is infact helpful and beneficial, that it helps them in concentration, in digestion, or for relaxing.
- iii) They may be fully aware of the risks but may value tobacco use for other reasons that thus do not wish to stop.
- iv) Unaware of help available and previous failed attempts at changing make them believe they cannot stop.

Stage-2) Contemplation

Here the patient knows the risks and problems, but is undecided and is unaware of the benefits on quitting smoking. In this stage the person is torn two ways, i.e., aware that he ought to stop but still feeling attached or drawn to smoking.

Stage-3) Preparation stage

Here the patient expresses a desire to quit. Those in preparation stage are planning to take action soon. They are beginning to make small changes and trying out different ways of behaving. They may tell others about their intention to stop and make clear plans on how they are going to do it.

Stage- 4) Action stage

Here patient makes visible changes and puts considerable effort for leaving the habit. This is often the time when they seek professional help.

Stage-5) Active period

During this stage, quit status is continued and strengthened, temporary stoppage becomes part of a more settled pattern. Unless this takes place the person may move into relapse, and return from there to pre-contemplation or contemplation stage.

It may be noted that a person may move from one stage to another in the forward or backward direction and may require continuous effort to move towards the direction to quit smoking. The emphasis of all the above points is to make the patients aware that tobacco use is affecting their physical health, family and social life. The guiding principle here should be to involve the patient in discussion and providing information on the consequences and risks of tobacco use.[21]

Barriers to tobacco cessation:-

A number of barriers to the provision of tobacco cessation services at a dental clinic have previously been reported. These include practitioner opinions, attitudes and perceptions toward tobacco cessation counseling.[13] Other reported barriers include lack of time, and concern about upsetting the patient¹⁵⁰ and lack of reimbursement. [22] Although, the most significant barrier still remains a lack of education of dentists and hygienists on cessation activities during their formative years of training. [23]

Nevertheless, there are a number of important activities that could hamper this: [24]

1. Training of more dental students, dental hygienists, and dental practitioners to provide tobacco cessation counseling;
2. Increase in the number of dental practices routinely monitoring tobacco use and providing tobacco cessation programs;

3. Increase in the utilization of the available procedures for tobacco cessation, (whether it is a covered service or not) so as to familiarize insurance companies with this practice; and
4. Stimulating the demand for more tobacco cessation coverage by employees. Such activities could go a long way towards encouraging employers to cover tobacco cessation without any differentiation by provider, as long as the outcomes are reasonably successful.

Conclusion:-

Tobacco imposes a colossal burden of disease and death leading to catastrophic health, social, economic and environmental effects. Prevalence and practices of tobacco use in India are varied and disparate. Tobacco consumption continues to grow at 2–3% per annum, and by 2020 it is predicted that it will account for 13% of all deaths in the country. [25] Tobacco dependence displays many features of a chronic disease. Only a minority of tobacco users achieve permanent abstinence in an initial quit attempt. The majorities of users persist in tobacco use for many years and typically cycle through multiple periods of remission and relapse. A failure to appreciate the chronic nature of tobacco dependence impedes clinicians' consistent assessment and treatment of the tobacco user over time. [26]

Effective interventions exist that can produce long-term cessation at up to double the rate achieved by smokers without treatment. It is important for clinicians to know that assessing and treating tobacco use generally leads to greater patient satisfaction with health care. Members of the dental profession have a unique opportunity to influence tobacco use by their patients.[27]

Dentists are obligated to help patients quit because tobacco use is the most common underlying cause of periodontal and many other oral diseases and conditions, apart from the risks to quality of life and to life itself. [28]Evidence is overwhelming that tobacco damages oral health and that clinical cessation methods work in dental practice. Although not specifically addressed in any guideline, professional education institutions, including dental and dental hygiene schools, need to identify tobacco use among all patients.[29]

The time that the dental team spends with patients usually exceeds that occurring in medical practice. The very brief (i.e., less than three minute) intervention clinicians should use to advise and assist patients need not be dedicated time but can be easily woven into other diagnostic and treatment services. As with other chronic diseases, repeated assistance is useful. Unlike treatment of other conditions, such support is particularly important to counter incentives by a powerful industry that constantly attempts to encourage previous users to relapse. [30]

Patients who quit must learn how to cope with environmental triggers. Resisting the rituals surrounding tobacco use, resisting offers of tobacco and encouraging support from those who live, work, and play with the patient are at least as important as using pharmacotherapy's for reducing discomfort associated with withdrawal and recovery. Human interaction with a respected health care provider has a powerful influence and can be tailored to individual patient interests, conditions, and culture. Pharmacotherapy should be reinstated as often as needed, that is, as often as relapsing patients are willing to make another attempt to quit, because tobacco dependence is a chronic, relapsing condition that usually requires multiple attempts before long-term abstinence is achieved. [31]

Repeated contacts and a longer duration of contact are often more easily provided in dental practice than in many medical care environments. Compensation for cessation services in dental practice may be by patient payment, by integration into another dental service, or as part of a reimbursement program. [32] Effective interventions exist that can produce long-term cessation at up to double the rate achieved by smokers without treatment. It is important for clinicians to know that assessing and treating tobacco use generally leads to greater patient satisfaction with health care. Members of the dental profession thus have a unique opportunity to influence tobacco use by their patients.

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RESEARCH ARTICLE

COMPARATIVE EVALUATION OF ANTIMICROBIAL POTENTIAL OF GINGER, GARLIC AND CINNAMON EXTRACTS AGAINST STREPTOCOCCUS MUTANS AND LACTOBACILLUS ACIDOPHILUS.

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Antimicrobial efficacy, Agar disk diffusion test, S.mutans, L.acidophilus.

Abstract

Aim and Objective: To evaluate the antibacterial potential of herbs and spices extracts in efficiently reducing cariogenic pathogens in oral cavity, thereby incorporating them into various dentifrices and topical preparations which are safe, economical, free from side effects and could act as the new source of antimicrobial agents.

Methods: Ethanolic extracts (10% each) of ginger, garlic and cinnamon were prepared and subjected to microbiological assay along with 0.2% chlorhexidine gluconate (CHX), to determine its zone of inhibition at volumes of 10 µl, 20 µl, and 30µl each using Agar disk diffusion test against Streptococcus mutans (S.mutans) and Lactobacillus acidophilus (L.acidophilus). One-way ANOVA test and Post hoc test were used to compare the antibacterial potential of herbs and spices extracts in efficiently reducing cariogenic pathogens in oral cavity by incorporating them into various dentifrices using SPSS software version 14. The significance level for all the statistical tests utilized in this study was set at $p < 0.05$ %.

Results: There was a significant correlation between the control and experimental groups. The highest antimicrobial efficacy was observed in positive control group and by 10% ethanolic Cinnamon extract in comparison to 10% ethanolic extracts of Ginger and Garlic.

Conclusion: The plant extracts such as Cinnamon extracts in 10% concentration could be used as an alternative antibacterial compound for effective reduction of cariogenic pathogens in oral cavity.

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Introduction:-

Dental caries is one of the most common infections of all oral diseases. Cariogenic micro-organisms, especially *S.mutans* plays an essential role in the pathogenesis of dental caries. Other microbes such as *Lactobacillus* species are also associated with dental caries by fermenting sugars into lactic acid and grow readily at low pH values. Among the chemotherapeutic agents, CHX is the “gold-standard” but has side effects like brown discoloration of the teeth, oral mucosal erosion, dryness, burning sensation and undesirable taste.¹ Thus there is a need for an alternative, safe, efficacious and cost effective treatment options and biologic products for oral diseases.

Natural products such as herbs and spices have been used since ancient times as antimicrobial agents that have fewer side effects, better patient tolerance, cost effective and readily available.^{2,3} Some common herbs such as cilantro, basil, ginger, turmeric, garlic, cinnamon etc., offer great health benefits by virtue of their powerful phytochemical and antioxidant properties.⁴

Thus, the aim of this study was to comparatively evaluate the antimicrobial potential of 10% ginger, 10% garlic and 10% cinnamon extract against *S.mutans* and *L.acidophilus*. To the best of our knowledge no study has been carried out to compare the antimicrobial potential of ginger, garlic and cinnamon extract against *S.mutans* and *L.acidophilus*.

Methods:-

The study was carried out in four steps:-

Phytochemical Standardization:-

According to the WHO guidelines⁵ the phytochemical standardization of the herbal drugs were performed by the Department of Pharmaceuticals, Rajiv Academy for Pharmacy, Mathura (U.P)

Preparation of Ethanolic Extracts:-

Fresh ginger, garlic and cinnamon (500 grams each) was procured from the local market and cleaned using distilled water, minced into fine pieces and suspended in sterile jars containing 1000ml of 70% ethanol each. They were subjected to the process of cold maceration for 48 hours in a sterile jar after which the process of filtration using sterile muslin cloth was done. The filtrates obtained were placed over steam bath apparatus for 5 days to facilitate evaporation of ethanol content. After 5 days, 25g of ginger, 25g of cinnamon and 28g of garlic extracts were obtained.

Preparation of 10% Ethanolic Extracts:-

For the preparation of 10% stock solution of ethanolic extracts, 10 grams of obtained extracts were dissolved in 100ml of dimethyl sulfoxide to obtain 10% ethanolic extracts of ginger, garlic and cinnamon respectively.

Determination of Zone of Inhibition:-

The stock solution of 10% ethanolic extracts prepared were subjected to agar disk diffusion test along with a positive control (0.2% CHX) at volumes of 10 µl, 20 µl and 30 µl each to determine the zone of inhibition against *S.mutans* and *L. acidophilus* respectively. Interpretation of diffusion results were carried out by the presence or absence of zone of inhibition. The standard strains of microorganisms used in this study were: *S.mutans* (Microbial Type Culture Collection-MTCC NO. 890) and *L. acidophilus* (MTCC NO. 10307, Chandigarh, India).

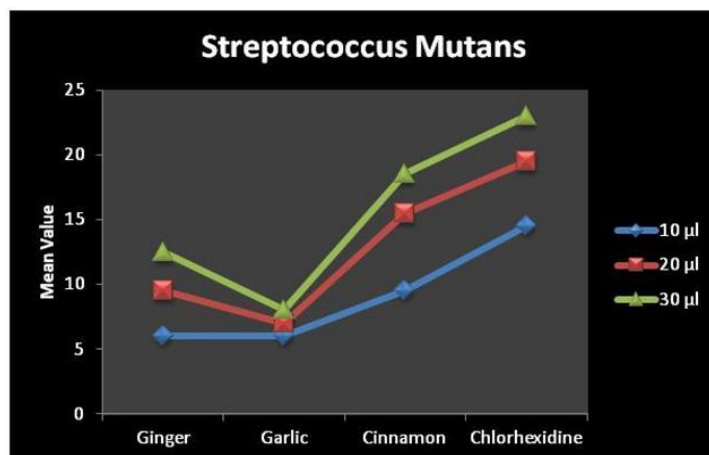
Statistical Analysis:-

One-way ANOVA test and Post hoc test were used to compare the antibacterial potential of herbs and spices extracts in efficiently reducing cariogenic pathogens in oral cavity by incorporating them into various dentifrices using SPSS software version 14. The significance level for all the statistical tests utilized in this study was set at $p < 0.05$ %.

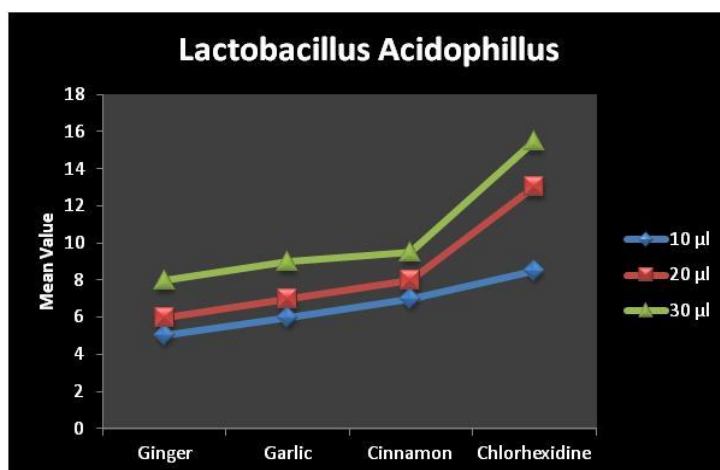
Result:-

Line diagram 1 showed the comparison of mean zone of inhibition values of 10% ethanolic extracts of ginger, garlic, cinnamon and 0.2% CHX at 10 µl, 20 µl, and 30 µl volume measured against *S.mutans*. The highest zone of inhibition was seen in 0.2% CHX followed by 10% ethanolic extracts of cinnamon and lowest by garlic extracts. Also, the zone of inhibition increased when the volumes of the extracts increased.

Line diagram 2 showed the comparison of mean of zone of inhibition values of 10% ethanolic extracts of ginger, garlic, cinnamon and 0.2% CHX at 10 μ l, 20 μ l and 30 μ l volume measured against *Lactobacillus acidophilus*. The highest zone of inhibition was seen in 0.2% CHX followed by 10% ethanolic extracts of cinnamon, and lowest by ginger extracts. An increase in inhibition zone was observed as the volumes of extracts increased.



Line Diagram 1:- Shows the comparison of mean values of disk diffusion test against *S. mutans*.



Line Diagram 2:- Shows the comparison of mean values of disk diffusion test against *L. acidophilus*.

Discussion:-

Initiation of dental caries and the microbial composition of plaque involves *S. mutans* or *Lactobacilli*. A prevalence of 87.4% and 36.7% were recorded for *S. mutans* and *L. acidophilus* for dental caries⁶ hence, these bacterial isolates were selected for this research work. Currently, CHX is the most potent chemotherapeutic agent against dental caries but due to its various side effects, it discourages patient to use as a mouth wash.⁷ The use of natural products such as ginger, garlic and cinnamon could be considered as a potential source of novel antimicrobial agents, used extensively for the treatment of cold, nausea, diarrhea and dental infections.⁸⁻¹¹

The purpose for this research was to compare and evaluate antimicrobial efficacy of 10% ethanolic extracts of ginger, garlic and cinnamon each with 0.2% CHX (positive control) at various volumes (10 μ l, 20 μ l, 30 μ l) using agar disk diffusion test. Dissolution of solvent based extracts was done in dimethyl sulfoxide (DMSO) which acts as a surfactant and easily gets penetrated in the agar. It is an oxidizing agent that neutralizes the antiseptic effect of ethanol and showed no antibacterial activity in the agar diffusion test.¹²

The results showed that maximum antimicrobial activity was exhibited by 0.2% CHX and 10% Cinnamon extract and least by 10% ethanolic extracts of Ginger and Garlic against both bacterial isolates but their effectiveness

varied. A mean zone of inhibition value showed by 0.2% CHX against *S.mutans* at 10 µl, 20 µl and 30 µl was 14.5 mm, 19.5 mm and 23 mm whereas against *L.acidophilus* it was 8.5 mm, 13 mm, and 15.5 mm respectively. The zone of inhibition was found to increase as the volume of extracts increased; this was in accordance with the findings of Nanasombat S.¹³

The mean zone of inhibition values of 10% ethanolic extract of Cinnamon against *S.mutans* was found to be 9.5mm, 15.5mm, 18.5mm at 10 µl, 20 µl and 30 µl and against *L.acidophilus* it was found to be 7 mm, 8 mm and 9.5 mm respectively. The inhibition value was not significantly lower than 0.2% Chlorhexidine. Mechanisms, where by Cinnamon extract inhibit growth of *S. mutans* are still unclear. The antibacterial effect of cinnamon were related to the inhibitory effect of its hydrophobic essential oils (cinnamaldehyde, eugenol and carvacrol) that enable them to partition the lipids of the bacterial cell membrane, disturbing the cell structures and rendering them more permeable. Extensive leakage of critical molecules and ions from bacterial cells leads to its death.¹⁴ Asmi OH et al also proved that cinnamon had showed the highest antimicrobial effect at maximum concentration on the growth of bacterial strains followed by garlic, ginger and turmeric.¹⁵

The highest mean zone of inhibition values of 10% ethanolic extract of cinnamon when compared to 10% ethanolic extracts of ginger and garlic against *S.mutans* and *L.acidophilus* could be attributed to the presence of phytochemicals like alkaloids, saponins and tannins in abundance as observed in the phytochemical screening of the spices. The results of this study showed that although the extracts were less potent than Chlorhexidine, still they were relatively effective in inhibiting the growth of oral bacteria. Hence, 10% ethanolic extract of cinnamon extracts could be used in the form of a natural dentifrices as an alternative for patients who wish for natural, economic products without artificial preservatives, flavors and colors. The evaluation of toxicity and allergic potential of these plant extracts was not included in this study. Hence, a conclusive comment on the toxicity and allergic potential of these plant extracts cannot be made.

Conclusions:-

The highest antimicrobial efficacy was shown by 0.2% Chlorhexidine followed by 10% ethanolic Cinnamon extract and lowest by 10% ethanolic Ginger and Garlic extracts at 10 µl, 20 µl and 30 µl against *S.mutans* and *L.acidophilus*. The antimicrobial efficacy increased with increase in volume. Thus, use of 10% ethanolic extract of cinnamon as an alternative antibacterial compound for effective reduction of cariogenic pathogens in oral cavity would not only reduce the clinical burden of drug resistance development but also lessen the side effects and cost of the treatment with allopathic medicine.

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RESEARCH ARTICLE

ANALYZING THE ROLE OF STRATEGIC ALIGNMENT OF BUSINESS STRATEGIES RELATED TO THE (MILES & SNOW) MODEL ON COMPETITIVE INTELLIGENCE. (A COMPARATIVE STUDY BETWEEN COCA-COLA AND PEPSI SOFT DRINKS COMPANIES)

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defenders' strategy, prospectors' strategy, analysts' strategy, respondents' strategy, and competitive intelligence.

Abstract

This study aims to identify the compatibility between a numbers of business alignment strategies related to the (Miles & Snow) model that represented by (defenders, prospectors, analysts, respondents) with competitive intelligence, represented by (planning and focus analysis, collection, communication, structure and processes, and organisational culture and awareness). The study population is two soft drinks company (Hayat Company for soft drinks and mineral water, Ltd. under license from PepsiCo Inc., and CC Company for the drinks industry, Ltd. under license from Coca-Cola) in the city of Erbil. The study pursues a descriptive analytical approach, along with examine a set of hypotheses for the purpose of achieving the goals of the study and its problem that have been tested by multiple statistical tools using statistical analysis software program (SPSS 20). A survey questionnaire prepared and distributed to managers and their assistant in addition to supervisors in the different units such as production, distribution, marketing and management, and in the various branches in the city of Erbil. The study sample numbered (63) managers. The study found a number of results based on the analysis of respondents' answers such as the both company have high-level rates of compatibility strategies comparable with excellence for Coca-Cola and comes second Pepsi. Based on the results the study presents a set of recommendations and suggestions to organizations.

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Introduction:-

Nowadays, markets which organizations operate characterized by a high degree of complexity and intense competition, which led to the emergence of many of the challenges that organizations have to adapt. Organizations become obligatory upon perception and analysis associated with the dimension of the competitive aspects in the markets that they were operates after becoming its survival and continuity in such markets depend on the ability of

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its managers to hire and direct available resources to have the greatest possible efficiency. Since helping that organizations to identify priorities and timing of strategic moves that should be completed in their activities efficiently in order to enhance its competitiveness and support along with take advantage of what has been waving profit from market opportunities (Hamwi and Abdullah, 2012, 886).

Competitive intelligence is not limited to the collection of intelligence information with pre-emptive nature of the competitors and their activities. It becomes recently extends to the external environment as a whole and analyzed after classified it, and then interpret and inference it on the various strategic decisions making that geared the future course of business organizations towards attaining strategic objectives to the extent that it is expected to rise competitive intelligence structure to a level of ability of these organizations in the future (Ismail and Jamil, 2016, 165).

Based on the above the present study will analysis the role of strategic alignment of business strategies related to the (Miles & Snow) model on competitive intelligence. This study has been divided into four sections, the first section deals with the scope of the study and its methodology. The second section will address the concept of strategies alignment to business strategies related to the (Miles & Snow) model, as well as the concept of competitive intelligence along with describes their characteristics and dimensions. Section three illustrates the results and determines the relationship between the two variables along with examines the impact of strategic alignment of business strategies on competitive intelligence. Finally, the fourth section will clarify the most findings and presents a set of recommendations.

Section one: the scope of study and Methodology:-

The scope of study:-

Study problem:-

In the era of globalization and rapidly changes along with the emergence of new environmental challenges constantly in all the areas of science made contemporary organizations face the competitive challenges. This organization cannot on its current based gain competitive advantage, strengthen and durability its survival. In addition to thinking to adopt more of a strategy or several strategies and compatibility between them to be able to acclimatize to these continuous and rapid changes and with different environments. In particular those environments that are dynamic, undependable, and intense competitive since competitors face the threats is a difficult subject. As a result, this organization requires to increased attention to competitive intelligence activities and to strengthening its dimensions to achieve competitive intelligence.

Based on the above, present study aims to identify the compatibility between a numbers of business alignment strategies related to the (Miles & Snow) model that represented by (defenders, prospectors, analysts, respondents) with competitive intelligence, represented by (planning and focus analysis, collection, communication, structure and processes, and organisational culture and awareness), along with testing the correlation in soft drinks company in Iraqi Kurdistan region Erbil city. In this regard the most important question that this study attempts to find proper answer may explains as following:

- Is there a strategic alignment of competitive business strategies followed by the organization and competitive intelligence?
- Is there is a correlation between strategic alignments of business strategies related to the (Miles & Snow) model and competitive intelligence?
- Is there is an impact of strategic alignments of business strategies related to the (Miles & Snow) model on competitive intelligence?

The significance of Study:-

The significance of the study can be explains under two aspects as following:-

- **The theoretical side:** the importance of this study embodied in enriches academic libraries in Iraqi Kurdistan region in particular, as well as Iraqi libraries in general. This study is an attempts to illustrates the theoretical concepts and conceptual framework of the role of strategic alignment of business strategies related to the (Miles & Snow) model on competitive intelligence at sample study that represent soft drinks organizations.
- **Applied side:** to clarify the relationship and the impact of of strategic alignment of business strategies related to the (Miles & Snow) model on competitive intelligence. Furthermore, directing the two organizations to research the importance of strategic alignment of business strategies and its role in improving the organizational performance thus improve the competitive intelligence.

The objectives of study:-**This study is an attempt to achieve the following objectives:-**

- To explain and describe the role of strategic alignment of business strategies on competitive intelligence in sample study.
- To clarify the correlation between strategic alignment of business strategies dimensions and the dimensions of competitive intelligence.
- To examine the impact of strategic alignment of business strategies dimensions on the dimensions of competitive intelligence.
- To illustrate the importance of strategic alignment of business strategies and the competitive intelligence to managers in all levels and to their organizations along with identify the benefits of this application.

Theoretical study Framework:-

The model (figure 1) illustrates the theoretical study framework. The model assumed a correlation and an impact of strategic alignment of business strategies related to the (Miles & Snow) model on competitive intelligence. In this study, strategic alignment of business strategies related to the (Miles & Snow) model is independent variable represents by four dimensions (defenders strategy, prospectors' strategy, analysts' strategy, and respondents' strategy). On the other hand, competitive intelligence is dependent variable represents by six dimensions (planning and focus analysis, collection, communication, structure and processes, and organisational culture and awareness).

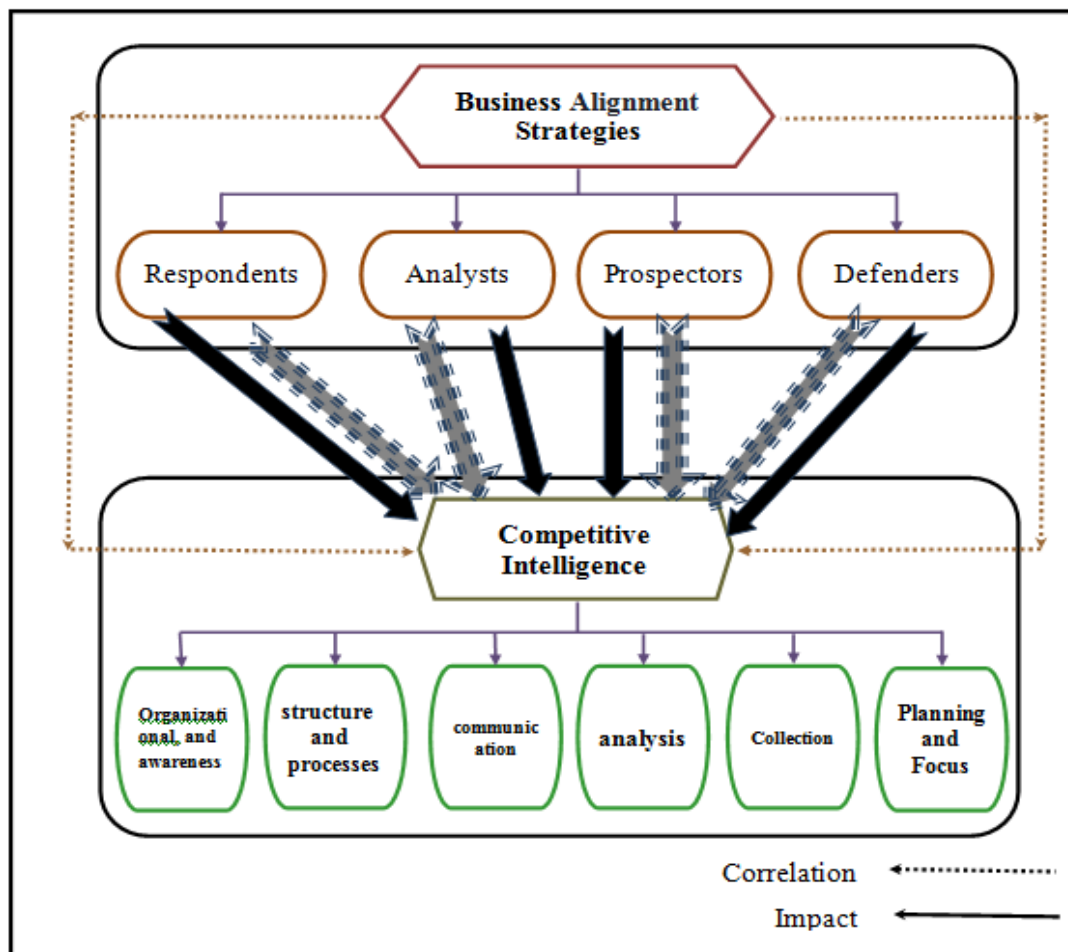


Figure 1:- Theoretical Study Framework "Source: prepared by researchers"

Research Hypothesis:-

To achieve the objectives of this study and testing the theoretical framework of the study, a number of hypotheses administrated and as following:

The First Hypothesis:-

There is a measurable positive correlation between strategic alignments of business strategies related to the (Miles & Snow) model and competitive intelligence.

From the first main hypothesis there are four sub-hypothesis as follows:-

- There is a measurable positive correlation between defenders strategy and competitive intelligence.
- There is a measurable positive correlation between prospectors' strategy and competitive intelligence.
- There is a measurable positive correlation between analysts' strategy and competitive intelligence.
- There is a measurable positive correlation between respondents' strategy and competitive intelligence.

The Second Hypothesis: there is statistically a significant impact of strategic alignments of business strategies related to the (Miles & Snow) model on competitive intelligence.

From the second main hypothesis there are four sub-hypothesis as follows:

- There is statistically a significant impact defenders strategy on competitive intelligence.
- There is statistically a significant impact prospectors' strategy on competitive intelligence.
- There is statistically a significant impact analysts' strategy on competitive intelligence.
- There is statistically a significant impact respondents' strategy on competitive intelligence.

Methodology:-**Study population and sample:-**

The study population is all managers in the two soft drinks companies in the city of Erbil that numbered (71) managers and assistant managers. The questionnaire distributed to all managers, their assistants, and supervisors in the units of production, distribution, marketing, and management and in various branches of the two soft drinks companies. 63 questionnaires returned and thus, the research response rate was (88.7%).

Study approach:-

In order to achieving the objectives of this study, descriptive and analytical approach was adopted. Since this approach describes theoretically study variables and then statistically analyzed in order to reach the objectives of study.

Data collection methods:-**In this study, data collected through secondary data and primary data as following:-**

- **Secondary data:** in this study various available source used to collect secondary data such as: books, scientific studies and research, theses and dissertation that relevant to present study in order to illustrates and explain to the theoretical framework of study variables.
- **Primary data:** the researchers relied on the survey questionnaire to achieve the study objectives through testing hypotheses and access to their results. The questionnaire consists of three sections. Section one presents the personal Characteristics of respondents such as: gender, age, level of education, years of experience, and position. Section two presents 20 items related to independent variable "strategic alignment of business strategies related to the (Miles & Snow) model". Finally, section three that presents 30 items on competitive intelligence, the table below gives more details about the instruments of the questionnaire

Table 1:- Questionnaire Instruments

Study Variables	Sub variables	No. of Items	Items	Sources
General Data	Personal data	4	4	Prepared by researchers
Strategic business alignment (Miles & Snow) model	Defenders	20	X1–X5	(Kess, Isoherranen, 2014), (Al-Lami, 2007), (Suleiman, 2010)
	Prospectors		X6–X10	
	Analysts		X11–X15	
	Respondents		X16–X20	
Competitive intelligence	Planning and focus	30	Y21–Y25	(Ismail and Hussein, 2015), (Srouf, and Mohammed, 2013)
	Collection		Y26–Y30	
	Analysis		Y31–Y35	
	Communication		Y36–Y40	
	Structure and processes		Y41–Y45	
	Organisational culture and awareness		Y46–Y50	

"Source: prepared by researchers"

The reliability of the questionnaire:-

Reliability means the measure's ability to show the same results when they are re-applied to the individuals themselves again. The reliability of this study tested through the use of scale (Cronbach's alpha), the total alpha coefficient was (94.7%) and this percentage is high for the purposes of the study.

Methods of data analysis:-

This study relied on a set of statistical tools as following:-

- Means, standard deviations, rate of agreements and the coefficient of variance in order to describe the study variables, analysis, and discussion the results.
- Simple correlation coefficient (Pearson) for the purposes of measuring the strength of the correlation between the two variables.
- Multiple linear regressions in order to measure the independent variable on dependent variable.

Section Two: Literature Review

Strategic alignment of business:-

There are a number of models concerning strategic behavior and competition. In this area, many academics and researchers presented several models that contribute the efforts of them, such as: Anderson & Paine, 1978 model, Miles & Snow, 1978 model, Burgelman, 1984 model, and Ansoff, 1987 model. In this regards, Miles & Snow, 1978 model is consider one of the most important, common and widely used in research and studies (Al- Qadu et al., 2007, 55). Thus, the present study will rely on (Miles & Snow) model for its compatibility with the study problem and objectives along with the theoretical side. Furthermore, it complies with the applied side, which is the soft drink company in the city of Erbil.

The (Miles & Snow) Model:-

Gimenez (1994, 4) and Musa (2014, 67) indicates that the strength of this model being concentrated in defining the relationship between organizational structure, work processes and technology in a way that allows organizations to interact with their surroundings. Moreover, it is effective in diagnosing some of the strengths and weaknesses as well as estimate what the organization could face by competitors actions. In addition, the model is characterized by providing a classification of strategies at the business level along with at the corporate level. The model assumed that organizations reasonably develop and adopt a different strategic behavior in order to achieve excellent compatibility with its surroundings. In his part, Daft (2001, 60) states that this model based on the idea that managers are annoying to formulate the four strategies to adapt to with the external environment and organizations seeking to achieve compatibility between the internal characteristics, strategies, and the external environment.

Miles & Snow (1978) presented four strategies to organizations on the level of business units in their study that included four fields: Printing, food, health care, and electronics. (Sultan & Akkar, 2013, 81). This classification based on the Organization's response to the three main elements: the investor problem in product and market, the engineering problem, and administrative problem (Miles et al, 1978, 546), (Girmenez, 1994, 4), (Boyne & Walker, 2004, 233), (Al-rukabi, 2004, 256), (Simon, et al., 2005, 24), (Isoherranen & Kess, 2011, 575), (Walker, 2015, 202).

Defenders:-

Are those organizations that provide a particular good or service and trying to stick them in terms of price, quality and service (Kess, Isoherranen, 2014, 1115). Furthermore, this organizations seeking more to focus on limited markets and do not tend to look for new opportunities outside their marketing respective fields. As a result of this focus, it is rarely such organizations need to make significant changes in its structure or the technology that it owns or methods of operation. Nevertheless they always seeking to improve current efficiency and it is characterized by relatively stable surroundings as this strategy is slow to change, so the defenders often resort to long-term planning (Musa, 2014, 67). Moreover, the defenders strategy is distinctive by the following characteristics (Al-Yasri, 2005, 52):

- Interest in the current market, they are trying to maintain existing customers without expanding into new markets.
- Focus on providing services and products relatively constant.
- Do not tend to creativity and innovation or expansion in research and development.
- Avoid engaging in high-risk business.
- Depends on internal efficiency and standard procedures for operations to provide high quality products to customers.
- Depend on central management style in the planning and control.
- Characterized by stability and adapt to environmental changes.
- Compete on the basis of low cost, whenever possible, delivery times and efficient service.

Prospectors:-

Organizations that follow the prospectors strategy dealing with unstable environments and more dynamic compared to other types of organizations in the same industry (Al-Qadu et al, 2007, 56), (Isoherranen & Kess, 2011, 577). This strategy follows the opposite strategy to the defenders that search for new products and new markets (Aragón-Sánchez. & Sánchez-Marín, 2005, 290). The management of this strategy focused on innovation and on two types of it firstly provide new product and secondly development of existing products. Since management offering these products on the market quickly to exploit the available opportunities. To implement this strategy it requires from management the ability to examine constantly the environmental conditions as well as the ability to predict events (Robbins, 1990, 131). Furthermore, the prospectors' strategy is distinctive by the following characteristics (Al-Yasri, 2005, 51):

- Looking for new opportunities for production and expansion of the range of goods and services.
- Looking for new markets and techniques characterized by modernity and flexibility.
- Focus on creativity, innovation and care about activities, research and development.
- Avoid entering into long-term commitments.
- It willing to face the risk.
- Depends on decentralized management style in the planning and control.
- Calls for Learning Orientation that is associated with the flexibility to face the prospect rapid changes.

Analysts:-

it is organizations that are seeking to bring a mix between the philosophy of defenders and prospectors as they work under the stable and unstable markets in order to monitor the behavior of competitors (Hamdi, 2015, 87), (Isoherranen & Kess, 2011, 578). This strategy is based on maintaining the current business and the efficiency of the work and defends on it as well as searching for new opportunities and markets across the behavior of prospector (Sultan & Akkar, 2013, 83). This strategy takes advantage of the opportunity on the basic product range and remains unchanged (Habtoor, 2007, 114). In addition, the analysts' strategy is characterized by the following characteristics (Al-Yasri, 2005, 51):

- Organization works routinely and efficiently through the formal structures and processes to retain existing customers.
- The organization works to monitor competitors along with adoption of innovation and new ideas in production to achieve growth.
- Organization relies on centralization and decentralization style to meet the requirements of its stable and changeable.
- The organization achieve a balance between efficiency that is characterized by defenders organizations and learning that is characteristic by prospectors organizations as it attempts to find a balance between stable or fixed technology requirements and flexible technologies requirements.
- Organization is thinking before entering into new business to make sure their profitability and ensuring its ability to maintain its current market share.

Respondents:-

Organizations adopt this strategy when they do not have the capacity to harmony and stability with the characteristics of the above three strategies (Robbins, 1990, 132), (Aragón-Sánchez. and Sánchez-Marín, 2005, 290). They are organizations that are trying to respond to the market in the light of possible fluctuations or achieve the reaction to the actions of the other competitors in the market (Al-Hamdi, 2015, 87). The respondents' strategy is distinctive by the following characteristics (Habtoor, 2007, 114), (Sulaiman, 2010, 72):

- The lack of a clear formulation of the organization's mission.
- Failure in the formation of strategies, policies and needed plans to develop the organization's mission into practice.
- The existence of unsuccessful attempts to modify or develop strategies appropriate to the nature of the organization environment in which it operates.
- The need for huge changes in the organizational structure that make enormous efforts to develop a strategy to cope with environment changes.
- Managers realize continuously change and the status of unsure that occur in organizational environments. However they are unable to respond to it effectively.
- The organization operates without a plan to respond to threats or when exposed to environmental stresses and thus behave randomly.

Competitive intelligence:-

In the world of business complex and permanently changing the competitive intelligence along with understanding the plans of competitors is a well-established base in the field of strategic decision-making that offers a distinctive competitive position to organizations. Moreover, identifying the competitive needs is the actual beginning of the competitive intelligence cycle that determine the priorities of the intelligence requirements for organizations that complete by the competitive intelligence path process in terms of planning, collection, analysis and dissemination of information (Abulqader, 2013, 25).

Jain (1997, 79) defines competitive intelligence as the available public information about existing or potential competitors which are used as inputs to the task in the operation of the marketing strategy of the organization. Moon (2000) states that competitive intelligence is a process of monitor and control of the competitors and their intentions in a legal and ethical ways as well as analyzed it according to the sequence of steps starts with planning and then selecting competitors then the definition of intelligence requirements and the ability to diagnose and analysis of competitors then find the two-way communication process (Al-Azawi, 2012, 10). Parker & Nites (2006, 44) confirms that competitive intelligence is the process of gathering knowledge about the external environment and turn them into competitive intelligence that provides actual assistance to the management organization in a good strategic selection. On his part (Qasim, 2011, 23) pointed out that competitive intelligence is a organized process to obtain general information about competitors and analysis it in order to distinguished targeting them in the market. Ismail & Hussein (2105, 161) concluded that competitive intelligence is a systematic ethical process for data collection and managing external information, analyzed it, and used to make decisions and formulate appropriate strategy for the organization along with strengthen its competitive advantages.

Competitive intelligence Models:-

There are three main models that presented competitive intelligence process as following (Salih et al, 2010, 360):

- Kahaner (1996) model.

- The model of professionals association.
- Calof & Dishman (2002) model.

This study will depend on Calof & Dishman (2002) model due to its comprehensiveness and the possibility of its application in the field as well as compatibility with the nature of the study problem. The model consists of six stages and they planning and focus analysis, collection, communication, structure and processes, and organisational culture and awareness (sewal, 2000, 3), (Salih et al, 2010, 362), (Hamwi & Abdullah, 2012, 893), (Ismail & Hussein, 2015, 161) and the figure below (figure 2) illustrates Calof & Dishman, 2002 model that call competitive intelligence cycle or competitive intelligence operations.

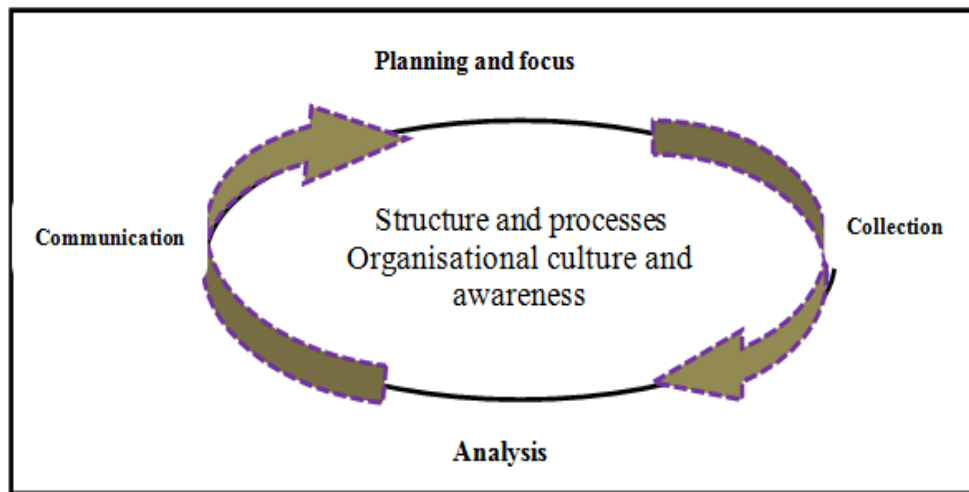


Figure 2:- competitive intelligence tool "Source: Sewall, R. (2004)"

Planning and focus:-

This stage includes identifying the information needs and bring these needs to the key intelligence topics along with giving priority to top management in that process including intelligent decision makers requirements (Ismail & Jamil, 2016, 170). This focus means that the concentration of competitive intelligence is only on the critical aspects and extremely important for the organization such as the activities and plans of competitors, the culture that competitors use, market trends, and demand (Ismail & Hussein, 2015, 162).

Collection:-

Information is the basis of competitive intelligence. In this stage information is gathering from a variety of different sources that published and unpublished legally and ethically from all possible sources. The collected data sources are storage and then tested to determine the degree of reliability and credibility (Salih et al, 2010, 365). The most important functions in this stage can be explains as following: (Srou and Mohammed, 2013, 376)

- Distribution the roles and tasks on the competitive intelligence team.
- Identify information sources.
- Determine the methods of competitive intelligence team work.

Analysis:-

This stage consider as the essence of the competitive intelligence process since at this stage the information will be analysis and interpretation by using many methods such as business environment analysis PEST (Policy, Economic, Social, Technology), and SWOT analysis (Strength, Weakness, Opportunities, Threats) (Ismail & Jamil, 2016, 171). Furthermore, this stage includes the following steps (Srou and Mohammed, 2013, 377):

- Determine the degree of reliability to the various sources of information.
- Determine the degree of accuracy of information sources.
- Connect between information and targets of competitive intelligence.
- Coordination between the various sources of information.

Communication:-

Is the process of transfer information from a person to another that the receiver can understand this information (Weihrich & Kootz,1993,712). It is transfer of meaningful information to relations' individuals (Byers & Rue,2004,13). Communication is the final results of the process of competitive intelligence as in this stage the information will be used along with present the results to decision-makers in the form of a report or in a meeting because the results of the competitive intelligence process must delivery to those individuals that owns authority and responsibility in order to interpret or converted it to final results. Furthermore, communication that link to intelligent can be done also by (E-mails, presentations, briefings, rival files, and private memoranda) (Salih et al, 2010, 367).

Structure and processes:-

Competitive intelligence requires appropriate policies and procedures as well as formal and informal infrastructure enable workers to contribute effectively to the competitive intelligence system as well as gaining benefits resulting from it (Salih et al, 2010, 367). There must be some kind of integration to the components of generating competitive intelligence process in order to not rely entirely on the technical side and ignoring the role of the human element. Since generating competitive intelligence process that not include human element cannot be able to provide managers the competitive knowledge-based on information that managers need and they expect (Hamwi & Abdullah, 2012, 895). While, operations are a mechanism that organization can accomplish its goals that include how managers address the problems of employees and problems of quality control operations (Ismail & Hussein, 2015, 162).

Organisational culture and awareness:-

In order to organization to successfully obtain benefiting from the efforts of competitive intelligence and the intelligence practice becomes a successful, organizations must support this culture within its organizing process that help it to establish in addition to the exchange of knowledge and ideas between individuals and departments within the organization. Moreover, organizations must have a high degree of organizational awareness to create a culture of competition since the high awareness of the organization environment competitive is consider being one of the basic rules of the organizational learning theory. Besides, instilling awareness / organizational culture must needs to apply the behavior and ethical standards. In order to happening this organizational must create a favorable environment for competitive intelligence that requires a continuous training to its employees. Moreover, one of the factors that helping to spread the culture of intelligence within the organization is the extent of the continuing demand of intelligence information of the various administrative levels in order to making decisions (Hamwi & Abdullah, 2012, 895).

Section Three: Results and Findings:

Section three presents a description of results and findings as the following:

Describe the personal characteristics of the study sample:-

Gender: table (2) shows the gender of the selected participants. The majority of the participants at Coca-Cola N=21 (77.8%) were male and N=6 (22.2%) were female. At the same time, the majority of the participants at Pepsi N=32 (88.9%) were male and N=4 (11.1%) were female.

Table 2:- Distribution of respondents- Gender wise.

		Coca-Cola		Pepsi	
		Frequencies	Percentage%	Frequencies	Percentage%
Gender	Male	21	77.8	32	88.9
	Female	6	22.2	4	11.1
Overall		27	100.0	36	100.0

Source: prepared by researchers based on the results of SPSS software.

Age:-

The figure bellow (3) shows the age of the participants that have been categories within four groups. The majority of the participants at Coca-Cola N=15 with (55.6%) their age are between 31-40 years, N= 9 (33.3%) their age are between 21-30 years, finally N= 3 (11.1%) are between 41-50 years. On the other hand, the majority of the

participants at Pepsi N=24 with (66.7%) their age are between 21-30 years, N=10 (27.8%) their age are between 31-40 years, finally N=2 (5.6%) are between 41-50 years.

Table 3:- Distribution of respondents - Age wise.

		Coca-Cola		Pepsi	
		Frequencies	Percentage%	Frequencies	Percentage%
Age	21 –30 years	9	33.3	24	66.7
	31 -40 years	15	55.6	10	27.8
	41 -50 years	3	11.1	2	5.6
	More than 51	0	0.0	0	0.0
Overall		27	100.0	36	100.0

Source: prepared by researchers based on the results of SPSS software.

Level of Education:-

The figure bellow (4) shows the level of education of the participants that have been categories within six groups. The majority of the participants at Coca-Cola N=24 with (88.9%) are holding bachelor degree, and the rest N= 3 (11.1%) are holding technical diploma degree. Furthermore, the majority of the participants at Pepsi N=21 with (58.3%) are holding bachelor degree. Comes second N= 7 (19.4%) are holding technical diploma degree, N= 5 (13.9%) are holding high diploma degree, N= 2 (5.6%) are holding high school degree, and finally N= 1 (2.8%) is holding PhD. degree.

Table 4:- Distribution of respondents- Level of Education wise.

		Coca-Cola		Pepsi	
		Frequencies	Percentage%	Frequencies	Percentage%
Level of Education	PhD.	0	0.0	1	2.8
	MSc.	0	0.0	0	0.0
	High Diploma	0	0.0	5	13.9
	BSc.	24	88.9	21	58.3
	Technical Diploma	3	11.1	7	19.4
	High School	0	0.0	2	5.6
Overall		27	100.0	36	100.0

Source: prepared by researchers based on the results of SPSS software.

Years of experience:-

The figure bellow (5) shows the years of experience of the participants that have been categories within six groups. The majority of the participants at Coca-Cola N=21 with (77.8%) their years of experience are between 6-10 years, N= 3 (33.3%) their years of experience are between 1-5 and 21-25 years as well. On the contrary, the majority of the participants at Pepsi N=21 with (58.3%) their years of experience are between 1-5 years, comes second N=14 (38.9%) their years of experience are between 6-10 years, and finally N=1 (2.8%) are between 11-15 years.

Table 5:- Distribution of respondents- years of experience wise.

		Coca-Cola		Pepsi	
		Frequencies	Percentage%	Frequencies	Percentage%
Years of experience	1 –5 years	3	11.1	21	58.3
	6 -10 years	21	77.8	14	38.9
	11 -15 years	0	0.0	1	2.8
	16 -20 years	0	0.0	0	0.0
	21 –25 years	3	11.1	0	0.0
	More than 26	0	0.0	0	0.0
Overall		27	100.0	36	100.0

Source: prepared by researchers based on the results of SPSS software.

Position:-

The figure bellow (6) illustrates the position of the participants. The majority of the participants at Coca-Cola N=31 with (48.1%) their position are managers and assistant managers as well, and N=1 with (3.8%) their position is supervisor. However, the majority of the participants at Pepsi N=22 with (61.1%) their position are managers, comes second and N=11 with (30.6%) their position are assistant managers, and finally N=3 with (8.3%) their position are supervisors

Table 6:- Distribution of respondents- position wise.

		Coca-Cola		Pepsi	
		Frequencies	Percentage%	Frequencies	Percentage%
Position	Managers	13	48.1	22	61.1
	Assistant managers	13	48.1	11	30.6
	Supervisors	1	3.8	3	8.3
Overall		27	100.0	36	100.0

Source: prepared by researchers based on the results of SPSS software.

Descriptive Statistics of questionnaires variables:-

The following is the details to describe the questionnaires variables that consist of business strategies related to the (Miles & Snow) model and competitive intelligence. Statistical analysis software program (SPSS, V. 20) was used in order to find the means, standard deviations, rate of agreements, and the coefficient of variance for all the questionnaires items as the following:

Description the dimensions of business strategies related to the (Miles & Snow) model:-

The table below (7) explains the means, standard deviations, rate of agreements, and the coefficient of variance of the dimensions of business alignment strategies related to the (Miles & Snow) model. The dimensions of business alignment strategic have a high means for Coca-Cola and Pepsi (4.18) (3.988) respectively, along with the total standard deviation of (1.063) (0.894) respectively. The rate of agreement for both company Coca-Cola and Pepsi was (%83.6) (%79.76) respectively, with a small dispersion in the respondents' answers as the coefficient of variance was (22.42) and (25.43) respectively. This results means that most of the respondents agree with the dimensions of business strategies related to the (Miles & Snow) model since the results came at a high level rates of compatibility strategies comparable with excellence for Coca-Cola and comes second Pepsi.

Defenders strategy:-

The table below (7) explains the means, standard deviations, rate of agreements, and the coefficient of variance of the defenders strategy. Defenders strategy for Coca-cola has a total high mean (4.15) with standard deviation of (1.039) with a rate of agreement reached (%83). On the other hand, Pepsi has a total high mean (4.02) with standard deviation of (.908) with a rate of agreement reached (%80.4) with excellence for Coca-Cola comparing to Pepsi

company. This result shows a high level of reliance to this dimension. Regarding to the items, the most elements that have contributed to enrichment the defenders strategy relating to Coca-Cola company depending on the percentage of the respondents agreement is X2 which states that "our company is seeking to produce high quality products outweigh products of the competitors." that supports the values of mean and standard deviation of (4.48, 1.014) respectively. However, relating to Pepsi company X1 that states "our company adopts defensive technique to curb the actions of competitors in the markets." is the most elements that have contributed to enrichment the defenders strategy that supports the values of mean and standard deviation of (4.44, .735) respectively

Prospectors' strategy:-

prospectors' strategy for Coca-cola has a total high mean (4.20) with standard deviation of (1.065) with a rate of agreement reached (%84). In contrast, Pepsi has a total high mean (4.03) with standard deviation of (.940) with a rate of agreement reached (%80.6) with excellence for Coca-Cola comparing to Pepsi company. This result shows a high level of reliance to this dimension. Regarding to the items, the most elements that have contributed to enrichment the prospectors' strategy relating to Coca-Cola company depending on the percentage of the respondents agreement is X8 which states that "the basic philosophy of our company is to win new customers to provide a variety of products constantly." that supports the values of mean and standard deviation of (4.56, 1.013) respectively. On the other hand, relating to Pepsi company X6 that states "our company is looking for the environmental market opportunities constantly through the knowledge of the demand for their products trends." is the most elements that have contributed to enrichment the prospectors' strategy that supports the values of mean and standard deviation of (4.33, .894) respectively

Analysts' strategy:-

Analysts' strategy for Coca-cola has a total high mean (4.27) with standard deviation of (1.019) with a rate of agreement reached (%85.4). On the contrary, Pepsi has a total high mean (4.002) with standard deviation of (.817) with a rate of agreement reached (%80.04) with excellence for Coca-Cola comparing to Pepsi company. This result shows a high level of reliance to this dimension. Regarding to the items, the most elements that have contributed to enrichment the analysts' strategy relating to both Coca-Cola and Pepsi company depending on the percentage of the respondents agreement is X12 which states that "our company is interested by all systems that will achieve a reduction in the cost of their products." that supports the values of mean of (4.56, 4.47) respectively, along with standard deviation of (1.013, .560) respectively.

Respondents' strategy:-

Respondents' strategy for Coca-cola has a total high mean (4.12) with standard deviation of (1.131) with a rate of agreement reached (%82.4). On the contrary, Pepsi has a total high mean (3.90) with standard deviation of (.912) with a rate of agreement reached (%78) with excellence for Coca-Cola comparing to Pepsi company. This result shows a high level of reliance to this dimension. Regarding to the items, the most elements that have contributed to enrichment the respondents' strategy relating to Coca-Cola company depending on the percentage of the respondents agreement is X19 which states that "our company is interested in following up customers needs to identify their complaints as a prelude to respond to them at right time" that supports the values of mean and standard deviation of (4.30, 1.103) respectively. However, relating to Pepsi company X18 that states "our company attaches great importance to follow up the technological changes in order to respond to the changes of customers' needs." is the most elements that have contributed to enrichment the respondents' strategy that supports the values of mean and standard deviation of (4.08, .937) respectively.

Table 7:- Means, standard deviations, rate of agreements, and the coefficient of variance of the dimensions of business alignment strategies related to the (Miles & Snow) model.

Items	Pepsi					Coca-Cola				
	Mean	standard deviation	Rate of Agreement	Coefficient of variance%	rank	Mean	standard deviation	Rate of Agreement	Coefficient of variance%	Rank
X1	4.44	.735	88.8	16.55	1	3.70	1.353	74	36.56	5
X2	4.36	.683	87.2	15.66	2	4.48	1.014	89.6	22.63	1
X3	3.89	1.141	79.6	29.33	3	4.30	.993	86	23.09	3
X4	3.67	.986	73.4	26.86	5	4.37	1.006	87.4	23.02	2
X5	3.75	.996	75	26.56	4	3.93	.829	78.6	21.09	4
Defenders	4.02	.908	80.4	22.58		4.15	1.039	83	25.26	
X6	4.33	.894	86.6	20.64	1	4.37	1.006	87.4	23.02	2
X7	3.94	.984	78.8	24.97	3	4.37	1.006	87.4	23.02	2
X8	4.08	1.079	81.6	26.44	2	4.56	1.013	91.2	22.21	1
X9	3.89	.887	77.8	22.80	4	4.19	.962	83.8	22.95	3
X10	3.94	.860	78.8	21.82	3	3.52	1.341	70.4	38.09	4
Prospectors	4.03	.940	80.6	23.32		4.20	1.065	84	25.35	
X11	4.06	1.094	81.2	26.94	2	4.22	.974	84.4	23.08	3
X12	4.47	.560	89.4	12.52	1	4.56	1.013	91.2	22.21	1
X13	4.03	.609	80.6	15.11	3	4.07	1.141	81.4	28.03	4
X14	3.78	.898	75.6	23.75	4	4.30	.993	86	23.09	2
X15	3.67	.926	73.4	25.23	5	4.22	.974	84.4	23.08	3
Analysts	4.002	.817	80.04	20.42		4.27	1.019	85.4	23.84	
X16	4.06	1.013	81.2	24.95	2	3.81	1.302	76.2	34.17	4
X17	3.94	.955	78.8	24.23	3	4.26	1.059	85.2	24.85	2
X18	4.08	.937	81.6	22.96	1	4.26	.984	85.2	23.09	2
X19	3.75	.770	75	20.53	4	4.30	1.103	86	25.65	1
X20	3.69	.889	73.8	24.09	5	4.00	1.209	80	30.22	3
Respondents	3.90	.912	78	23.38		4.12	1.131	82.4	27.45	
Strategic business alignment	3.988	.894	79.76	22.42		4.18	1.063	83.6	25.43	

Source: prepared by researchers based on the results of SPSS software.

Based on the above, table (8) illustrates the arranging of strategic alignment of business strategies related to the (Miles & Snow) model for both company Coca-Cola and Pepsi. Coca-Cola depends first on analysts' strategy, then prospectors' strategy, after that defenders strategy and finally respondents' strategy. On the other hand, Pepsi depends first on prospectors' strategy, then defenders strategy, after that analysts' strategy and finally respondents' strategy.

Table 8:- Arranging of strategic alignment of business strategies related to the (Miles & Snow) model for both company Coca-Cola and Pepsi.

Items	Pepsi					Coca-Cola				
	Mean	standard deviation	Rate of Agreement	Coefficient of variance%	rank	Mean	standard deviation	Rate of Agreement	Coefficient of variance%	Rank
Defenders	4.02	.908	80.4	22.58	2	4.15	1.039	83	25.26	3
Prospectors	4.03	.940	80.6	23.32	1	4.20	1.065	84	25.35	2
Analysts	4.002	.817	80.04	20.42	3	4.27	1.019	85.4	23.84	1
Respondents	3.90	.912	78	23.38	4	4.12	1.131	82.4	27.45	4
Strategic Business Alignment	3.988	.894	79.76	22.42		4.18	1.063	83.6	25.43	

Source: prepared by researchers based on the results of SPSS software.

Description the dimensions of competitive intelligence:-

The table below (9) explains the means, standard deviations, rate of agreements, and the coefficient of variance of the dimensions of competitive intelligence. The dimensions of competitive intelligence have a high means for Coca-Cola and Pepsi (4.21) (3.88) respectively, along with the total standard deviation of (1.003) (0.839) respectively. The rate of agreement for both company Coca-Cola and Pepsi was (%84.2) (%77.6) respectively, with a small dispersion in the respondents' answers as the coefficient of variance was (23.82) and (21.62) respectively. This results means that most of the respondents agree with the dimensions of competitive intelligence since the results came at a high level rates of compatibility strategies comparable with excellence for Coca-Cola and comes second Pepsi.

Planning and focus:-

This dimension for Coca-cola has a total high mean (4.11) with standard deviation of (1.040) with a rate of agreement reached (%82.2). On the contrary, Pepsi has a total high mean (4.01) with standard deviation of (.812) with a rate of agreement reached (%80.2) with excellence for Coca-Cola comparing to Pepsi company. This result shows a high level of reliance to this dimension. Regarding to the items, the most elements that have contributed to enrichment the planning and focus relating to Coca-Cola company depending on the percentage of the respondents agreement is Y5 which states that "our company owns a number of ways to provide the requirements of competitive intelligence plans." that supports the values of mean and standard deviation of (4.26, .984) respectively. On the other hand, relating to Pepsi company Y1 that states "our company is largely concerned with strategies to organizations that compete with them" is the most elements that have contributed to enrichment the planning and focus that supports the values of mean and standard deviation of (4.22, .637) respectively.

Collection:-

This dimension for Coca-cola has a total high mean (4.08) with standard deviation of (1.000) with a rate of agreement reached (%81.6). On the other hand, Pepsi has a total high mean (3.88) with standard deviation of (.885) with a rate of agreement reached (%77.6) with excellence for Coca-Cola comparing to Pepsi company. This result shows a high level of reliance to this dimension. Regarding to the items, the most elements that have contributed to enrichment this dimension relating to Coca-Cola company depending on the percentage of the respondents agreement is Y7 which states that "our company relies on efficient staff to work in the marketing intelligence system to collect data and information from competitors." that supports the values of mean and standard deviation of (4.26, .984) respectively. On the other hand, relating to Pepsi company Y6 that states " is the most elements that have contributed to enrichment this dimension that supports the values of mean and standard deviation of (4.14, .833) respectively.

Analysis:- This dimension for Coca-cola has a total high mean (4.17) with standard deviation of (.998) with a rate of agreement reached (%83.4). In contrast, Pepsi has a total high mean (3.87) with standard deviation of (.847) with a rate of agreement reached (%77.4) with excellence for Coca-Cola comparing to Pepsi company. This result shows a

high level of reliance to this dimension. Regarding to the items, the most elements that have contributed to enrichment this dimension relating to Coca-Cola company depending on the percentage of the respondents agreement is Y12 which states that "our company adopts the analysis of strengths, weaknesses, opportunities and threats to identify the company's competitive situation." that supports the values of mean and standard deviation of (4.56, 1.050) respectively. On the other hand, relating to Pepsi company Y14 that states "our company is following the information published on the internet for the analysis of research and development programs of the competitors." is the most elements that have contributed to enrichment this dimension that supports the values of mean (4.17).

Communication:-

This dimension for Coca-cola has a total high mean (4.31) with standard deviation of (.997) with a rate of agreement reached (%86.2). On the other hand, Pepsi has a total high mean (4.05) with standard deviation of (.775) with a rate of agreement reached (%81) with excellence for Coca-Cola comparing to Pepsi company. This result shows a high level of reliance to this dimension. Regarding to the items, the most elements that have contributed to enrichment this dimension relating to both company Coca-Cola and Pepsi depending on the percentage of the respondents agreement is Y17 which states that "our company is seeking to develop an effective communications system to gain potential customers." that supports the values of mean and standard deviation of (4.48, 1.014) respectively for Coca-cola and (4.31, .668) for Pepsi.

Structure and processes:-

This dimension for Coca-cola has a total high mean (4.27) with standard deviation of (.993) with a rate of agreement reached (%85.4). On the contrary, Pepsi has a total high mean (3.71) with standard deviation of (.892) with a rate of agreement reached (%74.2) with excellence for Coca-Cola comparing to Pepsi company. This result shows a high level of reliance to this dimension. Regarding to the items, the most elements that have contributed to enrichment the structure and processes relating to Coca-Cola company depending on the percentage of the respondents agreement is Y25 which states that "our company depends on efficient teams to explore, develop, and improve our product portfolio and operations." that supports the values of mean and standard deviation of (4.37, 1.006) respectively. On the other hand, relating to Pepsi company Y21 that states "our company relies on a flexible organizational structure in line with contemporary developments in the field of competitive intelligence." is the most elements that have contributed to enrichment the structure and processes that supports the values of mean and standard deviation of (4.03, .810) respectively.

Organisational culture and awareness:-

This dimension for Coca-cola has a total high mean (4.32) with standard deviation of (.992) with a rate of agreement reached (%86.4). On the contrary, Pepsi has a total high mean (3.81) with standard deviation of (.827) with a rate of agreement reached (%76.2) with excellence for Coca-Cola comparing to Pepsi company. This result shows a high level of reliance to this dimension. Regarding to the items, the most elements that have contributed to enrichment the organisational culture and awareness relating to Coca-Cola company depending on the percentage of the respondents agreement is Y26 and Y29 which they states that "our company sees competitive intelligence as the core values of its culture." and "in our company beliefs encourages most of the activities related to competitive intelligence" that supports the values of mean (4.41). On the other hand, relating to Pepsi company Y27 that states "the organizational culture in our company based on the consideration to workers as customers and makes the satisfaction as their priorities" is the most elements that have contributed to enrichment the organisational culture and awareness that supports the values of mean and standard deviation of (3.94, .826) respectively.

Table 9:- means, standard deviations, rate of agreements, and the coefficient of variance of the dimensions of competitive intelligence

Items	Pepsi					Coca-Cola				
	Mean	standard deviation	Rate of Agreement	Coefficient of variance%	Rank	Mean	standard deviation	Rate of Agreement	Coefficient of variance%	rank
Y1	4.22	.637	84.4	15.09	1	4.11	1.121	82.2	27.27	3
Y2	3.94	.754	78.8	19.13	4	4.15	1.064	83	25.63	2
Y3	4.03	.910	80.6	22.58	3	4.00	1.000	80	25.00	5
Y4	4.19	.749	83.8	17.87	2	4.07	1.035	81.4	25.42	4
Y5	3.67	1.014	73.4	27.62	5	4.26	.984	85.2	23.09	1
Planning and focus	4.01	.812	80.2	20.26		4.11	1.040	82.2	25.30	
Y6	4.14	.833	82.8	20.12	1	4.07	.997	81.4	24.49	3
Y7	3.78	.939	75.6	25.37	4	4.26	.984	85.2	23.09	1
Y8	3.81	1.009	76.2	26.48	3	4.11	.934	82.2	22.72	2
Y9	3.61	.688	72.2	19.05	5	3.89	1.050	77.8	26.38	4
Y10	4.08	.937	81.6	22.96	2	4.07	1.035	81.4	25.42	3
Collection	3.88	.885	77.6	22.81		4.08	1.000	81.6	24.50	
Y11	3.86	.899	77.2	23.29	3	4.26	.984	85.2	23.09	2
Y12	3.58	.996	71.6	27.82	4	4.56	1.050	91.2	23.02	1
Y13	3.92	.732	78.4	18.67	2	4.11	.934	82.2	22.72	4
Y14	4.00	.717	80	17.92	1	3.78	1.050	75.6	27.77	5
Y15	4.00	.894	80	22.35	1	4.22	.974	84.4	23.08	3
Analysis	3.87	.847	77.4	21.90		4.17	.998	83.4	23.93	
Y16	3.92	.770	78.4	19.84	4	4.15	.949	83	22.86	4
Y17	4.31	.668	86.2	15.49	1	4.48	1.014	89.6	22.63	1
Y18	4.17	.561	83.4	13.45	2	4.30	.993	86	23.09	3
Y19	4.08	.841	81.6	20.61	3	4.30	1.031	86	23.97	3
Y20	3.81	1.037	76.2	27.21	5	4.33	1.000	86.6	23.09	2
Communication	4.05	.775	81	19.14		4.31	.997	86.2	23.13	
Y21	4.03	.810	80.6	20.09	1	4.22	1.013	84.4	24.00	4
Y22	3.97	.654	79.4	16.47	2	4.19	.962	83.8	22.95	5
Y23	3.50	.737	70	21.05	4	4.33	1.000	86.6	23.09	2
Y24	3.58	1.105	71.6	30.86	3	4.26	.984	85.2	23.09	3
Y25	3.47	1.158	69.4	33.37	5	4.37	1.006	87.4	23.02	1
Structure and processes	3.71	.892	74.2	24.06		4.27	.993	85.4	23.25	
Y26	3.89	.785	77.8	20.17	2	4.41	1.010	88.2	22.90	1
Y27	3.94	.826	78.8	20.96	1	4.37	1.006	87.4	23.02	2
Y28	3.72	.779	74.4	20.94	4	4.11	.934	82.2	22.72	4
Y29	3.69	.856	73.8	23.19	5	4.41	1.010	88.2	22.90	1
Y30	3.81	.889	76.2	23.33	3	4.33	1.000	86.4	23.09	3
Organisational culture and awareness	3.81	.827	76.2	21.70		4.32	.992	86.4	22.96	
Competitive Intelligence	3.88	.839	77.6	21.62		4.21	1.003	84.2	23.82	

Source: prepared by researchers based on the results of SPSS software.

Based on the above, table (10) illustrates the arranging of competitive intelligence for both companies Coca-Cola and Pepsi. Coca-Cola depends first on organisational culture and awareness, then communication, after that structure

and processes and finally collection. In contrast, Pepsi depends first on communication, then planning and focus, after that collection and finally structure and processes.

Table 10:- the arranging of competitive intelligence

Items	Pepsi					Coca-Cola				
	Mean	standard deviation	Rate of Agreement	Coefficient of variance%	Rank	Mean	standard deviation	Rate of Agreement	Coefficient of variance%	rank
Planning and focus	4.01	.812	80.2	20.26	2	4.11	1.040	82.2	25.30	5
Collection	3.88	.885	77.6	22.81	3	4.08	1.000	81.6	24.50	6
Analysis	3.87	.847	77.4	21.90	4	4.17	.998	83.4	23.93	4
Communication	4.05	.775	81	19.14	1	4.31	.997	86.2	23.13	2
Structure and processes	3.71	.892	74.2	24.06	6	4.27	.993	85.4	23.25	3
Organisational culture and awareness	3.81	.827	76.2	21.70	5	4.32	.992	86.4	22.96	1
Competitive Intelligence	3.88	.839	77.6	21.62		4.21	1.003	84.2	23.82	

Source: prepared by researchers based on the results of SPSS software.

Testing the correlation between study variables:-

The first hypothesis states that there is a measurable positive correlation between strategic alignments of business strategies related to the (Miles & Snow) model and competitive intelligence. To test the correlation among the study variables, Pearson's correlation (r) was calculated. Table (11) illustrates that business alignment strategies related to the (Miles & Snow) model were positively correlated with competitive intelligence as the value of the correlation coefficient related to Coca-Cola was high (.927**) reached the value of significant at (0.000) and at the level of significance (0.01). Moreover, Table (11) illustrates that business alignment strategies related to the (Miles & Snow) model were positively correlated with competitive intelligence as the value of the correlation coefficient related to Pepsi company was above moderate (.631**) reached the value of significant at (0.000) and at the level of significance (0.01) based on managers respondents. Consequently, the first hypotheses accepted.

Table 11:- correlation between strategic alignments of business strategies related to the (Miles & Snow) model and Competitive Intelligence.

	Competitive Intelligence		
	Statistics	Pepsi	Coca-Cola
Strategic alignment of business strategies related to the (Miles & Snow) model	The correlation coefficient	.631**	.927**
	Potential value	.000	.000
	The sample size	36	27

**. Correlation is significant at the 0.01 level (2-tailed).

Source: prepared by researchers based on the results of SPSS software.

A number of sub-hypotheses is derived from the main first hypotheses that looking to test the correlation between dimensions of business alignment strategies related to the (Miles & Snow) model that represented by (defenders, prospectors, analysts, respondents) and competitive intelligence.

- **Coca-Cola:** table (12) shows that defenders strategy were positively correlated with competitive intelligence as the value of the correlation coefficient was high (.909^{**}). Table (12) shows that prospectors strategy achieved the highest positive correlation with competitive intelligence as the value of the correlation coefficient was high (.935^{**}). Furthermore, table (12) shows that analysts strategy were positively correlated with competitive intelligence as the value of the correlation coefficient was high (.903^{**}). Finally, respondents' strategy has the weakest positive correlation with competitive intelligence (.789^{**}). The value of significant for the four correlation reached at (0.000) and at the level of significance (0.01).
- **Pepsi:** table (12) shows that defenders strategy were positively correlated with competitive intelligence as the value of the correlation coefficient was bellow moderate (.463^{**}). The value of significant reached at (0.004) and at the level of significance (0.01). Prospectors strategy were positively correlated with competitive intelligence (.402^{*}). The value of significant reached at (0.015) and at the level of significance (0.01). In addition, table (12) shows that analysts strategy has the weakest positive correlation with competitive intelligence as the value of the correlation coefficient was below moderate (.391^{*}). The value of significant reached at (0.018) and at the level of significance (0.01). Finally, table (12) shows that Respondents' strategy achieved the highest positive correlation with competitive intelligence as the value of the correlation coefficient was high (.520^{**}). As a result, the four sub-hypotheses that derived from the main first hypotheses were accepted as well.

Correlation between the dimensions of business alignment strategies related to the (Miles & Snow) model and competitive intelligence

	Competitive Intelligence		
	Statistics	Pepsi	Coca-Cola
Defenders strategy	The correlation coefficient	.463 ^{**}	.909 ^{**}
	Potential value	.004	.000
Prospectors strategy	The correlation coefficient	.402 [*]	.935 ^{**}
	Potential value	.015	.000
Analysts strategy	The correlation coefficient	.391 [*]	.903 ^{**}
	Potential value	.018	.000
Respondents strategy	The correlation coefficient	.520 ^{**}	.789 ^{**}
	Potential value	.001	.000

^{**}. Correlation is significant at the 0.01 level (2-tailed).

^{*}. Correlation is significant at the 0.05 level (2-tailed).

Source: prepared by researchers based on the results of SPSS software.

Testing the impact hypotheses between study variables:-

This study conducted a multiple linear regression analysis in order to examine the impact of business alignment strategies related to the (Miles & Snow) model that represented by (defenders, prospectors, analysts, respondents) with competitive intelligence.

Coca-Cola:-

Table (13) demonstrates that business alignment strategies related to the (Miles & Snow) model illustrates 86% of the competitive intelligence as characterized by the (R^2). The significance value is 0.000 which is less than 0.05 at the degree freedom of (1, 25). Thus, the model is statistically significance in predicting how the business alignment

strategies related to the (Miles & Snow) model impacts on competitive intelligence. At 5% level of significance, the F calculated was 153.839 and it is greater than F tabulated (4.24), which explains that the overall model was significant.

Table 13:- the impact of business alignment strategies related to the (Miles & Snow) model on competitive intelligence (Coca-Cola)

	Competitive Intelligence				
	R ²	T		F	
		Tabulated	Calculated	Tabulated	Calculated
Strategic alignment of business strategies related to the (Miles & Snow) model	.860	2.78	12.403 (.000)	4.24	153.839 (.000)

Source: prepared by researchers based on the results of SPSS software.

Pepsi: table (14) shows that business alignment strategies related to the (Miles & Snow) model illustrate 39.8% of the competitive intelligence as characterized by the (R²). The significance value is 0.000 which is less than 0.05. Therefore, the model is statistically significant in predicting how the business alignment strategies related to the (Miles & Snow) model impacts on competitive intelligence. At 5% level of significance, the F calculated was 22.464 and it is greater than its F tabulated (4.24), which explains that the overall model was significant. Consequently, the second main hypothesis is accepted that states "there is a statistically significant impact of business alignment strategies related to the (Miles & Snow) on competitive intelligence".

Table 14:- the impact of business alignment strategies related to the (Miles & Snow) model on competitive intelligence (Pepsi)

	Competitive Intelligence				
	R ²	T		F	
		Tabulated	Calculated	Tabulated	Calculated
Strategic alignment of business strategies related to the (Miles & Snow) model	.398	2.42	4.740 (.000)	4.11	22.464 (.000)

Source: prepared by researchers based on the results of SPSS software

A number of sub-hypotheses is derived from the main second hypotheses that looking to test the impact of dimensions of business alignment strategies related to the (Miles & Snow) model that represented by (defenders strategy, prospectors strategy, analysts strategy, respondents strategy) on competitive intelligence.

Coca-Cola: table (15) shows that defenders strategy illustrates 82.7% of the competitive intelligence as characterized by the (R²). The significance value is 0.000 which is less than 0.05. At 5% level of significance, the F calculated was 119.469 and it is greater than its F tabulated (4.11). Besides, prospectors' strategy illustrates 87.4% of the competitive intelligence as characterized by the (R²) that consider being the strongest dimensional impact on the competitive intelligence. The significance value is 0.000 which is less than 0.05. At 5% level of significance, the F calculated was 173.617 and it is greater than its F tabulated (4.11). As well, analysts' strategy illustrates 81.5% of the competitive intelligence as characterized by the (R²). The significance value is 0.000 which is less than 0.05. At 5% level of significance, the F calculated was 110.197 and it is greater than its F tabulated (4.11). Finally, respondents' strategy illustrates 62.2% of the competitive intelligence as characterized by the (R²) that consider being the weakest dimensional impact on the competitive intelligence. The significance value is 0.000 which is less than 0.05. At 5% level of significance, the F calculated was 41.206 and it is greater than its F tabulated (4.11).

Table 15:- the impact of dimensions of business alignment strategies related to the (Miles & Snow) model on competitive intelligence (Coca-Cola)

	Competitive intelligence				
	R ²	T		F	
		Tabulated	Calculated	Tabulated	Calculated
Defenders strategy	.827	2.78	10.930 (.000)	4.24	119.469 (.000)
Prospectors strategy	.874	2.78	13.176 (.000)	4.24	173.617 (.000)
Analysts strategy	.815	2.78	10.497 (.000)	4.24	110.197 (.000)
Respondents strategy	.622	2.78	6.419 (.000)	4.24	41.206 (.000)

Source: prepared by researchers based on the results of SPSS software.

Pepsi: table (16) shows that defenders strategy illustrates 21.4% of the competitive intelligence as characterized by the (R²). The significance value is 0.004 which is less than 0.05. At 5% level of significance, the F calculated was 9.260 and it is greater than its F tabulated (2.42). In addition, prospectors' strategy illustrates 16.2% of the competitive intelligence as characterized by the (R²). The significance value is 0.015 which is less than 0.05. At 5% level of significance, the F calculated was 6.569 and it is greater than its F tabulated (2.42). Additionally, analysts' strategy illustrates 15.3% of the competitive intelligence as characterized by the (R²) that consider being the weakest dimensional impact on the competitive intelligence. The significance value is 0.000 which is less than 0.05. At 5% level of significance, the F calculated was 6.138 and it is greater than its F tabulated (2.42). Finally, respondents' strategy illustrates 27.1% of the competitive intelligence as characterized by the (R²) that consider being the strongest dimensional impact on the competitive intelligence. The significance value is 0.000 which is less than 0.05. At 5% level of significance, the F calculated was 12.617 and it is greater than its F tabulated (2.42). Consequently, the four sub-hypotheses that derived from the main second hypotheses were accepted as well.

Table 16:- the impact of dimensions of business alignment strategies related to the (Miles & Snow) model on competitive intelligence (Pepsi)

	Competitive intelligence				
	R ²	T		F	
		Tabulated	Calculated	Tabulated	Calculated
Defenders strategy	.214	2.42	3.043 (.004)	4.11	9.260 (.004)
Prospectors strategy	.162	2.42	2.563 (.015)	4.11	6.569 (.015)
Analysts strategy	.153	2.42	2.478 (.018)	4.11	6.138 (.018)
Respondents strategy	.271	2.42	3.552 (.001)	4.11	12.617 (.001)

Source: prepared by researchers based on the results of SPSS software.

Section four: Conclusions and recommendations

Conclusions:-

- The Coca-Cola Company focuses on analysts' strategy first then Prospectors after that defenders' strategy and finally respondents' strategy among the four available strategies alignment of business strategies related to the (Miles & Snow) model. On the other hand, Pepsi Company focuses on Prospectors' strategy first then both in the same level analysts' strategy defenders' strategy and finally respondents' strategy in response to the market in the light of possible fluctuations or achieves the reaction of the other competitors actions in the market which can be notice from the means of the four available strategies alignment of business strategies related to the (Miles & Snow) model.
- The results of answers respondents analysis in both companies was positive and they agree with strategies alignment of business strategies related to the (Miles & Snow) model and the dimensions of competitive intelligence. As the results of means for both companies have high-level rates of compatibility strategies comparable with excellence for Coca-Cola and comes second Pepsi.
- Among the companies sample study competitiveness for the exercise of its business. This is an indication of its ability to deliver varied products and services even though the existence of competition in the environment in the city of Erbil in Iraqi Kurdistan region in this area.
- Results of the analysis found a positive correlation between strategies alignment of business strategies related to the (Miles & Snow) model and competitive intelligence as well as between the four dimensions of strategies alignment of business strategies related to the (Miles & Snow) model (defenders, prospectors, analysts, respondents) and competitive intelligence.
- Results of the analysis found an impact of strategies alignment of business strategies related to the (Miles & Snow) model on competitive intelligence that have a reflection to the organizations help them able to conditioning potential according to the changes that occur in the environmental elements.

Recommendations:-

- Companies have operate to develop specialized units to environmental analysis attempts to examine and evaluate changes in environmental trends in general and competitive environmental in particular in order to cope with the changes that occur in the environment that have effects on its products and services along with its profit and survive.
- The need of the company's management in increased interesting in competitive intelligence in the context of being an integrated system since it has a positive impact in the organizations sample study.
- Organizations should gives more attention to environmental information and managing it along with directing efforts towards access to the requirement quality as it proves in the applied fields that have a huge impact in determining the future strategic direction for organizations through its contribution in defining business strategies alignment related to the (Miles & Snow) model.
- The need to focus on competitive intelligence within the standards, legal and ethical dimensions in order to complete the capabilities of this type of intelligence as it own the ability to seize opportunities and avoid environmental threats in addition to work according to data imposed by the competitive environment.
- Organizations should paying more attention to search about the customer's needs and desires about offered products and services and that offered by its competitors and their suitability for their desires along with the most important customer's notes as it has a role in discovery of competitors plans and take advantage of it in adopting strategies alignment of business strategies related to the (Miles & Snow) model in the future.

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Questionnaire:-

Personal Characteristics	
Gender	Male
	Female
Age	21 –30 years
	31 -40 years
	41 -50 years
	More than 51
Level of Education	PhD.
	MSc.
	High Diploma
	BSc.
	Technical Diploma
	High School
Years of experience	1 –5 years
	6 -10 years
	11 -15 years
	16 -20 years
	21 –25 years
	More than 26
Position	Managers
	Assistant managers
	Supervisors

	Strategic alignment of business strategies related to the (Miles & Snow) model	Strongly agree (5)	Agree (4)	Uncertain (3)	Strongly Disagree (2)	Disagree (1)
Defenders Strategy						
1	Our company adopts defensive technique to curb the actions of competitors in the markets.					
2	Our company is seeking to produce high quality products outweigh products of the competitors.					
3	Our company keeps on its products unchanged.					
4	Our company directed its business in order to be a leader in specific areas of the market.					
5	Our company is seeking to offer a limited range of products to customers, compared with competitors.					

Prospectors strategy						
6	Our company is looking for the environmental market opportunities constantly through the knowledge of the demand for their products trends.					
7	Our company bears high risks to provide better products for customers.					
8	The basic philosophy of our company is to win new customers to provide a variety of products constantly.					
9	Our company develops its products through the schemes of providing the service by adopting modern techniques.					
10	Our company would like to reschedule competitive acts continuously.					
Analysts strategy						
11	Our company analyzes in-depth the analysis ways by the decision-making related to changing technologies to provide the product.					
12	Our company is interested by all systems that will achieve a reduction in the cost of their products					
13	Our company keep pace with the case of competition with similar organizations across align the prices of their products and the ability of customers to the acquisition.					
14	Our company adopts all available methods to obtain information about customers' attitudes toward its product.					
15	Our company adopts the means of analysis that contribute to the assessment of its competitive position compared to similar organizations.					
Respondents strategy						
16	Our company is interested in making a change in its product in response to the effects of environmental pressures.					
17	Our company is characterized by possessing the ability to reduce its product prices in response to a change in the prevailing market prices.					
18	Our company attaches great importance to follow up the technological changes in order to respond to the changes of customers' needs.					
19	Our company is interested in following up customers needs to identify their complaints as a prelude to respond to them at right time					
20	Our company put its products at competitive characteristics in order to respond to environmental change.					

Competitive intelligence		Strongly agree (5)	Agree (4)	Uncertain (3)	Strongly Disagree (2)	Disagree (1)
planning and focus						
1	Our company is largely concerned with strategies to					

	organizations that compete with them.					
2	Our company constantly monitoring the technology used by the competing organizations.					
3	Our company is interested to monitor and evaluate the activities and plans of competing organizations.					
4	Our company holds regular meetings with workers to explain the competitive intelligence plans.					
5	Our company owns a number of ways to provide the requirements of competitive intelligence plans.					
Collection						
6	Workers are back to the information base through computers to solve problems and discover opportunities.					
7	Our company relies on efficient staff to work in the marketing intelligence system to collect data and information from competitors.					
8	Our company is keen on updating its information continuously on the movements of competitors.					
9	Our company collects information about the administrative style adopted by competitors.					
10	Our company adopts in its decisions on strategic marketing intelligence.					
Analysis						
11	Our company is keen on a regular basis in the formulation of research and development and analysis of new products and plans of its competitors					
12	Our company adopts the analysis of strengths, weaknesses, opportunities and threats to identify the company's competitive situation.					
13	Our company analyzes the strategies and directions that competitors may adopt in the future.					
14	Our company is following the information published on the internet for the analysis of research and development programs of the competitors.					
15	our company analyze the information foundation that achieve the organization a competitive advantage					
Communication						
16	Our company provides information relating to the competitive intelligence to all employees in our company.					
17	Our company is seeking to develop an effective communications system to gain potential customers.					
18	Our company has effective staffs that communicate with customers and they deal with them.					
19	Our company is seeking to improve communication technology as a key to achieve superiority over its competitors.					
20	Our company is seeking to develop effective communications systems to gain potential customers.					
Structure and processes						
21	Our company relies on a flexible organizational structure in line with contemporary developments in the field of competitive intelligence.					
22	Our company depends on based organizational structure that enhances the performance of employees.					
23	The company provides requirement competitive intelligence operations to employees.					
24	Our company used technology operations to help the					

	understanding the relevant customer.					
25	Our company depends on efficient teams to explore, develop, and improve our product portfolio and operations.					
Organisational culture and awareness						
26	Our company sees competitive intelligence as the core values of its culture.					
27	The organizational culture in our company based on the consideration to workers as customers and makes the satisfaction as their priorities.					
28	The culture of our company encourages creativity based on competitive intelligence.					
29	In our company beliefs encourages most of the activities related to competitive intelligence					
30	Our company promotes a culture of teamwork and cooperation between workers in the field of competitive intelligence.					

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RESEARCH ARTICLE

A CLINICAL STUDY TO COMPARE THE EFFICACY OF AUTOLOGUS PLATELET RICH PLASMA (PRP) VERSUS NORMAL SALINE DRESSING IN CHRONIC NON-HEALING ULCERS

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Abstract

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Key words:-

Chronic non healing ulcers; Autologous platelet rich plasma (PRP); platelet derived growth factors; wound reduction.

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Introduction:-

Skin ulcers are defined by the loss of tissue that includes the epidermis and the dermis, at times affecting the adipose tissue and the muscle fascia. There is not spontaneous recovery and the tissue lesion many times results in a fibrous scar [1]. Skin ulceration is a clinical problem, with an incidence of 0.78 %. The financial cost of treatment of such wounds is enormous [2]. Several etiological factors are associated to the development of this condition, such as: chronic venous disease, peripheral arterial disease, neuropathy, physical trauma, hematologic disorders, skin infection, inflammatory diseases, neoplasia, nutritional and iatrogenic alterations. The healing process is dynamic and involves complex events that include hemostasis, inflammation, granulation tissue formation, epithelialization, neovascularization, collagen synthesis, and wound contraction. It involves a sequence of events that begin at the moment of injury and continue for several subsequent months and can be divided into three stages: inflammation, proliferation and remodeling [3]. Several experimental clinical studies have demonstrated the reduction of growth factors of chronic wounds. Platelet aggregation has the leading role in the process of skin healing since it is responsible for releasing growth factors, adhesion molecules and lipids, which regulate migration, proliferation and function of keratinocytes, fibroblasts and endothelial cells [4, 5]. The analysis of supernatant of chronic wounds compared to acute wounds revealed a considerable decrease of the growth factors in the former, observing a quick metabolism of the growth factors due to proteases found in the wound, of bacterial or cellular source. On the other hand, in diabetic ulcers and due to venous insufficiency, the decrease of the growth factors occurs due to a mechanism of sequestration by fibrin around the capillaries [6]. Therefore, biological stimulation of the wound with platelet rich plasma (PRP) (which drives the most important factors for skin and tissue repair) has been recently introduced as a possible alternative to complex surgical methods.

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Autologous Platelet-rich plasma (PRP):-

The application of PRP has been documented first promoted by M. Ferrari in 1987 as an autologous transfusion component after an open heart operation to avoid homologous blood product transfusion.

PRP can be defined as the volume fraction of blood plasma contains high concentration of platelets, from a baseline serum level. PRP is mainly a platelet concentrate with minor proportions white blood cells and other blood components [10]. A concentration of 1,407,640 cells/ μ l (with a standard deviation of 320,100) has been suggested as ideal for the definition of PRP [10]. This number corresponds to a platelet count about 4-5 times higher than that contained in the blood, it usually ranges from 150,000 for the 350.000/ μ l (approximately an average value around 200.000/ μ l) [11].

PRP also called as platelet-enriched plasma, platelet-rich concentrate, autologous platelet gel, and platelet releasate [7]. Platelet rich plasma have been used to treat wounds since 1985 [9]. PRP serves as a growth factor agonist [10] and has both mitogenic and chemotactic properties [8,9]. PRP contains a high level of clotting and growth factors [7].

The mechanism of action for PRP is thought to be the molecular and cellular induction of normal wound healing response similar to that seen with platelet activation [12]. PRP accelerate all phases of wound healing (most prominent in angiogenesis) [12].

PRP used in the treatment of chronic skin and soft tissue ulcerations in addition to the use of PRP include dental and oral surgery faciomaxillary surgery orthopedic surgery and trauma], plastic and cosmetic surgery spinal surgery heart bypass surgery and burns surgery

Autologous PRP is a new biotechnology that has stimulation and acceleration of soft-tissue healing.

PRP provides as a surgical hemostatic agent that is biocompatible, safe, and effective. PRP enhances the hemostatic response to injury and accelerates epithelial endothelial and epidermal regeneration, stimulates angiogenesis, enhances collagen synthesis, promotes soft tissue healing, decreases dermal scarring and reverses the inhibition of wound healing caused by glucocorticoids. The high leukocyte concentration of PRP has antimicrobial effect. Since PRP is an autologous blood product, it carries no risk of transmitting infectious disease.

In PRP with chronic non-healing wounds, it has also been used as an anti angiogenic agent and as a carrier for growth factors. In surgical settings, PRP decreases the frequency of intraoperative and postoperative bleeding at donor and recipient sites, accelerates soft-tissue healing, supports the initial stability of grafted tissue at recipient sites as a result of its cohesive and adhesive nature, promotes rapid vascularization of healing tissue by delivering growth factors and, when used in combination with bone replacement materials, induces regeneration.

Growth Factors:-

PRP exerts effects via the degranulation of the alpha granules in platelets that contain growth factors. When the platelets in PRP are activated by thrombin, they release growth factors and serve to accelerate the wound-healing process by promoting cellular proliferation, matrix formation, osteoid production, connective tissue healing, angiogenesis, and collagen synthesis.

The active secretion growth factors begins within minutes of the start of the coagulation sequence and more than 90% are secreted during the first hour, remaining growth factors for the 7 days of their viability. . The rate of wound healing is determined by the number of platelets in the blood clot and PRP increases that initial number.

Methodology:-

This is a prospective randomized controlled study to test the efficacy of autologous platelet rich plasma versus normal saline dressing in chronic non-healing. This study was conducted in our medical college hospital for a period of one year from January 2015 to December 2015 in the department of plastic Surgery 20 cases were studied for chronic non healing ulcers with autologous platelet rich plasma and 20 cases for conventional method who received normal saline as dressing for chronic non healing ulcers. Patients were randomized according to random number table.

20 patients received autologous platelet rich plasma and 20 patients received conventional method (Wet normal saline dressing). The Inclusion criteria includes

Patients in the age group of 18 to 80 years with long standing non-healing ulcers (pressure sores, diabetic ulcers, venous ulcers, other non-specific ulcers),
Non-diabetic patients and patients with type 1 or type 2 diabetes mellitus,
Ulcer ≥ 4 weeks duration and ,
Ulcer $\leq 100\text{cm}^2$ in size,
Hb $\geq 10\text{ g\%}$.

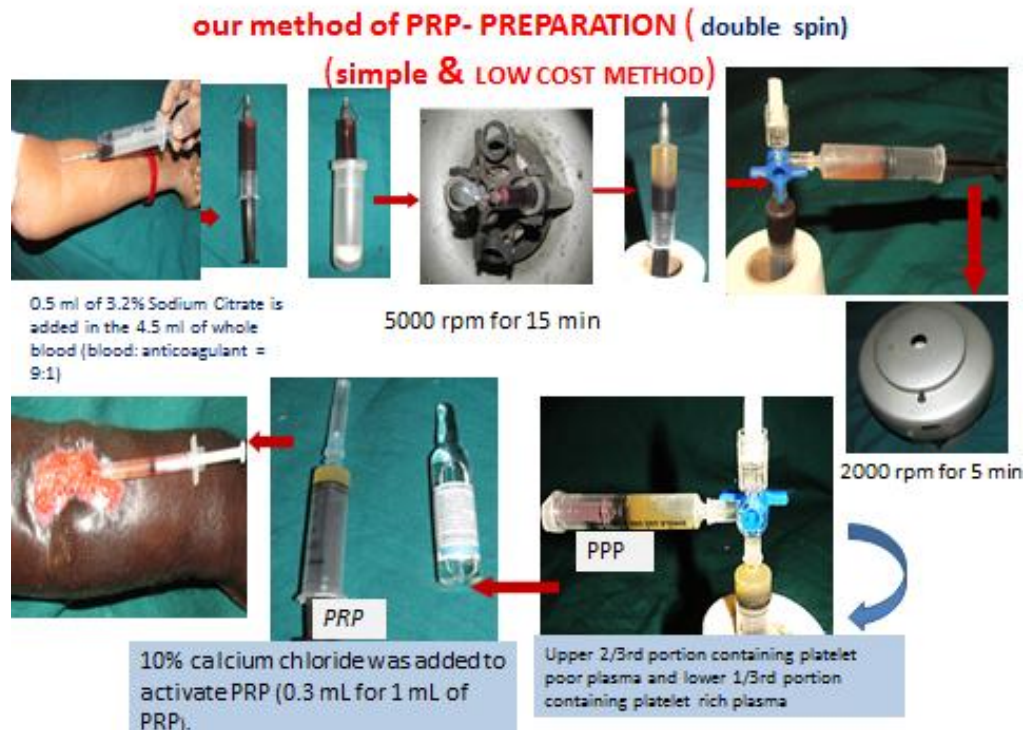


Fig.1 PRP preparation

our method of PRP- PREPARATION
Blood-platelet -3 laks/cumm
PRP- 12 laks/cumm
Concentration factor 4 time

Study Design And Measurements:-

This study is designed as a prospective, randomized-controlled clinical study. Patients would be randomly assigned to two groups, A (experimental group, n = 20) and B (control group, n = 20). Detailed history was taken in all cases regarding the duration, mode of onset, progression, and associated symptoms. Also the etiological factor responsible was elicited in the history.

Ulcer examination was done in all this patients and wound was assessed of its characteristics and photographed. Ulcer was assessed by the investigator at the beginning of the study and at the end of the study; Size of the wound was charted by tracing the edge of wound over the graph on day 1, 7, 14, and 21 day.

Dressing Technique:-

After allotting the dressing with the help of random number table, the following procedure was performed.

For conventional Dressing:-

A chronic non healing ulcer with no active pus discharge and slough was cleaned with the 0.9% saline solution and was covered with pad and roller bandage

For Autologous Platelet rich plasma:-

Autologous platelet rich plasma was prepared from patients own blood and injected periphery and over the ulcer bed using insulin needle(26 G) after cleaning with 0.9% saline and dressing done using pad and roller bandage.

The dressing was changed every 3rd day; similar dressings done to all the patients to both the groups and the outcome is measured on 7th 14th day 21st day.

Wounds will be assessed and measured (length and width, using a metric tape). Outcome is measured in terms of wound reduction between the two groups. Other wound characteristics, i.e., presence or absence of granulation tissue, pain, color, edges, etc. will be documented. The end point of the study will be complete wound closure either surgery in the form of skin graft or spontaneous closure

Statistical analysis:-

Unpaired students "t" test and chi square tests were used to find out the statistical significance. A 'P'<0.05 was taken as significant

Results:-

This Study was conducted in Coimbatore medical college hospital Coimbatore and the findings are tabulated. During the study year from January 2015 to December 2015, 40 patients with chronic non healing ulcers were randomized into study (PRP) and control (normal saline) group. These groups were studied for the effect of conventional dressing versus PRP on epithelialization and wound reduction. A total of 40 patients satisfied the selection criteria, analysis was done by using students paired 't' test for continuous variables within the groups and unpaired 't' test for continuous variables between cases and controls.

The efficacy of PRP was compared with control group as the percentage of ulcer surface area covered by granulation tissue. In PRP group had 73.86% mean granulation tissue their ulcer. The control group had 49.1% mean granulation tissues. The Split Skin Graft uptake was much better in the PRP dressing group $84.789\% \pm 4.391$ than the normal saline dressing group $68.75\% \pm 5.718$ which was statistically significant. The mean hospital stay was also significantly lesser in the study group than the control group.



Fig 2:- Clinical Case-acase of non healing ulcer over leg treated with PRP.

chronic unstable ulcer-2 yrs duration, treated with PRP with skin graft



Fig 3:-

Diabetic foot right. Side- 6 months duration
treated with PRP



Fig 4:-

chronic venous ulcer-2 yrs duration
treated with PRP and skin graft



Fig 5:-

Chronic unstable ulcer 3 yrs duration, failed skin
graft two times, treated with PRP and skin graft



Fig 6:-

Discussion:-

Chronic wounds are a major health problem in developing countries like India, and these wounds lack the necessary GFs for healing, they are often difficult to heal and associated with super added infection. PRP contains various necessary growth factors for wound healing. In addition, the high concentration of WBC's present in PRP is also helpful in preventing infections.[13]

In the present study, an attempt has been made to establish better healing rates with use of PRP in chronic non healing ulcers. In this study the baseline characteristics such as age, sex and location of the ulcer were similar in the patients who received PRP dressing in study group and in patients who received control (NS) dressing.

In the present study the mean platelet count was 2, 00,000 in both PRP group and NS group but the PRP yield 3-4 times higher than baseline value of platelet counts.

In this study, initial area of the ulcer (in mm²) was similar between the two groups. However, the final area of the ulcer (in mm²) was significantly reduced in patients with PRP group as compared to the patients in NS group at the end of the study. The percentage reduction in the area of the ulcer was more in the PRP group as compared to the NS group and this difference was statistically significant.

A study conducted by Frykberg et al.[14] on 49 patients with 65 non healing ulcers showed that 63 of 65 ulcers responded with a reduction in area, volume of the ulcers in a mean duration of 2.8 weeks with 3.2 treatments.

Steen Voorde et al.[15] conducted a study on 12 patients with 13 wounds, showing that 7 of 13 wounds required more than 1 application, with a mean number of 2.2 applications and a mean treatment period of 4.2 weeks.

Kakudo et al.[16] treated five cases of intractable skin ulcer with autologous PRP, among which three ulcers healed completely within 4 weeks and epithelization of wound occurred within 6.6 weeks on average.

In our study, 20 patients were treated with PRP and the mean reduction of the ulcers was 30.56%(SD5.6) in three weeks. We also noticed increase granulation tissue of the wound around 73% in three weeks and reduced pain and infection

Till now there is no standard method of preparation of PRP in literature. According to Marx, the device must use a double centrifugation technique.[17] Regardless of the rate of centrifugation or the time of centrifugation, a single spin cannot adequately concentrate platelets, because the red blood cells will interfere with their fine separation. Currently, PRP was found to be useful in treating chronic leg ulcers.

Limitations of our study:-

1. Follow up is short to derive conclusion on long term healing of ulcers.
2. Need further controlled, randomized prospective clinical trials are necessary to definitively demonstrate its efficacy.
3. There is also a need for the development of a standard protocol for the preparation of PRP, as literature currently there is no standardization of the procedure.

Conclusion:-

We believe that the application of of AUTOLOGUS PLATELET RICH PLASMA (PRP) dressings in comparison with the control group (normal saline group) for the treatment of chronic ulcers of the lower limb, the following conclusions were derived

1. PRP showed faster and better healing rates among the study group.
2. Ulcer area reduction and percentage reduction of ulcer was better in PRP group.
3. Formation of granulation over the ulcer better with PRP group
4. Uptake of skin graft better in PRP group than NS group
5. Skin graft stability and compliance better in PRP group.
6. There were no adverse effects or reactions seen when autologous PRP was applied over the ulcer.

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RESEARCH ARTICLE

COMPLICATIONS AFTER POSTERIOR ACETABULAR WALL INTERNAL FIXATION – AVASCULAR NECROSIS AND NONUNION.

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Acetabular fractures, Avascular Necrosis, Retrospective study, Ficat classification

Abstract

Introduction: Serious orthopaedics injuries caused by high impact trauma due to road traffic accidents are more prevalent in Saudi Arabia. Fractures of posterior wall are the most common type of acetabulum fractures. The outcome after surgical management of acetabular fracture is primarily related to the quality of articular reduction. This management is the most challenging task for surgeons.

Methodology: A retrospective cohort study was conducted in a tertiary care hospital, Riyadh, Saudi Arabia. All patients who underwent internal fixation for posterior wall acetabular fracture from October 2013 to September 2015 were included in the study. Post-surgical outpatient evaluation was done clinically by Merle D' Aubigne Y Postel - hip functional evaluation score and radiological assessment using Ficat classification for staging of avascular necrosis of femoral head.

Principal results: With increase in age, the function of hip declined. The step in fixation was found significantly associated with avascular necrosis ($p = 0.020$) and nonunion ($p = 0.020$). The gap was found significantly associated with nonunion only ($p = 0.001$).

Conclusions: Delay in surgery should be avoided if surgical management is decided for posterior acetabular wall fracture. Adequate surgical reduction of fracture of posterior acetabular wall is necessary. Screw seems to be a better choice of implant until further new evidence is received. A randomized controlled trial is needed to evaluate and compare different management options for fracture of posterior acetabular wall and to set standard guidelines for future.

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Introduction:-

The incidence of Acetabular fractures is increasing in developing countries because of rise in road traffic accidents (Robert S. Sterling, 2011). Acetabulum fracture counts about 0.5 to 8% of orthopaedic ward admissions (Grubor P, 2015) and out of this about 24 to 32 % have posterior wall involvement (Vincenzo Giordano et al., 2009). Currently about 1.5 million hip fractures occur every year (Robert S. Sterling, 2011). By the year 2050 the total number of hip fractures is expected to surpass 6 million worldwide (Robert S. Sterling, 2011).

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Fracture of acetabulum is not very common being related to high impact trauma always. The most common cause of such trauma is a road traffic accident, dashboard injury or a fall from height. Fall is the second commonest cause of acetabulum injury occurring in about 10% (Grubor P, 2015).

Sciatic nerve is the thickest and longest nerve of the body which contains both sensory and motor fibres. The location of sciatic nerve is prone to injury when an acetabulum fracture occurs, especially with posterior dislocation of hip. The common involvement is of common peroneal part (Ebraheim NA et al., 2007). The head of femur receives its blood supply from an arterial anastomosis. In this anastomosis, compromise of medial femoral circumflex artery, a branch of profundafemoris artery is most important cause of avascular necrosis of the head of femur due to fracture acetabulum and especially in dislocation of hip. Although a small portion of femoral head has alternate blood supply from a branch of obturator artery that runs in the ligamentum teres (Birhane A et al., 2015).

Rationale:-

In Saudi Arabia, cars are the main mode of transportation with approximately more than 6 million cars on road (Farah A. Mansuri et al., 2015). 20% beds in Ministry of Health hospitals in Saudi Arabia are engaged by Road traffic accident victims (Farah A. Mansuri et al., 2015). In the past 2 decades, 86,000 casualties and 611,000 injuries have been recorded in Saudi Arabia in road accidents (Farah A. Mansuri et al., 2015). Out of these, 7% had ended in disabilities (Farah A. Mansuri et al., 2015). The economic effects of road traffic accidents in Saudi Arabia projected in terms of potential productive years life lost (PPYLL) were examined in a study that has reported a 31.6% increase in deaths due to RTA among males in 1997-2002 compared with a 1.3% increase in deaths due to RTA among females. In Saudi Arabia, road traffic accidents are the major cause of hourly consultation to the health care facilities (Farah A. Mansuri et al., 2015).

When compared to other arab countries, Saudi Arabia has the second highest casualty rate due to road traffic accidents. Globally it is the 23rd country to have high number of casualties due to road accidents. World Health Organization has reported that in Saudi Arabia, a car accident occurs every second; while on average 17 people are killed in crashes daily.

Aim:-

The aim of this study was to assess the outcome of acetabular fracture in Saudi population after the surgical fixation procedure. The main objective was to note the complications of acetabular fixation with special focus on avascular necrosis and nonunion.

Materials and Methods:-

The study design was a retrospective cohort study. All male patients, Saudi nationals, who underwent surgical fixation for fracture of posterior acetabulum wall due to any cause at King Saud Medical City, Riyadh, Saudi Arabia were included. The period of surgical intervention was from 1st October 2013 to 30th September 2015. This study was approved by the Institutional Review Board and informed consent was taken from all study participants.

The data collected from medical records included patient's age, nationality, sex, cause and duration of fracture prior to admission, date of admission and surgery, any delay in surgery, percentage joint involvement, side of fracture, presence of dislocation, type of implant used, any associated fractures and co morbidities. Radiological investigations including x-rays and CT scan of pelvis were reviewed in hospital-based PACs system. Patients are routinely followed six weeks after surgery in the outpatient department. At follow up in the orthopaedic research clinic, clinical assessment was done using Merle D-Aubigne Y Postel (hip functional) score (David J. Biau et al., 2009). Sciatic nerve function was assessed clinically. Conventional x-rays of pelvis and hip joint were reviewed for any presence of step and gap at the site of surgical fixation. CT scan of pelvis was requested if there was suspicion of nonunion or avascular necrosis on plain x-rays and Ficat classification (Jawad et al., 2012) was used to stage the avascular necrosis of femoral head.

Totally 36 patients underwent surgery for posterior acetabular wall fracture during the study period. Approximate duration of outpatient follow-up for all patients was between nine months to two and half years. 1 non-Saudi national patient and 2 females were excluded from the study according to pre-decided exclusion criteria. 2 patients were noted to have avascular necrosis & total hip replacement before the start of study and were therefore excluded. 6 patients had lost their follow up earlier and were not accessible. 3 patients had provided incorrect contact information and 10 patients refused to participate in the study. Finally evaluation was done for 12 patients.

Statistical Analysis:-

The software used for data analysis was SPSS 21 and the results are presented as descriptive Statistics: frequency, percentage, range, means, median, 95% confidence interval and the inferential statistics: student-t-test for independent samples. Pearson's coefficient of Correlation, one way anova and χ^2 were used for testing the statistical significance at 5% level with 5% type I error and 20% power of the study. (Betty R.K and Jonathan A.C.S, 2009)

Results:-

The baseline characteristics of the patients are given in **Table 1**. Age of patients ranged from 18 to 65 years with mean \pm SE (mean) of 37.0 ± 4.4 years. The fracture for eleven patients (91.7 %) was due to road traffic accident and one (8.3%) had a history of fall. The fracture of posterior wall of acetabulum was divided in three grades according to the percentage of damage. One patient (8.3%) had < 25percent, 5 patients (41.7%) had 25-50 percent and 6 patients (50%) had >50 percent joint involvement. Six (50%) patients had an additional fracture (Upper limb(1), Lower limb(1), Both limbs(2), Pelvis(1), Ribs(1)). 4 patients (33.3%) had co-morbid illness (Lymphoma(1), Wegner's disease (1), Diabetes Mellitus & Hypertension (1), Diabetes, Hypertension and Ischemic heart disease (1)).

The Merle D - Aubigne Y Postel score was ranging from 6 to 12 with a mean \pm SE of 10.17 ± 0.57 and Pearson's Coefficient of Correlation of this score with Age in years showed a statistically significant negative high magnitude with $r = -0.849$ ($P=0.000$) as observed in **Figure 1** with 95% confidence interval, but delay in surgery (days) did not show much correlation with the hip function score.

Based on Ficat classification, one patient (8.3%) was classified in category IIB, one patient (8.3%) in category IV and ten patients (83.3%) in category zero. Two patients (16.7%) had nonunion and two patients (16.7%) had developed avascular necrosis.

For the patient who had step, Sciatic Nerve was not involved after surgery and the radiological Ficat classification was observed to be IV with $\chi^2=12.0$ ($P=0.002$), experienced avascular necrosis, $\chi^2=5.45$ ($P=0.02$) and had nonunion $\chi^2=5.45$ ($P=0.02$).

Among 10 patients who had no gap, one patient (10.0%) had sciatic nerve involvement after surgery. One out of ten patients (10.0%) who had no gap, the Ficat classification was observed to be IIB, 1 patient (10.0%) had avascular necrosis. One patient (100.0%) who had step and 1 out of 11 patients (9.1%) without step had avascular necrosis. 2 out of 10 patients with gap had non-union. One patient (8.3%) had poor hip function score (<7), 2 patients (16.7%) had fair score (8), 1 patient (8.3%) with medium (9) and good score (10) in each group, and 7 patients (58.3%) had very good score (11-12).

Merle score was compared to all variables shown in the **Table 2**. It was seen significantly related to AVN ($p = 0.017$) and percentage of fractured wall ($p = 0.05$) only. Among the 11 patients who had road traffic accident as the cause of fracture, one patient (9.1%) had a poor score, 2 patients (18.2%) had fair score, 1 patient (9.1%) had good score and 7 patients (63.6%) had very good score and a significant $\chi^2 = 12.0$ ($P=0.017$).

Among 8 patients with dislocation, 1 patient (12.5%) had POOR Merle score, 2 patients (25.0%) had FAIR score and 5 patients (62.5%) had VERY GOOD score. Of 2 patients whose sciatic nerve was found injured, 1 patient (50.0%) had a POOR score and 1 patient (50.0%) had VERY GOOD score. There were 8 patients without systemic illness and their average Merle score was 9.87 ± 0.81 . One patient was diabetic & hypertensive with a score of 11 ± 0.0 . One patient with diabetes, hypertension and ischemic heart disease had a score of 9.0 ± 0.0 . One patient with lymphoma had a score of 12 ± 0.0 and one patient with Wegner's disease had a score of 11.0 ± 0.0 . There was an insignificant difference in the mean Merle scores $F=0.322$ (0.855).

Two patients had avascular necrosis and both had POOR Merle score. Of two patients with nonunion, 1 patient (50.0%) had a FAIR score and the other patient had GOOD score. Seven patients were implanted with plate. Of them 1 patient (14.3%) had a FAIR score, 1 patient (14.3%) had MEDIUM score, 1 patient (14.3%) had GOOD score and 4 patients (57.1%) had VERY GOOD score. Out of 3 patients who were implanted with both plate and screws, 1 patient (33.3%) had POOR score, 1 patient (33.3%) had FAIR score and 1 patient (33.3%) had VERY GOOD score. 2 patients who were implanted with screws, both (100.0) had VERY GOOD score with $\chi^2 = 6.204$ ($P=0.624$) and is given in **Figure 2**. There was no significant difference in the category of delay in surgery.

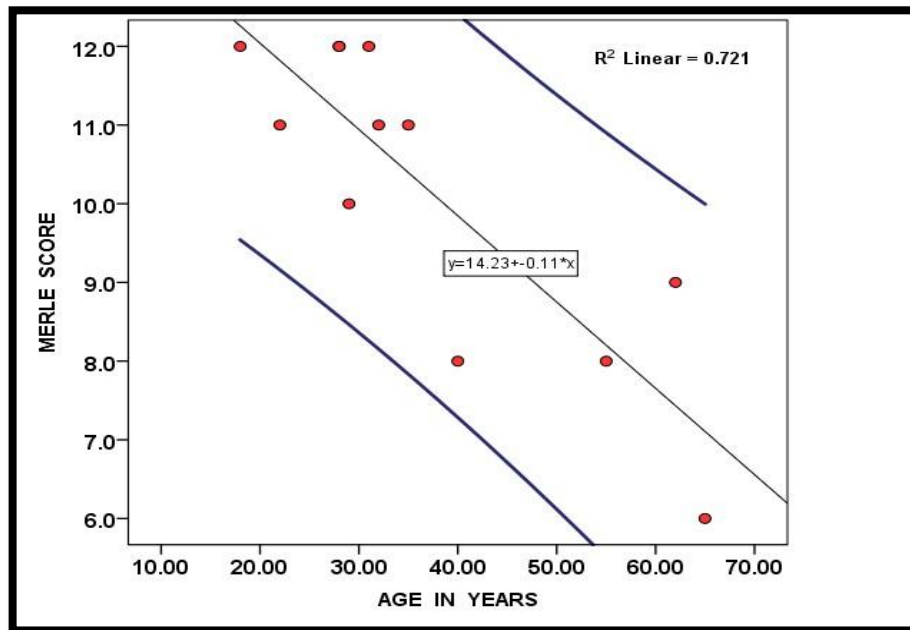
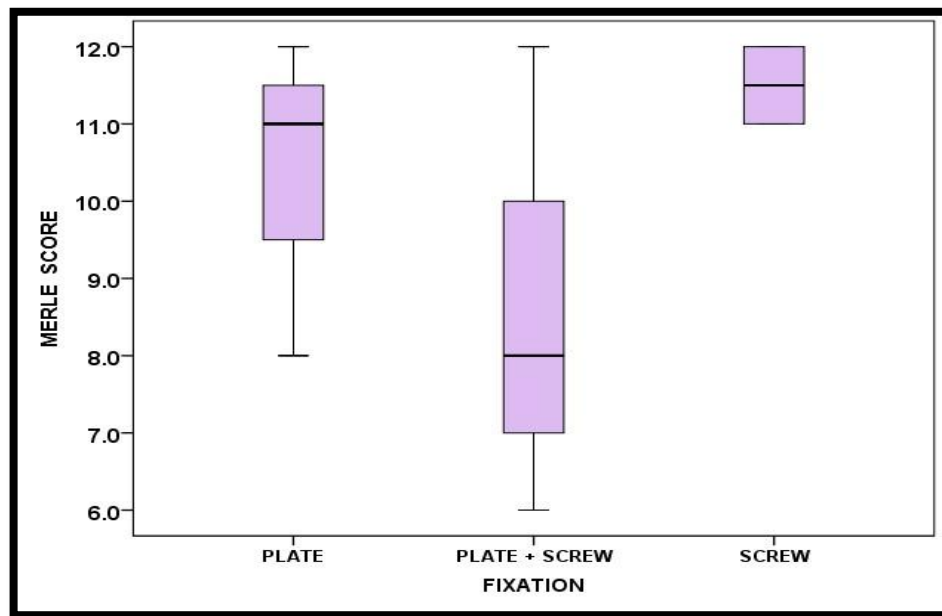
Table 1:- Descriptive Statistics of study participants.

Baseline Characteristics	n=12	(%)	Baseline Characteristics	n=12	(%)
Age in Years - 18-25	2	16.7	Fractured Wall - <25%	1	8.3
26-45	7	58.3	25-50%	5	41.7
46-65	3	25.0	>50%	6	50.0
Cause of Fracture - RTA	11	91.7	Sciatic Nerve Injury - Pre Op	2	16.7
Fall	1	8.3	Post Op	1	8.3
Fracture Side - Right	8	66.7	Normal	9	75.0
Left	4	33.3	Delay in Surgery - <7 Days	2	16.7
Dislocation - Yes	8	66.7	>7 Days	10	83.3
No	4	33.3	Fixation - Plate	7	58.3
Poly trauma - Yes	6	50.0	Screws	2	16.7
No	6	50.0	Plates + Screws	3	25.0
Co-morbid - Yes	4	33.3	Gap - Yes	2	16.7
No	8	66.7	No	10	83.3
			Step - Yes	1	8.3
			No	11	91.7

Table 2:- Inferential Statistics of Merle Score for patients under study.

Characteristics	Mean \pm SE	Test (P-value)	Characteristics	Mean \pm SE	Test (P-value)
Cause - RTA	10.2 \pm 0.61	t = 0.59	Sciatic N. Damage		
Fall	9.0 \pm 0.00	(0.56)	Pre Op - Yes	9.0 \pm 3.00	t = 0.89
Affected Side			No	10.4 \pm 0.49	(0.39)
Right	10.6 \pm 0.53	t = 1.14	Post Op - Yes	11.0 \pm 0.00	t = 0.42
Left	9.2 \pm 1.37	(0.28)	No	10.0 \pm 0.62	(0.68)
Dislocation			Fixation - Plate	10.4 \pm 0.57	
Yes	10.0 \pm 0.82	t = 0.39	Screws	11.5 \pm 0.50	F = 1.47
No	10.5 \pm 0.64	(0.70)	Plate + Screws	8.6 \pm 1.76	(0.27)
PolyTrauma			Step - Yes	8.0 \pm 0.0	t = 1.15
Yes	9.5 \pm 0.95	t = 1.18	No	10.3 \pm 0.59	(0.27)
No	10.8 \pm 0.60	(0.26)	Gap - Yes	9.0 \pm 1.00	t = 0.89
Wall Fracture (%)			No	10.4 \pm 0.65	(0.39)
<25	6.0 \pm 0.00				
25 - 50	11.0 \pm 0.77	F = 4.10			
>50	10.1 \pm 0.60	(0.05)*			

*Statistically significant at 5% level

Figure 1:- Scatter Diagram for Merle Score.**Figure 2:-** Whiskers Box Plot for Merle Score.**Main findings of this study were:-**

1. Step in fixation was significantly associated with avascular necrosis ($p = 0.020$) and nonunion ($p = 0.020$).
2. The gap was significantly associated with nonunion only ($p = 0.001$).

Discussion:-

Being rare, fracture of posterior acetabular wall has not been studied extensively in past. There are no randomized controlled trials available comparing the management options. In literature we mostly have case reports or case series which have not comprehensively reviewed the complications of different management modalities. In an older

study the percentage of avascular necrosis was reported to range from 18 to 32% (Michael R. Baumgaertner, 1999). Another recent study showed 11.53% incidence of avascular necrosis (Magu NK et al., 2014) while in our study it was 17%. We, therefore, present new and recent data related to the complications of posterior acetabular wall fracture and their relation to surgical intervention.

Avascular necrosis and nonunion of posterior acetabular wall fracture are related to poor surgical reduction. As noted in our study that absence of step and gap after surgical fixation is related to higher hip function scores. Similarly post-fixation step and gap will provide poor hip function and possibility of nonunion. There is no recommended protocol for radiological evaluation other than conventional x-rays after surgical fixation but some surgeons recommend an immediate postoperative CT scan of hip before discharge, to observe the quality of reduction (P.V. Giannoudis and V.S. Nikolaou, 2008). It is already recommended to consider total hip replacement as primary procedure in patients who have comminute fracture of posterior acetabular wall (James L. Guyton and Edward A Perez, 2012). Until we have more data comparing post OP radiological assessment to no assessment; we suggest performing a confirmatory CT scan of hip soon after procedure. It will aid in earlier detection of potential complications.

The choice of implant used in surgery is based on fracture anatomy and surgeon's choice. In one review some benefits of different implants over each other i.e. plate, screws or both together have been postulated (Zhang Y et al., 2013). There are no RCT's comparing the two options and no recommendation of using one method over other. Nevertheless at times surgeon has to use screws to hold the broken bone pieces in addition to plate fixation, to achieve complete reduction. Also at times two plates should be used to achieve reduction (Ebraheim NA et al., 2007). In our study, the clinical outcome was compared to the type of implant used at the time of surgical fixation. As we can see that the screw is a better option when compared to plate or plate plus screw (Rosario Spagnolo et al., 2009). However the availability of implant and the learning curve of surgeon also play a role in choosing the type of implant.

The presence of dislocation has an impact on sciatic nerve function and avascular necrosis (Iselin LD et al., 2013) observed an average delay of seven days from trauma to surgery. Most of our patients were operated later than 7 days. We recommend early intervention for patients who are candidates for surgery as it will help in reduction of late complications.

Most of our patients were young drivers below age of 40 years. The accident rate cannot be controlled except efforts to be done at national level but appropriate and timely surgical intervention for such patients soon after fracture can prevent future complications. If they have to undergo hip arthroplasty after few years, it is associated with related complications. Careful selection of choice of intervention and proper complete follow up will markedly reduce permanent disability in our patients.

There is essential need of a randomized control trial to compare and evaluate different management options for fracture of posterior acetabular wall, types of implants used, best time for surgery and post-operative clinical and radiological assessment. The trial should follow the patients for longer duration to compare the long term complications among different groups - mainly avascular necrosis and nonunion.

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RESEARCH ARTICLE

COMPARISON OF MITOTIC EFFECT OF AQUEOUS, AICOHOL AND PETROLEUM ETHER EXTRACT OF THE LEAVES OF *BOERHAAVIA DIFFUSA* L.ON ROOT MERISTEM OF *ALLIUM CEPA* L.

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Key words:-

Boerhaavia diffusa L, Abnormal cells, Mitosis

Abstract

In the present study the effects of various extracts of *Boerhaavia diffusa* L. on mitotic cells of *Allium cepa* L. is investigated. The concentrations are set as 0.05, 0.1, 0.15 and 0.2 using water, petroleum ether and ethyl alcohol extract of stem and leaves. The duration of the treatment is 30 minutes prior to mitotic peak. The mitotic index and index of various stages of mitosis are scored along with the number of abnormal cells. The mitotic index is found to be reduced after higher concentration of treatments while at lower concentrations there is an increase in the number of dividing cells. The percentage of abnormal cells is found to be increased with increase in the concentration of the treatment. The abnormalities showed a predominance of stickiness than fragments, bridges, unoriented and tropokinesis. The results indicated that the active toxic element is an ethyl alcohol soluble fraction as evidenced by higher frequency of abnormal cells. Thus it is recommended that excessive use of ayurvedic preparations like purnavasasa, a preparation made entirely from *Boerhaavia diffusa* L. may harm the epithelial lining of stomach and intestine.

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Introduction:-

Boerhaavia diffusa L is a perennial creeping weed, prostrate or ascending herb, up to 1 m long or more, having spreading branches. The roots are stout and fusiform with a woody root stock. The stem is prostrate, woody or succulent, cylindrical, often purplish, hairy, arranged in unequal pairs, green and glabrous above and usually white underneath. The shape of the leaves varies considerably-ovate-oblong, round, or sub cordate at the base and smooth above. Margins of the leaves are smooth, wavy or undulate. The upper surface of the leaves is green, smooth, and glabrous, where as it is pinkish white and hairy beneath. Leaves are up to 5.5×3.5 cm² in area.

Flowers are minute, sub capitates, present 4-10 together in small bacteriolate umbels, forming axillary and terminal panicles. These are hermaphrodite, and white, pink, or pinkish red in color. Bracts are deciduous and involucre. A perianth is present in the place of a calyx and corolla, which is tubular in shape, the tube being short and narrow at the base and funnel. Shaped at the top and constricted above the ovary. There are five lobes, which are small and acute. Two or three stamens are present and are slightly exerted. The stigma is peltate. The achene fruit is detachable, ovate oblong, pubescent, five ribbed and glandular, anthocarpous, and viscid on the ribs (Thakur *et al.*, 1989). The seed germinate before the onset of the monsoon.

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The plants grow profusely in the rainy season, and mature seeds are formed in October- November. Due to its sticky nature, the plant gets stuck on clothes of human beings and on the legs of animals, which helps in its dispersal from one place to another. It has a large root system bearing rootlets. The tap root is tuberous, cylindrical to narrowly fusiform to conical or tapering, light yellow, brown or brownish gray. Some workers have studied the regeneration of this plant through tissue culture. Bhansali *et al.*, (1978) reported induction of adventitious shoots using stem explants of *Boerhaavia diffusa* L. Roots were also regenerated from leaf segments of *Boerhaavia diffusa* L when cultured in vitro. These roots contained 0.15% alkaloid punarnavine. Increase in leaves of indoleacetic acid (IAA) in MS medium reduced the number of roots regenerate from the leaf segment, their length and alkaloid content (Shrivastava and Padhya, 1995).

In purulia (West Bengal), tribal eat this plant as vegetable. *Boerhaavia diffusa* L leaves are cooked and eaten in Assam, where it is commonly found in the markets. Its roots are used in the treatment of piles by the inhabitants of the Garhwal Himalaya (Uttaranchal). The root paste is used to cure bloody dysentery by the Bhils of the Jhabua district in Madhya Pradesh. The decoction of plant is given in the treatment of nodules in the body. The root juice is treating asthma, scanty urine, and internal inflammation disorders. *Boerhaavia diffusa* L is used for curing ailments such as leucorrhea, rheumatism, and stomach ache by the sahariya tribe in the Lalitpur district of Uttar Pradesh. This plant is also used by the tribes of Ambikapur district (Madhya Pradesh) for the treatment of elephantiasis. In the Indo-Nepal Himalayan terrain region, the tribal harvest this plant for medicinal purposes, mainly for flushing out the renal system, and to treatment seminal weakness and blood pressure (Mitra and Gupta, 1997).

The *Boerhaavia diffusa* L plant contains a large number of compounds as flavonoids, alkaloids, steroids, triterpenoids, lipids, lignins, carbohydrates, proteins and glycoproteins. Punarnavine $C_{17}H_{22}N_2O$, (Surange and Pendse, 1972), boeravinone A (Kadota *et al.*, 1989; Lami *et al.*, 1992), hypoxanthine 9-L-arabinofuranoside (Ahmad and Hossain, 1968), ursolic acid (Mishra and Tiwar, 1971), punarnavoside (Jain and Khanna, 1989), liiodendrin (Aftab *et al.*, 1996) and aglycoprotein having a molecular weight of 16-20 KDa have been isolated and studied in detail for their biological activity. Chopra *et al.*, (1923) reported that the plant contained large quantities of potassium nitrate, besides punarnavine. The herb and roots are rich in proteins and fats. The herb contains 15 amino acids, including 6 essential amino acids, while the root contains 14 amino acids. Seth *et al.*, (1986) isolated a new antifibrinolytic compound 'punarnavoside' from the roots of *Boerhaavia diffusa* L. Phytochemical screening of the roots from garden – grown in vivo plants of *Boerhaavia diffusa* L of different ages revealed that the maximum alkaloid content (2%) accumulated in the roots of 3 year old mature plants.

Biological Activity:-

As medicine in the traditional system: Different parts of the *Boerhaavia diffusa* L plants have been widely used by indigenous tribes in the traditional system of medicine. The roots have been widely used for treatment of dyspepsia, jaundice, and enlargement of spleen, abdominal tumors and cancers (Kirtikar and Basu, 1956). The root powder, when mixed with mamira (*Thalictrum foliolosm* DC), is used to eye diseases. It cures corneal ulcers and night blindness (Gupta *et al.*, 1962), and helps restore virility in men. People in tribal areas use it to hasten childbirth (Shah *et al.*, 1983). The juice of *Boerhaavia diffusa* L leaves serves as a lotion in ophthalmia. It is also administered orally as a blood purifier and to relieve muscular pain.

As medicine in the ayurvedic system: In old Indian books of medicine such as the Charaka Samhitha and Sushrita Samhitha, it mentioned that the Ayurvedic preparations made from punarnava – namely, punarnavastaka kvath, punarnava kshar, and punarnava taila – were used for the treatment of various ailments. The whole plant of *Boerhaavia diffusa* L is very useful source of the drug punarnava, which is documented in Indian pharmacopoeia as a diuretic (Chopra, 1969). The active principle contained in the herb is an alkaloid, known as punarnavane. The roots and leaves with flowers have been found to be highly potent (CSIR, 1988). In Ayurvedic medicine, different parts of this plant were reported to have various medicinal properties. It was used in renal ailments as diuretic (Anand, 1995); and to treat seminal weakness and blood pressure (Gaitonde *et al.*, 1974). It is also used in the treatment of stomach ache, anemia, cough, and cold, and as a diaphoretic, laxative, expectorant, and a potent antidote for snake and rat bites (Chopra *et al.*, 1956), in the treatment of nephritic syndrome (Singh and Udupa, 1972), hepatitis, gall bladder abnormalities, and urinary disorders (Cruz, 1995). The flowers and seeds are used as contraceptive (Chopra *et al.*, 1956).

Pharmacological and clinical properties: Pharmacological studies have demonstrated that punarnava possesses punarnavoside, which exhibits a wide range of properties- diuretic (Gaitonde *et al.*, 1974); anti-inflammatory (Bhalla *et al.*, 1968); antifibrinolytic (Jain and Khanna, 1989); anticonvulsant (Adesina, 1979); antibacterial (Olukoya *et al.*, 1993); antistress agent; antihepatotoxic, anthelmintic, febrifuge, antiscabies, and anti-asthmatic, antileprosy, and anti-urethritis and anti-nematodal activity. An aqueous extract of thinner roots of (*Boerhaavia diffusa* L) at a dose of 2 ml kg⁻¹ exhibited marked protection of various enzymes such as serum glutamic oxaloacetic transaminase, serum glutamic-pyruvic transaminase, and bilirubin in serum against hepatic injury in rats. Punarnava possesses diuretic and anti-inflammatory activities and the maximum activity was observed in samples collected in the rainy season. Due to the combination of these two activities, punarnava is regarded therapeutically as highly efficacious for the treatment of inflammatory renal diseases and common clinical problems such as nephritic syndrome, oedema, and ascites resulting from early cirrhosis of the liver and chronic peritonitis.

The plant is reported to be efficacious in abdominal tumors and cancers. The drug proved useful as a hematinic and as a growth in children fed with milk fortified with the drug. In the form of a powder or an aqueous decoction, the drug was found to be useful in the treatment of nephritic syndrome and compared well with corticosteroids. The drug decreased to albuminuria, the serum protein was increased and serum cholesterol level was lowered.

Singh and Udupa (1972) reported that dried root powder showed curative efficiency when administered orally for one month to children or adults suffering from helminths infection. The subjects became worm-free within five days of treatment. The drug singly or in combination with other drugs, was found to be effective in liver disorders, heart diseases (hypertension, angina, cardiac failure, etc), respiratory tract infections, leucorrhea, spermatorrhea, etc. The purified glycoprotein from *Boerhaavia diffusa* L exhibited strong antimicrobial activity against RNA (ribonucleic acid) bacteriophages with much of clinical research validating its long history of different uses in natural medicine; the commercial bulk of punarnava in India represents heterogeneous medicinal uses.

As food: The entire plant including the roots is eaten as vegetable, in curries and soups. The roots and seeds are added to cereals, pancakes, and other food stuffs. They are also served as bird feed or poultry feed. The plants are grazed by sheep, goats, and cows, and in West Bengal, it is believed that the plant enhances lactation period and also the amount of milk in cattle.

Significance of the Study:-

Boerhaavia diffusa L have been used in ayurvedic preparation for millennia. The scientific studies as well as the exploration of active principles are developed in recent years. In recent year of research have to reveal that many of such potent and effective during sources contain mutagenic or cytotoxic principles. This raises concern about the genotoxic hazards resulting from long term use of such plants. This is because of the fact that plant extract contains mainly alkaloids, glycoside, flavonoides, tannins etc. Because of the low cost of the ayurvedic preparations as well as its local availability, people can afford the long term use of such preparations. In view of these, the cytotoxic effect of the various fraction of *Boerhaavia diffusa* L is essential, and is attempted in this study.

Objectives of the Study:-

The present study is focused on to find out the effect of various fractions of extract of *Boerhaavia diffusa* L on mitotic index and variations in different phases of cell division in root meristem cells of *Allium cepa* L. This study also find out the nature and intensity of aberrations of dividing cells caused by various fractions of the extract of *Boerhaavia diffusa* L and the LC50 of the extract of *Boerhaavia diffusa* L with respect to the cell damage in *Allium cepa* L meristem.

Materials and Methods:-

The material:-

Boerhaavia diffusa L grown in wild condition were collected for the study. The plants were identified up rooted and bring to the laboratory.

Preparation of the plant material:-

The leaves of mature flowered twigs are separated and shade dried and oven dried at 60°C up to attaining a constant weight. The dried material was powdered and sieved.

Soxhlet extraction:-

The powdered material was extracted using petroleum ether, ethyl alcohol and water in a Soxhlet for 8 hours. The fractions were collected and dried.

The onion culture:-

Commercially available bulbs of *Allium cepa* L were used for the study well cleaned bulbs were sown in well ministered sterile sand, in a tray of size 30x26cm. The germinated bulbs with 0.5-1cm roots were selected and treated with different concentrations of extracts (0.05%,0.1%,0.15%,0.2%) of *Boerhaavia diffusa* L 5 bulbs were treated for each concentration.

Collection of root tip and storage:-

The root tip after treatment are cut out from the bulbs and washed well in distilled water. Transfer the root tip in to Carnoy's fixative (3 alcohol: 1 acetic acid) for 24 hours; and then stored at 4⁰C.

Squashing and staining:-

Stored root tips are hydrolyzed in 1N HCl for 3 minutes and washed well in water. Squash preparation of these root tips were made with 2% acetocarmine stain, and observed under microscope. Five slides are scored for evaluation. Three randomly selected fields are scored from each slide. Control is maintained for each treatment in distilled water.

Observations taken:-

The total number in cell in each field along with the number of dividing cells was scored. The total number cells pertaining to each phases of division was also noted.

The mitotic index were calculated rising the formula,

$$\text{Mitotic index} = \frac{\text{Number of dividing cells}}{\text{Total number of cells}} \times 100$$

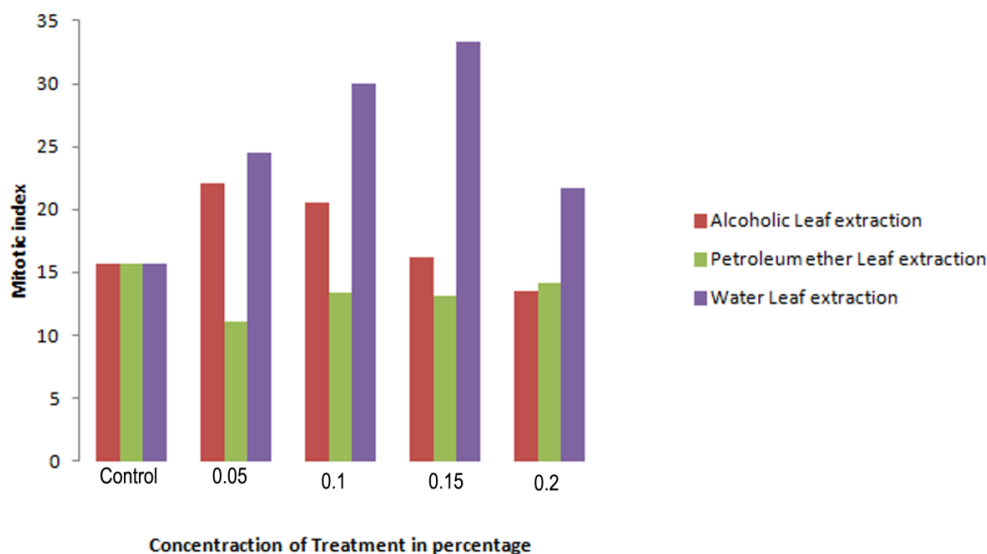
Percentage distribution of normal and abnormal cells, percentage of various abnormalities met with are presented in Tables and compared with control.

Results and Discussion:-

A careful analysis of the data revealed that the treatment with aqueous extracts of leaves of *Boerhaavia diffusa* L on mitotic cells of the *Allium cepa* L showed an increase in mitotic index when compared to control. The increase was not depended on the concentration of the extract treated. The control posses of an index of 15.70% while in treated cells possess 24.51, 30, 33.3 and 21.67 percent of dividing cells after treatments with 0.05, 0.1, 0.15 and 0.2 percent of extract respectively. Similar concentrations of alcoholic extracts yielded 22.05, 20.56, 16.14 and 13.47percent of mitotic cells. The petroleum ether extracts of leaves of *Boerhaavia diffusa* L mitotic cells of the *Allium cepa* L yielded 11.13, 13.37, 13.12 and 14.09 percent mitotic index after treatment with 0.05, 0.1, 0.15 and 0.2 percent of extract respectively.

Table 1:- Mitotic index of root meristem of *Allium cepa* L after treatment with extract of leaves of *Boerhaavia diffusa* L.

Concentration of Treatment in percentage	Mitotic Index		
	Alcoholic Leaf extraction	Petroleum ether Leaf extraction	Water Leaf extraction
Control	15.70	15.70	15.70
0.05	22.05	11.13	24.51
0.1	20.56	13.37	30.00
0.15	16.14	13.12	33.30
0.2	13.47	14.09	21.67



Graph 1:- Mitotic index of root meristem of *Allium cepa* L after treatment with extract of leaves of *Boerhaavia diffusa* L.

The distribution of various stages of mitosis is also considered after treatment with various concentration of extract from leaves of *Boerhaavia diffusa* L. The distribution of dividing cells in control population is 80percent prophase, 4.87 percent metaphase, 7.80 percent anaphase and 7.30 percent of cells at telophase. After treatment with various concentration of ethyl alcohol extracts of leaves of *Boerhaavia diffusa* L this distribution is disturbed and resulting in 26.3 prophase, 47.36 metaphase, 10.52 anaphase and 15.79 telophase percentages treated with 0.2 percent extract. After treatment with 0.15 percent extract the distribution is found to be 54.83 prophase, 22.58 metaphase, 10.90 anaphase, and 9.60 telophase percents. After treatment with 0.1 percent extract the distribution is found to be 51.72 prophase, 31.03 metaphase, 17.26 anaphase and no percnages telophase at all. However treatment with 0.05 percent of extract of leaves of *Boerhaavia diffusa* L showed 82.22 percent prophase, 8.80 percent metaphase, 6.66 percent anaphase and 2.20 percent telophase.

With respect to the presence of abnormal cells the treatment with alcohol extracts of leaves of *Boerhaavia diffusa* L on *Allium cepa* L induces 63.15% after treatment with 0.15 extract, which forms the highest percentage of abnormal cells observed in the present study. The increase in the number of abnormal cells was not depended on the concentration of the extract treated. The percentage of abnormal cells observed after various concentrations of treatment is presented in Table. 3.

The percentages of various abnormalities produced by extract in different solvents as well as from leaves of *Boerhaavia diffusa* L are given Table.4. The most observed abnormalities observed were stickiness, fragment, unoriented, bridge and tropokinesis. Metaphase stickiness was found to be the most common anomaly (42.10 in alcoholic extract of leaves) this was followed by unoriented chromosomes in metaphase (22.58% in alcoholic extract of leaves), prophase stickiness (3.70 percent in petroleum ether extract of leaves), anaphase tropokinensis (10.34 percent in petroleum ether extract of leaves), anaphase bridge (9.67 percent in alcoholic extract of leaves), telophase stickiness (6.896 percent in petroleum ether extract of leaves). Apart from these metaphase fragments (2.86 percent) due to treatment with alcoholic extract of stem, telophase bridge (1.31 percent due to treatment with water extract of leaves) and telophase tropokinesis (1.18 percent) due to treatment with alcoholic extract of stems are also observed (Table 4).Other extract treatments results are also seen in Table 4.

In general, the mitotic index decreases with increasing distance from the root cap junction. Cells of the root cap protect the root and must be constantly replaced as they are damaged or scraped away. The apical meristem, just beneath the root cap, contains most of the root's dividing cells. Therefore, cells in this area must complete the cell cycle often. Some daughter cells become part of the root cap, others differentiate and elongate into primary tissues of the root. If we allow growing roots to take up a radioactive precursor of DNA, almost all of the labeled cells lie in the meristem.

The mitotic index can also be used to quantify differences in cell division when an environmental parameter is changed. Plants grown in space in microgravity had a greater mitotic index than control plants grown on the ground. In zero-gravity, the gravity sensing cells in the root cap are unable to send the proper orientation signals. These signals normally inhibit growth in cells that are more distant from the root cap junction, and direct elongation of the primary root. In the absence of these signals, cells begin dividing to produce secondary roots, leading to a greater number of cells in mitosis.

Table 2:- Distribution of percentage dividing cells at various stages of mitosis of *Allium cepa* L. after treatment with aqueous, alcoholic, petroleum ether extracts of leaves of *Boerhaavia diffusa* L.

Concentration of treatment in percentage	Percentages of dividing cells at											
	Prophase in leaves extraction			Metaphase in leaves extraction			Anaphase in leaves extraction			Telophase in leaves extraction		
	A	PE	W	A	PE	W	A	PE	W	A	PE	W
Control	80.00	80.00	80.00	4.87	4.87	4.87	7.80	7.80	7.80	7.30	7.30	7.30
0.05	82.22	41.94	57.89	8.80	17.74	11.84	6.66	29.03	9.21	2.20	11.29	9.21
0.1	51.72	46.29	70.67	31.03	18.52	10.67	17.26	25.92	8.00	0	9.26	8.00
0.15	54.83	36.21	66.67	22.58	10.34	15.48	12.90	41.38	8.93	9.60	12.07	8.93
0.2	26.31	29.63	63.07	47.36	24.07	13.85	10.52	33.33	10.77	15.79	12.96	10.77

A- Alcoholic extraction of *Boerhaavia diffusa* L. leaves, PE- Petroleum ether extraction of *Boerhaavia diffusa* L. leaves, W- Water extraction of *Boerhaavia diffusa* L. leaves.

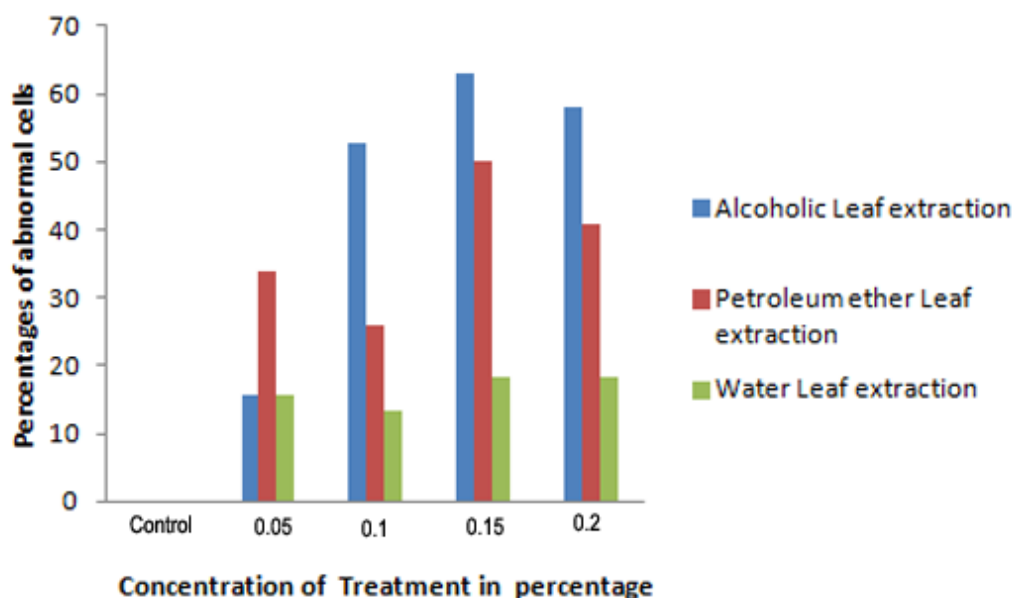
According to Badr and Ibrahim (1987) decrease of mitotic index level shows that experimental material had mitodepressive effect resulting in the inhibition of cells access to mitosis. It means that the treatment disturbs the normal cell cycle process by preventing biosynthesis of DNA and/or microtubule formation. Hence, this effect could be formed by decreased ATP level or suppression of the engine of energy production. Reduction in the mitotic activity could be due to inhibition of DNA synthesis or a blocking in the G2-phase of the cell cycle, preventing the cell from entering mitosis.

The common abnormalities are stickiness, fragment, unoriented chromosomes and bridges. Stickiness was seen in prophase, metaphase, anaphase and telophase. Fragment was seen in metaphase. Unoriented chromosomes were observed in metaphase and anaphase. Bridges was seen in anaphase and telophase. The types of abnormalities scored in this investigation resemble those reported by Somashekhar and Goda (1984). It has been suggested by them that stickiness is a type of physical adhesion involving mainly the proteinacious matrix of chromatin material.

Table 3:- Percentage of abnormal dividing cells at mitosis of *Allium cepa* L after treatment with various concentrations of aqueous, alcoholic, petroleum ether extract of leaves of *Boerhaavia diffusa* L

Concentration of Treatment in percentage	% of abnormal cells		
	Alcoholic Leaf extraction	Petroleum ether Leaf extraction	Water Leaf extraction
Control	0	0	0
0.05	15.78	33.87	15.78
0.1	52.63	25.92	13.33
0.15	63.15	50.00	18.45
0.2	57.89	40.74	18.46

The fragments may be a result of breaks since many reports point out that elevated frequency of aberrations is in the form of chromosomal breaks. The sticky bridges resulted from difficulty in anaphase separation due to adhesion or clumping of chromosomes or to depolymerization and cross linking of the DNA of the chromosomes. El-Ghamery, (2000) report that chromosome bridges may be due to the chromosomal stickiness and subsequent failure of free anaphase separation or may be attributed to unequal translocation or inversion of chromosomal segments leading to mutations.

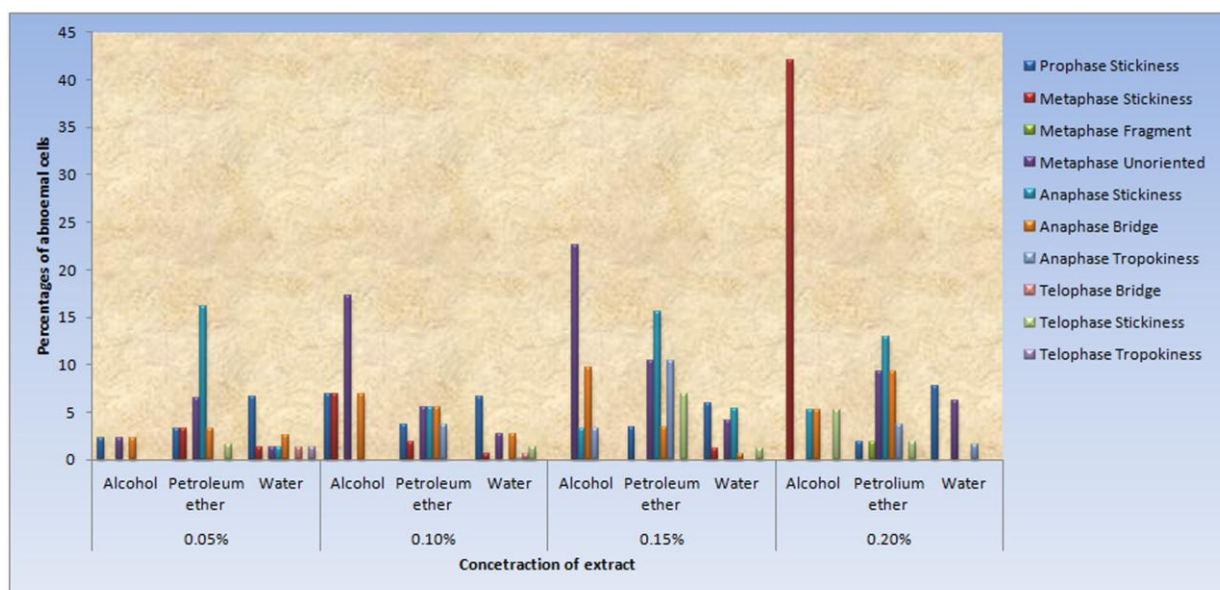


Graph 2:- Percentage of abnormal dividing cells at mitosis of *Allium cepa* L after treatment with various concentrations of aqueous, alcoholic, petroleum ether leaf extract of *Boerhaavia diffusa* L

Table 4:- Percentage of abnormalities at various stages of mitotic division in *Allium cepa* L after treatment with alcoholic, petroleum ether and water extract of leaves of *Boerhaavia diffusa* L.

Concentration of treatment (%)		0.05%			0.1%			0.15%			0.2%		
Stage	Abnormality	A	PE	W	A	PE	W	A	PE	W	A	PE	W
Prophase	Stickiness	2.22	3.22	6.58	6.89	3.70	6.67	-	3.45	5.95	-	1.85	7.69
Metaphase	Stickiness	-	3.22	1.31	6.89	1.85	0.67	-	-	1.19	42.10	-	-
	Fragment	-	-	-	-	-	-	-	-	-	-	1.85	-
	Unoriented	2.22	6.45	1.31	17.24	5.56	2.67	22.58	10.34	4.17	-	9.26	6.15
Anaphase	Stickiness	-	16.13	1.31	-	5.56	-	3.22	15.52	5.36	5.26	12.96	-
	Bridge	2.22	3.22	2.63	6.89	5.56	2.67	9.67	3.45	0.59	5.26	9.26	-
	Tropokinesis	-	-	-	-	3.70	0.067	3.22	10.34	-	-	3.70	1.54
Telophase	Bridge	-	-	1.31	-	-	0.67	-	-	-	-	-	-
	Stickiness	-	1.61	-	-	-	1.33	-	6.896	1.19	5.26	1.85	-
	Tropokinesis	-	-	1.31	-	-	-	-	-	-	-	-	-

A- Alcoholic extraction of *Boerhaavia diffusa* L. leaves, PE- Petroleum ether extraction of *Boerhaavia diffusa* L. leaves, W- Water extraction of *Boerhaavia diffusa* L. leaves.



Graph 3:- Percentage of abnormalities at various stages of mitotic division in *Allium cepa* L. after treatment with leaves extract of *Boerhaavia diffusa* L.

Conclusion:-

In the present study the effects of various extracts of *Boerhaavia diffusa* L. on mitotic cells of *Allium cepa* L. is investigated. The mitotic index is found to be reduced as the treatment concentration increases. But at low concentration there showed an enhancement in the number of dividing cells. The percentage of abnormal cells is found to be increased with increase in the concentration of the treatment. The common abnormalities observed are unoriented chromosomes at metaphase, fragments, tropokinesis, and bridges along with a predominance of stickiness. The results indicated that there exists a toxic principle in *Boerhaavia diffusa* L. and excessive use of its preparation may lead to anomalies in epithelial linings. Further studies are inevitable for understanding the component present in the extract which is responsible for cellular damage produced in root meristem of *Allium cepa* L. and prolonged use of *Boerhaavia diffusa* L. leaves as vegetable, good or badly affect epithelial linings of human.

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RESEARCH ARTICLE

COOPERATIVE LEARNING AND ETHNIC RECONCILIATION IN ESL CLASSROOM.

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Abstract

This study explores the efficacy of using Cooperative Learning (CL) in enhancing ethnic reconciliation among students while improving their L2 competency. According to Coelho (1994), students learn languages best when they are engaged in real interaction and CL is especially appropriate for multi-cultural classrooms. Sri Lankan education has its partly contribution to linguistic nationalism that gradually led to a disastrous ethnic conflict (Bannon 2003). Having understood that, recent research has found that English as a link language that binds cultures together can support reconciliation and greater economic prosperity throughout the country. Similarly, orienting students towards a new teaching-learning environment and fulfilling the objective of socializing with multicultural and multiethnic communities and respecting diversities, the English Language Teaching Unit of Sri Lanka Institute of Information Technology too conducts the L2 module of the Orientation 2016/17 for the new intake using cooperative learning activities. Thus, through this case study, it is aimed at providing students' and teachers' perceptions towards feasibility of CL for ethnic reconciliation. The students are divided into groups of 12 irrespective of their ethnicity or competency. The data gathered through questionnaires and interviews from a sample of 48 students and 3 teachers involved in the Orientation program highlight that CL in multiethnic ESL classroom encourages students' participation and promotes positive interactions between students. Further, as per the qualitative data, the interaction among students during cooperative tasks facilitates their L2 competency. In contrary, some groups of students gain attention more than others as some mostly focused on their individual performance despite collaboration among team-mates. However, the results indicate that CL allows each student to play an important role in doing the assigned tasks, resulting in enhancing team-spirit and ethnic reconciliation.

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Introduction:-

This paper presents an overall evaluation on the integration of cooperative learning in the ESL classroom with special reference to the Orientation 2016/17 offered by Sri Lanka Institute of Information Technology (SLIIT), Malabe, Sri Lanka in order to uplift the ethnic reconciliation among the freshers from diverse backgrounds while improving their L2 competency.

After being exposed to a long period of civil war, the government of Sri Lanka is focusing on ways to develop the school curriculum to integrate multicultural education into its peace education curriculum in order to foster intercultural understandings⁽⁷⁾. In terms of the 2011 Lessons Learnt and Reconciliation Commission Report in Sri Lanka⁽⁶⁾, a National Youth Policy has been presented and one of its goals is to strengthen social interaction among youth. The Ministry of National Languages and Social Integration has taken measures to implement an action plan to recognize diversities in Sri Lanka promoting the Trilingual Sri Lanka National Plan which emphasized the importance of Sinhalese, Tamil and English proficiency to achieve ethnic harmony. As per the UNDP report, youth has been victimized due to the long lasted ethnic conflict and steps should be implemented to make youth not to be ethnic extremists, make wiser non-violent choices, and invest their energy in peace and reconciliation. When the medium of instruction has changed to vernacular languages in 1956, the schools were separated. In order to reverse this division, there is a necessity of promoting a common language among youth and English has been identified as the primary medium which clears ethnic or cultural separations.

As Gnanaseelan⁽⁵⁾ (2011) identifies English language education as a good method which has the potential to help overcome a wide variety of social and cultural tensions despite the controversial opinions emerging time to time. However, realizing the great need for students to be armed with English language proficiency, it has been understood by various studies that English language can empower binding cultures together. Having realized this, corresponding to the necessity of being fluent in two official languages in Sri Lanka (Sinhala and Tamil), English language has also been integrated in the school curriculum as the Second Language (L2) and even the English medium education has been promoted. When it comes to tertiary education, the medium of instruction in most of the modules is L2. Thus, almost all the tertiary educational institutes conduct ESL courses; thereby the students' L2 proficiency is enhanced. However, it is necessary to notice whether the measures taken so far have been complied with the emerging demands in the education in Sri Lanka i.e. to cater a group of students from multiethnic and multicultural backgrounds. Bannon⁽¹⁾ (2003) has identified that any curriculum should consider how diversity is represented in the context of teaching so that the ethnic harmony is spread. Thus, as I mentioned previously, the tool for this is the proper utilization of English language or lessons in ESL classroom where all the students in the institution are gathered despite their respective disciplines.

Moreover, it has been observed by the Faculty of Computing of Sri Lanka Institute of Information Technology that the undergraduates tend to form groups who share the same characteristics or perceptions without being mingled with other students from diverse backgrounds. As per the observations made by the researcher, most often, the students tend to form groups or 'clicks' on their own and sometimes, certain minorities are discriminated due to many reasons such as ego, wealth, religions, attitudes or the cultural influences. Yet, in order to achieve the aspirations of the country, the students should grow peace and harmony within them. When it considered the Orientation Program 2015/16, the faculty understood that teaching the students technical context or grammar and writing during their orientation had not generated expected unity among themselves as the students focused only on the lessons rather than making contacts or interactions within a diverse context. It was noticeable that the number of students attending classes dropped towards the end of the program and those who with keen interest in learning English and those who wanted to win the good will or the attention of the teachers came to the lectures and this lead to ruin the intended aim of the ESL unit. Further the ultimate goal of the degree should not be only producing a graduate with good scores but also a quality team-player who can succeed in the competitive markets. Thus, there emerged a necessity of implementing a new orientation program that caters the values and ethics to students while enhancing their soft skills and English language proficiency. The result of this was the introduction of the Orientation 2016/17 – 'Bootstrap', making it a boot to enhance students' achievements. The priority was given to ESL lessons throughout and the ESL work plan was woven around the objective of orienting students towards a new teaching-learning environment and fulfilling the objective of socializing with multicultural and multiethnic communities and respecting diversities while improving their L2 proficiency.

In fulfilling the above requirements, the popular methodology in language classrooms; the cooperative learning method was adopted to the proposed Orientation 2016/17.

The researchers consider cooperative learning as a method to help improve students' academic performance as well as it encourages student connections and promote literacy skills. The basic theory of cooperative learning in ESL classroom is to make them work cooperatively to strengthen language and personal bonds.

As per the Education Research Consumer Guide⁽³⁾ by the U.S. Department of Education, cooperative learning is, 'a successful teaching strategy in which small teams, each with students of different levels of ability, use a variety of learning activities to improve their understanding of a subject. Each member of a team is responsible not only for learning what is taught but also for helping teammates learn, thus creating an atmosphere of achievement'.

This definition is clearly covered by the cooperative learning strategies. Further as Coelho⁽²⁾ (1994) explains the students from different cultural backgrounds learn best through cooperative activities and such activities enhance the learning of all students. The students will be working in groups to achieve the task assigned to them. Felder and Brent⁽⁴⁾ (1996) have conducted in depth research on cooperative learning and proven that CL is effective in a variety of instructional settings. As they describe, when it compared to individualistic tasks, individual performances are better when cooperative methods are utilized in the classroom. It further results in a variety of cognitive and affective outcomes. The cooperative learning methodology has been greatly examined by Coelho (1994, *ibid*) in her book: 'Learning together in the multicultural classroom'. She argues that students learn best when they are in a multicultural setting. As she highlights, students spend a limited time in the ESL classroom within their timetable. Thus the group interaction for ESL students is important. When the students are in heterogeneous groups, it makes them cohesive and cooperative and it leads to better language acquisition. However, an ample of studies pertaining to cooperative learning has been conducted so far. Yet, there is a dearth of research related to achieving ethnic harmony through cooperative learning in tertiary ESL classroom in Sri Lanka.

Considering the above literature, understanding the urge for ethnic reconciliation in the country and realizing the requirements of the Sri Lanka Institute of Information Technology, the English Language Teaching Unit of SLIIT carried out the Orientation Program 2016/17 in order to fulfill the objective of socializing with multicultural and multiethnic communities and respecting diversities while improving their L2 competency and making it a lifetime experience for them.

This qualitative study is an evaluation of the said Orientation program which answers the research question; Will the utilization of cooperative learning in ESL lessons enhance ethnic reconciliation among students while improving their L2 competency?

Methodology:-

The students were divided into groups of 12 irrespective of their language proficiency, discipline, ethnic or cultural diversities. This division is done by the institution. Due to the large number of students enrolled in the first year batch and having considered the limited number of classrooms and time slots to occupy all the students, the number of members per group was made 12. The sample of this study was consisted of 4 groups (48 students) and 3 teachers who have experienced teaching in the previous Orientation program as well. The diversity of the student sample was as follows.

Ethnicity	Religion
Sinhala – 28	Buddhist – 22
Muslim – 11	Islam – 11
Tamil – 8	Hindu – 5
Other – 1	Catholic – 9
	Other – 1
Total – 48	Total – 48

Unlike the Orientation 2015/16 which focused on enhancing all four language skills through more individual tasks and classroom teaching, the new program was consisted of group activities improving social/ethnic interactions and active team-wise participation that would equally result in the improvement of four language skills.

It is vital to emphasize that the cooperative learning techniques presented by the researchers had been adopted and amended to suit the said Orientation program. The LOs were communicated to students properly at the beginning: socializing with multicultural and multiethnic communities and respecting diversities and enhancing L2 proficiency, so that they would know the expected outcome of the program and how the program was run.

This study focused on the data gathered based on four main activities conducted throughout the program. It is necessary to highlight that apart from the list below, mini cooperative learning activities were constantly conducted in the classroom leading to the following major activities.

Food Festival:-

The aim of this activity was to improve students' day-to-day conversational skills, negotiation skills and unity across different cultures. The student groups were supposed to put up a food stall selling both oriental and western food. There were sellers, buyers and students to welcome and greet the visitors to the stall. They were in costumes which reflect different cultures.

Tasks and skills: Practicing dialogues/ conversations – Speaking Writing invitations and posters – writing Searching and discussing about different food and reading recipes – reading and speaking

Post-activity (in-class) – Students were asked to discuss about the variety of food they consumed, different costumes/cultural symbols witnessed and their overall experience with their group members. The students were guided and observed by the relevant facilitator.

Literary Carnival of SLIIT:-

The aim of this activity was to improve students' presentation skills, ability to describe people, places or things using relevant vocabulary and sentence structures and also to make them carryout some research on various cultural/literary icons/figures to be demonstrated at the carnival. As a group, the students were asked to pick a cultural or literary figure and read about his/her background. Then they were asked to demonstrate the figures and do a presentation explaining them in a creative way with the participation of all the team-mates.

Tasks and skills: Searching and discussing about different costumes – speaking and reading Role-play – speaking Doing presentations – presentation skills Watching a video clip and answer the questions on the worksheet – listening, writing Post-activity (In-class) – Students were instructed to talk about different figures/icons they witnessed and to explain why they are unique. It made them to realize that every culture has its own uniqueness so that we should respect them. Further they were encouraged to actively participate in group discussions based on the demonstrations of figures at the carnival and raise their own thoughts. The students were guided and observed by the relevant facilitator.

The four corners:-

The aim of this activity was to enhance students' skills in giving opinions using relevant vocabulary and phrases and supporting and opposing ideas in an appropriate manner. The students were asked to listen to the statements on diversity, peace and tolerance read by the respective facilitator and run to the corners of the class which were labeled as Strongly agree, Agree, Disagree and Strongly disagree. Next they had to build up an argument justifying their point of view. All the group members were given a chance to present thoughts in front of the task and the cooperative effort is observed.

Tasks and skills: Listening to the statements – listening, comprehension Giving opinions using appropriate language – speaking, vocabulary Building up arguments – conversations, questioning and answering

Waving Flag:-

The aim of this activity was to prepare themselves for explaining and justifying facts while promoting the importance of unity and harmony among people. They were encouraged to discuss and correct their grammar and sentence structures in writing. The student groups were provided with stationary and asked to design a flag and write a poem or a prose for each group based on one of the themes: Peace, Tolerance and Diversity.

Tasks and skills: Discussing about each student's culture – speaking Poem, prose – writing, reading, comprehension Presentation of the flag and role-play – speaking, presentation skills.

Post-activity – the groups presented their flags to the class explaining its specific symbols and the theme. Further they recited the poem or read the prose and explained why they selected such a theme and a set of vocabulary. The facilitators guided the students so that each group member was not neglected.

After two months of activities, the students were given a semi-structured questionnaire as some students would not convey genuine feelings when they are interviewed in person. Some students and three teachers were interviewed in order to seek in depth views and attitudes of them towards the new Orientation program. The next section of this paper presents the feedback gained from the students and teachers and some data have been presented verbatim.

Discussion:-

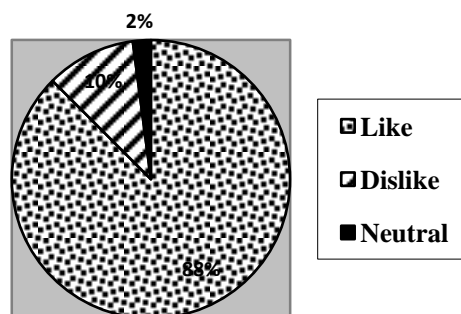
The cooperative Learning has been a popular learning technique among many teaching/learning settings and the first part of this research discussion will focus on the effectiveness of cooperative learning towards ethnic reconciliation among students as it is the primary objective of this study. Next the students' and teachers' feedback on improving L2 will be discussed.

As per the questionnaire data, the students had indicated their level of agreement or disagreement with the statements as follows.

Table 1: Responses of the students on Cooperative learning in ESL classroom					
Statements	Strongly Disagree	Disagree	Neutral	Agree	Strongly Agree
1. When I work together in a mixed ethnic group I achieve L2 proficiency more than when I work individually.	10.41%	6.2%	12.5%	27%	41.6%
2. When I work with the group, it prevents me from demonstrating personal abilities in activities.	27%	25%	20.8%	10.41%	16.6%
3. Even though the students' cooperativeness is expected when doing activities, some students do not help each other.	27%	27%	14.5%	14.5%	14.5%
4. Cooperative learning helps me to socialise more and encourage me to take part in lessons.	10.41%	12.5%	12.5%	29.1%	35.4%
5. Some students always try to be mingled with their own ethnic groups only.	20.8%	22.9%	22.9%	14.5%	18.7%
6. Cooperative learning improves my attitude towards ethnic reconciliation.	12.5%	10.4%	6.2%	31.2%	39.5%

Moreover, most students have shown a positive attitude towards the new learning system they experienced after the school time as shown in Figure 1 below. .

Figure 1: Students' overall attitude towards the new Orientation Program



It is noticeable that 86% of the sample drew positive attitudes towards the new Orientation program which was consisted of cooperative learning activities. 10% of the students did not like the program and 4% of the students didn't either like or dislike the program.

The students who liked the program admitted that this was a new system than what they experienced in school. As per the qualitative data, the positive feedback gained was that they were so curious to follow the program as new and exciting activities had been arranged for them. For most of them it was the first time that they engaged in cooperative learning in the English class with a specific objective of enhancing ethnic reconciliation and enhancing English proficiency. Further, they have been impressed by the four main activities I mentioned in the methodology. They appreciated the fact that they were in mixed cultural groups so that they could make friends from different backgrounds and those friendships were seem to be everlasting.

Response 1: "I could build up many good relationships with my other ethnic friends."

Response 2: "I thought Sinhala people are proud. I didn't like them earlier."

The students admitted that the Waving Flag activity gave them a chance to get to know each other. As the final product of the making of Flag activity was followed by a series of cooperative learning tasks such as getting to know each other, discussing and understanding similarities between them, discussing and reading about their different cultures paved them the way to be exposed to a range of vocabulary and different speaking patterns which improved their desire to use English. One Tamil student emphasized that even though he didn't know English much, English was the only language that could be used as the medium of communication between each other and he was forced to do so. Finally it has taken off his fear of speaking English and now he does his best to speak in English as much as possible.

Response 3: "This is the first time I am working with friends from other races. It is a nice experience"

As observed by the teachers, initially the students had tried to get together with the same race, yet eventually they had mingled with the other students as well.

Response 4: "I didn't like Tamil people at all. But now I think I have found a best friend. He is Tamil."

This is a key response by a student in the sample as the expected outcome of ethnic reconciliation has been a success due to the fact that he has found his best from another ethnicity.

Response 5: "We are waiting to eat 'Watalappan' prepared by our Muslim friends. They made delicious Watalappan for the food fiesta. We want them to make more again."

One student explained that the Food Festival was a wonderful experience as he learnt many things engaging in cooperative learning activities such as searching about different food from different cultures which enhanced reading skills while growing respect towards different cultures, activities to practice small dialogues which drew him to use language, and writing posters and invitations for the Food Festival which enhanced their writing skills.

It is clear that certain misperceptions grew in students' minds had been changed due to the fact that that they experienced working with people with other religions or races. They seemed to have had certain prejudices and misconceptions on working together and being together with people from different ethnicities and the program has let them mould their attitudes and grow awareness on peace and harmony. Thus, when it considered the qualitative data, it is clear that overall the learning context and the cooperative learning integrated had been impressed those 88% of students.

However there were 10% of students who did not like the program. The major reason for this was that they were from different ethnic contexts and mingling together was a bit difficult task at the very beginning. Some said that they preferred to be with a group of students who speak the same language or who belong to the same ethnicity so that they would not feel uncomfortable or insecure.

Response 6: "I feel so insecure... I don't like others. They always talk in Sinhala."

Response 7: "I don't have anything to do with them. I like my friends only."

Response 8: "Whenever teacher gives an activity, only they do. I don't know what's happening. I have only two more Sinhala Speaking friends in the groups. Others always talk in Tamil. So we skip the lecture sometimes."

As per the above statements made by the students, some still have the idea that they should work with people who belong to the same category. The cooperative activities are a burden to them as they have to be socialized with other ethnicities in the group. This marginalization has created a gap between them. As per Table 1, it is evident that about 42% of students acknowledge that students tend to befriend with people from their own ethnicities and other students are neglected.

Response 9: “Sometimes one specific ethnic group only take the decisions and don’t give chance to do to activities to others.”

Response 10: “They make decisions. We do nothing. Just listening to them. And I am ok with it. But not always.”

Response 11: “Some people want to show-off. Or over act. A headache.”

As the sample suggested there were students who tried to draw attraction of the others towards themselves so that the expected LOs were not achieved. As it should be a cooperative learning task, other students found such actions to be unpleasant and a burden.

Response 12: “I learnt nothing from the program.”

Response 13: “I am not social. I don’t want to be. I like lonely...”

Response 14: “I don’t mind group works. But I like to work on my own.”

It is noticeable that there is an average of 15% of students (see Table 1) has neutral attitudes towards working in cooperative activities in ESL classroom and achieving expected outcomes. In further interview it is indicated that they are the students who are able to adjust themselves to any situation and remain unnoticed and unattended to a certain extent when any discrimination occurred or any cooperative task is assigned to them. They simply let others to be shown off and wear neutral attitude towards the tasks or objectives. This has been clearly stated by the teachers too at their interview. In addition, the teachers suggested there were certain students who make troubles and cause disciplinary problems. Yet, the teachers strongly acknowledged that such behavior can be settled if this cooperative learning system had been continued for some more time.

As per the teachers’ view, the integration of cooperative learning in the orientation program was an overall success even though there are minor shortcomings. The key responses made by the teachers have been presented in Table 2 below.

Table 2:- Responses of the teachers on Cooperative learning in ESL classroom

Positive comments	Negative comments
<p>The students are happy to be with diverse people.</p> <p>Students improve their social skills when engaged in cooperative learning.</p> <p>It enhances long lasting friendship among students</p> <p>It helps to improve their English language usage.</p> <p>The students build up ethnic harmony through cooperative learning.</p>	<p>Sometimes, cooperative tasks make disciplinary problems.</p> <p>Some students tend to use their vernacular language when doing cooperative tasks which make certain minorities uncomfortable.</p> <p>Some students try to draw attraction than others when doing activities.</p> <p>Some students do not actively engage in activities and sometimes skip lectures.</p> <p>Some students’ attitudes cannot be changed within a short period in the cooperative learning setting.</p> <p>Content is too big to be covered with a limited time.</p>

According to the teachers, the main positivity they had identified was that the energy and willingness of the majority of students to take part in cooperative learning activities designed for them. As per the first main activity of designing a flag and a poem, the students were enthusiastically speaking and discussing about their cultures in order to find one particular aspect or an agreement in common. On the contrary, certain students have tried to use either Tamil or Sinhala to communicate with each other from the same race which made others uncomfortable.

Response 15: “Students actively took part in the role play activities and four corners which made introverts to be turned into extroverts.”

This is important that the students have become more socialized which makes them to unveil their true opinions and be open making it a more strong and healthy relationship.

Response 16: “The students feel they are valued and not neglected.”

As per the research by Yahya and Huei⁽⁸⁾ (2002), cooperative language tasks are useful to upgrade students’ proficiency levels and further it allows each student to take an important part in doing the group’s assigned tasks. It shows that they are valued as each student’s expertise is taken into consideration when completing the group task. This has been clearly pointed out through the Food Festival. In order to make such a big event a success, each student’s contribution was a must. This activity involves more speaking/ conversations which improves their speaking and strengthens their relationships.

However, the teachers highlighted the fact that some students had skipped some lectures. As I mentioned earlier, this is due to the fact that those students were not willing to take part in discussion sessions very much due to their poor proficiency. Yet as some students suggested, they had realized cooperative learning environment as the ideal platform to improve their proficiency while helping each other. This further broadens unity among students.

Finally the students were questioned of their feeling to be a member of their respective ethnic groups. In spite of the 10% of the students who disliked the program, the rest was highly impressed to be in a diverse group and had realized the essence of ethnic reconciliation leading to peace and harmony.

All in all the teachers stated that the cooperative learning platform was a success as the student made friends irrespective of other cultures, races, languages or religions. Further they had started to communicate in English to their level best despite their fear for English, introversion or poor proficiency making the learning outcomes of the orientation program a success.

Conclusion and Recommendations:-

Coelho’s (1994) research study has proven that cooperative learning helps students in multicultural classrooms to develop more appropriate skills for their futures. The proper guidance and instruction by the teacher drive the students towards proper learning and working together.

According to the Orientation 2016/17 of Sri Lanka Institute of Information Technology, the students were expected to be united as one family while improving their English proficiency. As it was explained in Coelho’s book, these skills will lead the students to productive and quality graduates in the future broadening their capacity to work in any environment full of diversities.

As per the results of this study, majority of the students (88%) were highly motivated by the teaching learning through cooperative learning strategies. Further the grouping of the students by the institution before the commencement of the classes was a success as the students were given an opportunity to explore a new diverse learning environment and get to know new friends. If the grouping had been done by the students themselves, the expected outcome would not have been achieved.

As per the suggestions made by the students, a request for the same type of a cooperative learning setting for their semester studies as well had been made. This can be considered a positive feedback from students. Yet, as per recommendations, it is needed to notify certain drawbacks of the program highlighted by the students. 18% of students still emphasized that the students from the same ethnic group tend to form their own relationships with their respective comfort zones. Some find it really difficult to step out of their comfort zone even if they do have a desire to do so. Moreover, while other students are actively engaged in cooperative learning tasks, some are still neglected. These issues should be taken into consideration when conducting the semester classes. If certain students miss the opportunity to demonstrate their abilities due to other students’ behavior, the teacher should be attentive and strategic enough to identify such circumstances and conduct lectures giving equal opportunity to all of them. Thereby the team-spirit can be further enhanced.

According to the research data, more than 85% of the sample ensured that the overall program was a success as the expected learning outcomes are achieved. All the three teachers too stated that the overall program was a success as the majority of the students were able to attain the expected level of performance and attitudinal change. They confirmed this based on their class observations and discussions held with other students on the final day of the

class. Further this has been confirmed by the teachers as compared to their prior teaching experiences gained in 2015/16 or before working at SLIIT, students were more active and willing to do the tasks in order to be successful as a team.

Deviating from the monotonous style of teaching, cooperative learning provides a student friendly platform to perform in an interactive way while helping each other. As the teachers suggested, cooperative ESL tasks should be planned appropriately so that each individual is encouraged to involve in the lessons. When this methodology is applied in a multicultural setting, the reconciliation can be promoted as each student is involved in fulfilling tasks cooperating with the rest of the team-mates. As witnessed by the researcher, the Literary Carnival and the Food Festival were of an utmost success as the students' participation in activities was noteworthy and admired by the audiences. However, it is recommended that the teachers should have freedom to amend or adopt cooperative learning activities according to their own preferences and classroom settings in order to cater the best to the students.

In conclusion, Sri Lanka Institute of Information Technology offers degree programs in Computing, Engineering, Architecture, Quantity Surveying and Business which are of highly technical aspects and the students are lectured in English. Thus, apart from promoting ethnic reconciliation, to accomplish the aim of implementing English only speaking environment for students facilitating their L2 usage to be utilized in higher education, the said Orientation 2016/17 was a successful approach.

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RESEARCH ARTICLE

CLINICAL PROFILE OF BLADDER OUTLET OBSTRUCTION {BOO} IN FEMALE.

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Key words:-

bladder outlet obstruction , curable illness , urinary tract symptoms.

Abstract

Aim:- To evaluate female patients of bladder outlet obstruction and find out common causes and clinical presentation of boo . To understand diagnostic modalities, treatment modalities and complications of boo in female. **Study Design:-** This is a prospective study of 50 female patients presented with bladder outlet obstruction in a single centre. including only female patients > 15 years age and excluding females with prior surgery of urinary tract / perineal surgery / spine surgery / known spine pathology .

Result:- According to age: the maximum number of cases (22%) is seen in the age group of 56 years to 65 years , body mass index(bmi) majority (52%) of the patients were in the bmi group of 26 - 29 kg/m². Symptoms in female boo majority (96%, n: 48) had sensation of incomplete emptying at the end of micturation. etiology most common causative pathology was urinary tract infection 28% (n: 14) surgical treatment offered 30% (n: 15) and received urethral dilatation/ urethrotomy.

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Introduction:-

Bladder outlet obstruction (BOO) in females, a relatively uncommon condition has recently attracted more attention and has got better identified. The International Continence Society has defined BOO as a 'generic term for obstruction during voiding and is characterized by increased detrusor pressure and reduced urine flow rate.^{1,2}

The International Continence Society (ICS) states that: 'Normal voiding is achieved by a voluntarily initiated continuous detrusor contraction that leads to complete bladder emptying within a normal time span and in the absence of obstruction³.

The prevalence of Bladder outlet obstruction is unknown. Though, large retrospective review trials have estimated that, of the 19.4%~25.5% women who present with lower urinary tract symptoms, 6.5%~9.6% were classified as Female BOO. BOO may be induced by specific functional and anatomic causes, the resulting obstruction produces lower urinary tract symptoms (LUTS). Categorizing and understanding these symptoms and causes is crucial when proceeding with evaluation for specific diagnostic modalities to be used to fully delineate the degree of BOO.

The dynamics of voiding in females is more complex than in the males, presumably due to mobility of the bladder neck and proximal urethra as well as due to the action of pelvic floor movements and fasciae over the urethra. In addition, anatomic differences allow many women to empty their bladders by simply relaxing the pelvic floor, some augment voiding by abdominal straining.

Due to the diverse characteristics of Female Bladder Outlet Obstruction and the lack of Gold Standard in diagnosis the management of bladder outlet obstruction in females is perhaps as complex as its diagnosis.¹ However determining outlet obstruction in females still remains an intriguing proposition, requiring clinical studies and research⁴. Once the cause is determined, appropriate treatment will lead to resolution of symptoms in most patients.^{5,2}

Material and Method:-

This is a prospective study of 50 female patients presented with Bladder Outlet Obstruction. The study was conducted at Padmashree Dr. D.Y.Patil Hospital and Research Institute, Kolhapur from May 2012 to April 2015 at the Department of General Surgery.

Inclusion Criteria:-

Female Patients > 15 years age

Exclusion Criteria:-

Females with prior surgery of urinary tract / perineal surgery / spine surgery / known spine pathology.

Methods:-

All the patients were evaluated as per the proforma. Special attention to the voiding symptoms, associated gynaecological disorder, history of previous catheterization and operation were noted.

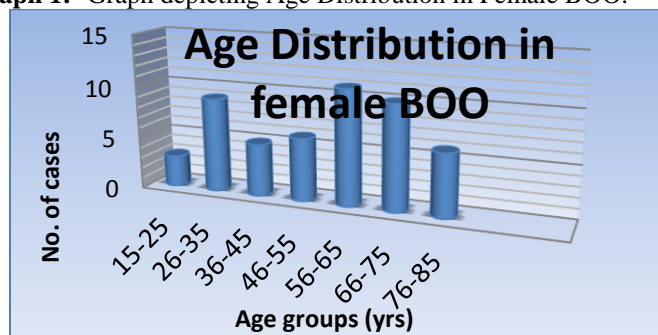
- Female patients presenting with lower urinary tract symptoms (LUTS) like increased frequency, urgency, nocturia, poor stream, hesitancy, straining, and incomplete voiding were screened..
- All relevant investigations (Routine hematological tests, urine for routine and cytology, culture and sensitivity, Renal Function Tests, Ultrasonography of abdomen and pelvis, Uroflowmetry and Cystoscopy) were done in these cases to establish the diagnosis.
- Maximum Flow rate (Qmax) of >15ml/second and voided volume of 150 ml is considered normal. Patients with Qmax<15 ml/second were suspected as a case of BOO.
- Depending on the findings and diagnosis, cases were considered for treatment accordingly.
- The proposed procedure was explained to the patient and relatives and a written consent was obtained prior to surgery. After appropriate anaesthesia Cystoscopy was carried out in the operation theatre. A rigid cystoscope (17 French) / Ureteroscope with fiberoptic light source and video camera fitted to it gently passed. Pan endoscopic examination was done from the meatus, urethra, bladder neck and the interior of urinary bladder. Appropriate procedures like Urethral dilatation, Otis urethrotomy, visual internal urethrotomy in stricture urethra, cauterization of pseudomembrane, bladder neck incision was done. Foley catheterization (No. 16 French) was done.
- Patients with no demonstrable cause and suspected to have neurogenic/ dysfunctional voiding were either subjected to Uro-Dynamic Studies or were prescribed medical management.
- Patients were advised regular follow up.
- A wide variation in prevalence from 2.7-46.4% is reported depending on the methodology adopted. This underlines the need for more reviews and research in this area.

Observations and Results:-

During this study a total of 50 female cases with Bladder Outlet Obstruction were studied. The following are the observations:

Age Distribution:-

The mean age of the patient in our study is 53.9 years (15years -85years). The maximum number of cases (22%) is seen in the age group of 56 years to 65 years.

Graph 1:- Graph depicting Age Distribution in Female BOO.**Body Mass Index(Bmi):-**

Majority (52%) of the patients were in the BMI group of 26 - 29 kg/m². The mean BMI was 25.2 Kg/m².

Lower Urinary Tract Symptoms in female BOO:-

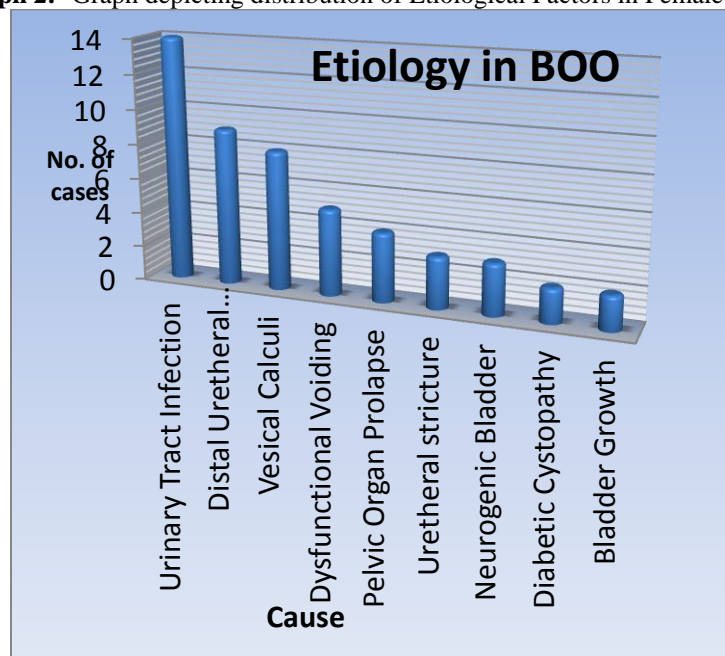
Considering all the voiding urinary complaints, majority of the patients (96%, n: 48) had sensation of incomplete emptying at the end of micturition. Most of them had poor stream (92%, n: 46) or straining for micturition (66%, n: 33). Similarly, common symptom was day time or night time frequency (92%, n:46)

Table 1:- Distribution of Lower Urinary Tract Symptoms in female BOO.

Symptoms	No. of cases	Percentage %
Urgency	15	30
Nocturia	42	84
Frequency	46	92
Straining	33	66
Incomplete emptying	48	96
Poor stream	46	92
Hesitancy	24	48

Etiology in female Bladder Outlet Obstruction:-

The most common causative pathology was Urinary Tract Infection 28% (n: 14), followed by Distal Urethral Stenosis in 18% (n: 9), Vesical Calculi in 16% (n: 8), Dysfunctional voiding in 10% (n: 5) of cases. Rest formed 28% (n: 14) of the cases.

Graph 2:- Graph depicting distribution of Etiological Factors in Female BOO.**Organisms Isolated on Urine Culture in female Bladder Outlet Obstruction:-**

In our study, 41 (82%) patients out of 50 had Urinary Tract Infection. On urinary culture the most common organism found was *E. coli* in 82.9% (n: 34), followed by mixed cultures in 4 patients 9.7% (n: 4). *Proteus*, *Klebsiella* and *Pseudomonas* were found in one case each (2.4%).

Distribution of Maximum Flow Rate 'Qmax' at Uroflowmetry:-

The maximum urine flow rate (Qmax) at Uroflowmetry in our cases was less than 12 mL/sec in 92% of the cases. 4 patients (8%) had a Qmax in the range of 13-15 mL/sec.

Maximum Flow Rate (Qmax) mL/sec	No. of cases	Percentage %
4-8	12	24
9-12	34	68
13-15	4	8

Table 8:- Distribution of Maximum Flow Rate 'Qmax' at Uroflowmetry.**Uro-dynamic study:-**

In this study, urodynamic study was performed in 20% (n: 10) cases. The flow rate is <15 mL/sec. The detrusor pressures in underactive bladder are <16 cm H₂O.

In neurogenic bladder there is poor bladder compliance with large bladder capacity. In cases of diabetic cystopathy there is poor bladder compliance with large bladder capacity. While in dysfunctional voiding there is varied bladder compliance and bladder capacity. In the cases of dysfunctional the detrusor pressures are >35 cm H₂O.

Blood Urea Levels:-

In our study 96% of the cases had Blood Urea levels in normal range. While 4% (n: 2) had deranged Blood Urea levels.

Serum Creatinine Levels:-

96% of the cases in our study had Serum Creatinine levels in normal range. 4% (n: 2) had deranged levels Serum Creatinine levels.

Co- morbidities:-

In our study 20% (n: 10) cases of 50 had Diabetes Mellitus, 28% (n: 14) cases had Hypertension. 12% (n: 6) had both Diabetes Mellitus and Hypertension.

Treatment:-**Treatment modality offered:-**

In our study, 52% (n: 26) of the cases were treated surgically, 28% (n: 14) cases given medical line of management, and 20% (n: 10) cases required combined (medical and surgical) modality of treatment.

Table 3:- Distribution of treatment modalities offered in female BOO.

Type	No. of cases	Percentage %
Surgical	26	52
Medical	14	28
Combined	10	20
Total	50	100

Surgical Treatment offered:-

In our study, 30% (n: 15) cases received urethral dilatation/ urethrotomy, 16% (n: 8) required Cystolithotripsy, 4% (n: 2) required Trans-Urethral Resection of Bladder Tumour (TURBT), and in 8% (n: 4) Pelvic Organ Prolapse (POP) repair was done.

Table 4:- Distribution of surgical treatment modality offered in female BOO.

Treatment modality	No. of cases	Percentage %
Urethral dilatation/ Urethrotomy	15	30
Cystolithotripsy	8	16
TURBT	2	4
POP repair	4	8

Discussion:-

The study on clinical profile of Bladder Outlet Obstruction in females (BOO) when compared to the previous studies carried out throughout the world, in most of the aspects there was a wide range of difference.

Age Distribution in Female Bladder Outlet Obstruction:-

In our study of BOO in females, mean age of the patients is 53.9 which is similar to some but different from many other studies.

- Our study shows that Bladder Outlet Obstruction is common in the post-menopausal women. The reason being changes in the hormonal status and age related anatomical derangements. Nadir et al.⁶ in his case study of 221 patients has found, the mean age group associated with BOO was 51.8 years(range: 22–91), which correlates to our study.
- Oravanet al.⁷ in his study shows that, mean age of the study population was 53.7 ± 13.6 years. Nitti et al.⁸ showed in their study of 76 obstructed cases the mean age was 57.5 years. Metinet al.⁹ in his study found the mean age is 52.6 years.

Incidence of Bladder Outlet Obstruction in Females:-

The incidence of BOO in our study is 6.5%. 72 cases were found to be obstructed and labeled BOO on the basis of uroflowmetry and significant post-void residual urine. Which also is closely in the range of various previous studies. Groutz et al.¹⁰ conducted a large retrospective review of women with lower urinary tract symptoms (LUTS), and recognized BOO in 6.5% of the cases⁵ which is similar to our study.

Lower Urinary Tract Symptoms (LUTS) in BOO.

- In our study of **Lower Urinary Tract Symptoms**, more than 84% cases presented with nocturia, frequency, straining, incomplete emptying, poor stream
- **Urinary tract infection(UTI)** is the most common etiology observed in our study.78% Urinary tract infection was the sole cause attributed to BOO.

- **Distal Urethral Stenosis (DUS)** is the 2nd most common etiology found in our study affecting 18%(n: 9) of cases.
- **Urethral Stricture** was the cause in 6% (n: 3) of the cases. They underwent Urethral Dilatation to 30 Fr. with Otis urethrotomy.
- There were 6% cases (n: 3) of **Neurogenic bladder** and **Pelvic Organ Prolapse (POP)**.

Organisms at Urine Culture in cases of BOO:-

E. coli was predominantly found in 82.9 % (n: 34) of the cases.

Uroflowmetry in evaluation of female BOO.

- In our study the mean Qmax was 9.6 mL/sec (4-14mL/sec). 92% of cases in our study had Qmax <12 mL/sec. Therefore our study includes severely obstructed group. Axelrod et al.¹¹ identified bladder-neck obstruction in a group of women, using and Qmax of <12 ml/s as cutoff. Yong et al.¹² have defined female Bladder Outlet Obstruction as Qmax <15 mL/sec. on Uroflowmetry.

Uro-Dynamic Studies in female BOO:-

In cases where identifiable anatomical cause for BOO was not found, Uro-Dynamic Study (UDS) was performed. The studies showed: maximum flow rate (Qmax) <12 ml/s, detrusor pressure at maximum flow (pdet.Qmax) of >20 cm H₂O in dysfunctional voiding cases

Co-morbidities associated with female BOO:-

- ❖ Of the 50 cases 20 % (n: 10) of the total cases were diagnosed cases of diabetes mellitus and 28% (n: 14) of the cases were a known case of hypertension. 4 % (n: 2) of the diabetics were diagnosed with changes of Diabetic Cystopathy.

Post Void Residue Study:-

All the cases of Bladder Outlet Obstruction in females underwent Pre-void and immediate Post-void residual urine study by abdominal Ultrasonography. 54% of the cases (n: 27) had a Post Void urine residue of >100ml. The mean PVR in our study is 161.5mL.

- Patients with functional BOO had a mean PVR urine of 243mL, while those with anatomical BOO had a mean PVR urine of 141.1mL. It is observed that, the patients with Dysfunctional Voiding (DV) had post void residue of more than 210ml. The mean Post void residue of DV cases was 298.2 mL of urine in our study of Bladder Outlet Obstruction in females. Haylen et al.¹³ have concluded that postvoid residual volumes higher than 30mL are significantly associated with increasing age, higher grades of prolapse, and increased prevalence of recurrent Urinary Tract Infection, this is evident in our study.
- Nittiet al.⁸ found a mean post-void residual urine (157 versus 33 ml, p <0.0001) in their study which correlates the value in our study.

Treatment Of Female Bladder Outlet Obstruction:-

To summarize, the treatment was mainly surgical, medical and in some cases combined approach was utilized. 52% (n: 26) cases were treated surgically, 28% (n: 14) cases received medical line of management while 20% (n: 10) cases required combined modality of treatment approach.

In our study, surgical treatment included urethral dilatation/ urethrotomy in 30% (n: 15) cases, cystolithotripsy in 16% (n: 8) cases, Trans-Urethral Resection of Bladder Tumour (TURBT) in 4% (n: 2) cases, and in 8% (n: 4) cases of Pelvic Organ Prolapse (POP), POP repair was done.

Thorough evaluation of the cases enables identification of the cause of BOO and to determine the treatment plan. The most important part of its identification is a high index of suspicion. Once the cause is determined, appropriate treatment will resolve the symptoms in most of the patients.

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RESEARCH ARTICLE

FACILE SYNTHESIZE OF GRAPHENE OXIDE BY MODIFIED HUMMER'S METHOD AND DEGRADATION OF METHYLENE BLUE DYE UNDER VISIBLE LIGHT IRRADIATION.

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Abstract

Graphene is a very huge and ultimate material for composite with metals, semiconductors and non metals in recent years, due to its has a unique atom-thick with two-dimensional (2D) structure, excellent physical properties like high conductivity and charge mobility, huge specific surface area, excellent mechanical, thermal and electrical properties. Thus, it has been regarded as an important component for functional materials, especially for developing a variety of catalysts and it has been considered widely as a prominent precursor and a starting material for the synthesis of this processable material. This work describes the synthesis of Graphene oxide (GO) by Modified Hummer's method and characterization of GO by Ultra violet visible spectroscopy (UV-Vis), UV-visible diffuse reflectance spectroscopy (UV-DRS), X-ray diffraction (XRD), Field emission scanning electron microscopy (FESEM) and Energy dispersive spectroscopy (EDX) analysis. The results obtained from the characterization techniques mentioned above is also explained in detailed and evaluation of catalytic application to environmental remedies, such as water purification of degradation of methylene blue under visible light irradiation.

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Introduction:-

Graphene oxide (GO) represents an emerging field of interdisciplinary science that spans a variety of disciplines, including chemistry, physics, materials science, device fabrication, and nanotechnology [1-3]. The current graphene boom started in 2004 when Geim, Novoselov et al. published the deposition and characterization of single sheets of graphite on solid supports. Their groundbreaking experiments on graphene were honored with the Nobel Prize in Physics in 2010 [4-7]. Exceptional electronic, optical and mechanical properties were discovered in quick succession as a consequence of the experience gained from other carbon allotropes [8-9]. In particular, the high charge carrier mobilities, the electrical and thermal conductivity, combined with transparency and mechanical strength make graphene highly attractive for future high-tech applications [30].

The current status of graphene oxide with respect to prototype applications has been extensively reviewed.[10] Many graphene-based devices outperform reference systems, for example in high-frequency transistors, foldable and stretchable electronic or photodetectors,[11] capacitors,[12] transparent electrodes,[13] sensors,[14] H₂ generation,[15] pollution management,[16] energy applications,[17] biomedical applications and in composite materials [30].

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Graphene is a two-dimensional (2D) carbon allotrope which can be viewed as both a solid and a macromolecule with molecular weights of more than $10^6 - 10^7 \text{ g mol}^{-1}$. In natural graphite, the graphene layers stick together through very pronounced π - π stacking interactions [30]. This noncovalent interlayer binding contributes significantly to the high thermodynamic stability of graphite. As a consequence, the wet chemistry of graphene is always concerned with overcoming these interactions. For example, a targeted exfoliation of graphite or the stabilization of solvent-dispersed graphene sheets always competes with reaggregation. It should be pointed out that a solid sample of graphene can only be stabilized on a support, such as a surface. A nonsupported graphene powder can not be expected to exist, since at least a partial restacking to graphite will take place! Another possibility of stabilizing individualized graphene is to “mask” the surface through chemical functionalization[18,30]. This paper we reported catalytic activity of graphene oxide (GO) under visible light irradiation and comparison the catalytic activity with graphite flake.

Properties of Graphene:-

Electronic properties:-

The electronic properties of graphene have received the most attention. Suspended graphene shows extremely high low-temperature charge carrier mobility, with a value approaching $200\,000 \text{ cm}^2 \text{ V}^{-1} \text{ s}^{-1}$ [19], making it an outstanding conductor of electricity. This excellent conductivity is due to the remarkable property of graphene that its charge carriers behave as mass less relativistic particles or Dirac fermions, and electrons can travel distances in the order of micrometers without scattering under ambient conditions [21]. This unique behavior has led to other phenomena in graphene. UV-DRS spectra revealed the graphene oxide has a zero-band gap 2D semiconductor due to its like as a semi metal, graphene exhibits a strong ambipolar electric field effect and the concentration of charge carriers can be as high as 10^{13} cm^{-1} , with room temperature mobilities of up to $15\,000 \text{ cm}^2 \text{ V}^{-1} \text{ s}^{-1}$ having been measured[22]. Moreover, an unusual half-integer quantum Hall effect (QHE) in graphene has been observed at room temperature[23,29].

Thermal properties:-

The thermal conductivity of suspended single-layer graphene at room temperature has been measured in the range $(4.84 \pm 0.44) \times 10^3$ to $(5.30 \pm 0.48) \times 10^3 \text{ W m}^{-1} \text{ K}^{-1}$ [24]. The superior thermal conduction property of graphene is beneficial for making graphene as an excellent material for thermal management [29]. The thermal conductivity of large-area, monolayer graphene grown by chemical vapor deposition (CVD) on a Au-coated SiNx porous membrane can reach about $2500 \text{ W m}^{-1} \text{ K}^{-1}$ at 350 K [25,29].

Mechanical Properties:-

According to Changgu Lee, Graphene is the strongest material ever tested, with a Tensile strength of 130 GPa and a Young's Modulus (defines stiffness) of 1 TPa [22,32]. Apart from this, Graphene is unbelievably light, weighing about only 0.77 mg/m². According to a Nobel announcement which illustrates that 1 m² of Graphene hammock would support a 4 kg cat, but would weigh only as much as one of the cat's whiskers [23,31].

Optical Properties:-

Graphene produces a highly opaque atomic monolayer in vacuum, as it has an ability to absorb approximately 2.3% of the white light. Adding another layer of Graphene increases the amount of white light absorbed by approx. the same value (2.3%) [24]. Once optical intensity reaches a certain threshold saturable absorption takes place [33]. Due to Graphene's properties of wavelength-insensitive ultrafast saturable absorption, full band mode locking has been achieved [34].

Experimental:-

Materials:-

Graphite Flakes (Sigma Aldrich), Sodium nitrate [NaNO_2], Potassium permanganate [KNO_3], Hydrogen peroxide 30% [H_2O_2], Sulphuric acid 98% [H_2SO_4], Hydrochloric acid 35% [HCl], Ortho phosphoric acid [H_3PO_4], Bismuth Nitrate [$\text{Bi}(\text{NO}_3)_3 \cdot 5\text{H}_2\text{O}$], Nitric acid [HNO_3 78%], Ammonium molybdate [$(\text{NH}_4)_6\text{Mo}_7\text{O}_{24} \cdot 4\text{H}_2\text{O}$] and Ammonium hydroxide [NH_4OH] and Methylene blue [$\text{C}_{16}\text{H}_{18}\text{ClN}_3\text{S}$]. All chemicals received from Merck India except Graphite flake received from Sigma Aldrich USA and used without further purifier.

Synthesis of Graphene Oxide by modified Hummer's method:-

Graphite flakes (2 g) and NaNO_3 (2 g) were mixed in 80 mL of H_2SO_4 (98%) and 10 ml of H_3PO_4 in a 1000 ml round bottom flask kept under at ice bath ($0-5^\circ\text{C}$) with continuous stirring. The mixture was stirred for 4 hrs at $0-5^\circ\text{C}$ temperature and potassium permanganate (12 g) was added to the suspension very slowly. The rate of addition was carefully controlled to keep the reaction temperature lower than 15°C . The mixture is diluted with very slow addition of 200 ml double distilled water, the ice bath was then removed and kept under stirring for 2 hrs at room temperature. The above mixture is kept in a reflux system at 90°C for 1h. After 1h, change the temperature to 30°C which gives brown colored solution. Again after 10 min, change it to 25°C , and maintain the temperature for 2 hrs. The solution is finally treated with 40 ml H_2O_2 by which color changes to bright yellow, 200 ml of water is taken in two separate beakers and equal amount of solution prepared is added and stirred for 1 hr. It is then kept without stirring for 3-4 hrs, where the particles settles at the bottom and remaining water is poured to filter. The resulting mixture is washed repeatedly by centrifugation with 5% HCl and then with distilled water several times until it forms gel like substance (pH- neutral). After centrifugation the gel like substance is vacuum dried [35].

Photocatalytic test:-

Organic pollutant such as methylene blue were chosen for photocatalytically activities evaluate of the as synthesized GO powder and Graphite flake (Sigma Aldrich USA). A 400 W HgX lamp was used as the light source to provide the simulated solar light. The experiments performed at an ambient temperature. The photocatalysts (GO or Graphite flake) 0.05 g were added into 50 mL of organic pollutant 5 ppm solution, and suspension was magnetically stirred for 30 min in the dark to reach an adsorption-desorption equilibrium with the photocatalysts. At a every 30 min time intervals, a 3 mL solution collected and then analyzed on UV-Vis spectrophotometer during the photo degradation process and same experiment perform as blank (without GO or Graphite flake). The concentration of methylene blue were determined by monitoring the variations in the main absorption centered at 664nm.

Characterization:-

The crystalline structure of the Graphene Oxide (GO) was investigated by powder X-ray diffraction (XRD, PanAlytical, X-Pert pro, Netherland) with $\text{Cu K}\alpha$ ($\lambda=1.54218 \text{ \AA}$) radiation . The size and morphology of the product were obtained with a Scanning electron microscope (FESEM-CARL Zeiss Germany, Model Ultra 55 FESEM, Gemini column, 1nm Resolution, detector is INLENSE), Energy dispersive spectroscopy (Oxford company, model 20 nm X Ma) UV-Vis & UV-Vis diffuse reflectance spectra of the sample were obtained in the range 300 – 800 nm using a UV-vis spectrophotometer (UV2550, Shimadzu, Japan) barium sulphate (BaSO_4) was used as a reflectance standard.

Results and Discussion:-

Figure.1. shows the UV-Visible spectra of Graphene Oxide, the peak at 215nm is $\pi-\pi^*$ transition due to $\text{C}=\text{C}$ bonding is an aromatic ring, where as the broad shoulder peak at 291nm is $n-\pi^*$ transition due to $\text{C}-\text{O}$ bonding. The absorption over 291 nm is expected to be caused by the conjugated fused ring plane; this indicates a $\text{sp}^2-\pi$ conjugated network.

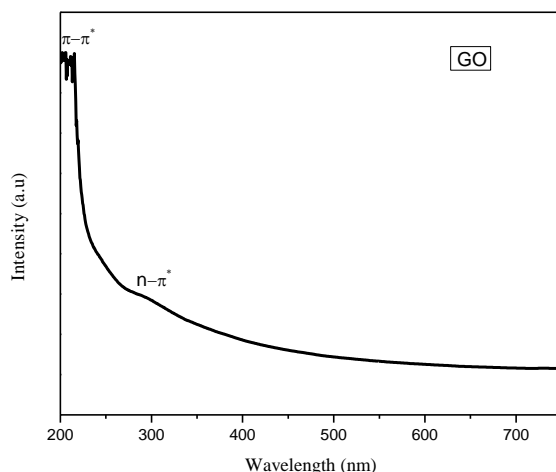


Fig.1:- UV-Visible spectra of Graphene Oxide

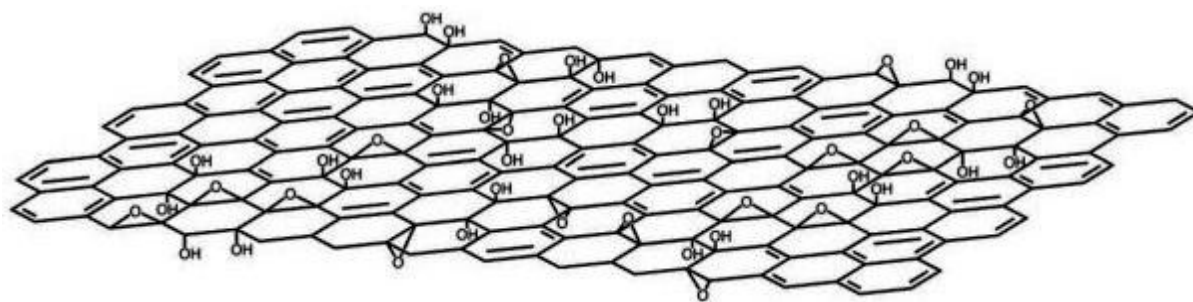


Fig 2:- Possible structure of Graphene Oxide

The XRD pattern obtained for as synthesized GO nanoparticles by Hummer's method is shown in Figure 3b. It shows the diffraction peak at $2\theta=10^\circ$, which is mainly due to the oxidation of graphite. The diffraction peak of pure graphite is found around 26° , corresponding to the highly organized layer structure with an interlayer distance of 0.34 nm along the (002) orientation is shown as inset in Figure 3a. The XRD pattern for synthesized GO by Modified Hummer's method is shown in Figure 3b. The disappearance of the peak at 26° and appearance of the peak at 10° , shows that the product is completely oxidized after the chemical oxidation and exfoliation, indicating an increase in d-spacing from 0.34 nm to 0.82 nm.

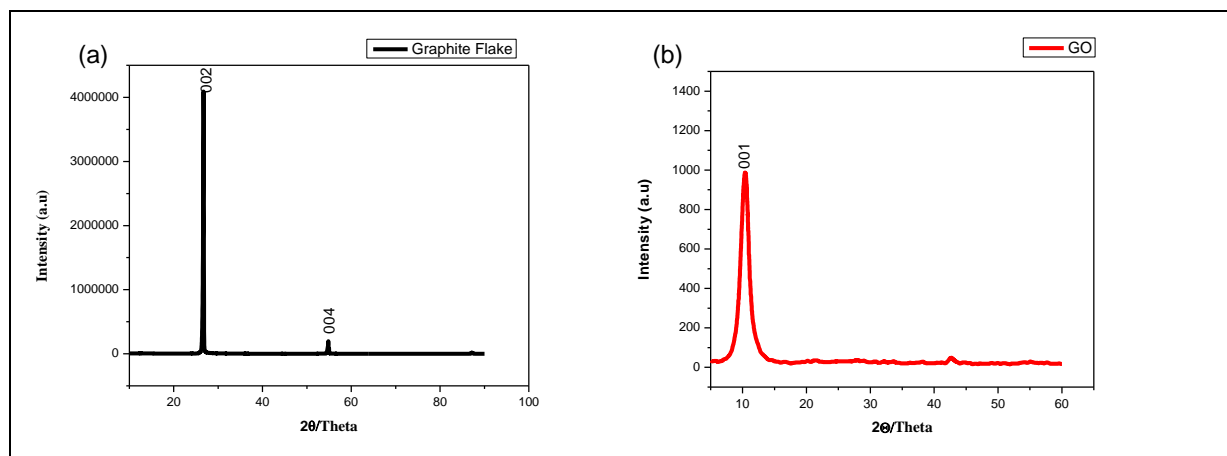


Fig.3:- XRD pattern of (a) Graphite flake, (b) Graphene Oxide.

Figure .4. Shows the FESEM images of Graphene Oxide. It shows single flakes of Graphene Oxide may be observed. Graphene Oxide flakes have relatively large surface and well defined and interlinked three-dimensional Graphene Oxide sheets, forming a porous network that resembles a loose sponge like structure with 49.38 nm range in size. Fig.5.shows the EDX spectra of Graphene Oxide, which revealed to purity of C,H,O elemental analysis and some impure of the elemental with Potassium (K) and Sulphur (S) also identified due to KMnO_4 and H_2SO_4 are precursor for the oxidation of GO..

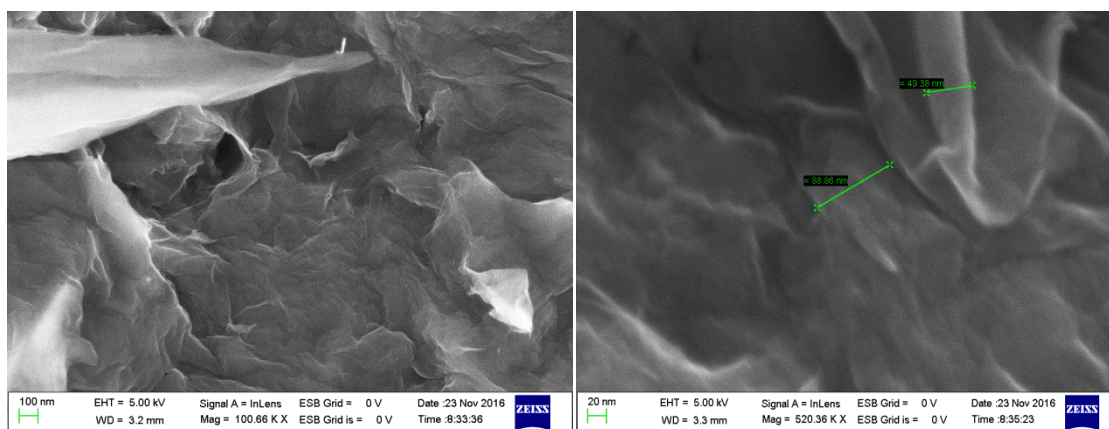


Fig 4:- FESEM images of Graphene Oxide

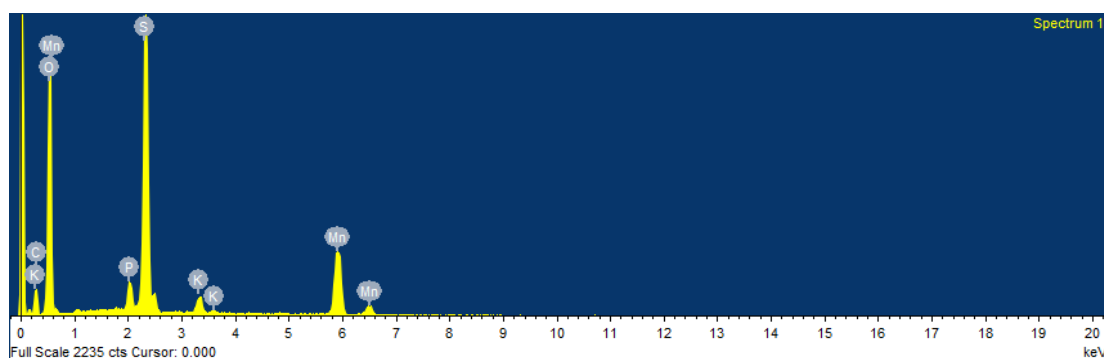


Fig 5:- EDX spectra of Graphene Oxide.

Fig.6. shows Ultra Violet Diffuse-Reflectance Spectroscopy (UV-DRS) is an important method for charactering the electronic state, optical state and calculates the band gap of semiconductor materials. The DRS spectra of as synthesized of Graphene Oxide and the band gap calculated and the band gap is zero, due to its revealed Graphene Oxide as a non semiconductor, its like metal so Graphene Oxide as a semi metal.

Band gap energy (E_g) = $h*c/\lambda$ ----- (1)

Where E_g , h , c , λ are band gap energy, plank constant, speed of light, cut of wavelength respectively.

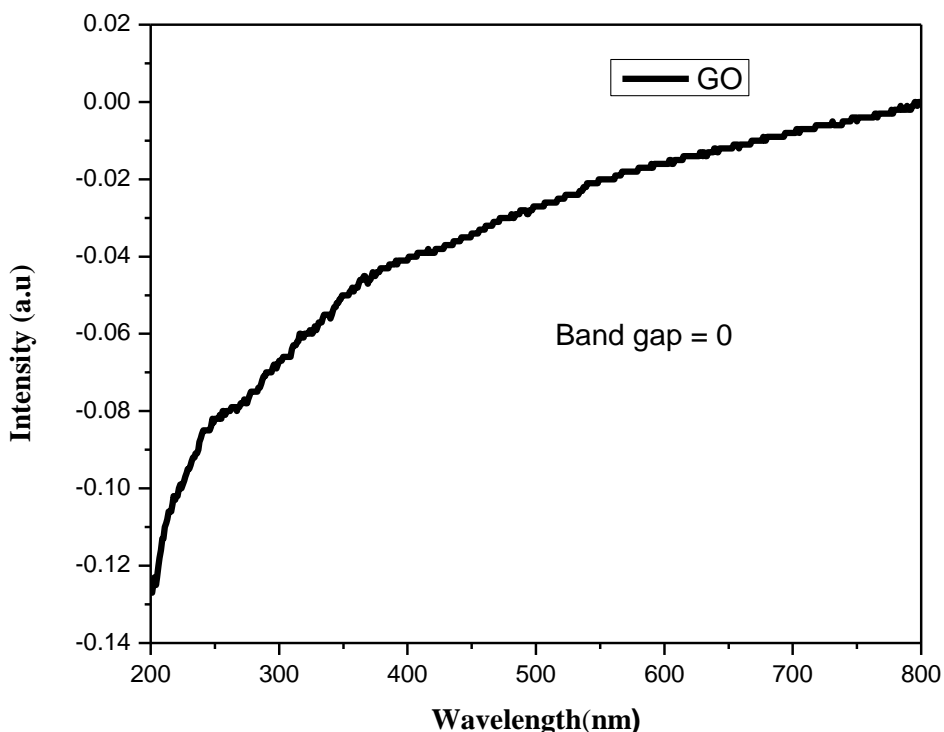


Fig 6:- UV-DRS spectrum of Graphene Oxide

Photocatalytic Effect:-

Generally photocatalytic degradation of dyes by a photocatalyst is critically depends on particle size, crystallinity, morphology, band gap energy, charge recombination and efficient charge separation. Graphene Oxide as a good efficiency for charge separation. Fig.7. shows the photocatalytic activity under visible light irradiation over degradation of Methylene blue were chosen as representative model pollutant. The establishment of adsorption – desorption equilibrium was obtained under continuous stirring 30 min before the degradation reaction was carried out, evaluated degradation under visible light irradiation measured to UV- Visible spectrometer to the cut off filter 664nm. Fig. 7a. shows 95.69 % degradation of MB with GO, Fig.7b shows 58.02 % degradation of MB with Graphite Flake and Fig.7c shows the 7.6 % degradation of MB in absence of catalyst within 120 min. Fig 7d shows a plot between time verses % of degradation of MB under GO, Graphite flake and in absence of catalyst. Among them GO responds more towards the degradation of MB under visible light irradiation and the GO curve indicate with increase the time degradation also increased, the increases of dye degradation below 30 minutes is rapidly after 30 minutes the degradation of dye is slow due to Graphene Oxide has large surface area so the degradation of starting 30 minutes is rapidly. So the effect of dye degradation not only depend on photo light (visible or ultra violet) also depend on surface area of catalyst.

$$\% \text{ of degradation} = (A_0 - A_t) / A_0 * 100 \text{ ----- (2)}$$

Where A_0 and A_t are respectively initial absorbance.

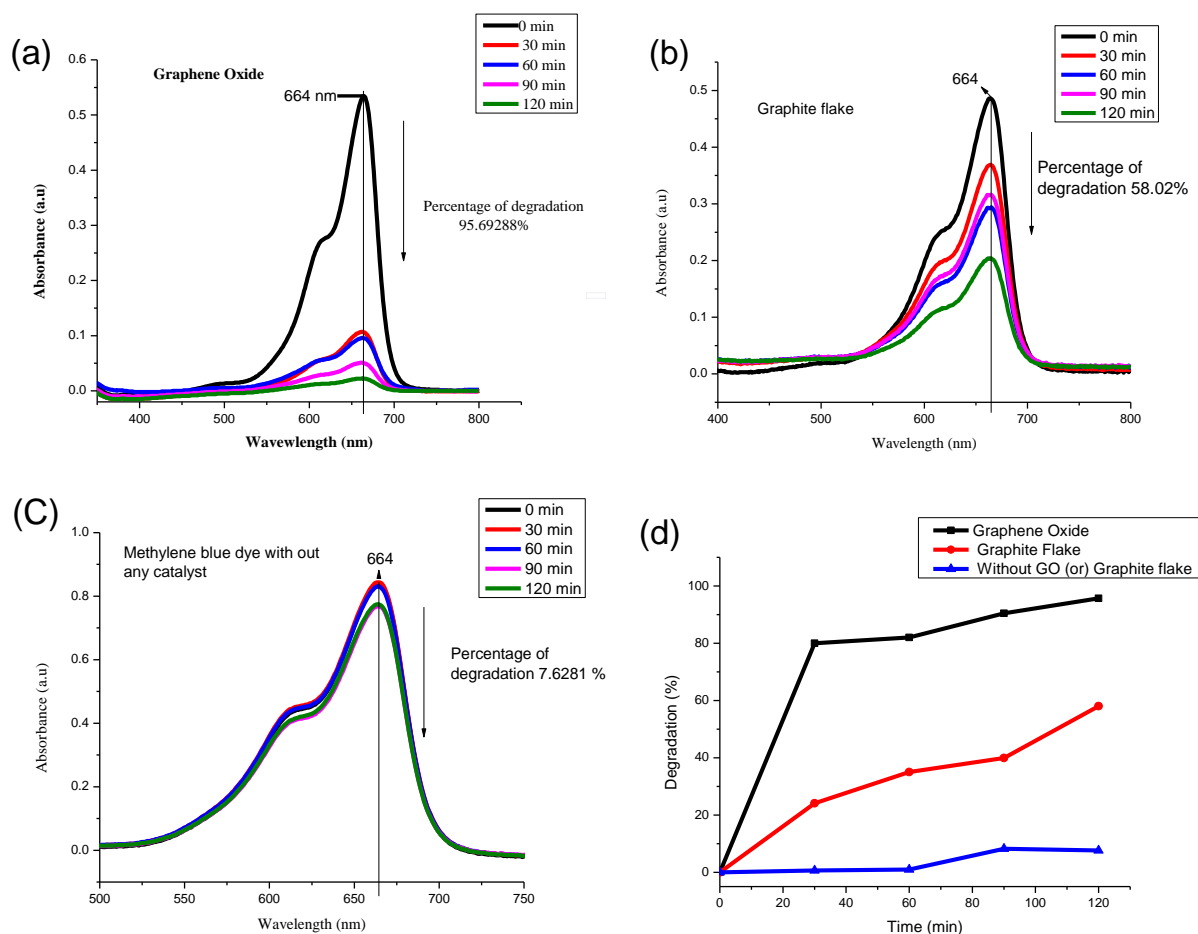


Fig.7:- Degradation of methylene blue dye under Visible light irradiation (a) MB dye with GO (b) MB dye with Graphite flake (c) MB dye without catalyst (d) Photocatalytic comparison between GO, Graphite flake and without catalyst under visible light irradiation as a function of time 't' verses % of degradation.

Conclusion:-

The synthesis of Graphene Oxide successfully synthesized by modified hummer's method. The synthesized of Graphene Oxide confirmed by XRD for the graphite flake disappearance of the peak at 26° and appearance of the peak at 10° , shows that the product is completely oxidized after the chemical oxidation to formation of Graphene Oxide and presence of C-O($n - \pi^*$ transition), C=C($\pi - \pi^*$ transition) bonds analysis by UV-Visible spectroscopy, FE-SEM image revealed sponge porous with large surface area and photocatalytic experiment gave good information for degradation of methylene blue dye with Graphene Oxide as a catalyst i.e. the effect of dye degradation not only depend on photo light (visible or ultra violet) also depend on surface area also due to this reason Graphene Oxide as only one best material for composite/hybridized to semiconductors and metals.

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RESEARCH ARTICLE

ANALYSIS OF PHYSICOCHEMICAL QUALITY PARAMETERS OF GROUND WATER NEAR MUNICIPAL SOLID WASTE DUMPING SITES AT PIPARIYA.

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Abstract

Ground water contamination is generally irreversible i.e., once it is contaminated it is difficult to restore the original quality of water. The effect of municipal solid waste dump on ground water quality at Pipariya, Madhya Pradesh was investigated. Physicochemical quality parameters such as temperature, total dissolved solids, pH, electrical conductivity, alkalinity, total hardness, phosphate, chloride, residual chlorine, were analyzed in groundwater samples. Water samples were collected from many sample stations (hand pumps and bore-wells) from different locations near to a municipal solid waste dump site. The present study is an attempt to observe the existence of pollution in ground water quality and its ill effects on human beings who settled near the dump site at Pipariya Municipal Council (PMC). During the study it was found that total dissolved solids (TDS), electrical conductivity (EC), exceeded the World Health Organization (WHO) tolerance levels for drinking water.

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Introduction:-

Ground water is the principal source of drinking water in both urban and rural areas. The importance of ground water for the existence of human society cannot be overemphasized¹⁻³. The quality of ground water depends on various chemical constituents and their concentration, which are mostly derived from the geological data of the particular region. Landfills used for *dumping* refuses and *municipal wastes*, contribute ground water pollution⁴⁻⁷. Water pollution refers to any type of aquatic contamination rendering the water body poisoned by toxic chemicals which affect living organisms. The contamination occurs through leakage, which is formed when rain water infiltrates the landfill and dissolves the solute fraction of the waste and the soluble product formed as a result of the chemical and biochemical processes occurring within the decaying wastes⁸⁻¹².

Pipariya is a Municipality city in district of Hoshangabad, Madhya Pradesh. Due to the uncontrolled and unscientific dumping of municipal solid wastes, all the water bodies near to the dumping sites at Pipariya gets polluted with toxic materials. The high rate of exploitation of ground water than its recharging, inappropriate dumping of solid and liquid wastes are the main causes of deterioration of ground water quality. Therefore the present study deals with assessment of ground water (drinking water) quality near municipal solid waste dumping sites in Pipariya.

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Materials and Methods:-

Study area:-

The study area selected was the urban area of the city. The present solid waste disposal site of PMC is located at Sandia Road which the extreme north-east area of Pipariya. Municipal wastes dumped on this site are largely from domestic and commercial sources. No scientific method of waste disposal is adopted.

Sampling:-

Selection of the sampling sites was done on the basis of the overall objective of the present study. In all total five sampling sites were selected within 0.50 km radius of the landfill site from where the groundwater samples were taken to assess the status of ground water quality at Pipariya. Water samples were collected in clean 500 ml plastic bottles after the extraction of water either from a bore-well or a hand pump. All the collected samples were immediately transported to the laboratory, stored at 4°C and analyzed the same day.

Physicochemical Analysis:-

It is very essential and important to test the water before it is used for drinking, domestic, agricultural or industrial purpose. All the samples were analyzed for relevant physicochemical parameters according to internationally accepted procedures and standard methods¹³. The samples were analyzed for following physicochemical parameter, temperature, electrical conductivity, turbidity, pH total hardness, total dissolved solids, dissolved oxygen, total alkalinity, chloride, calcium, magnesium, iron, and sulphate (mg/l) etc. All The reagents used for the analysis were AR grade and double distilled water was used for preparation of solutions.

Results and Discussion:-

The results of the physicochemical analysis of the groundwater samples are presented in Table-1. Temperature of the Ground Water sample was recorded as to be normal as per the desirable range. Temperature of groundwater sample slightly varied ranged from 26.0 to 27.5

Table 1:- Physicochemical Parameters.

SN	Parameters	Sample Sites									
		Site -1		Site -		Site -3		Site -4		Site -5	
		I	II	III	IV	V	VI	VII	VIII	IX	X
1	Temperature	26.2	26.0	27.1	26.6	26.1	26.8	27.5	27.2	27.0	26.9
2	pH	7.82	7.79	7.91	7.84	7.93	7.85	7.78	7.93	7.83	7.87
3	Turbidity	5.3	6.1	5.5	5.6	5.4	5.7	5.5	5.8	6.0	5.6
4	EC	885	910	883	890	892	889	908	904	894	888
5	TDS	901	888	898	891	903	887	890	900	907	904
6	TA	138	140	138	138	142	139	136	134	140	135
7	Cl ⁻	245	253	250	249	251	248	246	252	244	251
8	TH	383	375	378	380	384	386	379	391	389	394
9	DO	2.6	2.8	2.4	2.6	3.0	2.9	2.5	2.7	2.8	2.8
10	NO ₃ ⁻	4.1	3.9	4.0	3.8	3.7	3.9	3.6	4.0	3.8	3.5
11	F ⁻	0.11	0.09	0.12	0.10	0.08	0.11	0.10	0.09	0.10	0.9
12	SO ₄ ²⁻	112	109	129	118	127	122	116	119	99	108
13	PO ₄ ³⁻	10.1	9.9	9.6	10.0	9.7	10.2	9.8	10.1	9.5	9.8
13	Ca ²⁺	150	147	153	148	152	154	146	152	151	153
14	Mg ²⁺	143	147	150	144	145	149	146	151	154	147
15	Fe ⁺⁺	0.16	0.14	0.12	0.13	0.17	0.20	0.18	0.14	0.16	0.15

The pH of the groundwater samples is useful for the calculation of acidity and alkalinity, and important factor in maintaining the carbonate and bicarbonate system. The pH of the groundwater samples were from 7.79 to 7.93. The pH value of all the samples shows the water is neural in nature. The values of pH are within desirable limits (6.5-8.5).

Turbidity of groundwater samples obtained from 5.1 to 6.1NTU. EC is a measure of total salt content in water¹⁴. EC ranged between 885µs/cm to 910 µs/cm. TDS indicates the general nature of water quality or salinity¹⁵. During the study TDS is found between ranged 887 mg/l to 907 mg/l. The TDS concentration was found to be above the

permissible limit may be due to the leaching of various pollutants into the ground water which can decrease the potability and may cause gastro-intestinal irritation in human.

The total alkalinity was found to be in the range of 134 to 142 mg/l in ground water samples which are caused mainly due to OH^- , CO_3^{2-} , HCO_3^- ions¹⁶.

The total hardness of ground water samples were found in the range of 375 to 394 mg/l which is further compared with the standard value ranged 300 mg/l. Water hardness is usually due to the multivalent metal ions, which comes from minerals dissolved in the water. The hardness of water is not a pollution indicator parameter but it indicates water quality mainly in terms of calcium and magnesium. The desirable limit for hardness is 300 mg/l and the permissible limit in the absence of alternate source is 600 mg/l.

The value of chloride obtained 244 to 253 mg/l as presented in table which is further compared with the standard values 250 mg/l. Chloride is not harmful to human at low concentration but could alter the taste of water at concentration above 250 mg/l.

DO of ground water samples were found in the range of 2.4 to 3.0 mg/l. due to the capacity of water to hold oxygen¹⁷. The concentration of nitrate was found in water sample up to 4.1 mg/l. have also reported increase in nitrate concentration in ground water due to waste water dumped at the disposal site and likely indicate the impact of leachate.

The concentration of fluoride in the studied water samples ranged from 0.08 to 0.12 mg/l. The concentration of fluoride at low concentration in ground water has been considered beneficial but high concentration may causes dental fluorosis (tooth mottling) and more seriously skeletal fluorosis¹⁸.

Concentration of sulphate in water sample ranged from 99 mg/l to 129 mg/l. Sulphate is a non toxic anion but ailment like catharsis, dehydration and gastrointestinal irritation have been linked with it when concentration is high¹⁻²⁰.

Phosphate may occur in surface and ground water as a result of domestic sewage, detergents, and agricultural effluents. The phosphate content in the study area was found in all sites from 9.5 to 10.2 mg/l.

Calcium and Magnesium are measured by complexometric titration with standard solution of EDTA. The values of Calcium in water samples were found in the range of 146 to 153 mg/l. and Magnesium ranged from 143 to 151 mg/l.

Iron occurs in waters in varying degrees depending on the geology of the area. The iron content of water samples ranged between 0.12 and 0.2 mg/L.

Conclusion:-

The study assessed the evolution of water quality in ground water of Pipariya near MSW. On the basis of current investigation we can conclude that the ground water near the MSW dumping areas most of the under permissible limit of WHO but some of the achieving near permissible limit. A comparative study of ground water i.e. bore well and hand pump water carried out by taking certain important parameters like pH, TDS, TA, Nitrate, Cl^- , PO_4^{3-} , F^- etc. In this present investigation it was found that the maximum parameters were not at the level of pollution except few parameters like nitrate, TDS and TH in ground water.

Therefore, the best accepted option is to avoid the possibility of polluting the ground water resources and municipal solid wastes dump in controlled and scientific way at dumping sites.

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RESEARCH ARTICLE

IMMEDIATE REATTACHMENT OF FRACTURED TOOTH FRAGMENT USING PRE FRABRICATED GLASS FIBER POST IN A MAXILLARY CENTRAL INCISOR - A CASE REPORT.

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Crown fractures, fiber
Post, reattachment

Abstract

Anterior crown fractures are a common form of injury that mainly affects children and adolescents due to their position in the oral cavity especially in the maxillary arch. Reattachment of original tooth fragment to the fractured tooth forms a relatively quick, biologic and esthetic restoration. 32 years old male patient reported to the Department of conservative Dentistry & Endodontics with a crown fracture of maxillary left central incisor and mid root fracture of maxillary left lateral incisor one day after the trauma had occurred. This case report represent the maxillary left central incisor treated with reattachment of natural crown fragment using glass fiber post and dual core composite resin and in maxillary left lateral incisor treated with endodontic therapy.

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Introduction:-

Dental fractures account for 26-76% of dental injuries in the permanent dentition. Coronal fractures of permanent incisors represent 18-22% of all trauma to dental hard tissues, 28-44% being simple (enamel and dentin) and 11-15% complex (enamel, dentin, and pulp). Traumatized anterior teeth require quick functional and esthetic repair. This is particularly true in case of young patients as it not only causes physiologic impairment, but also esthetic disfigurement leading to a psychological impact.¹

Primary goal of the treatment of traumatically injured teeth remains esthetic and functional rehabilitation. Several therapeutic procedures are available to restore fractured anterior teeth. Reattachment of the fractured fragment is an excellent biological approach for restoration, when the fragment is available.²

The concept of reattachment began in 1964 when Chosak and Eidelman used a cast post and conventional cement to reattach an anterior crown segment. Recent developments in restorative materials, placement techniques, and adhesive protocols allow reattachment using resin based composites.³ Tennery was the first to use acid etch technique for the reattachment of fractured tooth fragment.

With the recent improvements in resin based restorative materials, tooth colored fiber posts along with resin luting cement are of choice because of several advantages such as esthetics, bonding to tooth structure and low modulus of elasticity similar to that of dentin. In some cases where the fracture extends subgingivally, but is supra alveolar, a full-thickness envelope flap provides the required isolation and visibility.¹

This is a case of a permanent incisors with crown and middle root fracture treated using glass fiber post in left maxillary central incisor and Root canal treatment in maxillary left lateral incisor.

Case Report:-

A 32-year-old male patient reported to the Department of Conservative Dentistry and Endodontics with fractured maxillary anterior tooth. The tooth had fractured 1 day ago on account of a minor two wheeler accident. His medical history was noncontributory. Patient complained of severe pain and swelling with 21 & 22. On clinical and radiographic examination, it was diagnosed as a complicated crown fracture of the maxillary left central incisor and crown root fracture in maxillary left lateral incisor (Figure-1). The oblique fracture line was supragingival both labially and palatally with 21. The treatment plan decided for the patient was reattachment of the fragment with 21 and root canal treatment in reallion 22.



Figure 1:- Ellis class III fracture with 21 and middle root fracture with 22.

Patient was informed of long term prognosis of the teeth and the treatment was initiated. The teeth were anaesthetized using local infiltration technique (2% lidocaine with 1:100,000). Single visit root canal treatment of 21 and 22 was carried out (figure-2). The post space was prepared by using peeso reamers retaining 5 mm of apical gutta percha and No.2 prefabricated glass fiber post was selected. After checking the fit of the post, it was trimmed leaving about 2 mm of post beyond the tooth with 21. The pulpal aspect of the fractured fragment was prepared using a straight fissure bur (internal groove). The post and the fragment were adjusted until the fragment was approximated to its original position to obtain retention for the fragment. The post was luted using dual core resin luting cement (RelyX™ U200, 3M ESPE, Germany) with 21 [Figure-3].

Both the fragment and tooth were etched with 37% phosphoric acid etchant gel for 15 seconds. Then the bonding agent (3M ESPE, USA) was applied and light cured for 10 seconds. Flowable composite resin (Filtek Z 350XT flowable restorative, 3M ESPE, USA) was applied to both fragment and tooth surfaces. The fractured segment was then accurately attach on the tooth, paying special attention to the fit between the segments. When the original position had been reestablished, excess resin was removed and the bonded area was light cured for 40 seconds on each surface.

The occlusion was carefully checked and adjusted. The margins were properly finished with finishing burs and polished with polishing paste. Final radiograph was taken to confirm the proper alignment of the attached fragment. The patient was given oral hygiene instructions and was recalled for follow-up visits. The six month follow up (Figure-4) shows satisfactory result esthetically and functionally.

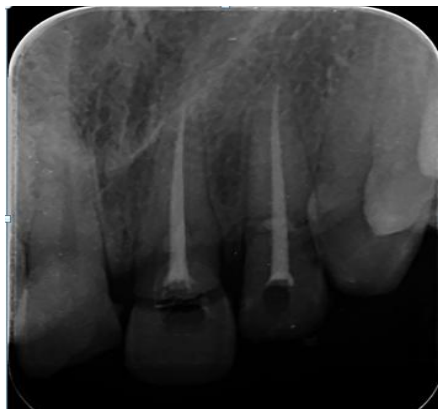


Figure 2:- Root canal treatment was carried out with 21 and 22.



Figure 3:- Reattachment with help of glass fiber post i.r.t.21 and MTA Fillapex i.r.t. 22.



Follow up after 6 months

Discussion:-

Traumatic dental injuries in the permanent dentition necessarily involve function, aesthetics and psychological aspects particularly in young patients. Factors determining the treatment plan and prognosis for coronal teeth fractures include the extent of fracture, endodontic involvement, periodontal status, maturity of root, biological width invasion, presence / absence of fractured tooth fragment and the fit of the fragment and remaining tooth, occlusion, esthetics and finances. Depending on these factors, various treatment options are composite restorations, orthodontic / surgical extrusion, crown lengthening followed by post and core supported restorations and reattachment of fractured fragment.^{2,4}

Conventional methods employed in the restoration of fractured teeth include partial and full coverage crowns, laminate veneers, and composite resins all of which are time consuming, high priced, and not conservative.⁴ It is beneficial to quickly restore the function and the aesthetics of a fractured tooth. The reattachment of a fractured crown fragment may be the most conservative and desirable treatment of choice for anterior teeth. The reattachment of the crown fragment to a fractured tooth is the best method to reinstate the natural shape, contour, surface texture, occlusal alignment and color of the fragment, which offers excellent esthetic and functional result.⁵

Post and core supported crowns are recommended in cases of major tooth loss where the fractured fragment is not available. With the fracture line extending below the alveolar crestal bone orthodontic extrusion or surgical extrusion is recommended before the restoration. But with the fracture lying above the alveolar bone crest reattachment of the fractured fragment is a more viable option. There is sufficient evidence to show that the reattachment of the fractured coronal fragments using modern adhesive systems results in predictably successful short- and medium-term outcomes.¹

However, this technique can be used only when the intact tooth fragment is available. As the fractured fragments were intact in the cases described above, use of natural tooth substance clearly eliminated problems of differential wear of restorative material unmatched shades and difficulty of contour and texture reproduction associated with other restorative techniques.⁷

The common complications in the post and core system are debonding and root fracture. These are more common with cast metal posts used earlier which can cause wedging forces coronally that may result in irreversible failure because of fracture of an already weakened root. The newer variety of nonmetallic posts is made of either ceramic or fiber reinforced materials like carbon, quartz or glass in an epoxy matrix. Fiber reinforced composite resin post offers many advantages for reattachment procedure such as conservation of tooth structure, bonding to the tooth structure, low modulus of elasticity equal to that of dentin, esthetics, simplicity of procedure requiring less chair time and cost-effectiveness.⁸

By using glass fiber post with composite core and with recent advances in adhesive techniques and materials one can create a Monoblock, a multi-layered structure with no inherent weak interlayer interfaces. Early retrospective studies indicate that the clinical performance of fiber post is promising and the failure rate recorded is 3.2% over a period of up to 4 years. Trope *et al.* in 1985 showed that Endodontically treated teeth can be reinforced with the use of resin composite restoration. The flowable composite reinforces the tooth, helps in achieving higher bond strengths and minimizes the inclusion of air voids.¹

Tooth fracture reattachment allows restoration of the tooth with minimal sacrifice of the remaining tooth structure. The use of natural tooth substance clearly eliminated problems of differential wear of restorative material, unmatched shades and difficulty of contour and texture reproduction associated with other restorative techniques.²

Conclusion:-

Reattachment technique is the most conservative and biological method of restoring a fractured anterior tooth. Reattaching a tooth fragment with newer adhesive materials may be used successfully to restore fractured teeth with adequate strength, but long term follow up is necessary in order to predict the durability of the tooth-adhesive-fragment complex. Patient cooperation and understanding of the limitations of the treatment is of utmost importance for good prognosis. The need of the day is to educate the population to preserve fractured segment and seek immediate dental treatment.

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RESEARCH ARTICLE

DENTO-ALVEOLAR EFFECTS OF MEMORAX VERSUS HYRAX RAPID PALATAL EXPANDERS: A COMPARATIVE CBCT STUDY.

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Abstract

Background: Rapid maxillary expansion usually associated with dental tipping of the anchor teeth, but the aims of the treatment are always to achieve minimal dental and maximum skeletal effects. The objective of the present study was to evaluate and compare the effect of memory palatal split screw (Memorax) to that of conventional Hyrax rapid palatal expanders on dento-alveolar structures, which were measured and recorded by Cone beam computed tomography (CBCT).

Materials and Methods: It was conducted over 14 subjects of adolescent patients with bilaterally constricted maxillary arch, with age ranged from 12ys to 15ys, with a mean of 13.6 ± 1.4 divided equally into two groups; the Memorax group and the Hyrax group. For all subjects, maxillofacial CBCTs were taken before expansion (T_1) and after 3 months at the time of removal of the expanders (T_2). 10 dental measurements were taken (5 linear and 5 angular). Data were explored for normality using Kolmogorov-Smirnov test of normality. Paired t test was used to compare mean values of before and after treatment. Independent (unpaired) test was used to compare between the two groups.

Results: There was no significant difference in the percent change after using both expanders, except in the inter-premolar apex width, where the percent change was significantly greater in the Memorax group ($p=0.0082$), and in the inter-premolar angle, where the percent change was significantly greater in the Hyrax group ($p=0.0096$).

Conclusion: In the premolar area, Memorax produced bodily movement expansion pattern, while Hyrax showed more tipping of the anchor teeth.

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Introduction:-

Interest in rapid maxillary expansion (RME) has increased during the past four decades. In spite of the recommended use of the RME for different aims, its fundamental objective is to increase the transverse width of the upper dental arch at the apical base (Sarver and Johnston, 1989). Although the major treatment aim of RME could be observed in the transversal expansion of the skeletal facial structures, dento-alveolar changes may be considered as an additional contribution or side effect.

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Usually, appliances with fixed jack screw are used for RME and they produce heavy forces which when applied to the maxillary anchor teeth and alveolar structures exceed the limits required for orthodontic movement (Isaacson and Ingram, 1964, and Zimring and Isaacson, 1965). Pressure caused by the applied forces compresses the periodontal ligament of the anchorage teeth and subsequently produces orthopedic movement by opening the mid-palatal suture (Haas A J, 1961) and orthodontic movement by tipping the upper posterior teeth and bending the alveolar bone (Da Silva Filho et al, 1991, Ciambotti et al, 2001, Oliveira et al, 2004, Kılıç et al, 2008, Braun et al, 2000, Asanza et al, 1997 and Garrett et al, 2008) with both slow maxillary expansion (Hicks, 1978) as well as RME. Hence, the aim of the RME treatment is to achieve minimal dental and maximum skeletal effects (Haas, 1961). Many different RME appliances have been developed for this purpose.

A study (Wichelhaus A, Geserick M and Ball J, 2004) described and evaluated a new maxillary memory palatal split screw (Memorax) that includes nickel–titanium open coil springs in the screw bed in order to lessen the massive expansion forces. The screw could be activated six times a day to produce a constant force level of 12-14 N (1224-1428 grams) providing effective and rapid expansion. According to the authors, this new screw could produce rapid, constant, and physiological expansion forces, thus making the expansion procedure more effective, more physiological, and well tolerated by patients. However, another study (Halicioğlu et al, 2010) showed that memory palatal split screw produced significant increase in the inter-premolar and intermolar distances and reduced the nasal airway resistance.

In other studies, the authors investigated effects of conventional Hyrax screw (Timms D J, 1981) and compared it with the memory screw on skeletal and dento-alveolar structures (Halicioğlu and Yavuz, 2011). They used plaster models and postero-anterior cephalograms at pretreatment (T_1), after expansion (T_2), and after retention (T_3). They found that the use of memory screw may be advantageous because it shortens the maxillary expansion period, provides additional expansion in the retention period, and generates light forces relative to the conventional Hyrax screw. Although studies about memory screw have increased, further investigations are still needed to compare the effects of RME caused by memory screw and Hyrax screw on the dento-facial structures. The aim of the present study is to investigate and compare the transversal dento-alveolar changes in subjects treated with a Memorax screw and conventional Hyrax screw using CBCT.

Methods:-

The material of this study consisted of CBCT radiographs of 14 patients, with bilateral maxillary cross-bite caused by basal apical narrowness, who were planned for RME at the Department of Orthodontics, Faculty of Dental Medicine for Girls, Al- Azhar University, Cairo, Egypt.

The research proposal was approved by the research ethics committee (REC), Faculty of Dental Medicine for Girls, Al- Azhar University, Cairo, Egypt. The detailed procedure was discussed with the patients and their guardians who signed an informed consent before the start of the clinical work.

The subjects were randomly divided into two equal groups: memory-screw group (Memorax group) treated using Memorax RME appliance (Product number: 167M1529—Memory expander type 'N'; Forestadent, Pforzheim, Germany). This group comprised 7 subjects (6 females and 1 male) with ages ranged from 13.6 ± 1.4 years, and Hyrax-screw group treated using Hyrax RME appliance (Product Type: 602-808-10 Hyrax® Medium, Dentaureum, Ispringen, Germany). This group also comprised 7 subjects (6 females and 1 male) with ages ranged from 13.5 ± 0.4 years. Maxillary first premolars and first molars were banded and four-armed expansion screws were soldered to the bands in both groups. In the conventional Hyrax-screw group, the patients were instructed to activate the screw two quarter-turns a day ($0.2 \times 2 = 0.4$ mm) one in the morning and one in the evening, while in the Memorax group, the patients were instructed to activate the screw six quarter-turns a day ($0.2 \times 6 = 1.2$ mm); two in the morning, two after lunch, and two in the evening. Both screws were activated until the occlusal aspect of the maxillary lingual cusp of the upper first molars contacted the occlusal aspect of the facial cusp of the mandibular first molars, thus producing the desiring expansion. The mean expansion period was 7 days in the memory-screw group and 22 days in the Hyrax-screw group with average total screw quart-turns in both group was 42 (8.4 mm) and 44 (8.8mm) respectively. Then, patients were left with the appliances as retainers for 3 months with the fixation of the screw of the hyrax group according to manufacturer instructions (Fig 1).

CBCT imaging: the RME achieved in this study was evaluated using Next Generation Scanora 3D CBCT scanner (Soredex, Tuusula, Finland). Images were taken prior to treatment (T_1), and 3 months after removing the appliance

(T2). The machine was set to the following protocol for all the scans of the study: 90kVp, 4mA, Voxel size 0.35mm, Scanning time was 45 seconds, with Field of view 13 x 13 cm.

After acquisition, data were exported and transferred in DICOM format to be secondarily re-evaluated for linear and angular measurement, where Invivo Dental software (version 5.3; Anatomage, San Jose, California, USA) was used for that purpose.

Invivo software was employed to superimpose preoperative and postoperative scans. The set of DICOM data of the preoperative scan was loaded into the software, and then the set of the postoperative scan of the same patient was loaded over it at the superimposition module. According to variation in positioning of both scans, a second adjustment was needed to ensure perfect superimposition, hence guaranteeing measuring linear and angular measurements at the exact level. Superimposition module was used to superimpose the postoperative scan over the preoperative one, where four landmarks, at different planes, were chosen at each scan, and then registration of these landmarks was automatically performed by the software. Superimposition sequence was repeated for each patient individually (**Fig 2**).

After completion of superimposition, the two scans (preoperative and postoperative) were regarded as a one unit and moved in the same manner. Orientation of the whole volume was made to ensure that the orthogonal reference lines (axial, coronal and sagittal) were following certain planes. Axial line was made parallel to maxillary plane (ANS-PNS) at sagittal view and parallel to the nasal floor at the coronal view respectively, and the sagittal line was parallel to the mid-sagittal plane at the coronal view (**Fig 3**). Ten dental measurements were recorded; five linear and five angular measurements (**Table 1**). For linear measurements, coronal view was utilized, where coronal reference line was moved to intersect at the palatal roots of the upper first molars at both sides. At the produced view, maxillary base width and inter-molar width (between apices of palatal roots and mesio-buccal cusp tips bilaterally) were measured at pre-operative scan then it was hidden then the same measurements were taken at the same view for the post-operative scan (**Fig 4**). At the same view, angulation of right and left first molars were measured in the same manner, and also the inter-molar angle as well. Molar angulation was measured by drawing the angle connecting the mesio-buccal cusp tip to the palatal root tip intersecting with the axial line which was made parallel to the nasal fossa for this measurement (**Fig 5**). Same measurements were repeated at the first premolars level after moving the coronal view to that level.



Figure 1:- Memorex rapid palatal expander during active expansion period.

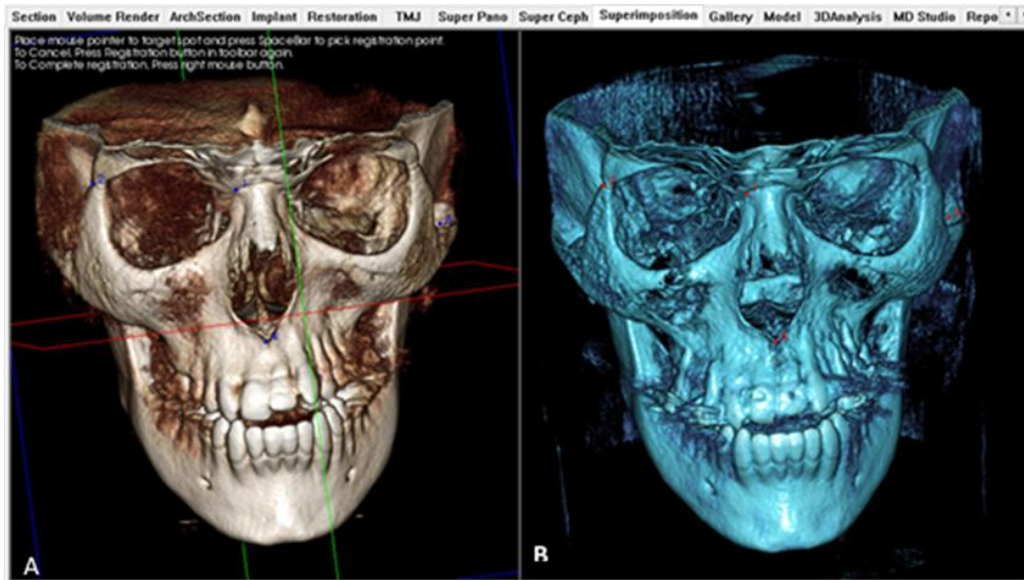


Figure 2: four landmarks, at different planes, numbered (1,2,3,4) were chosen at each scan (A: Blue colored numbers in the Preoperative scan), and then registration of these landmarks was automatically performed by the software (B: Red colored numbers in the postoperative scan).

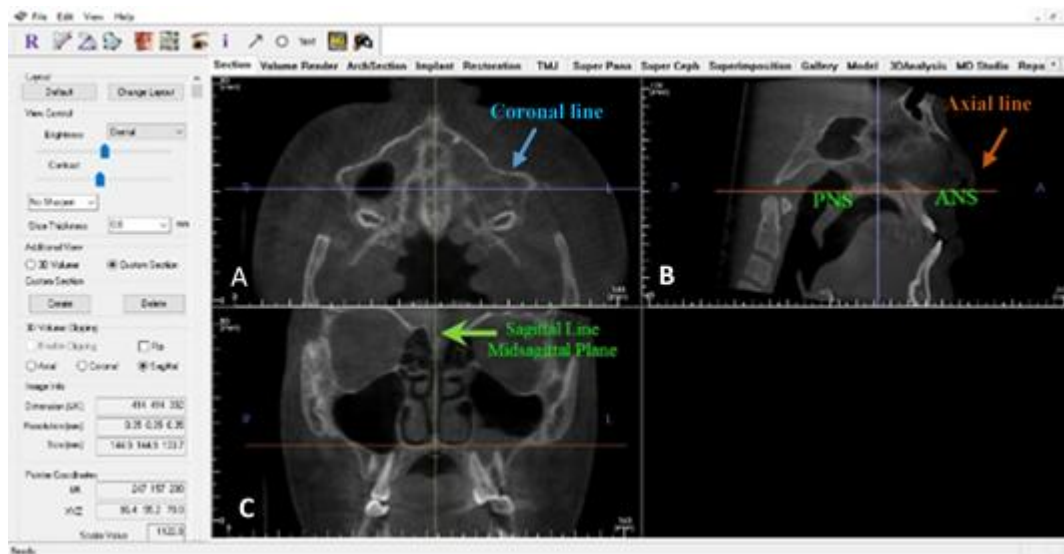


Figure 3:- orthogonal reference lines. (A: coronal line, B: Axial line, and C: sagittal line)

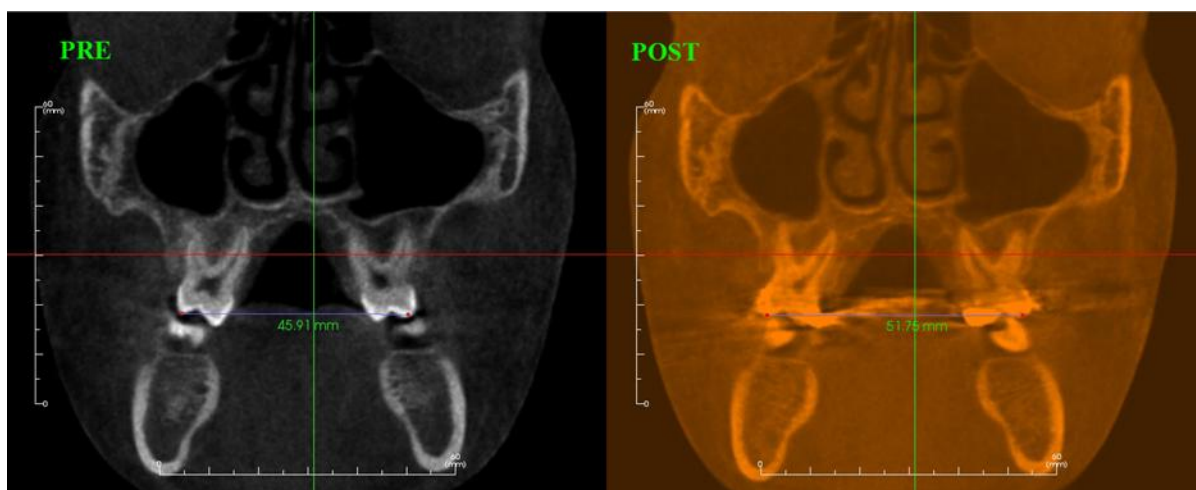


Figure 4:- coronal reference line was moved to intersect at mesio-buccal cusp tips bilaterally to measure the inter-molar width in the pre- and post-operative scans.

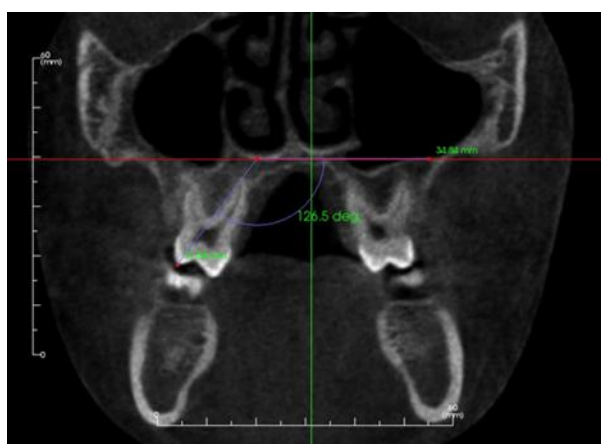


Figure 5: Molar angulation measurements.

Results:-

Values of all parameters were presented as mean and standard deviation (SD) values. Data were explored for normality using Kolmogorov-Smirnov test of normality. The results of Kolmogorov-Smirnov test indicated that most of data were normally distributed (parametric data), so parametric tests were used for the comparisons. Paired t-test was used to compare mean values of before and after treatment. Independent (unpaired) t-test was used to compare between the two groups. The percentage change after treatment was calculated as follows:

(after-treatment value – before-treatment value) X100/ before-treatment value.

The significance level was set at $p \leq 0.05$. Statistical analysis was performed with SPSS for Windows v.16.0 (SPSS Inc., USA).

Both linear (mm) and angular (degree) values increased after treatment in the Memorex group. Paired t test revealed that the difference was statistically significant for all measurements, except for the right premolar angle degree, where the increase after treatment didn't reach the level of statistical significance ($p=0.0682$) (**Table 1**). Both linear and angular values increased after treatment in the Hyrax group. Paired t test revealed that the difference was statistically significant for all measurements (**Table 2**). Unpaired t-test revealed no significant difference in the mean pretreatment values of both groups. Moreover, there was no significant difference in the mean post-treatment values of both groups. Unpaired t-test revealed no significant difference in the percent change after using both expanders, except in the inter-premolar apex width, where the percent change was significantly greater in the

Memorax group ($p=0.0082$), and in the inter-premolar angle, where the percent change was significantly greater in the Hyrax group ($p=0.0096$) (Table 3).

Table 1:- Pre-treatment (T_1) and post-treatment (T_2) values, percent change and significance of the difference using paired t-test in the Memorax group

Memorax	T_1			T_2			Percent change			t value	P value
	Mean	SD	SE	Mean	SD	SE	Mean	SD	SE		
Maxillary base width	61.41	4.74	1.69	63.20	4.80	1.81	2.93	2.22	0.84	3.44	0.0137*
Inter-molar crown width	48.81	2.88	1.09	54.86	2.59	0.98	12.48	3.03	1.14	12.44	<0.0001*
Inter-molar apex width	26.93	4.62	1.74	29.19	3.89	1.47	9.05	5.54	2.09	5.078	0.0022*
Right molar angulation	119.17	7.51	2.83	125.86	5.11	1.93	5.76	3.18	1.2	5.165	0.0020*
Left molar angulation	120.11	6.41	2.42	124.57	7.45	2.81	3.71	3.12	1.18	3.189	0.0188*
Inter-premolar root angle	58.57	13.55	5.11	60.94	12.48	4.71	13.06	4.68	5.54	4.735	0.0032*
Inter-premolar crown width	36.96	6.62	2.5	44.77	1.58	0.6	21.21	2.80	1.06	24.229	<0.0001*
Inter-premolar apex width	24.94	6.31	2.38	28.86	6.04	2.28	16.94	9.52	3.59	4.248	0.0053*
Right premolar angle	105.34	12.64	4.77	109.47	9.38	3.54	4.38	5.71	2.15	2.219	0.0682 ^{ns}
Left premolar angle	108.09	6.91	2.61	113.37	5.97	2.25	5.01	3.93	1.48	3.3361	0.0156*

SD: Standard Deviation, SE: Standard Error, ns: non-significant, *: significant at $p<0.05$

Table 2:- Pre-treatment (T_1) and post-treatment (T_2) values, percent change and significance of the difference using paired t test in the Hyrax group

Hyrax	T_1			T_2			Percent change			t value	P value
	Mean	SD	SE	Mean	SD	SE	Mean	SD	SE		
Maxillary base width	58.16	4.46	1.68	60.63	5.30	2	4.19	1.63	0.62	6.1177	0.00087*
Inter-molar crown width	49.69	4.21	1.59	56.73	6.26	2.36	14.14	6.97	2.63	5.2158	0.00198*
Inter-molar apex width	28.31	3.11	1.17	29.90	4.18	1.58	5.33	4.65	1.72	2.896	0.0274*
Right molar angulation	117.93	1.91	0.72	125.69	4.78	1.8	6.55	2.80	1.06	6.0813	0.00089*
Left molar angulation	123.83	1.79	0.68	131.96	8.34	3.15	6.51	5.39	2.03	3.1857	0.0189*
Inter premolar root angle	69.20	5.14	1.94	79.41	8.94	3.37	14.49	4.59	1.73	6.9434	0.0004*
Interpremolar crown width	39.64	6.40	2.42	46.96	5.79	2.18	18.11	7.73	3.02	5.4499	0.00158*
Interpremolar apex width	27.43	7.17	2.71	28.57	7.27	2.74	4.38	4.47	1.69	3.0044	0.02387*
Right premolar angle	103.64	6.54	2.47	115.23	5.87	2.22	11.26	1.57	0.59	32.588	<0.0001*
Left premolar angle	107.53	3.71	1.4	114.76	3.69	1.44	6.84	5.27	1.99	3.566	0.00118*

SD: Standard Deviation SE: Standard Error *significant at $p<0.05$

Table 3:- Comparison of percent change in dento-alveolar effects in both Memorax and Hyrax group

	Percent change [Mean (SD)]		t value	P value
	Memorax	Hyrax		
Maxillary base width	2.93 (2.22)	4.19 (1.63)	1.2104	0.249 ^{ns}
Inter-molar crown width	12.48 (3.03)	14.14 (6.97)	0.5779	0.574 ^{ns}
Inter-molar apex width	9.05 (5.54)	5.33 (4.65)	1.3608	0.198 ^{ns}
Right molar angulation	5.76 (3.18)	6.55 (2.80)	0.4933	0.603 ^{ns}
Left molar angulation	3.71 (3.12)	6.51 (5.39)	1.1895	0.2572 ^{ns}
Inter premolar root angle	13.06 (4.68)	14.49 (5.59)	1.4742	0.1662 ^{ns}
Interpremolar crown width	21.21 (2.80)	18.11 (7.73)	0.9976	0.3382 ^{ns}
Interpremolar apex width	16.94 (9.52)	4.38 (4.47)	3.1596	0.0082*
Right premolar angle	4.38 (5.71)	11.26 (1.57)	3.0738	0.0096*
Left premolar angle	5.01 (3.93)	6.84 (5.27)	0.7365	0.4756 ^{ns}

(SD): (Standard Deviation), ns: non-significant, *: significant at $p < 0.05$

Discussion:-

Generally, RME screw is turned twice a day during expansion. Isaacson and Ingram (1964) and Zimring and Isaacson (1965) suggested turning the screw twice until the suture opens in adolescent people and recommended that slower rates of expansion would allow for physiologic adjustment at the maxillary articulations and prevent residual force. In this study, the Hyrax screw was activated twice daily even after the suture has opened to stabilize the force applied to the appliance from the start till the end of expansion period. While in the Memorax group, screw was activated six times a day during RME. The difference between the screw activation protocols results from the different levels of forces produced by screws. Isaacson and Ingram (1964) measured the forces produced by conventional RME appliance and found that a quarter turns of the screw produced heavy and intermittent forces ranging from 1.5 to 4 kg, and may accumulate up to approximately 10 kg. However, Wichelhaus et al. (2004) evaluated that memory screw activated six times a day could generate a continuous force ranging from 1.225 to 1.425 kg. Additionally, Zimring and Isaacson (1965) hypothesized that if the expansion procedure was carried out at lower force level in a shorter treatment time, the total expansion might be physiologically stable. In the current study, adequate maxillary expansion was accomplished with the Memorax screw in 7 days with relatively lower forces than that produced by conventional expanders. However, the results of the present study shared to support the hypotheses of Zimring and Isaacson (1965) and Isaacson and Ingram (1964).

Both linear and angular values increased after treatment with both Memorax and Hyrax. Paired t-test revealed that the difference was statistically significant for all measurements, except for the right premolar angle degree in the Memorax group, where the increase after treatment did not reach the level of statistical significance. While there was no significant difference in the percent change after using both expanders, except in the inter-premolar apex width, where the percent change was significantly greater in the Memorax group ($p=0.0082$) which revealed a bodily movement pattern in the premolars area. In the Hyrax group, the percent change in the inter-premolar angle was significantly greater ($p=0.0096$) than the Memorax group which revealed a more tipping movement in the premolar area. These results could be regarded to the fact that the memory screw provided additional expansion in the retention period, which might have probably resulted from the activation of the nickel–titanium springs. In other words, in the retention period, nickel–titanium springs integrated in the screw might have not only resisted the residual forces that are considered to be the source of relapse, but according to Halıcıoğlu et al (2010), it also produce further expansion during the retention period. It was also observed that expansion with Hyrax led to a more tipping movement in the right rather than the left premolar, which may be attributed to the design of the Hyrax appliance used in the study. Braun et al (2000) demonstrated that the amount of dento-alveolar tipping depends on factors, such as the type of expansion appliance, age of the patient, resistance of the surrounding tissues, and schedule of screw activation. It has been observed that the rigidity of the expansion appliance is one of the factors responsible for decreasing the buccal tipping of the anchorage teeth (Timms, 1981). Although many different RME appliances have been developed to minimize the dento-alveolar inclination (Memikoglu and İşeri, 1999 and Başçiftçi and Karaman, 2002), occurrence of dento-alveolar tipping is unavoidable (Wertz, 1970 and Bishara and Staley, 1978). Asanza et al (1997) and Ciambotti et al (2001) found less molar tipping in patients treated with a Hyrax expander according to their studies. This difference may owe to the different measurement techniques employed for the determination of inclination. While Garrett et al (2008) showed that dento-alveolar inclination lacks significant correlations with the schedule of screw activation but correlate well with the amount of expansion. In the

present study, both the screws produced statistically significant dento-alveolar molar tipping after expansion. According to Hicks (1978) the anchorage teeth are expected to tip during expansion and remain upright at retention. In our study, this occurred in the Hyrax group showing significant increase in dento-alveolar tipping in the anchor premolars (inter premolar angle) ($P=0.0096$). This did not occur in the Memorax group, as an almost bodily movement of the anchorage teeth took place during the expansion period. This was due to the decreased alveolar bending occurred as a skeletal response to the rapid physiological distribution of the force applied by the appliance, which resulted from the nickel–titanium springs in the screw. This produced a significant increase in the inter-premolar apex width ($P=0.0082$). We believe that the continuous force exerted during retention, is the main reason behind preventing molar teeth to remain upright, producing non-significant dentoalveolar tipping in the anchored molars, which was decreased significantly in the premolar area. Hence, Memorax might increase the dento-alveolar tipping during the retention period especially in the molar area which might exceed the planned overexpansion if left more than 3 months. Halicioğlu K and Yavuz (2011) reported similar results when using Memorax for expansion, where the inter-premolar and molar width increased in the retention period. So they recommended before completing the active phase of RME, to take into account that some amount of increments will be observed in intermolar and interpremolar widths during the retention period. So according to previous findings which were emphasized during our study, The Memorax is not recommended to be left more than 3 months retention in the patient mouth.

Conclusions:-

Both appliances produced dento-alveolar tipping in the anchored teeth. But for the first premolars, Memorax produced a bodily movement expansion pattern with less dento-alveolar tipping. On the other hand, Hyrax showed a more tipping movement of the anchor premolars and molars.

Recommendations:-

- The Memorax is not recommended to be left more than 3 months as a retainer in the patient mouth as it increases the dento-alveolar tipping of the anchored teeth in the retention period.
- Further investigations are needed to analyze the forces and stresses produced by the Memorax in both active expansion and retention periods.

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RESEARCH ARTICLE

IN VITRO PROPAGATION OF SPATHOGLOTTIS PLICATA BLUME VIA ASYMBIOTIC SEED GERMINATION.

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In vitro propagation, callus induction, protocorms, regeneration and *S. plicata*

Abstract

The loss of plant genetic resources has necessitated the development of many *ex situ* conservation techniques. The application of plant tissue culture techniques in orchid conservation and propagation requires an efficient *in vitro* regeneration protocol. This study reports the development of such highly efficient protocols for the *in vitro* asymbiotic seed germination of *Spathoglottis plicata*. The seeds were successfully germinated asymbiotically on Gamborg B5 (1968). Various growth regulators such as 2,4-Dichlorophenoxyacetic acid (2,4-D), Indole acetic acid (IAA) Benzyladenine (BA) and Kinetin individually were used for callus induction and multiple shoot initiation from the protocorms. B5 medium supplemented with 2,4-D (9.03 μ M) was suitable for callus induction. Calli developed a route of production of protocorm-like bodies and eventually develop into plantlets on transfer to growth regulator free half strength basal medium. The well rooted plants were hardened successfully in the potting mixture containing coconut husk, sand, charcoal, and brick pieces in the ratio 1:1:1:1.

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Introduction:-

Orchidaceae is a highly successful family, with representatives capable of occupying almost every ecological situation, apart from marine environments and habitats characterized by extreme cold throughout the year. Orchids exhibit an incredible range of diversity in size, shape and colour of their flowers and as a result, these plants have great ornamental value (Kasulo *et al.*, 2009). In India, orchids form 9% of the flora. It is estimated that about 1300 species belonging to 140 genera are present in the Himalayas with others scattered in Eastern and Western Ghats (Jain, 1980). Around 10% (3000) of the world's total orchid species are believed to be endangered in their native habitats. Orchids are subject to high levels of threat, through both natural and anthropogenic causes. The greatest threat to orchid diversity is habitat loss. Clearance of natural vegetation for ranching, monocrop agriculture, mining, logging, burning and urban development, has decimated many orchid species. Habitat destruction triggers the loss of pollinators, other plants and fungi on which the orchids mostly depend for their survival (Hagsater and Dumont, 1996).

Micropropagation has major advantages over conventional methods of plant propagation. It is an invaluable aid in the multiplication of elite clones of recalcitrant species, and is important in terms of multiplying plants throughout the year, with control over most facets of production. It is possible to generate pathogen free plants, even from explants of infected mother plants, plant materials such as male sterile, fertility maintainer and restorer lines can be cloned; and it enables the production of large number of plants in a short time from a selected number of genotypes,

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where the traditional methods of multiplication are either not available or are ineffective in large scale multiplication systems (Debergh and Zimmerman, 1991).

The genus *Spathoglottis* is a member of the subfamily Epidendroideae. All members of this genus are terrestrial and most grow at low moderate altitudes, although a few taxa occur at high altitudes in grasslands and open forests in moist areas (Dressler, 1993; Beltrame, 2006). Whole plants as well as their different parts, viz., roots, rhizomes, pseudobulbs, stems and leaves are used as medicinal products. These are used for treatment of different diseases such as general debility, stomachache, bone fractures, colds, and wound healing, general weakness and to cure various other diseases (Subedi et al., 2013; Pant and Raskoti, 2013). *S. plicata* is a handsome terrestrial orchid species with nodes borne thickened bulb like stem (pseudobulb). The ground orchid *S. plicata* is one of the most commonly cultivated orchids in the Southeast Asia and could be found in gardens throughout the region. It is suitable as a potted orchid because it is very attractive, fast growing, easy to grow and with rapid fruit maturation. At maturity, plants flower almost throughout the year (Kheawwongjun and Thammasiri, 2008).

Materials and Methods:-

Explants source:-

Asymbiotic seed germination:-

The immature capsules of *S. plicata* (naturally pollinated) collected from Kakkavayal, Wayanad, Kerala were used for asymbiotic seed germination. The plant was authenticated and identified by Dr. K. Kishore Kumar, Head, Department of Botany, Farook College, Calicut, Kerala

Callus induction:-

40 days old protocorms (before the emergence of first leaves) of *S. plicata* were used as explants for callus.

Sterilization:-

The freshly collected capsules were surface sterilized in sodium hypochlorite solution (NaClO, 0.6 % w/v) for 2 minutes, rinsed thrice with sterile distilled water, dipped in 70 % (v/v) ethanol for 60 seconds and flamed. Seeds from the surface sterilized capsules were extracted by splitting the capsule longitudinally with a sharp sterilized surgical blade. The seeds were then spread as thin film in the test tube containing 10 ml of solid culture medium.

Culture media and initiation of culture:-

Five different basal media such as Murashige and Skoog (1962), Linsmaier and Skoog (1965), Gamborg B5 (1968), Schenk and Hildebrandt (1972) and Knudson C (1946) were tested to select a suitable medium for asymbiotic seed germination of *S. plicata*. Gamborg B5 with growth regulators such as 2,4-D (2.26, 4.52 and 9.03 μM), IAA (2.9, 5.7 and 11.4 μM) BA (2.22, 4.44 and 8.88 μM) and Kinetin (2.32, 4.64 and 9.29 μM) individually were used to assess the effect on callus induction.

All the media were supplemented with 3 % (w/v) sucrose and solidified with 0.8 % (w/v) agar (Hi media-India). The pH of the media was adjusted to 5.6–5.8 with 1 N NaOH or HCl. Around 10–15 ml of the medium was dispensed into 250 mm \times 150 mm culture tubes (Borosil). The mouth of the tubes was covered with aluminium foil and was autoclaved at 1.06 kg pressure for about 20 minutes at 121°C. The autoclaved medium in the culture tubes was cooled and allowed to solidify as slants. The inoculations were done after four days to ensure that the media were free from contamination. Five replicates were used for each treatment and maintained at $25 \pm 2^\circ\text{C}$ in culture room under a 12 hrs photoperiod of $50 \mu\text{mol m}^{-2} \text{s}^{-1}$ irradiance provided by white fluorescent tubes and with relative humidity of 70 %. All the experiments were repeated three times with 5 replicates per treatment.

Rooting:-

For root development, individual shoots were inoculated on half strength liquid basal medium supplemented with auxins such as IAA (2.9, 5.7, and 11.4 μM), IBA (2.46, 4.92, and 9.84 μM) and NAA (2.69, 5.38 and 10.76 μM) individually. The microshoots were supported on filter paper bridges (Whatman's No. 1).

Hardening of plantlets and transferring to community potting mix:-

About 2–4 cm long rooted plantlets (with 2–3 roots) were placed in perforated plastic cups containing charcoal pieces (\approx 2–4 cm), brick pieces (\approx 2–4 cm), coconut husks, and sand in the ratio of 1:1:1:1 and covered with holed transparent poly bag. The plantlets were maintained for 3–4 weeks in normal laboratory condition and irrigated at regular intervals. The potted plants were exposed to normal day light for about 1 hr in a day for the first week and

subsequently the exposure period was increased by 2 hrs and finally after 1 month the plantlets were placed in full day light condition.

Data Analysis:-

Asymbiotic seed germination:-

Bursting of the seed coat and emergence of the enlarged embryo i.e. the protocorm was considered as germination. The germination of seeds was recorded and percentage of seed germination was calculated. The seeds were scooped out and scrutinized randomly and observed under the microscope. The seeds were classified as germinated/non-germinated and the germination frequency was calculated by counting the total number of seeds germinated with that of total number of seeds observed.

$$\% \text{ of germination} = \frac{\text{Total number of seeds germinated}}{\text{Total number of seeds observed}} \times 100$$

Callus induction:-

The induction period, frequency of callus induction survival rate and conversion frequencies on various hormonal treatments were tabulated.

Results:-

Asymbiotic seed germination:-

The immature pods of *S. plicata* showed differential response (Table 1) when cultured on different basal media. Greening and swelling of seeds was found to be the first significant change after 8 weeks of culture.

Among the five different basal media used, B5 medium was found to be most effective in inducing a germination frequency of 95 % in *S. plicata* (Fig. 1) where as KC and MS induced 90 % and 25 % of seed germination.

Callus induction:-

80 days old protocorms were inoculated on the B5 basal medium supplemented with 2,4-D and IAA alone. Callus was initiated from the protocorms became visible within 30 days. Two morphologically distinct types of callus are observed. The first type was pale yellow in colour and compact in texture. The second type of callus was appeared slightly later and developed more rapidly than the first with translucent in colour and friable.

Callus induction was observed directly from seed derived protocorms of *S. plicata*. B5 medium supplemented with various concentrations of 2,4-D (2.26, 4.52 and 9.03 μM) were found to be efficient in inducing callus from protocorms of *S. plicata* (Table 2). 2,4-D (9.03 μM) supplemented B5 basal medium promoted 70 % callus induction within 45 days (Fig. 2 A&B). The well developed calli were transferred to hormone free basal medium for further differentiation. After 50 days with an intermediate subculture, the calli transformed into PLBs. These PLBs eventually develop into plantlets. The protocorm derived calli induced in the lower concentration of 2,4-D (2.26) shows 60 % frequency of plantlet conversion with the maximum of 4.6 ± 0.50 plantlets.

Rooting and Hardening:-

IAA significantly promoted rooting in *S. plicata* within 15 days. Number of days required for initiation was less, frequency of root induction was maximum, roots were longer and number of roots was more when compared to other hormonal treatments (Table 3; Fig 3 A&B). Higher root numbers were recorded in 5.7 μM of IAA (4.4 ± 0.92) with the maximum root length of 1.49 ± 0.34 . The well rooted shoots were washed thoroughly under sterile distilled water and transplanted into pots with a potting mixture of charcoal pieces, brick pieces and chopped mosses (at 1:1:1 ratio). The cultures were maintained for 3–4 weeks in normal laboratory condition before transferring to perforated plastic pots with potting mixture containing charcoal, brick pieces, coconut husks, and sand (at 1:1:1:1 ratio) for hardening (Fig 3 C,D&E).

Table 1:- Effect of various basal media on asymbiotic seed germination of *S. plicata*.

S.no	Basal Medium	No. of days taken for seed germination	Frequency of germination [%]
1	MS	30	25
2	LS	-	-
3	B5	30	95
4	SH	-	-
5	KC	30	90

MS - Murashige and Skoog (1962) medium, LS - Linsmaier and Skoog (1965), B5 - Gamborg B5 medium (1968), SH - Schenk and Hildebrandt medium (1972) and KC - Knudson C medium (1946).

Table 2: Effect of plant growth regulators on callus induction from 40 days old protocorms of *S. plicata*

Hormone (μM)		Number of days taken for callus induction ^a	Frequency of callus induction (%) ^a	Frequency of plantlet conversion (%) ^a	Average number of plantlets ($\pm\text{SE}$) ^{ab}
2,4-D	IAA				
2.26	-	30	64	60	4.6 \pm 0.50
4.52		30	44	45	3.4 \pm 0.38
9.03		30	70	30	3.6 \pm 0.38
	2.9	30	-	-	7.0 \pm 1.2
	5.7	30	-	-	7.2 \pm 1.2
	11.4	30	-	-	7.2 \pm 1.1

^a Data represents the mean of five replicate

^b \pm SE: Standard error

Table 3: Effect of auxins on rooting of *S. plicata* in B5 liquid medium

Growth regulators (μM)			Time taken for root initiation (days)	Frequency of root formation (%)	Average number of roots \pm SE ^a	Average root length (cm) \pm SE ^a
IAA	IBA	NAA				
2.9			15	50	3.2 \pm 0.73	0.82 \pm 0.22
5.7			15	55	4.4 \pm 0.92	1.49 \pm 0.34
11.4			15	40	3.0 \pm 0.31	1.06 \pm 0.10
	2.46		15	30	2.6 \pm 0.50	1.14 \pm 0.08
	4.92		15	27	2.0 \pm 0.31	0.69 \pm 0.12
	9.84		15	29	2.5 \pm 0.41	0.96 \pm 0.19
		2.69	15	25	2.0 \pm 0.44	0.68 \pm 0.14
		5.38	15	32	3.0 \pm 0.54	1.09 \pm 0.29
		10.76	15	40	3.4 \pm 0.4	1.14 \pm 0.17

Only the significant treatments are computed here

Data represents the mean of five replicates

^a \pm SE: Standard error

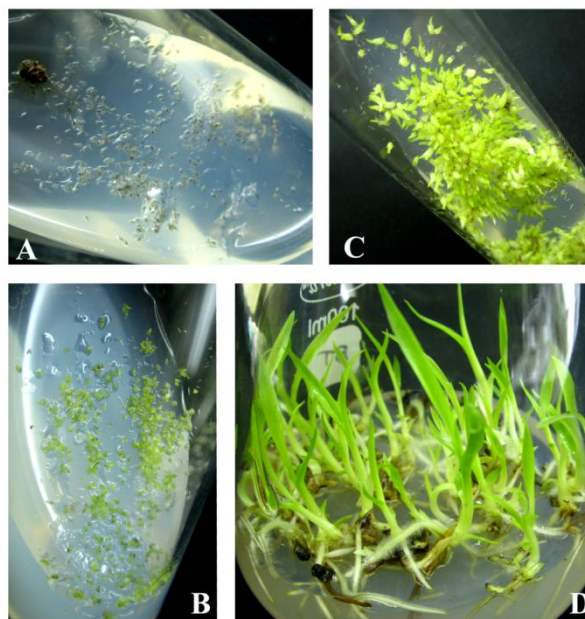
Figure 1. Asymbiotic seed germination of *S. plicata*

Figure 1:- Asymbiotic seed germination of *S. plicata*, A-Seeds of *S. plicata* in B5 medium, B- Protocorm formation, C- First leaf emergence from protocorms, D- Plantlet development.

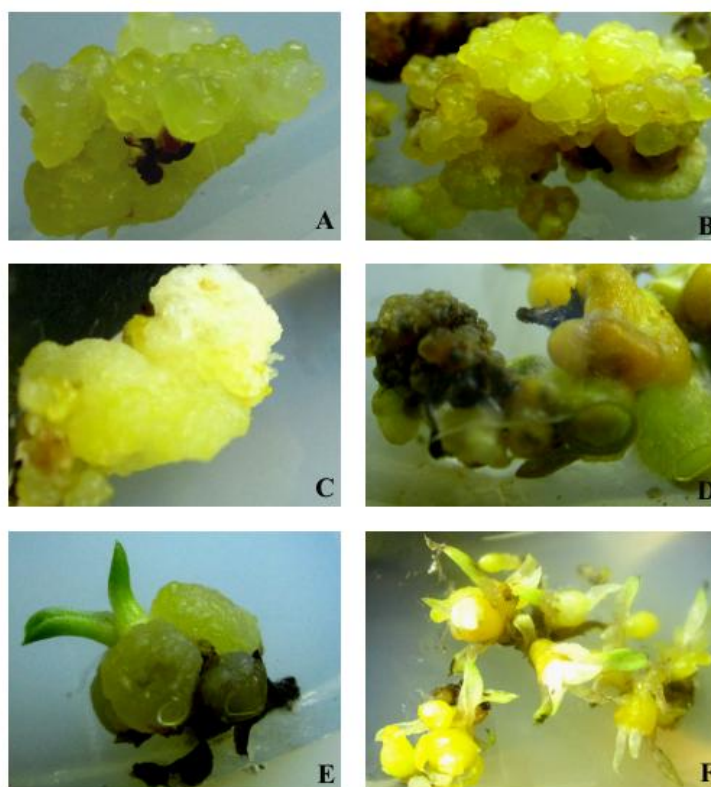
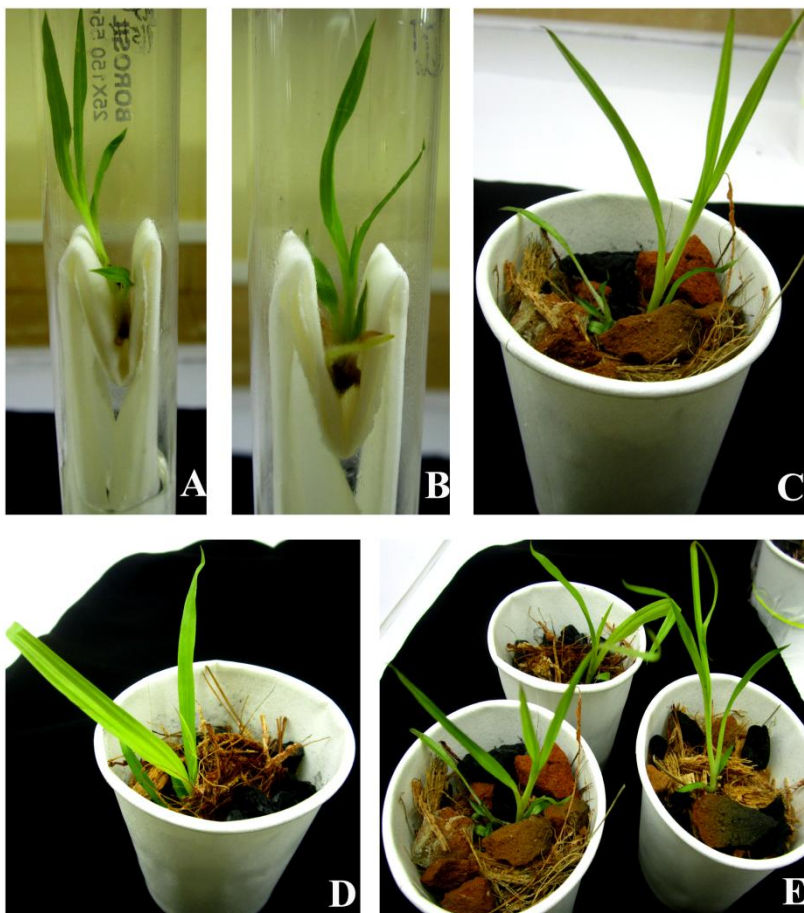
Figure 2. Callus induction of *S. plicata*

Figure 2: Callus induction of *S. plicata*, A&B - B5 medium+ 2,4-D (9.03 μM), C&D- B5 medium + 2,4-D (2.26 μM), E&F- B5 medium+ 2,4-D (2.26 μM)

Figure 3. Rooting and Hardening of *S. plicata***Figure 3:-** Rooting and Hardening of *S. plicata*, A&B - Rooting [B5 liquid medium + IAA (5.7 μ M)], C, D&E – Hardening.**Discussion:-**

Many terrestrial and epiphytic orchids have been successfully propagated using *in vitro* asymbiotic seed germination techniques (Malmgren, 1992; Arditti and Ernst, 1990; Chou and Chang, 2004). Species specific media for germination of seeds have been reported in many orchids (Arditti and Ernst, 1984; Johnson *et al.*, 2007). This study reports the standardization of suitable seed germination medium for *S. plicata*. In the present study, B5 medium was found to be the best suitable medium for the germination of *S. plicata*. Similar results were observed with the early findings in *Calopogon tuberosus* (Kauth *et al.*, 2006) and *Habenaria macroceratitis* (Stewart and Kane, 2006). The nutrient requirement for orchid seed germination differs from species to species as well as there is no universal medium for all the orchid species (Stewart and Kane, 2006). The germinated seeds continued to develop into complete plants on the same respective seed germination medium (Fig. 1 C and 1 D).

Malmgren (1992) found that the asymbiotic seed germination of terrestrial orchids were higher on media containing amino acids because this form of nitrogen can more readily be assimilated by the germinating seeds than inorganic nitrogen due to its simplified form. However, glycine, an amino acid added to MS medium was reported to inhibit germination compared to ammonium nitrate (Spoerl and Curtis, 1948). Hence, in the present study, it is inferred that the low germination frequencies observed for *S. plicata* (25 %) on MS medium may be due to the inhibitory effects of glycine (2 mg/L) present in the medium. Effects of various amino acids on germination frequencies differ among orchid species (Kauth *et al.*, 2006). The nutritional requirement for seed germination and seedling development varies with genus, species and locality. A number of standard media such as KC, Vacin and Went, Raghavan and

Torrey, MS, Hyponix medium etc have been formulated for orchid culture. Several species of orchid show specific requirement.

Generally callus induction in orchids is rather difficult due to its slow growth and necrotic tendency (Naing *et al.*, 2011A). Recently, combinations of 2,4-D and TDZ have been reported for the callus induction of ornamental plants including some orchid genera, *Cypripedium formosanum* (Lee and Lee, 2003), *Vanda coerulea* (Lang and Hang, 2006). However, BA alone or in combination with 2,4-D totally inhibited callus induction in *Paphiopedilum* hybrid (Lin *et al.*, 2000). Similarly Ishii *et al.* (1998) mentioned that the combination of 2,4-D and BA could not effectively induce callus from leaf segment in *Phalaenopsis*. This shows that 2,4-D at certain concentration induced the callus formation in orchids. Similarly the ratios between concentrations of 2,4-D, BA and Kinetin were significantly associated with the percentage survival of protocorm and callus formation. Of the combinations tested, the lower concentration of 2,4-D (2.26 μ M) was found as optimal concentrations for the best callus induction in both the orchids. Finally it revealed that addition of exogenous hormones to the medium is quite important for callus induction.

In the present study higher concentration of 2,4-D (9.03 μ M) successfully induced callus from the protocorms within a short period of time. When PLB were transferred into different kinds of basal media, formation of shoots appeared on all media but conversion frequencies of PLBs to shoot and average number of shoots were ultimately low (Naing *et al.*, 2011B). In this study, transfer of callus obtained from lower concentration of 2,4-D (2.26) to hormone-free medium stimulated more PLB development and eventually allowed to produce plantlets.

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RESEARCH ARTICLE

DECODING THE FORMULA FOR EVERGREEN BUSINESS SUCCESS.

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Abstract

The goal of this article is to understand deeply what the most important elements of success of a business. To achieve this objective, the study extensively collected, identified and analyzed best management practices, decode them into qualified factors that contribute most to sustained business performance. The findings suggest that the talented team & innovation are the main concern in determining the success. Other remarkable findings include that culture & structure, Execution & Leadership were all interrelated to each other and essentially need to be satisfied for attaining success. The mechanism through which organizational culture impacts the design of structures and vice versa is explained at the conceptual level. This article provides formula which when executed correctly will lead to increase value for both customer & company as well as sustain in this dynamic world. A schema for understanding the different potential domains for innovation is also added

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Introduction:-

For the initial 50 years of the twentieth century the path for profit optimization of firms was clear and unambiguous. In light of microeconomic hypothesis, this equation was to make an undifferentiated item at any rate conceivable unit cost and to offer it to the market at the most minimal price.^[1] Over the years number of substitutions for the microeconomic theory has showed up in business and scholarly writing. The Figure 1 represents the driving forces for success in 20th Century. An expanding number of firms found that the microeconomic theory no longer enhanced development and profitability. Subsequently, managers and academics began to search for alternative formulas for profit optimization.

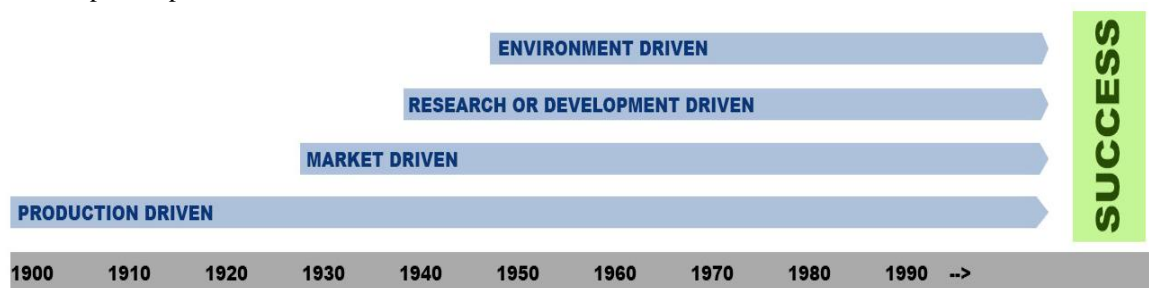


Figure 1. Driving forces for success.^[1]

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It is sensible that we have entered new age described by low development, quick change and complex. ^[2] Markets, products, and organizations have become global, diverse, knowledge intensive and multicultural. The view of the world and its systems as stable and predictable has been replaced with uncertainty, nonlinearity, and chaos.

Innovation – Need of the Hour:-

As per Forbes, the normal life expectancy of an effective Service and Product 500 Company was 67 years in the 1920's. Today it is 15 years. ^[3] More organizations need to innovate to enhance these declining numbers. We ought to be worried about many organizations that are presently effective don't have innovation instilled in the texture of their organizations. They are the ones that need to maintain a strategic distance from the FDH (fat, dumb, and happy) disorder, attempt new things and not rest on their laurels. They need to take risk to keep on achieving great success. They ought to realize that survival today requires more than treading water, and that a considerable lot of the organizations that were once extraordinary are currently gone or on out to a great extent since they quit innovating. Wherever advancements originate from, in any case they are done, and in whatever piece of the business they happen, organizations need to persistently innovate or risk dying. The problem with innovation improvement efforts is rooted in the lack of an innovation strategy. The remainder of this paper is devoted to presenting a formula for such strategic innovation

Evergreen Formula:-

Based on various literature studies, related analysis, personal observation and experience, a set of four dimensions appear to be dominant factor for business success. It is framed into equation as follows:

$$BS = e^L \times \int_s^c (wT + sI)$$

Where,

BS	= Business Success
wT	= Winning Team
ΔsI	= Triangle of strategic Innovation
Limits: C&S	= Culture & Structure (5F)
e ^L	= Execution & Leadership (3S)

In words the formula is stated as follows: “The degree of Business Success is equal to the integration of winning Team & strategic Innovation within the cultural & Structural boundaries, the whole executed under an effective leadership”. Let's take a look at each element briefly.

Winning Team (wT):-

Organizations realize that they should have the best talent keeping in mind the end goal to prevail in the hypercompetitive and progressively complex worldwide economy. The achievement of organization is directly proportional to the nature of the talent they can draw in and keep in their group for which talent and reward management plays import role.

A. Talent:-

Alongside the comprehension of the need to employ, create, and hold skilled individuals, organizations must oversee talent as a basic asset to accomplish the most ideal outcomes. The qualities of individuals who can possibly be the sort of "talent" are:

- Honesty/Integrity,
- Attitude of appreciation, energy and enthusiasm.
- Joy/Desire to be of service to others,
- At minimum a mid-level of competence combined with a high level of aptitude.

One should likewise realize that it's less expensive and more dependable to create stars than to get them. So first class training programs must be made to grow right skills and to hold top performers.

B. Reward:-

The best asset of an organization is people, on whom the greatest investment ought to be made. ^[4] Employee performance relies upon a substantial number of elements, for example, motivations, appraisals, work fulfillment, training and development, etc. Monetary incentive measures, for example, wages, and other benefits have traditionally been utilized to motivate employees to improve performance, particularly labors from production companies. ^[5] Incentives and related frameworks are fundamental to developing capacities and to translating them into better performance. However, studies have demonstrated that compensation does not help efficiency on the long run and cash does not enhance performance significantly (Whitley, 2002). ^[5] Moreover, concentrating just on this perspective may deteriorate workers' attitude, as they may seek after just monetary benefits. Luckily, there are other non-money related benefits that influence inspiration, for example, luxury gifts, travel, social acknowledgment and performance feedback.

Reward strategies are executed through measurements. But, if mere numbers alone considered as assessment for rewards, then it prompts to distorted systems. ^[6] Hence adequacy of these rewards systems additionally relies on upon various social, psychological and economic components. To actualize a genuine performance based reward system, organizations need to build up a proper reward strategy. There are many methodologies that can be utilized. It's vital to understand that there is no 'one-size-fits-all' way to deal with building up performance based reward culture. To be successful, one has to customize and tailor a system that's unique to their business needs and existing HR processes.

Strategic Innovation (ΔI):-

Innovation gets to be "strategic" when it is a deliberate, repeatable process that makes a noteworthy distinction in the value delivered to buyers, clients, partners and the company. A composite framework introduces the potential domains for successful innovation processes built within three boundaries or disciplines. Schema is given in figure:2

A. Boundaries:-

1. **Increasing value to Customers:** Strategy begins with the customer, not the company's assets. Choices must be taken whether to give superior price value (as lower costs) or superior non-price value (as superb usefulness, grand customer service, or an extraordinary customer relationship). Organization ought to include voice of clients in each progression of product development and catch their input throughout the customer journey. 85% of surveyed corporate leaders believe that strategies aiming at increasing value have more potential to lead to long-term success than strategies aiming at reducing prices ^[2]
2. **Increasing value to Company:** While watching out for operational efficiency, high performing companies ceaselessly search for better approaches to reinforce their top line. Innovative partnerships with academics, vendors, and organizations from different and/or comparable sectors are often considered to exploit the partners' offerings, processes, and resources, while profiting by internal strengths. Two-thirds of surveyed corporate leaders believe that strategies aiming at increasing revenue have more potential to lead to long-term success than strategies aiming at reducing costs. ^[2]
3. **Experimenting with new ideas:** It highly matters, what we change when the world around us changes. High-performing organizations understand the dynamics of today's economy. They urge employees to innovate and search for rapid ways to experiment with different ideas and models. Think big, begin little, learn and adjust quick, act smart. They also expect and accept failure.

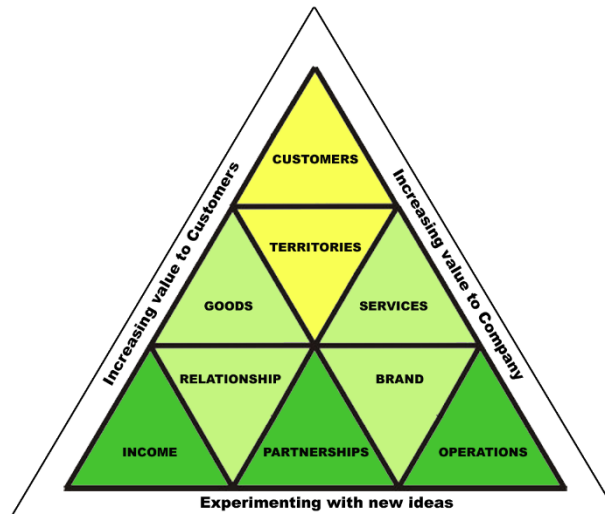


Figure 2. Strategic Innovation Triangle.

B. Potential Domains:-

1. **Market Domain:** Marketing related growth strategies express at whom the business creates value for, running from existing to potential customers or customer segments, and where a business has or could have critical importance:
 - a. **Customers:** Customers are fundamental to any business model. Without them, no organization can survive. So-called customer strategies go for characterizing which segment of people or associations an organization ought to reach and serve, and which ones it ought not.
 - b. **Territories:** Similarly, strategies around sales territories give choices to organizations to create and enter particular markets, regardless of whether local or global.
2. **Value Addition:** Strategies that encompass an organization's value offer concentrate on an organization's customer confronting components, for example, how an organization creates recognizing elements and functionalities, how it upgrades the value of its offering through additional services, how it supports convincing cooperation with customers, and how it exhibit its offering and business on the market.
 - a. **Goods:** A product-based growth strategy comprises of creating an outstandingly grand product or structure of product: by creating unique elements and functionalities, or by being more straightforward or simpler to use than rival products. This permits an organization to win a considerable premium and gain a higher market share.
 - b. **Services:** A service-based growth strategy comprises of improving the value of the core product or structure of product through additional services, for example product guarantees, maintenance plans, or consumer support.
 - c. **Relationship:** Enhancing client experience is about creating and supporting convincing and meaningful interactions with client to engage them and create a sustainable emotional bond with them
 - d. **Brand:** Brand strategies comprise of improving the portrayal of an organization's identity, goods and services to the market, including its clients, prospects, vendors, rivals, and other key business associates.
3. **Operating Model:** An organization can also gain growth by concentrating on its business and operating models, including how it make profit, how it ties with business associates, and how it supports the business over the long run.
 - a. **Income:** Organization can grow by building up better approaches to generate revenue and increment edges.
 - b. **Partnerships:** Partnerships strategies give a path to an organization to bring superior value by benefiting from its own strengths while utilizing the abilities of others. It can occur at numerous levels of the value chain and with various business associations, extending from vendors and consumers to even rivals
 - c. **Operations:** Business can also develop by ensuring their process, procedures, methodologies and other operational activities completely support their value-creation desire.

According to corporate leaders, using a mix of many potential domains is expected to lead to higher performance levels and delivers more sustainable results than isolated improvements, and does so in ways competitors cannot

easily spot or copy.^[2] However, even when multiple domains are used, sustained success is possible only when all three boundaries are satisfied.

Culture & Structure (5F):-

The most imperative factor pulling in top talent is organization's culture. In some quarters of the business world, culture is still viewed as delicate—it's not considered as important as, say, operations.^[7] In accentuating issues and critical thinking, we fail to sustain trust, discovery, and creative ability. In making numbers, measurements, and material performance the methods and the objective, we disregarded our aggregate requirement for deep engagement, motivation, qualitative inquiry, pattern arrangement, narrative and meaning. We fragmented our values, our conduct, our families, our spirituality, our lives, and our work. We separated our bodies from our minds, our minds from our hearts, our hearts from our soul, and our souls from one another. We suffer now from the lack of individuals equipped to perform the new functions.^[8]

As indicated by a Bain survey of 365 organizations in Europe, Asia and North America, even those organizations that figure out how to cultivate high-performance cultures regularly discover them difficult to maintain. They additionally found that almost 70% of business pioneers concur: Culture gives the best source of competitive advantage. The progress of a sustainable culture that expects and boosts innovation at each level and capacity of the organization really undergirds every component of the innovation framework. The common elements of this winning culture are as follows.

- A. Fun:** making the way of culture where individuals enjoy the work they do and the people that they do it with. The most straightforward approach to check whether you've done this well, is just to walk around the place and check whether individuals are smiling. Smiling workers normally show people who are having a good time at work, making the most of their occupation, and having fun around with their associates. At the point when employees live it up will probably stay faithful to the organization.
- B. Family:** the way that employees characterize a "family like atmosphere" is that they realize that their superiors and companions truly care about them as an individual. This is the kind of culture where individuals demonstrate true concern and affection for each other, both as esteemed members from the team and as esteemed members from the community.
- C. Friends:** individuals who are exceptionally connected with and faithful to their organizations often consider much of their colleagues to be their close friends, some of the time even their best friends. This is not something one can command, it develops naturally from the initial two elements of fun and family.^[9]
- D. Freedom:** in organizations with a remarkable culture, individuals are given the training, resources, time and support to carry out their tasks amazingly well – and afterward they are given the freedom (empowerment) to go beyond and succeed all alone. Work satisfaction assumes a noteworthy part in stimulating new and beneficial ideas. Workers must be given the chance to do what they observe to satisfy in their roles to harness the creativity important to set up a innovative thought process. One of the quickest ways to kill the imagination and engagement of skilled worker is to attempt to micromanage their work which must be maintained a strategic distance from.
- E. Felicitate:** In an organization that has greatly engaged and satisfied employees, one of the fundamental things that inspires them is getting sincere, honest and genuine praise at least once every seven days.^[9] This compliment does not need to originate from the supervisor, it can be gotten from any of the colleagues, and the key is that it must be particular and honest to goodness. If people feel acknowledged for their work and are included in decision-making, their upgraded eagerness and inspiration will prompt to better efficiency and loyalty.

This kind of culture (5F) is what employees are looking for where they would give 110% of their discretionary effort. In order to embrace this new corporate philosophy, organization structures will also need to become more agile and flexible. If organizational culture and structure are not in accord, there will be serious tensions and problems which will affect the organization's functioning and its results. Organizational culture justifies the behavior and rules imposed on workers and administration by the organizational structure within the context of legitimate qualities and standards of behavior. On the other way, organizational structure standardizes the culture, i.e., mirrors its qualities, norms, and attitudes. However, the organizational structure can reinforce or even change the current organizational culture. Therefore, the relation of organizational culture and structure is dual as illustrated in figure 3.^[10]



Figure 3:- Mutual relations between organizational culture & structure.

Speed of decision has become increasingly important, with strategy development cycles becoming shorter and shorter. In light of a dynamic external environment, organizations must learn quickly and create a fast, flexible, flat structure that reduces bureaucracy and simplifies work. Departmental boundaries must be broken that avoid data sharing and collaboration. Nothing wrong with bureaucracy fundamentally. Processes and protocols are essential for any organization to function well. However, an excess of formality can hinder progress, diminish employee's excitement, and drain their energy. Additionally we need from our leaders Structural readiness. This denotes to the company structure's flexibility that backing innovation and to assign existing, qualified staff to high-priority operations – without question – one of the main obstructions to implementation.^[11]

Execution & Leadership (3S):-

Recommendations are meaningless unless you can act on them effectively. Creative minds are of no use, if you cannot instill a high level of disciplined execution where your great ideas are turned into massive action. It takes strong leadership at every level of an organization, determination and a willingness to make innovative culture a top priority. Leadership is a philosophy. It's an attitude. It's a state of mind. And it's available to each one of us. Successful leaders will perform following creative roles in the new leadership paradigm

- A. Spiritual Visionaries:** Spiritual visionaries are people who decipher the universe and individuals' roles in that—or if organization is taken, its part in business and society and its worker's roles in accomplishing its mission. An optimistic effective leader defines a very clear picture of the future—a vision for the team and blueprints for action to achieve those goals
- B. Social Artists:** Social artists are individuals who continually work on themselves to develop skills & wisdom to provide consultation, leadership, and guidance on changing paradigms. Social artists work in collaborative networks to create social innovation. They use powerful conversations to build trust, encourage forward thinking and create energy within the team. The behaviour displayed by the leader also influences the subordinate's mindset
- C. Scientific reflector:** leaders are proactive, seize opportunities before competitors do and tackle problems early. They work to change the organizational culture by implementing new ideas; eliminate waste, increase productivity. Delegate decision making wherever required.

Execution is a derivative of very clearly communicated expectations, visibility around key performance indicators, high levels of accountability and high levels of employee engagement. Investments in technologies and systems like ERP, CRM or SCM etc also influences success. The right tools clear the path for smoother execution and may improve the end product. But tools mean nothing without the right expertise and the right ingredients behind them. Software should support the process, but it can't stand alone.^[12] Disciplined attention & flawless operational execution is what really counts.

Limitations & Future Work Considerations:-

This fundamental research is mainly concerned with generalizations and with the formulation of a theory; can also be iterated as “Gathering knowledge for knowledge's sake”. While each factor of business has its own specific objectives, the formula as a whole has one overarching goal: Evergreen business success. It can be used as practical management tools which assures the firm's profitability & sustainability. It is primarily intended for strategy formulation or validation, redefining of existing strategies to make sure it's successful. The formula can be highly effective to entrepreneurs & decision makers – at all levels and in all functions of an organization – anyone who is struggling to make choice among multiple options. This vital decision is what turns the company from a nonprofit into a moneymaking endeavor in long run, whether across growing or declining market environment. Formula given will be relevant across industries like Automotive; Communications, Media; Chemicals; Building; Nutrition; HR Services; Hospitals; Retail, and Fabrics, but not limited to. Although this qualitative research addressed a major limitation of past studies, that being the lack of explicating the relationship/critical-link between all major variables,

it does not consider these factors in an empirical contextual, rather provided as a 'conceptual umbrella'. But this is an excellent base from which subsequent research will be undertaken on validation of these relationships in diverse environmental contexts. It would also summarize key trends, challenges/opportunities, and illustrates how this formula is applicable, through real-life examples in follow-up studies.

Conclusion:-

In the business world, the mandate for innovation is inescapable. If a company doesn't adapt to fit changing realities, it risks losing its position in the market. The formula provided is intended to tell managers which management practices they need to focus on and how they can improve it. The triangle of strategic innovation helps to grasp the full potential of future growth opportunities, and also corporate leaders will need to reinvent their business with above stated 5F & 3S techniques to achieve evergreen success.

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RESEARCH ARTICLE

COMPARATIVE STUDY OF CONSUMER'S INVOLVEMENT FACTORS IN FASHION CLOTHING.

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Key words:-

Fashion clothing involvement, consumer behavior, confidence, knowledge about purchasing and consumer's involvement.

Abstract

For many years, Fashion clothing has been an area of interest to many consumer researchers. This study examines the effect of age, gender, materialism and self-image product image congruency on consumer's involvement in fashion clothing. Today everyone has become materialist to get success. Thus people use fashion clothing as an important key to express their self to others and fit themselves in social environment. As fashion clothing has become the essential part of consumer's life. So, it is important to investigate consumer's involvement in fashion clothing to understand their behavior. The main purpose of this study is to measure the fashion clothing involvement of youth, adults, working people selected in Sargodha city. Fashion clothing is the center of this research. The sample size chosen for this study was 150. All samples were selected from Universities, Job Organization and Business establishment in Sargodha city. Factor analysis using principle components with varimax rotation was used. Also, the KMO and Bartlett's test was conducted in order to find or examine the interactions and relationships between variables.

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Introduction:-

The term "Fashion" is accounted for through the attributes of change, ambivalence, novelty and added value when associated with an object (**Kawamura 2005, pp.4-5**). Clothing actually reflects the identity of wearer. **On Kivisit and Shaw (1987)** found that consumers select and buy products according to self-concept. Fashion involvement is used mainly to predict behavioral involvement and characteristic. Clothing sector firms are competing to increase their profit share in the market and among these firms; branded clothing has shifted the conventional clothing interest of people. Brand awareness have gained importance among consumers.

Mostly the choice of brands and products is based upon family acceptance. In Pakistan modernism and fashion clothing is a symbol of happiness and well-being. A large no. of Pakistan families adopt any fashion style but keeping their values and religious rules in their minds. Clothing may be seen as symbolizing status not only to the individual but also to the family and society.

Research reveals that the brands use to present a good image for customers. Brands are not that important for marketing but still they are viewed as improving the lifestyle of the consumers. The main benefit of brands is to give

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status, value and quality to consumer. So that the consumer use more brands to enhance his personality. Similarly, we have chosen fashion clothing to examine the level of involvement of consumers with its purchase decision.

The need for the study is to check whether the purchase knowledge self-confidence fulfillment, social status, materialism, brands and fashion involvement and demographic characteristics of the youth, adults, job holders and business establishment consumers in Sargodha city can effect on their fashion involvement and also income.

Zaichkowsky (1986) defines involvement as “a person’s perceived relevance of the object based on inherent needs values and interest”. From this definition we can say that values play a key role in forming a consumer’s involvement to any product.

Review of Literature:-

The main purpose of this study was to examine the involvement of all the selected consumers in fashion clothing and whether the female consumer ranging age from 30 to 59 prefer fashion advertising models more closely to their age. The sample for this study was consisted of 182 females. A questionnaire designed to examine the response of consumers according to the given scales measuring participant’s belief about the model’s appearances and attractiveness. And how it effects on the purchasing that of clothes. Participants who perceived more similarity to the models were found to have more positive beliefs about the model’s appearance and attractiveness and the fashion ability of the model’s clothing. Hence the researcher finds in this study that the marketers should pay more attention to the advertisement of their clothes and should consider the age of the model used in their promotional materials.

(Kozar 2010):-

McFatter (2005) studied on fashion involvement to understand consumer behavior about purchasing and also to develop improved marketing strategies. The purpose of this study was to examine that the prediction of fashion involvement of female consumers can be determine by knowing the personality likes, media usage, price perception and selected demographic characteristics of the female consumers. Relationships to fashion involvement are discussed including the following variables: media usage, self-confidence, self-fulfillment, price perceptions and selected demographic characteristics. At the end of this study the researcher fond that the consumer’s media usage and price perceptions were significant of fashion involvement, while personality traits were not.

Browne and Kaldenberg (1997) presented a casual relation between materialism and involvement. In this context, it could be inferred that fashion clothing, as possession, may be seen for its role as a code. For instance, **Noesjirwan and Crawford (1982)** make this convergence saying that clothing is primarily a means of communicating, not personal identity, but social identity to others.

The study by **Grant and Stephen (2005)**, examine the impact of brands on the behaviors of younger teenage girls and their purchasing decision for fashion clothing. This group is highly Fashion-sensitive. The main purpose of study is to determine the role of branding and purchasing influences from the perspective of these young people in order to gain a richer and deeper understanding of their behavior. The findings revealed that the respondents were prepared to pay a premium for a branded product. This research also provided the basic difference of buying behavior of parents and children.

Kawabata and Rabolt (1999), compared the clothing purchase behavior of US and Japanese female university students. Evaluative criteria when buying clothing, an interest in clothing and fashion and source of fashion information were studied. Relationships between variables were investigated and the difference and similarity between the two countries were discussed. US students tended to give higher scores for fit quality, fashion and branded than Japanese students. We found the difference in the meaning of fashion between the two countries by cluster analysis. Japanese students were more interested in fashion when they spent more money on clothing. US students were more active in collecting information about fashion. In both countries, students who were selected for information tended to have more interest in fashion clothing.

Research Methodology:-

Statement of the Problem:-

To know the fashion clothing involvement among consumers in Sargodha city. To determine the fashion involvement of different genders (Students, Job holders, Businessman and housewives) from Sargodha. To identify the significant factors which are important to understand the involvement of customers in fashion clothing.

Objectives of study:-

The objectives of the present study are,

1. To identify the factors of fashion clothing involvement.
2. To identify the no. of people who prefer fashion clothing and also identify their demography.

Sampling Technique:-

The sample for determine the involvement of consumer in fashion clothing was collected with the help of given questionnaire from 150 consumers in Sargodha city. Samples were drawn from universities, job organizations and business establishments in Sargodha city.

Method of Data Collection:-

The primary data was collected by explaining the structured questionnaire to the respondent. Secondary data was collected by internet, news-paper, books and articles.

Analysis:-

Factor analysis and reliability tests:-

This study consists of five dimensions: confidence, quality interest, materialism, purchase knowledge, social consciousness in fashion involvement. To test and verify all these dimensions and reliability of these constructs including principal components factor analysis, items to total correlation analysis, and Cronbach's alpha analysis were conducted.

Questionnaire items with higher factor loadings were selected.

KMO and Bartlett's Test.

Kaiser-Meyer-Olkin Measure of Sampling			.650
Adequacy.			
Bartlett's Test of		Approx. Chi-Square	409.004
Sphericity		df	105
		Sig.	.000

The Bartlett test for Sphericity was significant at 105 degree of freedom ($\chi^2 = 409.004$, $p < 0.01$), which also showed the presence of significant correlations among variables. Another index of appropriateness of factor analysis is the Kaiser-Meyer-Olkin (KMO) measure of sample adequacy, which measures whether the distribution of values is adequate for conducting factor analysis. In this case, KMO was as high as 0.650.

Rotated Component Matrix:-

	Component				
	1	2	3	4	5
Fashion clothing is a medium to fit into the social environment.	.705				
Wearing clothing is the most satisfying and enjoyable thing to me.	.700				
Fashion clothing is central to my identity as a person.	.657				
I read the fashion news Regularly and try to keep.	.325				
I like being involved in making buying things of fashion clothing.		.707			

i like to purchase things Of fashion clothing to impress other people.		.529			
I think ads about fashion clothing are important to me to buy.		.495			
Consuming latest clothes fashion increases my self-confidence.			.754		
I'm more confident that I buy good quality of product.			.708		
Fashion clothing gives me a sense of self fulfillment.			.564		
Color is very important Influential on my choice of thing.				.807	
Knowledge concerning the new fashion trend is important.				.685	
People notice when you materials most					.781
Expensive brands of product.					

I purchase things of new fashion clothing					.686
Only when they are well accepted.					
My social status makes me think of buying materials of fashion clothing.					.489

Extraction Method: Principal Component Analysis. Rotation Method: Varimax with Kaiser Normalization.^a a. Rotation converged in 8 iterations.

There are five factors extracted from the exploratory factor analysis. Now we name all the factors according to the properties of their variables. By using these factors we'll check the reliability of the variables By finding Cranach's alph

Fas	Statement	Loading	Alpha
	Fashion clothing is		
	medium to fit into	0.705	
	the social		
Social	environment.		
Conscious	Wearing fashion		
	clothing is one of		
	the most satisfying	0.700	
	and enjoyable thing		
	to me.		
	Fashion clothing is		
	central to my	0.657	
	identity as a person.		
	I read the fashion		
	news regularly and		0.680
	try to keep.	0.325	
	I like being involved		
	in marking buying	0.707	
	things of fashion		
Purchase	clothing.		
Knowledge			
	I like to purchase		
	things of fashion		
	clothing to impress	0.529	
	other people.		
	I think ads about	0.495	
	fashion clothing are		0.333
	more important to		
	me to buy.		
	Consuming the		
	latest fashion	0.754	
	clothes increase my		
Confidence	self-confidence.		

	I'm more confident			
	that I buy good	0.708		
	quality of product.			
	Fashion clothing			
	gives me a sense of		0.613	
	self fulfillment.	0.564		

	Color is very			
	important influential	0.807		
	on my choice of			
	things.			
Materialism				
	Knowledge			
	concerning the new			
	fashion trends is	0.685		
	important.			
			0.453	
	People notice when			
	you purchase	0.781		
	materials of fashion			
	clothing.			
Quality				
Importance	I purchase things of			
	new fashion			
	clothing when they	0.686		
	are well accepted.			
	My social status			
	makes me think of	0.489		
	buying materials of			
	fashion clothing.		0.45	

This table gives the name of all the factors of fashion clothing involvement. This table also gives the values of loading for all variables and the last column shows the Cronbach's alpha values of each of the factors.

The values of Cronbach's alpha represent the reliability of the factors. If their values are more than 0.5, than the factors are assumed to be more reliable.

The major factors of fashion clothing involvement are

Social consciousness (Cronbach's alpha =0.68) Social consciousness is the most dominant factor. Fashion clothing is medium to fit into the social environment and central to consumer's identity as person. Consumer selects such type of clothes which are good to move in society and the other would like to carry that too. Furthermore, only those clothes will be acceptable for them which are accepted by their environment.

Purchase knowledge (Cronbach's $\alpha=0.333$) Purchase knowledge is the second dominant factor. This shows, consumers feel that they should have sufficient level of purchase knowledge about the product and new brands. For that, they think ads are very helpful to have purchase knowledge.

Confidence (Cronbach's $\alpha = 0.613$) Confidence is the third dominant factor. "Consuming the latest fashion clothes increases my self-confidence and more fashion clothing is important for their self-fulfillment. According to consumer, carrying new brands and trends gives them confidence to move in society

Materialism (Cronbach's $\alpha=0.453$) Materialism is the fourth dominant factor. Materialism indicates the purchasing as status, color or happiness. It shows that the consumer often used some brands or product to show off, therefore, the people that have Materialism values are more prone to be highly involved in fashion clothing.

Quality importance (Cronbach's $\alpha=0.450$) The fifth factor is called quality interest." People notice when you purchase the materials most expensive brand of product". Young consumers that are highly involved with fashion clothing, behavior reveals more interest in fashion clothing and also the product quality.

Finding:-

From the heading no.4, Analysis shows the analysis of demographic variable showed the homogeneity and heterogeneity between the consumers of different age group given in questionnaire. It shows that nothing is like in the choices of different generations in social consciousness, purchase knowledge, confidence, materialism and quality importance. The young generations carry new brands and expensive products to gain social approval.

In next table, KMO and Bartlett have showed the significant of correlation among variables and also showed that the values are adequate for conducting factor analysis. The Bartlett's Test of Sphericity was significant at 105 degree, of freedom (Chi-square= 409.09).

"Rotated component matrix" table gives the five factors which are highly involved in fashion clothing. All the factors contain different no. of variables.

Last table is "Naming of factors", in this table all the factors got names according to their properties. Social consciousness is the most dominant factor in this analysis which shows that the consumers adopt new fashion trend to survive in society. The respondent given in conducted in the questionnaire carry fashion and brands to make their personality in social environment. After social consciousness, purchase knowledge, confidence, materialism and quality importance are the factors which are highly involved in fashion clothing. In second column loadings are given against each variable. The Cronbach's α values represent the reliability of the factors. Since the Cronbach's α values for two factors are more than 0.5, so these factors are more reliable than others. Social consciousness is the most dominant factor than others factors.

Conclusion:-

After all the research work it can be concluded that this study can be used full for the consumers as well as the marketers who are trying to promote their product to the people because it gives them knowledge about the liking and thinking of consumers. This research based on consumer in Sargodha city. The researchers conclude that the selected demographic characteristic, the analysis showed that young people and females consumers involved more than the adults and men's. It could be credited to the fact that the most of the respondent were teen agers, young and females. Although the literature indicated that confidence might have some relationship to fashion involvement, they were significant in this study. The majority of the correspondences were female and student this could have contributed to confidence. By using this research, the marketer can gain benefit by developing pricing and could branding. Because this study have conclude that ads and branding effect on the involvement of consumers in fashion clothing. Retailers can also get benefit from the study by knowing what to consider when pricing products and promoting to consumers.

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RESEARCH ARTICLE

STATISTICAL ANALYSIS OF DETERMINANTS OF CUSTOMER'S SATISFACTION ABOUT SUNDAY BAZAR AT SARGODHA, PAKISTAN.

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Abstract

The study attempts to discover out the determinant of customer satisfaction towards Sunday bazaar in 49 Tail, PAF Road. In this study 145 responses was surveyed by useful methods. For the literature review and expert opinion it has been found that customer satisfaction are based on products, price, services, atmosphere and personal interaction. Regression Analysis was used to identify the impact of factors on customer satisfaction. The research finding out that these factors were positively impacted on customer satisfaction towards studied Sunday bazaar.

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Introduction:-

In today highly competitive marketing environment the positioning decision of Sunday bazaar from store point getting goods from store point of view play important or in marketing management. Now a days shopping behavior of users has experienced a strange hanged and is putting many obstacles for market today. Sunday bazaar need to have not only the market designs to give them a better chance over the competitors but also need to develop these in a manner that uniqueness can be mainland international.

Traditionally price and differentiation of the products were consider as one of the most important factor to maintain the customers and satisfy them because Sunday bazaar owner provide their customers new varieties every Sunday. It is very important for the firms growing and development process to have customer satisfaction. Customer have become an important part of any firm especially those in the grocery retails sector and many researcher have force on the importance of the customer. Zairi (2000) cited in Singh (2006) said, "customers are the purpose of what we do and rather than depending on us, we very much depend on them. The customer is not source of a problem; we should not perhaps make that customer, should go away" because our future and our security will be put in jeopardy". That is the main reason why firms today are forcing on the customers satisfaction and loyalty.

According to Hnasemark & Albinsson (2004) cited in Singh (2006), "satisfaction is an overall attitude towards a product provider or an emotional reaction to the difference between what customers expect and what they actually receive regarding the fulfillment of a need".

We undertook survey of Sunday bazar in Sargodha and conclude that most of people shop here because it satisfied them. As the quality of product is better, not only better quality but also at reasonable price. It satisfied the customers as the get different varieties under same roof like. It saves there lot of time and also give the advantage of

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low price. Especially ladies prefer to shop here because they get advantage of getting different things like grocery, clothing, readymade food and many others.

These benefits are passed on to consumers in the form of savings. The number of the customers increase, it is more profitable for the owner of bazar as it achieves benefit of cost. As the buyers increase it also increases the quality of the different items. It promotes the international market. Progress of Sunday bazar is although slow but even then customers prefer to buy the goods of daily use because it provides advantage of low price and personalized service. That is the main reason why customers prefer to shop here.

Literature Review:-

Evans and Lindsay (1996) stated that companies with satisfied customers have a good opportunity to convert them into loyal customers who buy from these companies over an extended period of time. Today "in the highly competitive and dynamic business environment requires financial institutions to have satisfied customers and keep them in order to survive and compete with other players in the market successfully.

According to **Sivadas and Baker-Prewitt (2000)**, satisfaction also influences the likelihood to recommend a bank and redemption. It promotes loyalty to the extent that it is a prerequisite to maintain an attitude favorable report and recommend a bank takeover. Once customers recommend a financial institution, it promotes both redemption and loyalty to this financial institution. Thus, the key to generating loyalty is to get customers to recommend to other service provider. In addition, customers are likely to recommend a service provider when they are satisfied with the services and when they have a relatively favorable attitude to this service provider.

According to **Hansemark and Albinsson (2004)**, satisfaction is an overall attitude of clients to a service provider, or an emotional response to the difference between what customers expect and what they get, regarding the achievement of a need, purpose or desire.

Nguyen et. al, (2007). Studied on exploring the impact of hedonic shopping motivations and attributes on a supermarket loyalty shopper. In addition, the study focuses on loyalty attitudes. Loyalty behavior should be considered in future research. Found that as supermarkets attributes and hedonic shopping motivations had positive effects on shopper loyalty. And also found that the impact of hedonic motivations on shopper loyalty was different between younger and older, as well as low-income groups and higher customer. However, no difference was observed between male and female buyers.

Jayant Anand (2009). In this study evaluated the reason behind number of supermarkets in developing countries increasing and found that large and small retailers can survive profitably by separating their markets on the basis of income groups and their product grouping.

Skallerud and Gronhaug (2010) investigated the positioning strategies supermarket Chinese food and found some important factors related to the nature of the relationship between food supply and demand for food could be influenced by the positioning strategies of supermarkets.

According to **Mohsan et. al, (2011)** studied the best-performing financial institutions always seek the needs and requirements of their customers in order to survive and compete successfully in a dynamic business environment of today. That is why researchers' organization worldwide have consistently stressed the importance of customer satisfaction, loyalty and retention. The results of the study revealed that customer satisfaction is positively correlated with customer loyalty and negatively correlated with customer intentions to pass. Some recommendations for future research are also made.

Nguyễn Thị Thu Thương (2016). Studied on attempts to find out the determinants of customer satisfaction towards supermarkets in Thai Nguyen city. In this study 200 responses of the 4 selected supermarkets in Thai Nguyen city were surveyed by convenient method. The research findings showed that product, price, personal interaction, convenience, services and physical appearance were positively impacted on customer satisfaction towards studied supermarkets. Among them price had a strongest influence on customer satisfaction.

Research Methodology

The research process consist of five main steps. The first step consist of information to relate customer satisfaction with specific reference to Sunday bazaar. The second step focuses on questionnaire based on information. The third step consist of collecting data. The forth step contains data interpretation and analysis using appropriate statistical tools. The fifth step contains an outcome of the analysis.

Likert-type scale ranging from strongly agree (1) to strongly disagree (5) was related point to each of the identified selection attributes.

Exploratory factor analysis (EFA) offers an effective way of assessing construct measures. Accordingly, item-total correlations and Cronbach's alpha were first used to assess reliability, followed by principal components analysis along with varimax rotation to explore dimensionality for each scale. In this process, the items don't meet evaluating criteria are eliminated. Criteria of refinement of item included factor loading > 0.50 , item-total correlation > 0.3 , Cronbach alpha > 0.60 , % of variance $> 50\%$ (Hair et al., 1998). Using SPSS 21 software we were able to assess and refine measurement scale. Regression Analysis was chosen to identify the impact of the independent variables on dependent variable (Customer Satisfaction). Primary data was collected primarily through survey method using a structured questionnaire which was designed to capture the factors impacting customer satisfaction in Sunday bazar.

Sampling:-

Samples were selected by convenience method. The interviews were conducted in person at the Sunday bazar, household, offices, schools etc. Samples were selected as students, teachers, office staff, housewives etc. Based on the number of parameters to be estimated, the sample size targeted in this study was 145.

Hypothesis:-

In order to explore factors influenced significantly customer satisfaction and define the influenced level of these factors, sets of hypotheses were developed.

H1: There is a positive impact of product on customer satisfaction.

H2: There is a positive impact of price on customer satisfaction.

H3: There is a positive impact of personal interaction on customer satisfaction.

H4: There is a positive impact of convenience on customer satisfaction.

H5: There is a positive impact of services on customer satisfaction.

H6: There is a positive impact of physical appearance on customer satisfaction.

Analysis And Discussion Of Findings:-

Cronbach Alpha for Scale of Factors:-

Following the procedure and criteria described above, the Cronbach alpha results show that PR1 (Price of goods is appropriate to their quality) should be deleted due to items-total correlation of .365 under the cut-off value Cronbach alpha (0.4). The result Cronbach alpha is showed in the Table 1.

EFA for all Scale of Factor Together:-

The result of Bartlett's Test of Sphericity and KMO measure indicated that the degree of inter correlation among the items were suitable for EFA procedure ($KMO=0.678 > 0.5$, $Sig = .000$). After establishing the multidimensional scale and reliability of each scale, all 19 items were jointly subjected to a common factor analysis the result of this procedure shows that 6 factors consist 19 items together explained 64.667% of the total variance. In this testing, rotation method: Varimax with Principal Component Analysis was applied providing better reliability.

Table 1:- Cronbach Alpha Reliability Test Result.

Construct/items	Item	Description	Corrected item- Total correlation
Product-PROD (Alpha=.559)	PROD1	Products in Sunday bazaar is durable.	.558
	PROD2	I find product with high quality in the Sunday bazaar.	.490
	PROD3	Variety of product is good.	.651
	PROD4	The brands sold by the Sunday bazaar are good ones.	.634
Price-PR (Alpha=.299)	PR1	Price of goods is appropriate to their quality.	.365
	PR2	The Sunday bazaar provides special discount for potential byers on different occasions.	.448
	PR3	Prices in the Sunday bazaar are competitive.	.783
Personal interaction-PI (Alpha=.473)	PI1	The salesmen have enough knowledge to answer my question.	.761
	PI2	The salesmen assistance during purchase is good.	.720
	PI3	Billing is always correct.	.687
	PI4	Shopkeepers are friendly and polite.	.722
Convenience-CO (Alpha=.388)	CO1	Sunday bazaar location is convenient.	.737
	CO2	Sunday bazaar has good return policies.	.714
	CO3	Convenient and safe parking facility is available.	.658
Services-SER (Alpha=.655)	SER1	Security of Sunday bazaar is very good.	.688
	SER2	Delivery of goods is available.	.701
	SER3	Sunday bazaar's after sales services are good.	.629
Physical Appearance-PA (Alpha=.275)	PA1	Cleanliness of the Sunday bazaar is good.	.479
	PA2	A layout that allows me to easily find the products I need.	.431

Table 2:- Rotated Component Matrix^a

	Component						
	1	2	3	4	5	6	7
Delivery of goods is available.	.823						
Sunday bazaar's after sales services are good.	.638						
Security of Sunday bazaar is very good.							
Overall I am very satisfied with the factor influencing customer in Sunday bazaar.	.625						
Price of goods is appropriate to their quality.	.485						
The Sunday bazaar provides special discount for potential buyers on different occasions.	.483						
Sunday bazaar has good return policies.							
Convenient and safe parking facility is available.							
A layout that allows me to easily find the products I need.	.355						
Products in Sunday bazaar is durable.							
The salesmen have enough knowledge to answer my question.		.806					
The salesmen assistance during purchase is good.		.640					
Cleanliness of the Sunday bazaar is good.		.573					
I find product with high quality in the Sunday bazaar.		.338					
Variety of product is good.			.792				
The brands sold by the Sunday bazaar are good ones.			.712				
Shopkeepers are friendly and polite.			.507				
Billing is always correct.			.374				
Sunday bazaar location is convenient.				.741			
Prices in the Sunday bazaar are competitive.				.713			
					.802		
					.762		
						.840	
							.862

Extraction Method: Principal Component Analysis

Rotation Method: Varimax with Kaiser Normalization.^a

Rotation converged in 10 iteration

Testing Model and Hypotheses:-

Based on the revised model, the following function was established to present the relationship between independent variables and customer satisfaction (CS).

$$CS = b_0 + b_1*PROD + b_2*PR + b_3*PI + b_4*CO + b_5*SER$$

Regression Result:-

As shown in table 3 and 4 the result indicated that R square = .360, F value was 12.962, sig. = 0.000. These demonstrated that the liner multiple regressions between factors and customer satisfaction are appropriate with data and able to be used. The Sig. of factors was shown that four factors among five factors have statistical significant impact on customer satisfaction (table 5). VIF<2, it means that there is no multicollinearity in the research model.

They are product, personal interaction, service and physical appearance with standardized coefficient range from 0.227 to 0.236. Through the value of R square, the explanatory levels of the model is 36.0%. Its mean 36.0% can be explained by the four independent varieties.

Table 3:- Model Summary^b

Model	R	R square	Adjusted R square	Std. Error of Estimate	Change Statistics					Durbin-Watson
					R square Change	F Change	Df1	Df2	Sig. F Change	
1	.600 ^a	.360	.333	.372	.360	12.962	6	138	.000	1.468

a. Predictors: (constants),PHY,PRI,PROD,PER,SER,CON

b. Dependent variable: SAT

Table 4:- ANOVA^a

Model	Sum of Squares	df	Mean Square	F	Sig.
1 Regression	10.753	6	1.792	12.962	.000 ^b
Residual	19.081	138	.138		
Total	29.834	144			

a. Dependent variable: SAT

b. Predictors: (constants),PHY,PRI,PROD,PER,SER,CON

From the equations above it can be seen that one unit change in service increased by 0.236 times the level of customer satisfaction similarly one unit change in product increased by 0.227 times the level of personal loyalty.

Hypothesis 1:-

There is a positive impact of product on customer satisfaction.

The standardized regression coefficient of product on customer loyalty is 0.227 (sig. = 0.004 < 0.05). This means that product is directly proportional to on customer satisfaction with 95% level of confidence.

Therefore, the hypothesis 1 is supported.

Hypothesis 2:-

There is a positive impact of price on customer satisfaction.

The standardized regression coefficient of price on customer loyalty is 0.084 (sig. = 0.251 > 0.05). This means at the level of confidence of 95%, price do not contribute to explanation of level of customer satisfaction.

Therefore, the hypothesis 2 is rejected.

Hypothesis 3:-

There is a positive impact of personal interaction on customer satisfaction. The standardized regression coefficient of personal interaction on customer satisfaction is 0.126 (sig. = 0.089 < 0.05). This means that personal interaction is inversely proportional to customer loyalty with 95% level of confidence.

Therefore, the hypothesis 3 is rejected.

Hypothesis 4:-

There is a positive impact of convenience on customer satisfaction.

The standardized regression coefficient of convenience on customer loyalty is 0.088 (sig. = 0.290 > 0.05). This means that convenience is inversely proportional to customer satisfaction with 95% level of confidence.

Therefore, the hypothesis 4 is rejected.

Hypothesis 5:-

There is a positive impact of services on customer satisfaction.

The standardized regression coefficient of services on customer loyalty is 0.236 (sig. = 0.003 > 0.05). This means that services is directly proportional to customer satisfaction.

Therefore, the hypothesis 5 is supported.

Table 5:- Coefficients^a

Model	Unstandardized coefficients		Standardized coefficients	t.	Sig.	Collinearity Statistics	
	B	Std. coefficients	Beta			Tolerance	VIF
1 (Constant)	-.147	.182		-.805	.422		
PROD	.150	.050	.227	2.967	.004	.790	1.266
PRI	.058	.051	.084	1.154	.251	.865	1.156
PER	.081	.047	.126	1.714	.089	.857	1.166
CON	.048	.046	.088	1.061	.290	.675	1.482
SER	.108	.036	.236	2.991	.003	.742	1.348

a. Dependent variable: SAT

Conclusion:-

In Sargodha Sunday bazar is doing well and we think this types of bazars should be organized in different cities. We surveyed from different people around these areas and conclude that most of housewives, office holders and students overall they are very satisfied with the quality of product and price.

As the number of customer increase, they get more profit. The result, it is great advantage for the owners. They will provide new product and of better quality at reasonable price. This will promote international economy.

These types of bazar should be set in more cities so that every citizen can buy the different item of daily use. The manager should also focus on the negative impacts and try to improve them. Price has the highest impact on Sunday bazar, followed by product. an important conclusion from study, customers are satisfied with almost all factors.

The manager and retailers focus on the said attributes to propose appropriate promotion and positioning of Sunday bazar.

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RESEARCH ARTICLE

ARAB SPRING AND THE ARAB -ISRAELI CONFLICT.

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Abstract

The Arab Spring as waves of protests brought down several dictator regimes that some of them were a pro-Israeli government. Israel, even if they wanted to, they could not stay away from Arab neighbors, especially with the continuation of the historical conflict with the Arab world, because of the Palestinian issue and the consequent tensions and animosities. Pledge Israel and its leaders are aware that what was happening in the Arab neighbors have impact in various fields and levels, which is a historical issue has been to prepare for it by setting up scientific institutions aim to monitor development in the Arab world, analyze and understand the meanings of these reflections on Israel and the Arab-Israeli conflict. The fear of Israel was real, when the people were attacking of Israel's embassy in Egypt, and the long peace treaty between Egypt and Israel has threatened. In opposite, signing an understanding deal between Fateh and Hamas, and re-opening of the Rafah crossing at the border between Egypt and the Gaza Strip that Israel has seen it as a threat to its national security. In addition, escalating Syrian civil war and growing terrorist groups in Syria has made the situation more complicated for Israel. The paper tries to evaluate the importance of the political uprising in the region on Israeli situation. Likewise, illustrate the impact of the Arab spring on the Arab-Israel conflict, and then how their relation has an impact on the security stability in the Middle East.

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Introduction:-

Mass demonstration and popular revolts have been an outbreak with began in Tunisia at the end of 2010, and in other countries in the earlier 2011. Thus, a great wave of the uprising has started in the Islamic world generally and Arabic countries, particularly. This wave of protest has to lead into bring a political upheaval and eliminating some authoritarian Arab regimes which four presidents ultimately lost their power in the region. Additionally, such upheaval has opened a door to Islamist parties to come to power and have been a major player in those countries.

However, Arab spring marks the end of the stability of secular authoritarian regimes in the Arab world. Those countries that have experienced revolution, have stepped toward democratic and elective government, warm and active participation by the public in political activities, peaceful rivalry among political parties and prevent electoral fraud such as double voting and so on.

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Furthermore, the elections in those countries have been proven that the Arab spring is also an Islamist spring, not only because of their results which they have gotten from the post-dictatorship election but also due to that role they have played in overthrowing secular dictatorship regimes. The initiated waves of change have affected every aspect of life in the region, including the Israel-Palestinian conflict; the primary reaction of Palestinians to the Arab spring was reconciliation between two major parties in Palestine both Hamas and Fatah.

The political changes in the Arab peninsula have definitely had a fundamental effect on Palestinian's and the Arab public way of thinking toward Israel. In the new Arabic world, public opinion is much more vital than the president or individual decision, and in the case of Arab-Israel conflict, it apparently seems public opinion will decide how the strife will be ended. And this is made a serious anxiety for Israel decision-makers about the future of their security and relations with the Arab world. Thereby, building security barriers on the borders by Israel with its neighbors such as Egypt and Jordan, enlarging the security budget and abstaining from any gesture toward Gaza Strip and West Bank have only been some of the steps taken by Israel in response to the Arab Spring as well.

This paper will discuss some questions such as, to what extent rising Islamic parties are creating an opportunity to minimized danger the conventional war between Arab-Israel? Or it might become escalating Arab-Israel conflict? Does Arab spring would inspire greater support for Palestinian conflict with Israel? I also discuss how the political upheaval in the Arab countries affects the Arab – Israeli conflict, and to what extend Israeli have concerned about the Arab spring? Here, there are some objectives, which are motivated to write this work, and those are clarifying what Arab spring is? How is it started? Similarly, an illustration of the impact of the Arab spring on the Arab-Israel conflict, and how their relation has an influence on the security stability in the Middle East will be discussed?

The Arab spring in the Middle East and North African countries has erupted in the result of existing of several internal factors, which became collapse some leadership who's had a strong relation with West and Israel, and they could keep the peace for a long time. Notwithstanding, shifting in some of those regimes have pushed the United States and Israel to rethink about their relations in such a way, which keep peace with Arab countries. In order to figure out the recent political upheaval in Arab peninsula and Arab-Israel conflict, we shall point out the background of this conflict and its roots, particularly the post-Cold war area.

Additionally, this work will be structured like this, first; point out the Arab spring as recent incidents and its factors. The next, impact of Arab spring on the current relations of Arab-Israel strife, through study Arab public version toward Israel and readiness of Israel to live peacefully with some new Arab regimes around its border. Subsequently, the discussion will be about implicating of Arab-Israel issues on the security conditions in the region.

Political Upheaval and Its Causes in the Arab Worlds:-

Up to late 2010, the Arab world boasted a long list of some leaders, such as Qaddafi, who took charge of Libya in 1969; Saleh became president of Yemen in 1978; Mubarak took power of Egypt in 1981; Ben Ali rose to Tunisia's presidency in 1987, and the Assad family has ruled Syria since 1970. (Gause III 2011) These leaders had controlled all aspects of life in their own countries and have banned all political activities rather torturing people under different reason in order to maintain on power. (Gause III 2011)

Arab spring, however, has become a turning point in the history of the Arab countries, which was a revolutionary wave of protests and wars occurring that began on 18 December 2010, and characterized by civil uprisings in most of those countries and transferred to civil war later in some of them. Therefore, whether we like it or not the Arab peoples' aspirations for dignity and voice are satisfied, and how smoothly transitions to democracy proceed, are not just great moral questions they will also determine the region's stability and its economic forecasts for decades to come. (Dadush and Dunne 2011)

Since Bouazizi set himself on fire in Tunisian in December 2010, the flames that ended his life have spread across the entire Middle East. The protests and upheavals originally stimulated by his action have developed a variety of terms such as Arab spring, awakening or uprising, and they have defeated, threatened, or at least alarmed almost every ruling regime in the region. (Brym, Godbout et al.) The terms used to describe this event clearly indicate a sharp break from the decades of political stagnation and oppression there. (Brym, Godbout et al.) Political upheavals have also received by the US and EU when they express their support to the revolution in Tunisia and Egypt, Obama and European leaders have backed the protestors to overthrow of authoritarian leaders in favor of transitions to

democracy (Dadush and Dunne 2011) The political upheaval in the region has not come from the vacuum but has derived from some reasons, for example, economic reason. The economic problem has played a key role in rising unrest situation such as the financial weakness, poverty, high level of joblessness, low wages and rising food costs; a great proportion of redundancy was mainly dominant among the rising numbers of young people in those countries have experienced Arab spring. (Goldstone 2011) Tunisia had very difficult economic and financial issues to deal with; GDP growth was at about 1.5% during the protest, and there was a blueprint for rising to nearer to 4% in 2012. (Taylor 2011) In addition, although neoliberal economic policies affected all states in the Arab world, Tunisia and Egypt hold a special place in the hearts of those advocating economic reform. Egypt was the site of the first test of neoliberalism in the region, and the president of the World Bank once hailed Tunisia as the "best student ... in the region" when it came to economic restructuring. Both states, application of neoliberal policies exacerbated the divide between rich and poor, creating tensions that played out during the uprising. (Gelvin, J. L. 2015) These hard conditions have affected the Arab-Israel conflict as Israeli president referred poverty and oppression in the region have motivated resentment against Israel, he said, it better our neighbors will have it; we shall have better neighbors.(Press 2011)

Demography factor; according to the United Nations development program in 2009, the region's population more than doubled between 1975 and 2005 to 314 million, and in 2010, over 50% of the population was under 25, with large numbers living in poverty. However, economic growth is not keeping pace with population growth and so does not provide sufficient jobs for those entering the labor market. Thus, regional youth unemployment stands at 23%, nearly double the world average of 13%. As an example, unofficial estimates say that young people make up 83% of Egypt's unemployed. (Spring 2012) Furthermore, since 1990, the population aged between 15 and 29 has increased by 50% in Tunisia and Libya, 65% in Egypt and 125% in Yemen, making a substantial lack of employment opportunities and leading to mass dissatisfaction and disappointment toward their own government. (Goldstone 2011)

Moreover, corruption is another reason; both Egypt and Tunisia were enabled through the exclusion of the majority of the population from the economy. Family members of the ruling elite boasted their wealth in the streets of Tunis and Cairo as standards of living for the majority of the population stagnated. (Dodge 2011) Thus, there was an enormous proportion of corruption and lack of transparency, for instance, in Egypt Mubarak's family seemingly built up a wealth of between \$40 to \$70 billion, and 39 officials and businessmen close to them are alleged to have made fortunes averaging more than \$1 billion each.(Goldstone 2011)

Moreover, Arab spring has not expanded to all Arab countries, actuality; just six out of twenty-two Arab League member states have experienced genuine upheaval in the region, which are Egypt, Tunisia, Libya, Bahrain, Yemen and Syria that have experienced various levels of serious unrest. (Say, East et al.) The political turmoil in some of those countries have has a direct impact on Israel, especially Egypt and Syria.

Israel Faces the New Reality:-

Israel's relationship with the Arab world has taken a variety of forms ranging from the stated position between the executive and the policies that are trying to take advantage of the changes on the ground in the region to serve their interests. Altogether, it is possible to point out that the Israeli position of the Arab spring movement between the three stages: the stage of shock, at which point stunned by the Arab spring, Israel and there was an Israeli fear of serious changes in Egypt specifically. And the phase out of the shock, a stage that followed the outbreak of the Syrian revolution, and specifically where Israel graduated from the shock and to re-examine its strategic location in the region began and trying to understand what is going on around them. And phase adjustment, the current phase, which is trying to Israel, which benefits as much as possible from the preoccupation with the Arab world, the Interior for privacy and faltering Arab spring in order to achieve political and strategic interests were useful in the long run with this transition passage in the Arab world, and these interests the issue of Syria's Weapons Mass Destruction, weakening Hamas and Hezbollah, situation of Sinai peninsula, Iran and a political settlement with the Palestinians. The following discussion will shed light on these issues.

The root of Arab-Israel conflict has returned to a time before waging World War one and weakness Osman Empire, since that time British has thought to establish Jewish state in the Arab peninsula, moreover, this process has started practically after 1918, and by 1929, enough population had gathered for that state. (Fraser 1995) Israeli state declared formally on 14 May 1948, and then the ending British mandate over era declared, conflict had started since that time, and the First Arab-Israeli War occurred in 1948. (Fraser 1995)

Successively, the second war happened when conflict increased as Egypt closed the Straits of Tiran for Israeli shipping, and then Syria, Jordan, and Iraq had based an enormous number of troops along the Israeli boundary. After diplomatic efforts to solve the crisis failed, Israel attacked in June 1967 and occupied the Gaza Strip; the Sinai Desert from Egypt, the Golan Heights from Syria and the West Bank and East Jerusalem from Jordan, afterward; in 1973, and another war was an outbreak. (Conflict 2008)

The conflict was on going during the cold war, and after a collapse communist bloc, Israel has persisted on hostility against Palestinian. Apparently, since Hamas took control of Gaza in 2007, Israel has tried to surround and damage the Islamist movements with a mix of diplomatic isolation, economic pressure, and occasional military strikes, to terrorize Hamas, specifically, Israel also retains the threat of a more massive military response, such as the 2008-2009 "Cast Lead" operation, which generated more than 1,000 Palestinian deaths. (Byman 2011) Now we are moving on current relation between both side and impact of Arab spring on this relationship.

The positions of Israel on the Arab revolutions have divided into two separate tracks. The first is led by Israeli President Shimon Peres, who adopted a stand in favor of democratic change and called on decision makers in his country to understand what happened and understand it and maybe the initiative to take advantage of the atmosphere of openness to wave again its vision based on the call for the Middle Eastern economic and political market share of its population, according to the claim Peres, democratic values and openness that as long as Israel has claimed that it owned, unlike the Arab world. The second track, however, has the position adopted by Israeli Prime Minister Benjamin Netanyahu's exciting suspicion of what is happening in the Arab world, considering what is going on around its country in preparation for the control of Islamist forces believed that the most important projects are to pounce on Israel and destroyed. He, therefore, called for caution and urged the world to be alert to the danger coming after the Arab spring.

Binyamin Netanyahu has launched a scathing attack on the uprisings in the Middle East and North Africa, said that Arab countries moved not forward, but backward ... the Arab spring was becoming an "Islamic, anti-western, anti-liberal, anti-Israeli, undemocratic wave". (Harriet Sherwood 2011) Israel's preliminary reaction to the beginning of the Arab spring could be, frankly, depicted as one of the fears bordering on high anxiety, and some say such a response was not completely groundless. When popular protests were widespread all over the region, and probably with their inspiration, gatherings were held in the West Bank and Gaza Strip for putting an end to the split between Fatah and Hamas, which Fatah has controlled the West Bank, and the government led by Hamas, which controls Gaza since 2005. (Heller 2012) The political uprising in the region, particularly in Egypt, encourages Palestinians to avoid their leaders with the intention of end up the Israeli occupation and unification of the two parts of the country. The success of the revolution in Tunisia and Egypt in 2010 and 2011 respectively, led to breakdown the barrier among the public across the region. Even, the people in the West Bank and Gaza Strip did not resistant from this wave of people power flowing through the region, and demonstrated on the streets in solidarity people in their neighbors' countries. In Palestinian Authority the government tried to conquer the demonstrations dreading the effects of people power for their own governments. The most emphasis had given to the settlement and reconciliation between Hamas and Fateh. The Collapse of Mubarak in Egypt and public pressure on the street pushed the both party to enter in a negotiation talk.

Protesters were called to regulate relations between both parties and "national reconciliation" which was presented both as a national goal and a means of helping the end of Israeli rule in the West Bank.(Heller 2012) Because, after won parliamentary election in January 2006 by Hamas, with a good record of non-corruption in comparison to Fatah, with 74 seats. However, Israeli refused to accept a Hamas dominated government, as a result, became a split between both parties. (Winstone 2006) Nevertheless, initiating of reconciliation has made a real fear for Israel in the case of controlling West Bank by Hamas. These changes were making Israeli been concern about the political upheaval and following the developments in the Arab countries, because what was happening in the region directly related to their national security. The following graph shows the reaction of Israeli toward political upheaval in the region.

Graph (1): Mixed Reaction among Israelis toward the Arab Spring.

Mixed Reaction Among Israelis			
	Total	Arab	Jew
<i>Did you follow news about the political demonstrations?</i>	%	%	%
Yes	71	93	68
No	29	7	32
Don't know	0	0	0
<i>Were you excited about the demonstrations?*</i>			
Very excited	21	40	18
Somewhat excited	37	33	38
Not too excited/Not excited	41	26	44
Don't know	0	2	0
<i>Will the demonstrations lead to more democracy?</i>			
Yes	43	70	39
No	43	22	46
Don't know	14	8	15
*Asked of those who followed news about political demonstrations.			
PEW RESEARCH CENTER Q91, Q92, & Q93.			

Source: Pew Research Center

As it can be noticed from the above graph, Israelis are evenly divided about the legacy of the popular uprisings: 43% think the political demonstrations will expand democracy in the region, while the same percentage disagrees. However, Israeli Arabs are much more optimistic, with fully 70% believing the protests will lead to more democracy. Just 39% of Israeli Jews share this view. This means that they have seen the Arab spring as a new dynamic that promote more instability and conflict between Arabs and Jew.

In addition, in regard, Israel's neighbors the civil war that has waged in Syria, and Assad's rule have shaken and lost control in the country. Later, the Supreme Military Council that replaced Mubarak's government has promised to defend its foothold in the Gaza Strip from Israeli attack.(Heller 2012) In these situations, Hamas agreed to sign a "reconciliation agreement" with Fatah, which drafted by Egypt in 2009 and deferred by the Hamas leadership under pressure from Syria and Iran.(Heller 2012) This is led to make the uncertain situation for Israel.

Furthermore, Israel is concerned about Hamas, because for Israel, reaching a deal with the Palestinians would mean making territorial concessions. Many here note that after Israel withdrew from Gaza in 2005, Hamas won Palestinian parliamentary elections in 2006, and then held control over Gaza in 2007. The Muslim Brotherhood also gained power in Egypt for the first time and experienced a very short rule in the country, observers also say, it may be a matter of time before Hamas takes over the West Bank, thus coming Islamists to power and possess an inclination to uphold Hamas has made Israel felt a hazard and expected an unstable security. (KERSHNER 2011)

Thus, the Arab spring played a role in the signing of the reconciliation pact, and the document asserted a willingness to hold elections for the presidency and Legislative Council. (Heller 2012) Therefore, the probability of exploding clashes between Palestinian and Israeli security forces as a result of outdoor condition still foreseeable. Hamas activists would try to place themselves at the forefront of the first fights since Arab spring. Furthermore, Hamas has repeatedly proven its capability to incentive a war against Israel, which would make Israel respond militarily action against the civilian. (Heller 2012)

New conflict via demonstration, if it started, will not be a direct product of the Arab spring, because Palestinian people would be conversant with the popular uprisings against Israel, but renewed conflict would erode the chance of the diplomatic process to settle the conflict, this development would be added to the other consequences of the turmoil in the region.(Heller 2012) Israel, for its part, will be cautious and worry of any new territorial deployment

in the West Bank because of fears about elevated security threats as a result of what they called radicalization among the Palestinians and throughout the region.(Heller 2012)

Rising Islamist parties in Arab countries is another concern point for Israel, which it believes, they would limit the inclination of resumption of negotiations for promoting peace and stability, accordingly, all of these would reduce the capacity and the desire, of international players, chiefly the US and EU to put forward a new initiative for reopening of dialogue. Thus, elevating mass protest among the Palestinians, may lead into an all-out strife against the hostility of Israel.(Heller 2012) Thus, Israel alarmed both Palestinian parties should take seriously the peril of the lack of diplomatic dialogue for managing conflict.

This shows the probability that Israel will take hostility action against Hamas and jihadist organizations in Gaza as occurred in November 2012, when Israel tanks shelling the area in response of Hamas operation, which generated four civilian casualties in Israel.(Ramadan 2012) In November, 14, 2012, the seven-day-long Gaza operation of Israel which called " Pillar of Defense" started; however, the changing balance in the region, the involvement of US administration and Islamic countries, including Egypt in the matter and the ability of Hamas to do damage in Tel Aviv due to its new missile's ability have all prevented a great catastrophe and the casualties in the recent Israeli military operation have remained relatively limited by 145 deaths from Palestinian compared to the level of destruction and number of casualties in the Operation Cast Lead in 2008.(BİLİCİ 2012)

This operation has some motivated, such as to influence US foreign policy after the re-election of Obama in 2012, an assessment of the Arab reaction after Arab spring and challenging Egyptian government, however, Egypt condemned the attack and withdrew its ambassador from Tel Aviv, and sent its foreign minister to Gaza.(Sayed 2012) Moreover, Israel considers the new political reality in Egypt as one of the most significant challenges facing Israel, due to the possibility of helping Hamas in Gaza. (Heller 2012) When the head of the Higher Military Council was vividly alarmed that "the threat against the Palestinian government has enraged Arab nations, which may augur disastrous results".(Levy 2012) Under Sisi authority also, regardless of its objective, any military operation in Gaza would inevitably prompt a harsh response in Egyptian public opinion and a demand to react against Israel. In anticipation of such a response, Israel will likely deny the relative freedom of action it enjoyed during Operation Cast Lead, instead, Israel will probably go to carry out a rapid campaign.(Heller 2012) Israel also put up security fences on the border with Egypt and Jordan, increasing the security budget and abstaining from any gesture toward the Palestinians have been only some of the steps taken.(Ravid 2011).

Furthermore, the Egypt-Israeli relationship could consider also in providing security of Sinai Peninsula and the Israeli-Egyptian border, recently despite an increased troop presence there; the Egyptian government apparently seems unable to control the area, and jihadist organizations are increasingly free to operate there.(Heller 2012) Islamist militants frequently attacking Egyptian military in North Sinai province and wounding more in an attack on a security checkpoint and military bases, and the Islamic State in Iraq and Syria (ISIS) group's Egyptian branch in the Sinai Peninsula claimed responsibility for the most incidents in the area. (Al Jazeera 2017). Apparently, this is a severe concern for the Israeli national security. The Islamic State-linked terrorist groups in the Peninsula have threatened Israel and Jews among its belligerent messages, as mentioned "Oh Jews, wait for us. The punishment [we have prepared for you] is severe and soon you will pay a high price." (JPOST 2016) Therefore, security officials in Israel have kept a close eye on their border with Egypt, concerned about possible attacks from Sinai Province. As the Israel Defence Forces (IDF) mentioned that "the enemy is Daesh ... we believe that these terror cells will try to hit our forces in order to prove themselves." (Derek Stoffel 2016)

The Islamic state from Sinai Province has launched several rocket attacks against Israeli territory in 2016. IDF declared that terrorist cell attacked an army patrol along the security fence that marks the Israeli-Egyptian border; the force was able to respond and to hit back at those who tried to infiltrate the border into Israel. (Derek Stoffel 2016)

Consequently, the situation along the Egyptian border, which includes the southern border with Gaza, is even more sensitive. Post-Mubarak period there were serious incidents along the Sinai border, leading to a crisis in relations. Gunmen regularly crossed the Sinai border into Israel and killed several Israelis. Israeli security forces then gave chase and killed some of the gunmen and also, Israel blamed Egypt for failing to control the Sinai. (Taylor 2011) Therefore, Israel believed that shifting of power in Egypt and instability in Jordan could have profound consequences for Israel, which depends on the peace accords, they were only two Arab countries, which regarded as

a cornerstone of Israeli security, And so, Israeli government claimed instability in Egypt requires Israel to prepare the response to a more extreme scenario in which the Egyptian army moves into Sinai in breach of the peace treaty. (Heller 2012)

Moreover, in Jordan, there is discontent on the Jordanian street, thus, Israel has worried about its border with Jordan, which thinks that it is largely unsecured. Therefore, Israel has trying to prepare for reaction; because it has already a peace agreement with Jordan so breaking that treaty will make Israel take aggressive action. (Heller 2012) Similarly, "The Daily Beast" America magazine reported that the US sources have confirmed the keenness of "Israel" for military intervention in Jordan, the event that a military attack by the fighters of ISIS. (Eli Lake 2014)

In addition, The Golan Heights security, it is vague where the unrest in Syria will end, but the current situation is likely to end at some point in political change. Assad also has supported Hamas and Hezbollah which they rejected peace or at least has not embraced negotiations and is a close friend of Iran.(Byman 2011) Still, Israel's artillery has fired back at Syria after gunfire hit an Israeli military vehicle in the Golan Heights. The incident comes amid high tensions in the region as Israel is waging an offensive against Gaza while civil war continues in Syria. Moreover, International community is really a concern about Syrian military use of chemical weapons against civilian and which they are waiting for final orders from Assad, as the US has officials reported. And Netanyahu says Israel, and the international community is watchfully observing developments in Syria regarding its stores of chemical weapons. (Benari 2012)

Furthermore, after rising the Islamic States in Iraq and Syria (ISIS) in Syria, the security threats of this terrorist group have increased to Israel. The Islamic States has released a video which threatens the Israeli occupation forces, and it appeared fighters, speaking in Hebrew, directed a message to "the Jews." The broadcast terrorist organization video under the name "break boundaries and slaughter the Jews" began to talk about the Jews and how they arrived in Palestine, and how they had occupied, and initiated some of the verses and the hadith related to the presence of Jews in the land of the Arabian Peninsula, as well as in the Levant.

Therefore, Arab uprising targeted Bin Ali in Tunisia and Mubarak in Egypt were the central pillars of a regime holding a critically important Arab state to a political course that completed Israel's vital security interests.(Heller 2012) Mubarak was scarcely the Zionist supporter, for instance; he did not allow anti-Israel and American movements and even propaganda in Egypt; Egyptian media were not only prohibited to critics Israel's hostility but also to speculate about the state of the President's health.(Heller 2012) Thus, from US standpoint, what Roosevelt said about Anastasio Somoza, could have said with equal validity about Mubarak: "He may be a son of a bitch, but he's our son of a bitch," because his significance service to Israel and American, his overthrow in the American circle necessarily augured badly for the US and Israel's interests.(Heller 2012) Mubarak has been a fixture of Israel's geostrategic landscape for three decades. Israel elected eight prime ministers, conducted several wars and engaged in peace talks with multiple partners, and Mubarak was always there. He personified regional stability.(Benn 2011)

Under Mubarak's rule, Egypt also kept the Rafah crossing point between Egypt and Gaza closed, helping Israel restrict the flow of goods and people into and out of Gaza.(Byman 2011) Therefore, his policy was based consistently on interpreting Egypt's national interest necessity for maintenance of the Israeli-Egyptian Peace Treaty, fight off Islamist movements, active suppression of what he called extremists, and close coordination with the U.S.(Heller 2012) However, Israel treaties with Egypt in 1979 and Jordan in 1994 remain unpopular among public, particularly during the Arab spring and in post-Mubarak era, while Israel was relied on the strength of Mubarak's regime and the Jordanian monarchy to keep them intact, but collapsing Mubarak increased the possibility of rising protest in Jordan led to Israeli disappointed. (Zacharia 2011) In consequence, Netanyahu expressed his anxious and asked the international community to oblige "any Egyptian government preserves the peace accord with Israel".(Zacharia 2011) Due to; a political leader of Egypt's Muslim Brotherhood had called on any government that replaces Mubarak's regime to withdraw from the 32-year-old peace treaty with Israel.(Lake 2011)

Nevertheless, Dan Gillerman, a former Israeli ambassador to the UN in respond to some threat by Muslim Brotherhood said, "if they do it, it will be a very unfortunate development for Egypt and Israel, both countries have enjoyed a peace for thirty years, undoubtedly not a very warm peace, but a cold peace is better than a hot war". (Lake 2011) Moreover, the revolution in Egypt and unrest elsewhere in the Arab world has shaken Arab leader loyalty for Israel, nevertheless, sympathy for Hamas in Egypt is getting high, and Hamas' resistance to Israel is also popular, already the new Egyptian government declared it will open Rafah and no longer collaborate with the

economic isolation of Gaza.(Byman 2011) Likewise, Cairo (Under Muslim Brotherhood Rules) had declared opening Rafah for putting an end to Palestinian suffering and breaking long term siege. Egyptian Foreign Minister has also added that "Egyptian national security and Palestinian security are one".(Levy 2012) Subsequently; the Muslim Brotherhood also called on the government to prevent the continuing supply of gas to Israel out of "solidarity with the Palestinian people besieged in the West Bank and Gaza".(Levy 2012)

Afterwards, the experienced of Arab spring by Libya, Yemen and Syria seemed uncomfortable for Israel because of weakening and overthrowing those regimes were hospitable to Israeli interests, thus, Israeli concerns were further intensified when it began to appear as though the forces most likely to benefit from and exploit the fall of established regimes were not the secular.(Heller 2012) Netanyahu also warned, "if extreme elements are allowed to exploit democratic processes to attain power and promote anti-democratic goals, as happened in Iran and Egypt under Morsi and elsewhere, the result will be harm to peace, and harm to democracy".(Zacharia 2011)

For that reason, the real fears among Israel's are that a new Arab government has might be dominated by Islamists, would it break the peace treaty between them and Israel? Would it seek to go to war again?(Miller 2011) In response, the spokesman for the Brotherhood asserted that they "would respect the peace treaty with Israel as long as Israel shows real progress on improving a lot of the Palestinians." (Lake 2011) However, the Israeli government regards its main approach to the Arab Spring is that Israel must prepare itself to deal with and respond whatever the upshots might be. Additionally, the US has illustrated its compulsion of the Israeli security and asked Israel to work for peace with new actors in the region, when US administration said, "as for Israel, our friendship is rooted deeply in a shared history and shared values; our commitment to Israel's security is unshakeable...It's important that we tell the truth: The status quo is unsustainable and Israel too must act boldly to advance a lasting peace."(President Barack Obama speech, Washington, D.C., May 19, 2011) Therefore, supporting Israel's security and pursuing Arab-Israeli peace by the US, require the US efforts to reshape the structure of the region in order to fulfill its own interests there.(Terrill 2011)

Also, the former US peace negotiator A. D. Miller contends "the irony is that the challenges a new Egypt would pose to America, and Israel would not come from the worst-case scenarios imagined by frantic policymakers and intelligence analysts an extremist Muslim takeover, an abrogation of peace treaties, the closing of the Suez Canal, nevertheless, from the very values of participatory government and free speech that free societies so cherish."(Byman 2011) Because, there's no doubt, a new Egyptian government and president, more responsive to public opinion, indeed, legitimized by the public in free elections, will be, by necessity or inclination, far more critical of Israeli actions and policies and far less likely to give Israel the benefit of any doubts.(Miller 2011) Moreover, Netanyahu has highlighted his fears in public, particularly the threat of Egypt turning into a new Iran. Nonetheless, yet a more optimistic analysis suggests that the Israeli government could be leverage the Egyptian crisis to seek new opportunities, a window to restart the peace process with the Palestinians and try to support the spread of democracy in the region.(Benn 2011) Seemingly, the current government would not want to withdraw from the peace treaty with Israel; nobody in Egypt wants war with Israel and there is every sign right now that the military will still play a strong role in the next. (Miller2011) In sum, the Arab Spring has not only shaken some dictator regimes in the region, but also threatened Israeli security. The consequences of the political uprising have made the situation much complicated, particularly after rising jihadist terrorists and hijacking the demands of people.

Conclusion:-

The Arab Spring has changed the structure of the regime and pushed Arab world toward semi - democracy, human dignity, transparency and so on. Several factors have played the role in such political and social upheaval such as economic, demography. Furthermore, political upheaval has brought Islamist to power (even for a short period), that creates concern for Israel and pushed Telavi to test the first operation on Gaza after Arab spring, which made a real concern for the regional and international security. The Arab Spring also has strengthened the conflict between Israel and some other Arab countries, when the peace treaty between those countries and Israel threatened. However, the question is remains is that whether future war between Arab and Israel will be limited between Israel and Palestinian or will engage other Arab countries and (jihadist groups like ISIS), specifically those countries that have experienced the Arab Spring?

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RESEARCH ARTICLE

ANALYSIS OF METEOROLOGICAL DROUGHT IN IRAQ USING THE RECONNAISSANCE DROUGHT INDEX (RDI).

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Abstract

The drought characteristics were analyzed in Iraq dependent on the observed climate data from ten meteorological stations during 1980-2011. Reconnaissance Drought Index (RDI) was used to studying and analyzing of droughts. The variable of interest in RDI is precipitation over potential evapotranspiration (ET_o), the ET_o is calculated using Penman-Monteith method. The results showed that the 2008 year was the worst dry year had passed in all parts of Iraq. It was observed that the maximum severity of drought (S_{max}) was identified during the periods (2007-2009) for the most stations. The Mild drought (Extremely drought) conditions are the highest (lowest) number of drought event than other conditions for all stations. Most of study region experienced the strongest drought events through the last period from 1999 to 2009 as compared with the previous period.

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Introduction:-

In the last decades, drought has become a frequent phenomenon in many parts of the world, which in turn affects heavily on agriculture and water sources [1]. Analysis and monitoring of drought can be done with drought indices such as the Palmer Drought Severity Index (PDSI) widely used in the United States [2], the decile index operational in Australia [3], Z-Index used by the National Metrological Center of China [4], the Standardized Precipitation Index (SPI) [5], Reconnaissance Drought Index (RDI) [6], and the Standardized Precipitation Evapotranspiration Index (SPEI) [7]. The climate of Iraq classified as dry and semi-dry, so the impact of evapotranspiration on the drought considers a significant impact. Therefore, the Reconnaissance Drought Index (RDI) was used among these indices in this study, which is based on both precipitation and potential evapotranspiration.

Several studies on drought assessment have been carried out in different parts of the world can be summarized as follows: [8] attempted an assessment and monitoring of regional drought using two well-known indices, namely, SPI and RDI, during the period 1971-2008 in Cyprus. They found that both indices had been given a similar form in analyze drought conditions and monitor drought events. [9] used the SPI to evaluate the spatial and temporal characteristics of meteorological drought for the period 1980 to 2010 in Iraq. They showed that the worst dry year was observed at 2008. 30%, 36%, 22% and 12% of the area were classified under extreme, severe, moderate and near normal drought, respectively. [10] investigated the similarities or differences in SPI and RDI indices using data from a variety of climatic zones in Iran. Although their results showed that both indices had the same behavior, RDI index depends on ET was more sensitive to climatic variability than SPI index. The Iraq region is also prone to

drought. Nevertheless, the drought condition and its characteristics in Iraq has received limited attention. [11] calculated the SPI for 12-month using rainfall data records observed from 1941 to 2002 in the north of Iraq. He found that the percentage of wet (drought) conditions were up to 44 % (56%) of the study period. He declared that moderate wet and drought conditions were more common than others. [12] used the SPI method for 9, 12-month time scales. The results showed that the Northern of Iraq exposed to the severe drought for SPI 9- month during the study period as compared with other parts of the region. Whilst, the Baghdad, and Diwaniya regions experienced the highest degree of drought according to SPI 12-month.

The main aim of this paper is to analyze the drought characteristics using Reconnaissance Drought Index (RDI 9-month) for ten stations in Iraq. A MATLAB software codes were written by M-File Programming to calculate the potential evapotranspiration and drought characteristics.

Materials and Methods:-

Study area and Meteorological data:-

Iraq region is located in southwest Asia at the crossroads of the Middle East. It covers an area of 435,052 km², which lies between the latitudes of 29° 5' and 37° 22' north and the longitudes of 38° 45' and 48° 45' east. Historical records of monthly precipitation, temperature, humidity, wind speed, and sunshine data from 1980 to 2011 were obtained from the Iraqi Meteorological Organization and Seismology (IMOS) for ten stations. The location of the Meteorological stations are presented as shown in Fig.1.

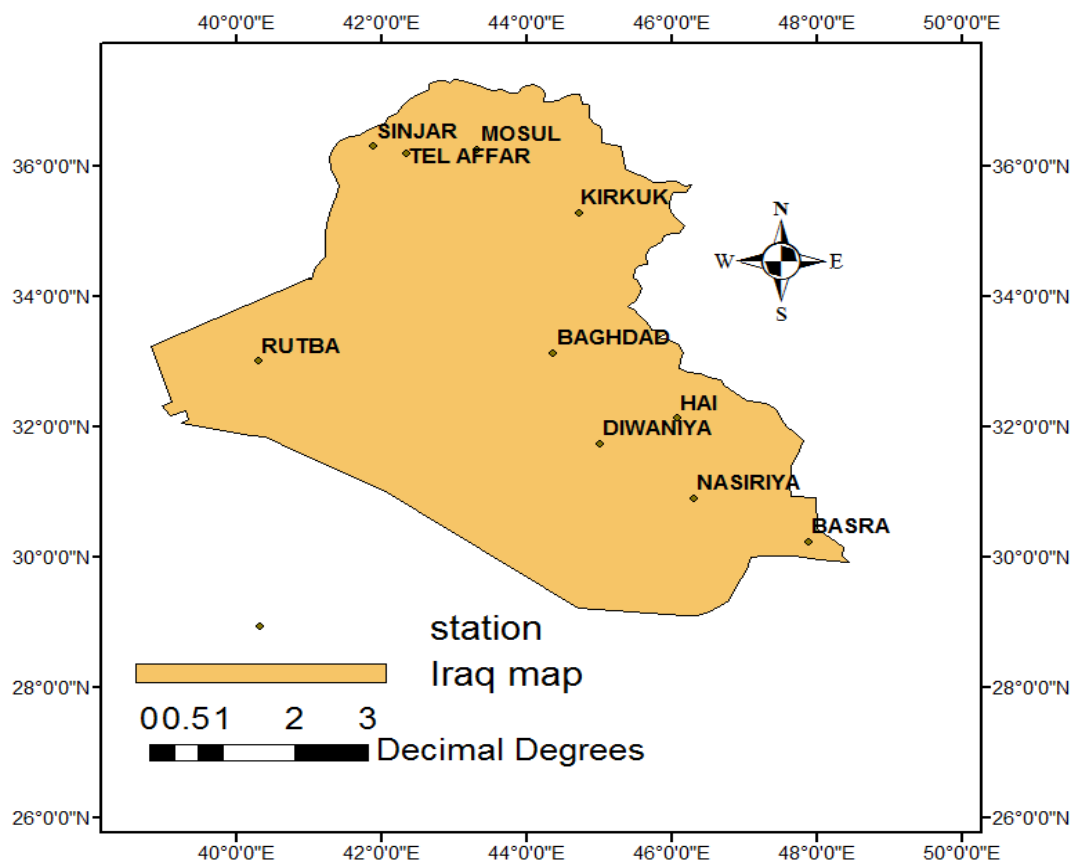


Fig.1:- locations of meteorological stations in Iraq

Methodology:-

Reconnaissance Drought Index (RDI):-

The Reconnaissance Drought Index is based on the ratios of precipitation over potential evapotranspiration values for different time scales [13]. The potential evapotranspiration is computed by the Penman- Monteith formula. Initially, α_k is presented as the coefficient of the i th year in an aggregated form using a monthly time step and can be calculated on a monthly, seasonal or annual basis as following.

$$\alpha_k^{(i)} = \frac{\sum_{j=1}^k P_{ij}}{\sum_{j=1}^k ET_{ij}}, \quad i = 1:N \quad \text{and} \quad j = 1:k \quad (1)$$

Where P_{ij} is precipitation, ET_{ij} is potential evapotranspiration in j^{th} month of i^{th} year.

The next step, the Normalized RDI (RDIn) is calculated, utilizing $\bar{\alpha}_0$ is the arithmetic mean of α_0 values calculated for the N years of data.

$$RDI_n^i = \frac{\alpha_0^i}{\bar{\alpha}_0} - 1 \quad (2)$$

After that, the Standardized RDI (RDIST) is calculated, using the following equation for each year.

$$RDI_{st}^i = \frac{y^i - \bar{y}}{\hat{\sigma}_y} \quad (3)$$

Where y_i is the $\ln(\alpha_0^{(i)})$, \bar{y} its arithmetic mean and $\hat{\sigma}_y$ is its standard deviation. In this study RDI is applied to represent RDIST. Drought category classification suggested for the RDI [13] as illustrated in Table-1.

Table 1:- Drought classification for RDI value and corresponding event probabilities.

RDI	Classification	Probability (%)
0 to -0.99	Mild drought (Mi.D)	34.1
-1 to -1.49	Moderately drought (Mo.D)	9.2
-1.5 to -1.99	Very drought (S.D)	4.4
-2<	Extremely drought (E.D)	2.3

Calculation of potential evapotranspiration:-

The Penman - Monteith formal is one of the most common used ones, defined as equation (4), recommended by the FAO in 1998 as reference ET, depended on observed meteorological data, such as the temperature, relative humidity, sunshine hours, and wind speed. Many studies found that the penman-monteith is more appropriate for arid and semi-arid regions [14];[15];[16]. In present study, total potential evapotranspiration (ET0) has been estimated for the study stations using Penman – Monteith equation [17] as follows

$$ET_0 = \frac{0.408\Delta(R_n - G) + \gamma \frac{900}{T + 273} U_2 (e_s - e_a)}{\Delta + \gamma(1 + 0.34U_2)} \quad (4)$$

Where ET0 is the potential evapotranspiration rate (mm d⁻¹), and the monthly value of ET0 will be used in this paper, G is the soil heat flux density (MJ m⁻² day⁻¹); T is the mean daily air temperature at 2 m height (°C); U_2 is the wind speed at 2 m height (m s⁻¹); e_d is the saturation vapor pressure (KPa); e_a is the actual vapor pressure (KPa); Δ is slope of vapor pressure curve (KPa 0C-1); γ is the psychrometric constant (KPa 0C-1); R_n is the net radiation at the crop surface (MJ m-2 day-1).

Results and Discussion:-

Drought characteristics for the RDI from 1980 to 2011 periods were presented in Table-1. It can observe from this table, the value of the maximum severity of drought (S_{max}) was identified during the periods (2007-2009) for the most stations. The highest value of S_{max} was found at Kirkuk station, and had the longest duration drought, began in Jan-2007 and ended in Oct-2009, lasted 34 months. The maximum value of the severity of peak drought (S_p) was observed in BAGHDAD station, occurred in Nov-1990 and relative frequency of drought during the whole periods is 43.4 %. While the relative frequency (RF) for all stations ranged between 39.6% - 52.1%.

As shown in Fig.2, the longer duration of drought for Mosul was observed between Sep-1998- Jul-2002 (47months), with the intensity of 1.3. The years in 1998, 1999, 2000, 2007, 2008, and 2009 were the worst drought years as compared with other years. Baghdad (Rutba) station exposed to worst dry through the years 1986, 1987, 1997, 1999, 2007, 2008, and 2009 (1983, 1984, 2001, 2004,2005,2006 and 2007), experienced the longer duration

of drought between Dec 1998- Jan2001 (Jan2004-Jun2005), with intensity 1(1.5). In Basra station, the longer duration of drought was determined between Jan2010- Dec2011. It exposed to worst dry years in 1994, 2003, 2008, 2010, and 2011.

Table 2:- Drought characteristics for the RDI 9-month time scale from 1980 to 2011 periods

Station	The severity of peak drought (Sp)	The relative frequencies (RF)%	The minimum severity of drought (Smin)	The maximum severity of drought (Smax)
MOSUL	-2.65 / (April 2008)	44.95	-0.01 / (May.1997)	-58.80 / (Sept. 1998 –July 2002)
SINJAR	-2.80 / (Nov. 2005)	45.21	-0.001 / (Aug. 1986)	-40.11 / (Nov. 2007 –Dec. 2009)
TEL AFFAR	-2.71 / (Oct. 2000)	39.63	-0.01 / (Apr. 1992)	-47.73 / (Sept.1998- April 2001)
KIRKUK	-3.09 / (Nov. 1999)	42.82	-0.02 / (Apr. 1992)	-52.55 / (Jan. 2007-Oct. 2009)
BAGHDAD	-3.81 / (Nov. 1999)	43.35	-0.16 / (Jan. 2003)	-35.80 / (Dec. 2007-Oct.2009)
RUTBA	-3.78 / (Sept. 2004)	44.15	-0.01 / (Sept. 1986)	-28.74 / (Jan. 2004-July 2005)
HAI	-3.63 / (Dec. 2003)	47.34	-0.001 / (Sept. 2010)	-45.31 (Jan. 2003-Dec.2004)
DIWANIYA	-3.01 / (Dec. 1998)	52.13	-0.001 (Oct. 2000)	-31.42 / (Jan. 2007-Nov.2009)
NASIRIYA	-3.07 / (Dec. 1990)	50.00	-0.06 / (Mar. 1991)	-29.04 / (Dec. 2007-Sept. 2010)
BASRA	-3.48 / (Dec. 1998)	42.55	-0.001 (Sept. 1995)	-31.86 (Jan.2008-Nov.2009)

Table-3 summarize the frequency of each wet / drought conditions for all stations. The results showed that the Mild wet / drought (Extremely wet /drought) conditions are the highest (lowest) number of wet / drought event than other conditions for all stations. In drought condition, the highest (lowest) number of drought events occurred in Diwaniya and Nasiriya (Basra, Kirkuk, and Baghdad) (the number of drought events between 188-196 (160-163) times). Sinjar (Tel Affar) recorded the highest numbers of moderately (severely) drought events, while the highest numbers of extremely drought events existed in the Mosul and Tel Affar.

Table-3 the numbers of wet / drought event results for the RDI 9-months.

Station	E.W	S.W	Mo.W	Mi.W	Mi.D	Mo.D	S.D	E.D
MOSUL	4	15	32	156	112	20	19	18
SINJAR	2	16	33	155	101	36	22	11
TEL AFFAR	7	6	28	186	85	16	29	19
KIRKUK	1	12	44	158	104	23	17	17
BAGHDAD	1	11	45	156	101	29	23	10
RUTBA	9	13	23	165	111	27	14	14
HAI	0	14	47	137	117	35	14	12
DIWANIYA	3	24	41	112	144	28	15	9
NASIRIYA	0	20	41	127	136	23	16	13
BASRA	5	14	31	166	102	26	16	16

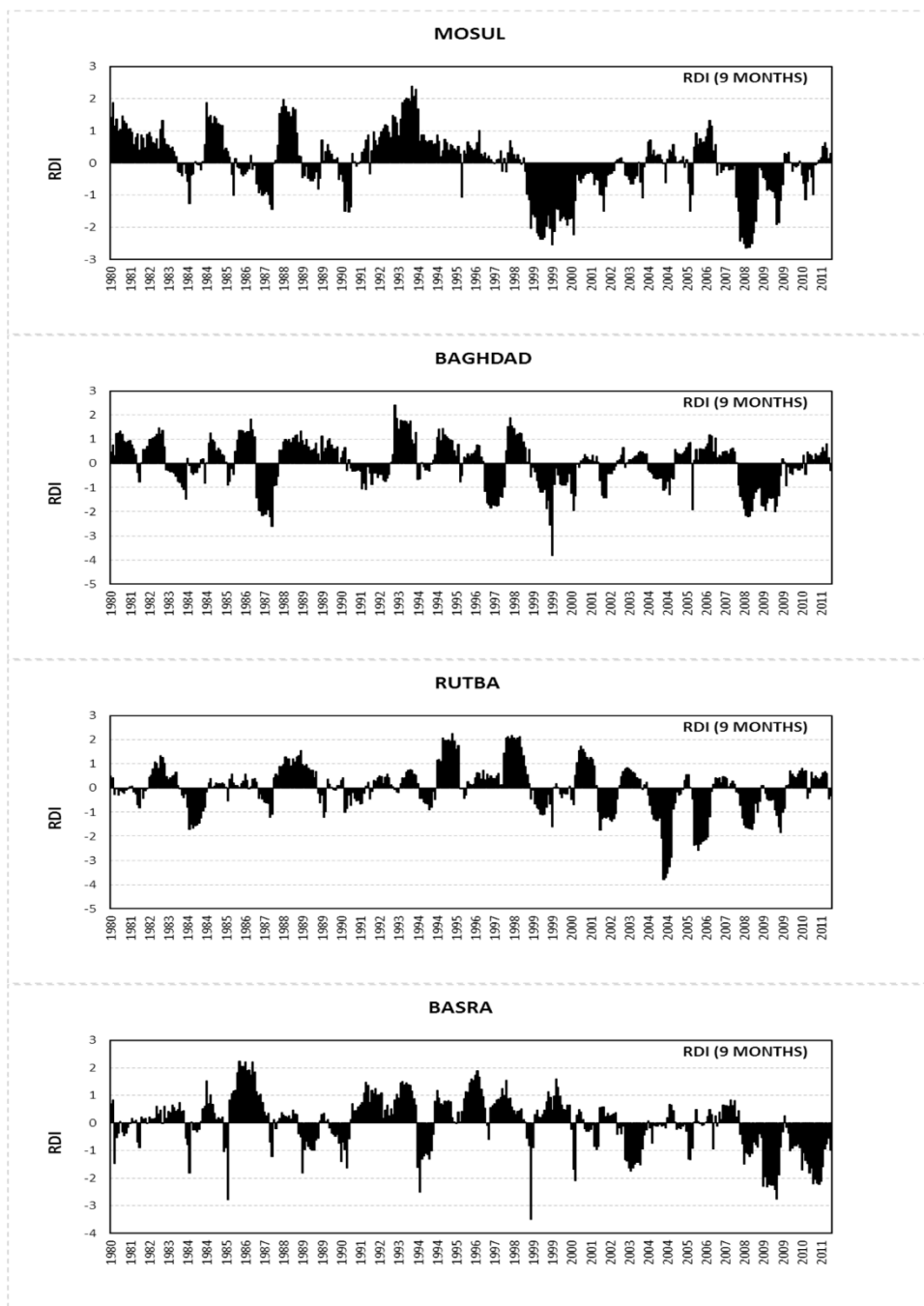


Fig 2:- The time series of RDI for some stations.

The duration of period (30) years was divided into three period each period represents by ten years. The percentage of drought events at three periods for all stations was calculated as shown in Fig.3. It was observed that the last period had the maximum percentage of drought events for all stations as compared with the previous periods except three stations. Indeed, the Kirkuk and Basra experienced the higher value of drought event at the first period, while the Nasirya station exposed to the stronger drought at second period.

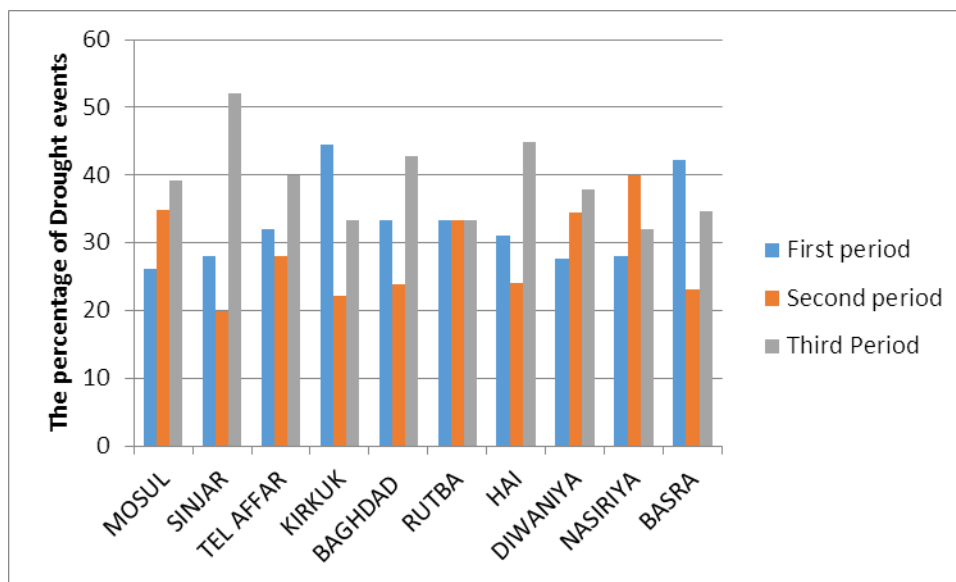


Fig 3:- the percentage of drought events for all stations at three periods.

The RDI characteristics were used to generate drought maps for Iraq region using GIS software as shown in Fig.3. The spatial pattern of the average duration of drought (D_{avg}) shows an increasing trend from eastern part toward southwestern of the Iraq region, ranged from 2.62 to 10. The spatial distribution of the average severity of drought (S_{avg}) ranged from -2.12 to -7.87. The highest value of the average severity of drought (S_{avg}) dominated in the central Iraq decreases toward northeastern, and southwest of the study area. It can be seen from figure the Baghdad and Nasirya regions experienced the highest magnitude droughts and had the longer durations as compared with other regions.

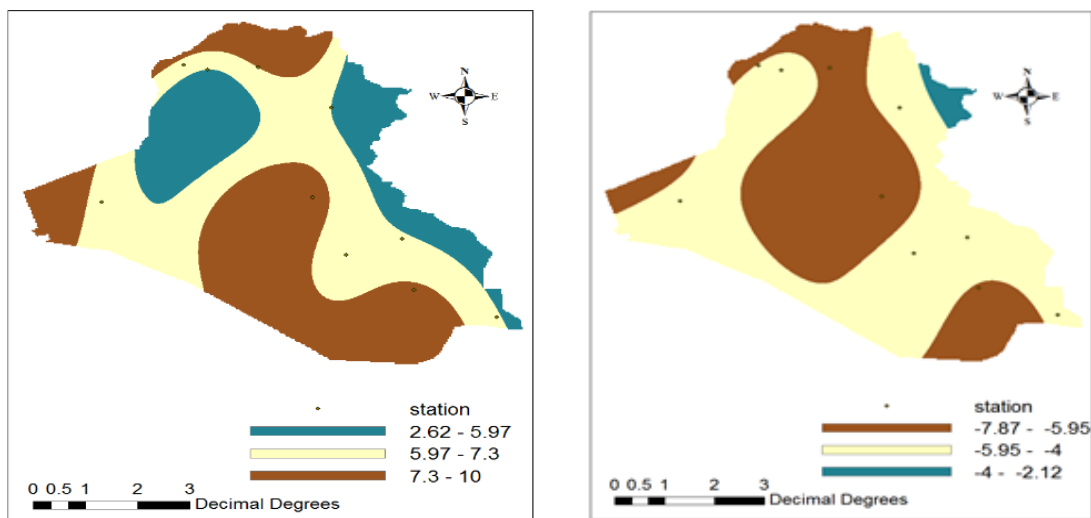


Fig 4:- The D_{avg} and S_{avg} drought using RDI.

Conclusion:-

The aim of this study is to analyze the drought characteristics in Iraq region. The Reconnaissance Drought Index (RDI) was used to analyze the drought based on the historical data for ten meteorological stations during the years from 1980 to 2011. The results showed that the 2008 year was the worst drought years had passed in all parts of Iraq, which was consistent with the finding of [9] and [12]. It was found that the frequency of each wet / drought conditions indicated that the Mild wet / drought (Extremely wet /drought) conditions are the highest (lowest) number of wet / drought event than other conditions for all stations. Also, most study regions were exposed to the highest drought events during the period from 1999 to 2009. As spatial pattern results, the Baghdad and Nasirya regions experienced the highest magnitude droughts and had the longer durations as compared with other regions through the study period.

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RESEARCH ARTICLE

QUALITY OF HEALTH SERVICE AND ITS EFFECT ON PATIENT SATISFACTION IN DR. SYAIFUL ANWAR HOSPITAL OF MALANG INDONESIA.

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Abstract

Service quality dimension covering reliability, responsiveness, assurance, empathy, and tangible is the ultimate goal a public hospital should achieve as organization productivity through patients' satisfaction. This study investigated the influence of service quality on patients' satisfaction of Dr Syaiful Anwar Hospital in Malang, East Java, Indonesia. Assigning 50 patients hospitalized during the research, this study collected data using questionnaires, interview and document analysis and used SPSS 17.0 as a tool of analysis. Findings of t-test showed that variables of quality service gave positive effect on patients' satisfaction, each of which is: Assurance (46%), Reliability (34,3%), Tangible (42%), Responsiveness (67,1%), and Empathy (43,3%). Determinant test showed all variables affected 46.1% ($R^2 = .464$) on public satisfaction and responsiveness performed the most dominant effect on patients' satisfaction to 67.1% based on beta test.

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Introduction:-

Satisfaction is feeling happy or disappointed someone has after comparing a perception or impression of the performance of a product and expectations. Satisfaction is a function of perception or impression of performance and expectations (Kotler, 2002). The quality of health care services to patients can not be separated from the employee's role as a provider of services to patients. Management commitment to quality of service is the first step process of providing excellent service to patients. Likewise, employees' commitment to the quality of service is standard, it is the continuous effort of the employee to provide service beyond the minimum standards set. Employee satisfaction to their job will be a positive influence on the quality of services received by users.

The fast development of human resource management that regarded nurses or clerks as a burden or cost, is currently changing the value to asset health. Therefore, almost all large and small health institutions will make every effort to develop human resources as much as possible through improving the quality and performance of the nurses to provide job satisfaction for employees. In conditions of fierce competition, each hospital attracts and retains patients, as well to apply right strategy to achieve patient's satisfaction. This means that no matter how small the patient's complaint, the hospital must respond positively, because the handling of complaints could provide an opportunity to turn disgruntled customer into a satisfied customer or even become immortal customer (Schnaars, 1991). According Artanti (2015), the impacts of dissatisfied customers vary: 90% of dissatisfied customers will not return to the hospital; a customer that is unsatisfied will tell at least to 9 other friends or relatives; a customer that is satisfied with the services will tell to others and direct them into customers; 13% of customers who were disappointed will tell to

more than 20 people; the cost to recruit new customers requires five times higher than if we maintain relationships with existing customers.

Dr. Saiful Anwar Public Hospital Malang is one of the spearheads of local government providing public health service. Management makes improvements for the sake of reform, particularly to improve service quality through evaluating the standard of service and formulation of policy services. By doing so, the hospital can make improvements on service quality and give satisfaction to public users, especially the patients.

To determine the patient's perception of service quality and satisfaction an empirical study is required. Evaluations carried out in general are not enough for management of the hospital, because the information can not be generated for all patients in the hospital. For the sake of the future strategic decisions, only research activities are appropriate to provide answers.

The main problem of the hospital as an institution of health services that challenges many competitors is whether its service has been devoted according to the expectations of patients or not? As local hospital, Dr. Saiful Anwar Hospital should always maintain confidence and satisfaction of patients as customers through improving quality of its services to increase patient satisfaction. Dr. Saiful Anwar Hospital needs to determine carefully the needs of patients as an effort to meet the desires and improve satisfaction services provided. Building relationships and conduct a study of patients are required to adjust services as expected. This is the concept of customer focus. The creation of quality of service will surely create user satisfaction with the service. The implication would be able to provide many benefits, including establishment of a harmonious relationship between providers of goods and services to customers, provide a good foundation for the creation of customer loyalty and make the patient as an agent of promotion.

The hospital has a system of health care standards set by the government, that is a standard service-oriented to customer satisfaction and a comprehensive standard for enterprise services. One effort of the hospital to win the competition is to improve the quality of products or its services. Therefore, hospitals are trying to win competition by improving the quality of products and services, so as to provide customer satisfaction.

In the future, a hospital should have a goal of achieving ISO certification. ISO provides advantages, such as: a better reputation, level of awareness of need to maintain quality, apparent procedures and responsibilities, better documented, eliminating unnecessary work, easier to track and audit, better service to customers, improved customer satisfaction and employee, performing continuous improvement, increasing profits, opportunities for expansion and change the bad image of society to government health services better. Accordingly, implementation of quality management system (ISO 9001:2000) makes service better, all activities are carried out procedurally, documented and regularly and an analysis of previous activities is possibly conducted.

The first step is to know what quality dimensions that influences patient satisfaction, especially patients in the hospital. According to Zeithaml, Berry and Parasuraman (2012) dimensions of quality include: reliability, responsiveness, assurance, empathy, and tangible (real) to qualify customers' satisfaction. Appropriate service policies of patient care should be examined through dimensions of quality services of hospital patients. The problem is whether there is an effect of the quality dimension variables. Thus, orientation of this study is to prove whether there is influence quality of service to client satisfaction based on the dimensions of reliability, responsiveness, assurance, empathy, and tangible at the hospital, to identify dimensions of service quality that gives the most influence on patient satisfaction.

Another problem is how much the influence of dimensions of service quality that consists of reliability, responsiveness, assurance, empathy and tangible to the satisfaction of patients. This study was done to contribute ideas to the management of health care providers to improve services to patients and to give consideration in decision making about the relationship improved service performance at the hospital. The so complex problems of life today that cause health problems makes health care is really critical.

Research Problems:-

The formulation of the problem of this research are:

- a. How are descriptions of quality activities of health service and patient satisfaction in the hospital of Dr Saiful Anwar Malang?

- b. How does dimension quality of health services that include reliability, responsiveness, assurance, empathy, tangible affect partially level of patient satisfaction in the hospital of Dr Saiful Anwar Malang?
- c. How does dimension quality of health services that include reliability, responsiveness, assurance, empathy, tangible affect simultaneously patient satisfaction in the hospital of Dr Saiful Anwar Malangd.
- d. What is dimension quality that affect the dominant influence on patient satisfaction of Dr Saiful Anwar Hospital in Malang?

Review of Literature:-

Quality:-

Quality is a zero defect, completeness and conformance to requirements. Quality is conformance to specifications, when viewed from perspective of manufacturer. Juran, (in Yamit, 2005:337) defines quality as a standard specifically where the ability (availability), performance, reliability, ease of maintenance (maintainability) and its characteristics can be measured. Davis, (in Yamit, 2005:8) makes the definition of quality broader in scope, as a dynamic condition related to products, services, people, processes, and environments that meet or exceed expectations. The approach used by Davis asserts that quality is not only emphasized on aspect of final result, i.e. products and services but also human quality, and quality of the environment. It is impossible to produce qualified products and services without qualified people and processes.

To provide product services is in contrast to produce manufactured products in some ways. Such differences have important implications in quality management. Zeithaml, Berry and Parasuraman, (in Yamit, 2005:10-12) has conducted various studies on types of services, and managed to identify five dimension characteristics used by customers in evaluating quality of service.

The fifth dimension of service quality characteristics are:

- a. Reliability, the ability to provide services quickly and satisfactorily and in accordance with what is already promised.
- b. Responsiveness (comprehension), desire of the staff to help customers and provide service with a full response.
- c. Assurance (collateral), the ability, courtesy and trustworthiness owned by the staff, free from danger, risk or hesitations.
- d. Empathy, ease in the relationship, good communication, and attention sincerely to customers' needs.
- e. Tangibles (direct evidence), physical facilities, equipment, employees, and means of communication.

Quality of services is strongly influenced by consumer expectations. Consumer expectations can vary from one consumer to another consumer even though the service provided is consistent. The quality may be seen as a disadvantage if consumers' expectations are too high, even good services have been served. According Wyckof in Lovelock (quoted from Purnama, 2006:19-20) notion of service quality is perfection level to meet wishes of consumers. Quality of service is a comparison between perceived service (perception) of consumers with quality of service the customers expect. If the perceived service quality is equal or exceeds from the quality of service being expected, the service is qualified and satisfactory. According to Grönroos (in Purnama, 2006:20) quality of service includes:

- a. Quality function, how service is implemented including dimension of contact with consumers, attitudes and behavior, internal relationships, appearance, ease of access, and service mindedness.
- b. Technical quality and output quality perceived by costumers, that is price, punctuality, speed of service, and aesthetic output.
- c. Company's reputation, reflected by the company's image and reputation in the eyes of consumers.

In sum, quality of service is level of service of excellence to meet the desires of consumers or customers by an organization. Quality of service is measured by five indicators of service: reliability, responsiveness, assurance, empathy, and tangibles.

Customer Satisfaction:-

To define actual patient satisfaction is not easy, because the patient has a wide range of characteristics: good knowledge, social strata, experience, income and expectations. For example, a patient coming just wants to try certain health services from a health center. Before transaction, this new patient must have had hopes that he would be better served, waiters are friendly, responsive, and he will be served with therapeutic treatment. If his expectations is consistent with what is experienced and perceived, it exceeds from his expectations and certainly he

will feel satisfied. Conversely, if he experienced and felt the service is beyond the expectations; for example, the service is unfriendly, unresponsive and ineffective treatment, it is ascertained the patient is not satisfied.

The example above shows patient satisfaction can be seen after the use of products and services. A patient satisfaction is an after-purchase evaluation or evaluation results after comparing what is felt to his expectations. Patient satisfaction is the result (outcome) perceived on the use of products health services, that is equal or exceeds from the expectations (in Zulian Yamit, 2005:78). Patient satisfaction has become a central concept in the management discourse (Tjiptono and Chandra, 2005:192). Public Health Service patients generally expects the health care services enjoyed with good service or satisfactory (Assauri, 2003). Patient satisfaction can shape perceptions and further enhance positioning sub-district health services in the public eye.

Patient satisfaction is the result of perceived control over the use of products and services, equal or exceed the desired expectations. Kotler (2003) suggests several methods to measure patient satisfaction, as follows:

- a. The complaint system. This system provides the opportunity for patients to give suggestions, complaints and other forms of dissatisfaction by providing a suggestion box. Any suggestions and complaints entry should be a concern of management, for suggestions and complaints are generally guided by experiences of patients to reflect their love to the an organization.
- b. Survey of patients. Survey of patients is a common method used in measuring patient satisfaction, for example, by postal mail, telephone or direct interviews.
- c. Panel patients. Management invites loyal patients and patients who have moved to other hospital. The faithful patients will provide information on the level of satisfaction they feel and the former patients will inform why they move to other hospital. If patients who are not faithful (customer loss rate) is increasing, this indicates a failure in satisfying the patient's management.

Parasuraman (1998) identifies five dimensions of service quality (Servqual) as follows: (1) Tangible, appearance of physical facilities, including equipment, personnel and means of communication. Tangible could include elements of physical, personnel equipment, and communications materials. The aim is to strengthen quality, comfort, and safety of services offered to consumers. (2) Reliability, company's ability to keep and fulfill the promises given to consumer confidence. According Yasid (1999:113) reliability is the ability of a reliable, accurate and consistent in providing appropriate services that consumers want. (3) Responsiveness, quick response in reading and serve the desires or needs of consumers. The response is the willingness to help customers and provide services immediately. Some organizations choose to focus on the response position. They give attention to the desire of consumers to show "willingness to help" and serve the desires as soon as possible. (4) Assurance, knowledge, reliability, courteous of employees, trustworthy and confident. These dimensions can be used for the positioning by a number of industry effectively, especially if the reliability and confidence of service providers is very important. (5) Empathy, giving genuine concern and a private individual given to consumers by trying to understand the desires of consumers. A company is expected to have knowledge and understanding of consumers, understanding the needs of consumer needs specifically.

Consumers' attitude is actions by individuals, group or organization pertaining to the decision making process to achieve and use goods or services that are affected by the environment including decision making process. One of the fundamental factors on the study of consumers' attitude is people often buy products not for what they do, but for what they mean. This way, consumers buy a product nit to achieve its functions, but for its certain purposes, e.g. image, pride, and personality. Smart marketers tend to create certain relation between offered product and consumers.

Mowday, et. al., (1979:230) and Roberts (2003:48) developed a model of service quality in the organization connecting factors that affect quality of service to organization. The model compares quality of service in organization with output or consequence of quality of service to the organization, including: personal characteristics (e.g. age and tenure), characteristics of role and work (e.g. identity and feedback), structural characteristics (e.g. formalization and decentralization) and work experience (including cohesiveness group) that will produce output and consequent quality of services organizations in the form of a will to survive in the organization, level of attendance, a feeling of belonging and being part of the organization and a willingness to try as much as possible for the organization.

Customer satisfaction is determined by the quality of customer services desired, so quality assurance is a top priority for every hospital, which is currently used as a measure of competitive advantage of the hospital. According to Oliver and Doner cited by Kotler et al. (2003) customer expectation is a trust before trying (pre-trial belief) about a product that is used as a standard to evaluate performance of a future product or experience. Based on the description above, the conceptual framework of this study as follows:

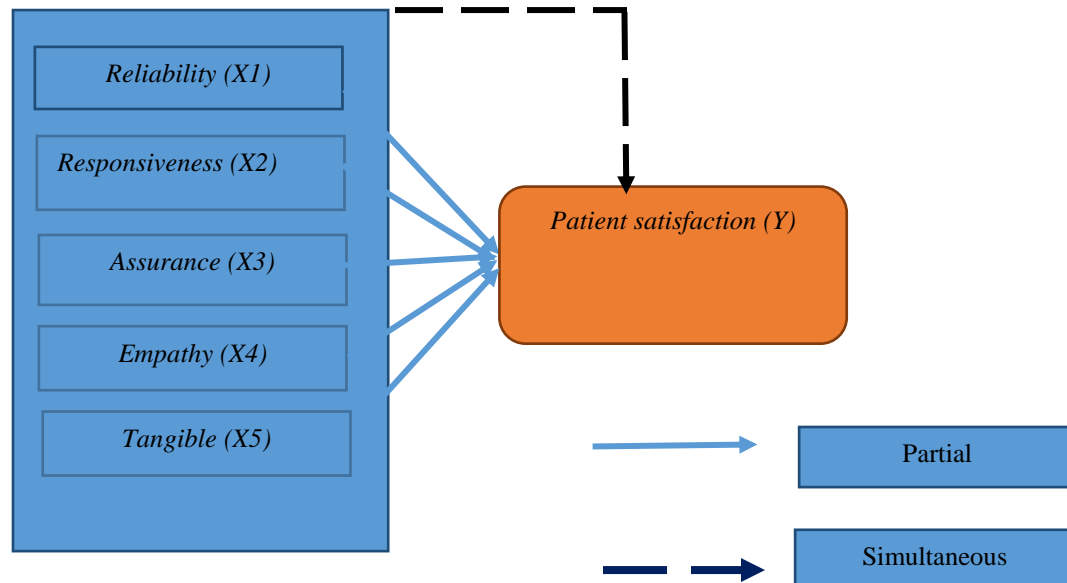


Figure 1:- Conceptual Framework of this Research

Method:-

This research employed quantitative design with explanatory non-experimental approach. The dependent variables included: reliability, responsiveness, assurance, empathy and tangible; and the independent variable was patient satisfaction. It described the influence of variables of service quality to client satisfaction through hypothesis testing without providing treatment. This study was conducted at Dr. Saiful Anwar Hospital in Malang, Indonesia from April to July 2016, which involved 50 patients as sample. Data were collected using questionnaires, observations and interviews, and were analyzed through descriptive and inferential statistics with SPSS version 17.

Results and Discussion:-

Regression Analysis:-

Test of multiple linear regression analysis is to examine and analyze the influence of Reliability, Responsiveness, Assurance, Empathy and Tangible to the satisfaction of the patients of Dr. Saiful Anwar Hospital in Malang. The magnitude of these effects can be seen from the recapitulation of regression testing in table 1.

Table 1:- Result of Regression Analysis

Model	Un-standardized Coefficients		Standardized Coefficients	T-value	T Table	Sig.
	B	Standard Error	Beta			
Constant	-21.198	2.080		-10.203	1.645	.000
X1	.343	.278	.305	2.551		.000
X2	.671	.412	.340	3.448		.016
X3	.266	.401	.257	2.463		.010
X4	.433	.315	.189	2.834		.022
X5	.420	.299	.277	2.321		.001
Multiple Correlation (R)				0.681		
Coef. Determinant (R ²)				0.464		
Ftest				16.287		
Ftable				2.400		
Probability				0.000		

Effect of Partial (t test):-

The ability of dimensions of service that affects patient satisfaction partially is explained by the regression equation. Based on data on Table 1, a regression equation was prepared as: $Y = -21.198 + 0.343 X_1 + 0.671 X_2 + 0.266 X_3 + 0.433 X_5 X_4 + 0.420 + 2.080$ and be explained as follows:

Effect of Reliability:-

Dimensions of reliability that had a positive impact on patient satisfaction at 0.343 meant if service quality dimension of reliability increased by 1 score it lead to increase patient satisfaction 34.3%. Evaluation of the effect of reliability through t-test showed t-value = 2.551 greater than t-table = 1.645 (df: 50, α : 5%) or $p < .05$, proving that reliability influenced significantly quality of patient care.

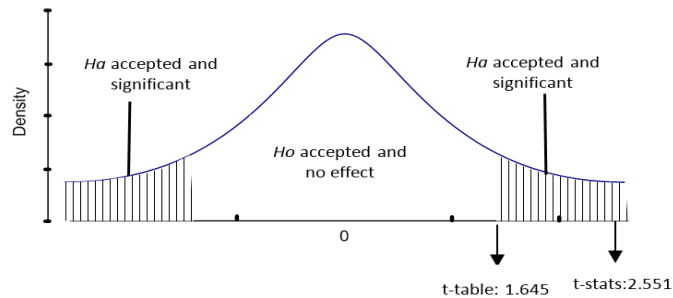


Figure 2:- Partial significance of *Reliability*

Effect of Responsiveness:-

Responsiveness dimension that had a positive impact on patient satisfaction = .671 meant if responsiveness increased 1 score it improved patient satisfaction 67.1%. Evaluation of significance through showed t-value = 3.448 bigger than t-table = 1.645 (df: 50, α : 5%) or ($p < .05$). Evidently, responsiveness affected significantly quality of patient care.

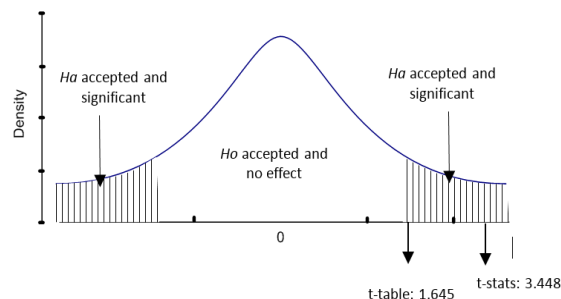


Figure 3:- Partial Significance of *Responsiveness*.

Effect of Assurance:-

Dimension of assurance that had a positive impact on patient satisfaction by 0.266 meant if assurance services increased 1 score it signified patient satisfaction 26.6%. Evaluation of significance through t-test showed t-value = 2.463, greater than t-table = 1.645 (df: 50, α : 5%) or $p < .05$. Evidently, assurance affected significantly quality of patient care.

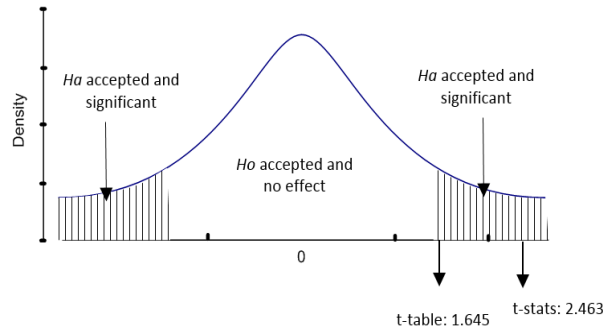


Figure 4:- Partial Significance of *Assurance*

Effect of Empathy:-

Empathy dimension that contributed a positive impact on patient satisfaction at 0.433 meant if an empathy increased by 1 score, patient satisfaction increased 43.3%. Evaluation of significance through t-test showed t-value = 2,834, greater than t-table = 1645 (df: 50, α : 5%) or $p=0.002$. It is evident that empathy affected significantly quality of patient care.

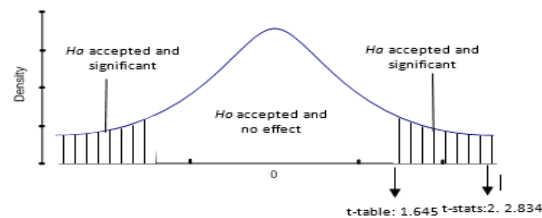


Figure 5:- Partial Significance of *Empathy*

Effect of Tangible:-

Tangible dimension that gave a positive impact on patient satisfaction at 0.420 meant if the tangible increased 1 score, patient satisfaction lead by 42%. Evaluation of significance through t-test = 2,321 that was greater than t-table - 1,645 (df: 50, α : 5%) or $p=0.001$ evidently proved that tangible affected significantly quality of patient care.

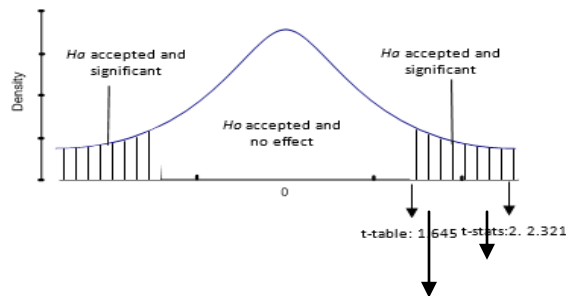


Figure 6:- Partial Significance of *Tangible*.

Simultaneous Influence (Test F):-

Simultaneous effect of dimensions of Reliability (X1), Responsiveness (X2), Assurance (X3), Empathy (X4) and Tangible (X5) on patient satisfaction (Y) was described by F-test. Data on Table 1 showed t-value = 16 287 and F-table = 2.400. If it was equal or exceeds the F-table, it was significant. The figure showed that F-value was greater than F-table; showing the null hypothesis (H_0) was rejected and the alternative hypothesis (H_a) was accepted. That is, the variable of reliability, responsiveness, assurance, empathy and tangible jointly affected significantly patient satisfaction in Dr Saiful Anwar Hospital Malang ($p=0.000$).

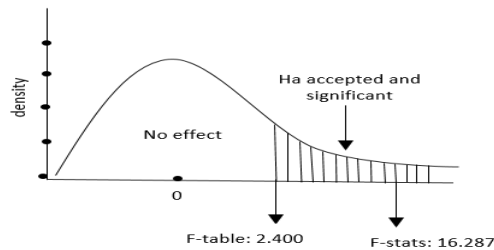


Figure 7:- Determinant Coefficient Significance

Beta Test:-

Beta test was to examine independent variables that had the most dominant effect on dependent variable, the variable that had the highest regression coefficient. Result of analysis as seen in Table 1 was that among service dimension variables, i.e. Reliability (X1), Responsiveness (X2), Assurance (X3), Empathy (X4) and Tangible (X5) to client satisfaction (Y), Responsiveness (X2) gained the highest value of standardized beta coefficient = 3,448. Thus, responsiveness was a service variable that had the most dominant influence on patient satisfaction in Dr. Saiful Anwar Hospital in Malang.

Determinants (R^2) Test:-

Determinant test was used to determine the amount of free variable to variable terminated bound. Table 1 showed coefficient of determination (R^2) = .464. This meant all variables of quality service dimension covering reliability, responsiveness, assurance, empathy, tangible contributed patient satisfaction in Dr. Saiful Anwar Hospital Malang up to 46.40%. The remaining 53.60% was influenced by other variables not analyzed in this study.

Conclusion:-

1. Analysis of overall service gap dimension, the fit between services provided by Dr. Saiful Anwar Hospital in Malang with patient expectations shows there is a gap or level of satisfaction at (-10.54). This score was categorized as moderate; yet it fully meets expectations of patients, the services provided is quite good.
2. The variables of service quality dimensions (Reliability, Responsiveness, Assurance, Empathy, Tangible) jointly have positive effect and significant impact on patient satisfaction of Dr Saiful Anwar Hospital Malang. F-value (16.287) is greater than F-table (2.640)
3. The variables of service quality dimensions (Reliability, Responsiveness, Assurance, Empathy, Tangible) partially have positive influence and significant impact on patient satisfaction of Dr Saiful Anwar Hospital Malang. This is supported by the t-value of each variable is greater than t table (1.645). The t-values of each of these variables are: Assurance (2463), Reliability (2551), Tangible (2321), Responsiveness (3448), Empathy (2834). Furthermore, the value of R^2 = (0464) showed that 46.4% variable of customer satisfaction is contributed by the service quality that is Reliability, Responsiveness, Assurance, Empathy, and Tangible. Other remaining 53.6% is contributed by other variables that are not included in this research model
4. Results of Beta Test shows that responsiveness variable achieves regression coefficient = .671; that means responsiveness has a dominant influence on the patient satisfaction of Dr. Saiful Anwar Hospital Malang.

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RESEARCH ARTICLE

IMPACT OF RISK-RETURN ANALYSIS ON STOCK FUTURES AND OPTIONS TRADING IN INDIAN EQUITY DERIVATIVES MARKET WITH SPECIAL REFERENCE TO NSE.

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Manuscript Info

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Key words:-

Futures, Call and Put option, Closing price, Strike price, Bullish and Bearish Market, Risk and Return.

Abstract

Futures & options is one of the most important segments in derivatives market in India. Financial derivatives have emerged as one of the largest market of the world during the past two decades in terms of trading volume, number of index and stock options available for trading, participation of investors in derivatives market. It is also observed that investors are showing lot of interest in the derivatives market. However, investors have lost lot of money in the derivatives market due to lack of knowledge about the product and investment strategies etc. The risk involved in futures and options trading can be minimized / return on futures and options trading can be improvised through designing suitable investment strategies. So, investor need to develop risk management as well as risk analysis tool which is the key to limiting futures and options risk/maximizing profit. The derivatives contract is standardized contract. In India, the BSE Sensex and Nifty-50 are the popular indices on futures and options trading. The everyday price changes will occur on stock index futures and options. Some of the major factors such as weather, war, Debt, refugee displacement, land reclamation and micro & macro economic factors will affect the index prices. So, this study challenge to test the precariousness of stock index prices in Indian Derivatives market and also this study have to focus on software industry **Infosys & TCS** during the period of April to June 2016.

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Introduction:-

The futures and options market have been the fastest growing segment of the securities industry. With this rapid growth has come to debate over the purpose served by futures and options in the capital markets. As we see it, the principal function of futures and options is to provide a significant expansion of the patterns of portfolio returns available to investors. Such expansions make investors better off and add to the liquidity and efficiency of the capital markets.

The emergence of the market for derivative products are forward, future and option are traded in the derivatives market. Comparing to stock market, derivatives markets are more risky because everything should be in a contract format. Derivatives are risk management instruments, which derive their value from an underlying asset. Banks,

Securities firms, companies and investors to hedge risks, to gain access to cheaper money and to make profit, use derivatives. Derivatives are likely to grow even at a faster rate in the years to come.

Price fluctuations make it hard for businesses to estimate their future production costs and revenues. Derivative securities provide them a valuable set of tools for managing this risk. This article describes the progress of Indian derivatives markets, the popular derivatives instruments, and the main users of derivatives in India.

Introduction to Derivatives Market in India:-

Derivative trading in India takes place either on a separate and independent Derivative Exchange or on a separate segment of an existing Stock Exchange. Derivative segment function as a Self-Regulatory Organisation (SRO) and SEBI acts as the oversight regulator. By their very nature, the financial markets are marked by a very high degree of volatility. Through the use of derivative products, it is possible to partially or fully transfer price risks by locking in asset prices. As instruments of risk management, these generally do not influence the fluctuations in the underlying asset prices. However, by locking in asset prices, derivative products minimize the impact of fluctuations in asset prices on the profitability and cash flow situation of risk-averse investors. In the last decade, many emerging and transition economies have started introducing derivative contracts.

National Stock Exchange of India Ltd:-

The National Stock Exchange of India Limited (NSE) commenced trading in derivatives with the launch of index futures on June 12, 2000. The futures contracts are based on the popular benchmark Nifty 50 Index. The Exchange introduced trading in Index Options (also based on Nifty 50) on June 4, 2001. NSE also became the first exchange to launch trading in options on individual securities from July 2, 2001. Futures on individual securities were introduced on November 9, 2001. Futures and Options on individual securities are available on 173 securities stipulated by SEBI. The Exchange has also introduced trading in Futures and Options contracts based on Nifty IT, Nifty Bank, and Nifty Midcap 50, Nifty Infrastructure, Nifty PSE indices. This section provides you with an insight into the derivatives segment of NSE. Real-time quotes and information regarding derivative products, trading systems & processes, clearing and settlement, risk management, statistics etc. are available here. Today NSE's share to the total equity market turnover in India averages around 72% whereas; in the futures and options market share is around 99%.

Basics of Equity Derivatives:-

According to the Securities Contract Regulation Act, (1956) the term Derivative is a contract or a product whose value is derived from value of some other asset known as underlying.

Equity derivative is a class of derivatives whose value is at least partly derived from one or more underlying equity securities. Options and futures are by far the most common equity derivatives. It provides you with an insight into the daily activities of the equity derivatives market segment on NSE. Two major products under Equity derivatives are Futures and Options, which are available on Indices and Stocks.

Derivatives Product:-

Index Futures: The contracts are traded on an Index futures or financial index. For each index there may be a different multiple for determining the price of the futures contract.

Index Option:- These options have the index as the underlying asset. In India, Index options are European option. For example options on Nifty, Sensex, etc.

Stock Futures:- Stock futures are agreements to buy or sell a specified stock, i.e., the equity share of a specified company, in the future at a specified price.

Stock option: These options have individual stocks as the underlying asset. For example, option on ONGC, NTPC etc.

Futures Contract:- A future contract is an agreement between two parties to buy or sell an asset at a certain time in the future at a certain price. Futures contracts are special types of forward contract in the sense that the former are standardized exchange-traded contracts.

Options Contract:-

An option represents the right to buy or sell a security or other asset during a given time for a specified price. Options are of two types:

Call option: It gives the buyer the right but not the obligation to buy a given quantity of the underlying asset.

Put option: It gives the buyer the right, but not the obligation to sell a given quantity of the underlying asset.

European option: The owner of such option can exercise his right only on the expiry date/day of the contract. In India, Index options are European.

Positions in derivatives market As a market participant, you will always deal with certain terms like long, short and open positions in the market. Let us understand the meanings of commonly used terms:

Long Position Outstanding/ unsettled buy position in a contract is called "Long Position".

Short Position Outstanding/ unsettled sell position in a contract is called "Short Position".

Open position Outstanding/ unsettled either long (buy) or short (sell) position in various derivative contracts is called "Open Position".

The Derivatives Market Participants:-

There are broadly three types of participants in the derivatives market - hedgers, traders (also called speculators) and arbitrageurs. An individual may play different roles in different market circumstances.

Hedgers They face risk associated with the prices of underlying assets and use derivatives to reduce their risk. They always wanted to reduce their investment risk.

Speculators They try to predict the future movements in prices of underlying assets and based on the view, take positions in derivative contracts. Derivatives are preferred over underlying asset for speculation purpose, as they offer leverage, are less expensive (cost of transaction is generally lower than that of the underlying) and are faster to execute in size (high volumes market). They are accepting their investment risk with view to more return.

Arbitrageurs Arbitrage is a deal that produces profit by exploiting a price difference in a product in two different markets. Arbitrage originates when a trader purchases an asset cheaply in one location and simultaneously arranges to sell it at a higher price in another location. Such opportunities are unlikely to persist for very long, since arbitrageurs would rush in to these transactions, thus closing the price gap at different locations. They don't want to take risk and always play with safe.

Review Of Literature:-

Phil Holmes (2006), Stock index future hedging: Hedge ratio estimation, duration effects, expiration effects and Hedge ratio stability this study focused the impact of hedge duration, and the time of expiration and hedge ratio stability over the study period and it helps to find with the help of some statistical tools like OLS, ECM, Risk & Return Analysis, Descriptive Statement and GARCH models.

Antonios Antoniou and Phil Holmes (2000), Futures trading, information and spot price volatility: evidence for the FTSE-100 stock index futures contract using GARCH This paper examines the impact of trading in the FTSE-100 Stock Index Futures on the volatility of the spot market. It ensures that the relationship between information and volatility of the spot and future with the use of GARCH analysis. The results suggested that futures' trading has a more volatile than spot market.

Stock returns volatility in the Tokyo stock exchange(2002), **Y.K. Tse** This paper examines the stock returns volatility in the Tokyo Stock Exchange. Structures of returns volatility are estimated and forecasted. The model of ARCH/GARCH and exponential weighted moving average (EWMA), this three tools used to measure the volatility of stock index future. The results show that the EWMA method gives the best forecasts. These findings have

implications in forecasting movements of market volatility, with applications to option pricing and control for variation margin risk in stock index futures.

Statement of the problem:-

The research in derivatives market awareness is comparatively less in India, when compared to other developed countries. The globalization of financial markets have been increasing the large number of traders over the decade by providing a wide variety of market and trading investment options. However, it makes the trading investment decisions process much more complex. The investors generally consider their investment needs, goals, objectives and constraints while making investment decisions. But it is not possible for them to make a successful investment decision at all times. Their attitude is influenced by a variety of factors such as speculative income, get rich quickly strategy, stories of successful traders, online trading, investor derivatives awareness programme, etc. A better understanding of equity derivatives options strategies, valuation processes and outcomes are important for financial planners because an understanding of how traders generally respond to market movements would help them in devising appropriate asset allocation strategies for clients.

Objectives of the Study:-

- The main objective of the study is to evaluate futures and options in equity derivatives of selected software companies of Infosys and TCS.
- To find out the risk and return relationship in future and option contract
- To have an in-depth awareness about future & option market in India.
- To assess risk management tools and its strategies.
- To analyze the precariousness of Future and Option market.

Scope for the Further Study:-

- The major scope of the study is to diminish the risk in derivatives and generate better returns in trading.
- A derivative contract is the off-balance sheet financing so no records maintain for the future reference.
- This study should spotlight on Future and option segment only so some changes happening in future.
- The different participants also obtain good returns in derivatives contract i.e. Hedger, speculator, Arbitrageur.

Limitations:-

- The study is based on secondary data and some inherent limitations of the secondary data would have exaggerated the study.
- This study should focus on stock future and options on particular companies only.
- The result of the investigation may change depending on the time period.
- This study we have to consider only the short term decision making.
- There is no stretchy trading in future contract because it is a standardized contract.

Methodology of the study:-

a.) Sample Selection, Sample size and Period of the study

- Underlying asset of Infosys and Tcs stock futures and options to be used for the study
- The availability of financial data required for calculating futures and options trading.
- Underlying assets must be in National stock exchange in the derivatives segment
- Selections of strike price have to be selected on contract's first day of options premium based on spot price.
- The study covers three contracts ranging from April 2016 to June 2016

Sources of Data:-

The study is exclusively based on secondary data. The base price of the contracts on subsequent trading days will be the daily close price of the futures and options contracts. The major sources of data collected from the www.nseindia.com.

Tools Used for Analysis:-

The statistical tools used to measure the futures and options price movements are; Risk (alpha & beta), Return and Descriptive Statement Analysis.

Jensen's Alpha is a risk-adjusted performance benchmark that tells you by how much the returns of an actively managed portfolio of market returns.

Originating in the late 1960s, Jensen's Alpha (often abbreviated to Alpha) was developed to evaluate the skill of active fund managers in stock picking.

- A positive Alpha means that a portfolio has beaten the market, while a negative value indicates underperformance
- A fund manager with a negative alpha and a beta greater than one has added risk to the portfolio but has poorer performance than the market

Careful stock picking and financial engineering means that investors can add alpha to a portfolio without adversely affecting beta.

Jensen Model ALPHA is defined by this equation

Expressed as a formula:

$$= R_f + \beta \times (R_m - R_f)$$

Where:

R_m = Return on Market

R_f = risk free rate of return.

β = Beta coefficient of portfolio

Beta describes the volatility of the portfolio with respect to that of the wider market, and is calculated with this equation

$$\beta = \text{Cov}_{xy} / \text{Var}_x$$

x = Market Returns

y = Portfolio Returns

The market return is usually described by the expected return of an index, like the Nifty 50 Index.

Returns:-

Long

$$R_t = (P_t - P_{t-1}) / P_{t-1} \times 100 \text{ or } (\text{Current Stock price} - \text{Previous day stock price}) / \text{Previous day stock price} \times 100$$

$$\text{Short} = (\text{Current Stock price} - \text{Next day stock price}) / \text{Next day stock price} \times 100$$

Where,

R_t = Return at the time

P_t = The Closing price of the day

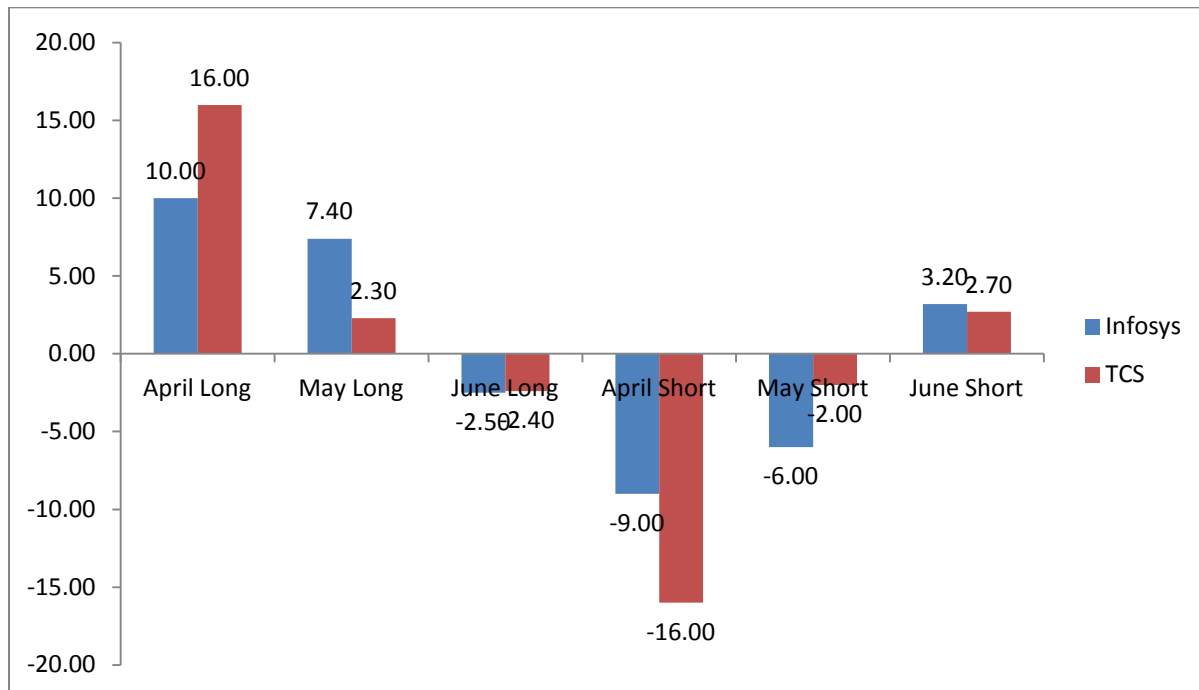
P_{t-1} = The Closing price of the day $t-1$

Descriptive Statement Analysis of daily return, standard deviation, Skewness Kurtosis Jarque-Bera test were analysed.

Analysis and Interpretation:-**Return Analysis:-**

Return table for stock futures (Apr-Jun 2016)

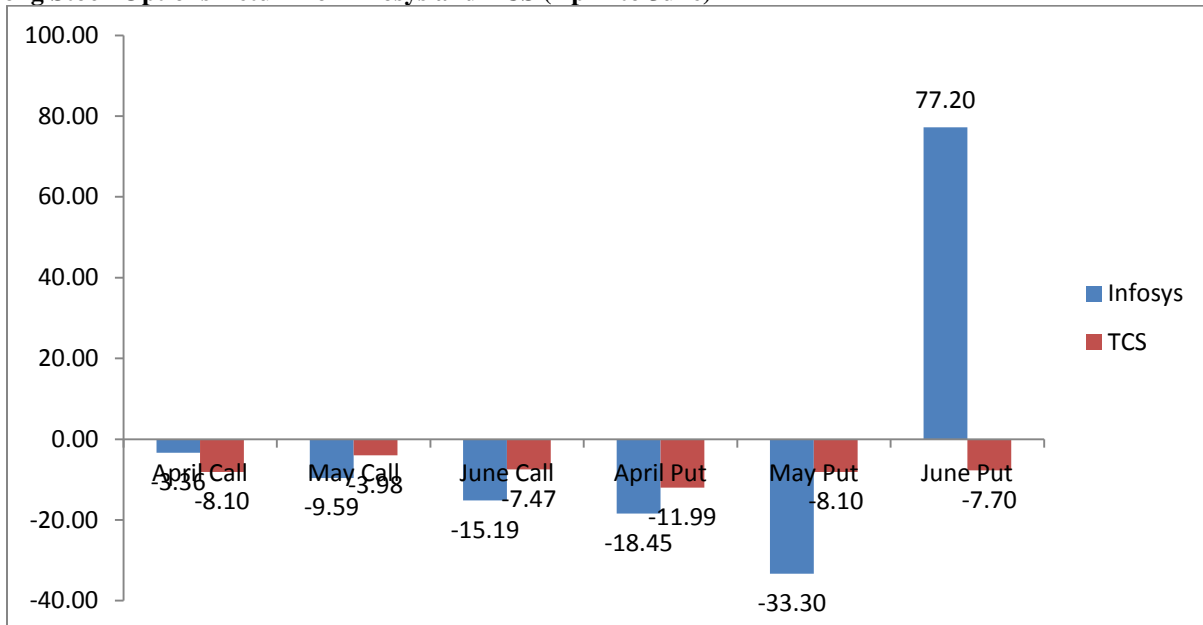
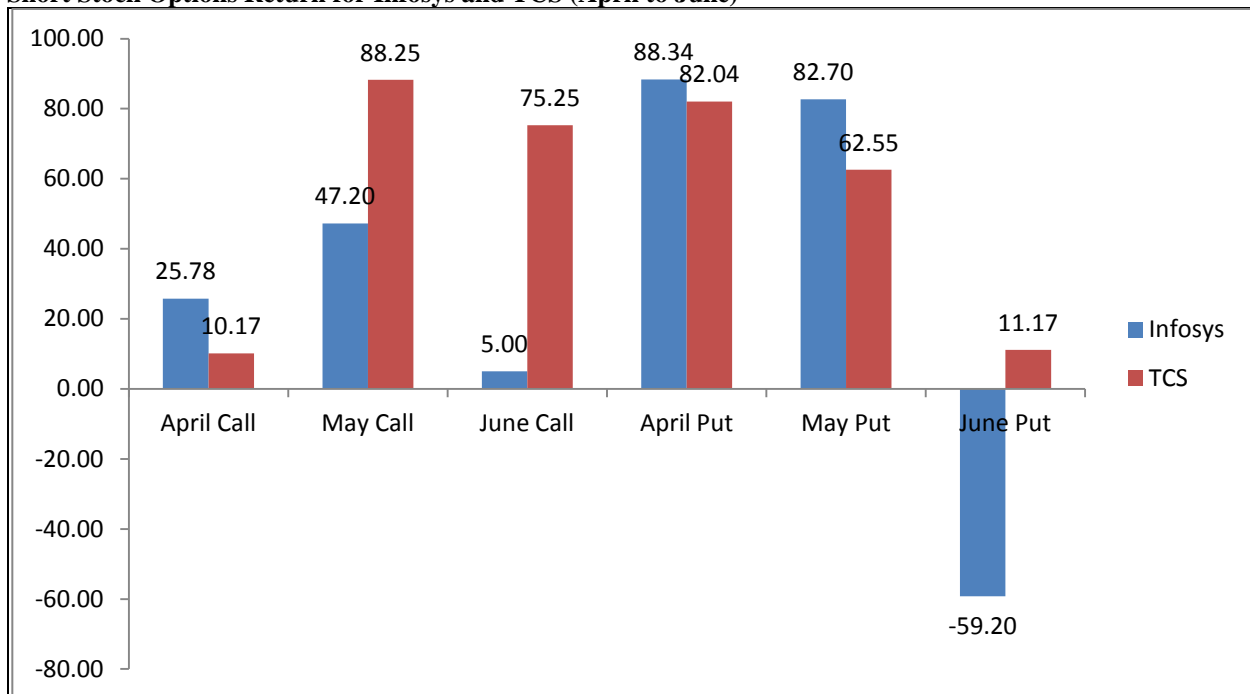
Long	Month	Infosys Fut	TCS Fut	Short	Infosys Fut	TCS Fut
	April	10.00	16.00		-9.00	-16.00
	May	7.40	2.30		-6.00	-2.00
	June	-2.50	-2.40		3.20	2.70

Long & Short Stock Futures Return for Infosys and TCS (April to June)

The above table indicates comparing the long and short return percentage of two software industries stock future of Infosys and TCS. This analysis indicates that TCS has maximum return in the month of April 2016 (16%) and Infosys has maximum return in the month of April 2016 (10%). At the same time short return percentage almost negative for the selected period with little positive return for both Infosys and TCS.

Return table for stock options (Apr-Jun 2016).

Long					
Month		Infosys CE	TCS CE	Infosys PE	TCS PE
April		-3.36	-8.10	-18.45	-11.99
May		-9.59	-3.98	-33.30	-8.10
June		-15.19	-7.47	77.20	-7.70
Short					
Month		Infosys CE	TCS CE	Infosys PE	TCS PE
April		25.78	10.17	88.34	82.04
May		47.20	88.25	82.70	62.55
June		5.00	75.25	-59.20	11.17

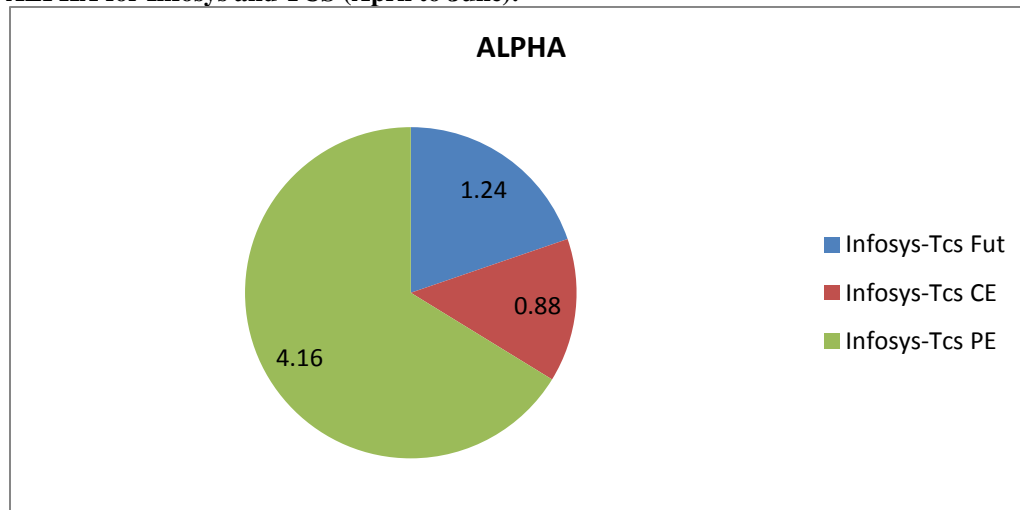
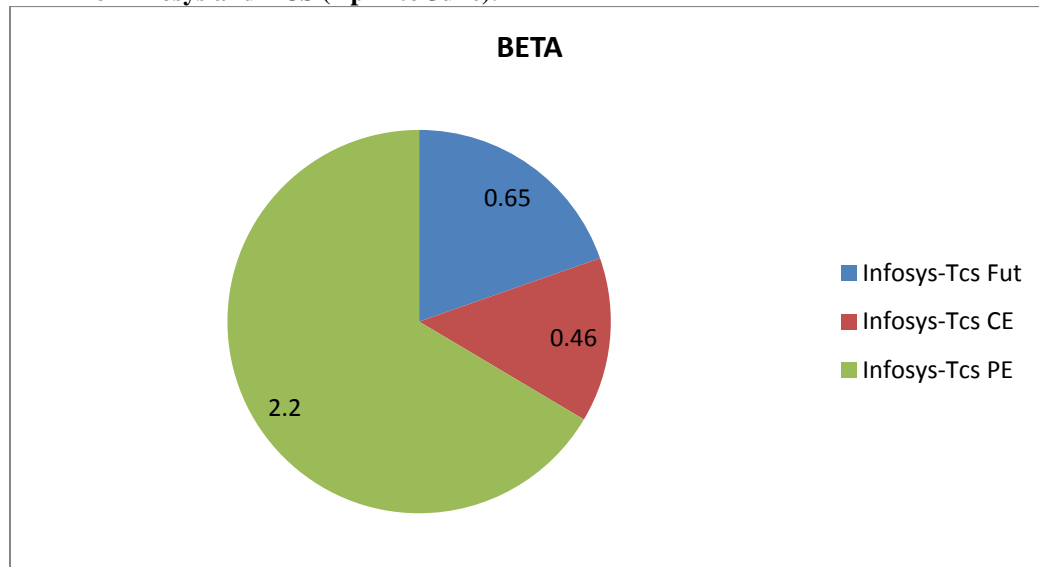
Long Stock Options Return for Infosys and TCS (April to June)**Short Stock Options Return for Infosys and TCS (April to June)**

The stock option table indicates comparing the return percentage of Infosys and TCS, this opposite to futures return, long return percentage came with negative return at the same time short return stood with huge positive. This analysis indicates Infosys and Tcs showed maximum return in the month of April & May 2016 (88.34% & 88.25%) and only Infosys has down in the month of June put option. The above table indicates that short return has given more positive than long return because of mounting uncertainties for H1B Visa factor in software industries so inversely impacted.

Risk Analysis (Alpha & Beta):-

Alpha & Beta table for Infosys & TCS Futures (Apr – Jun 2016):

Long	Infosys -Tcs Fut	Infosys -Tcs CE	Infosys -Tcs PE
Beta	0.65	0.46	2.20
Jensen Expected Return (ALPHA)	1.24	0.88	4.16
Short	Infosys -Tcs Fut	Infosys -Tcs CE	Infosys -Tcs PE
Beta	0.65	0.65	2.20
Jensen Expected Return (ALPHA)	1.24	5.47	4.56

ALPHA for Infosys and TCS (April to June).**BETA for Infosys and TCS (April to June).**

The positive alpha means the stock has over priced and the negative alpha means the stock has underpriced. Here both stock Infosys & TCS has overpriced.

The beta of less than one means that the security prices will be less volatile than the market price. A beta of greater than 1 indicates that the security's price will be more volatile than the market. Here the beta of stock futures (.65) has less volatile than the market and the beta of stock option (2.20) has more volatile than the market.

Descriptive Statement:-**Long Stock futures and options for Infosys & TCS (Apr – Jun 2016)**

Long	INFY_FUT	INFY_OP_CE	INFY_OP_PE	MARKET_RET_URN	TCS_FUT	TCS_OP_CE	TCS_OP_PE
Mean	-0.1384	-0.4070	0.0928	0.0311	0.0341	-0.1769	-0.2818
Median	-0.1000	-0.3600	0.0000	0.0300	0.0000	-0.2700	-0.3500
Std. Dev.	0.2399	0.3730	0.7342	0.1333	0.1776	0.5408	0.4046
Maximum	0.2900	0.3900	1.7900	0.3500	0.3900	0.9600	0.8300
Minimum	-0.6600	-1.0000	-1.0000	-0.2200	-0.3900	-1.0000	-1.0000
Skewness	-0.2958	0.1544	0.4647	0.3414	-0.0463	0.3684	0.6009
Kurtosis	2.2789	2.0473	2.3818	2.6696	2.4329	2.1910	3.0334
Jarque-Bera	2.2111	2.5491	3.1671	1.4625	0.8391	3.0433	3.6741
Probability	0.3310	0.2796	0.2052	0.4813	0.6574	0.2184	0.1593
observations	61	61	61	61	61	61	61

The above table presents the result of descriptive statistics of standard deviation, Skewness, Kurtosis and Jarque-Bera for Infosys and Tcs during the study period in the month of Apr – Jun 2016. It helps to summarize the overall performance of the business to understand the risk and reward of stock futures and options trading.

Descriptive Statement:-**Short Stock futures and options for Infosys & TCS (Apr – Jun 2016)**

Short	INFY_FUT	INFY_OP_CE	INFY_OP_PE	MARKET_RET_URN	TCS_FUT	TCS_OP_CE	TCS_OP_PE
Mean	0.14	0.56	-0.09	-0.03	-0.03	0.18	0.28
Median	0.10	0.61	0.00	-0.03	0.00	0.27	0.35
Std. Dev.	0.24	0.48	0.73	0.13	0.18	0.54	0.40
Maximum	0.66	1.58	1.00	0.22	0.39	1.00	1.00
Minimum	-0.29	-0.51	-1.79	-0.35	-0.39	-0.96	-0.83
Skewness	0.30	-0.25	-0.46	-0.34	0.05	-0.37	-0.60
Kurtosis	2.28	2.36	2.38	2.67	2.43	2.19	3.03
Jarque-Bera	2.21	1.65	3.17	1.46	0.84	3.04	3.67
Probability	0.33	0.44	0.21	0.48	0.66	0.22	0.16
bservations	61	61	61	61	61	61	61

The above table presents the result of descriptive statistics of standard deviation, Skewness, Kurtosis and Jarque-Bera for Infosys and Tcs during the study period in the month of Apr – Jun 2016. It helps to summarize the overall performance of the business to understand the risk and reward of stock futures and options trading.

Summary of Findings and Suggestions of the Study.**Findings:-**

The following are the major findings of the Study.

- The study found that the highest return was gained from short positions in put options in the both the stocks.
- Indian derivatives markets were more unstable throughout the study period.
- From the above study illustrates the comparative return analysis of Infosys stock future and TCS stock future has maximum return in the month of April (10% & 16%)
- The comparative return analysis of Infosys & TCS stock option has highest return in the month of May & June 2016 (88.34% & 88.25%).
- The comparative risk analysis of Stock Future & option Infosys & TCS has utmost minimum risk in stock option 0.46 & less market volatility, the maximum risk in stock option is 2.20 & more market volatility is 2.20.
- The stock futures volatility is normal of 0.65. They are a lesser amount of volatile than market.
- Descriptive statement explains that are constructive in summarizing data.

Suggestion:-

The following are major suggestions of the study

- The investors are advised to create the short positions in the beginning of the monthly contract of the selected Futures and options.
- Trading positions should create beginning of the contract to capture the more premium with a view of the eroding in the end of the contract.
- Normally, derivatives have certain risk so the investors should not take immediate investment decision and instead they should wait and watch the market movement.
- This risk and return analysis helps the investors to take appropriate investment decision.
- The Descriptive statement helps the investors invest their securities in the foremost company.
- Updating of market information help the investors to get maximum return.
- The investors should aware about the stock market conditions (e.g. Macro and micro data's monetary policy, GDP, inflation, war, social and political issues, currency fluctuation, etc.)
- The study suggests that the Regulatory Authority (SEBI) must monitor the reliability or the truth of information released by Companies.
- The present study suggests that whenever the performances of sectors declines in response to crisis announcement, the share holders should obtain instant decision either to buy or sell the stocks.
- Investors should enter in to the market with right direction with view to gain potential return.

Conclusion:-

Investors need to see the overall on an average the value of beta and the value of expected returns. There are several different types of risks involved in financial derivatives transactions. Achieving the right balance between risk and return will ensure that traders achieve their financial goals with proper decisions. Risk in futures and options investment is about what might cause a loss of money on those investments. Deciding what risk is to take high or low as well as looking at chronological data are two examples of understanding risks and returns of future financial planning. The recent financial derivatives growth will definitely lead to boost the investors' self-belief in the Indian derivative market and carry an overall progress in all the segments of this market. Futures and Options traders need a very firm knowledge of the basics of Futures and options trading before even thinking of ways to make the money out of it. So, this article would give a broad outlook for the new investors to learn about the Futures and options trading in India.

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RESEARCH ARTICLE

SURVEY ON AWARENESS ABOUT ORTHODONTIC TREATMENT IN GENERAL POPULATION OF NAGPUR DISTRICT.

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Key words:-

Malocclusion, Awareness, questionnaire survey.

Abstract

Context: Malocclusion is not a disease in itself but a variation in the arrangement of teeth, morphology of jaws, face and cranium. Awareness is the state or ability to perceive, to feel or to be conscious. Awareness forms the basis for planning oral health which is an inseparable part of general health.

Aims & Objectives: - To assess the awareness of orthodontic problems & its treatment in Nagpur District.

Materials & Methods: A cross sectional, observational (non-experimental) descriptive questionnaire survey was conducted in urban population of Nagpur District. Two age groups were studied i.e. adolescence and adult. According to the prestructured questionnaire (15 objective questions) data was formulated for assessment of knowledge and awareness of orthodontic treatment.

Results: There were no statistically significant differences in general information and awareness about the proper age of orthodontic treatment.

Conclusion: Awareness of malocclusion and the need to make corrections has increasingly become prevalent among our population. As growing public interest in oral health increased, the demand for orthodontic treatment also became more noticeable in dental practices.

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Introduction:-

Oral health can affect general health & well being of childhood through development. Malocclusion is not a disease in itself but a variation in the arrangement of teeth, morphology of jaws, face and cranium. Desire and need for orthodontic treatment is influenced by awareness, socioeconomic factors, and availability of the experts, ethnic trends and values. Awareness is the state or ability to perceive, to feel or to be conscious. Awareness forms the basis for planning oral health which is an inseparable part of general health. With the availability of meristic criteria and

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evolutions of indices of malocclusion, information on malocclusion and treatment needs is now being made available from all around the world.

Aims & Objectives:-

There is a paucity of data about the awareness of orthodontic treatment among school children in the Indian context. Therefore the rationale for this study was to assess the awareness of children towards orthodontic problems & its treatment. In this context, the objective of the present epidemiological survey was to assess the awareness of orthodontic problems & its treatment in Nagpur District.

Materials & Methods:-

A cross sectional, observational (non- experimental) descriptive questionnaire survey was conducted in urban population of Nagpur District. The sample size of 2000 people represented the rural population of Nagpur District. The sample was constituted from patients reporting to college OPD, patients from satellite clinics and students from school and college. In this modern era, though general population is aware of a dentist yet many people are unaware of Orthodontics as a speciality dealing with misalignment of teeth. Even if people are aware of orthodontics they hesitate to seek the treatment because of high cost of treatment, long duration of treatment, esthetic concern about placement of orthodontic appliance, extraction of some teeth if necessary. This survey has been formulated to evaluate the concern of general population in seeking orthodontic treatment.

Two age groups were studied i.e. adolescence and adult. People in age group 13-19 years constituted adolescent group & and People in age group of 20-40 years constituted adult group. Children who obtained written informed consent from parents to participate in the study were included. Exclusion criteria used were- history of previous orthodontic treatment, rampant caries, multiple missing teeth, mutilated malocclusion and other craniofacial anomalies like cleft lip and palate, facial hemiatrophy, cleidocranial dysplasia etc. An oral health lecture was given to all the children in the school to create awareness about Dental health and Orthodontic treatment.

Statistical data was analysed and categorised according to age and gender. Responses of the people were recorded on a 3 point Likert scale from poor, fair and good. According to the prestructured questionnaire (15 objective questions) data was formulated for assessment of knowledge and awareness of orthodontic treatment.

Questions included were:-

- Q.1) Have you ever visited a dentist?
a) Yes b) No
- Q.2) Have you heard of an orthodontist?
a) Yes b) No
- Q.3) Are you aware that an Orthodontist can bring your teeth in proper position?
a) Yes b) No
- Q.4) Did you notice improper positioning of your teeth by yourself or was it told to you by society/others?
a) Yourself b) Society/others.
- Q.5) Have you ever noticed people with improperly positioned teeth?
a) Yes b) No
- Q.6) Do you think that teeth should be properly positioned for a better facial appearance?
a) Yes b) No
- Q.7) Do you know at what age orthodontic treatment should be started?
a) At school b) Junior college
- Q.8) Do you know the side effects of improperly positioned teeth?
a) Yes b) No
- Q.9) Do you know that taking proper orthodontic treatment at an early age would improve your facial appearance?
a) Yes b) No
- Q.10) Do you know the cost of orthodontic treatments?
a) Yes b) No
- Q.11) Do you know how much time, orthodontic treatment takes?
a) <1 year b) 2-3 year
- Q.12) Are you aware that few teeth may have to be removed for proper positioning of irregular teeth?
a) Yes b) No
- Q.13) Do you know different types of habits that can cause improper positioning of teeth?

- a) Yes b) No

Q.14) Do you know about orthodontic braces and its types?

- a) Metallic Braces , Ceramic Braces
b) Lingual Braces , invisalign braces

Q.15) Are you aware that the improperly positioned teeth can be corrected at the age of 35-40 yrs also?

- a) Yes b) No

Statistical analysis:-

Data was coded and entered into excel sheet. To maintain the data quality (validity) rechecking and cross checking was done during data entry phase. Later, data was transferred into SPSS windows version 16, where cleaning, coding, recording, crosschecking, processing and analysis of the data were done. Simple

Descriptive statistics was applied to describe the study variables. The t- test procedure was used to compare means for two groups.

Results:-

There were no statistically significant differences in general information and awareness about the proper age of orthodontic treatment. Gender & Age distribution of the sample participating in the study is represented in Tables 1 and 2.

Table 1:- Distribution of sample size

Gender	Frequency	Percentage
Female	1010	50.5 %
Male	990	49.5 %
Total	2000	100 %

Table 2:- Age wise distribution.

Age	Frequency	Percentage
15-24	650	32.5
25-34	800	40
35-44	440	22
45-54	110	5.5
Total	2000	100

Table 3:- Response to Q1 –Q6.

	Q1		Q2		Q3		Q4		Q5		Q6	
	a	b	a	b	a	b	a	b	a	b	a	b
Frequency	550	1450	550	1450	660	1340	550	1450	700	1300	790	1210
Percent	27.5	72.5	27.5	72.5	33	67	27.5	72.5	35	65	39.5	60.5

Table 4:- Response to Q7 – Q12

	Q7		Q8		Q9		Q10		Q11		Q12	
	a	b	a	b	a	b	a	b	a	b	a	b
Frequency	1570	430	700	1300	1010	990	230	1770	320	1680	230	1770
Percent	78.5	21.5	35	65	50.5	49.5	11.5	88.5	16	84	11.5	88.5

Table 5:- Response to Q13 – Q15.

	Q13		Q14		Q15	
	a	b	a	b	a	b
Frequency	1450	550	1210	790	700	1300
Percent	72.5	27.5	60.5	39.5	35	65

Table 6:- Gender wise evaluation of response

		GENDER		Total
		Female	Male	
Evaluation	FAIR	240	310	550
	GOOD	640	580	1220
	POOR	130	100	230
Total		1010	990	2000

Chi Square=1.55 p=0.459

Table 7:- Age wise evaluation of response.

		AGE				Total
		15-24	25-34	35-44	45-54	
Evaluation	GOOD	370	500	240	110	1220
	POOR	90	80	60	00	230
	FAIR	190	220	140	00	550
Total		650	800	440	110	2000

Chi Square=8.55 p=0.2

According to their survey answers we found out that Majority of the sample included in this study had never visited a dentist before (Q1) neither the orthodontist (Q2) & his area of specialization (Q3). These people were told about their mal-alignment of teeth by other people of the society (Q4) & were not yet aware of the functioning of the orthodontist (Q6). These subjects were not aware of the side effects of improperly positioned teeth (Q8). They believed that this kind of treatment should be started at an early age (Q7) in order to improve their facial appearance (Q9), even though they were not aware about the cost of such treatment (Q10). They showed awareness that such treatment could take more than 2 years (Q11), but were not aware that few sound teeth had to be sacrificed for the same (Q12). The survey revealed that they had a knowledge about various habits causing malocclusion (Q13), metallic & teeth colored ceramic braces (Q14) but were not about adult orthodontic treatments (Q15). (Table 3-7)

Discussion:-

“Why people do not seek orthodontic treatment?” is the driving question among all staring orthodontist & a driving force for this study. The answer to this question would vary from, “Lack of Awareness, Metal display, Expensive treatment, Duration of treatment or Extraction of sound teeth”. The exact satisfactory response to this question was evaluated through this study.

Globally, there has been an increase in awareness of Orthodontics as a dental specialty in children as well as adults^[1-2]. The level of dental health knowledge, positive dental health attitude, and dental health behavior are interlinked and associated with the level of education and income as demonstrated by various studies^[3-8]. Attitudes and perceptions towards dental appearance differ among populations and among individuals^[9, 10].

Self-awareness is a dynamic process, not a static phenomenon^[11]. Although; imparting oral health education begins from the footstep of awareness, evaluation of its implementation is an important indicator of the success of the education imparted^[12]. Age-related changes in malocclusion concerns ideally should be studied longitudinally because with the progress in age, the awareness to nature & complexity of malocclusion increases.

In a similar study by Siddegowda¹³, High school children showed a higher level of awareness about orthodontic treatment when compared to middle school children. Urban children had shown more positive attitude like visiting a dentist in a study done by Singh *et al.* in 2012.^[14] Considering the influence of age on awareness of orthodontics, it was found that it increased significantly. Similar findings were seen in a study conducted by Friedman *et al.*, in 1976.^[14] The aim of orthodontic procedure is to improve dental occlusion, which results in better smile and good functioning in harmony with the face.^[13] Overall, it has been seen as an increase in awareness of orthodontic treatments as a dental specialty among children as well as adults.^[1]

Conclusion:-

Enhancing appearance and improving psychosocial status have been identified as important motivating factors behind the decision to initiate orthodontic treatment. This survey created awareness of orthodontic treatment among

various patients & children's. Awareness of malocclusion and the need to make corrections has increasingly become prevalent among our population. As growing public interest in oral health increased, the demand for orthodontic treatment also became more noticeable in dental practices.

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RESEARCH ARTICLE

OMEGA-3 FATTY ACID IMPROVE NON ALCOHOLIC FATTY LIVER INDUCED BY HIGH FAT DIET IN RATS.

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NAFLD, Omega-3 fatty acid, glucose, triglycerides, NEFA

Abstract

Dietary composition can influence the development of Nonalcoholic fatty liver disease (NAFLD). The amount and type of dietary fat may directly affect liver fat content, with high-fat diets being potentially harmful. A higher dietary fat intake is associated with liver inflammation and NAFLD. An Omega-3 fatty acid used in this study as a treatment to modulate these metabolic disturbances. This study was designed to elucidate the effect of Omega-3 fatty acid on high fat diet induced NAFLD in albino Wister rats. NAFLD induced by allowing rats to feed on the high fat diet for 20 weeks. Omega-3 fatty acid was used in doses of 0.8 mg/Kg body weight. Plasma glucose, triglycerides and non-esterified fatty acids (NEFA) were measured by enzymatic method. It can be concluded that administration of high fat diet caused NAFLD in rats via abnormal or extensive fat accumulation when energy intake exceed energy expenditure. Omega-3 fatty acid succeeded in ameliorating the deleterious effects of high fat diet induced NAFLD.

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Introduction:-

Nonalcoholic fatty liver disease (NAFLD) is rapidly becoming a serious global health problem, as its prevalence has risen rapidly in concert with the prevalence of obesity and type 2 diabetes. Estimates of the prevalence of NAFLD range widely from 11 to 46 % . It can progress to advanced liver disease in approximately 15–20% of patients. Clinical characteristics associated with NAFLD include obesity, hyperlipidemia, diabetes mellitus, and hypertension, all of which have been associated with underlying insulin resistance. [1,2]

Aside from weight gain and obesity, dietary composition can influence the development of NAFLD. The amount and type of dietary fat may directly affect liver fat content, with high-fat diets being potentially harmful[3]. A higher dietary fat intake is associated with liver inflammation and NAFLD[4,5]

While no human studies have linked NAFLD and diets high in saturated fat, evidence from experimental animal studies demonstrates that high dietary saturated fatty acid consumption worsens insulin resistance, NAFLD, and cardiovascular disease in rodents[6].

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Omega-3 fatty acids have recently been proposed as a potential treatment for NAFLD. These fatty acids have proven benefit in lowering serum blood glucose, serum triglycerides nonesterified free fatty acids (NEFFA) and in the treatment of cardiovascular disease. Interest in their potential in the treatment of cancer, mood disorders and cognitive disorders has also emerged.[7]

n-3 polyunsaturated fatty acids (PUFAs), such as docosahexaenoic acid (DHA), have been found to decrease liver fat content in children with NAFLD and possess anti-inflammatory effects. Moreover, recent studies indicated the benefits of *n*-3 PUFAs in lipid metabolism modification, dyslipidemia improvement, and hepatic inflammation mitigation, which may contribute to the amelioration of NAFLD.[8,9]

The aim of this study is to investigate the biochemical effect of omega-3 fatty acid involved in treatment of NAFLD induced by high fat diet in rats.

Materials and Methods:-

Materials:-

Omega-3 fatty acid was purchased from Sedico Co. (Giza, Egypt). Glucose kits were purchased from Spinreact Co. (Santa Coloma, Spain), triglyceride kits obtained from Spinreact Co. (Santa Coloma, Spain).

Animals:-

The present study was carried out on Thirty male Wistar rats of 96 ± 10 gm as body weight range. They were obtained from animal house of research institute of ophthalmology (Giza, Egypt). They were housed in groups of five rats per cage under controlled environmental condition of air and temperature with a 12 h light- dark cycle. Animal were allowed the diet and water in a free manner. Body weight of rats was recorded every two weeks.

Diet:-

In this experiment there were the standard normal rat chow diet and the high fat diet for induction of obesity in rats. The standard normal chow diet (ATMID Company, Egypt) consists of soybean, Corn, soybean oil, calcium carbonate, dicalcium phosphate, sodium chloride, lecithin, methionine, and vitamin/mineral mixture.

The high fat diet consists of 25% fat "beef tallow" + 10% sucrose + 20% corn starch + 45% normal chow with 30% sucrose in drinking water according to [10] with modification.

Experimental design and animal grouping:-

The rats were divided randomly into three groups of ten rats each and treated as follows:

Group I:- (normal control): rats were fed on a normal chow diet for 20 weeks and administered water as a vehicle via oral gavage.

Group II:- (High fat diet group): rats were fed on the high fat diet for 20 weeks and administered water as a vehicle via oral gavage.

Group III:- (omega-3-fatty acid treated group): rats were fed on the high fat diet for 20 weeks and administered fish oil daily for the last two consecutive weeks via oral gavage in a dose accounting for (0.8 gm/ kg body weight) of Omega-3 fatty acid [11, 12]. The rats were weighed then sacrificed at the end of the experiment after about 18 hours fasting to minimize variation in lipid pattern during two successive days after blood samples have been collected from retro - orbital veins into tubes containing EDTA and centrifuged at 1500 RPM for 30 minutes. An aliquot of the separated plasma was used for immediate estimation of fasting blood glucose. Second aliquot was kept on -20°C for triglyceride and NEFA analysis. All animal procedures were performed upon approval from the Ethics Committee of Beni-Suef University and in accordance with the recommendations of the proper care and use of lab animals.

Biochemical Analysis:-

Fasting blood glucose was carried out by enzymatic method (Spinreact, Santa Coloma, Spain). The triglyceride level was determined according to the enzymatic method (Spinreact, Santa Coloma, Spain). And NEFA levels were determined according to the (Duncombe) method described by (Duncombe 1964 [13]).

Statistical Analysis:-

Data were presented as means \pm SEM values. The results were analyzed statistically by one-way analysis of variance (ANOVA) with subsequent multiple comparisons using Tukey multiple comparison Post-Hoc test. The p-values less than 0.05 were considered significant. Correlations between variables were assessed by Pearson's correlation test. All calculations were made using the computer program SPSS 16.0 (SPSS, Chicago, III, USA). The data were graphed using GraphPad Prism 6 (GraphPad Software, Inc., USA).

Results:-**Metabolic Markers:-****Fasting plasma glucose level:-**

The fasting plasma glucose level was significantly increased in high fat diet treated group when compared to that of the normal control group as shown in table (1). Treatment with omega-3 fatty acid significantly reduced fasting plasma glucose elevation as shown in table (1) and fig. (1).

Table 1:- fasting plasma glucose, triglyceride and non-esterified fatty acid levels in the rat of various studied groups.

	Normal control	High fat diet group	Omega-3 fatty acid treated group
Fasting plasma glucose level	93.01 \pm 3.89	135.26 \pm 2.54 [#]	102.69 \pm 4.12 [*]
Plasma triglyceride	84.01 \pm 15.80	145.26 \pm 18.78 [#]	85.55 \pm 7.12 [*]
Plasma NEFA	52.07 \pm 5.89	95.26 \pm 4.54 [#]	62.69 \pm 4.12 [*]
❖ Data are means \pm SEM. ❖ Data are expressed as mg/dl. ❖ #p < 0.05 compared with the normal control group. ❖ * p < 0.05 compared with the high fat fed group.			

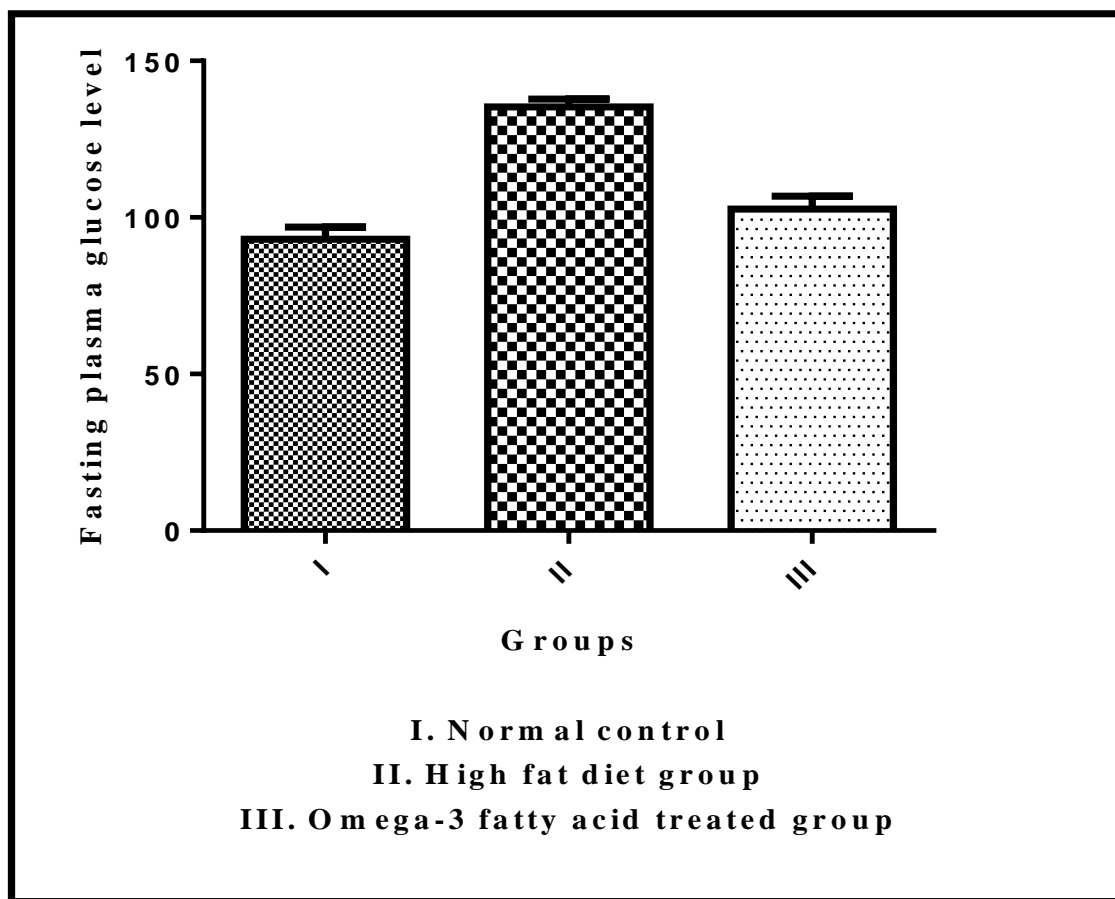


Figure 1:- Changes in fasting plasma glucose level in different groups.

Plasma triglycerides level:-

The plasma triglycerides level was significantly increased in high fat diet treated group when compared to that of the normal control group as shown in table (1). Treatment with omega-3 fatty acid succeeded in significantly reducing plasma triglycerides level as shown in table (1) and fig. (2).

Plasma non- esterified fatty acid level:-

The plasma **non-esterified fatty acid** level was significantly increased in high fat diet treated group when compared to that of the normal control group as shown in table (1). Treatment with omega-3 fatty acid succeeded in significantly reducing plasma non- esterified fatty acid level as shown in table (1) and fig. (3).

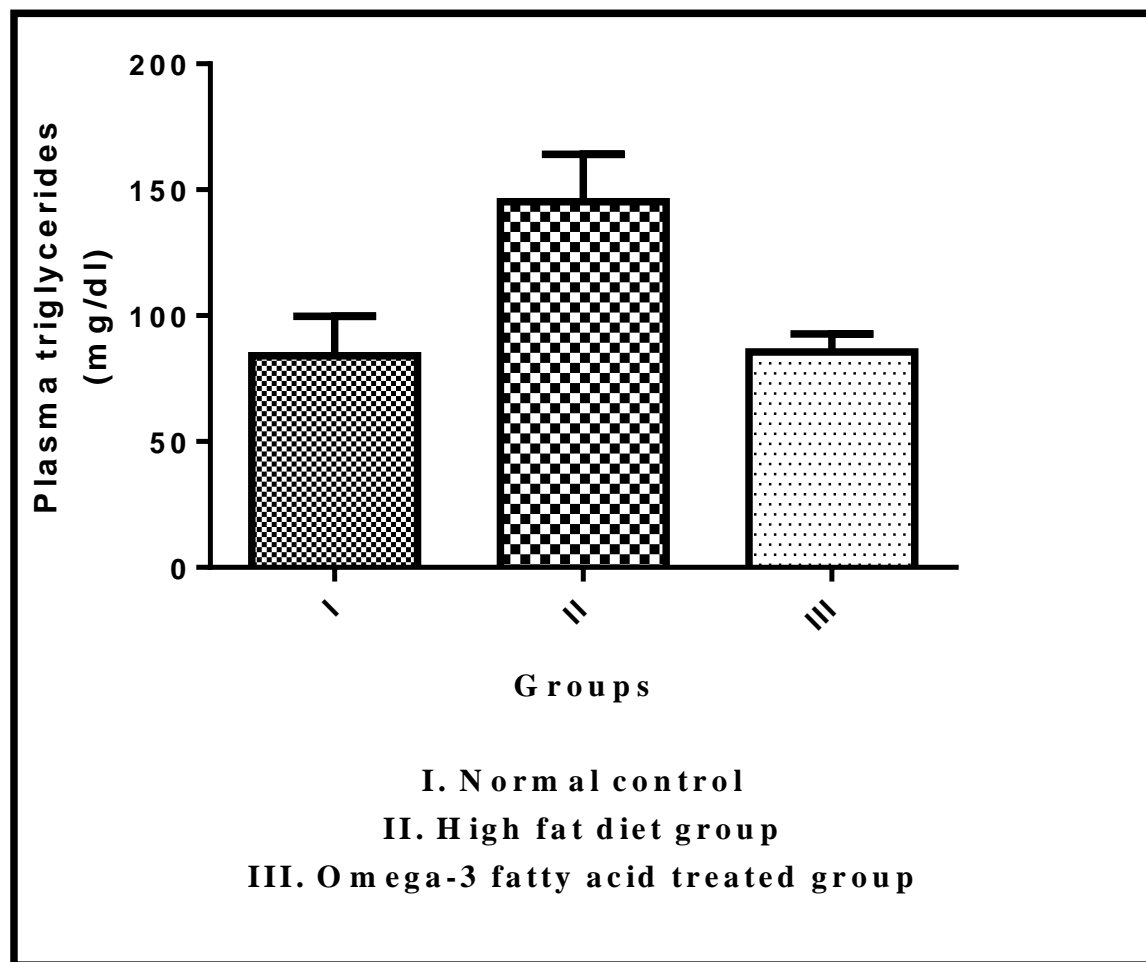


Figure 2:- Change in plasma triglycerides in different groups.

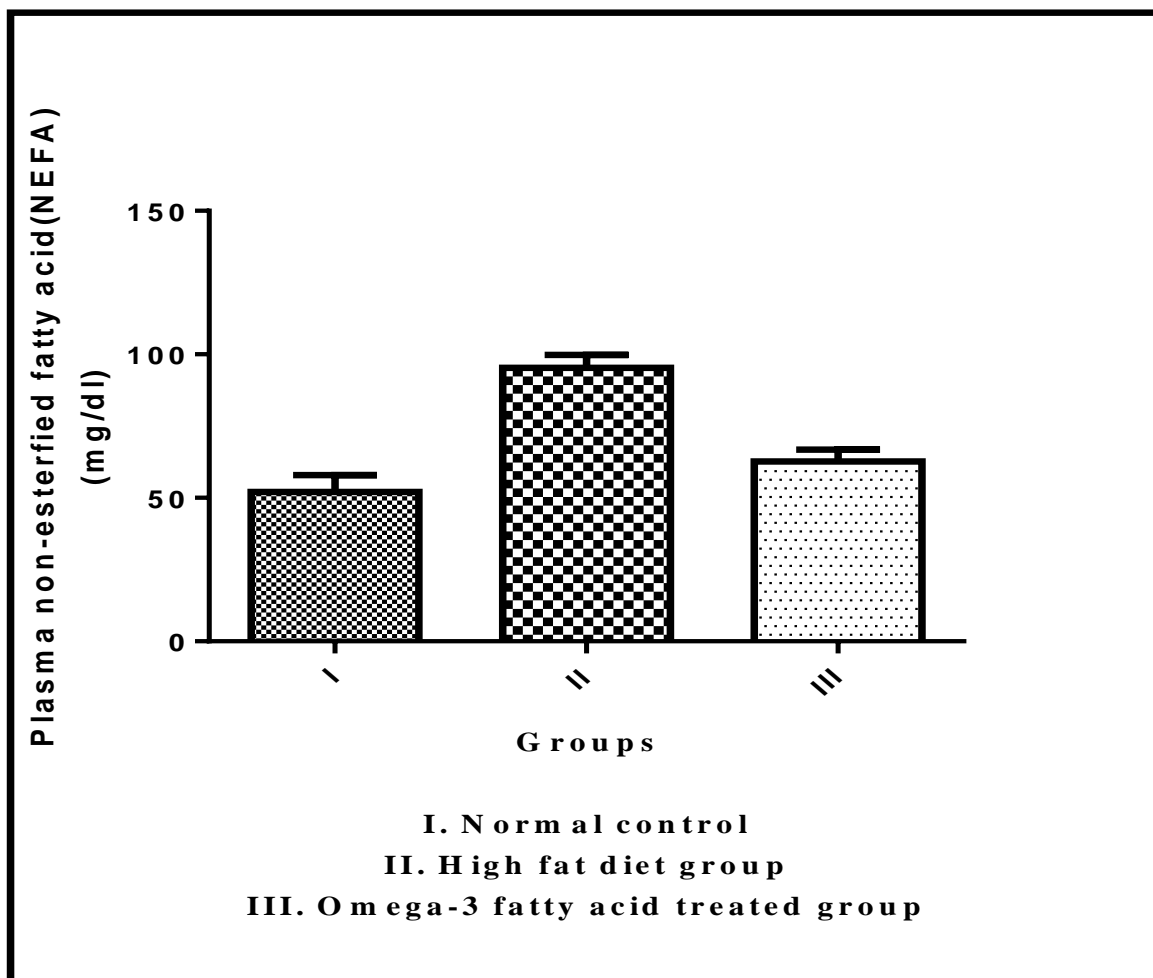


Figure 3:- change in plasma non -esterified fatty acid in different groups.

Discussion:-

NAFLD can be considered the hepatic manifestation of the metabolic syndrome. Component of metabolic syndrome include insulin resistance, dyslipidemia, type 2 diabetes, hypertension and hypertriglyceridemia[13,14]. When the liver gets fatty because of NAFLD, the ability of insulin to inhibit hepatic glucose production is impaired. This hepatic insulin resistance leads to a slight increase in plasma glucose concentrations and stimulation of insulin secretion. The present study demonstrated that the level of fasting plasma glucose the high fat diet treated group was significantly increased compared to a normal control group. These data were in harmony with the previous study [15] which reported that a significance increase in fasting plasma glucose level with a defect in insulin signaling occurred in adipose tissue after HFD intake in rats compared with of control rats.

The present study found that treatment with omega-3 fatty acid, there was a significant decrease in the raised level of plasma glucose compared to HFD treated group. This in agreement with the previous studies [16,17], which reported that omega 3 fatty acid enhanced glucose uptake and improve insulin resistance in HFD-induced NAFLD in rats as omega -3 fatty acid may improve insulin signal transduction in adipocytes, affecting in turn, insulin-stimulated glucose transporter (GLUT4) in both skeletal muscle and adipose tissue accompanied with lower glycemia and insulinemia [18]. Moreover, omega-3 fatty acid could regulate both the activity and expression of the liver glucose-6-phosphatase, which could explain the protective effect with respect to the excessive hepatic glucose output induced by a high fat diet [19].

Hypertriglyceridemia and high levels of non-esterified fatty acids (NEFA) in the MetS promote abnormal accumulation of lipids within the liver, in a form of steatosis or non-alcoholic fatty liver disease (NAFLD)

(12). Thus, NAFLD affects up to 90% of obese people and nearly 70% of the overweight subjects, and it is thought to be the hepatic event in the MetS. In addition, NAFLD is commonly associated with MetS risk factors such as obesity, IR, hypertension and dyslipidemia [20]. Data presented in this study indicated that HFD intake resulted in a significant increase in the level of triglycerides and NEFA in rats compared to those of normal control. The observation of the current study was in accordance with those of the previous studies [21,22] which reported that there is an elevation of both triglycerides and NEFA in NAFLD in rats. The mechanisms governing the accumulation of TGs in hepatic cells include changes in hepatocellular metabolism, [23,24] propagating an imbalance between uptake and de novo fatty acid synthesis, VLDL formation and subsequent export, and oxidation capacity [25]. The present study also revealed that treatment with omega-3 fatty acid showed significant reduction in the level of triglycerides and NEFA in plasma compared to those of HFD treated group. This is in agreement with the previous [26,27] which supports the triglyceride-lowering properties of omega-3 fatty acid and decreased NEFA level in plasma. This could be explained by changes in transcription of several nuclear receptors are reported to mediate the TG-reducing effects of omega-3 fatty acid: sterol regulatory element binding proteins (SREBP), liver X receptor-alpha (LXR α), retinoid X receptor alpha (RXR α), farnesoid X receptor (FXR), and peroxisome proliferator-activated receptors (PPARs), and each play prominent roles in controlling lipid metabolism. Also the reduction of NEFA by omega-3 fatty acid due to suppressing in gene expression involved in new fatty acid synthesis and by inducing fatty acid oxidation in different tissues, such as, liver, skeletal muscle and white adipose tissue [28,29]

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RESEARCH ARTICLE

LES STRUCTURES DE GESTION DES DESTINATIONS TOURISTIQUES ET LEURS FONCTIONS FACE AUX DEFIS DE LA COMPETITIVITE : UNE SYNTHESE DE LA LITTERATURE.

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Abstract

Il n'est plus d'intérêt de rappeler l'importance du tourisme pour le développement socioéconomique des pays et des territoires. Le secteur devenant primordial dans les stratégies de développement de nombreux pays va attirer depuis près de trois décennies l'intérêt des chercheurs de tous les horizons qui vont chercher à le comprendre, l'analyser et identifier les facteurs clés de son développement. De même, le fort taux de croissance du secteur, sa forte résilience et résistance aux crises ainsi que les perspectives très encourageantes de son développement futur vont enflammer la course à la compétitivité des destinations touristiques, devenues les unités de base et centrales dans cette quête de compétitivité vont être au centre des politiques et des stratégies de développement touristiques, mais aussi, faire foisonner la recherche scientifique sur les conditions, les formes et les dimensions de cette compétitivité et sur les défis qu'elle impose et les formes, rôles et fonctions des structures de management de ces destinations pour y répondre efficacement et asseoir la compétitivité des destinations. Malgré, le caractère très récent de la recherche managériale sur ces différentes questions un nombre assez important de recherches ont été produites et un corpus théorique est entrain de se constituer et de s'établir.

Cet article, de nature exploratoire, se propose d'étudier, à l'aune de la littérature, le rôle et les fonctions des structures de gestion des destinations touristiques dans le cadre des nombreux défis imposés par la recherche de la compétitivité dans un marché touristique mondial évolutif, mutants et hautement concurrentiel. Plus particulièrement et à travers une synthèse de la littérature existante tant sur la compétitivité et ses implications pour les destinations touristique que sur les théories et modèles de management des destinations touristiques, nous cherchons à faire un état de la littérature sur ces questions et à relever le degré d'intégration des défis posés par la compétitivité dans les rôles et fonctions des structures de gestion des destinations touristiques.

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Introduction:-

Depuis que le tourisme est reconnu comme secteur économique important, la question de la compétitivité des destinations touristiques est devenue à l'ordre du jour et intéresse aussi bien les gouvernements, les chercheurs que les organismes internationaux. En effet, le secteur touristique de part sa croissance continue et ses apports aux équilibres socioéconomiques de nombreux pays tant développés, émergents qu'en voie de développement est devenu un objet majeur au centre des politiques de développement prônées par les gouvernements et les organismes internationaux qu'un objet central dans les axes de recherche de plusieurs chercheurs à travers le monde. La nature mondialisée et hautement concurrentielle du marché touristique international fait que la réussite dépend largement de la position de ce secteur dans le marché international du tourisme (Gomezelj et Mihalic, 2008). De même, au centre du secteur, les destinations se pointent comme les unités centrales et importantes de l'analyse et de la mise en œuvre des politiques et stratégie touristiques. Et le succès d'une destination dans le tourisme international dépend en grande partie de sa compétitivité (Enright et Newton, 2004). Ainsi, La compétitivité des destinations devient un objet d'études central tant des chercheurs en tourisme que des organismes internationaux intéressés par le secteur qui s'intéressent tous à comprendre les dimensions et les enjeux de cette compétitivité et à étudier les aptitudes des entreprises, des territoires et des pays à soutenir la concurrence sur un marché mondial de plus en plus évolutif, saturé et intégré et à discuter plus particulièrement des fonctions et tâches fondamentales que doivent remplir les structures de management des destinations pour maintenir et améliorer leur compétitivité (Gomezelj et Mihalic, 2008).

Si la question des effets des structures organisationnelles pour la performance et le succès des organisations est un sujet largement discuté et débattu dans les sciences du management, la pertinence des structures organisationnelles pour la compétitivité des destinations touristiques est un sujet récent et qui depuis son initiation par Ritchie et Crouch en 1993 est devenu au centre de l'intérêt de la recherche scientifique sur le tourisme. Cette position centrale prise par cette thématique émane de la congruence de plus en plus constatée de la perception des destinations, par les chercheurs et les gouvernements, comme les unités compétitives du tourisme sur les quelles doivent ce centrer les questions de recherche et d'analyse et de mise en œuvre des actions de promotion et de développement visant l'amélioration de la compétitivité. Poser la question des effets des structures organisationnelles du management des destinations touristiques sur la compétitivité des destinations touristiques se heurte à plusieurs difficultés liées en premier lieu à la nature complexe et polysémique des concepts de compétitivité et de destination touristique et en second lieu à la problématique même de ces structures de gestion et de leurs fonctions.

Dans cet article de nature exploratoire nous chercherons à mettre en lumière, sur la base d'une étude documentaire relative aux différentes avancées théoriques sur la question, les fonctions, tâches et rôles que doivent assurer les structures de gestion des destinations pour faire face aux défis imposés par la compétitivité dans le secteur touristique. Cette quête nous mènera tout d'abord à s'interroger sur la question de la compétitivité des destinations touristiques et relever les principaux défis qu'elle impose aux organes de gestion des destinations touristique pour ensuite s'interroger sur ces différentes structures et leurs fonctions et enfin dégager les conclusions et les perspectives.

Sur les Destinations Touristiques et leur Compétitivite:-

Dans cette section nous tenterons de mettre en relief tous les aspects de la littérature relatifs aux concepts clés de notre recherche à savoir notre champ d'investigation – les destinations touristiques, le concept de compétitivité et celui de compétitivité des destinations touristique et les théories et modèles qui ont tenté de l'expliquer pour ensuite essayer identifier les principaux défis que cette compétitivité impose au management des destinations.

La destination touristique, un concept complexe et insuffisamment définie:-

Le succès du tourisme est étroitement associé au mot destination (Kadri et al., 2011). Toutefois ce concept de destination reste difficile à définir (Haywood, 1986). En effet, il existe une multitude de définition qui ont été données à la destination touristique. La littérature d'origine institutionnelle la définit généralement en référence à un espace géographique relativement cohérent, doté d'une certaine autonomie décisionnelle et présentant des caractéristiques attractives pour les visiteurs extérieurs auxquels des produits adaptés sont proposés (Commission européenne, 2000). L'Organisation Mondiale du Tourisme (OMT) la définit comme «tout territoire touristique accessible, incluant des infrastructures d'hébergement, de transport, ainsi que des activités, animations et services, constitue une destination. Cette destination, qui peut être définie par des limites physiques, thématiques ou administratives, adopte un ensemble d'images et de qualités distinctives qui lui donnent une identité.». Si l'on se réfère à présent à la *Resource-based view* (RBV), une destination peut être définie comme un ensemble de

ressources productives et humaines (Penrose, 1959) dont le stock disponible à un instant t résulte d'une économie d'accumulation des ressources (Cool, 2000) réalisée au fil des siècles par les générations successives (Dierickx et Cool, 1989). De même de nombreux chercheurs émanant de plusieurs disciplines ont tenté de cerner ce concept en lui apportant des définitions. Ainsi, GEE et al (1989) considèrent que les destinations touristiques représentent des domaines spécifiques que les voyageurs choisissent de visiter et où ils passent une période de temps significative. Mathieson et Wall's (1992) avancent qu'une destination touristique correspond à un endroit qui présente des caractéristiques qui sont connues par un nombre suffisant de visiteurs potentiels et qu'ils considèrent comme une identité qui attire les voyageurs quelques soient les attractions d'autres endroits. Pour Jenkins et Tosun (1996) les destinations touristiques sont des domaines ayant des caractéristiques naturelles ou créées par l'homme et qui attirent les visiteurs locaux et étrangers. De leur côté, Morant et al (1996) conçoivent la destination touristique comme une combinaison d'éléments interdépendants (ressources, infrastructure, transports, ...) qui produisent de la satisfaction des touristes. Kim (1998), lui, présente la destination touristique comme un produit exceptionnellement complexe de l'industrie du tourisme qui comprend, entre autres, le climat, l'infrastructure et de la superstructure (services et attributs naturels et culturels). D'autres auteurs adoptant des points de vue très étroits considèrent la destination comme une localité (Scaramuzzi, 1993), une station touristique, une zone touristique ou une région (Sancho, 1998).

De toutes ces définitions, il ressort qu'il ya un certain nombre de points communs partagés par les auteurs notamment la référence à l'aspect géographique (endroit, lieu, domaine, territoire, zone, région, localité), à l'aspect du fonctionnement en terme économique (marché, produits, services), l'aspect ressources et disponibilités (climat, infrastructures, ressources naturelles et culturelles,...) et à l'aspect psychologique (motivation du touriste et visiteurs).

Toutefois, malgré cette multitude de définition nous pouvons avancer que le concept de destination, au cœur de la compréhension du phénomène qu'est le tourisme (Botti et Séraphin, 2011), possède également un caractère complexe et souffre d'une insuffisance définitionnelle et de théorisation (Boualem et al, 2011) et ne fait pas l'objet d'un consensus commun, unanime et complet de tous les chercheurs. Il est ainsi, claire que le concept de destination pris dans sa totalité et sa complexité pose un problème de définition et d'approche. Une lecture de la littérature nous informe que le concept de destination est devenu en quelque sorte un mot-valise mythique (Hazebroucq, 2009 : 8) dont la signification est fortement corrélée à des considérations contextuelles (Kadri et al., 2011). Et cela est dû, selon les chercheurs, au fait de la domination scientifique de deux principales disciplines qui sont la géographie et le marketing qui ont été les premières à s'intéresser au concept de destination et qui l'on adapté à leur objet et imposer leur façon de l'aborder. La géographie, ayant été la première à s'accaparer le concept et l'étudier, a mis toujours en avant la dimension spatiale. Ainsi, les définitions telles que celles de l'Équipe MIT (2005, p. 11) et de Hayllar et al. (2008), montrent clairement le poids de la variable spatiale dans l'interprétation de ce concept (kadri et al, 2011). Pour l'Équipe MIT, «La destination est un lieu d'interactions territoriales qui relient ensemble les données socioculturelles, naturelles, patrimoniales et techniques» et pour Hayllar et al., « l'étude des territoires touristiques concerne d'abord l'étude des espaces.». Ainsi, nous pouvons relever que le concept de destination tel que abordé par cette discipline est resté, compte tenu de fait que les pratiques touristiques supposent mobilité et déplacement, attaché à une dimension spatiale malgré la nature de totalité et de complexité du phénomène touristique (Violier, 2009). Cet auteur, revient ensuite pour critiquer le réductionnisme de cette approche qu'il juge positiviste dans le sens où elle ne définit le lieu touristique que par la présence d'objet naturels ou culturels ; alors que pour lui, la présence d'objets ne suffit pas à assurer la présence des touristes. Selon cet auteur « la destination serait ce que les touristes perçoivent comme l'espace où ils mettront en œuvre des pratiques qui définissent alors le territoire touristique » (Violier, 2009). C'est dans le même sens que Caccomo (2006, p.136) avance que «c'est la fréquentation qui confère sa valeur à la destination» ou Ceriani et al. (2008) qui considèrent qu'un «lieu est touristique par les touristes qui le pratiquent et que, dialogiquement, il correspond plus ou moins au projet formé par les touristes». C'est une fois que la discipline du marketing va s'intéresser au concept de destination qu'un élargissement de sa conception va se constater. Sans occulter la dimension spatiale cette discipline va approcher la destination touristique comme un produit auquel peuvent s'appliquer les stratégies et les techniques de marketing applicables aux produits industriels. C'est déjà un géographe en la personne de Richard Butler qui a été le premier à présenter en 1980 un modèle, inspiré de la théorie du cycle de vie du produit, mettant en perspective la naissance et le déclin de la destination en fonction de l'évolution du nombre des visiteurs dans le temps. D'autres auteurs vont suivre cette nouvelle voie ouverte par Buttler et s'intéresser à la destination comme produit, notamment Tocquer et Zins (1999) qui définissent la destination touristique comme un type de produits et services touristiques issus de nombreux opérateurs privés et publics et Davidson et Maitland (2002) qui considèrent la destination comme un lieu de production de produits touristiques concentrant l'attention et les motivations des visiteurs. D'autres auteurs

comme Kye-Sung (1990), Etchner et Ritchie (1991), Stabler (1988), Telisman-Kosuta (1989) et Gallarza et al. (2002), quand à eux vont aborder la définition de la destination à partir de l'image que cette dernière renvoie à ses visiteurs. Dans cette perspective, la destination peut être un site, un village, une ville, une région, un pays ou une zone et la territorialité de la destination dépendra à la fois de l'effort de communication entrepris par cette dernière en sus des expériences vécues. De ce point de vue, le concept acquiert une dimension vitale dans la construction identitaire du territoire, dans la mesure où «la destination devient un lieu qui prend la dimension de destination», la marque de ce lieu (Gibson et Tiard, 1999, p. 79).

Comprendre le concept de destination touristique implique donc de faire la synthèse entre une approche géographique (la destination est avant tout un lieu qu'un ou plusieurs touristes ont décidé de visiter) et une approche marketing pour laquelle la destination est un territoire commercialisé. Dans cette perspective, la destination est appréhendée comme un territoire qui peut-être en lui-même un produit touristique, c'est-à-dire un ensemble de services supports (hôtellerie, restauration, transport...) qui gravitent autour d'éléments attractifs et qui, dans son ensemble, peut répondre aux besoins d'un segment de touristes (Botti et al., 2008).

Les travaux modernes issus des disciplines du management et de la gouvernance quant à eux ont élargi le champ de conceptualisation de la destination pour la rehausser au stade de système (Beritelli et al, 2007, Ruhanen et al, 2010, Laws et al. 2011, Zhang et Zhu, 2014). A ce titre Hazbroucq (2007) considère la destination comme «un « macroproduit » touristique qui repose sur un système complexe composé d'acteurs hétérogènes et disséminés sur le territoire» (Hazbroucq, 2007, p.121). Scott et al., (2008) avancent, de leur côté, qu'une destination peut être considérée comme «un groupe d'acteurs inter-reliés intégrés dans un réseau social. Ce groupe d'acteurs, malgré leur indépendance sont interdépendants dans leur recherche de la réponse aux besoins des touristes et visiteurs et participent tous à la fourniture de l'expérience clients. L'interaction de ces acteurs est complexe, dynamique et soumise à des chocs externes».

La compétitivité, un concept polysémique:-

Comme l'affirment Tixier et Mathe (1996), le souci de compétitivité occupe une position centrale parmi les éléments fondateurs des orientations politiques de l'entreprise. Pour cette raison, toute entreprise dont les activités veulent s'inscrire dans la durée doit intégrer cette notion dans son vécu quotidien. Le concept de compétitivité est un concept central dans la recherche économique et managériale. Issu en premier lieu du monde de l'entreprise où il s'appliquait à un processus industriel, à un produit ou à une firme et où il désignait la capacité de l'entreprise à réaliser des gains de parts de marché par rapport à ses concurrents ou à sa "capacité à réaliser des performances supérieures à la moyenne » (Marniesse et Filipiak, 2003) et comme « résultant de la capacité de l'entreprise à maintenir ses performances sur le long terme » (Muchielli, 2002). La compétitivité de l'entreprise est ainsi fonction de ses performances commerciales et se définit par rapport à sa position sur les marchés vis-à-vis de ses principaux concurrents. La notion de compétitivité fait allusion directe à celle de concurrence. Ainsi, être compétitif, c'est «être apte à affronter dans des conditions favorables la concurrence qui s'exerce dans un domaine de la vie économique et sociale».

Sur le plan des corpus théoriques managériaux ayant admis l'importance de la compétitivité, notre examen de la littérature nous montre que les principales approches se sont plus focalisées sur l'identification et l'analyse des sources et des facteurs de la compétitivité des entreprises. Ces corpus peuvent être distingués en deux principales approches : l'approche unidimensionnelle et l'approche pluridimensionnelle. La première approche est représentée par les travaux du célèbre bureau de consulting le Boston Consulting Group (B.C.G.) dont les adeptes voyaient en les *coûts* la principale source de compétitivité. En effet, pour le BCG, «dans un milieu concurrentiel, l'entreprise compétitive est celle qui a les coûts les plus bas, c'est à dire celle qui a su parvenir à l'utilisation la plus efficace des facteurs pour des coûts de facteurs équivalents et ce par le biais de l'expérience » (BCG, 1988). On peut déduire de cela que pour ce courant la compétitivité d'une entreprise se traduit exclusivement par la compétitivité de ses produits. Cette approche a été vivement critiquée par les chercheurs qui la considéraient d'une part réductrice pour avoir ramené la compétitivité à la seule dimension coût alors que d'autres dimensions telles que la qualité des produits, l'image du marque, les facilités de paiement sont à envisager dans une lutte concurrentielle et d'autre part sa non pertinence en matière d'analyse des coûts qu'elle aborde de manière séquentielle sans tenir compte des liaisons entre les activités et leur impacts sur ceux-ci. Ainsi, à la dimension coûts il est judicieux de tenir compte d'autres dimensions hors coût pour représenter la compétitivité. De l'autre côté, la lecture de la littérature concernant l'approche pluridimensionnelle nous renseigne que les tenants de cette approche ont développé plusieurs schémas d'analyse des sources, des facteurs et des déterminants de la compétitivité. En premier lieu, M. Porter

(M.E. PORTER 1985) dans son modèle de la chaîne de valeur a mis l'accent sur l'importance des liaisons entre les différentes activités comme sources de l'avantage concurrentiel de l'entreprise. De leur côté les adeptes de l'approche basée sur les ressources (RVC), (Wernerfelt, 1984 ; Rumelt, 1984 et Barney, 1986 et 1991), considèrent que l'avantage concurrentiel est à rechercher en interne. Ainsi, en concevant l'entreprise comme un portefeuille de ressources et non comme un portefeuille de Produits/Marché les tenants de l'approche ressources et compétences avancent que ce ne sont plus les besoins des clients qui déterminent la stratégie, mais se sont les ressources et les compétences que l'entreprise possède et l'acquisition de l'avantage compétitif provient de la combinaison des ressources au sein de l'entreprise et leur traduction en termes de stratégie à mettre en œuvre sur les marchés des biens. Cette approche considère que les entreprises sont capables d'accumuler des ressources et des compétences qui se transforment en avantage sur les concurrents si elles sont rares, créatrices de valeur, non substituables et difficiles à imiter (Barney, 1991; Dierickx et Cool, 1989). Ces ressources désignent généralement des actifs possédés de façon permanente ou contrôlés par l'entreprise en vue de concevoir et de mettre en œuvre sa stratégie. Barney (1991) les distingue en trois principales catégories: le capital physique, le capital organisationnel et le capital humain. Plus tard, trois autres catégories se sont venues s'ajouter en l'occurrence les ressources financières, les ressources technologiques (savoir-faire, brevets...) et les ressources réputationnelles (marques, notoriété...). Les compétences désignent de ce fait, la capacité organisationnelle de l'entreprise à pouvoir déployer ces ressources ou actifs sous forme de combinaisons efficaces et efficientes pour atteindre son objectif.

Certaines autres approches se sont attaquées à la question à travers un autre angle de vision notamment en mettant l'accent plus sur le processus causal de création de l'avantage compétitif plutôt que sur les ressources, les compétences ou les activités. A ce titre Lawrence et Lorsh (1986) estiment que la compétitivité d'une entreprise est principalement associée au degré de flexibilité de sa structure et sa capacité d'adaptation aux exigences de son environnement. Pour ces auteurs, rester compétitif exige une grande capacité d'adaptation aux changements de l'environnement qui se manifeste à travers d'une part l'adaptation de la structure organisationnelle, l'adaptation des compétences des dirigeants et l'insertion compétitive de l'entreprise dans son environnement. De leur côté, Hamel et Prahalad (1994), stipulent, contrairement aux avancées de porter qu'ils ont jugé trop défensives et à usage exclusif des entreprises dominantes et aux travaux de l'approche ressources qu'ils ont jugé trop centrés sur l'interne négligeant les mutations de l'environnement et des bases de la concurrence mais aussi de l'approche basée sur l'adaptation prônée par Lawrence et Lorch, que la compétitivité suppose une vision volontariste et offensive de la stratégie d'entreprise. Ce qu'ils qualifient d'*intention stratégique* (Hamel et Prahalad, 1994). En effet, selon ces auteurs, pour réussir et détenir un avantage compétitif, une entreprise ne doit pas s'adapter à son environnement mais le changer, le transformer à son profit. L'art du stratège est de modifier les règles du jeu concurrentiel et non de s'y conformer et le changement n'est plus une contrainte mais un objectif. Pour ces deux auteurs la stratégie ne se conçoit pas sans intentionnalité et l'entreprise doit avoir une ambition permanente de compétitivité partagée par tous ses membres, non directement reliée aux ressources actuelles détenues et permettant d'avoir un point de repère face à un environnement turbulent tout en créant une tension créatrice de valeur et de changement basée sur une vision claire des dirigeants permettant d'identifier les ressources et compétences manquantes afin d'atteindre le développement future souhaité.

Cette notion de compétitivité après avoir conquis le monde de l'entreprise va voir s'élargir son horizon dans les années 1980 pour s'appliquer à d'autres entités et dimensions, notamment le secteur, la branche, le territoire, la région et la nation. En effet, s'il est possible de s'interroger sur la compétitivité d'une entreprise, il est aussi possible de s'interroger sur celle d'une branche industrielle, d'un territoire ou encore de l'économie d'une nation, etc. Ainsi, de plus en plus va émerger une approche que l'on peut qualifier de macroéconomique de la compétitivité qui s'intéresse aux territoires et à la nation tout entière. Et de nombreuses définitions vont être données par les chercheurs à cette compétitivité. Ainsi, Debonneuil et Fontagne, 2003, p. 8) définissent la compétitivité d'une nation ou d'un territoire comme la « *Capacité à améliorer durablement le niveau de vie de ses habitants (...) et à leur procurer un haut niveau d'emploi et de cohésion sociale* ». Dans la même veine D'Andrea Tyson la conçoit comme l' « *Aptitude à produire des biens et des services qui satisfont au test de la concurrence sur les marchés internationaux et à augmenter de façon durable le niveau de vie de la population* ». De sa part l'OCDE présente la compétitivité comme « *la latitude dont dispose un pays évoluant dans des conditions de marché libre et équitable pour produire des biens et services qui satisfont aux normes internationales du marché tout en maintenant et en augmentant simultanément les revenus réels de ses habitants dans le long terme* ».

Cette transposition du terme «compétitivité» pour qualifier des entités géographiques a été très critiquée par plusieurs auteurs, en particulier par le prix Nobel Paul Krugman et M.E. Porter. Pour Krugman, «parler de la

compétitivité des pays n'aurait pas de sens » et « la plupart de ceux qui utilisent le terme de compétitivité ne s'y sont même pas arrêtés pour réfléchir à la chose » (Krugman, 2001, p. 20). De son côté, Porter estime que « chercher à expliquer la 'compétitivité' au niveau national est incorrecte. Ce qu'on doit comprendre, ce sont les déterminants de la productivité et le taux de croissance de la productivité. Pour trouver des solutions, on devrait focaliser, non pas sur l'ensemble de l'économie, mais plutôt sur des industries spécifiques ou encore des segments d'industrie" (Porter, 1990). Et il ajoute des années plus tard qu'« il n'y a même pas de définition admise du terme compétitivité appliquée à un pays. La notion de compétitivité est claire quand elle s'applique à une entreprise pas quand elle s'applique à un pays » (Porter, 1999, p.167). La majorité des auteurs ayant critiqué cette transition remettent en cause principalement l'incompatibilité des définitions de la compétitivité des territoires et des nations, elles mêmes porteuses de contradictions, avec les types de compétitivité de l'entreprise mais surtout les difficultés de l'utilisation d'outils conceptuelles identiques et les difficultés méthodologique d'établir des critères identiques pour mesure (Delaplace, 2011). Malgré tout cela, Aiginger, (2006) estime que même en présence de certaines controverses sur le concept, les travaux théoriques et les débats qui entourent la compétitivité des nations ont permis de faire émerger, une définition consensuelle

Somme toute, notre examen de la littérature traitant du concept de compétitivité nous révèle la nature polysémique, ambiguë, complexe et multidimensionnelle du concept et l'absence d'une définition unique et unanime du terme. Marniesse et Filipiak (2003) considèrent que « la compétitivité est une notion encore mal cernée ». Porter (1990) stipule que « son ambiguïté découle de l'existence d'une variété de définitions qui rend difficile une définition complète et indiscutable de la compétitivité ». Quand à Scott et Lodge (1985), ils expliquent cette complexité par la nature multidimensionnelle et relative du concept même de compétitivité. Par ailleurs, il ressort aussi que le concept est relatif et dynamique. Relatif du fait qu'il change avec le niveau de l'analyse menée (nation, secteur, entreprise), le bien analysé (bien homogène ou différencié), et enfin l'objectif de l'analyse et dynamique dans le sens où la compétitivité n'est pas un concept statique et est étroitement lié aux conditions économiques du pays ainsi que les conditions du marché international.

La compétitivité des destinations touristiques : entre recherche académique et modèles opérationnels:-

Malgré que le tourisme soit devenu un secteur économique très important dans la compétitivité des pays et des territoires, il est paradoxal de relever qu'il est resté un des parents pauvres de la recherche scientifique et un domaine d'étude récent. Certes, les difficultés à l'appréhender compte tenu de l'absence d'une définition unanime et universelle, les mythes et les préjugés qui lui sont associés et les difficultés de mesure de sa productivité ont fait que les économistes ont longtemps tardé à s'y intéresser et à lui accorder la place qu'il mérite comme secteur économique à part entière et comme sujet de recherche. C'est ainsi, que c'est à partir des années 1970 que les économistes et après eux les géographes ont commencé à l'étudier après que les services et plus particulièrement le tourisme sont devenus très importants dans les PIB des pays développés. Les sciences de gestion vont emprunter la voie à ces disciplines mais plus tardivement vers la fin des années 1980. Reconnu comme multiforme et multisectoriel est donc multi-produit et multi-métier (Botti et Peypoch), le tourisme nécessite pour sa compréhension et son développement de faire appel à plusieurs facettes du management (stratégie, politique, marketing, gestion des ressources humaines, gestion financière,...). Cet engouement vers le secteur va rapidement fait naître un foisonnement de travaux de recherche d'abord en Amérique du Nord (Bornhorts, Brent Richie, et Sheehan, 2010; Minguzzi, 2006) puis en Europe (Escadafal (1997), Dewailly (2006), Lozato Giotart et Balfet (2004) et Maunier (2007)). Rapidement l'intérêt pour la gestion et la compétitivité des destinations touristiques va passer au premier plan de l'intérêt des chercheurs après que celles-ci vont être reconnues d'une part, comme les unités de bases sur lesquelles repose la compétitivité du tourisme en général et des territoires en particuliers et d'autre part, de la prise de conscience que la concurrence internationale dans le secteur se fait de plus en plus entre destinations et non entre entreprises. En effet, Dwyer et al. (2000) nous enseignent qu'il est très utile pour l'industrie et le gouvernement de comprendre quels sont les points forts et les points faibles d'un pays par rapport à ses concurrents en matière de tourisme et de savoir comment et pourquoi la compétitivité d'une destination change. Teece et al. (2008), pour leur part, soulignent que la nature dynamique de la concurrence en matière de tourisme exige des destinations de combiner et gérer leurs ressources touristiques de manière à développer un avantage concurrentiel. Par conséquent, les destinations doivent faire face au défi de gérer et organiser leurs ressources rares de manière efficiente afin de fournir une expérience de vacances supérieure à d'autres expériences dans d'autres destinations (Cracolici et Nijkamp, 2008). Cet engouement pour la compétitivité des destinations touristiques va se traduire par de nombreuses recherches et travaux académiques mais aussi par le développement de modèles opérationnels sur la question.

La compétitivité des destinations touristique dans la recherche académique:-

Au niveau académique, notre examen de la littérature portant sur le sujet nous montre que de nombreux travaux de recherche ont porté sur la compétitivité touristique (Chon et al., 1991 ; Ritchie et Crouch, 1993 ; Hu et Ritchie, 1993; Gartner, 1993 ; De Keyser et Vanhove, 1994, Evans et Johnson, 1995 ; Dann, 1996 ; Pearce, 1997 ; Kozak et Rimmington, 1999 ; Crouch et Ritchie, 1999 ; Ritchie et Crouch 2000, Hassan, 2000 ; Mihalic, 2000 ; Thomas et Long, 2000 ; Gallarza et al., 2002 ; Dwyer et Kim, 2003; Kozak, 2002; Ritchie et Crouch 2003 ; Gonzalez et Falcon, 2003 ; Enright et Newton, 2004 ; Ruhanen, 2007 ; Cortes et al. 2007; Gomezel et Mihalic, 2008, Cracolici et Nijkamp, 2008.). Crouch l'une des références incontestée du domaine, a relevé dans sa revue de la littérature sur le sujet que l'analyse de la compétitivité des destinations touristiques ne peut se faire qu'en termes relatif et qu'elle peut viser l'étude d'un territoire particulier (par exemple Hong Kong pour Enright et Newton – 2004) ou l'analyse d'une des modalités de la compétitivité (la compétitivité prix pour Dwyer, Forsyth et Rao, 2002 ou le marketing de destination pour Buhalis, (2000) et Crouch, (2011).

Plusieurs constats peuvent être dégagés de cette littérature. Le premier est que ces travaux n'ont pas porté sur le développement d'un cadre théorique global définissant la compétitivité générale de la destination touristique mais ils se sont intéressés plus particulièrement à certaines destinations, à certaines formes de tourisme ou à certaines formes de tourisme dans certaines destinations. Le deuxième est la variation de l'échelle de destination retenue qui pouvait être des pays (ex : Omerzel Gomezel et Mihalic, 2008), des régions (ex : Cracolici et Nijkamp, 2008) ou des villes (ex : Clavers-Cortés et al., 2007). Le troisième est que certains travaux ont également eu des apports significatifs quant à l'opérationnalisation de la compétitivité touristique en proposant des méthodologies de mesure sophistiquées de cette compétitivité (ex : Enright et Newton, 2004). Le quatrième est que certains travaux de recherches ont porté sur des aspects particuliers de la compétitivité des destinations, y compris le positionnement des destinations (ex : Chacko 1998), les systèmes de gestion des destinations (ex: Baker, Hayzelden et Sussmann 1996), la commercialisation des destinations (ex : Buhalis 2000), la compétitivité des prix (ex :Dwyer, Forsyth et Rao 2000a, 2000b, 2000), la gestion de la qualité (ex : Go et Govers 2000), l'environnement (ex : Hassan 2000, Mihalic 2000), le tourisme de nature (ex :Huybers et Bennett 2003), la gestion stratégique (ex : Jamal et Getz 1996; Soteriou et Roberts 1998) et des voyages à forfait (ex :Taylor1995), l'image et l'attractivité des destinations au niveau touristique (ex : Dann, 1996 ; Chon et al., 1991 ; Hu et Ritchie, 1993 ; Gallarza et al., 2002 ; Gartner, 1993). Malgré ce foisonnement de travaux de recherche sur le concept de compétitivité des destinations touristiques peu d'auteurs ont tenté de lui donner une définition précise et les définitions qui existent ne montrent pas l'existence d'une définition standard, universelle et unanime. Ainsi, selon (Hassen, 2000), la compétitivité d'une destination touristique signifie «la capacité d'une destination à créer et intégrer des produits à valeur ajoutée en maintenant les ressources locales et préserver sa position sur le marché par rapport aux concurrents». Dans le même sens, Dwyer et al. (2011) retiennent que « la compétitivité d'une destination est liée à sa capacité à proposer des produits et services plus «performants » que ceux d'autres destinations pour satisfaire les besoins des visiteurs. ». De leur côté, Ritchie et Crouch (2000) la définissent comme «la capacité de la destination à créer de la valeur ajoutée et l'augmentation, par conséquent, du bien être national par la gestion des prestations et des processus en intégrant les relations entre eux dans un modèle économique et social». Adoptant une optique marketing D'Hauteserre (2000) parle de la capacité d'une destination de maintenir sa position sur le marché et de chercher l'amélioration au fil du temps afin de faire face à la concurrence. D'autres chercheurs comme Cracolici et Nijman (2008), Barney (1991); Grant, (2005) et Wernerfelt, 1984, sont allés s'attaquer à la question en se basant sur le mouvement des ressources et compétences pour expliquer et définir ce concept. Ces auteurs considèrent ainsi les destinations comme une combinaison de ressources physiques, naturelles, culturelles et humaines qui sont rares, inimitables et non-substituables et qui peuvent générer des compétences utiles pour créer et développer un avantage concurrentiel (Barney, 1991). Selon ces auteurs, la compétitivité d'une destination dépend de ses ressources tangibles et intangibles, de son management et de ses décisions.

De notre examen de la littérature académique, il paraît bien évident que la compétitivité des destinations touristique est un sujet qui a retenu fortement l'attention des chercheurs mais il apparaît également fort évident que les concepts de compétitivité en général et de compétitivité touristique en particulier, aisés à définir en apparence, posent d'importants problèmes de mesure (Cracolici et Nijkamp, 2008). Porter (1990) soulignait déjà l'ambiguïté du concept de compétitivité. Du fait de l'ambiguïté et du caractère multiforme de ce concept, les auteurs ont ainsi utilisé des méthodologies diverses pour mesurer la compétitivité touristique. Kim (1998) notait déjà que les chercheurs, pour caractériser l'attractivité d'une destination, ont utilisé une grande variété de critères, même si certains d'entre eux étaient communs à la plupart des travaux. Dwyer et al. (2011) notent pour leur part qu'un problème majeur, caractérisant toute tentative d'établir des indices de compétitivité, est lié à l'intégration de critères

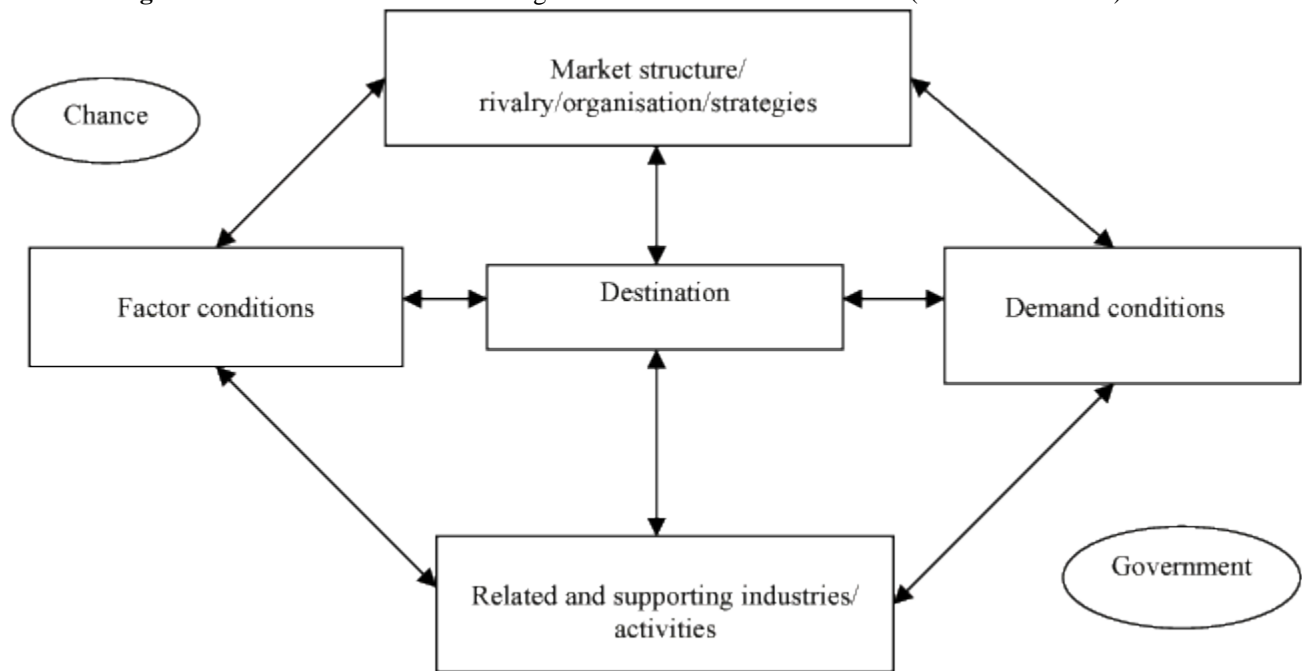
«objectifs» de compétitivité (ex: évolutions des parts de marché, nombre d'emplois issus du tourisme...) et «subjectifs (ex : richesse de la culture, qualité du service, beauté des paysages...). Il n'existe pas ainsi actuellement de méthode unanimement reconnue permettant d'intégrer simultanément ces deux types de critères (Dwyer et Kim, 2003).

Les modèles de compétitivités touristiques:-

En réalité il n'existe pas à proprement dit de modèles de compétitivités qui ont été élaborés spécifiquement aux destinations touristiques mais certains chercheurs qui ont été aussi reliés par certains organismes internationaux voulant modéliser la compétitivité des destinations touristique, sont allés, sur la base des théories économiques de l'avantage comparatif et des travaux de Porter (1990) sur les avantages compétitifs, proposés des modèles génériques visant à cerner l'ensemble des particularités et des dimensions de la compétitivité des destinations touristiques. Nous nous contenterons dans ce point de présenter l'application du modèle de Porter et de présenter l'un des modèles les plus connu, le plus influent et dominant dans la littérature à savoir celui développé par Crouch et Richie en 1999.

Le Diamant de M. Porter:-

Le modèle mis au point par M. Porter (1990) dans son ouvrage la compétitivité des nations est celui qui va être plus tard appliqué au secteur du tourisme (THR - Barcelone). Porter affirme que le succès d'une entreprise ne dépend pas uniquement de sa stratégie ni de son positionnement, mais également de son intégration au sein de son environnement. Sa thèse centrale repose sur l'idée que les régions, les destinations, les « clusters » connaissent un certain succès au niveau d'activités ou de secteurs partiels, car leur environnement local est le plus dynamique et le plus stimulant et encourage les entreprises à préserver leur avantage. Il s'agit de la thèse centrale. Dans destinations touristiques il existe des groupes de sociétés directement ou indirectement liés au tourisme et qui concentrés sur une zone géographique spécifique et fonctionnent comme des clusters. La nature cosmopolite du secteur touristique, nécessitant différentes entreprises de taille et d'activités différentes mais liées par le même marché, stimule ce processus de regroupement. Dans son modèle Porter explique que la l'avantage concurrentiel d'une nation ou d'un territoire repose sur quatre facteurs interdépendants qui, ensembles, forment un « **diamant** » selon le terme utilisé par Porter pour faire référence à ces points déterminants. Pour Smeral (1996) un point de départ pour le développement de stratégies pour l'amélioration de la position concurrentielle d'une destination touristique est à chercher dans le système dynamique constitué par les facteurs déterminant développé par Porter. Ainsi par référence au modèle de porter nous pouvons bâtir les fondements de la compétitivité de la destination. Appliqué aux destinations touristiques le modèle se présentera comme suit :

Fig. 1:- Points déterminants des avantages concurrentiels des destinations (Diamant de Porter).

Ces points déterminants sont:-

1. **Les conditions de facteurs.** Renvoient à la détention de dotations factorielles naturelles, culturelles ou artificielles (attractions naturelles, ressources culturelles et historiques, infrastructures d'hébergement, de transport NTI,..., ressources humaines, prix des facteurs, efficacité de la production,...) fortes et nécessaires à la concurrence dans le secteur du tourisme et sans les quels il ne peut y avoir d'activité touristique.
2. **Les conditions de la demande.** Élément majeur et déterminant important de l'avantage concurrentiel les conditions de la demande renvoient pour d'une destination à plusieurs éléments dont principalement, la taille du marché local, la structure et la diversité du marché, la position sur les marchés à croissance rapide, le renforcement de la culture touristique des consommateurs et des sociétés d'accueil (Cooper et al., 2001), la protection du touriste-consommateur,...
3. **Les secteurs d'activité associés et impliqués.** La position concurrentielle d'une destination dépend et peut être renforcée également par la diversité et la qualité des fournisseurs installés dans le territoire (transporteurs, services connexes de haute qualité (système financier, de télécommunication, de santé), industrie agroalimentaire, installations sportives, de loisirs et culturelles, etc.
4. **Structure du marché, rivalité, organisation et stratégie.** Ces conditions sont très importantes et renvoient à la manière dont la destination et ses éléments sont organisés et gérés notamment la disponibilité d'un plan stratégique soutenu par toutes les parties impliquées, à la fois des secteurs public et privé, d'une stratégie marketing et promotionnelle claire et efficace, une image de marque forte, des mécanismes de coordination efficaces, des organes de gestion bien structurés et gouvernés.

A ces quatre facteurs déterminants viennent s'ajouter deux autres facteurs non moins importants pour la compétitivité de la destination à savoir la chance et le gouvernement. Ces deux facteurs peuvent avoir une influence significative sur le degré de compétitivité de la destination et son succès notamment à travers d'une part les actions publiques qui peuvent être soit un facteur stimulant ou au contraire nuisant au développement du tourisme (ex : réglementation sur les hôtels, change, fiscalité, accueil et information des touristes, plan de circulation et d'aménagement, ...) et d'autre part, à travers la qualité de la gouvernance (implication ou non de toutes les parties prenantes).

A noter que le «diamant» se présente comme un système de renforcement mutuel dans le sens où il y a interdépendance entre les différents déterminants. Ainsi, par exemple, des conditions favorables de la demande n'engendreront un avantage concurrentiel que si l'état de la rivalité est suffisant pour que les entreprises (par ex., les hôtels) puissent leur répondre. De même, les avantages qui existent au niveau d'un point déterminant peuvent également créer ou améliorer les avantages d'un autre.

Le Modele De Crouch Et Ritchie:-

Le modèle considéré le plus influent en matière de compétitivité touristique au regard des chercheurs et praticiens reste celui développé par **Crouch** et **Ritchie** en 1999. Ces deux auteurs, en se basant eux aussi sur le modèle du diamant de Porter mais en soulignant les limites des travaux de recherche précédents et des modèles généraux qui ont été élaborés sur la compétitivité des destinations qui selon eux malgré qu'ils ait produits une liste assez exhaustive des déterminants de la compétitivité des destinations ces déterminants n'ont pas tous une importance ou une influence égale pour cerner toute les dimensions concurrentielles de la destination. Ces auteurs estiment que le niveau de recherche sur la question de la compétitivité des destinations est assez maintenant développée pour pouvoir passer du simple listage des attributs et déterminants de cette compétitivité pour se recentrer sur l'étude de l'importance relative de chaque attribut et déterminant. Pour eux, l'impact d'un attribut de compétitivité sur la performance relative d'une destination est fonction à la fois de l'importance de l'attribut ainsi que du degré de variation des destinations sur l'attribut et bien qu'un attribut puisse être considéré comme important, il ne sera pas un déterminant de la compétitivité s'il ya peu de différence entre les destinations sur cet attribut. Sur fond de ces limites ils vont développer un modèle conceptuel de la compétitivité des destinations touristiques qui repose sur quatre composants majeurs : « les ressources et attractions centrales », « les facteurs et ressources supports », « le management de la destination » et les « déterminants ». Les « ressources et attractions centrales » comprennent les éléments fondamentaux de l'attractivité d'une destination. Ce sont les raisons fondamentales qui expliquent pourquoi un visiteur choisit une destination plutôt qu'une autre. Les facteurs inclus dans cette composante sont la physiographie, la culture et l'histoire, les liens avec la destination, les activités, les événements et les infrastructures touristiques. La physiographie correspond au paysage et au climat, les liens avec la destination renvoient aux relations personnelles avec des résidents du pays émetteur et les infrastructures touristiques comprennent essentiellement le logement, les services de restauration, les moyens de transport et les attractions principales. L'image étant généralement considérée comme une composante importante de la compétitivité d'une destination, des attributs utilisés pour caractériser l'image comme par exemple le climat, les paysages ou l'hôtellerie, sont ainsi fréquemment utilisés pour mesurer la compétitivité touristique (Enright et Newton, 2004).

Les «facteurs et ressources supports» sont les facteurs susceptibles de soutenir le succès d'une destination au niveau touristique. Ils incluent notamment les infrastructures globales d'une destination, les établissements d'enseignement et les facteurs liés à l'accessibilité de la destination.

La troisième composante de la compétitivité touristique, à savoir le « management de la destination», se focalise sur les activités pouvant influencer les autres composantes, premièrement en améliorant l'attractivité des ressources et attractions centrales, deuxièmement en renforçant la qualité et l'efficacité des facteurs supports et enfin en adaptant les contraintes imposées par la quatrième composante à savoir les «déterminants».

Alors que les aspects de cette composante les plus étudiés sont le marketing de la destination, des auteurs estiment qu'un spectre plus large d'activités liées au management devrait être pris en compte comme l'organisation et la maintenance des ressources et attractions clefs, ainsi que les services liés à ces ressources et attractions.

La quatrième composante est les « déterminants ». Elle inclut les facteurs qui peuvent modifier, éventuellement d'une manière négative, l'influence des trois autres composantes.

Ces facteurs sont par exemple la localisation, la structure des coûts et la sécurité. Ces facteurs ne sont pas contrôlés par les acteurs du tourisme mais peuvent jouer un rôle crucial dans la compétitivité d'une destination (Enright et Newton, 2004). Crouch et Ritchie (1999) prennent en compte également dans leur modèle de la compétitivité touristique le micro et le macro environnement de la destination qu'ils estiment être inter-reliés et affectant simultanément "le cœur de la compétitivité" de la destination. Le microenvironnement renvoi aux aspects propres à la destination touristique et à ceux des acteurs principaux du tourisme dans la destination (T.O, Agences de voyages, les résidents, les touristes visiteurs, les employés des professions touristiques, les médias, les groupes de pression,...) pouvant faire l'objet de comparaison entre destinations concurrentes., les groupes d'action et de pression, les médias, les institutions financières et sociétés d'investissement, les administrations publiques, et l'environnement immédiat). Tandis que le macro-environnement concerne les éléments extérieurs à l'environnement immédiat de la destination mais pouvant exercer une certaine influence sur sa compétitivité. Il s'agit notamment, de l'attention portée par les autorités à l'environnement naturel et à sa protection, de la conjoncture économique internationale, des problèmes démographiques, de l'évolution technologique, etc.

En définitive, nous pouvons retenir que le modèle de Crouch et Ritchie donne une indication claire sur les différents éléments sur lesquels repose la concurrence entre destinations touristiques et enseigne que la compétitivité et la supériorité d'une destination sur le marché touristique dépend de sa capacité à gérer et organiser ses ressources de manière rationnelle et efficace.

Les Structures de Gestion des Destinations : Nature, Typologie et Fonctions:-

Parallèlement au développement des travaux sur la compétitivité des destinations touristiques, la question des structures organisationnelles et de leur pertinence pour la compétitivité des destinations a été au centre de la recherche sur le tourisme. Ritchie et Crouch ayant discuté cette question en 1993 (Ritchie et Crouch, 1993), puis l'ayant reprise dans leur modèle en 2003, (Crouch et Ritchie, 2003) considèrent qu'à part dans le cas spécifiques des *resorts*, aucun prestataire et acteur du territoire de la destination ne peut prétendre répondre à lui tout seul à la demande des touristes et prendre lui-même en main leur expérience globale. Selon eux, comme système d'offre la destination, même s'elle ne constitue pas une entité commerciale légale, elle est composée d'un ensemble d'entreprises, d'organisations et de groupes qui œuvrent ensemble de manière semi-organisée, en partie coopérative, mais qui sont finalement motivés en grande partie par leur propre intérêt et doit être exploitée comme une entreprise (Ritchie et Crouch, 2003, p : 96). Dans cette perspective, les deux auteurs ont mis en lumière l'impact du management de la destination et le rôle de la Destination Management Organization (DMO) dans la compétitivité des destinations (Botti et Peypoch, 2011).

Après avoir jeté les éclairages nécessaires sur la problématique de la compétitivité des destinations touristiques et les défis qu'elle impose au management de la destination, nous chercherons dans cette section, toujours sur la base d'une recherche documentaire, comment la littérature aborde les structures de management des destinations et d'identifier le degré de correspondance et de concordance de ces structures et plus particulièrement de leur rôles et fonctions par rapport aux défis de la compétitivité soulevés et des exigences d'un management efficace des destinations touristiques.

Panorama des structures du management des destinations touristiques:-

Notre propos ici n'est pas de traiter des structures de gestion dans le cadre institutionnel distinguant les organes intervenants dans la gestion du tourisme aux échelles nationales, régionales et locale mais il s'agit de présenter et de discuter des structures de gestion des destinations touristiques telles qu'elles ont été définies et présentées dans le point précédent. L'examen de la littérature sur les structures de management des destinations touristiques nous montre qu'il existe cinq types de structures allant des structures basées sur le marché aux structures de type hiérarchique. Ces différents types de structures et leurs caractéristiques sont présentés dans le tableau suivant :

Tableau n°1:- Les structures idéales des destinations touristiques.

		Structures marché	Structures en réseaux			Structures hiérarchiques
			Contrats implicites	Contrats explicites		
				Système décentralisé	Système centralisé (Entreprise Focale)	
Nombre d'acteurs		Élevé	Moyen	Moyen	Faible	
Taille des acteurs		PME	PME	PME et un grand groupe	Un grand groupe ou entreprise	
Coûts de transaction		Très élevés	Elevés	Moyens à faibles	Très faibles	
Objectifs des acteurs		Indépendant s	Interdépenda nts	Interdépenda nts	Dépendants	
Leadership de la destination		Aucun Leader dominant	Réseaux	Réseaux	Entreprises dominantes dans le réseau	
Marketing de la destination		Individual	RTO/ individuels	RTO fort / individuel	Entreprise dominante/ individuel	
Management de la destination		Absente	Collectivités locales	RTO/ gouverneme nt local	Entreprise dominante/ gouvernement local	
Coordination de la communication entre les parties prenantes		Absente	Limitée	Peu de coordination	Coordination entre les grands acteurs	
					Fortement intégrée/ stratégique	

Source : Réalisé sur la base des travaux de Powel (1990) et de Bieger (2006)

Comme mentionné ce tableau distingue cinq types de structures de gestion des destinations allant de la structure du type marché, caractérisée par la présence d'un grand nombre d'entreprises pour l'essentiel de petites taille offrant individuellement leur services aux touristes, poursuivant leur propres objectifs, menant leur propres actions marketing et commerciales et se livrant à une concurrence intense sans aucune coordination à l'autre extrême de type corporate modèle dominant en Amérique du Nord et caractérisé par l'existence d'une société ou groupe privé («business corporation» selon Flagestad, 2001) dominant qui, en agissant au nom de la collectivité, cherche à atteindre des objectifs d'efficience et de profit à travers la réduction des coûts de transaction en internalisant tous les services nécessaires aux touristes et opère par intégration ou par contractualisation avec les autres sociétés de services qui sont généralement de petites tailles. Ce groupe détient le pouvoir politique de prise de décision en matière de gestion, de développement et de commercialisation de la destination et tous les autres acteurs et parties prenantes de la destination, y compris la sphère publique, dépendent de ce groupe qui opère à leurs égard par un marketing et une communication stratégiques et hautement intégrés. Entre ces deux types de structures extrêmes, il existe tout un continuum de configurations possibles épousant des formes de réseaux plus ou moins formalisé du point de vue opérationnel et de coordination. Dans ces formes en réseau la forme des contrats implicites caractérise les destinations dirigées par un grand nombre d'acteurs sous la coordination d'un organisme touristique régional (OTR). Dans ce type de structure, l'absence d'une réelle coordination formelle fait que les coûts de transaction restent assez élevés et le comportement coopératif des acteurs est dicté plus par la nécessaire interdépendance de leurs activités et la peur de sanctions en cas de concurrence déloyale et non par l'engagement volontaire dans une stratégie de coopération (Williams, Harkins et Latane, 1981). Egalement dans ce type de destination c'est en général l'autorité publique locale qui assure la fonction du management alors que le marketing et la commercialisation sont assurés par les entreprises et le cas échéant par l'organisation touristique régionale. Dans d'autres destinations les relations entre les différents acteurs peuvent être formalisées par une contractualisation explicite. Cette contractualisation favorise la coopération et la coordination réduisant les risques et les coûts de transaction et rendant la gestion et la commercialisation de la destination plus efficace (Granovetter, 1973). Ce type de structure admet deux principales variantes. Soit un système de nature décentralisé dans le quel les entreprises, dont la grande majorité sont des PME, se constituent en réseau qui prend en charge la gestion et la promotion de la destination en s'aidant ou non par organisme touristique régional. Soit un système de nature centralisé avec la présence d'une entreprise dominante qui peut mobiliser assez de ressources et de pouvoir et qui joue le rôle de leadership et fixe les stratégies de développement et de promotion de la destination qui sont suivies par les autres acteurs. Dans ce type de structure étant donné que cet acteur stratégique ne peut à lui tout seul assurer tous les services et toute l'expérience aux touristes et visiteurs il reste dépendant en partie des autres prestataires avec lesquels il doit collaborer et coordonner les actions de communication et de marketing.

Fonctions principales de gestion des destinations:-

En retenant uniquement les travaux de recherche et les études qui ont porté sur le management des destinations, les principales fonctions de ce management répertoriées selon l'ordre de leur nombre et fréquence de citation par les différents auteurs sont reprises dans le tableau suivant.

Tableau n° 2:- Fonctions du management de la destination.

Fonctions	Auteurs
Marketing de la destination, branding et positionnement	Aberg (2014), Anderson (2000), Crouche et Richie (1999), Harril (2005), Jamieson (2006), Lasser et Beritelli (2013), Longjit et Pearce (2013), Morrisson (2013), Pearce et Schänzel (2013), Pearce (2014b), Pechlaner, herntrei et Kofink (2009) et OMT (2007)
Planification, suivi et évaluation des destinations	Aberg (2014), Anderson (2000), Crouche et Richie (1999), Dweyr et Kim (2003), Fuchs et Weiermair (2004), Jamieson (2006), Lasser et Beritelli (2013), Morrisson (2013), Pearce (2014b), Rygllová (2008) et OMT (2007).
Développement des produits/services	ARTN (n.d), Ivanis (2011), Jamieson (2006), Lasser et Beritelli (2013), Morrison (2013), Pearce (2014b), Pechlaner et al (2009), Risteski, Kocevski et Arnaudove (2012) et OMT (2007).
Recherche, gestion de l'information et renforcement des connaissances	Anderson (2000), ATRN (n.d.), Crouch et Richie (1999), Harrill (2005), Morrison (2013), Pavlovitch (2003), Pearce (2014b) et OMT (2007).
Gestion des ressources, gestion de l'environnement	Crouch et Richie (1999), Dwyer et Kim (2003), Fuchs et Weiermai (2004), Longjit et Pearce (2013), Pavlovitch (2003), Risteski et al. (2012) et Rygllová (2008).
Gestion des visiteurs, gestion de l'expérience des visiteurs, gestion des risques d'aventure, gestion de la sécurité	Anderson (2000), Crouch et Richie (1999), Longjit et Pearce (2013), Pavlovitch (2003), Pearce et Schänzel (2013) et Risteski et al. (2012)
Construction de relations	Crouch et Richie (1999), Dwyer et Kim (2003), Jamieson (2006), Morrison (2013) et OMT (2007).
Développement des ressources humaines, formation	Dwyer et Kim (2003), Longjit et Pearce (2013), Pearce (2014b) et OMT (2007).
Responsabilité organisationnelle, leadership et partenariat	Anderson (2000), Crouch et Richie (1999), , Jamieson (2006), Risteski et al. (2012)
Lobbying	Lasser et Beritelli (2013)
Coordination du service	Lasser et Beritelli (2013),
Fourniture et diffusion d'Informations	Pearce et Schänzel (2013)
Régulation et canalisation de la pression touristique	Laws (1995)
Gestion du cycle de vie	Sainaghi (2006)
Gestion de problèmes particuliers (par exemple, capacité de charge)	Sainaghi (2006)
Soutien aux entreprises	OMT (2007)

Source: Elaboré sur la base des travaux de Douglas G. Pearce (2015)

De la lecture de ce tableau nous pouvons relever en premier que le management des destinations touristiques, compte tenu de la complexité et de la multi-dimensionnalité de celle-ci exige un grand nombre de tâches et de fonctions portant sur plusieurs aspects. Ces fonctions, telles qu'elles ressortent de ces travaux et études, vont du marketing et de la commercialisation, à la planification de développement de la destination et de la formulation des stratégies, à la gestion des ressources et de l'environnement, au développement de l'offre et des produits et activités, la recherche et l'innovation, le suivi et l'évaluation et d'autres éléments spécifiques, la gestion de l'information et des connaissances, la gestion des touristes et visiteurs mais aussi à la gestion et de la collaboration et de la coordination entre les différents acteurs et les différentes parties prenantes agissant dans le territoire de la destination. Certaines autres dimensions aussi importantes dans la gestion des destinations ont été peu citées par les auteurs, tel est le cas du lobbying, l'information des touristes et visiteurs, la gestion du cycle de vie de la destination, la gestion de la capacité de charge, la régulation et la canalisation de la pression touristique et le soutien aux entreprises des différentes branches du secteur touristiques.

L'autre constat fondamental à relever de cette lecture de la littérature sur la question est que malgré l'existence d'un certain accord entre les chercheurs sur nombre de fonctions du management de la destination il reste qu'aucun de

ces travaux et de ces recherches n'a discuté ni la question de la pertinence, de la viabilité et de l'efficacité de telle ou telle de ces fonctions, ni celles qui sont effectivement réalisées dans les destinations.

Les fonctions de l'organisme de gestion de destination (OGD ou DMO):-

Plusieurs autres chercheurs se sont concentrés non pas uniquement sur les fonctions du management de la destination mais sont allés se poser des interrogations sur celles qui devraient être assurées par les organes ou structures de gestion de la destination, appelées généralement Organisme de Gestion des destinations (OGD) ou par son équivalent anglais *Destination Management Organisation* (DMO). Recherchant à identifier les rôles, formes et fonctions de ces structures, les auteurs se sont heurtés en premier lieu à la problématique de cette appellation de DMO. En effet, dans sa conception anglo-saxonne le 'M' dans le terme DMO renvoi au marketing plutôt qu'au management. Alors que dans son acception la plus courante, notamment celle de l'OMT, le terme DMO est désigné comme Organisme de Gestion des Destinations. D'autres auteurs sont même allés utiliser les deux acceptations pour parler d'organisme de gestion et de marketing des destinations. Il est à noter qu'au fond cette querelle entre auteurs ne porte pas sur le côté sémantique du terme mais plutôt sur l'intérêt de préciser exactement ce que reflète l'appellation en termes de fonctions et d'activités de base assurées par cette organisation dans le cadre d'une destination. Comme pour le cas des fonctions du management des destinations nous présenterons les fonctions de l'OGD de la destination en fonction de l'ordre et fréquence de citation par les différents auteurs. Les résultats de notre lecture de la littérature sur cette question sont repris dans le tableau suivant.

Tableau n° 3:- Les fonctions de l'OGD à l'aune de la littérature.

Fonctions	Auteurs
Marketing de la destination, Branding et positionnement	Baggio (2008), Bieger et al (2009), Bornhorst, Ritchie et Sheehan, Bramwell et Rawding (1994), Crouch et Richie (1999), Elbe et al (2009), Gretzel et al (2006), Heath et Wall (1999), Osmankovic et al (2010), Pechlaner et al. (2012), Presenza et al. (2005), Prideaux et Cooper (2002), ROS Développement et planification (2008), Sheehan, Richie et Hudon (2007) et Socher (2000))
Renforcement des relations / coordination / facilitation	Bornhorst et al. (2010), Crouch et Richie (1999), Elbe et al. (2009), Heath et Wall (1992), Presenza et al. (2005), Prideaux et Cooper (2002), Sheehan et al (2007) et OMT (2007)
Développement des produits/ développement des activités	Baggio (2008), Bieger et al. (2009), Bornhorst et al. (2010), Osmankovic et al (2010), Pechlaner et al. (2012), Socher (2000) et OMT (2007)
Planification, formulation de la stratégie, suivi et évaluation	Baggio (2008), Bornhorst et al. (2010) , Heath et Wall (1992), Pechlaner et al. (2012), Jenkins et al (2011) et OMT (2007).
Management des ressources, management de l'environnement	Bornhorst et al. (2010), Presenza et al. (2005) et ROS Développement et planification (2008)
Développement des ressources humaines, Formation	Presenza et al. (2005), ROS Développement et planification (2008) et OMT (2007).
Management de la destination	Bornhorst et al. (2010), Crouch et Richie (1999) et Gretzel et al (2006).
Assurance qualité	Osmankovic et al (2010) et ROS Développement et planification (2008).
Information et réservations	Bieger et al (2009), ROS Développement et planification (2008) et OMT (2007).
R&D, Système d'information, management et développement des connaissances	Osmankovic et al (2010) et Presenza et al. (2005)
Gestion des visiteurs, gestion de l'expérience clients	Bornhorst et al. (2010) et Presenza et al. (2005)
Fourniture de services, coordination	Bornhorst et al. (2010) et ROS Développement et planification (2008);
Soutien aux entreprises	Prideaux et Cooper (2002) et ROS Développement et planification (2008)
Prise de décisions ou exécution	Baggio (2008) et Bornhorst et al. (2010)

Management des crises	Presenza et al. (2005)
Aide à l'accès au financement	Presenza et al. (2005)
Améliorer le bien-être des résident	Bornhost et al. (2010)
Animation	ROS Développement et planification (2008)

Source: Elaboré sur la base des travaux de Douglas G. Pearce (2015)

Nous pouvons à la lumière de ce tableau constater que la fonction marketing de la destination se distingue comme le propre du travail et de la mission d'un OGD, suivie de l'établissement des relations et de la coordination entre acteurs et par la suite de l'activité développement des produits et de l'offre. Les autres tâches relatives à la planification, la stratégie de développement de la destination, la gestion des ressources, le soutien aux entreprises et l'aide à l'accès au financement,..., ne sont que peut traitées par les différents auteurs quand il s'agit du rôle et des fonctions de l'OGD. Cela nous amène à formuler la conclusion que, malgré qu'elle est citée par quelques auteurs, la fonction du management de la destination apparaît comme une fonction distincte ne relevant pas des attributions d'un OGD.

Un autre constat à relever de ces études est que, comme pour le cas du management des destinations, hormis quelques tentatives, les auteurs sont restés muets sur les fonctions effectives exécutées par les OGD et se sont plus intéressés à ce qu'ils devraient ou pourraient faire. Les quelques études empiriques menées en Amérique du Nord et en Europe (Presenza, Sheehan et Ritchie, 2005 ; Getz et al., 1998 et Gretzel et al., 2006, ROS Development and Planning, 2008) ont toutes montré que l'amalgame entre la fonction du marketing de la destination et l'intégration de la gestion de la destination par un OGD ont abouti à l'échec jusqu'au point où Pike et Page (2014, p. 205) sont allés affirmer que: «L'utilisation de l'organisation de gestion des destinations comme un descripteur générique est inutile dans l'ajout de clarté et de but à la discussion du rôle des OGD, car il confond le besoin perçu de gestion avec la fonction largement de commercialisation qu'ils entreprennent effectivement... un DMO est un organisme responsable de la commercialisation d'une destination touristique identifiable avec une frontière géographique explicite». Cette affirmation nous montre une fois encore que la fonction de gestion de la destination et à distinguer de celle du marketing de la destination et donc elle ne fait pas partie de la mission et des attributions de l'OGD. Cette conclusion contredit aussi la conception de l'OMT qui dans l'une de ses publications a défini l'OGD comme "une entité de premier plan qui coordonne la gestion de tous les éléments intégrant une destination (lieux d'intérêt, équipements, facilités d'accès, moyens de transport, hébergements, etc.) et des activités telles que le développement de produits, leur marketing et l'établissement de leurs prix. L'OGD fait le lien entre les diverses autorités, les parties prenantes et les groupes d'intérêt. Il facilite également les partenariats et les synergies dans le cadre d'une stratégie cohérente visant à améliorer le développement, la gestion et la promotion de la destination. Il existe de nombreux types d'OGD en fonction des besoins et des spécificités de chaque destination à l'échelon national, régional et/ou local." (OMT, 2007)

A ce stade est sur la base des conclusions de notre lecture de la littérature existante sur la question, il nous semble pertinent de se poser la question sur qui devrait assurer et exercer les fonctions de gestion de la destination ?

Certains éléments de réponse à cette interrogation sont à chercher dans les travaux et études d'un certains nombre d'autres chercheurs qui ont essayé d'étudier la question en interpellant le rôle joué par la sphère publique dans le développement et la gestion des destinations. En effet, ces auteurs mettant l'accent sur la prise de conscience, par les acteurs publics territoriaux, de l'importance du tourisme pour le développement et la promotion des territoires et régions et leur implication de différentes manières dans le secteur (Aménagement, réalisation d'infrastructures, élaboration de stratégies, gestion de biens collectifs, incitations et soutien aux investisseurs et aux entreprises,...) ont commencé à s'intéresser aux fondements, formes et modalités de cette implication et aux rôles et fonctions que devrait assurer cette sphère publique dans le management de la destination et le degré de leur efficacité. L'intérêt porté à cette nouvelle variable va élargir le champ des investigations sur la gestion des destinations touristiques du simple contexte sectoriel et professionnel vers un contexte plus large intégrant tous les acteurs et toutes les parties prenantes présents sur le territoire de la destination. Ainsi, une importante littérature va se développer sur les modalités juridiques de cette gestion (Dredge et al., 2011 et Jenkins et al., 2011), les fonctions à assurer par la sphère publique (Bramwell et Rawding, 1994), le partage des responsabilités (Longijt et Pearce, 2013) et la problématique de la coordination (Palmer, 1996) et de la gouvernance (Barrado Timón, 2004, Gerbaux et Marcelpoil, 2006 et Marsat et al., 2010), de la durabilité (Dredge et al. 2011). Toutefois, la littérature note que, mise à part l'étude de Marsat et al. (2010), qui ont pu développer une approche globale et multifonctionnelle de la gestion de la destination et ont examiné les liens entre les fonctions de la sphère publique locale et le management territorial, le produit de ce

courant de pensée est resté très diffus et cloisonné sur les fonctions spécifiques (marketing et commercialisation, aménagement, développement durable,...) et n'a pu se soulever au stade d'une véritable théorie de gestion des destinations.

Conclusions et implications theoriques:-

Notre revue de la littérature nous a montré que la question de la compétitivité des destinations touristiques est une question centrale dans les travaux de recherche en tourisme. Toutefois, les concepts de destination, de compétitivité que de compétitivité de destination, malgré la foisonnante littérature de recherche qui leur a été consacrée restent encore entachés d'un certain flou et ambiguïté. La compétitivité de la destination apparaît ainsi comme la finalité recherchée mais qui partage encore les chercheurs et les organismes intéressés par le tourisme sur les dispositifs et les stratégies à mettre en œuvre ainsi que les formes et les styles de gestion à adopter pour la booster. Une première réalité qui émerge clairement de cette littérature est que face aux défis imposés par la compétitivité le management de la destination touristique n'est pas une tâche aisée et nécessite une multitude de fonctions et fait intervenir un grand nombre d'acteurs. Vu comme système, la destination est composée d'un complexe d'acteurs indépendants mais en même temps interdépendants dans leurs activités et participant tous à la délivrance de l'expérience aux touristes et aux visiteurs et générant de la valeur pour le territoire et ses différentes parties prenantes. L'autre conclusion majeure qui se dégage de notre examen de la littérature est l'absence d'un modèle unique et unanime relatif aux fonctions à assurer et les structures qui devraient les assurer. En effet, nous avons ainsi pu relever que si les chercheurs identifient une palette très large de fonctions, tâches et activités que nécessite le management de la destination, ils ne reconnaissent qu'un nombre limité de ces fonctions, tâches et activités lorsqu'ils abordent les organismes de gestion des destinations ou la sphère publique. S'il est reconnu que les défis imposés par la compétitivité exigent pour une destination touristique la nécessité de coordonner et d'intégrer de multiples fonctions et d'acteurs aux intérêts fortement divergents, il est aussi de relever la grande variété des situations et des types de destinations touristiques et des contextes de leur émergence et développement et la nature et le nombre des acteurs impliqués dans leur gestion et promotion qui sont autant de contraintes pour les chercheurs les poussant à adopter des postures et des approches différentes ou de se concentrer sur certains aspects plutôt que d'autres.

La troisième conclusion importante à tirer de cette synthèse de la littérature et qui est directement liée à notre problématique principale de cette recherche est que les principaux défis imposés par la compétitivité même s'ils ne semblent pas être des préoccupations primordiales parmi les fonctions des structures de management des destinations touristiques comme c'est le cas par exemple des fonctions marketing, sont plus cités comme des fonctions de management de la destination et dans une moindre mesure parmi celles de l'OGD. Nous pensons que les considérations relatives à l'expérience client, à l'image de marque, au numérique, à la gestion des connaissances et au développement durable, malgré qu'elles soient au centre des préoccupations de nombreux chercheurs et qui les considèrent comme des déterminants de l'attractivité et de la compétitivité des destinations touristiques, elles sont encore insuffisamment modélisées pour le cas de ces dernières et qu'il s'agit de préoccupations incombant à toutes les parties prenantes de la destination prise comme système ou comme une entreprise virtuelle au sens de Botti (Botti, 2009).

En définitive, nous pensons que l'absence d'un corpus théorique complet et bien établi, aussi bien sur la gestion et la compétitivité des destinations touristiques que plus particulièrement sur les structures de gestion et leurs fonctions, traduit le caractère récent de l'intérêt pour ces domaines de la part de la science du management et dénote de leur caractère complexe et multidimensionnel toutefois, ils laissent aussi la porte grandement ouverte pour les chercheurs pour développer des recherches et études plus poussées sur les différentes dimensions du management stratégique et opérationnel des destinations touristiques.

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RESEARCH ARTICLE

BANANA PEEL EXTRACTS FOR THE PRODUCTION OF SINGLE CELL PROTEIN BY USING *SACCHAROMYCES CEREVISIAE*.

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Banana peels, protein deficiency,
Saccharomyces cerevisiae, single cell
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Abstract

Protein deficiency is a state of malnutrition which results from eating a diet in which proteins are not enough or present in an inadequate amount. It is a major factor responsible for a variety of ailments including mental retardation, kwashiorkor and even death, with millions of affected people as it increases an individual's susceptibility to and severity of infections. The escalating prices of traditional protein sources have intensified this problem. The present study aims to produce low cost protein by using waste material like banana peels which can alleviate protein deficiency of masses while considerably reducing environmental pollution. Bioconversion of waste banana peels into valuable product like single cell protein (SCP) has the potential to solve the problem of protein deficiency worldwide. Here, banana peels were used as substrate; peels were treated with hydrochloric acid 10% (w/v), followed by inoculation with pure culture of the yeast i.e. *Saccharomyces cerevisiae* and left for eight days for fermentation. Temperature was maintained at 30° C for the growth of the yeast. Chemical analysis to quantify the protein content in the fermented media revealed an increase in the protein content as compared to the unfermented media. SCP is rich in certain essential amino acids which are limiting in most plant and animal sources. So, the protein thus produced is considered to be beneficial for the mankind for reducing illness and death. It is a method that can aid in reducing the burden of waste materials on the mother Earth.

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Introduction:-

Protein is a nutrient needed for normal growth and maintenance of human body ^[1]. Proteins (made up of amino acids) are important dietary constituents because they are required for the structure, function, and regulation of the body's major tissues and organs. Protein deficiency in food may pose a problem because essential amino acids, which can't be synthesized by the body itself, are not replenished ^[2]. Single cell protein can be a potential alternate to solve the global protein deficiency problem ^[3]. SCP or microbial proteins are edible unicellular microorganisms, used especially as a substitute for protein-rich foods. It is rich in certain essential amino acids like lysine and methionine which are limited in conventional sources of protein like animal proteins ^[4]. India grows the second largest quantity of fruits and vegetables ^[5]. Each fruit generates about 40% of its total mass as waste that includes peels, pulp and seeds. Banana (*Musa sapientum*) is a popular fruit consumed worldwide. Once the peel is removed,

the fruit can be eaten raw or cooked and the peel is generally discarded. It is an abundant and low cost agricultural waste residue and is easily available in large quantities^[6]. Banana peel constitutes about 18-20% proportion of fruit which is wasted^[7]. These peels are sometimes used as feedstock for cattle, goats, pigs, poultry etc. as they are rich in carbohydrates, and various vitamins and mineral elements^[8]. Various microorganisms that are used for the production of SCP are bacteria (*Cellulomonas*, *Alcaligenes*, etc.), algae (*Spirulina*, *Chlorella*, etc.), molds (*Trichoderma*, *Fusarium*, *Rhizopus*, etc.) and yeast (*Candida*, *Saccharomyces*, etc.). Yeast is a suitable candidate for single cell protein production because of its superior nutritional quality and efficiency to use low cost raw materials^[9]. Moreover, yeast cells are easy to harvest because of their large size (than bacteria), high malic acid and lysine content and their ability to grow at acidic pH.

Thus, in the present study, submerged fermentation has been carried out to produce single cell protein of *Saccharomyces cerevisiae* by using banana peels as a source of energy to convert into a high quality of protein. The banana peels were treated with acid to obtain monosaccharide as *Saccharomyces cerevisiae* lack the enzymes that hydrolyze the polysaccharide into simpler glucose units. In this study, the effect of addition of dextrose as carbon source when added to banana peel extracts is compared with banana peel extract without dextrose supplementation.

Material and Methods:-

Requirements: Lyophilized culture, inoculating loop, incubator shaker, laminar air flow, Erlenmeyer flask (500ml), measuring cylinder, distilled water, autoclave, ethanol (70%), cotton, refrigerator, centrifuge, colorimeter, etc.

Culture revival of *Saccharomyces cerevisiae*:-

Medium components: Pure culture of yeast (*Saccharomyces cerevisiae*) used was MTCC170. It was revived on Yeast extract potato dextrose (Yeast Extract 3.0g; Peptone 10.0g; Dextrose 20.0g; Agar 15.0g; distilled water 1.0 L) at 30⁰ C for 48 hrs and was stored at 4⁰C^[10].

Procedure:-

- Pure culture was obtained in lyophilized form from MTCC (microbial type culture collection), IMTECH.
- Culture was revived in Yeast extract potato dextrose broth medium.
- Culture was spread on the Yeast extract potato dextrose agar (YEPDA) medium plates using spreader, under sterile conditions and incubated at 30⁰C for 24hrs. Optical density of the culture was checked in colorimeter at 680nm.
- Sub culturing was done to maintain culture for further use.

Collection of Banana peels:-

The banana peels were collected from the local market in Chandigarh, India and washed several times with sterile water.

Sample preparation and Pre-treatment:-

Medium components- Banana peels (60gm), hydrochloric acid 10%(w/v), Erlenmeyer flask (500ml), measuring cylinder, distilled water, water bath at 100⁰C, Whatman filter paper no. 1, autoclave, dextrose (4gm/100ml).

Procedure:-

- The Banana peels were degraded to convert cellulose content into more available sugars by chemical treatments with little modification to the procedure of Lenihan 2010^[11].
- 100 ml of 10% (w/v) Hydrochloric acid was added to 60gm of banana peels in conical flasks.
- The mixture was placed in water bath at 100⁰C for one hour.
- After being allowed to cool, Banana peel extract was filtered through Whatman filter paper no: 1. The filtrates (98ml) were used as substrates and dextrose was added (4gm/100ml) in one group i.e. banana peel dextrose (abbreviated as BPD) and one was used without dextrose supplementation i.e. Banana peel extract (abbreviated as BPE).
- The two media were autoclaved at 121⁰C for 15 min.

The sterile broths thus prepared were used as carbon and nitrogen sources for biomass production.

Fermentation and harvesting of single cell protein:-

- Submerged fermentation was carried out in different Erlenmeyer flasks containing 98ml of the media.
- Media were aseptically inoculated with 2ml of inoculum of *Saccharomyces cerevisiae* (2×10^7 cfu/ml).
- Static fermentation was carried out at 28°C in a laboratory incubator for 8 days.

Drying of the final sample after fermentation:-**Procedure:-**

- After 8 days of fermentation, media (BPE and BPD) were poured into different Eppendorf centrifuge tubes.
- The Eppendorf tubes containing media were centrifuged at 10,000rpm for 15min.
- Supernatant was removed and pellet along with Eppendorf tube was weighed (wet biomass).
- Chemical analysis of the wet pellet was done to find the protein content.
- Hot air oven was set at 60°C . The Eppendorf tubes containing pellet were kept in pre-heated oven (60°C) for 24hrs.
- After drying, the dry weight of the sample was weighed.

Results:-**Biomass content of *Saccharomyces cerevisiae*:-****Table 1:-** Final percentage increase in the Biomass content after fermentation of 8 days:

Growth media	Biomass content increase (%age)
Banana Peel Extract	72.4%
Banana Peel Ext+Dextrose	84.6%

In submerged process ^[12], the banana peel substrate used for fermentation contains the nutrients to support the growth of microorganisms. The results obtained in the study showed that there was difference in dried biomass and the protein content when comparing the BPE and BPD media after fermentation.

The results showed that there was an increase in the biomass content in the banana peel extract with and without dextrose supplementation after fermentation. The utilization of sugar from the extract corresponded to the biomass production. There was a constant increase in total biomass content in banana peel extract supplemented with dextrose and banana peels without dextrose supplementation, during fermentation. The biomass content of BPD was found to be more than BPE. Although, it was also observed that the difference between the percentage biomass content of the two media was not too big. This demonstrated that BPE and BPD both have the nutrients that could be needed by an SCP microorganism for its growth.

Protein estimation:-

Protein was estimated in the sample by the method of Lowry et al (1951).

Table 2:- Protein content in banana peel extract at concentration of 60% after fermentation.

Growth media	1 st day (mg/g)	8 th day (mg/g)
Banana Peel Extract	2.5	11.7
Banana Peel Extract + dextrose	3.1	12.9

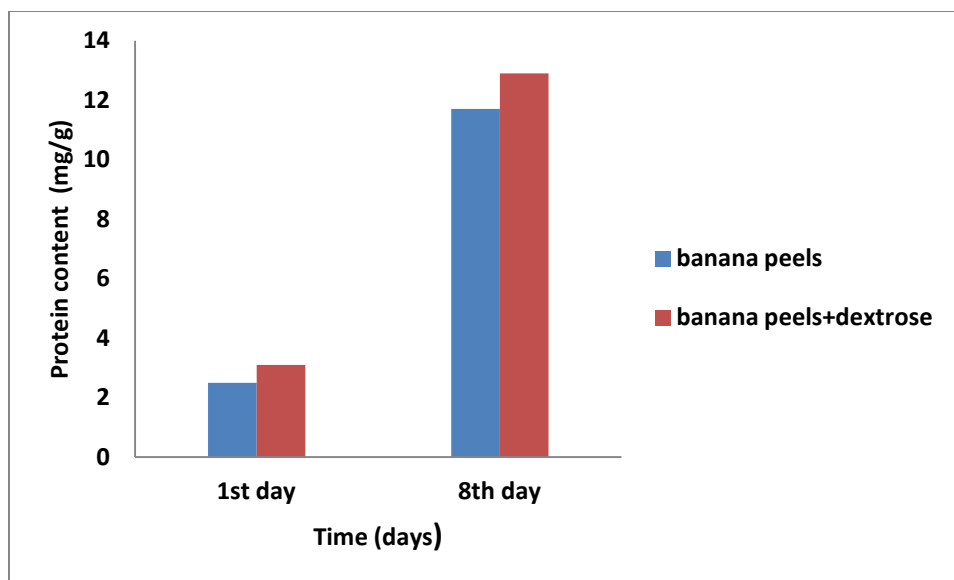


Figure 1:- Concentration of protein in banana peel extracts after fermentation.

SCP from yeast provides superior and better nutritional value in diets than other sources. This may be due to its acceptability, palatability and digestibility compared to the other SCPs^[13]. Crude protein content of yeast cells is one of the major factors to study in different growth media.

During the 8 days of fermentation, the protein content has significantly increased in the media. When examined the two different media on first day of fermentation, a low content was found in both the media. However, the protein content in BPD (3.1mg/g) was little higher than that of BPE (2.5mg/g) on the first day of fermentation as shown in Figure-1. While on the final day of fermentation the resultant protein content of BPE (11.7mg/g) and BPD (12.9mg/g), when tested, uncovered that supplementation with dextrose resulted in the enrichment of protein content in BPD as compared to BPE. Nevertheless, the difference between the media was not very large. This showed that supplementation of dextrose supported the growth of *Saccharomyces cerevisiae* in the banana peel substrate and made source of carbon and energy readily available for the yeast during the fermentation process.

In the present study, the experimental results clearly showed that high residual sugar content in the banana peels could be a suitable substrate for multiplication of yeast in the form of single cell protein leading to considerable increase in protein content of the bio processed product^[10].

Discussion:-

The results obtained from this study revealed that fermentation can bring about desirable changes in the nutrient composition of banana peel extracts. Yeast showed potential to increase the protein content of the banana peel extracts. The peels fermented with *Saccharomyces cerevisiae* had an increase in the protein content to 12.9mg/g from 3.1mg/g (for BPD) and 11.9mg/g from 2.5mg/g (for BPE). This implied that yeast had significant ($P < 0.05$) effect on protein content. Both the media i.e. BPE and BPD had a considerable surge in the biomass and protein contents. Since the difference between biomass and protein contents of the two media was not colossal and both the media produced relatively good results, banana peel extract can be used for the SCP production, with or without dextrose supplementation. This observation, further confirmed that banana peels had enough nutrients for the growth of *Saccharomyces cerevisiae*.

Conclusion:-

The data obtained in this study indicated that banana peel provided necessary nutrients for the *Saccharomyces cerevisiae* to grow and synthesize proteins. In conclusion, a promising yield of SCP production by *Saccharomyces cerevisiae* was achievable by fermentation using banana fruit peels. The present findings suggested that banana peels can be used as a potential substrate for products with relatively high protein content as produced by submerged state bio processing of banana waste by *Saccharomyces cerevisiae* that successfully enriched its protein content by utilizing various ingredients available in banana peels and there is a scope of converting waste fruit peels to

proteinaceous feed and food. Thus, banana fruit peels should be exploited properly as a substrate for the production of cellular biomass of edible or food grade yeast instead of dumping them, so that they can be used as animal feed supplement and for human consumption with minimal expense of money.

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RESEARCH ARTICLE

Homogeneous Finsler Square Metrics of Douglas Type.

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Abstract:

In this paper, we study homogenous Finsler square metric $F = \frac{(\alpha+\beta)^2}{\alpha}$ of Douglas type, and we investigate the necessary and sufficient conditions for the homogenous Finsler square metric to be Douglas metric, then it has following properties:

- (1) it is a Berwald metric or Randers type, and
- (2) it is a Riemannian metric.

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1. Introduction

A Finsler metric F on a manifold M is a homogeneous continuous function $F : TM \rightarrow [0; +\infty)$ where F is smooth on the slit tangent bundle TM_o satisfying nonnegativity ($F(y) > 0$ for any $y \neq 0$) and strong convexity (the fundamental tensor $g_{ij} := [\frac{1}{2}F^2]_{y^i y^j}$ is positive definite on TM_o). Here $(x^i; y^i)$ denote the natural system of coordinates of TM .¹

The notion of (α, β) -metric in Finsler spaces was introduced by M. Matsumoto [4] as a generalization of Randers metric $L = \alpha + \beta$, where α is a regular Riemannian metric $\alpha = a_{ij}(x)y^i y^j$, i.e., $\det(a^{ij}) \neq 0$ and β is a one-form $\beta = b_i(x)y^i$ and studied by many authors ([5], [6], [8], and [9]). A Finsler metric $L(\alpha, \beta)$ on a differentiable manifold M^n is called an (α, β) -metric, if L is a positively homogeneous function of degree one in α and β . There are several important (α, β) -metrics, namely Randers metric $L = \alpha + \beta$ Kropina metric $L = \frac{\alpha^2}{\beta}$, Matsumoto metric $L = \frac{\alpha^2}{(\alpha-\beta)}$, generalized Kropina metric $L = \frac{\alpha^{n+1}}{\alpha^n}$ and Z. Shen's square metric $L = \frac{(\alpha+\beta)^2}{\alpha}$.

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In 1929, L. Berwald constructed the following famous Finsler metric[10]

$$F = \frac{(\sqrt{(1-|x|^2)}|y|^2 + \langle x, y \rangle^2 + \langle x, y \rangle)^2}{(1-|x|^2)^2 \sqrt{(1-|x|^2)}|y|^2 + \langle x, y \rangle^2}.$$

This metric, defined on the unit ball $B^n(1)$ with all the straight line segments as its geodesics, has constant flag curvature $K = 0$. In a modern point of view, Berwald's metric belongs to a special kind of Finsler metrics called Berwald type or square metrics given as the form

$$F = \frac{(\alpha + \beta)^2}{\alpha}, \quad (1.1)$$

where α is a Riemannian metric and β is a 1-form[1]. It is known that above equation is a regular Finsler metric if and only if the length of β with respect to α , denoted by b , satisfies $b < 1$.

(α, β) -metrics form an important class of Finsler metrics that can be expressed in the form

$$F = \alpha \phi\left(\frac{\beta}{\alpha}\right),$$

where $\alpha = \sqrt{a_{ij}(x)y^i y^j}$ is a Riemann metric and $\beta = b_i(x)y^i$ is a 1-form with $\|\beta\|_\alpha < b_0$ on a manifold. It is well known that $F = \alpha \phi(\frac{\beta}{\alpha})$, is a positive definite Finsler metric if and only if $\phi = \phi(s)$ is a positive C^∞ function on $(-b_0, b_0)$ satisfying the following condition[3]

$$\phi(s) - s\phi'(s) + (b^2 - s^2)\phi''(s) > 0, \quad |s| \leq b < b_0. \quad (1.2)$$

In 1927, J. Douglas introduced the Douglas curvature for Finsler metrics[11]. Douglas curvature is an important projectively invariant in Finsler geometry. It is also a non-Riemannian quantity, since all the Riemannian metrics have vanishing Douglas curvature inherently. Finsler metrics with vanishing Douglas curvature are called Douglas metrics. Roughly speaking, a Douglas metric is a Finsler metric which is locally projectively equivalent to a Riemannian metric[12].

Douglas metrics form a rich class of Finsler metrics including locally projectively at Finsler metrics and Berwald metrics, the later are those metrics whose Berwald curvature vanishes[7].

In this present article, we study homogenous Finsler square metric $F = \frac{(\alpha + \beta)^2}{\alpha}$ of Douglas type, and we investigate the necessary and sufficient conditions for the homogenous Finsler square metric to be Douglas metric, then it has following properties:

- (1) it is a Berwald metric or Randers type, and
- (2) it is a Riemannian metric.

2. Preliminaries

Definition 2.1. A locally projectively flat (α, β) -metric $F = \alpha \phi(\frac{\beta}{\alpha})$ is said to be trivial, if α is locally projectively flat and β is parallel with respect to α .

A result of Z. Shen and G. Civi Yildirim [8] says that a Berwald's metric $F = \frac{(\alpha + \beta)^2}{\alpha}$ is locally projectively flat if and only if the spray coefficients of α are given in an adapted coordinate system by

$$G_\alpha^i = \xi y^i - 2\tau \alpha^2 b^i, \quad (2.1)$$

for some 1-form $\xi = \xi_i(x)y^i$ and some scalar function $\tau = \tau(x)$, and at the same time the covariant derivative of β is given by

$$b_{i|j} = 2\tau\{(1 + 2b^2)a_{ij} - 3b_i b_j\}. \quad (2.2)$$

Later on, B. Li, Z. Shen and Y. Shen, found a sufficient and necessary condition for (α, β) -metrics to be locally projectively flat in dimension $n \geq 3$ [12]. It says that for a projectively flat (α, β) -metric $F = \alpha \phi(\frac{\beta}{\alpha})$ on an open subset $U \subseteq R^n$ with $n \geq 3$, if we add

Theorem 2.1. Let $s = \frac{\beta}{\alpha}$ and let $F = \alpha\phi(\frac{\beta}{\alpha})$ be an (α, β) -metric on an open subset $U \subseteq R^n (n \geq 3)$, where $\alpha = \sqrt{a_{ij}y^i y^j}$ and $\beta = b_i(x)y^i \neq 0$. Let $b := \|\beta\|_\alpha$. Suppose that the following conditions hold:

- F is not of Randers type, i.e., $F \neq \sqrt{c_1\alpha^2 + c_2\beta^2} + c_3\beta$ for any constants c_1, c_2 and c_3 ,
- β is not parallel with respect to α ,
- $db \equiv 0$ or $db \neq 0$ everywhere or b is constant on U .

Then F is a Douglas metric if and only if $\phi(s)$ satisfies the following ODE,

$$\{1 + (k_1 + k_3)s^2 + k_2s^4\}\phi''(s) = (k_1 + k_2s^2)\{\phi(s) - s\phi'(s)\}, \quad (2.3)$$

where k_1, k_2, k_3 are constants with $k_2 \neq k_1k_3$ and the covariant derivative $\nabla\beta = b_{i|j}y^i dx^j$ of β with respect to α satisfies equation (2.2).

We can see that the function $\phi(s) = (1 + s)^2$ satisfies equation (2.3) with $(k_1, k_2, k_3) = (2, 0, -3)$ or $(k_1, k_2, k_3) = (-3, 0, 2)$.

We used the following results which proved by G. Yang in [18].

Theorem 2.2. Let $F = \alpha\phi(s), s = \beta/\alpha$ be a regular (α, β) -metric on an open subset $U \subset R^2$, where $\phi(0) = 1$. Suppose that β is not parallel with respect to α and F is not of Randers type. Let F be a Douglas metric. Then one has one of the following two cases.

- $\phi(s)$ satisfies (2.3).
- F can be written as

$$F = \tilde{\alpha} \pm \frac{\tilde{\beta}^2}{\tilde{\alpha}}, \quad (\tilde{\alpha} := \sqrt{\alpha^2 - k\beta^2}, \quad \tilde{\beta} := c\beta), \quad (2.4)$$

where k, c are constants with $c \neq 0$

Corollary 2.1. Let $F = \alpha \pm \beta^2/\alpha$ be a two-dimensional (α, β) - metric. Then F is a Douglas metric if and only if β satisfies

$$r_{ij} = 2\tau\{(1 \pm b^2)a_{ij} \mp 3b_ib_j\} + \frac{3}{\pm 1 - b^2}(b_is_j + b_js_i), \quad (2.5)$$

where $\tau = \tau(x)$ is a scalar function. Note that

- $F = \alpha + \frac{\beta^2}{\alpha}$ is positive if and only if $b^2 < 1$,
- $F = \alpha - \frac{\beta^2}{\alpha}$ is positive if and only if $b^2 < \frac{1}{2}$.

Every Finsler metric F on a manifold M induced a spray $G = y^i \frac{\partial}{\partial x^i} - 2G^i \frac{\partial}{\partial y^i}$ which determines the geodesics. By definition, a Finsler metric F is a Berwald metric if the spray coefficients $G^i = G^i(x, y)$ are quadratic in $y \in T_x M$ at every point x , i.e., $G^i = \frac{1}{2}\Gamma_{jk}^i(x)y^j y^k$. Riemannian metrics are special Berwald metrics. In fact, Berwald metrics are almost Riemannian in the sense that every Berwald metric is affinely equivalent to a Riemannian metric, i.e., the geodesics of any Berwald metric are the geodesics of some Riemannian metric [2]. The Douglas metrics are more generalized ones than Berwald metrics. A Finsler metric is called a Douglas metric if the spray coefficients $G^i = G^i(x, y)$ are in the following form:

$$G^i = \frac{1}{2}\Gamma_{jk}^i(x)y^j y^k + P(x, y)y^i. \quad (2.6)$$

Douglas metrics form a rich class of Finsler metrics including locally projectively flat Finsler metrics. The study on Douglas metrics will enhance our understanding on the geometric meaning of non-Riemannian quantities.

Definition 2.2. Two (α, β) -metrics $F_1 = \alpha_1 \phi_1(\frac{\beta_1}{\alpha_1})$ and $4F_2 = \alpha_2 \phi_2(\frac{\beta_2}{\alpha_2})$ are said to be of same type if there is an element $\Pi \in G$ such that $\Pi(\phi_1) = \phi_2$. In this case, the functions $\phi_1(s)$ and $\phi_2(s)$ are said to be equivalent. G is called the representation group of (α, β) -metrics.

For example, all the functions equivalent to $(1 + s)$ will provide Randers type metrics. Conversely, if $F = \alpha \phi(\frac{\beta}{\alpha})$ is of Randers type, then $\phi(s)$ must be equivalent to $(1 + s)$. Actually, the functions for Randers type metrics, which are given by $\phi(s) = \sqrt{1 + us^2} + vs$, can be expressed as $\phi(s) = g_u \circ h_v(1 + s)$. Notice that all the functions are always asked to satisfy $\phi(0) = 1$. Suppose that a given locally projectively flat (α, β) -metric $F = \alpha \phi(\frac{\beta}{\alpha})$ is neither locally Minkowskian nor of Randers type, then $\phi(s)$ must be a solution of (2.3) according to Z. Shen's result. Due to the non-uniqueness, if we rewrite the metric as $F = \tilde{\alpha} \psi(\frac{\beta}{\tilde{\alpha}})$, then the new function $\psi(s)$, which is equivalent to $\phi(s)$, must be also a solution of (2.3) with some different parameters.

Theorem 2.3. A Finsler metric F on a manifold M ($\dim M \geq 3$) is locally projectively flat if and only if F is a Douglas metric with scalar flag curvature.

In [15] shows an (α, β) -metric $F = \alpha \phi(\frac{\beta}{\alpha})$ is a Berwald metric if and only if β is parallel with respect to α , i.e., $b_{i|j} = 0$, regardless of the choice of a particular ϕ , and [12] the authors obtained a characterization of (α, β) -metrics of Douglas type.

3. Homogeneous Finsler Square Metrics Of Douglas Type

Recall that the group $I(M; F)$ of isometries of a Finsler manifold $(M; F)$ is a Lie transformation group of M [14]. If $I(M; F)$ acts transitively on M , then $(M; F)$ is called homogeneous. Thus, the homogeneous Finsler manifold M can be written as the form $M = G/H$, where G is a Lie group acting isometrically and transitively on M , and H is the isotropy subgroup at a point in M . Moreover, if the Lie algebra of G , \mathfrak{g} , has a decomposition

$$\mathfrak{g} = \eta + m \quad (\text{direct sum of subspaces}),$$

where η is the Lie algebra of H and m is a subspace of \mathfrak{g} satisfying

$$\text{Ad}(h)(m) \subset m \quad \text{for all } h \in H.$$

Then the homogeneous Finsler manifold $(G/H; F)$ is called reductive. In this case, the tangent space $T_o(G/H)$, where $o = eH$ is the origin, can be canonically identified with m . Note that the isotropy subgroup $I_x(M, F)$ of $I(M; F)$ at a point $x \in M$ is compact [14], and M can be written as

$$M = I(M; F)/I_x(M, F).$$

Then $M = I(M; F)/I_x(M, F)$ is a reductive homogeneous manifold.

Let $(G/H; F)$ be a reductive homogeneous (α, β) -space of the form $F = \alpha \phi(s)$ where $s = \frac{\beta}{\alpha}$ with a Riemannian metric α and a 1-form β on G/H . In this section, we assume that α and β are both G -invariant. Consider the underlying homogeneous Riemannian manifold $(G/H; \alpha)$. Let $\langle \cdot, \cdot \rangle$ denote the corresponding inner product on m . According to [16], the form β corresponds to a vector u in the subspace

$$V = \{u \in m | \text{Ad}(h)u = u \quad \text{for all } h \in H\}.$$

We also assume that $\beta \neq 0$, or equivalently, $u \neq 0$.

We use the local coordinate system developed in [17], which is given as follow.

Let u_1, u_2, \dots, u_n be an orthonormal basis of m with respect to the inner product $\langle \cdot, \cdot \rangle$ and $u_n = \frac{u}{|u|}$. Then there exists a neighborhood U of $o = H$ in G/H such that the map

$$(\exp(x^1 u_1) \exp(x^2 u_2) \dots \exp(x^n u_n)) \longmapsto (x^1, x^2, \dots, x^n),$$

defines a local coordinate system on U .

Since α and β are both G -invariant, $b := \|\beta\|_\alpha = |u|$ is a constant. By [17], at the origin $o = H$, we have

$$\begin{aligned} a_{ij} &= \delta_{ij}, & b_i &= b\delta_{ni}, \\ b_{i|j} &= \frac{b}{2}(\langle [u_i, u_j], u_n \rangle - \langle [u_n, u_i], u_j \rangle - \langle [u_n, u_j], u_i \rangle), \\ s_{ij} &= \frac{b}{2}\langle [u_i, u_j], u_n \rangle, & r_{ij} &= -\frac{b}{2}(\langle [u_n, u_i], u_j \rangle + \langle [u_n, u_j], u_i \rangle). \end{aligned} \quad (3.1)$$

Note that $s_n = b^i s_{in} = b s_{nn} = 0$.

Theorem 3.4. *Let $F = \frac{(\alpha+\beta)^2}{\alpha}$ be a homogeneous Finsler square metric on G/H . Then F is a Douglas metric if and only if F is a Berwald metric or F is a Douglas metric of Randers type.*

Proof. Suppose that $F = \alpha\phi(\frac{\beta}{\alpha})$ is a homogeneous (α, β) -metric $F = \frac{(\alpha+\beta)^2}{\alpha}$ on G/H , where the Riemannian metric α and the 1-form β are both G -invariant. We only need to compute at the origin $o = H$. By (3.1), it is obvious that $b_{n|n} = 0$ at the origin. We now prove the theorem in the following two cases.

Case 1: $\dim(G/H) \geq 3$. Suppose that F is a Douglas metric, and F is neither a Berwald metric nor of Randers type. Since b is a constant, it follows from Theorem 2.3 and (2.2) that

$$b_{n|n} = 2\tau\{1 - b^2\} = 0. \quad (3.2)$$

Since α and β are both G -invariant, the scalar function $\tau = \tau(0)$ is a constant.

By the assumption that F is not a Berwald metric, it follows that $\tau \neq 0$. So we have

$$(1 - b^2) = 0. \quad (3.3)$$

By (2.3), we have

$$\phi''(s) = \{\phi(s) - s\phi'(s)\} \frac{k_1 + k_2 s^2}{1 + (k_1 + k_2 s^2)s^2 + k_3 s^2}. \quad (3.4)$$

Plugging (3.4) into (1.2), we get

$$\begin{aligned} \phi(s) - s\phi'(s) + (b^2 - s^2)\phi''(s) &= \{\phi(s) - s\phi'(s)\} \left\{ 1 + \frac{b^2 - s^2 + (k_1 + k_2 s^2)}{1 + (k_1 + k_3)s^2 + k_2 s^4} \right\} \\ &= \{\phi(s) - s\phi'(s)\} \left\{ 1 + \frac{1 + k_1 b^2 + (k_2 b^2 + k_3)s^2}{1 + (k_1 + k_3)s^2 + k_2 s^4} \right\}. \end{aligned} \quad (3.5)$$

By taking $b = s$ in (1.2) we can see that $\phi(s) - b\phi'(s) > 0$, is always positive as long as F is a Finsler metric. So the (1.2) implies the following

$$\frac{1 + k_1 b^2 + s^2(k_3 + k_2 b^2)}{1 + (k_1 + k_3)s^2 + k_2 s^4} > 0, \forall |s| \leq b < b_0. \quad (3.6)$$

Taking $s = 0$ we have $1 + k_1 b^2 > 0$ than $1 + k_1 s^2 \geq \min\{1, 1 + k_1 b^2\}$.

On other hand, we have $(k_1, k_2, k_3) = (2, 0, -3)$ and by taking $s = b$ in (2.3), then

$$\begin{aligned} \phi''(s) &= \{\phi(s) - s\phi'(s)\} \frac{2 + 0s^2}{1 + (2 + 0s^2)s^2 - 3s^2}, \\ &= \{\phi(s) - s\phi'(s)\} \frac{2}{1 - s^2}. \end{aligned} \quad (3.7)$$

And when $(k_1, k_2, k_3) = (-3, 0, 2)$ we have

$$\begin{aligned}\phi''(s) &= \{\phi(s) - s\phi'(s)\} \frac{-3 + 0s^2}{1 + (-3 + 0s^2)s^2 + s^2}, \\ &= \{\phi(s) - s\phi'(s)\} \frac{-3}{1 + s^2}.\end{aligned}\quad (3.8)$$

It is clear that the solution of (3.7) and (3.8) is given by

$$\phi(s) = \frac{c}{1 \mp s^2}, \quad (3.9)$$

for some constant c . This implies that is of Randers type, which is a contradiction. This completes our proof in this case.

Case 2: $\dim(G/H) = 2$. By Theorems 2.1 and 2.2, we only need to prove the theorem under the assumption that F is given by $F = \alpha \pm \frac{\beta^2}{\alpha}$, where the Riemannian metric α and the 1-form β are both G -invariant. We will also use the above local coordinate system and setting $n = 2$. Note that $b_2 = b$ and $s_2 = 0$. By Theorem 2.2,

- if $F = \alpha + \frac{\beta^2}{\alpha}$ is a Douglas metric, then at the origin $o = H$, we have

$$r_{22} = b_{22} = 2\tau(o)(1 - b^2).$$

Since $b^2 < 1$ when $F = \alpha + \frac{\beta^2}{\alpha}$ is positive definite, we conclude that $\tau(o) = 0$.

- If $F = \alpha - \frac{\beta^2}{\alpha}$ is a Douglas metric, then at the origin $o = H$, we have

$$r_{22} = b_{2|2} = 2\tau(o)(1 + b^2) = 0.$$

Thus we also have $\tau(o) = 0$. Since α and β are both G -invariant, the scalar function $\tau(x)$ is a constant. Therefore $\tau = \tau(o) = 0$, which implies that F is a Berwald metric.

This completes the proof of Theorem under the assumption that both α, β are G -invariant.

Let u be a vector corresponding to β in the subspace V given in the above. Then the condition for F to be a Berwald metric is equivalent to the following:

$$\langle [v, w]_m, u \rangle = 0 \quad \text{for all } v, w \in m, \quad (3.10)$$

$$\langle [u, v_1]_m, v_2 \rangle + \langle [u, v_2]_m, v_1 \rangle = 0 \quad \text{for all } v_1, v_2 \in m. \quad (3.11)$$

By Theorem 3.4 and a direct observation, we have

Theorem 3.5. Let $F = \frac{(\alpha + \beta)^2}{\alpha}$ be a homogeneous Finsler square metric on G/H , where the Riemannian metric α and the 1-form β are both G -invariant. Suppose the Lie algebra \mathfrak{g} of G is perfect, i.e., $\mathfrak{g} = [\mathfrak{g}, \mathfrak{g}]$, then F is a Douglas metric if and only if F is a Riemannian metric.

Proof. By Theorem 3.4, if $F = \alpha\phi(\frac{\beta}{\alpha})$ is a Douglas metric, then it is a Berwald metric or a Douglas metric of Randers type. Let $\mathfrak{g} = \eta + \mathfrak{m}$ denote a reductive decomposition of \mathfrak{g} . If F is a Douglas metric of Randers type, then F can be expressed in the form $F = \tilde{\alpha} + \tilde{\beta}$, where α and β are both G -invariant, and β is a closed 1-form. In this case, there exists a vector $u \in \mathfrak{m}$ satisfying (3.10), where the inner product $\langle \cdot, \cdot \rangle$ is corresponding to $\tilde{\alpha}$, and the vector u is corresponding to $\tilde{\alpha}$ with respect to the inner product. Now the condition that $\text{Ad}(h)(u) = u$ for all $h \in H$ is equivalent to

$$[v, u] = 0 \quad \text{for all } v \in \eta. \quad (3.12)$$

Since the inner product $\langle \cdot, \cdot \rangle$ is G -invariant, we have

$$\langle [v, w_1]_m, w_2 \rangle + \langle [v, w_2]_m, w_1 \rangle = 0 \quad \text{for all } v \in \eta, w_1, w_2 \in \mathfrak{m}. \quad (3.13)$$

Combining (3.12) and (3.13), we obtain

$$\langle [v, w]_m, u \rangle = 0 \quad \text{for all } v \in \eta, w \in \mathfrak{m}. \quad (3.14)$$

Now the assumption $g = [g, g]$ implies that there exists two vectors $w, v \in g$ such that $[w, v] = u$. Let $w = w_1 + w_2$ and $v = v_1 + v_2$, where $w_1, v_1 \in m$ and $w_2, v_2 \in \eta$. Then we have

$$[w, v] = [w_1, v_1] + [w_1, v_2] + [w_2, v_1] + [w_2, v_2]. \quad (3.15)$$

Therefore by (3.10) and (3.14), we have

$$\begin{aligned} \langle u, u \rangle &= \langle u, [w, v]_m \rangle \\ &= \langle u, [w_1, v_1]_m \rangle + \langle u, [w_1, v_2]_m \rangle + \langle u, [w_2, v_1]_m \rangle \\ &= 0. \end{aligned}$$

Thus $u = 0$, which implies that F is a Riemannian metric. If $F = \alpha\phi(\frac{\beta}{\alpha})$ is a Berwald metric with α and β both G -invariant, then there exists a vector $u \in m$ satisfying (3.10). Then a similar argument shows that F is also a Riemannian metric.

4. Conclusion

The important example of Finsler space are different type of (α, β) -metric are Randers metric, Kropina metric and other special (α, β) -metric. In [13] Ramesha M and S. K. Narasimhamurthy are devoted the necessary and sufficient conditions for a Finsler space with a special (α, β) -Metric $F = C_1\alpha + C_2\beta + \frac{\beta^2}{\alpha} : C_2 \neq 0$, to be a Douglas space and also to be Berwald space. H. Liu and S. Deng in [19] have shown the necessary and sufficient conditions for Randers homogeneous of Douglas type to be Berwald and Riemannian metric.

In this present article we consider homogenous Finsler square metric $F = \frac{(\alpha+\beta)^2}{\alpha}$ of Douglas type, and we investigate the necessary and sufficient conditions for the homogenous Finsler square metric to be Douglas metric, then it has following properties:

- (1) it is a Berwald metric or Randers type, and
- (2) it is a Riemannian metric.

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REVIEW ARTICLE

SIMVASTATIN AS A SOLE OSTEOPROMOTIVE MATERIAL IN CRITICALLY SIZED BONE DEFECTS: SYSTEMATIC REVIEW.

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Simvastatin, Osteoinductive, critical sized Bone defect.

Abstract

Objective: The aim of this study is to evaluate the effect of simvastatin as a sole osteoinductive material on bone formation by eliminating the role that may be attributed to the various bone grafts that have been used with simvastatin as drug carriers. **Material and methods:** The search terms (Simvastatin) AND (Bone Defect) were used to search the PUBMED electronic database. In addition to this online search, a hand search of the relevant major international journals was also performed. **Results:** The search results showed total of 46 articles, only 3 articles were found to fulfill the inclusion criteria. Two out of these 3 articles used the same simvastatin concentration (0.2ml simvastatin solution on either collagen or gelatin sponge) while in the third article, collagen graft of swine origin was used with two different simvastatin dose (2.2 mg and 0.5 mg). Simvastatin seems to have osteoinductive potentials when applied locally in bone defects when used in proper dose and with proper carrier. **Conclusion:** The safety and osteoinductive properties of simvastatin are directly affected by the used dose and carrier, thus further studies are needed in order to determine the ideal dose/carrier complex that maximize the osteoinductive capabilities and eliminate or minimize the associated inflammatory reaction.

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Introduction:-

Although considered as the gold standard for bone regeneration, yet, autogenous bone is far away from being the (ideal) graft material because of its disadvantages such as donor site morbidity, limited availability [1,2] and unpredictable resorption.[3]

This has led to the use of other graft material such as allografts, xenografts and alloplastic materials in order to avoid these drawbacks especially in elective surgeries in which limiting patient discomfort should be considered as a main priority. [4] However, the use of these bone substitutes is also associated with some disadvantages such as the lack of osteoinduction, antigenicity & risk of disease transmission. [5, 6]

In the past years, researchers investigated the visibility of combining the osteoconductive properties of alloplasts with the osteoinductive properties of the recombinant bone morphogenic proteins (BMP). [7, 8] Which in spite of

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showing promising results, yet it did not have its way to the common daily clinical practice due to the difficulties facing its application such as the high cost and the inability to deliver the accurate dose of BMP.[9,10]

These aforementioned drawbacks of the current reconstructive options led to the rise of a new concept **THE IN SITU TISSUE REGENERATION APPROACH** [11]"this is authors quoted text" which involves the use of an external stimuli that up-regulate the patient own cells. This can be achieved via the addition of certain pharmacological compounds to an osteoconductive bone substitute.[12]

The bone anabolic action of statins was first reported by Mundy et al in 1999 after he screened more than 30,000 pharmacological compounds. He found statins to be the only compounds that have the ability to up-regulate BMP-2 and stimulate osteoprogenitor cells proliferation .[13]In addition to their osteogenic properties , statins were found to have anti-inflammatory , anti-oxidant and angiogenic effects.[14,15]

However, on addressing the anti-inflammatory effect of simvastatin, it should be kept in mind that, simvastatin in high doses can actually induce inflammation around the site of local application, [16] that is to say, while a low dose can be inefficient in bone formation, high doses may stimulate an excessive inflammatory response [17] which is well recognized as a major contributing factor to delay healing in both animal and human models. [18, 19]

The effect of locally applied simvastatin on bone formation received the main attention of researchers in the last few years as it was used in combination with various carriers such as collagen sponge,[17,20] Alpha tricalcium phosphate (α -TCP),[12,21]Beta tricalcium phosphate(β -TCP),[21]hydroxyapatite (HA),[21] gelatin sponge[22]and bioglass.[23] The results of these studies showed that the local effect of Simvastatin on bone depends on 2 main factors which are the used carrier and the simvastatin dose.[11]

The aim of this study is to evaluate the effect of simvastatin as a sole osteoinductive material on bone formation by eliminating the role that may be attributed to the various bone grafts that have been used with simvastatin as drug carriers.

Materials and methods:-

Search strategy:-

The search terms (Simvastatin) AND (Bone Defect) were used to search the PUBMED electronic database. In addition to this online search, a hand search of the major international journals that covers the field of dental implants was performed to detect any overlooked articles (Clinical Implant Dentistry and Related Research, Clinical Oral Implants Research, Implant Dentistry, International Journal of Oral and Maxillofacial Surgery, Journal of Oral and Maxillofacial Surgery, Journal of oral implantology). Search was carried out on the 20th of November 2016

Inclusion criteria:-

1. Simvastatin applied in surgically created critical sized bone defects.
2. Simvastatin used as sole effective osteoinductive material.
3. Studies provide histomorphometric results.
4. In vivo studies only

Exclusion criteria:-

1. Studies in language other than English
2. Simvastatin applied in periodontal defects or on surface of intact bone.
3. Systemically administrated simvastatin.
4. Simvastatin used with specially designed carriers, bone substitutes or mixed with any other material than may induce new osteogenesis.
5. Presence of any systemic condition that may affect the osteogenesis process.

Study selection and data collection:-

The search results showed total of 46 articles which were subjected to a 3 staged filtering process in order to detect the articles that comply with the aforementioned inclusion criteria. In the first stage, 31 articles were excluded after screening their titles. In the second stage another 8 articles were excluded after screening their abstract. In the last stage the full text of the remaining 7 articles were screened which result in eliminating another 4 articles. In the end

of the filtering process, only 3 articles were found to fulfill the inclusion criteria {Table 1}. The screening process was accomplished by 2 reviewers. No additional articles were found through hand searching.

Results:-

The 3 selected papers were all animal studies which may reflect that simvastatin did not find its way to human application due to the diversity in the results which varies from success in some studies to failure in others.

Collagen sponges were used in 2 out of these 3 studies [17, 20] yet, they were of different origin and the simvastatin dose used in both studies was also different. In the third study gelatin sponge was used as a carrier for simvastatin. [22].

In the study performed by Wong et al in 2003, [20] the collagen graft used was of bovine origin. In this study 15 critical sized bone defects (10x5 mm) were created in the parietal bone of 9 New Zealand rabbits. The defects were divided into 3 equal groups, passive control group (received no graft), active control (grafted with 0.2 mg collagen graft) and experiential group (grafted with 0.2 mg collagen graft mixed with 0.2 ml simvastatin solution). All animals showed normal recovery without any complication. The histomorphometric results of the biopsies obtained after killing the animals in the 14th day showed that the amount of newly formed bone was 308 % more in the experiential group { mean $1.63 \text{ mm}^2 \pm 0.48 \text{ mm}^2$ } compared to the active group { $0.4 \text{ mm}^2 \pm 0.28 \text{ mm}^2$ }. The difference between the 2 groups was considered statistically significant.

The second study that used collagen sponge was performed by Calixto et al in 2011 [17] in which it was of swine origin due to the outbreak of bovine spongiform at this time. A bilateral critical sized bone defects were created in the parietal bones of 64 Wistar rats using a 5- mm diameter trephine. Animals were divided into 4 equal groups, Control group (received no treatment), carrier group (received 0.2 mg collagen sponge), Simvastatin -1 group (received collagen sponge mixed with 2.2 mg/50 μ l Simvastatin solution), Simvastatin-2 group (received collagen sponge mixed with 0.5mg/50 μ l Simvastatin solution).

During the recovery period, all animals of Simvastatin -1 group showed necrosis and crust formation which persisted until the 45th day after surgery. On the other hand, only 4 animals of the simvastatin -2 group showed such complication which lasts only for 30 days. The histomorphometric results of the biopsies obtained after killing the animals in the 60th day showed that the highest amount of newly formed bone was detected in the control group (mean $0.98 \text{ mm}^2 \pm 0.1 \text{ mm}^2$) followed by the simvastatin-1 group ($0.75 \text{ mm}^2 \pm 0.2 \text{ mm}^2$) while the least amount was in the simvastatin-2 group ($0.5 \text{ mm}^2 \pm 0.15 \text{ mm}^2$). The differences between these groups were considered statistically significant.

Unlike the previous studies, gelatin sponge was selected as a drug carrier by Özeç et al in 2007 [22] to perform his study in which he used a 3-mm round bur to create a 23 critically sized defects in the mandible of 23 Wistar rats. Animals were divided into 3 unequal groups, experimental group (include 9 animals that received 0.2mg gelatin sponge mixed with 0.2 ml simvastatin solution), active control group (include 8 animals that received gelatin sponge) and passive control group (include 6 animals that did not receive any graft).

All animals showed normal and complication free recovery. The histomorphometric results of the biopsies obtained after killing the animals in the 14th day showed that the amount of newly formed bone was 190 % more in the experiential group compared to the active group and 240 % more than the passive group { mean values were not available }. These results were both considered to be statistically significant.

Discussion:-

The direct effect of statins on bone was discovered for the first time by Mundy et al [13] in 1999. Their study, along with others, suggest that statins, administered either locally or systemically, act as potent stimulators of bone formation and regeneration [13,24,25] Simvastatin was found to up regulate BMP-2, [26] vascular endothelial growth factor (VEGF) gene expression and alkaline phosphatase expression in osteoblasts, [27] it was also found to inhibit osteoclastogenesis. [28]

On comparing the effect of locally applied versus systemically administered statin on stimulating bone healing, locally applied statin was found to be 50-80 times more potent than systemically administered statin. [29] The

minimal effect of systemically administered statin on bone is expected as statins are known for targeting liver to treat hyperlipidemia with only small amount that reach and accumulate in bone. [30]

The aim of this study was to highlight the anabolic potentials of Simvastatin on bone when used as a sole osteoinductive material; any study that used simvastatin with a carrier that may affect either osteogenesis or the rate of simvastatin release was excluded from this study.

As it was mentioned earlier, whether the locally applied statin will play a positive or negative effect on osteogenesis is mainly determined by the used carrier and the statin dose. The 3 papers selected in this research showed that 3 different carriers were used as well as 3 different doses of Simvastatin.

In two out of the three papers, the dose of simvastatin was the same (0.2 ml simvastatin solution); yet 2 different carriers were used. In the study conducted by Wong et al,[20] collagen graft of bovine origin was used while in the study conducted by Özeç et al,[22] gelatin sponge was used. The results of these two studies showed that collagen sponge may be preferred over gelatin sponge as a drug carrier for simvastatin. This was based on the fact that although in Wong's study the size of defect was larger than the size of defect in Özeç's study (10x15 mm vs. 3x3mm), yet when the experimental group in each study was compared to its relevant active control group, the amount of newly formed bone was higher in Wong's study than in Özeç's study (308 % vs. 190 %).

It is worth to be mentioned that in both studies, animals were killed at the 14th day after the surgical procedure which will eliminate the time factor in the process of bone formation. However, the limited number of the studies does not enable the author to reach a definite decision regarding the superiority of collagen sponge over gelatin sponge.

In the 3rd study conducted by Calixto et al, [17] two different doses of simvastatin were used (2.2 mg and 0.5 mg). The carrier used in this study was collagen sponge of swine origin. Unlike the previous studies, the passive control group showed the highest amount of bone formation when compared to the other 3 groups.

This was attributed to the higher inflammatory levels that were found in the carrier group compared to the control group. The inflammatory reaction was even higher in the simvastatin groups especially within the group that received higher simvastatin dose. The synergetic inflammatory reaction of both a high simvastatin dose and an antigenic carrier was found to have deleterious effect on bone formation.

Regarding the safety of using simvastatin, animals in studies conducted by Wong [20] and Özeç [22] showed normal and complication free recovery, whereas all animals in Calixto[17] study that received simvastatin showed necrosis and crust formation which was more evident in the animals that received 2.2mg simvastatin. In addition to this local complication 6 animals of the simvastatin groups died (4 animals from the 2.2mg group and 2 animals from the 0.5mg group). These results comply with the findings of Nyan [16] and Calixto[17] that statins may have a dual effect on osteogenesis based on the degree of inflammatory reaction which is affected by the used dose.

Conclusion:-

The safety and osteoinductive properties of simvastatin are directly affected by the used dose and carrier, thus further studies are needed in order to determine the ideal dose/carrier complex that maximize the osteoinductive capabilities and eliminate or minimize the associated inflammatory reaction.

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Disclosure statement:-

The authors report no conflict of interest. The Final version of this manuscript was revised and approved by all authors.

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RESEARCH ARTICLE

PREVALENCE OF REFRACTIVE ERRORS IN RURAL AND URBAN SCHOOL CHILDREN – A CROSS SECTIONAL STUDY.

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Abstract

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Introduction:-

The quality of life of any person depends on many aspects. One's ability to see things clearly can determine success or failure at work or in school. Normal vision helps a person to lead an independent life, without having to rely on others to fulfill their basic needs.

It has been estimated that about 180 million people worldwide are visually disabled. 45 million of them are blind. Developing countries account for four out of five of the visually disabled. One of the major causes of correctable blindness is refractive error. It has been estimated that refractive errors are prevalent in about 2.3 billion people. Out of these, 1.8 billion people have access for examination of the eyes and proper treatment. The remaining 500 million people have uncorrected refractive errors resulting in impaired vision and blindness.

Refractive errors are the commonest cause of impaired vision and they form the second important cause of treatable blindness in the world next only to cataract.

Refractive error is an intrinsic optical defect where parallel light rays are not focussed on the retina, resulting in a blurred image. Refractive errors develop during childhood and adolescence. If they are not treated children may develop amblyopia or strabismus.

Refractive errors may not only be associated with genetic factors but also with various other risk factors like prolonged near activity like reading, playing video games and watching television or computer for longer duration which considerably reduces the outdoor play time of children. This study has been undertaken to study the prevalence of refractive errors in urban and rural school children in Madurai. It is expected that this study will also give an insight into the magnitude of this problem and create awareness among the common public.

Aim and objectives:-

- (i) To find the prevalence of refractive errors in school children aged 10-14 years.
- (ii) To compare the prevalence of refractive errors in urban school children with that in rural school children.

Materials and methods:-

A cross sectional school based study was designed with 600 students within the age group of 10-14 years. A total of four schools, two from an urban area and two from a rural area were selected randomly.

300 students were selected from the schools located in the urban area and another 300 students were selected from the schools located in the rural area.

The principals, teachers and the concerned students were briefed about the study. Prior informed consent for participation in the study was obtained from the parents of these children.

A standardized questionnaire prepared in their local language was used to collect data on their age, sex, parental history of wearing glasses and other environmental factors like watching television, playing video games, outdoor activities and reading books.

Visual acuity (VA) test was performed using Snellen's E chart. Students with visual acuity $<6/6$ in one or both the eyes were tested for the presence or absence of refractive error by pinhole test. Those showing improvement with pinhole (indicating refractive error) were then subjected to retinoscopy and subjective refraction.

Emmetropia was defined as a spherical equivalent between -0.50 DS and $+0.50$ DS. A spherical equivalent less than -0.50 DS was taken as myopia. Hypermetropia was defined as a spherical equivalent greater than $+0.50$ DS. Astigmatism was defined as a cylindrical error less than -0.50 diopter cylinder (DC) in any axis.

Those children with both spherical and cylindrical errors were documented as having the spherical equivalent of myopia or hypermetropia respectively. The children with only cylindrical errors were documented as having astigmatism. Refractive error was categorized into three groups as i) myopia ii) hypermetropia and iii) astigmatism.

The particulars of each student were filled on a structured questionnaire specially designed for this purpose.

The data collected was analyzed by Pearson's Chi Square test using SPSS (Statistical Package for Social Sciences).

Observation:-

Mean age of the 600 children examined was 12.05 years. Out of these children, 297 were boys and 303 were girls.

The results of the analysis are presented in the following table.

TABLE1:- Prevalence of refractive errors in school children.

	Number of children (600)	Percentage
Refractive errors	97	16.2
No refractive errors	503	83.8

The over-all prevalence of refractive errors in school children was found to be 16.2%.

TABLE 2:- Distribution of various types of refractive errors in school children.

Type of refractive error	Number of children	Percentage
Myopia	67	69.1%
Hypermetropia	10	10.3%
Astigmatism	20	20.6%

TABLE 3:- Prevalence of refractive errors in urban and rural school children.

	Refractive errors (97)	Percentage
Urban	62	64
Rural	35	36

p value = 0.015

Out of the 97 children with refractive errors, 62(64%) children were from urban schools and 35(36%) children belonged to rural schools. This difference in prevalence was statistically significant with a p value of 0.015.

TABLE: 4:- Distribution of various types of refractive errors in school children in urban and rural areas.

Type of error	Urban children (62)	Rural children (35)	p value
Myopia (67) (69.1%)	42 (67.7%)	25 (71.4%)	0.066
Hypermetropia (10) (10.3%)	6 (9.7%)	4 (11.4%)	0.758
Astigmatism (20) (20.6%)	14 (22.6%)	6 (17.2%)	0.125

From the above observations it was found that the percentage of myopes was higher in rural areas (71.4%) compared to urban areas (67.7%). Similarly, the percentage of hypermetropes was higher in rural areas (11.4%) compared to urban areas (9.7%). These differences were not significant.

In contrast, astigmatism was more prevalent in urban children (22.6%), compared to rural children (17.2%) though it was not significant was found to be the least common error.

Discussion:-

School children form a vulnerable group affected by visual impairment. **Murthy GVS, Gupta SK, et al.**, have reported in their study that 81.7% visual impairment in school age children had been due to refractive error.

Both the children and their parents fail to recognize the vague symptoms produced by the presence of refractive errors. This creates subsequent detrimental effects on the educational potential, learning ability and the mental health of children.

Once these children leave school, the chances of getting themselves screened for vision is reduced. If diagnosed, refractive error can be treated safely and effectively by simple methods. Hence this study was aimed at school children aged 10-14 years.

In this present study, 97 out of the 600 school children had refractive errors. Of these 97 children with refractive errors, 62 belonged to urban schools and 35 belonged to the rural schools. The increase in the prevalence of refractive errors in urban school children compared to rural school children is statistically significant.

This result is in accordance with the results obtained by **Saw SM et al., 2001**. This higher prevalence of refractive errors in the urban school children may be attributed to a change in life style and improved socioeconomic conditions with increased duration of near work, better education facilities, television and computer usage and reduction in outdoor activities compared to their rural counter parts.

Studies by **Afghani T et al., 2003**, have showed that increasing literacy standards was associated with an increase in prevalence of refractive errors.

Ayub A et al, 2007 found out in his study that there was an increase in prevalence of refractive errors in private sector schools, compared to government sector schools due to presence of the above said similar reasons.

Myopia was found to be the commonest refractive error in our study being **69.09 %** of the total. **Astigmatism** was the next common error forming **20.62%** of the total. **Hypermetropia** was the least common of all the errors contributing **10.31%** of the total.

This is in accordance with the previous studies conducted on this subject by **Hussain AB, Ahmed EK**. The prevalence of refractive errors in school age children was 25.32% in their study. They also reported myopia in 63.5%, hypermetropia in 11.2% and astigmatism in 20.4% children.

Study of **Junaid SM et al**, shows an overall refractive error of 10 % in school children. Hypermetropia was present in 58% while myopia was present in 36% children. Astigmatism was identified in 6% children. However their study reported a higher prevalence of hypermetropia than myopia which is contradictory to this present study. Another

study carried out by **Ayub A et al** demonstrated myopia in 43%, hypermetropia in 21.5% and astigmatism in 35.5% children.

Conclusion:-

This study has confirmed the finding of others that there is an increase in the prevalence of refractive errors in children of urban areas compared to those in rural areas. Myopia was found to be the commonest error being followed by astigmatism. Hypermetropia was found to be the least common error.

With the advent of modern gadgets and improvement in technology, children are being constantly exposed more and more to the ill effects of these inventions. With increasing importance being given to academic activities and a reduction in duration of outdoor activities, due precautions must be taken by parents to ensure adequate visual hygiene in children.

Awareness on refractive errors and the importance of wearing glasses in children with refractive errors must be imparted in schools.

During the last few years, steps have been taken by the government to screen children in schools for early identification of refractive errors and their effective management by provision of spectacles.

Adequate screening procedures in schools, maintenance of visual hygiene and providing corrective measures will definitely reduce the burden of this problem in our society.

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RESEARCH ARTICLE

THE EFFECT OF FRAGMENTATION IN VEGETATIVE AND ECOLOGICAL DYNAMISM OF MYRISTICA SWAMP – A FRAGILE FRESHWATER ECOSYSTEM OF WESTERN GHATS.

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Abstract

A vast area of Western Ghats, coming under different forest types, are degraded and are threatened with ecological deprivation and biodiversity loss. Deforestation for urbanization, fragmentation due to developmental activities, over exploitation to meet the increased demand are the main anthropogenic threats. The freshwater swamps of Western Ghats dominated by the Myristicaceae members too face this loss of character. These ecosystems have unique biodiversity, bio wealth ecological functioning and supporting edaphic factors. Historically the natural landscape of Kerala is well represented by freshwater swamps and marshy lands which create a positive water regime and provide protection for unique flora and fauna. The high population density and high demand on space and livelihood of the state lead to the depletion or alteration of this natural landscape. In advanced stages, this leads to the fragmentation and isolation of the unique ecosystem. These changes in the ecological and edaphological characteristics will deprive these unique flora and fauna of the marshy condition to which it is adapted. The existing fragments of the freshwater swamp create a series of remnant patches, relic vegetation, surrounded by a matrix of different vegetation and by altered land use. Since *Myristica* swamps have been edaphically created with free flowing water stream, fragmentation initially results in the modification of velocity of water flow and changes in the silting or free flow of running water. The present investigation discusses the current distribution of swamps and its conservation status, ecological function and vegetational succession, by highlighting the effects of fragmentation and isolation of vegetation.

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1. Introduction:-

Myristica swamps are edaphic formations of fresh water evergreen forest type coming under type category 4C/FSI (Champion and Seth, 1968) represented by a unique phytogeographic condition with supporting characteristics like amphibian vegetation. The predominant arborescent members are from primitive families like Myristicaceae, Flacourtiaceae, Dipterocarpaceae, while the shrubby and herby flora is represented by members of advanced families like Araceae, Aricaceae, Zingiberaceae, Marantaceae, etc. The community status of Myristicaceae members

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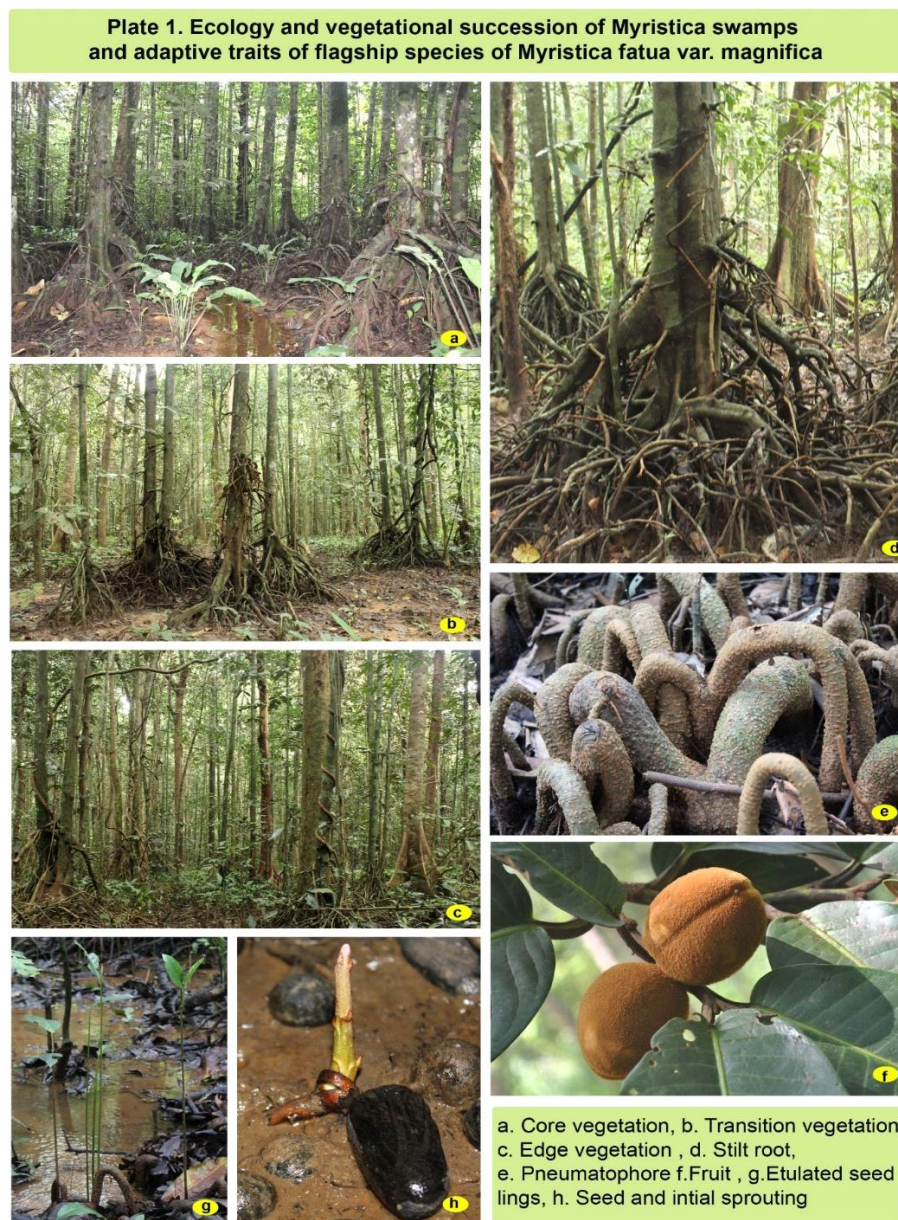
Address:-Plant Systematics and Evolutionary Science Division, Jawaharlal Nehru Tropical Botanic Garden and Research Institute, Karimancode P.O., Palode, Thiruvananthapuram- 695 562, Kerala, India.

is structurally and functionally dominant over other members, hence, the name *Myristica* swamps. This water logged edaphic environment and associated vegetations are distributed in the valleys or foot hills of Western Ghats and subjected to seasonal flooding and silting. The evergreen members are amphibian in nature specially adapted for surviving in marshy water logged condition as well as in terrestrial condition. These swamps were reported from different states along the Western Ghats (Saldanha, 1984; Gadgil&Chandran, 1989; Chandran&Gadgil 1989; Chandran&Mest, 2001; Vasudeva et al., 2001) Karnataka, Goa (Santhakumaran et al., 1995, 1999) and Kerala (Krishnamoorthy, 1960; Pascal, 1984). The ever green vegetation of swamps includes many endemic tree species and are well documented (Bourdillion, 1908; Sasidharan&Sivarajan, 1996; Ramesh and Pascal, 1997; Chandran&Mesta, 2011). The swamps are also inhabited by some of the red listed plants included in red data book of Indian plant (Nayar&Sastry, 1987, 1990). Hence, the *Myristica* swamps, with pristine vegetation are virtually live museums of ancient life of great biological interest. During the historical events of development and urbanization, these forest lands have been exploited and obviously perished. Land use changes like conversion to paddy field or plantation of areca, coffee, rubber or submergence under irrigation projects or extermination due to fire, all have had adverse impacts on these swamps (Krishnamoorthy, 1960). The existing swamps are also heading towards extinction. An example of such anthropogenic activity is evident in the Sasthanada swamp, Sankili section, Kulathupuzha Forest Range, Trivandrum Forest Division having 12 patches, extending to an area of 0.223 sq km (22.29 Ha). The fragmentation involves dividing up of continuous ecosystem into smaller areas called patches, and each patch has a relatively homogenous floristic, ecologic, and edaphic conditions. When fragments are created, plant dynamics is modified owing to changes in the forest spatial organization, such as the size, shape, and type of isolation barrier of forest patches. The functional dynamism of patches are affected with edge effects, migration and establishment of new species, decrease in the recruitment rate of specific species and increased recruitment rate of flexible species and disturbances in faunal interactions and disproportionally favoured and hence dominating species like *Lagerstromaeaspeciosa* and while others like *Myristica fatua* var. *magnifica* are facing local extinction. This vegetational, structural and functional changes are generating changes in the availability of water, nutrients, light, wind and other basic environmental factors by creating different degrees of ecological succession in the same area viz. Core vegetation, the unchanged original vegetation; Edge vegetation, representing transformed one. In between the core and the edge there is transitional vegetation, which are in different stage of succession. In short, the fragmentation due to this change in land use causes, the vegetation which once was supported by wetland (core vegetation) changing to the terrestrial area vegetation (edge vegetation). Thus the isolation and fragmentation of *Myristica* swamps lead to the breakup of these natural ecosystems into smaller patches of vegetation posing threat to their conservation status. Changes in the physical edaphology and micro climate will alter the reproductive phenology of the members of the original vegetation, especially biota like *Myristica fatua* var. *magnifica*. It is also noticed that this unique ecosystems are also seen as Sacred Grove, the patches of vegetation protected in the religious ground to avoid the perceived wrath of its resident God. The community status of flagship species viz. *Myristica fatua* var. *magnifica* fluctuates in advance with vegetational succession indicating past massive fragmentation of swamp during the urbanization of the state and thus these habitats can be described as “swampy relics” especially due to fragmentation and isolation followed by the vegetational succession, ecological and edaphological changes. The present study puts emphasis on the trends of vegetational succession due to fragmentation and effect of invasion of other species in the swamp.

Methodology:-

2.1.Study area:-

Kerala, the most south-westerly state of India, is located between latitudes 8° 17' 30" N. and 12° 47' 40" N. and longitudes 74° 27' 47" E. and 77° 37' 12" E. The state is isolated from the Deccan Plateau by the Western-Ghats, one of the biodiversity hotspots of India. The *Myristica* swamp of Sasthanada region, Sankili section of Kulathupuzha Forest Range (8°45'82"N.and 77°10'55"E.) is situated at an elevation of 155 MSL. The vegetation of this *Myristica* swamps is in the three stages of succession such as core (Figure legend A), transition (Figure legend B) and edge zone (Figure legend C).



2.2. Vegetation sampling:-

A detailed exploration was made in entire study area. The vegetational type, floristic composition and anthropogenic pressure were identified. The whole area is divided into three regions the core, transition and edge area. The line transects were laid across all types. The phytosociological features and floristic status were assessed by laying quadrates of size 15 x 10 in random. All the members were identified and enumerated. Height and girth at breast height (GBH) of all trees and shrubs (≥ 10 cm GBH) were measured using clinometer and metallic measuring tape respectively. Vegetation with less than 10 cm GBH were counted and enumerated as seedlings or herb species. The identification of the species was done by referring authentic books. The following parameters were calculated using this vegetation data.

2.3. Phytosociological analysis:-

Relative frequency (rF), Relative density (rd), Relative dominance (rD) were calculated and summed up to get the Important Value Index (Barbour et al., 1947), the calculation is done by using the following formulae.

$$\text{Relative frequency (rF)} = \frac{\text{Frequency of } i^{\text{th}} \text{ species}}{\text{Total frequency of all the species}} \times 100$$

$$\text{Relative density (rd)} = \frac{\text{No. of individuals of } i^{\text{th}} \text{ species}}{\text{Total frequency of all the species}} \times 100$$

$$\text{Relative dominance (rD)} = \frac{\text{Sum of the basal area of the individual of the } i^{\text{th}} \text{ species}}{\text{Total basal area of all the individuals}} \times 100$$

$$\text{Basal area} = \frac{(\text{GBH})^2}{4\pi}$$

$$\text{Important Value Index (IVI)} = rF + rD + rd$$

2.4. Density:-

The density of trees were found out by using point centred quadrat method (Barbour et al., 1971) Random points were located, the area around each point was divided into four 90° quadrates and the nearest of each quadrat was marked. Average distance for all the trees were taken together and computed for density as per the formula given,

$$\text{Density (Trees/ hectare)} = \frac{10000}{2(\text{Average distance in meter})^2}$$

2.5. Whiteford index:-

It is the ratio of abundance and frequency. In general, an index value of 0.025 to 0.05 indicates that the species is random, while the value higher than 0.05 indicates increase in aggregation and value lower than 0.025 indicates a tendency towards regularity (Whiteford, 1948). It was calculated using the formula.

$$\text{Whiteford index} = \frac{\text{Abundance}}{\text{Frequency}}$$

$$\text{Frequency} = \frac{\text{Total no. of quadrates in which species occurred}}{\text{Total no. of quadrates studied}} \times$$

$$\text{Abundance} = \frac{\text{Total no. individuals of species}}{\text{Total no. of quadrates occurrence}}$$

2.6. Similarity index:-

The value of similarity index indicates the affinity between each community. It is derived by the formula of Sorenson (1948).

$$\text{Similarity index} = 2 \times \frac{\text{No. of common species}}{\text{Total no. species}}$$

2.7. Floristic diversity indices:-

The species diversity within a species, community or within a habitat is calculated to assess the biodiversity. The diversity comprises of two components via, species richness and evenness or equitability. A number of indices have been reported which measure both components of species diversity, i.e. Species richness and evenness into a single index. They are discussed below.

2.7.1. Simpson's index (D):-

The index measures the probability that two individuals selected at random from a sample will belong to the same species and range from 0-1 (Simpson, 1949).

$$D = 1 - \sum_{i=1}^S [ni/N]^2$$

ni = Number of individuals

N = total number of species

2.7.2. Shannon- Wiener Index (H'):-

This index of diversity is based on information theory. The information content is measure of the amount of uncertainty. The index is zero and maximum when all the species are represented by the same number of individuals. It generally falls between 1.5 to 3.5 and rarely exceeds 4.5 (Shannon- Wiener, 1963).

$$H' = - \sum_{i=1}^S \left[\left(\frac{n_i}{N} \right) \ln \left(\frac{n_i}{N} \right) \right]$$

n_i = Number of individuals

N = total number of species

2.7.3. Equitability (J):-

The natural distribution of individuals among the species is called species evenness or equitability (Gupta and Shukla, 1991; Pielou, 1975).

$$J = H'/H \text{ (max)}$$

$$H \text{ (max)} = \log_2 S$$

S = Number of species in a quadrat

2.7.4. Species richness:-

The index of species richness (R) was calculated using the formula given by Menhinick (1964).

$$R = S/\sqrt{N}$$

R = Richness index

S = total number of species in the community

N = total number of individuals summed over all species.

2.7.5. Variety ratio:-

It indicates the diversity of each species with reference to the total number of individuals in unit area. The high value indicates high diversity of species.

$$\text{Variety ratio} = \frac{\text{Total number of species (S)}}{\text{Total number of individuals (N)}}$$

2.8 Regenerative status:-

The natural germination percentage, seedling establishment and casualty rate were calculated by counting methods. Randomly selected 10x10 meter quadrates were placed, and number converted into percentage.

Results:-

The *Myristica* swamps are the freshwater evergreen vegetated ecosystems predominated by the members of arborescent family Myristicaceae. The *Myristica fatua* var. *magnifica*, *Gymnacrantheracananica* are the upper canopy dominant species, which constitute thick and continuous upper strata. These two species are structurally modified to survive in the swampy condition, especially with knee roots and silt roots (Plate 1.d & e). The Myristicaceae members like *Myristica malabarica*, *Knema attenuta* are commonly seen as dominant associate species; hence the name *Myristica Swamps* was derived. The non- Myristicaceae tree members like *Lophopetalum wightianum*, *Lagerstroemia speciosa*, *Hydnocarpus pentandra*, *Holigarna arnottiana*, *Syzygium travancoricum* etc. are also well distributed in this forest type and sometime fall in the dominant category. The second and third strata are also well represented with younger members of these species with discontinuous canopy. The migrant and flexible species like *Vateria indica* and *Hopea parviflora*, are also distributed with different magnifications, accordingly with successional status of the swamp. The ground vegetation is mostly represented with *Lagenandra ovata*, *Calamus rotang*, *Pinangadicksonii*, *Pandanuskaida*. The lianas and climbers are mostly represented by *Gnetumula*, *Entadarheedei*, *Jasminum cordifolium*, and *Piper spp.* The floristic composition of ground vegetation and occurrence of the liana members are also dependent on degree of succession. Analysing the degree of similarity and dissimilarity between the isolated fragments of the swampy vegetation is typified into three categories according to the different degree of vegetational succession, namely Core vegetation, dominated with the species like *Myristica fatua* var. *magnifica* and *Gymnacrantheracananica*, the transition vegetation has also the same dominant members with less degree of community status, while the edge vegetation is represented by dominant

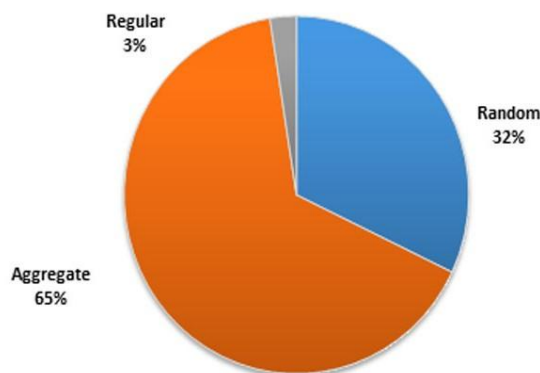
species *Lagerstroemia speciosa* and, *Myristica fatua* var. *magnifica*. The recorded relative occurrence and community status of *Myristica fatua* in terms of IVI are 86.45 in core area, 49.81 in transition zone and 22.34 in edge area. This also indicates a gradual decrease in occurrence and dominance of the *Myristica fatua* var. *magnifica*. *Gymnacranthera canarica* shows the value of 52.58, 27.02, and 19.89 respectively. Other associate species show different degrees of community status with respects to IVI value. (Table 1). As we look into the similarity of these three zones closely using the similarity index, the vegetative structure clearly represents an inverse relation. The core and the edge show very less similarity (29.5%) compared to the core and transition zone (45.9%). whereas the transition zone shows more affinity towards edge vegetation (72%). This represents the invasion of other species from edge to the transition and this trend will later result in the extinction of the swamp and the replacement of the terrestrial forest vegetation.

Table 1:- The Community Status of Important Tree species of Myristica swamps in Three Succession Zone

Sl. No.	Binomial	Core Area	Transition Zone	Edge Area
1	<i>Aporosa lindleyana</i>		16.75	05.12
2	<i>Chilocarpus denudatus</i>	13.92	06.82	08.53
3	<i>Deris pinnatum</i>		04.47	03.95
4	<i>Gymnacranthera canarica</i>	52.56	19.89	49.37
5	<i>Holigarna arnottiana</i>	16.10	06.38	24.15
6	<i>Hopea parviflora</i>	-	14.58	16.73
7	<i>Hydnocarpus pentandra</i>	11.74	10.00	13.85
8	<i>Lagerstroemia speciosa</i>	23.50	30.90	08.90
9	<i>Lophopetalum wightianum</i>	33.75	09.48	19.85
10	<i>Myristica fatua</i> var. <i>magnifica</i>	86.43	22.34	59.81
11	<i>Neolamarckia cadamba</i>	06.74	-	16.60
12	<i>Syzygium travancoricum</i>	-	06.30	10.50
13	<i>Vateria indica</i>	-	08.00	20.49
14	<i>Xanthophyllum arnottianum</i>	-	11.20	04.85

The floristic composition and diversity of Myristica swamps also vary with vegetational succession, due to the isolation and fragmentation. The floristic composition of swamp includes 82 species belonging to 72 genera of 44 families. The distribution pattern of swampy vegetation shows, 32% of species random in occurrence, 65% in aggregate distribution and the remaining 03% with regular distribution pattern. This indicates the conservation value of the system; hence any alteration in ecosystem will lead to the massive extinction of species (Fig.1).

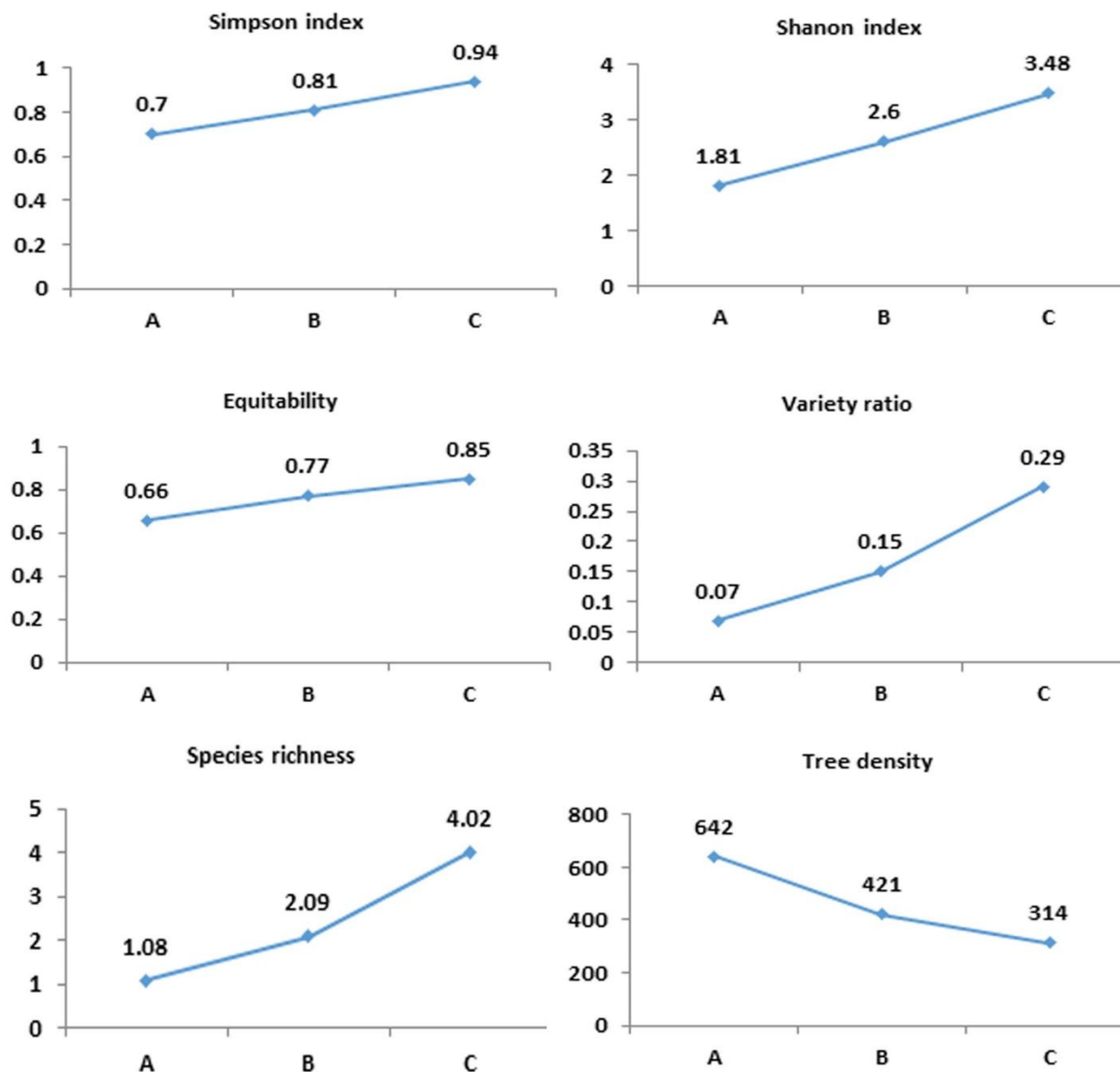
Fig.1: Vegetation Distribution pattern in Myristica Swamp



In the core area, the floristic diversity value, Simpson index is, 0.70, while in the transition zone, the value is, 0.81 and in the edge area the highest values which are about equal to that of the evergreen forests of Western Ghats i.e. 0.94 have been recorded. The same increasing trend has been recorded in the Shannon-Weinner index. The respective values are 1.81, 2.60, and 3.48. The Equitability value of core area is 0.66, transition zone value is 0.77

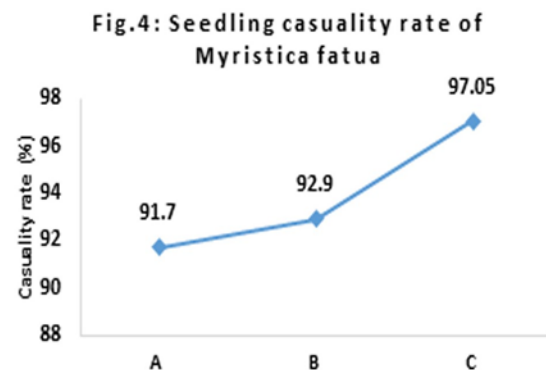
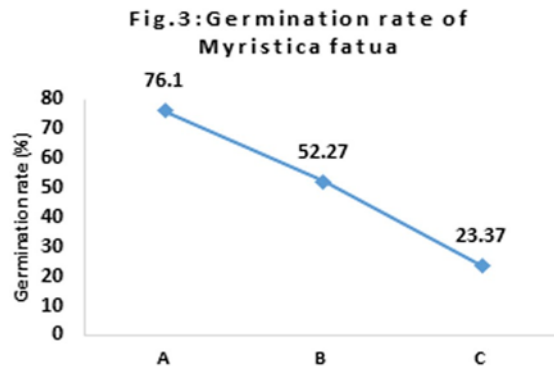
and edge area value is 0.85. The species richness also shows a decrease from 1.0 in the core area, 2.1 in the transition and 4.0 in the edge zone. The variety ratio also shows the standard of increasing the value by the advancement of ecological succession; the respective values are 0.07, 0.15 and 0.29. The density of tree species also shows changes with advancement of the ecosystem. In the core area of the swamp the tree density is 642 individuals per hectare, it decreases to 421 (68.5% of core area) in transition zone and further to 314 (48.9% of core area) in edge zone. The fragmentation and isolation of the specific ecological niche of the unique species of swamp ecosystems are declining or eroding gradually and lead to evolution and development of new vegetation and forest types (Fig.2).

Fig.2: Effect of fragmentation and isolation in florestic wealth and diversity of Myristica Swamps

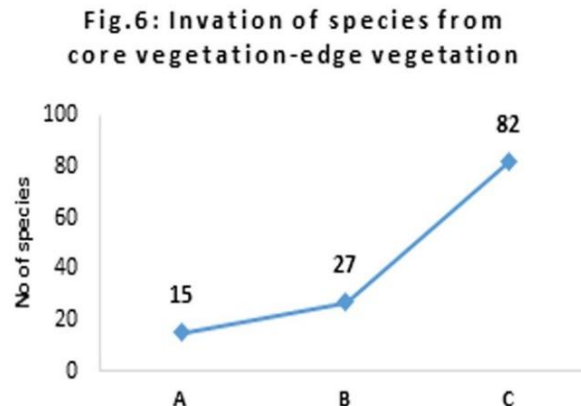
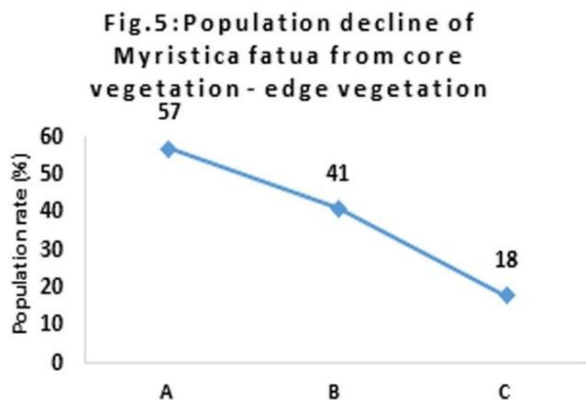


The study about the reproductive capacity and aggressive capacity of *Myristica fatua* shows that the species has high degree of reproductive capacity with gregarious flowering and good fruit setting. The average seeding is 56 seeds in one sq. meter area and 92% of these seeds are viable. The primary dispersal rate is 58-66 %, mainly by *Ocercerosgriseus* (Hornbill), *Phaethontidaepsittaciformes* (Parrot) and *Ratafaindica* (Malabar Squirrel). The premature shedding due to the feeding of avian fauna like *Phaethontidaepsittaciformes* (Parrot), *Ocercerosgriseus* (Hornbill) and mammalian fauna like *Ratafaindica* (Malabar Squirrel), *Rhesus macaque* (common Monkey) were

also noticed. 48% of secondary dispersal was also noticed. The main dispersing agents are the common crustacean *Barytelphusa guerinii* (tropical freshwater Crabs) and *Rattus rattus* (Rat). The germination percentage in natural condition varies from 76.10 % to 23.37 % (core to edge) with an average value of 57.00 % (Fig.3). The subsequent growth and establishment of seedlings up to two years were analysed and the same trend was observed as for *Myristica fatua* and it also varies in accordance with different degree of succession and fluctuation. In the core area, the casualty rate is 91.70 %, in the transition zone, 92.90 % and 97.05 % in edge area. (Fig.4)



The *Myristica* swamps are the edaphic variant of tropical freshwater swamps and are evergreen in nature. These succumb to many threats due to the destructive and interactive face created by the natural and artificial factors. In turn it formulates various ecological and edaphological units. The population of unique species *Myristica fatua* var. *magnifica* in its different age class shows aggressive trends of declining population size through the advancement of transition of vegetation. In core area, the rate of population of *Myristica fatua* is 57%. While in transition vegetation the rate of population of *Myristica fatua* is 41%, in edge vegetation, surviving with other forest intruders, the *Myristica fatua* population is drastically declines to 18% of the total population. The analysis shows that invading of other evergreen species to the swampy area will cause the switch over of the swampy vegetation to evergreen vegetation (Fig. 5&6). The core area shows high degree of germination percentage and initial survival rate. The increased mortality rate indicates the specific ecological and edaphological requirements of the species for successful establishment. The fragmentation and isolation of this ecosystem has resulted in the decline in the number of individuals of structurally and functionally dominant specific species, *Myristica fatua* var. *magnifica* and hence it is considered as flagship species.



Discussion:-

Human alteration of the global environment has triggered six major extinction event in the history of the life and has caused wide spread changes in the abundance and distribution of the organisms (Han Olff and M.E Ritchie, 2002). Land use changes have the greatest impact on the biodiversity of the tropical forest area (Sala et al., 2000). Habitat fragmentation on a large scale occurs in the freshwater swamp ecosystem of the Western Ghats, which progressively gets subdivided into smaller and more isolated vegetation having interrupted matter and energy circulation. The changes in the ecological distribution has subsequently affected the floristic composition, vegetational structure and functional dynamism across other biotic and abiotic components stimulating a backdrop of natural patches created by changing landform and natural disturbance. This has led to different degrees of vegetational succession replacing the species that have survived in the swampy habitats. The fragmentation and isolation of ecosystem followed by the disruption and discontinuation of the flora and fauna will interrupt the functional dynamism of the ecosystem by checking energy and matter flow. These also result in the formation of different plant sub communities within the swamps itself. The different floristic elements occupy different space with different association having different boundaries, each with a varying degree of uniformity in structure and composition. By analysing the degree of similarity and dissimilarity between the floristic elements of fragments and isolated segments, a gradual gradation indicates the progressive trends of floristic fluctuation and succession. The vegetation is Phanero-thero type, dominated by trees and herbs, among which 80% of the arborescent species are paleoendemic and members of primitive families. Considering this primitive compositions, vegetation type, and antiquity, this swampy vegetation may be probably the remains of the most ancient forest of the Western Ghats. These unique and vital forest types are on the verge of extinction due to natural and manmade reasons. Nevertheless the fragmented freshwater swamps preserve remnants of vegetation thereby maintaining the conservation status of rare ecosystems to some extent.

The changing community structure of *Myristica* swamps are the primary driving force of vegetational changes, while direct effect of human activities like artificial constructions, habitat destruction, alternation of the land use pattern, exploitation for livelihood and introduction of alien species are secondary forces of change. Hobbs and Yates in 2003 and Laurence et al. in 2002 observed the same in other forest ecosystems. The fragmentation is also creating modified plant dynamism owing to the changes in the atypical organization in the size, shape and isolation of the forest patches. The edge effect and following invasion of other species have altered the internal microclimate and also the nutrient status of the soil system. These changes, the disproportionate establishment of plant species immediately affect the establishment of flagship species, *Myristica fatua* var. *magnifica*, the dominant species of the core area is being locally extinct in the edge area. Even in the transition zone, the number of the individual are not enough to specify the characteristic features of the swamp. The gradation of the floristic diversity indices and tree density indicates that this process is causing difference not only in the diversity but also in the bio wealth of core, transition and edge area. The change in the important value index indicates the changes in the community status of the common species of the swamp. The fragmentation of the swamp has a tendency to lose original species like *Myristica fatua* var. *magnifica*, *Gymnacranthera canarica*, and gain new species like *Lophopetalum wightianum*, *Vateria indica*, *Hopea parviflora*, the species that are capable of establishing fragmented swamp having new ecology and edaphology, initially by altering the water regime due to the acceleration in the silting and breakage of water flow. This alternate pattern of water flow leads to changes in normal drainage pattern and creates a modified substrate and ultimately leads to reduction of new environment for the plant colonization. The modified water regime results in the removal of specific underground swampy amphibian vegetation and can act as a focus for invasion of new species of terrestrial vegetation. The requisite micro niche of some vital faunal association also disturbed. The secondary seed dispersal of *Myristica fatua* var. *magnifica* were affected, hence successful establishment of the flagship species is affected in recruitment stage. The changes will also lead to changes in the humus deposition and decomposition rate. The flood washed off most of the debris and decrease saprophytic faunal diversity. This has altered the fauna abdication as well as the seed bed characterization resulting in mineral inequilibrium of soil system. The changes in the floristic composition and vegetational structure, upon the fragmentation of the *Myristica* swamp, and the production of remnant vegetation patches are likely to result in more flexible species getting established. Lovejoy et al., 1986 had reported that fragmentation in the tropical forest resulting in the rapid growth of vine and secondary vegetation in a 10-25.m, strip around the remnant edge. The freshwater swamps of Kerala once continuously distributed in low elevated lateritic hill rocks of the Western Ghats, were fragmented due to urbanization and other anthropogenic or natural reasons and are now distributed as relic vegetation, both in natural forest areas of southern Western Ghats and in the midst of the populated areas as sacred groves. The plant species *Myristica fatua* var. *magnifica* can only survive and tend to occur only in this specific ecosystem, hence they termed as 'flagship species'. In addition to this an interactive faunal association of *Barytelphusa gujerini* affects the secondary seed dispersal of *Myristica fatua* var. *magnifica*. The fragmentation in

swamp ecosystem is altered and a modified energy flow is created by generating a dispersal barrier for different life forms especially for flagship species or the keystone species.

Conclusion:-

Myristica Swamps are unique fresh water phyto-geographic ecosystems of Western Ghats, once distributed in low laying areas of Kerala State in a continuous manner. The floristic composition, ecology and edaphology are specific with high occurrence and distribution of Myristicaceae members, especially of flag ship species, *Myristica fatua* var. *magnifica*. The members are structurally modified due to inundated during greater part of the year. Presently, due to natural and manmade changes these are under threat and in path of extinction. The existing treasure home of biowealth, fresh water swamps, requires more attention to conserve, especially to check the local extinction of a few vital species and its associate. The typical swamps are characterised by low floristic diversity and high stem density. The vegetation is amphibian features, adapted to survive in marshy conditions. The germination and recruitment of new members of the important species are related to general ecology and physical edaphology including some faunal associations. These forest relics are facing various levels of disturbance, among which fragmentation is in prime position, which would be causative factor regarding the loss of swamp ecosystems. The fragmentation and isolation alter floristic structure, composition, micro climate, and water regime. These initial floristic, ecological and edaphological changes ends with formation of new ecosystem with common floristic composition, terrestrial ever green forest.

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RESEARCH ARTICLE

POLYPHENOLS CONTENT AND EVALUATION OF ANTIOXIDANT ACTIVITY OF ANACYCLUSPYRETHRUM(L.) LAG. FROM TIMAHDITE A MOROCCAN MIDDLE ATLAS REGION.

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Abstract

As part of valorisation of medicinal and aromatic plants from Timahdite, a region located in Moroccan Middle Atlas, we focused our study on an endemic species which belongs to Asteraceae family: *Anacyclus pyrethrum* L. This species, widely used by local people in traditional pharmacopeia, was chosen to conduct phytochemical studies and to assess the antioxidant activity of its roots, leaves and flowers extracts.

Thus, the plant was harvested in May 2014. The selected organs (roots, leaves and flowers) of *Anacyclus pyrethrum* were sorted, cleaned and dried separately. Then, they were reduced into powder and stored away from light in a cool and dry place until use. Each organ was subjected to a phytochemical screening in order to qualitatively highlight its secondary metabolites.

This qualitative analysis shows that roots are rich in alkaloids while the aerial parts (leaves and flowers) are rich in tannins and flavonoids. Extraction of total polyphenols was performed by maceration using methanol / water (70/30) as solvent. Yields were 2.30% for roots, 3.3% for leaves and 3.8% for flowers.

Then, fractionation of hydro-methanol crude extract was carried out using successively three organic solvents of increasing polarity: chloroform, ethyl acetate and butanol. Quantitative analysis of total polyphenols conducted with Folin-Ciocalteu reagent showed that hydro-methanol crude extracts are richer in phenolic compounds than other fractions.

Antioxidant activity of crude extracts determined through DPPH test was spectrophotometrically quantified and values of the half-maximal inhibitory concentrations (IC₅₀) were graphically determined. These values are 77.62 µg gallic acid equivalent (GAE) / ml for flowers, 94.18 µg / ml for leaves and 145.81 µg / ml for roots, while 46.79 µg / ml was found for ascorbic acid which was used as standard.

Finally, a correlation was observed between total polyphenols content and antioxidant capacity determined by DPPH method: Aerial parts

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(leafy stem and flowers) richer in polyphenols exert a better antioxidant activity compared to roots.

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Introduction:-

These last decades have been marked by the widespread use of synthetic products in various industrial fields. Through years, this practice in the case of synthetic antioxidant molecules has often led to potential toxicological risks. Now, their use is reconsidered by international organizations and new plants are sought as a source of natural antioxidants [1,2]. Thanks to their properties, these natural antioxidants attract more interest in various fields. They are used to prevent and to treat cancer, inflammatory and cardiovascular diseases [3]. They are also used as additives in food, pharmaceutical and cosmetic products [1]. Thus, scientific research has been developed with the aim to extract, identify and quantify antioxidant compounds from natural sources such as medicinal plants and food products. In this quest for safe natural antioxidants, polyphenolics, which are widespread natural plant compounds, are of great interest because of their beneficial effects on health [4].

With the aim to find promising phenolic compounds, our research focuses on *Anacyclus pyrethrum* which is a Moroccan and Algerian endemic species belonging to *Asteraceae* family [5]. This perennial plant is widely prevalent in Moroccan Middle Atlas [6]. It is a 40 to 60 cm-height species which is characterized by long and thick roots. Its numerous simple or ramified stems lie on the ground and bear pinnate, alternate, finely cut, delicate and pubescent leaves. Its yellow-heart flowers are composed of white petals inside and purple petals outside. The species is well appreciated as a remedy against toothache [5], digestive disorders [8]. It also acts as a warming agent to enhance women fecundity and constitute a tonic agent for the nervous system [9]. *Anacyclus pyrethrum* is also used to treat cough, cold as well as heart, lung and digestive diseases [10]. For this medicinal value, this plant was selected for the present study. Moreover, review on this species showed that its chemistry remains insufficiently studied. According to our knowledge, no research work was conducted on the aerial parts of the plant. Thus, our work aims to contribute to a complete phytochemical study of *A. pyrethrum* and to contribute to the explanation of its various activities. We focused our interest on the wild species which particularly grows in Timahdite's forests, at altitudes ranging from 400 to 3100m. [11].

The aim of our work is to quantify *Anacyclus pyrethrum*'s phenolics in the extracts of three organs and to assess their antioxidant power by DPPH method.

Material and methods:-

plant material

Anacyclus pyrethrum was harvested in May 2014 at 1800m of altitude in Timahdite, a region located at 30km south from Azrou. Samples were harvested with their roots, stems, leaves and flowers in order to make their identification easier. This plant identification was conducted in the laboratory of Botany and Plant Ecology at the Scientific Institute of Rabat by Professor MOHAMED IBN TATOU. Samples of different organs (roots, flowers and leaves) were cleaned, shade dried at room temperature and then stored away from light until use.

phytochemical screening :-

Plants organs (roots, leaves and flowers) were powdered with an electric grinder. After this step, chemical composition of the drug was determined by a phytochemical study based on solubility tests, colour and precipitation reactions and also by observations under ultraviolet light.

The target phytochemical groups in these tests include: alkaloids, polyphenols (flavonoids, anthocyanins, tannins, ...), saponins, coumarins, sterols, terpenes, mucilage, carotenoids, reducing sugars (oses and holosides)

- Research on alkaloids was performed using Dragendorff and Mayer's reagents which are reagents for general characterization of alkaloids [12].
- Characterization of tannins was carried out with Iron chloride reagent. The differentiation between gallic and catechic tannins was shown thanks to Stiasny's reaction [13].
- Identification of flavonoids was performed by cyanidin test [14].
- Research of saponosides was quantitatively determined by calculating the foam index after shaking the aqueous extract [15].
- Anthraquinones were identified by Borntrager's test [16].

- Sterols and terpenes were highlighted by Liebermann-Burchard's test [17].
- Coumarins were demonstrated by UV fluorescence at 365 nm [13].
- Mucilages were indicated by the presence of a flocculent precipitate after adding ethyl ether in a test tube containing plant's decoction [18].

Phytochemical screening was carried out separately on plant's leaves, flowers and roots.

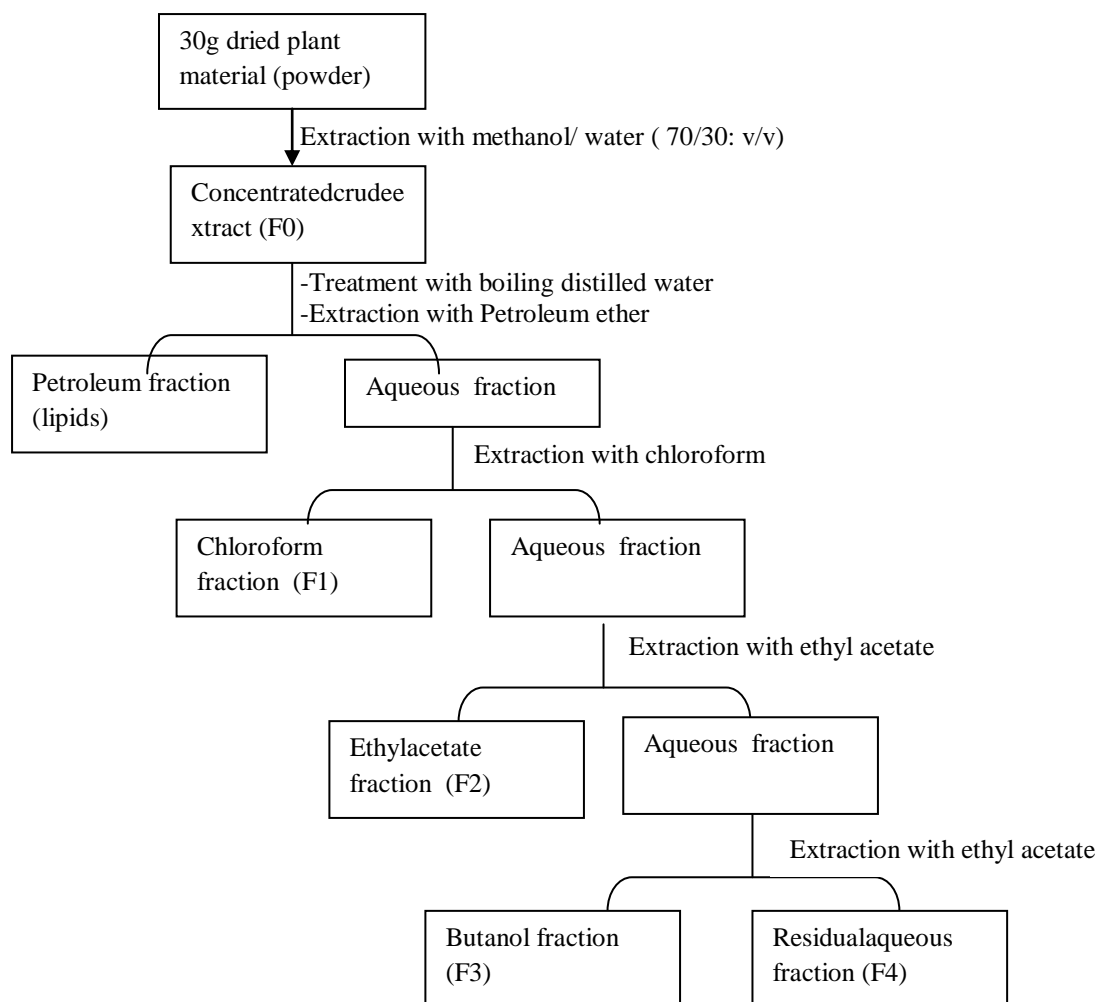
Preparation of extracts:-

Test samples composed of 30 g powder of root, leaves and flowers of *Anacyclus pyrethrum* was macerated in a hydro-alcoholic mixture (methanol / water 70/30: v / v) with magnetic stirring for 30 min. The mixture was then stored in the dark for 24 hours at room temperature. The obtained solution was filtered on filter paper and the filtrate was concentrated under vacuum at 50 ° C with a Büchi-type rotary evaporator [19]. The dry extract was then weighed, labelled and stored at 4 ° C until use.

Fractionation:-

Polyphenols extraction was performed according to Bruneton's method [15] with a slight modification. It is based on polyphenols' degree of solubility in organic solvents. Fractionation of crude hydro-methanol extracts was carried out by successive use of four organic solvents with different polarity: petroleum ether, chloroform, ethyl acetate and butanol.

In addition to the hydro-methanol crude extract (F0), four fractions were selected for further work: chloroform fraction (F1), ethyl acetate fraction (F2), butanol fraction (F3) and residual aqueous fraction (F4). The different extracts were kept cold away from light until use. Picture 1 summarises fractionation steps of the crude extracts.



Picture 1:- Fractionation steps of crude extracts.

Determination of Total Phenols Content :-

Total polyphenol content of *A. pyrethrum*'s crude extracts was determined by Folin-Ciocalteu's method [20]. In a 100 ml-volumetric flask, 40 ml of each extract were mixed with 1.5 ml of an aqueous solution of sodium bicarbonate (Na_2CO_3) at 10 % and 1.5 ml of Folin-Ciocalteu reagent (10fold diluted). Then the flask was supplemented with distilled water. The mixture was stirred and incubated in the dark at room temperature for 30 minutes. The reading step was performed against a blank using a spectrophotometer (UV mini-1240) at 765 nm. A standard curve with gallic acid as positive control was established in parallel under the same operating conditions. Results are expressed as milligrams of gallic acid equivalent per gram of extract (mg GAE / g).

Determination of flavonoids contents:-

Total flavonoids content of *A. pyrethrum*'s extracts was determined according to aluminumtrichloride method. The mixture was stirred and then incubated in the dark at room temperature for 30 minutes. A blank is prepared by replacing the extracts by methanol and the absorbance is read at 433 nm with a UV spectrophotometer. Results are expressed as mg of quercetin equivalent / gram of dry plant material. Quercetin calibration curve served as a standard.

Assessment of antioxidant activity :-**Dpph Test:-****Principle:-**

DPPH (2,2-diphenyl-1-picrylhydrazyl) is generally the most widely used substrate for rapid and direct assessment of antioxidant activity thanks to the stability of its free radical form and the simplicity of the analysis. This method is based on the measurement of antioxidants' ability to scavenge the DPPH radical [21]. Indeed, the latter is characterized by its ability to generate stable free radicals. This stability is due to the relocation of free electrons in the molecule, this relocation leads to a violet blue colour which is characteristic of the DPPH solution. Evaluation of antioxidant effectiveness is spectrophotometrically assessed at 515 nm through the decrease of the blue colour due to recombination of DPPH radicals [22]. In the presence of free radical scavengers, purple-coloured DPPH (2,2-diphenyl-1-picrylhydrazyl), is reduced into 2,2-diphenyl hydrazine -1- picryl which presents a yellow colour in the presence of free radical compounds [23].

Protocole:-

Ethanol solutions of ethyl acetate fraction were prepared. The following concentrations were obtained: 16; 32; 48; 64; 80; 96; 112; 128; 144; 160; 176 and 192 $\mu\text{g} / \text{ml}$. 200 μl of each ethanol solution were added to 1.8 ml of DPPH* solution which is also prepared in ethanol (0.024g / l). In parallel, a negative control was prepared by mixing 200 μl of ethanol with 1.8 ml of ethanol DPPH* solution. Absorbance was read against a blank at 515 nm after 30 min of incubation in the dark at room temperature. Ascorbic acid solution was used as a positive control. Absorbance was read in the same conditions for samples and controls. The test is repeated three times. Results are expressed as DPPH* reduction percentage. This percentage is calculated using the following formula:

$$\% P = [(A_{bl} - A_s) / A_{bl}] \times 100$$

P : DPPH* reduction percentage; A_{bl} : blank absorbance; A_s : sample absorbance

Results and discussion:-**phytochemical tests :-**

Results of phytochemical tests are gathered in table 1.

Table1:- Results of phytochemical screening of *Anacyclus pyrethrum*'s hydro-methanol extracts

Chemical class	Organs		
	Roots (R)	Leaves (LS)	Flowers (F)
Alkaloids	+++	+	+
Gallic tannins	+	+	+
Catechic tannins	+	++	+++
Flavonoids	-	++	+++
Anthocyanins	-	-	-
Leucoanthocyanins	+++	++	++
Saponosides	+	+	-
Free anthracenic compounds	-	-	-
Combined anthracenic compounds	+++	++	++

Coumarins	+	++	+
Carotenoids	-	-	-
Sterols and triterpenes	+	++	+
Reducing compound	+++	++	++
Oses et holosids	+++	++	++
Mucilages	+++	+	+
Narcotics	-	-	-

(+++): strong reaction; (++): moderate reaction; (+): weak reaction ;(-): no reaction

Results of phytochemical tests have shown that:

- Alkaloids are present in all three parts of the plant, but in small quantities in the aerial parts (leaves and flowers) compared to roots.
- Flavonoids are not detected in roots, but leaves contain mainly flavanols and flavanonols (red colour) while flowers essentially contain flavones (orange-pink colour).
- Tannins: Based on the colour obtained with FeCl₃ reagent, different parts of this species contain gallic tannin. Catechic tannins are less important in roots compared to leaves and flowers. Their presence is confirmed by Stiasny's reaction.
- Saponosides were detected and foams indices show that saponins content is low (1.1 cm) in leaves and roots. They are clearly absent in flowers.
- Sterols and terpenes are detected in a higher content in leaves compared to flowers and roots. A violet colour appears and then turns green.
- Coumarins presence is remarked thanks to fluorescent spots observed during qualitative analysis.
- Reducing compounds are present in large amount in the three parts of the plant.
- Finally, it should be noted that carotenoids, narcotics, free anthracenic compounds (anthraquinone) and anthocyanins are absent or rare in all studied parts of the plant.

The roots of *A. pyrethrum* mainly contain alkaloids and tannins. Similar results have been shown by Bellakhdar, 1996 [24], Hmamouchi, 1999,[25],Center for Mediterranean Cooperation, 2005[26], Selles et al., 2012[27], Limbachia&Jadhav, 2012 [28]). They also contain mucilage. The presence of mucilage in the roots of *A. pyrethrum* allowed us to note the viscous character of the extracts. During our phytochemical tests, we have demonstrated the presence of coumarins, sterols and triterpenes in the roots of *A. pyrethrum* but in small amount. These results are consistent with the previous works of Hamimed, 2009 [29]; Sellese et al., 2012[30]. The roots of *A. pyrethrum* also contain oses, holosides and reducing compounds. This was highlighted by (Selles C., 2012) [30]. Hence, the chemical composition of the roots of *A. pyrethrum* has proved to be interesting and rich in compounds known as bioactive molecules (Hamimed, 2009 [29]; Christian, 2013[31]). The overall results justify the various activities of this plant including the use of part of its chemical compounds as alternative to synthetic antioxidants.....

Extraction yields :-

Yields of crude hydro-methanol extracts from roots, leaves and flowers are respectively: 2.3%, 3.3% and 3.3% (Table 2).

For crude extracts fractionation, results show that butanol fraction has the highest yields (1.93%, 3.2% and 3.7%) followed by the aqueous fraction (1.33%; 2.9% and 3.2%) and then the ethyl acetate fraction (1.23%, 2.23% and 2.4%). On the other side, the lowest yield was obtained with the chloroform fraction: 0.76%, 1.8% and 2.1% respectively for roots, leaves and flowers (Table 2).

Table 2:- Yields of polyphenol extracts and fractions obtained from *Anacyclus pyrethrum* 'sorgans

Extracts and fractions from <i>A. pyrethrum</i> 'sorgans	Yields (%)		
	Roots (R)	leaves (LS)	Flowers (F)
Crude hydro-methanol extract	2.30%	3.3%	3.8%
Chloroform fraction	0.76%	1.8%	2.1%
Ethyl acetate fraction	1.23%	2.23%	2.4%
Butanol fraction	1.93%	3.2%	3.7%
Aqueous fraction	1.33%	2.9%	3.2%

total phénol content

UV / Visible spectrophotometry allowed to quantify polyphenols in all extracts and fractions. Results are shown in Table 3.

Table 3:- Polyphenol content of *A.pyrethrum*'s extracts and fractions expressed in milligrams equivalents gallic acid per gram of extract (mg GAE/g of extract)

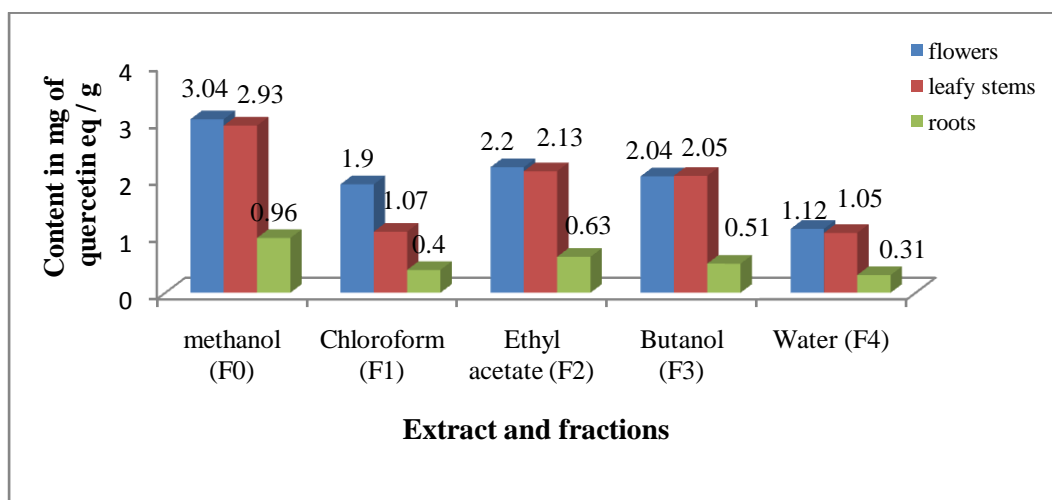
Extracts	Polyphenol content in mg GA eq /g of extract		
	Flowers	Leaves	Roots
hydro-methanol extract (F ₀)	21.84 ± 2.41	14.23 ± 1.44	7.83 ± 1.18
Chloroform fraction (F ₁)	13.49 ± 1.51	5.67 ± 0.48	4.94 ± 0.21
ethylacetate fraction (F ₂)	14.20 ± 1.40	10.23 ± 1.34	5.94 ± 1.21
butanol fraction (F ₃)	19.84 ± 1.81	12.15 ± 1.18	6.15 ± 1.70
residual fraction (F ₄)	16.12 ± 0.09	11.85 ± 1.09	5.88 ± 0.63

For all parts of the studied plant, we noticed the variability of total phenols contents (Table 3). The highest content was found in flowers. It varies between 13.49mg and 21.84mg of gallic acid equivalent per gram of extract followed by the leaves with a content ranging between 5.67 mg and 14.23mg GA eq / g of extract. Polyphenol content of roots varies between 4.94 mg and 7.83mgGA eq / g of extract.

We also found that methanol crude extracts contain relatively high concentrations of polyphenols compared to other fractions. Phenol content of a plant depends on a number of factors such as climatic conditions, time of harvest, solvent of extraction, storage conditions [24].

flavonoids content:-

Flavonoid content in hydro-methanol extracts obtained from *A.pyrethrum*'s organs are recorded in picture 2.



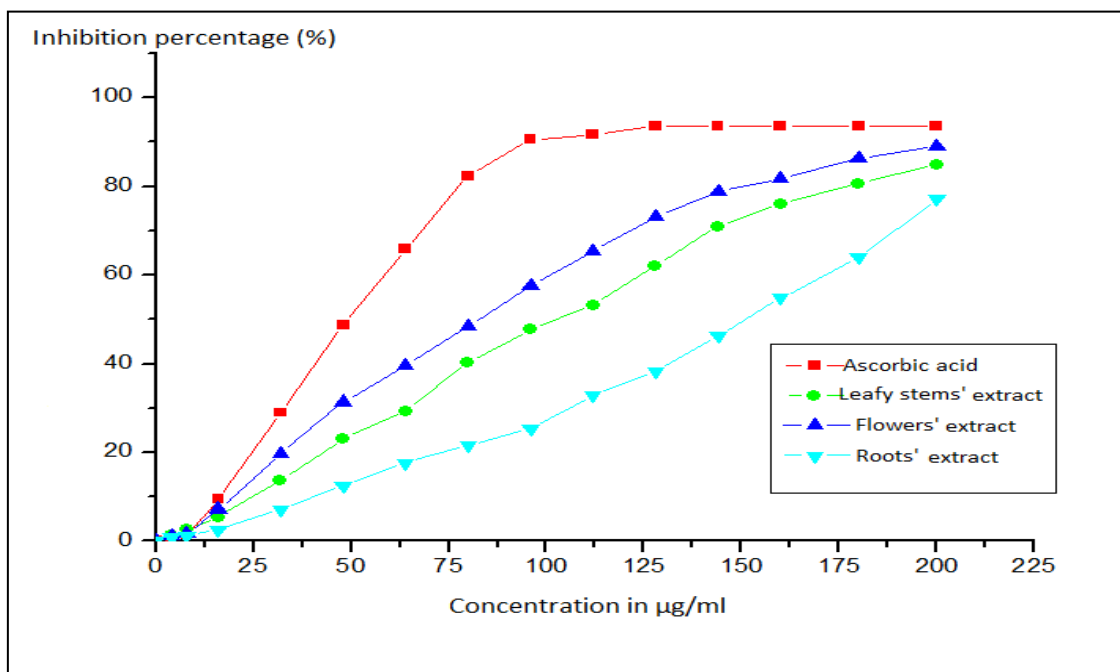
Picture 2:- Flavonoid contents in flowers', leaves' and roots' extracts and fractions of *Anacyclus pyrethrum*

Concerning flavonoid, the highest content is recorded in flowers' hydro-methanol extract with a value equal to 3.4 mg of quercetin eq / g of dried plant material followed by the leaves' hydro-methanol extract with 2.93 mg of quercetin eq / g and finally roots' hydro-methanol extract with 0.96 mg of quercetin eq / g dried plant material.

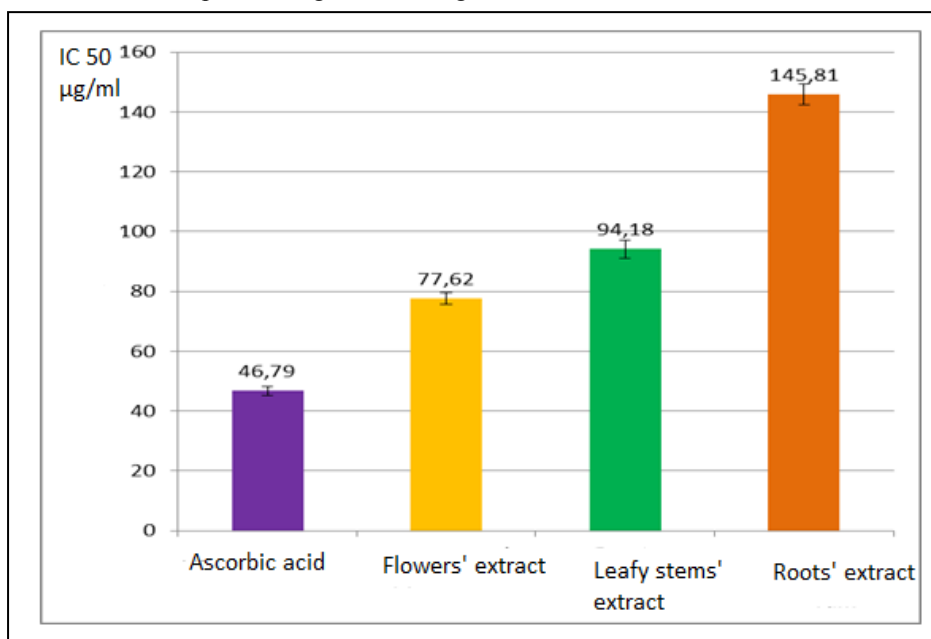
We can deduce that the crude extracts from flowers and leaves of *A. pyrethrum* are the richest in flavonoids.

Antioxidant activity:-

Values obtained from DPPH test allowed to establish curves on picture 3.



Picture3:- DPPH d'inhibitionpourcentages according to extracts concentrations



Picture4:- Half maximal inhibitoryconcentration (IC₅₀) of hydro-methanol extracts towards DPPH

The results obtained show that hydro-methanol crude extracts of *A. pyrethrum*'s organs have very important antioxidativeactivities, with IC₅₀ 77.62µg / ml for flowers, 94.18µg / ml for leaves and 145.81µg / ml for the roots. These levels are lower than that of ascorbic acid (IC₅₀ 46.79µg / ml) used as a positive control (Picture 4).

On the other hand, we note that there is a correlation between the concentration of polyphenols and antioxidant activity, confirming that polyphenols are powerful antioxidants that inhibit the formation of free radicals and oppose oxidation of macromolecules.

The antioxidant activity depends not only of the concentration of polyphenols, but also on the structure and nature of antioxidants in the extract [13]. Generally, polyphenols with high number of hydroxyl groups show higher antioxidant activity [25].

Conclusion:-

The phytochemical study of three organs of *Anacyclus pyrethrum* shows that the leaves and flowers are rich in tannins and flavonoids, while the roots are richer in alkaloids than both of the other parts of the plant.

Concerning the phenolic composition of the extracts, flowers and leaves showed high total polyphenol content in comparison with the roots.

This in vitro study of the antioxidant activity performed with the method of scavenging DPPH free radical showed that methanol crude extracts have a high capacity to trap the DPPH radical with 50% inhibitory concentrations (IC₅₀) about 77.62 µg / ml for flowers, 94.18 µg / ml for leaves and 145.81 µg / ml for roots. These extracts may therefore be an alternative to some synthetic additives.

It is therefore important to characterise and identify the bioactive molecules in all fractions of the three organs of *A. pyrethrum* by HPLC analysis.

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RESEARCH ARTICLE

Cone-beam computed tomography-A boon to dentistry.

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Abstract

Imaging is the most important and frequently used diagnostic tool in dentistry. Although the history and clinical examination are of prime importance when evaluating patients, the use and evolution of non-invasive technology from two-dimensional (2D) X-ray modalities to three-dimensional (3D) cone beam computed tomography (CBCT), for imaging is increasingly becoming popular. Interest in CBCT from all fields of dentistry is unprecedented because it has created a revolution in maxillofacial imaging, facilitating the transition of dental diagnosis from 2D to 3D images and expanding the role of imaging from diagnosis to image guidance of operative and surgical procedures by way of third-party applications software.

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Introduction:-

Since introduction in the late 1990s, cone beam CT (CBCT) has become a common modality to image the facial skeleton. CBCT has been described as the 3D method of choice for maxillofacial imaging due to the lower cost of the equipment and test, the reduction in radiation level in comparison with multi-slice computed tomography, high resolution for bones and teeth, and the possibility of obtaining the whole set of traditional orthodontic images in only one exposure.¹

The American Association of Oral and Maxillofacial Radiology has stated that cross-sectional views are recommended for planning dental implants, and this in combination with the easy accessibility, easy handling, and low-radiation dose of CBCT imaging will lead to the widespread use of CBCT imaging in implantology.² Oral implants require localized area of oral and maxillofacial area for radiation exposure; so, CBCT is an ideal choice. CBCT scans help in the planning of oral implants; they enable measurement of the distance between the alveolar crest and mandibular canal to avoid impingement of inferior alveolar nerve, avoid perforation of the mandibular posterior lingual undercut, and assess the density and quality of bone, and help in planning of the oral implant in the maxilla with special attention to the nasopalatine canal and maxillary sinus.³ If CBCT was equal to a full-mouth series in efficacy, it could be argued that, depending on machine type, the radiation risk would be considerably less, as would the time and effort it takes to image the patient.

Background:-

The imaging of CBCT is achieved by operating a rotatory gantry to which an imaging x-ray tube and image receptor are fixed. A deviating pyramidal or cone-shaped source of ionizing radiation is focussed through the center of the

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area of interest onto an area image receptor on the opposing side. The x-ray tube and image receptor rotate around a rotation fulcrum fixed within the center of the region of interest. During the rotation, multiple (150 - 600) sequential planar projection images of the field of view (FOV) are obtained in a complete or partial arc. This technique varies from a traditional medical CT machine, which uses a fan-shaped x-ray beam in a helical progression to acquire individual image slices of the FOV and then stacks the slices to acquire a 3D image. Each slice requires a distinct scan and separate 2D reconstruction. Since CBCT exposure incorporates the entire FOV, only one rotational sequence of the gantry is necessary to acquire enough data for image reconstruction.⁴

The four components of CBCT image production are (1) acquisition configuration, (2) image detection, (3) image reconstruction, and (4) image display. The dimensions of the FOV or scan volume able to be covered depend primarily on the detector size and shape, the beam projection geometry and the ability to collimate the beam. During the scan, single exposures are made at certain degree intervals, providing individual 2D projection images. The resolution, and therefore detail, of CBCT imaging is determined by the individual volume elements or voxels produced from the volumetric data set. In CBCT imaging, voxel dimensions primarily depend on the pixel size on the area detector, unlike those in conventional CT, which depend on slice thickness.⁴

Benefits of cone-beam CT in dentistry:-

CBCT equipment occupies less room space compared to conventional CT machine. Structural relationships of hard and soft tissues can be observed directly. The ability to rotate images and to add or subtract structural components permits relationships to be studied. Contiguous structures can be separated and normal hidden surfaces examined in detail. Accurate linear and volumetric measurements can be made. It eliminates superimposition of images of structures outside the area of interest. CBCT imaging produces images with sub millimetre isotropic voxel resolution, which achieve accurate level of spatial resolution for maxillofacial imaging. Images of the patient can be acquired in sitting, standing or supine position. CBCT volumetric data is isotropic, which means all three dimensions of the image voxels are the same. This makes it possible to reorient the images to fit the patient's anatomic features and perform real-time measurements.^{5,6}

Applications of CBCT in dentistry:-

Implant Dentistry:-

Conventional panoramic radiography is unable to image adequate bony support preoperatively. Obviously, having this information preoperatively greatly reduces the likelihood of the need to change the treatment approach intraoperatively. The reported indications for CBCT use in implant dentistry vary from preoperative analysis regarding specific anatomic considerations, site development using grafts, and computer-assisted treatment planning to postoperative evaluation focusing on complications due to damage of neurovascular structures. It will be difficult to prove a clear and statistically significant benefit of cross-sectional imaging (with special emphasis on CBCT) over conventional two-dimensional imaging such as panoramic radiography with respect to damage of the IAN or other vital neurovascular structures in the arches resulting in dysesthesia or pain in comparative prospective studies due to the high number of cases needed for such an evaluation.⁷

Orthognathic Surgery:-

A 3-dimensional model can be reliably adopted for orthodontic and orthognathic analysis and surgical prediction, extensive research is needed to characterize the landmarks and relationships that this technology allows us to measure. As useful as cephalometric analysis can be, its imaging accuracy is inadequate in such deformities as hemifacial microsomia, severe facial asymmetries, and occlusal cant. Volumetric analysis promises to offer better prediction in terms of the morphology of the defect, as well as the volume of graft material necessary for repair. Questions abound regarding the stability of the arch after grafting, the quality of the bone graft over time, and the effect on overall facial growth; CBCT provides a means to investigate these issues in depth. Only difficulty is how best to apply and manipulate that data for more accurate surgery and treatment planning.⁸

In 2009, Swennen et al. recommended the following three-stage sequence for imaging when evaluating surgical treatment outcomes using CBCT: In Stage 1 (3–6 weeks post-operatively): imaging is used to verify the transfer of bony parts. This time frame circumvents post-operative soft tissue swelling which might interfere in occlusion and is prior to bony consolidation, thereby providing proper visualization of osteotomy lines. In Stage 2 (6 months to 1 year post-operatively): imaging at this stage evaluates the soft tissue response and should preferably occur after the removal of orthodontic brackets. In Stage 3 (2 years or more post-operatively): this imaging is used to evaluate long-term changes in surgical treatment.⁹

Orthodontics:-

Orthodontists can use CBCT images in orthodontic assessment and cephalometric analysis. Today, CBCT is already the tool of choice in the assessment of facial growth, age, airway function and disturbances in tooth eruption.^{10,11}

While 3D CBCT images are most often used to assess skeletal contributions to malocclusion, researchers are now investigating the use of these images to assess dental relationships in orthodontic patients.¹² CBCT likely will provide information that could result in one or more of the following outcomes: (1) enhanced diagnosis, such as precisely localizing impacted and supernumerary teeth; (2) quantifying the magnitude of a defect or deformity, such as in patients with craniofacial anomalies; (3) improving differential diagnosis of skeletal, dental or combined malocclusions, including identifying the jaw(s) contributing to malocclusion and determining whether the discrepancy is bilateral or unilateral, such as in orthognathic surgery, asymmetry, craniofacial anomaly and open bite cases; and (4) helping to identify possible causes of malocclusions, such as the contribution of TMJ abnormalities to an open bite or asymmetry.¹³

The decision regarding the use of CBCT depends on the severity of malocclusion. The more severe the malocclusion, the more probability of needing the examination and similarly the milder the malocclusion, the less likelihood of needing a CBCT scans. Malocclusion severity is understood as the presence of vertical and sagittal skeletal discrepancies, facial asymmetry, craniofacial malformation and tooth eruptive disorders. There is no rationale in indicating CBCT for patients with Class I malocclusion and anterior crowding.^{14,15}

Airway morphology and obstructive sleep apnoea:-

Cone beam computed tomography can better assess the cross-sectional dimensions of the airway space than conventional 2D radiography. The drawing of airway circumferences and calculation of cross-sectional areas by computer also greatly reduces operator-dependent bias.¹⁶ Aboudara et al found much larger interindividual variations of the volume and area of the upper airway in cephalograms than with CT.¹⁷

Caries diagnosis:-

CBCT imaging appears to be the best prospect for improving the detection and depth assessment of caries in approximal and occlusal lesions. Recent work with benchtop-based local or limited CBCT (LCT) systems has demonstrated the potential for caries detection and depth characterization by high-resolution systems. Further studies are needed to evaluate CBCT for the detection of occlusal or pit and fissure caries, a task for which 2D imaging has been weak.^{18,19}

Periodontal applications:-

Mol et al observed that the CBCT images provided more accurate information on periodontal bone levels in three dimensions than the images of photostimulated phosphor plates. In a similar study, it was found that CBCT was better in morphological description of periodontal bone defects, while the images obtained by charged coupled device sensor provided more bone details.²⁰ Noujeim et al concluded that CBCT technique has better diagnostic accuracy than periapical films in the detection of interradicular periodontal bone defects.²¹

CBCT measurements were found to be equivalent to direct measurements and dehiscences were diagnosed with higher accuracy than fenestrations. CBCT images were used for alveolar bone width measurements. Intra-surgical furcation involvement measurements were compared by using CBCT images and it was reported that CBCT images demonstrated a high accuracy in assessing the loss of periodontal tissue and classifying the degree of furcation involvement in maxillary molars.²²

Endodontic applications:-

CBCT for endodontic purposes appears to be the most promising use of CBCT, in many instances instead of 2D images. Endodontic applications include the diagnosis of periapical lesions due to pulpal inflammation, visualization of canals, elucidation of internal and external resorption, and detection of root fractures. CBCT was found to be useful in differentiating solid from fluid-filled lesions (periapical granulomas from cysts) using grayscale values in the lesions.²³

For most endodontic applications, limited FOV CBCT is preferred to medium or large FOV CBCT because there is less radiation dose to the patient, higher spatial resolution and shorter volumes to be interpreted. Limited FOV CBCT should be considered the imaging modality of choice for initial treatment of teeth with the potential for extra canals

and suspected complex morphology, such as mandibular anterior teeth, and maxillary and mandibular premolars and molars, and dental anomalies. If a preoperative CBCT has not been taken, limited FOV CBCT should be considered as the imaging modality of choice for intra-appointment identification and localization of calcified canals. Limited FOV CBCT should be considered the imaging modality of choice if clinical examination and 2-D intraoral radiography are inconclusive in the detection of vertical root fracture. Limited FOV CBCT should be the imaging modality of choice when evaluating the nonhealing of previous endodontic treatment to help determine the need for further treatment, such as nonsurgical, surgical or extraction and for nonsurgical retreatment to assess endodontic treatment complications, such as overextended root canal obturation material, separated endodontic instruments, and localization of perforations.²⁴

Temporomandibular joint diagnostics:-

CBCT in general has an acceptable accuracy for diagnosing osseous TMJ abnormalities with fairly high sensitivity, although small abnormalities might be missed. CBCT most accurately depicted erosive changes of the bone cortex of the mandibular condyle.²⁵ The high detectability of CBCT images on bone morphology of mandibular condyles was observed. CBCT has also been found useful as an image-guided technique for safe puncturing of the superior TMJ space. CBCT has been applied in the evaluation of bifid mandibular condyles, coronoid hyperplasia and articular eminence morphology and to assess condylar remodelling accompanying splint therapy.²⁶ CBCT was applied in a series of cases of osteochondroma of the mandibular condyle and the value of 3D reconstructions when viewing expansive lesions was emphasized. CBCT has also been applied to cases with facial asymmetry and condylar hyperplasia, synovial chondromatosis, fibrous ankylosis in rheumatoid arthritis, metastasis of a bronchial carcinoma and juvenile idiopathic arthritis.^{26, 27, 28}

Oral and Maxillofacial Pathology:-

CBCT is useful in evaluating the intricate anatomy of many unusual presentations, measuring their exact dimensions and their association with surrounding structures. CBCT scan is the imaging method of choice in pre and postoperative diagnosis assessment of inflammatory odontogenic cysts together with alloplastic graft materials repairment of the osseous defects and dental rehabilitation by metallic implants, due to high specific abilities in bone tissue 3D evaluation. Three-dimensional imaging of cysts and tumors of the maxillofacial region can give the surgeon the vital information necessary for planning surgery; with volumetric analysis, this can help anticipate the need for and volume of a potential graft for reconstruction.^{8,29} Various effects of cysts and tumours on the anatomic structures can be studied using CBCT.³⁰

Oral and maxillofacial surgery:-

CBCT is the technique of choice in mid-face fracture cases, orbital fracture assessment and management and for inter-operative visualisation of the facial bones after fracture. Since it is not a magnetic resonance technique, it is the best option for intra-operative navigation during procedures, including gun-shot wounds. CT is the modality of choice for the evaluation of complex facial fractures, especially those involving the frontal sinus, nasoethmoidal region, and the orbits. CBCT acquires all projection images in a single rotation, therefore scan time is reduced enormously (20 seconds or less), often performed with the patient in a seated position. Because the data are acquired volumetrically, voxel size is isotropic, equal in all 3 dimensions of the voxel, whereas conventional CT voxels are anisotropic, where the height of the voxel (Z-axis) is determined by slice pitch, a function of gantry motion.³¹

Prosthodontics:-

Other than implant planning, CBCT has its role in prosthodontics as planning craniofacial defect reconstruction. Three-dimensional augmented virtual models of the patient's face, bony structures, and dentition can be created out of CBCT DICOM data by software volume rendering for treatment planning. The process of initial assessment to a follow-up during a 4 years review would be precise with the use of a CBCT, thereby improving the prognosis of over dentures.³²

Forensic dentistry:-

CBCT technology offers 3D visualization and more complex and more accurate imaging compared to analog and digital radiographs. Increasing availability of this technology provides the forensic odontologist with an imaging modality capable of providing a 3D representation of the teeth and the jaws whose benefits may be extended in several ways to the science of forensic odontology. Rai et al investigated age estimation based on the pulp-to-tooth area ratio (PTR) of maxillary canines measured in three planes (axial, sagittal, and coronal) obtained from CBCT image data.³³

Limitations of CBCT:-

The spatial resolution and the contrast resolution of CBCT is lower than that of conventional film-based or digital intraoral radiography. Radiographic artifacts are another problem in CBCT imaging. The two types of artifact that can reduce the diagnostic yield of the images are distortion of metallic structures, called “cupping artifact” and the appearance of streaks and dark bands between two dense structures, so that these artifacts may reduce the diagnostic yield of images. The patient has to stay absolutely still as his/her movement can adversely affect the sharpness of the final image during the scan.³⁴

Effective dose of CBCT:-

Heterogeneity in measurement methods and scanning protocols between studies made comparisons of effective doses of different CBCT units and scanning protocols difficult. Few studies related doses to image quality. Reported effective dose varied across studies, ranging between 9.7 and 197.0 mSv for field of views (FOVs) with height #5 cm, between 3.9 and 674.0 mSv for FOVs of heights 5.1–10.0 cm and between 8.8 and 1073.0 mSv for FOVs .10 cm.³⁵

Conclusion:-

In summary, with the continued decreasing cost of CBCT technology, it is only a matter of time until CBCT finds its way into the average dental practice. It is clear that the utility of the CBCT cannot be disputed. CBCT is capable of providing accurate, submillimeter-resolution images in formats allowing 3D visualization of the complexity of the maxillofacial region.

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RESEARCH ARTICLE

The evolution of GDP growth rate in the economy of Libya.

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Abstract

This study is purposing to studying the development of the growth rates in Libya's economy, as this study is purposing to know the real reasons of the decreasing or wiggling in the rate of the economic growth, beside to know the development that's happened in the main growth rates in the economic sector of Libya's economy and the change that happened to it during the period (1970-2015).

The economic growth is a process in which increased the real income an increase that cumulative and persistent during a period of time, where the investment according as most important factor in the investment process and the growth status materializewith a whole process of making capital, the process of expand the rate of the local production growth and that's by using the available local source, lie in taken two kind of decisions.

The first ones: Attaching in which sectors we should investing with.

The second ones: Blend of factors which must be using to product goods and services which rule the production technology, where the result pointed that the investment is the most important factor in the growth process and the status of growth materialize with the whole process of making capital, as the result showed that the Libya's economy had notice a rising with the growth rates in the product sectors during the study period.

It's arranged on it the speed of the living level improving, and that's considered a positive indicator which had service the path of the economic growth in Libya.

The Key Words: The Evolution of the Economy / The Rate of the Growth / The Libyan Economy.

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Introduction:-

The economic and socially development is the most important purpose which all the countries are seeking to achieve it in our contemporary world, whether the development ones or the ones that walking in the growth way, as after the second world war had finished in the first of the forties of this century, the birth of about two-thirds of the world which are known now as the developing countries, where it's came into being, carrying with it a lot of economic, social, politic, cultural, and administrative problems, as a result to the tardiness that it had suffering of for a long centuries, and as a result of the hard life condition that arrange on other different factors like colonization with its two kind the old and modern, so the development had becomes presenting one of the main cases which occupies a

special place on the two levels, the global and the national, and one of these signs is, the large numbers of the researches, studies, and articles which spreading in several parts of the world, and that's in purpose of understand the problem of the development countries, and face it with an accurate scientific facing, where the development in our time depended on a set of components or elements, that must have provided to start with march of growth and progress in any community of human societies, and these components or elements, are an interlocking circles that completed each others, where the absence of one or some of it affecting in the growth process in these communities, where these components are usually presented of, the political system, economy system, a natural sources, investment, accumulation of capital, manufacturing, human sources, and the efficient flexible management of that able to make maximum benefit of these elements, as it showing during several experiment that passed it the underdevelopment countries, with preparation and implementation the economic and socially development plan, the economic and technical feasibility study of plans and programs development, are no longer in itself enough to rule on the possibility of implementation these plans, without taking in consideration another important factor which is the administrative feasibility study, which means the range of possibility or ability of administrative device that based on undertaken to such vital task before thinking of doing these plans, and that doesn't usually done unless provided an effective device for planning, take task of coordination and connection between planning and implementation process, and that's now are not available for most of the development countries, and that's consider to the absence of the active manager that able to find system of administrative planning of policies, programs and helpful sources to selecting the aims of the plan and how to apply it, where the economic and socially development planning with the absence of the active devices to collect the information, and a new system of information and active units to planning and budget, are forming a heavy burden to any country that seeks to achieve the development, as the plan that doesn't building on a specific information, will not achieve its goals, as the absence of the capable central and sub devices will make the plan is building on policies, objectives, and programs that so far from the necessary scientific studies to prepare the sound plan of development.

The final purpose of the development is to achieving the fit growth rate in each productivity and service sectors in balance way, and that's what wasn't achieved it in the past during the experiments and attempts which had done in this field, as a result of a combination of factors that contributed directly in the weakness of the economic basic in Libya, and occurrence to economic and social development were unbalanced, and a changing in administrative structure had a negatively affected in the administrative system efficiency, of these factors the unbalance change which had happened in the socially building of community, by the fast social change that's unplanned, as a result of administrative and developmental experience that relatively short, compared to other of the advanced industrial countries, and as a result of the flowing of cash capital after the showing of the oil in the middle of ninety, and what had come with it of the expenses in making a lot of main projects in the same time, the rarity of the work element especially the technical competencies, the economic, socially and cultural, incapacity on absorb the modern technology, the instability of economic policies and its stability in a particular style, the weakness of the level of the education and training, as well as the weakness of the system of material and moral incentives that motivation to work and good performance.

The Problem of the Study:-

Libya according as one of the development countries, which passing today on a transformative stage of the traditional possession to redevelop possession, beside of the transformative stage to the socialism and what came with it of the application and its obstacles problems, like the low or weakness with the rate of economic growth and achieving a low rates which lead the country to put many of plans and programs which purposing to increase the economic growth rate, and that's by:

- The controlling on inflation rates at low levels.
- Working on achieving a real positive interest prices.
- Limiting of the deterioration of the market prices in the local coin.
- Achieving a high economic growth rates.

Though Libya had passed through the development plans and programs in achieving a great deal of the economic stability, but achieving an economic growth rate was not in the required level, which means there is another factors stand in front of achieving an economic growth rate that desirable, where the reasons of that go back to three factors which are:

- The inactivity of the investment policies that been applied to achieving that much of the accumulation of capital which achieving the desirable economic growth rate.

- Disruption the relation between the available savings and required investment.
- The existence of two elements together.

From up the summary of the problem that the Libyan economic had suffering of presented to the stability of the real economic growth rates in the levels (acceptable) to insure of high growth rates during applying a comprehensive and sustainable development plan.

The limits and the scopes of the study:-

The study purposes to know the development that happened in the rates of the main growth in the production sectors of economy in Libya and what happened in it of changing during the period (1970-2015).

The Aim of the Study:-

The study aims to:

- Studying the growth rates in Libya's economy.
- Knowing the real reasons of the decreasing or the wiggle in the economic growth rates in Libya.
- Studying the requirements and the possible ways to decreasing a high growth rates in Libya's economy in the future.

The Methodology of the Study:-

The method that been used in the study on using analytical method with its two parts, descriptive and quantitativeto suit the nature of the study subject especially the specific indicators of growth rates during the study period.

The Important of the Study:-

The important of the study came from the few studies in the field of the economic growth rates, where the economic growth according as the most prominent targets of any economic program or economic plan, beside to what it presenting of facts and analytical observations that helping in study the privacy of the subject of the economic growth and its important in any analytical economic study.

1-The Accumulation of capital

The accumulation of capital had magnified during the net investment process (**Rostow, 1960**), which present the different between net country incomes (the social income minus the extinction) and between what it consumed of that income during a specific accounting period. The accumulation of capital has expanding the ability of the country of production goods.

The term of capital is broader than the term of traditional capital goods, in other words, it's the goods that doesn't giving a real time benefit but producing a goods that achieving these benefit, and if the capital had definition of the exists that creating an extra channel in the future to the income that measurable in the society, as many goods and services as has been customary a consumer goods that must classified specifically as a part of the accumulation of capital to country, as an example the expenses on education which is continually reinforcing the mind ability of individuals beside that it is giving a direct satisfaction , these expenses must partly account as investment expenses, and the same thing for some consumer goods like clothes, the consumer chive goods and similar, considered necessary to stimulate the agricultural product to increase the production and it's necessary to classify as an accumulation capital(**A.B. Therol, translated by Dr. Kasem Al-Dujaili 1998, P219**).

So that the only way to build the productivity energies of country and rising the average of individual income, is with distribution the productivity energies of goods and there is no need to material product of capital but to product a different kinds of capital like consumer goods (Catalyzed) and expansion the investment field in human capital and all of that are contributing in the production increase and rise the living levels. The development process is connected with manufacturing and the manufacturing connected with making capital, and a large number of the economist also seeing that the investment is the most important factor in the development process, the professor Arthur Lewis(**A. Louis, (Allen and Unwin 1955)**) had described the development process as a process that leading to transformation the economic from saving and investment 5% to saving and investment 12%. As the professor Johnson(**J.J. Johnson, 1969, P9**)had also seeing the formation of capital with its expansion conception that is a special character for development process. The state of the growth shapes in a comprehensive process of the capital formation, the mechanical, economical, and social stay, to sustain and increase the accumulation of capital to the per capita in its different shapes, conversely the state of the retardation connected with possess a relatively littleaccumulation of the different shapes to capital.

1.1. The Capital and the Technological Progress

The rate of growth of the required capital to absorption of new technology in countries which are characterized by delay in the technological level is being bigger than it is in the development countries. The countries that delay technology are limited with producing technology, and in the economics of the extra work and low factor to the capital\the work, as the deepening of capital which means giving each worker more capital to work with it may create a big difference in the production level, bigger than countries that have deepened capital continues for a long time.

The development in the product technical which is considered as the most clear explanation of the technological advances is the outcome of the group operations of researches and innovation, as it's an activity creating a new knowledge, but the development and evolution processes are processes that apply the new knowledge in production activities and all these processes are economical processes, as the relative importance of activities to the different factors which contribute in the progress process and the speeding of the progress itself had changing from country to country according to the development stage which is passing in it, beside that the number of the springs of technological advances are not recouping ones to the other [Dr- Qasim Al-Dujaili, 1998, P (226-227)].

1.1.1. Scarcity of Capital

Of the main properties which are characterized by the economics of development countries are the scarcity of capital which is the inability of capital, the bad using of it, and the inadequate what it consists to finance the necessary investments to promote with its economic potential and the implementation of its development plans, that's because of the capital is of the important direct growth factors, when this factor that synergizes with the human natural sources, so it will give a direct productivity.

The relative scarcity takes of capital different shapes in all sectors of the economic activity in such of countries, and the dominant phenomenon is the scarcity of the capital sources with its kinds like industrial buildings and the modern machinery and equipment with high production levels, and also the social capital like schools, hospitals, paved roads, Bridges, sewers, tunnels, railways, road transport and maritime and air, and the means of irrigation, sanitation and other (Mansoor. Dr-Ali Hafez, P32).

The scarcity of capital, is a phenomenon that has a strong relation of showing capital and demand of it and misguided of capital formation in economy, as the show of capital is weak and that's because the weakness of saving because of the decreasing of the real income level as a result of decreasing the production level which back to the decrease of capital, and the demand of capital as well is weak and that's incentive weakness on investment because of the weakness of the purchasing power as a result of the decreasing of the real income level which back to the decrease of the production level and that's because of the decrease of using capital in production.

And that's how the empty circles in the show and demand sides of capital in the economics of the development countries (Caftan. Dr-Mohammed Fazel Mohammed Aziz Caftan, 1978, P46) and it is possible to say that the scarcity of the capitals assets in most of the development countries are a reality that can't be denial and may cause in the continuing underdevelopment.

1.2. Resource Allocation and Economic Reconstruction

1.2.1. The Resource Allocation

Proceeding from the fact that the scarcity of resources in the development countries and its relation with needing the development process, the central issue in the development economics are resource allocation on the different needs which compete ones each other on these source, that's in most of the development countries there are two main limited on the growth of product process, and they are: the ability of the investment and the ability of import.

The most theories that resource allocation and most of the investment standards reflex this truth, so that, the start point that is known in the resource allocation process are how to maximize the rate of product growth by using what's available of the local resources, and how to minimize of this process needs of foreign currency.

Regardless of the decisions that are connected with the size of the required investment, it can be recognizing two kinds of specialized decisions of resource: [A. Luis, (Allen and Unwin 1955)]

- The first one is linked to which sectors that we should investing.
- The second one is combination of factors which must be using to production goods and services which control of production technology (Murad. Dr-Mohammed Helmi 1968, P 123).

In the same time which seems that these two decisions are separated ones on each other but in reality they are not as of the scientific side we find that the overlap between the product decisions and the technologic decisions are a thing that can avoid it.

As the deal of customize resources between the projects can't be separated from considerations of broader economic and policy subjects which is industry versus Agriculture, the balance growth versus unbalance growth, the outside trade strategy ...etc, and the effect of all these decisions will be through these next targets development strategy, as if the target is maximize the current level of well-being or maximize the growth and the output in some point of time in the future.

1.2.2. The Economic Rebuilding

The study of world experiments which engaged to its economic building for period of after war can take advantage of it with rebuilding our national economic in face of the different of condition, the stage natural, and the economic and socially development stage between Libya and those countries, but it faced a problems with one kind of natural and engaged to it almost with similar ways with different of the kind of methods that had take it to get this target.

With the different of the nature of the economic and socially systems of these countries and to rebuilding purposes, it requires concentration the economic resource, the ways of planning, and the central Administration in the hands of the state, as in British after war had establishment of the Ministry it mission to providing advice and recommendations to rebuilding and by that we seen that there is a necessary to make such a Ministry in Libya as to finish it mission after removing the reason of creating it(Naser. Salah, 1965, P161).

From other side the national economic rebuilding after war required providing large capitals, and the countries had different with providing these resources as the Soviet Union as an example had depending mainly basic on the self possibilities, while British beside to its self possibilities had depending on the outside helps, and as it comes to Libya, the provision of material resources is not a hard thing to do as Libya owns a huge reserves of oil, as the economic rebuilding required makes a critical reform for period of after war and required intensifying of capital element and increasing the production by inter improvement in the management and planning.

2- The Definition of Economic Growth

It's a process that increasing the real income an continues and cumulative increasing during a period of time extended to (quarter of century) where these increasing being bigger of the growth rate of peoples with providing a production and social services, Protect renewable resources from pollution, and preservation of non-renewable resources depletion(<http://ar.wikipedid.org>).

There is a different definitions of economic development, and of these commons definitions that the process including achieving a high growth rate of averagereal per capita income during a period that expanded from time (three decades) that is not accompanied by this deterioration in income distribution or an increasing in the poor level in society.

As it also known as the increasing of the number of goods and services which a specific economic producing it, and these goods had been production by using the mainly product elements which are earth, work, capital, and organization.

Since the industrial revolutionhas done then the spreads of it and what came of it of expansion changes in the industrial structure and achieving a big economic and social growth, then these economic and social progress is what called of the economic growth¹

So that the economic growth is means in general, the increasing the income for particular State had been majoring by using the ratio of the gross domestic product growth, and it's been comparing the ratio in particular year with the previous one.

¹ The English dictionary for example between the two words (Development, Growth) both are synonyms, one of them may be using in a place of other, as they're two carry the meaning of change which is connecting of improvement, so that the meaning of development and growth that's been mention in the research are the most popular meaning in the economic writes.

The increase in capital, the technologic progress, and improving the level of education, considered as the mainly reason of economic growth, as the past years had showing the idea of the sustainable development another factors like the main environmental factor of economic growth in realist by exclusion the rate of the expansion and not count it when the goods and services had been accounted.

2.1. The Theories of the Economic Growth

When we speaking on the economic growth. We must go back to some of the theories of economic growth and the opinions which the first economists had put it or came with it which the books of economic had included it, and brevity we mention of it:

- Adam Smith the first of traditional economist who contributed with explain the growth foundation in the free capital economics, and his theory had specialized with interaction trait in front of the steady progress where Adam Smith had built his thought on the principle of dividing work and connecting it degree with the size of market and how it expands, make the income a main condition of the economic growth and insuring of its important to collecting the capital to considered as a main element in the theories of economic growth.
- As Tomas Robert Malts said that the human well-been and his happiness are stopping on first of all the increasing of resource in ratio that equivalent to the increasing of people at less, and Malts had built his theory in peoples based on two principle:
 - The first ones: the food is necessary for human life and necessary to keep him alive.
 - The second ones: The instinct of controlling of each sex to the other, and Malts summarizes his theories to see the people increasing on a form ergonomically successive where the resources doesn't increasing unless on a form calculations successive.
- As the theory of Dived Ricardo was the more inclined to pessimism as he moved from the thought of increasing expensive which control on the thought of Adam Smith to the thought of decreasing expensive his situation in that is the same with the traditional economic which had following Adam Smith and founded the theory of the economic growth which characterized by pessimistic to the future, that the economic growth is achieving as a result of the people growth and what lead to it of increasing in the output nationalist.
- As Walt Whitman Rostow had focused in his study about the idea of stage as most notably as a standalone unit [Caftan, Mohammed Fazel Mohammed Aziz, 1984, P(175-212)], and it based on that the national economy walk on hard way cross stage after stage surpassing the difficulties and hardships until to the high degree of the economic growth to insure that the modern style of development, including five stages that should be done, started with the consumer society, to reach the starting stage which is the crucial stage.
- As Marx had building his analysis of economic growth in the light of capital on the theory of extra value as he start with divided the total national result in to three elements which are: the stable capital, the changing capital, and extra value which mean by it profit, rent and interest that the capitalist had it by unlawfully, and the Marxist interpretation of economic growth mainly on the physical interpretation of history and as it is the important factor and the final with make the history and it development is economic factor by its nature.

2.2. The Analyses of Growth and Income Distribution

There are many ways that consists of identical to expression about the growth in the income or the result of any country as an example [A.B troll \ translated by Dr-Qasim Al-Dujaili, P(123-124)].

The growth can be expression about the Calculated by dividing the investment coefficient to the gross (0\1) and the investment production is (1\ 5) which is:

$$\begin{array}{ccccccc}
 & \text{Increase in production} & & \text{investment} & & \text{increase in production} & \\
 \text{Growth} = & \frac{\quad}{\text{Gross}} & = & \frac{\quad}{\text{gross}} & * & \frac{\quad}{\text{investment}} & \\
 & \text{O } \Delta & 1 & \Delta \text{O} & & & \\
 \frac{\quad}{1} & * & \frac{\quad}{0} & = & \frac{\quad}{\quad} & = \text{growth} &
 \end{array}$$

And by definition that the slow growth either a decreasing of investment factor, or decreasing of production of capital, or both of them. And this equation is the mainly of the theory that said, the rate of the growth is faster in the development countries demanded a more resource for investment, but itself doesn't formed a development theory.

As an alternative we find that the output income can be expressed as a foregone conclusion of the total workstrength (L) divided on the output of unit work (L/O). and of that we can expressing about the output growth by its foregone of collection growth rate in unit work ($L/\Delta L$) and the growth rate in the output of unit work or what can expressing by productivity of work which is : $(L/O) \setminus [(L/O)] \Delta$

$$\text{Growth} = \frac{\frac{\Delta}{\text{Increase in production}}}{\frac{\Delta}{\text{Increase of workstrength}}} = \frac{\frac{\Delta}{\text{Increase in production}}}{\frac{\Delta}{\text{work strength}}} * \frac{\Delta}{\text{work output}}$$

And by this formula, the slow growth attributed by definition either to the slowly of growth rate, or delay of growth rate in work production, or both of them. And the expressing about growth on this way highlights on adoption the income growth of work unit (and the most important is growth of per capita income) on the growth of work production.

The main thought behind production function to measure the growth rate is provide a better understanding of growth resource and expressing by quantitative form about the size of the contribute which perform by all source when the growth rate had been calculated.

This way had been used in extensive form in the development countries and it had a lot of benefit, where some development countries has starting in our time to use this way as when some of these countries don't find it useful in extensive form because of the resource of growth in these countries had institutional in nature or not able to major.

2.3. The Mainly Indicators of Economic Growth in Libya

2.3.1. The Growth Indicator of the Period (1962-1969)

The traditional social and economic structure that mainstream in Libya's economy had changed radically change, where it enabled the successive cash flows as a result of oil exports to get rid of the deficit, where it directed attention to development that founded by oil income, and work on appear many economic development in start of it the rising of the growth rate, the increasing of the public spending, the level of the real income, improving the public level to the prices, increasing the imports, and creating a work opportunities.

As where the growth rates pointed on the available data but these period had seen a high growth rates which described by it's of higher growth rates in the world where the local output had achieved a yearly growth as much as (22.6%), and as a result of that the real average income to the individual had rising in realty of (16%) yearly, as the gross fixed investment had achieved a yearly growth rate as much as (15.6 %).

As on the economic sectors level, the data pointed that the oil sector had achieved a yearly growth as much as (46.7%) then the building and construction sector with average (19.9%), then the transportation sector with average (16.6%), then the services sector with average of (13.4%) finally the trade sector with average (12.7%).

The oil had achieved an increasing income for country what make it able to create an accumulation process in capital which consider as an important element of the economic development elements, where the available data in this regard pointing that since (1963) the oil revenues start to form an increasing rate of public revenues, where the rate of oil revenues had rising in the gross revenues from (26%) during the fiscal year (1962-1963) to (80%) in (1968-1969), in face of it the public spending with rate (1.7%) during the period (1955-1962) with rate (3.17%) during the period (1963-1968), where came of it most of these increasing about planning development program which targeted ensure rapid improvement in the standard of living and reducing migration from rural areas to cities and that's during increasing the important of the agricultural sector on the ground that the main supplier of foodstuffs and a source of income for too much community sons, beside to the increasing depended on the public sector with provide public services like education, health, transportation, and housing, where the State had depended a transformative economic and socially plan (plan 1963-1968) targeted through it provide development requirements, achieving the economic stability, and improve the living level for individuals, where it specialize for it huge amounts of money reached (450) million dinar a big part of it had specialized (43%) for spending on the

agriculture sector and service sector, while the rest has specialize (57%) for investment in the oil sector, where the percentage contribution of oil has rise in the economic active from (6.9%) in the year (1958) to (53.9%) in the year (1964) then to (62%) in the year (1968).

And the table number (1) the rate of real yearly growth of Libya economic during the period (1962-2009).

Table number (1), the rate of the real yearly growth of Libya economic during the period (1962-2009)

Item 1	1962-1969	1970-1982	1983-1990	1962-2009
Local production	22.6	13.2	2.5	11.8
Agriculture sector	4.1	4.6	11.8	6.7
Oil and Minerals	46.7	14.3	7.8	17.8
Manufacturing	8.4	18.6	7.0	13.0
Construction	19.9	15.3	1.8	12.0
transportation	16.6	18.1	0.4	13.1
Wholesale and retail trade	12.7	17.0	2.1	10.9
Services sector	13.4	14.0	2.7	11.1

The increasing of the contribution of non-oil sectors (Agriculture and Industry) in the gross domestic product, where it depended on the three development plans, the three-year plan (1973-1975), the five-year plan (1976-1980) and the second five-year plan (1981-1986) the development strategy that target achieving the most rate of total growth, with the different in the production activity, but it's with all these efforts, the contribution of sectors in the local product had stay very modest, which conform on continued the dominance of oil on the Libya's economic.

2.3.2. The Indicators of the economic growth for period (2010-2015)

During the past period Libya had notice a long war (Chad War – American Aggression) and live a tough economic siege, as its development journey had go back effected by the condition and economic policies which had approach far a way of the principle of developmental differentiation supporting to battleship efforts and the Sustainability of counter the siege which limit of the positive development effects which achieved during the period of seventies support of the negative of the next period as the result was staying the Libya's economic goes on in a vicious circle.

Where the gross domestic product with underway prices in the Libya's economic had achieving a growth rate as much as (16.9%) during the year (2010) and that as a result of the refresh of Libya's economic in the wake of the global economic crisis that affected on it perform during the year (2009) where the rise of the pace of growth of the world economy had contributed with the rising of oil prices in the national market and increasing the revenues and exports, and each of these factor had a positive effect on the value of the output and the rate of its growth, but in the year (2011) the Libya's economic had noticed a go back in its perform and that's as a result of the politic development events where the local output had recorded a negative rate of growth as much as (53.1%) comparison with the year (2010) but in the year of (2012) the record oil prices during the year of (2012) had supporting the economic growth in Libya, where Libya achieved the highest rate of growth at current market prices during the year (2012) it reached about (126.8%) and that's as a result of covering the economic from the consequences of political circumstances and the inside development which Libya had notice it during the year (2011), and in the year of (2013) we find the rate on the real gross domestic product in Libya had notice big swings in the last time affected by the number of the outside and inside factors and on its head the fiscal crisis and also the economic fallout that result of the politic transformative which Libya had notice it, the thing leads to shrink the output in Libya with average (22.5%), where the growth economic data had shows for the period (2010-2015) that the gross domestic product with the ongoing and study prices as in the table number (2)

Table number (2) the development of the gross domestic product:

the year	The domestic product with the ongoing prices	The growth rate
2010	73,965,000	16.5%
2011	34,699,000	-53.1%
2012	78,697,332	126.8%
2013	60,990,432	-22.5%
2014	28,543,523	-53.2%

From the table number (2) that the gross domestic product had start to decrease in the year of (2011) comparison with the year of (2010) and with decreasing rate as much as (53%) when in the year of (2012) we find that the gross domestic product had grown with rate (126.8%) and that's as a result of the rising in the oil prices.

As the contribution of economic sectors with generate the gross domestic product had affected with the political and economic events which Libya has pass through it after the year of (2010) where the rate of contribution of crude oil and mining sector had increasing from (6.9%) in (1958) to (53.9%) in (1964) then to (62%) in (1968) when the rate of contribution of other sectors had stay modest.

2.3.3. The Population Growth Rates

As to the size of the population and the rates of its growth, the growth population in Libya and with it absolute image had notice a quick, continuous and regular development.

Where the absolutely number of the population increases didn't affecting with the advance and retreat that the Libyan's economic had notice trough the time successive stages, and that's what assured it the result of the residential census that's had done in Libya from the first census until the last one, where the statistical data had point to the clearly increases of the residential census in Libya as it showing in the table number (3).

Table number (3) the numbers of the population in Libya during the period of 2000-2015

The year	Number of population (million population)
2000	5.640.000
2001	5.842.000
2002	6.021.000
2003	6.221.000
2004	6.420.000
2005	6.629.000
2006	6.843.000
2007	7.065.000
2008	7.294.000
2009	7.530.000
2010	7.774.000
2011	6.288.652
2012	6.283.403
2013	6.265.987

This population increase is a normal result and that's because of the rising in the rate of population growth which maintained a high pace (1.56%) during the seventies decade and a rate of (2.52%) during the eighties decade of the past century, but the pace of this rate had take to decreasing after the year of (2010) with rate (1.56%) during the period (1995-2006), as the data are also showing that yearly net growth rate of the Libyan population that been achieved in the period of (1973-1984) that amount to (4.21%) had decreasing to (2.80%) during the period of (1984-1995) then to (1.8%) during the period of (1995-2006) which insure that the gross of the population number had grown with a yearly rate as much as (2.94%) during the period of (1973-2006), as the available statistics point that the Libyan population that living in Libya are constitutes a rate (91%) of the gross population number according to the results of the population census in (1973) where this rate had decrease (89%) of the gross population number.

2.3.4. The Use Index

The available data pointed that the size of the power worker in Libya had reached 433500 thousand worker in the different economic activity, where the patriots number of them had reached 383500 thousand worker which is almost (88.5%) of the gross use index, when the power worker which are not patriots among 50000 worker which is almost (11.5%).

As on the sector level, the available data pointed to the decreasing the rate of the contribution of the agriculture sector to (22.659%) faced to the increasing of the rate of contribution of the manufacturing industry to (3.7%).

But the next period (1980-1990) had characterized by volatile remarkable on the partly level, where the total of use since the beginning of 1980 had reached 780223 thousand worker, (72.9%) of them are national employment as the

rest rate had formed (27.1%) of the foreignworker, as the available data pointed to the increase of the workers rate in the industry manufacturing sector to (4.6%) when the number of the worker in the agricultural sector had decreasing in to (15.5%), as this wiggle in the size of the power worker had continue during the period (1990-2007) where the gross use had reached 1780710 worker during the year (2007), where the national direct power had formed a rate of (90.6%), as the rest rate is (9.4%) of the foreign workers.

2.3.5. The indictors of the growth of the gross domestic product and Composition of fixed capital and Investment Spending

The Libyan oil exports had formed since (1961) an inflection point in the economic history of Libya, where it's foundation a new stage known of the capital surplus stage and quick growth, and with the start of (1962) a substantial changes had happened on the structure of the Libyan economic come with achieving a high rates of the growth and also in the rise of the part of the individual of the gross beside to create kind of the relatively sophisticated activities, and we can show these indictors according to these stages:

- **The first stage (1962-1969)**

This stage describing as one of the faster periods of the growth in the history of Libya, where substantial changes had happened on the structure of Libyan economic comes with achieving high rates of growth and also a rise of the per capita of the gross domestic product.

- **The second stage (1970-1981)**

This stage described as one of the important periods of the capital accumulation that output by the first oil price shock (1973-1974), where this period had noticed an economic vogue present of increasing the growth rates where the real gross domestic product had achieving a growth amount an average (13%) yearly, where this increase come with the rising in the average of the per capita of the gross domestic product and a decrease in the public level of the price, where the inflation rate had decreasing with rate (7.5%) in average, as this period had characterized rise of the work level therefore decrease with the unemployment, and the upward trendthat the Libyan economic that go through during this periodback to unit a combination of factors as one of the important is the rise of the oil prices in the national market, which arrange on it.

A fast growth in the income and spending, which led the State to take many procedures, where the state had put an economic and social growth plans for the years (1973-1975) and the first five-year plan for the years (1976-1980), Targeted by that the infrastructure, urbanism movement,and push forward the economy wheel to diversify its revenue sources.

- **The third stage (1982-1988) the stage of the recession**

This stage had notice a retread with spending the American dollar, and a strong decrease in the world oil prices, where the price of the one oil barrel in the year (1986) reach in to less than (10) dollar to the barrel, which led to decrease the oil income, the thing that made the State to intervention and working on set the public spending and austerity policies approach, and stop execution many of the economic projects which was listed in the second five-year plan (1981-1986).

The thing that arrange on it log the economic to the recession where the most prominent features presented with decrease the rate of the economic growth, and raise the rate of prices growth, where the inflation had reached almost (5%) in average.

These factors lead to discredit of the Libyan economic therefore to the local and outside fiscal unbalance, and the deepening of structural imbalances in different economic sectors, where the black market had shows and also the exchange market that parallel, as the deterioration of the balance of payments leaving behind inability amount to 1377.9 million dinar in the year (1981), and increase the inability of the public budget to the total output with the current market prices to (13.6%), the thing that arrange on it set many correction procedures which was decreasing the dinar value in face of other coins, limited the public spending, applying the cash policy, canceling some of policies that own to customs protection and replace it with the consumption tax.

- **The fourth stage (1989-2015) the stage of economic correction**

This stage had noticed enter Libya to the economic correction stage, which target the restructuring of Libyan economic, addressing the structural imbalances and direction it with what fits on the economic, policy, region, and national development, and working on push the pace of the activity economic with more depended on the self possibilities, where the methodological framework of the economic correction had focused on working with the demand management policies that purposing to suppression themagnificationpressure,laying an ingredients of a

well-established for stability and economic growth in the shadow of achieving a relative stability in the local level prices, as it had also focused on strengthen the supply side policy through making the structural correction, and that's by minimize the rule of the State in the economic activity, and also applying some fiscal repairs which purposing increase flexibility of the fiscal sector, as the program based on achieving its aims by making the State withcontinue with adopt the rationalization approach in the public spending, and the perfect uses of the public sources in a manner insure achieving a steadily growth.

TheLibyan economic had achieved an relative average growth rate during the period (1998-2005) in the real gross domestic product, where this growth back to rising of the local demand pace for consumption and investment caused by the improving the oil prices, where the available datapointed to, although the real gross domestic product had achieving a real growth its rate reached in the average about (3.5%), the inflation rate continue with rises, where it value reached this high (4.3%) when the outstanding balance of public local debt had decrease as a percent of the local result of (70.8%) in the year (1988) to (25.5%) in the year (2002), and with that improve the balance of payment had been put where it continue with a good achieving except the year (2001) which achieved an inability as much as 75.8 million dinar, where that's back to the surplus that came of the outstanding balance to the improving of the oil prices which its percent is almost (99%) of the gross exports, when with connected with the total inability of the public balance as a percent of the gross domestic product had decrease from (11.2%) yearly during the recession period in to (2.7%) yearly during this period.

As with what connecting with the analyses the reality of the public spending in the Libyan economic during the period (1990-2015), as the first half of the nineties century had noticed a marked fall back in the public spending and that's to direct the public policy to decrease the global economic recession effects in the national economic by rationalization the size of the public spending and working on rebalance between the revenues, expenses, and the payment balance, and that's aware to the decrease that happened in the oil revenues because of the oil prices, but in the second half of the nineties century had noticed an marked increase in the size of the public spending of improving which happened in the oil prices, and also to the last direction of the State to the increase of the public services size that introduction to citizens, where the increase had continue with this spending until the yearly composite growth rate had reached (16.2%) during the years (2007-2009) when it was (5.3%).

But the consumer spending, the rate of it input in the gross domestic product was high in the beginning of the nineties but it had decrease during the last years to reached to the less value in the year of (2008) with rate (26.92%), until the yearly composite growth rate had become (2.4%) and the cause of that back to the State direction to encourage the industrial sector and customizing the big part of the spending to invest seeking to achieving a requirements of the development project, where the Libyan economic had noticed a marked investment expansion, where the rate of the contribution of investment spending in the gross domestic product had increasing to reach to (73.08%), as the rate of the composite growth of this spending reach to (20.89%) for years (2003-2007) and during the years (2007-2009) reach to (19.6%) the thing that make sure the politic and economic direction of the State to rise the sectors contribution rate non-speed in the gross domestic product during increase the investment spending.

2.4. The Challenges and Future Economic Policies

2.4.1. The Challenges that Faced the Economic Growth

The economic growth in Libya had faced a set of economic and social challenges, as what pointing of the notes on the Libyan economic during the main indictors of the growth rates during the past period, that economic had facing these challenges:

- Rentier of Libyan economic, where the sector of crude oil extraction and exported it contribution with rate (34.3%) in generating the local product and with rate (95%) of the exports gross.
- The rule that limited to the privet sector in the development project in terms of the decreasing its rate contribution in the economic active, generating a work opportunities, covering the increase in the local demand, and financing investments.
- The high rates of the unemployment, where the high rates had continues for almost (12.2%).
- The tumble of the investment plans in the execution stage which is back to the weakness of the coordinate between the public fiscal policy of the state and the investment policy, the things that led to big growth of the government consumer spending with out to come with it a big improving in the government services.
- The weakness of the Libyans sharing in the projects that been listed in the social development program and depended on the foreign item.

- The weakness of the local market which not allowed to develop the production sectors, and excessive in expansion with the size of the public spending during the period of seventies and beginning of eighties.
- The decrease of the efficiency of the work item, where the data pointing to depended on execution most of the development plans on the foreign workers.
- The inactivity of the industrialization policies, which means the strategy of industrialization that direct to import substitution doesn't work with active the rule of this sector in the national economic.
- The limited of the rule of the fiscal and monetary policy to processing the problem that Libyan economic had exposed to it.

2.4.2. The Future Economic Policy of the Economic Growth

On the light of diagnosis the reality, the available humans and material potential, and defined the problem and challenging that facing the development in Libya, a plan had been put to economic correct of the Libyan economic, selected the necessary means and policies to achieving it in purpose of get an advance rates in the economic growth, and when we meditate with what come in the plan of vision and future economic policies on the economic in Libya and closing that with the economic reality in Libya and the variables that came on it and what had recorded and recording on that reality of opinion and notes, as a reflex of what came of visions and future economic policy, it may be an important and reality way to advancement with the economic rates growth in Libya on the total level on the national economic and on the sector level, and that's according to:-

- Required with the targets of the economic plan to connect to specific economic activity, for a specific period of time, measured accurate measure with the necessary of clarify the wording of aims to cross the possibilities of interpretation and different hermeneutics, and to make the aims a reality building on the light of balanced estimates to the future of the economic activity and derived from the history reliable information by using a appropriate ways for economic prediction, that's beside to the important to make the aims homogeneous, integrated, and consistent without interference[Bodka, Dr-Abd Al-Kader Mohammed, 1979, P (285-287)].
- The Libyan economic in his rentier current circumstances is the most need to an economic rule to the State that make the partnership easier and the merger with the private activity through a look that seen in the economic alliance between the State active and the works sector, a decisive shift to a perfect economic and social performance and with high harmony and compatibility of generating a well-established pattern of sustainable development.
- As the Libyan economic always need to follow a total policy including find a stable environment on the total economic level that make sure to stimulate the economic growth through control on the swell rates and the public spending that unjustified economic and socially.
- In the Activity funding: the important of activate the government outlay development (specialist) to take on it shoulder a missions to grant soft loans to the privet sector, and also restructuring support in the public arbitrage of the State and depended a Priorities doesn't missed the product activity on the privet sector, as it should be activate the laws, orders, prevalent instructions and what can be add to it of improvement that service the private investment climate in Libya.
- Minimize the spending in the operational paragraphs for saving money that direction to the investment beside employs the operation spending o support the investment spending.
- The balance in the distribution of investment in the base of the balance between the principles of social justice (as for services and the infrastructure) and on the base of the economic efficiency (as for production activity).
- The needs become so urgent to improving and increase the product level and developing the competition level in all the national economic sectors and raise the absorptive capacity of the investment and the ability to creative a new sources for wealth.
- And what the investment need in Libya in the future (Saleh. Dr-Muzher Mohammed Saleh, 2010): the development situation of Libya pointed on delve into the fragmented patterns of the investment doesn't strength the result of the growth unless it depending on a strong push of the development presented the minimum of the investment, its rate doesn't less than (15-20)% of the gross domestic product that necessary for make sure a self and sustained growth, and by that the availability of the minimum of the investment with it broad form consider as a necessary condition to achieve the self sustained growth that's limited with availability an external economic savings.
- Guarantee an important fiscal riser of the private investment activity that ensure the success of projects of the privet activity and it's initiative in two directions:

The first ones: Renew the areas and the existing industrial parks which owned it the private sector by saving technology, work ways, production, marketing that leave the past technological era, with making (5-7) new industrial parks in all country.

The second ones: presented by supply an important much of the social capital or the infrastructure that prepares to the industrial areas a research subject which it's exist considered as a necessary complement of self sustained growth supplements.

At the level of the agricultural activity, depended on strategy agricultural areas that ensure the grain security in Libya by suggestion make (7) agricultural areas by reality of (1) million acres for each area of the new intensive agricultural form take care of it the private activity by help from the State, beside of the expenses of the residential complexes idea on the same standers.

The open field in front of the foreign investment must be subject to the laws and the national legislation (Iraqi) of the organization, control, areas, limits and what ensure the right of each parts and achieving the hops benefit.

2.5. The Conclusions and Recommendations

Of all what came before about the economic growth and its main indicators in the Libyan economic for period (1970-2016) and discussed it, we can say that the economic growth is accounts today a increase interest of the concerned and specialists and with all government levels, private sector and public peoples which looking forward to the achievements of development that fit with the stage natural.

2.5.1. The Conclusions

*The investment is the most important factor in the development process, and the state of the growth shaping in the total process of formation the capital and establishes an economic and social mechanism to sustain and increase the accumulation of the capital.

In the country that characterize with late of the technological level, the rate of the demand capital growth to accommodate a new technology is being bigger of what it's in the development countries and of these main specialize which characterizing of it the economic development countries are the scarcity of capital, the miss used of it, and the inadequate of what it consists to finance the necessary investment to rise.

*The start point in the resource allocation process is how to maximize the rate of the growth of the output by using the local resources and how minimize the needs of this process of the foreign coins.

*The economic growth is means in general the increase of the income of a particular state, the increase in the goods and services which produce by particular economic and this increase in the capital, the technological advances and improving the education state are considering as the main reason of the economic growth.

*In Libya the period (1970-1980) had noticed a rise in the growth rates in the main product sectors and during the period (1986-2010) the development march had fall back and the positive development effects that been achieving in the seventies had limited because of the two war, the siege, the exceptional circumstances that Libya been through in the eighties and the period that follow it.

*The popular growth had kept its high pace (4.21%) during the seventies and eighties of the past century, but the pace of this rate had noticed a decrease after the year of (1990) in to (2.80%) affecting with the conditional that Libya go through it, and therefore the decrease of the values of the demographic indicators after the year (2003) but the rate of the popular growth in Libya had reached in rises to a rate of (3.26%) on the long average range during the period (2000-2010).

*The gross domestic product had increased with composite growth rate as much as (13.0%) of the underway prices for a period of time (1970-1980) but it decrease to (2.5%) of the underway prices for period of time (1980-1990) and (3.7%) of the fixed prices, and that's decrease back to filling the big part of the economic resources to the war effort and to the partly stopping of the oil exports during this period.

And the data of the economic growth of the period (2003-2007) had showed that the gross domestic product had achieved a yearly growth as much as (11.8%) of the underway prices and (6.8%) of the fixed prices which reflex the existence of a high inflationary pressures.

*During the period (1970-1979) the size of the making the fixed capital had increase with composite growth rate as much as (15.6%) of the underway prices but it's achieve a negative yearly growth rate as much as (7.55%) during the eighties as what came of the war from the devastation of the asset of the production capacities.

And during the period that came after (2003) as the making of the fixed capital had achieved a rate as much as (128%) of the underway prices and (97%) of the fixed prices thanks to the development in the government investment.

*During the year (2003) the proportion of government consumer spending to the local product (9.5%) while the investment spending doesn't reached except (8.8%) for the same year.

After (2005) and as a result of the relative improvement in the investment activity , the rate of the fixed capital to the gross domestic product had return to rise to reached (15.6%) in the year (2007) and that's considered as a positive indicator that servant the economic growth path in Libya.

*The main indicators of the Libyan economic growth rates during the period (1970-2010) pointed to that economic is facing a different challenges that the research pointed to it, and as the important ones of it:

- The rentierof the Libyan economic as the extraction and export of crude oil sector contributes with rate (95%) with generated the gross domestic product.
- The limited rule of the privet sector in the development process as the rate of it contributes of making the gross domestic product was (13.4%) during the eighties of the past century, rising toalmost reached the limit of (30%).
- The high rates of the unemployment had continued and it was almost (18.2%) as the last statistical survey of it in the year (2007) with clearly focuses of it among young people and women.
- The big growth in the government consumer spending which had formed a competition to the investment spending which leading in its part to decrease the rate of the public growth of economic.

2.5.2. TheRecommendations

The recommendations that we see are more important and provide opportunities to take by it to increase and stability the rates of the economic growth.

- 1- The rebuilding is require a concentration of the economic resources, planning ways and the central administration between the State which requiring provide a hung of capital.
- 2- Working on increase the rate of the contribution of the productive sectors (Agriculture, Industry and tourism) in the gross domestic product and the possibilities of material and human in Libya are available for that if it's been advantage and development well.
- 3- Activate the product investment of the natural and human resource during customize the necessary fiscal resources with what make the investment politic is support to the aim of achieving a comprehensive and sustainable development of the national economic.
- 4- Support the privet sector and build a partnership between it and the public sector and strengthen its role in all the economic and social activity.
- 5- Open the way (studying) in front of the direct and indirect foreign investment with continue in development the laws and legislation which organizing that.
- 6- The formation of the new investment consider as a very important factor with select the economic growth rate and increase the income with quick rates to cross the population growth rates for rising the living level of individual as the investment consider ad the first condition to achieving a quick rates of the economic growth.
- 7- Beside making an economic plans and apply it, it's necessary that:
 - Using the economic account in way that more comprehensive and profoundto reach the perfect use of resource.
 - Continuing applies the plan and considered that as not separated partof the planning process and that's according to mechanics and procedures that clear and accurate.
 - Select the future development of the Libyan economic for the ten years at less during preparing for the future predictive calculations.
- 8- And as we see it important and the necessary of had intention on it is go far or cross the phenomenon that the human only find himself in reach gain or take of gain in all ways that available in front of him, as that consider of the very dangerous things, especially in the way of the development process, as the development experiment in the development state hadshows that the development, building, and evolution are need the human that find himself in the form of the society and the country level during new add, special product work and as a part of

the development situation and that's what we need for to live up with economic growth rates in Libya and its prosperity.

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RESEARCH ARTICLE

EXPERIMENTAL STUDY ON SORPTION BEHAVIOR OF DESICCANTS

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Key words:-

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Abstract

Moisture removal from air is a practice that is beneficial to many applications. One of the methods to separate water vapor from air is by using desiccants. Present work depicts water vapor sorption behavior of CaCl₂, Silica gel, Molecular sieve, Bentonite powder and Aluminum oxide desiccants. In comparison to open air conditions, superior water vapor sorption was exhibited by all the desiccants in closed glass casings containing water, due to availability of higher humidity. After 141 hours of absorption time, CaCl₂ absorbed 36.2% of its weight in open air conditions and 43.1% due to availability of higher humidity in closed glass casings having water. In comparatively lower humidity conditions prevailing in open air conditions, Molecular sieve exhibited superior adsorption capability than Silica gel while inside closed glass casings, Silica gel overpowered Molecular sieve during the later half of adsorption process. Regeneration experiments for desiccants showed that 100% regeneration of Silica gel and CaCl₂ could be obtained when heated to around 117.5°C and 134°C respectively for 5 hours. LiCl and Molecular sieve indicated 70.6% and 59.7% regeneration respectively when heated to 140°C.

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Introduction:-

Separation of moisture from air is a practice that is being used in many applications for achieving the desirable benefits. Some of the applications where practice of moisture removal from air is followed are in chemical industry to remove water produced by chemical reactions, air dehumidification air-conditioning system for comfortable and healthy environment [1, 2], humidity control and air drying for industrial and agricultural sector like textile mills, post-harvest crop storage etc. [3], extracting water from air [4], control of humidity in food packaging [5], Moisture control in pharmaceutical [6] and electronics industry [7].

One of the methods to separate moisture from air is by using materials having affinity to attract water vapor and retain it in the form of water. Some of the commonly occurring materials that have the property to attract moisture from air are clay, sand, wood, sugar, caramel, honey, glycerol, table salts, polymers material such as cellulose, nylon etc. Commercially available materials having the property to attract water vapor from air are known as desiccants and the mechanism by which these materials attract and hold water vapor is adsorption or absorptions [8, 9]. Some of the desiccants are Calcium Chloride, Lithium Chloride, Lithium Bromide, Triethylene Glycol, Silica Gel, Aluminum silicates (Molecular sieve or Zeolites), Aluminum oxides, etc. These desiccants have been reported in many research works for sorption of water vapor molecules from air. As energy saving alternative Lithium chloride

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has been used for air dehumidification and desiccant regeneration [10]. Molecular sieve has been used for dehydration of natural gas [11] and for understanding moisture adsorbing behavior [12]. It is also reported that hygroscopic Lithium bromide can be used for providing an attractive alternative to conventional cooling systems [13]. Triethylene glycol desiccant was used in cooling system for dehumidification and regeneration [14]. Water vapor adsorption by Silica gel has been studied for desalination [15]. Use of Calcium chloride is reported for absorption of water vapor from air [16].

The objective of the present work is to study through experimental trials water vapor sorption behavior of different desiccants and identify their regeneration capability for reusable purpose.

Theoretical background:-

Desiccants attract moisture from air and thereby facilitate mass transport of water vapor molecule due to low vapor pressure created at the surface. Desiccants have low surface vapor pressure; this is due to the fact that it contains large surface area as a result of availability of pores. When these water molecules migrate at the surface due to low vapor pressure, adsorption occurs physically or chemically and in some cases by both the process. Desiccants like Silica gel, Molecular sieve, etc exhibit adsorption mechanism, whereby the water vapor molecules bind on a solid surface as a result of surface force attraction such as physical, electrostatic force or chemical adsorption. The molecular distances inside the pores of the adsorbent are much shorter than in the gaseous phase for similar pressure and temperature conditions and thus the adsorbate density in adsorbed phase becomes liquid-like due to van der Waals attractive forces. When pore size is very small, another phenomenon called as capillary condensation occurs. Capillary effect is mainly due to intermolecular van der Waals forces, surface tension effect and wetting behavior of water on desiccant surface. Due to capillary effect there is lowering of vapor pressure over the surface of desiccant and thereby facilitates condensation process.

In case of metal chloride desiccants like LiCl, CaCl₂ and LiBr water vapor is attracted to these desiccants via absorption process. Due to this absorption process there is lowering of vapor pressure over the surface of these desiccants. During absorption, water vapor molecules completely dissolve and dissociate the ions of these desiccants. The driving force for absorption in this case is the negative enthalpy of reaction of water with these desiccants; in other words it is thermodynamically favorable to absorb water molecule. There is also a physical interaction between electropositive metal ions and lone pair of oxygen in water through ion-dipole force.

Materials and Methods:-

For studying water vapor sorption behavior, self indicating irregular granular Silica Gel of 3-4 mesh size [Merck Specialities Pvt. Ltd., India], Aluminium Oxide of particle size 0.05 to 0.15 mm [Merck Specialities Pvt. Ltd., India], 8 x 12 mesh Molecular sieve 4A (Sodium Alumino Silicate) [HiMedia Laboratories Pvt. Ltd., India], Bentonite powder (Aluminium Silicate) of size lesser than 25µm [HiMedia Laboratories Pvt. Ltd., India] and Calcium Chloride (fused) 95% [Loba Chemie Pvt. Ltd., India] were considered. The desiccants chosen were all of general grade. The photograph of these desiccants is shown in figure 1.

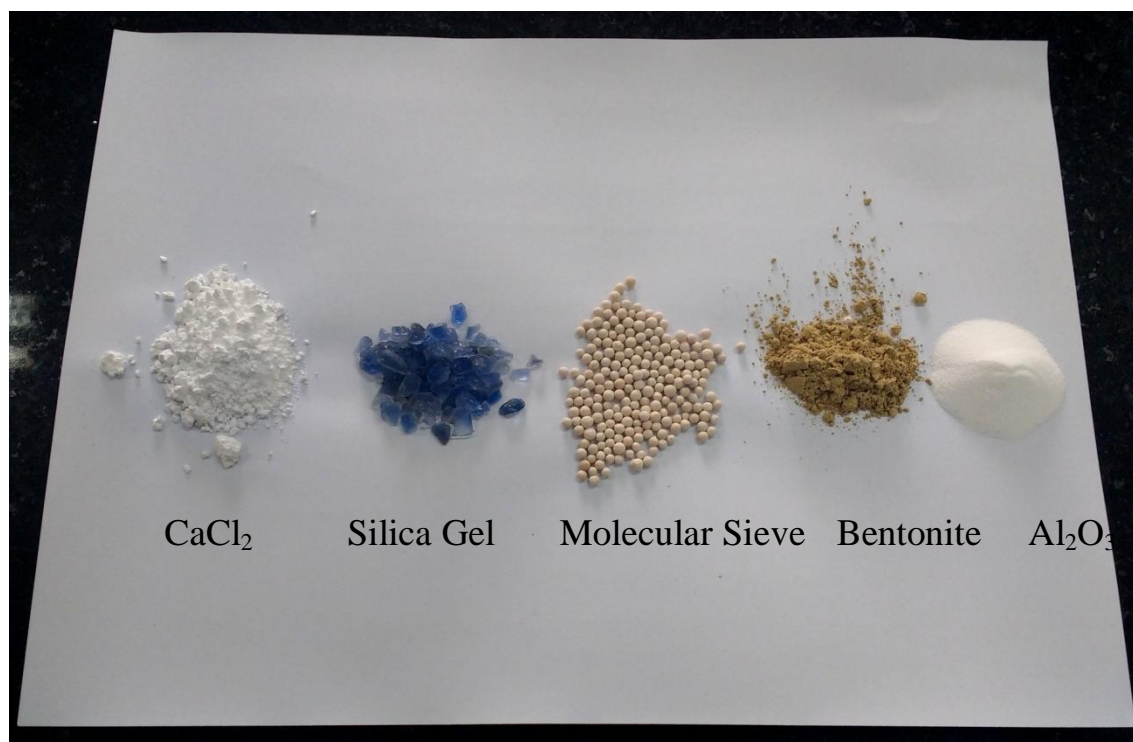


Figure 1:- Photograph showing Calcium Chloride, Silica Gel, Molecular Sieve, Bentonite powder and Aluminium oxide desiccants.

Two batches of experiments were carried out, one in open air condition and another inside closed glass casings containing disc with water, for higher humidity condition. In one batch of experiment 10g each of the desiccant materials were kept in glass beakers for 141hrs in open air condition as shown in figure 2. The air temperature and humidity were recorded to vary between 20°C to 26°C and 52% to 71% respectively during the duration of experiment. In another batch, same quantity of desiccants were kept inside glass beakers and the beakers were placed inside closed glass casings having disc containing water as shown in figure 3. In this case due to water contained in the disc, humidity was observed to be 100%.

Mass gained by the desiccants materials due to sorption of water vapor from air, in both the batches of experiments, were recorded at various intervals of time during the experiment and mass of water molecules held by desiccants were found out. In both the batches of experiments percentage of water collected, relative to the mass of desiccants, from atmospheric air were obtained for all the desiccants and graph showing variation of percentage sorption with time were plotted.

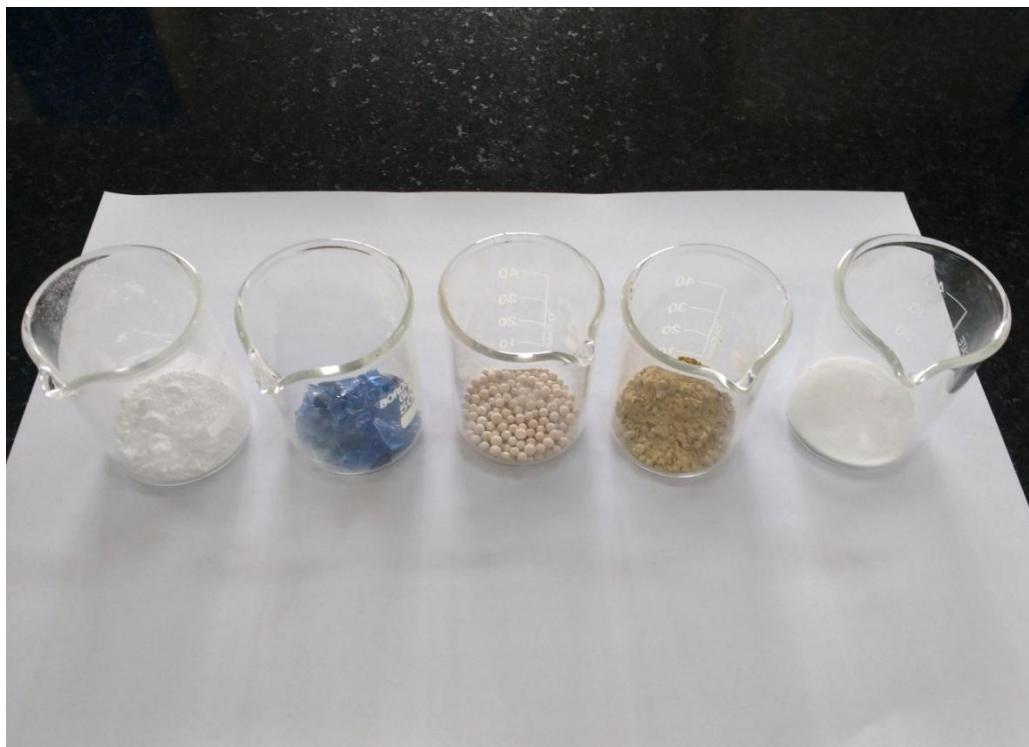


Figure 2:- Experimental setup for water vapor sorption in open air condition by desiccants.

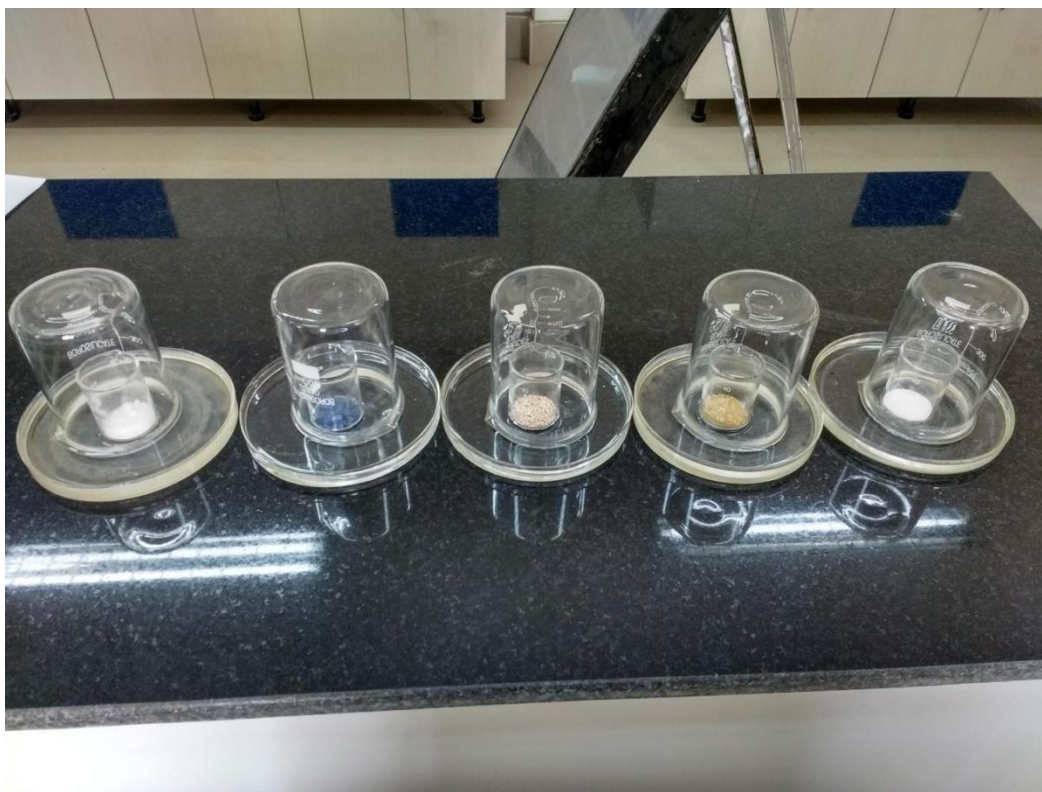


Figure 3:- Experimental setup for water vapor sorption in closed glass casings for higher humidity conditions.

For studying the regeneration temperatures LiCl, CaCl₂, Silica gel and Molecular sieve desiccants were taken into consideration. Four batches of 5g each of LiCl, CaCl₂, Silica gel and Molecular sieve were placed like before inside

a closed glass casings having disc containing water for 7 days and the relative humidity was observed to be 100% inside the glass casings. After 7 days of water vapor sorption by these desiccant materials, they were taken out from the glass casings and weighed in for quantifying the amount of water sorption. Immediately after sorption of water vapor, each batch of these desiccants was then heated for 5 hours in an oven at different temperatures. The temperatures chosen for heating the batches of desiccants were 110°C, 120°C, 130°C and 140°C. After heating in the oven, desiccants were weighed again to obtain the amount of water lost due to heat. Percentage of water removed due to heating effect from the desiccants was calculated as the ratio of mass of water removed during heating and mass of water vapor held during the sorption period of seven days. Since percentage regeneration of desiccants is considered as percentage of moisture removed from the desiccants, graph showing variation of percentage regeneration of desiccants with heating temperatures was plotted.

Results and Discussion:-

Sorption in open air conditions:-

Water vapor absorbed/adsorbed at different intervals of time by 10 grams of each desiccant material in open air condition is shown in table 1.

Table 1:- Mass of water vapor held by desiccants at different intervals time in open air condition.

Desiccants	Mass of water vapor absorbed/adsorbed					
	6 hrs	23 hrs	45 hrs	93 hrs	117 hrs	141 hrs
CaCl ₂	0.15	0.55	1.63	2.70	3.19	3.62
Silica Gel	0.13	0.44	1.09	1.56	1.74	1.90
Molecular sieve	0.17	0.64	1.80	2.07	2.09	2.09
Bentonite	0.05	0.16	0.29	0.30	0.32	0.33
Aluminium oxide	0.04	0.12	0.21	0.20	0.21	0.21

It is found that during the experiment intake of water vapor by Bentonite powders and Aluminium oxide was significantly less and this is due to the fact that surface area of these desiccants are much lesser than of other desiccants. In comparison to Bentonite and Aluminium oxide, Silica gel was found to be superior desiccant with higher moisture adsorption of 1.90 grams in 141 hrs. The reason behind this is due to the internal structure of Silica gel, consisting of network of interconnected microscopic pores formed by connections of the polysilica gel particles via oxygen[17]. This structure causes large surface area in the range of 500m²/g [18], producing a much reduced partial pressure of 0.13 Pa in an unsaturated state [19] and hence it has great affinity to adsorb water vapor. The adsorption process occurring in Silica gel is first by bonding of water molecule onto the hydroxyl groups of silica gel followed by further physical adsorption in additional layers by van der Waals forces, which are the decisive driving force for decreasing the vapor pressure [20]. At the end of experiment after 141 hrs of sorption process, CaCl₂ was found to absorb highest amount of moisture 3.62 grams, 1.9 times more than by Silica gel, followed by Molecular sieve with 1.90 grams. The reason behind CaCl₂ absorbing higher amount of moisture from air is due to thermodynamically favorable reaction of water molecule with CaCl₂, having enthalpy of formation for hexahydrate as -2218 kJ/mol. The bonding of water molecule with CaCl₂ is due to electrostatic interactions of Ca²⁺ ions with lone pair of oxygen. And once the absorption initiates, vapor pressure at the surface of the CaCl₂ lowers significantly facilitating higher absorption of water vapor.

Variation of percentage of water vapor intake with time by desiccants from air in open air condition at relative humidity between 52% and 71% is shown figure 4. After 141 hours CaCl₂ absorbs water vapor 36.2% of its weight, Molecular sieve and Silica gel adsorbs 20.9% and 19.0% of its weight. Whereas, Bentonite and Aluminum oxide

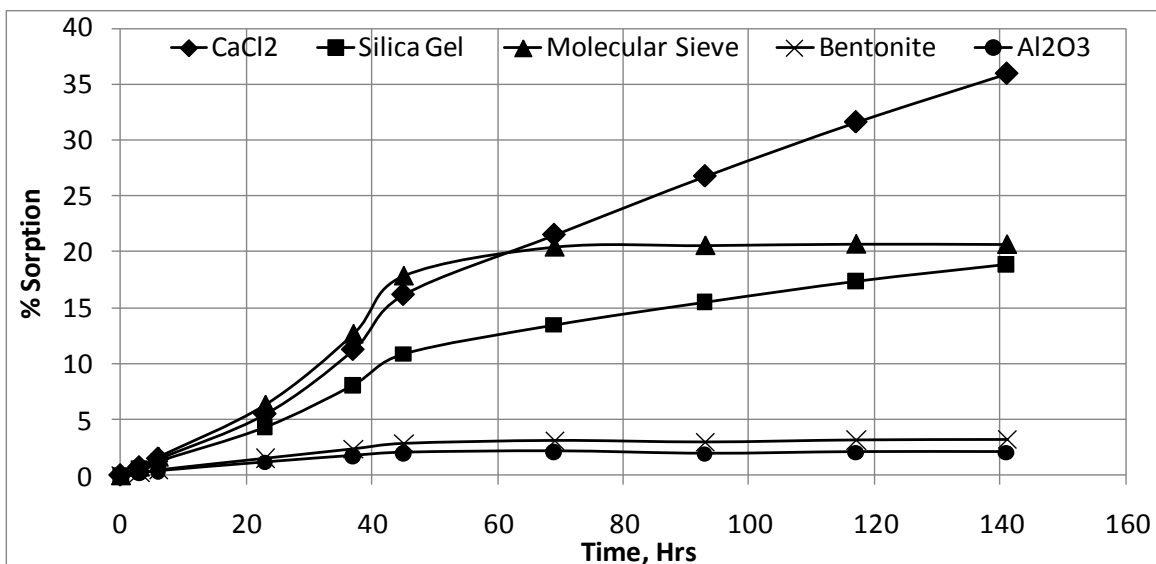


Figure 4:- Graph showing percentage sorption of water vapor by different desiccants in open air condition.

exhibits poor efficiency with water vapor adsorption of 3.3% and 2.1% respectively. It is observed that Molecular sieve in open air conditions that is in lower humidity, exhibited superior adsorption behavior than Silica gel with higher amount of water vapor adsorption at any stage during the experiment. Even when compared to other desiccants, Molecular sieve showed relatively better adsorption capability from initial to 60 minutes of adsorption time. The reason for molecular sieve showing superior behavior to other desiccants at lower humidity is due to the fact that molecular sieve has uniform network of pores with uniform pore size [21, 22].

Sorption in closed glass casings having higher humidity:-

In case of higher humidity conditions that is experiment conducted in closed glass casings, water vapor intake with respect to time by 10 grams each of desiccant materials is shown in table 2. In comparison to low humidity conditions in open air conditions, sorption of water vapor from air by all the desiccants were higher due to availability of higher humidity in closing glass casings having water disc. Like in open air condition here too CaCl₂ showed superior absorption capacity with 4.31 grams of water at 141 hrs. Further effect of higher humidity on adsorbing behavior of Molecular sieve and Silica gel was also observed whereby after 141 hrs of adsorption time, unlike in open air condition Silica gel with 2.72 grams of water exhibited superior capability than molecular sieve.

Table 2. Mass of water vapor absorbed/adsorbed by desiccants at different intervals time in closed glass casings with higher humidity.

Desiccants	Mass of water vapor absorbed/adsorbed					
	6 hrs	23 hrs	45 hrs	93 hrs	117 hrs	141 hrs
CaCl ₂	0.17	0.61	1.87	3.19	3.78	4.31
Silica Gel	0.15	0.52	1.39	2.14	2.45	2.72
Molecular sieve	0.17	0.66	1.92	2.16	2.18	2.19
Bentonite	0.09	0.29	0.67	0.92	1.11	1.09
Aluminium oxide	0.08	0.25	0.60	0.89	1.01	1.12

Variation of percentage intake of water vapor with time by desiccants in higher humidity conditions is shown in figure 5. It is observed that due to higher humidity percentage sorption of water by desiccants is improved with Calcium Chloride absorbing as high as 43.1% after 141 hrs.

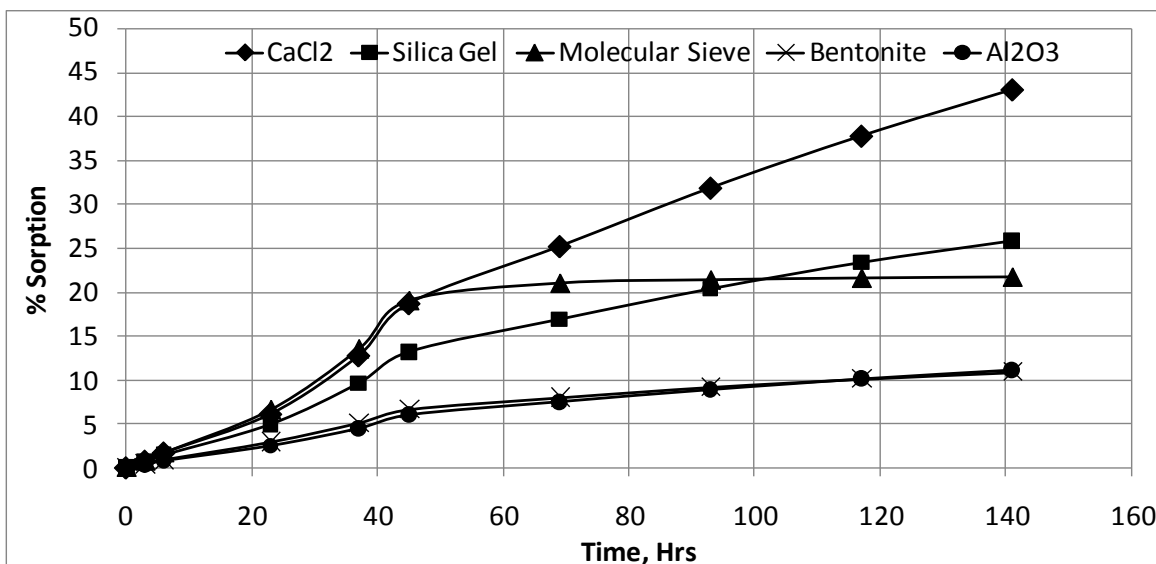


Figure 5:- Graph showing percentage sorption of water vapor by desiccants in closed glass casings having higher humidity condition.

Also compared to other desiccants, relatively higher improvement in adsorption of water vapor was seen in case of Aluminium oxide, 11.2% and Bentonite powder, 10.9%; the margin of improvement in adsorption for Aluminium oxide and Bentonite was 9.1% and 7.6% respectively. As in lower humidity conditions, water vapor sorption rate was highest for Molecular sieve in initial part of adsorption process but in later half unlikely Calcium chloride showed exponential increase in moisture intake.

Regeneration of desiccants:-

Experimentation for studying regeneration temperature showed water sorption by 5 grams each of LiCl, CaCl₂, Molecular sieve and Silica Gel. Quantity of water vapor held by desiccants in seven days is shown in table 3 and water vapor removed later on after heating these desiccants at different temperature is shown in table 4.

Table 3:- Mass of water vapor held by desiccants in seven days.

Desiccants	Batch no.1 (g)	Batch no.2 (g)	Batch no. 3 (g)	Batch no. 4 (g)
LiCl	5.48	6.93	7.08	6.52
CaCl ₂	4.84	6.03	6.18	2.97
Molecular sieve	1.13	1.32	1.17	1.24
Silica Gel	2.02	2.05	2.14	2.24

Table 4:- Mass of water vapor removed from desiccants during heating in grams and percentage.

Desiccants	Batch no.1 110°C		Batch no.2 120°C		Batch no. 3 130°C		Batch no. 4 140°C	
	(g)	(%)	(g)	(%)	(g)	(%)	(g)	(%)
LiCl	1.85	33.8%	3.70	53.4%	4.37	61.7%	4.60	70.6%
CaCl ₂	2.78	57.4%	4.73	78.4%	5.19	84.0%	3.74	125.9%
Molecular sieve	0.45	39.9%	0.69	52.3%	0.64	54.7%	0.74	59.7%
Silica Gel	2.20	108.9%	2.34	114.1%	2.38	111.2%	3.29	146.8%

It is observed in case of LiCl and Molecular sieve, water vapor removal gradually increases with heating temperatures; water removal at 110°C is 33.8% for LiCl and 39.9% for Molecular sieve and whereas at higher temperature of 140°C mass of water vapor removed is increased to 70.6% and 59.7% respectively. Calcium chloride indicates 125.9% of water vapor removal at 140°C which suggest that heating temperature has exceeded the regeneration temperature and regeneration temperature is apparently 140°C. While in case of Silica gel 108.9%

water removal is observed even at 110°C, which indicates that the regeneration temperature of silica gel is below 110°C. Variation of percentage regeneration of desiccants with heating temperatures is shown in figure 5.

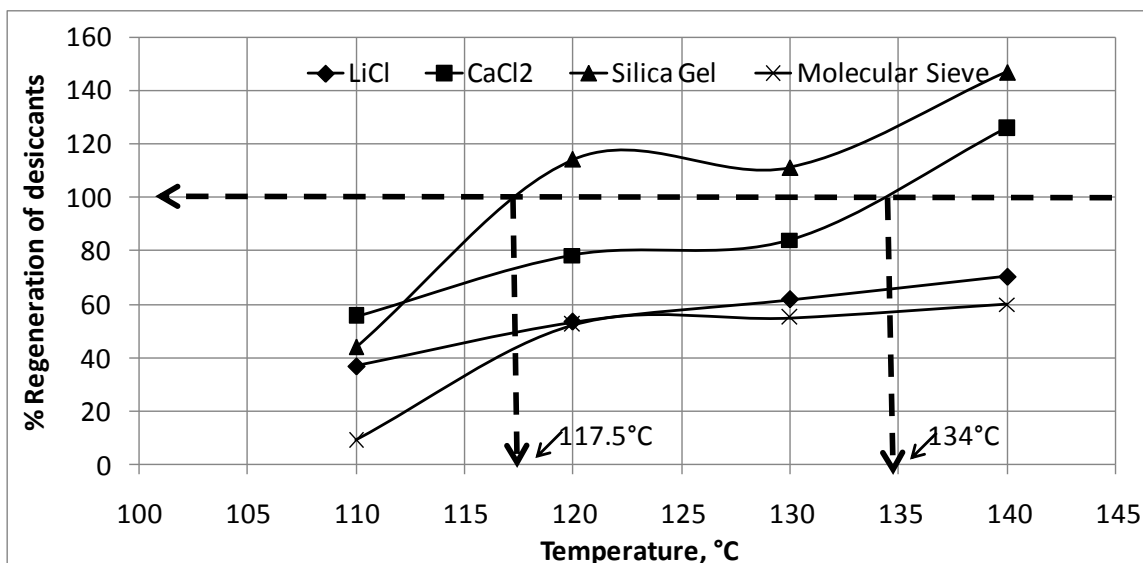


Figure 5:- Plot showing percentage regeneration of desiccants at different heating temperatures.

From the plot, it is observed that 100% regeneration of Silica gel and Calcium Chloride occurs when heated to around 117.5°C and 134°C respectively. These temperatures corresponding to 100% regeneration are regarded as the regeneration temperature of the Silica gel and CaCl₂ when heating duration is 5 hours. In case of LiCl and Molecular sieve, even at highest experimental heating temperature of 140°C it did not give away all the moisture and showed incomplete regeneration.

Conclusion:-

Moisture removal from air by desiccant route is a viable method. Compared to Aluminium oxide and Bentonite powders, CaCl₂, Molecular sieve and Silica gel are relatively more efficient for sorption of water vapor from air. Higher humidity condition generates higher sorption of water vapor from air by desiccants. Molecular sieve even at relatively lower humidity exhibits good adsorption capacity of water vapor molecules. The regeneration temperatures of Silica gel and CaCl₂ is in present experimental study is observed to be 117.5°C and 134.0°C respectively, whereas LiCl and Molecular sieve desiccant showed 70.6% and 59.7% regeneration respectively, when heated at 5 hours at 140°C.

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RESEARCH ARTICLE

Knowledge, Attitude and Practices towards Road Traffic Safety Regulations among Health Science Students in Uttarakhand: A cross-sectional study

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Abstract

Background: Every year, more than 20 million people are injured and 1.17 million are killed due to road traffic accidents. According to the WHO, this is the second most important cause of death among 15 to 29 years old. The objective of this study was to determine the knowledge, attitudes and practices of healthcare profession students in Uttarakhand regarding road traffic safety regulations.

Materials and Methods: This descriptive cross sectional study was conducted among 150 students of Medical (100) and Nursing Science (50) at All India Institute of Medical Sciences, Rishikesh, Uttarakhand. Stratified random sampling technique was used and data was collected using a self-structured questionnaire.

Results: Data reveals that majority of them drive two wheelers (84%) and only 3.3% of them had learned driving training from driving school. Furthermore, only 38.7% own the driving license and 20% of them had history of Road Traffic accident. Moreover, even less than one fourth of the students (18.7%) had high level of knowledge about Road Traffic Safety and mean attitude score was 24.12 ± 3.5 (Mean%, 75.4%). Fortunately, 78.7% students use indicators while turning, 77.3% always blow horn before overtaking, 85.3% do not prefer driving after drinking alcohol, 66.7% always obey all traffic signals. However, 33% of them endanger their lives by not using helmets, (36%) not wears seatbelt while driving/seating in car (34%) and sometimes using mobile while driving (32.7%).

Conclusion: This study reveals that only 18.7% of students has high level of knowledge regarding road traffic safety regulation and majority (81.3%) of them had moderate to low level of knowledge. However, they had good mean score of attitude but still they endanger their lives, probably this has contributed in their inappropriate practices of road traffic safety. Therefore, they require sanitizations program to improve the knowledge, attitude and practice of road traffic safety regulations to reduce the chances of road traffic accidents.

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Introduction:-

Every year the lives of almost 1.24 million people are cut short globally as a result of a road traffic accident. Around 20-50 million more people suffer from non fatal injuries. According to World Health Organization (WHO), road traffic accidents kill more people around the world than malaria, and are the leading cause of death for young people aged between 15-29 years old, especially in developing countries.ⁱ

The primary reason for the road accidents is increasing motorization, along with this contributing factors are over speed, poor following of traffic rules, driving while drinking, not wearing seat belts and helmets, using mobile phones while driving and poor road designs. In 2010, there were close to 5 lakh road accidents in India, resulting in more than 1.3 lakh deaths and inflicted injuries on 5.2 lakh persons. These translate into one road accident per minute and one road death every 4 minutes. Unfortunately, more than half the victims were in economically active age group. The loss of main bread winner can be catastrophic.ⁱⁱ **Error! Reference source not found.**

According to a report on accidental deaths in India 'Road Accidents' cases in the country have marginally increased by 0.7% during 2013 compared to 2012 while casualties in road accidents in the country decreased by 1.2% during 2013 as compared to 2012. It has also been observed that the rate of deaths per thousand vehicles has decreased marginally from 1.4 in 2009 to 0.9 in 2013, as the number of vehicles in the country has increased by 78.0% and the quantum of 'Road Accidents' has increased by 5.1% during the same period.ⁱⁱⁱ

In the state of Uttarakhand, 10.8 persons are killed per 10000 motor vehicles which is the very close to national average of 10.9 and during the year 2010, there were 773 fatal accidents causing 931 deaths and injured 1656 persons^{iv}. The state's capital Dehradun tops the list of persons injured in accidents followed by Haridwar, Udham Singh Nagar, Tehri and Chamoli. Hence we realized the need to assess the knowledge, attitude and practices of health sciences students regarding road traffic safety regulations in Uttarakhand.

Materials And Methods:-

A descriptive cross sectional study was conducted among the Medical and Nursing students of All India Institute of Medical Sciences, Rishikesh, Uttarakhand. A sample of 150 medical (100) and nursing (50) students was drawn using stratified random sampling technique. Data was collected through a self-structured questionnaire, which was distributed randomly among the students through Faculty In-charge of Medical and Nursing programs.

The questionnaire consisted of personal data and background information followed by a knowledge questions, attitude scale and expressed practice scale. The knowledge questionnaire consisted of 15 questions. Attitude scale consists of 8 statements, which were categorized as: strongly agree, agree, disagree and strongly disagree. The expressed practice scale consists of 12 statements categorized under always, sometimes and never practiced. The study was approved by Institutional Ethical Committee and confidentiality of information and anonymity of subjects was maintained. The data was collected after obtaining the verbal consent of the subjects. Data was analyzed using SPSS version 16 and data was presented using descriptive statistics.

Results:-

The data revealed that majority of the students (73.9%) were in the age group of 18-19 years and nearly equal number of them were males (52.7%) and females (47.3%). Furthermore, majority of them (75.3%) were either rural or suburban dwellers. Majority of them (61.3%) of them verbalized that their parents' monthly income is in between 10,001 – 50,000 per month (Table 1).

Table 1:-Socio-demographic characteristics of the students**N =150**

<i>Socio-demographic characteristics</i>	<i>f (%)</i>
<i>Age in years</i>	
17	07 (04.7)
18	51 (34.2)
19	58 (38.9)
20	23 (15.4)
≥21	10 (06.7)
<i>Gender</i>	
Male	79 (52.7)
Female	71 (47.3)
<i>Course of Study</i>	
Medical	100 (66.7)
Nursing	50 (33.3)
<i>Parent's monthly income in Rupees</i>	
Don't Know	14 (09.3)
≤ 10,000	16 (10.7)
10,001 – 50,000	92 (61.3)
≥ 50,000	28 (18.7)
<i>Place of residence</i>	
Urban	36 (24.0)
Rural	72 (48.0)
Semi-urban	41 (27.3)

Table 2 depicts that significantly good number of students (61.3%) did not owe a driving license, but still they drive two wheelers (84%) and four wheelers (16%). Moreover, only 3.3% of them learned driving from a formal driving school, rest of them (96.7%) learned it from family members/ friends. Furthermore, 20% of students verbalized the history of road traffic accident and in addition, 62% of them also mentioned the history of road traffic accident among their family members/ friends.

Table 2: Driving and Road Traffic Safety related information of students**N = 150**

<i>Variables</i>	<i>f (%)</i>
<i>Status of driving license ownership</i>	
Owes	58 (38.7)
Did not owe	92 (61.3)
<i>Type of vehicle drives</i>	
Two wheelers	126 (84.0)
Four wheeler	24 (16.0)
<i>Source of driving training</i>	
Family/ friends	145 (96.7)
Driving school	05 (03.3)
<i>H/o Road Traffic Accident (RTA)</i>	
Present	30 (20.0)
Absent	120 (80.0)
<i>H/o RTA among family/ friend</i>	
Present	93 (62.0)
Absent	57 (38.0)

Illustrated data in Table 3 shows that nearly one fourth of the students (23.3%) had low level, about half of them (58%) had moderate level and only few of them (18.7%) had high level of knowledge about Road Traffic Safety. The Mean attitude score of the students regarding the Road Traffic Safety was 24.12 ± 3.5 . The mean percentage of attitude score was 75.4% with range of 8-32.

Table 3:-Level of Knowledge and mean attitude towards Road Traffic Safety among students
N = 150

Knowledge& Attitude	Statistics
Level of Knowledge	
Low	35 (23.3)
Moderate	87 (58.0)
High	28 (18.7)
Attitude Score	
Mean±SD	24.12±3.5
Mean Percentage	75.4%
Range	8 – 32

*Attitude Score; Minimum= 8 & Max= 32; Figures given in parenthesis is percentage

It was found good number of students always practiced healthy road traffic safety regulation such as use indicators while taking turns (78.7%), blow horn before overtaking (77.3%), obey traffic signals, lights, signs (66.7%). However, many of them endanger their life by never practicing very crucial road traffic safety regulations such as never wearing helmet (36%), seat belts (34%), sticking to zebra crossing (24.7%) and following specified speed limits (10.7%).

Table 4:-Practices of Road Traffic Safety among students
N = 150

Road Traffic Safety Practices	Always f (%)	Sometimes f (%)	Never f (%)
Wear a seat belt when you are driving/ seating in a four wheeler?	54 (36.0)	45 (30.0)	51 (34.0)
Keep specified speed limit in mind while driving?	68 (43.0)	66 (44.0)	16 (10.7)
Use helmet when driving a two wheeler?	41 (27.0)	55 (36.7)	54 (36.0)
Obey all traffic signals, lights and signs?	100 (66.7)	38 (25.3)	12 (8.0)
Driving a vehicle when alcoholic?	---	22 (14.7)	128 (85.3)
Use mobile phone while driving a vehicle?	---	49 (32.7)	101 (67.3)
Stick to zebra crossing while crossing a road?	31 (20.7)	82 (54.7)	37 (24.7)
Use indicators while taking turns and look both sides before turning?	118 (78.7)	18 (12.0)	14 (9.3)
Use right side of road while driving?	---	44 (29.3)	106 (77.7)
Overtake from left side?	19 (12.7)	75 (50.0)	56 (37.3)
Blow horn before over take?	116 (77.3)	24 (16.0)	10 (06.7)

Discussion:-

Road traffic fatalities remain a major public health problem, with the highest fatality rates per 100000 population in low and middle income countries in Asia. These countries have higher road traffic fatality rates (21.5 and 19.5 per 100 000 population, respectively) than high-income countries (10.3 per 100 000). WHO predicts that road traffic injuries will rise to become the fifth leading cause of death by 2030.¹ Indian data shows that road traffic injuries account for 20-50% emergency admissions, 10-30% of hospital admissions, and 60-70% of people hospitalized with traumatic brain injuries.^v

We had nearly equal number of male and female participants. The majority (73.9%) were in the age group of 18-19 years, which is the most vulnerable group for road traffic accidents. Majority of our students were from middle class families, while few were having a monthly parental income <Rs 10000. This must be highlighted that persons from poor economic settings are disproportionately affected by road traffic injuries, even in high income countries. For instance a study in New South Wales, Australia^{vi}, found that children of relatively lower socioeconomic status were at highest risk of a road traffic injury. Another research from Bangalore^{vii} has found that mortality from road traffic injuries was 13.1 and 48.1 per 100000 in poorer socioeconomic groups of urban and rural populations respectively, compared to 7.8 & 26.1 among their more affluent urban and rural counterparts.

54.7% of students in study admitted about crossing the specified speed limits. Similar practices have been observed by another study from south India^{viii}, where 68% of students admitted about it. Many other Indian as well as

international studies have shown similar observations.^{ix,x,xi} An increase in average speed is directly related both to the likelihood of a crash occurring and to the severity of crash consequences^{xii,xiii}. A 5% increase in average speed leads to an approximately 10% increase in crashes that cause injuries, and a 20% increase in fatal crashes.^{xiv} Pedestrians have a 90% chance of surviving a car crash at 30 km/h or below, but less than a 50% chance of surviving impacts of 45 km/h or above^{14,xv}.

The risk of involvement in a crash increases significantly above a blood alcohol concentration (BAC) of 0.04 g/dl^{xvi,xvii}. Young or novice drivers are at a much increased risk of having a road traffic crash when under the influence of alcohol. Laws which establish lower BAC between zero and 0.02 g/dl for young/novice drivers can lead to reductions of between 4-24% in the crashes involving young people^{xviii}. 14.7% of our participants were indulging in drunken driving at times. Moreover, majority of our study participant were from age group between 18-19 years. This finding has been observed by others^{8,9}. A study from Haryana reported that 18% Road traffic accident victims gave history of having consumed alcohol within 6 hours before RTA^{xix}. However the percentage of accident victims under influence of alcohol was reportedly lower in older studies from India (4.6-8%)^{xx,xxi} showing that incidence of drunken driving is increasing in India.

Only 27% of students in our study always wore a helmet while driving. Wearing a motorcycle helmet correctly can reduce the risk of death by almost 40% and the risk of severe injury by over 70%^{xxii}. When motorcycle helmet laws are enforced effectively, helmet wearing rates can increase to over 90%^{xxiii,xxiv}.

66% of participants in our study use a seat belt (36 % always use it, another 30% consented that they use it off and on), while 34% told that they never use. A study from another medical college from India has shown 74% participants wearing the seat belts². Wearing a seat belt reduces the risk of a fatality among front seat passengers by 40-50%^{24,xxv,xxvi} and fatalities of rear seat occupants by 25-75%^{25,26,xxvii,xxviii}. Mandatory seat belt laws, their enforcement and appropriate public awareness campaigning have been shown to be very effective in increasing rate of wearing seat belts^{25,xxix,xxx}. 67% of study participants never use mobile phone while driving. Similar findings have been reported by Jogand S et al.^{xxxi}

A recent study on adolescent medical students from Maharashtra⁹ has found that knowledge about road traffic rules was good in 42.6%, average in 42.53%, and only 14% of students had poor knowledge. These findings are similar to our observations depicting that young student population is having an overall satisfactory knowledge but when it comes to actual use of this knowledge, many falter. In the above mentioned study, 80% donot use helmets, 68% donot have driving license and nearly two third do not always follow the traffic rules. Thus it is clear that though good attitude can play a major role in changing the behavior and practice of individuals, this is hardly enough. This must be accompanied by strict implementation of traffic laws. We also recommend that the knowledge about traffic rules may be included in basic school curriculum, so that children have an idea about road safety from a young age.

Conclusion:-

The present study assessed the knowledge, attitude and practice of health sciences students regarding road traffic safety regulations and found that students were having moderate level of knowledge. However, they had significant positive attitude towards obeying them. The human body is highly vulnerable to injury and that humans make mistakes. A safe road traffic system is therefore one that accommodates and compensates for human vulnerability. No country can afford to sit back and assume that its road safety work is complete and international community must continue to recognize road traffic injuries as an important health and development issue and intensify support for their prevention. Significant progress in national road safety requires close collaboration between relevant leaders and agencies. India being one the global leader in road traffic accidents has much work ahead. The knowledge of young individuals is moderately good but they still require frequent sensitization programs and implementation of traffic rules to develop practices which are actually safe on the road. Road safety measures through signboards, posters and mass media need to be strengthened to reduce the incidence of road traffic accidents. We also suggest addition of road safety knowledge as a part of curriculum of school children.

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RESEARCH ARTICLE

A STUDY ON PROBLEMS OF THE ELDERLY PARENTS OF MIGRANT WORKERS

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Aging, Migration, Life satisfaction

Abstract

Aging is a natural, universal and inevitable with the passage of time. It is a continuous process in end of our life. Old age is the end stage in the life journey and closing period in the life span of a man with decreased capacity for adaptation. The aged are otherwise known as elderly, senior citizen "Geriatric age group" and second childhood. The field of study was dealt with aging and the special problems of the elderly parents of migrant workers. The population of the older persons in India is continuously increasing due to the advancement in medical science, improvement in living conditions and public health facilities have prevented epidemics and brought the general diseases under control. This has increased the life expectancy. So, the children should extend their support. But due to urbanization, occupation the children are migrated from their own places. The parents are underwent many problems like, physical, psychological. The elderly persons need care and support of the family members. The main objectives of this study were psychological problems of the respondents and level of life satisfaction.

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Introduction:-

The term gerontology comes from the Greek word *geras* meaning 'old age' and *logos* referring to the study of a subject. The term, gerontology is very broad and encompasses the psychological, socio economic and physiological aspects of old age. Social gerontology is concerned with reciprocal relationship between the individual and society. The term geriatrics deals with causes and amelioration of physical pathology in old age. Senescence, a biological term refers to government that is ruled by elders. Ageism is discrimination against individual on basis of old age. The term geronto phobia is used to designate the fear of growing old or the fear of hatred of the aged. The term ageing refers to the process of growing old.

India has around 100 million elderly at present and the number is expected to increase to 323 million, constituting 20 per cent of the total population, by 2050.

Review of Literature:-

Daniel Doyle and Marilyn J. Forehand (2014) made a study on Life Satisfaction and Old Age a Re examination. Using data from a large, representative national sample, this article replicates some earlier analyses regarding life satisfaction and old age. Persons over age 65 were found to be only slightly less satisfied with their lives than persons aged 40 to 54 and 55 to 64. Poor health, loneliness, and money problems are the strongest correlates of life

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satisfaction across age groups. The small negative correlation between age and satisfaction is largely eliminated when controls for these three factors are imposed. A regression analysis is used to determine the relative strength of a number of possible predictors of life satisfaction within different age groups.

Prakash I.J., (2003) conducted a study on Aging, disability, and disabled older people in India. India is witnessing a demographic revolution, leading to a considerable increase in the proportion of older people in the population. Similarly, life expectancy of both the mentally and physically disabled has improved considerably. About 5% of Indian older people have problems with physical mobility. Aging has become a gender issue in India not only because more women are surviving into old age; they are also vulnerable and disadvantaged in many ways. In most cases they are the only caregivers available for the old and disabled. Older Indians are considered a high-risk group for multiple morbidity. It is estimated that nearly four million Indians suffer from mental problems. India has around 12 million people designated as "handicapped." However, little information is available about disabled people who grow older. The National Policy on Older Persons, which has been recently formulated, aims at providing an improved quality of life for millions of older Indians. However, the concerns of older disabled and of the disabled who grow old are still treated separately in both policy and practice.

Kalavathy M.C., Thankappan K.R., Sankara sarma P., Vasan R.S. (2000) made a study on prevalence, awareness, treatment and control of hypertension in an elderly community-based sample in Kerala, India. Hypertension is one of the most important causes of cardiovascular morbidity and mortality in the elderly. With the increase in the number of elderly in India, hypertension is likely to emerge as an important public health problem. The study was performed a cross-sectional survey of a random sample of 357 community-dwelling elderly individuals (191 women, 166 men; mean age 70 years) in Kerala. We measured blood pressure on all study participants using a standardized technique to assess the prevalence of hypertension. We compared the variations in prevalence, awareness, treatment and control of hypertension according to age, sex and place of residence of the subjects, and examined the socio-demographic correlates of hypertension using specific multiple logistic regression.

The overall prevalence of hypertension sample was 51.8% (95% CI; 46% - 56.8%), Which did not vary with sex but increased with age. Fewer than half of the hypertensive subjects were aware of their condition or were on treatment, and only a quarter of the treated hypertensive achieved adequate control of blood pressure. Rural elderly subjects were especially less likely to be aware of, and on treatment for hypertension. Smoking status and rural residence (in men) and marital status (in women) were important correlates of hypertension. The study findings emphasize the public health importance of hypertension in the elderly in Kerala at present, and point to a likely increase in burden of this problem in India in the near future. A National Hypertension Programme, targeted to meet the imminent public health challenge posed by hypertension seems warranted.

Material and Methods:-

Objectives:-

- To study the psychological problems of the respondents.
- To study the level of life satisfaction among the respondents.

Hypotheses:-

- There is significant difference between living arrangement of the respondents and health problem.
- There is significant difference between loneliness of the respondents and their satisfaction in life
- There is significant difference between economic status of the respondents and their standard of living.

Research Design:-

The researcher has used descriptive research design.

Universe

The universe of the study was Labbaikudikadu town panchayat, Perambalur district.

Sample Size:-

The researcher adopted simple random sampling to select the elderly people in Labbaikudikadu town panchayat, Perambalur district. So the sample was 800.

Tool for data collection:-

A self prepared Interview Schedule was used to collect primary data from the respondents.

Hypotheses Test:-

- There is significant difference between living arrangement of the respondents and health problem.

DIFFERENCE BETWEEN LIVING ALONE AND HEALTH PROBLEM

S. No	Living arrangement of the Respondents	Mean	S.D	Statistical inference
1	Yes (n 706)	2.46	1.738	T= 6.07 0.00<0.05 Significant
2	No (n 94)	1.35	.936	

Findings:

It is evident from the above table that there is significant difference between living alone and health problem. Hence, the calculated value Less than the table (0.00<.05).

2. There is significant difference between loneliness of the respondents and their satisfaction in life.

DIFFERENCE BETWEEN LONELINESS AND SATISFACTION IN LIFE

S.No	Loneliness of the Respondents	Mean	S.D	Statistical inference
1	Yes (n 706)	1.46	.499	T= -.141 0.79>0.05 Not Significant
2	No (n 94)	1.47	.502	

Findings:

It is evident from the above table that there is significant difference between living alone and satisfaction in life. Hence, the calculated value Less than the table (0.79>0.05).

3. There is significant difference between economic status of the respondents and their standard of living.

DIFFERENCE BETWEEN ECONOMIC STATUS AND STANDARD OF LIVING

S. No	Economic status of the Respondents	Mean	S.D	Statistical inference
1	Yes (413)	10998.84	10319.912	T= -1.34 0.003<0.05 Significant
2	No (369)	12065.04	12245.889	

Findings:

It is evident from the above table that there is significant difference between Economic status of the respondents and standard of living. Hence, the calculated value Less than the table (0.003<.05).

Conclusion:-

Proper planning for old age is very essential in order to make fuller, happier, richer and graceful aging. Today old age does not necessarily spell death, disease and despair, if one plans ahead for old age. Make efforts to get a steady income during old age. The elderly have been traditionally well-respected and well cared for in Indian culture; experts predict that younger generations in the future are less likely to be concerned and involved in the health care of their older relatives due to urbanization, industrialization or migration not giving much importance to the elderly people. They should consider like child and take care of them. Parents play the vital role to seed the value the next generation to give respect, care and support during the old age period.

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RESEARCH ARTICLE

REMOVAL OF HEAVY METALS BY SURFACE ASSIMILATIVE.

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Heavy metals, Bio sorption, Tea waste,
Textile Industrial waste water.

Abstract

Wastewater contamination is an ever increasing problem which the whole world is facing now. Industrialization & globalization has led to the tremendous increase in use of heavy metals over the past decades. This has gradually resulted in increased flux of metallic wastes in marine, ground, industrial & even treated waste waters. As far as the modern treatment processes are concerned, removal of heavy metals are very expensive, require high energy and generate toxic sludge which pose serious threat to the whole ecosystem. Nowadays, the uses of natural, low cost & effective adsorbents are being highly recommended. As well as the method of bio sorption can help in removing heavy metals to a greater extent. For this project, agricultural waste i.e. Tea waste is used as bio sorbent for the removal of heavy metal like, chromium, cadmium & lead. With this new combination, it is possible to produce pollutant-free water & pollution-free ecosystem thereby reducing metallic pollution at low cost, simple, efficient & by natural means. In the present study, heavy metal contamination in textile industrial waste water has been discussed. The conventional methods of treatment are very costly, energy intensive and generate toxic sludge. Thus, the bio-sorption has been investigated as cost effective method for removal of heavy metals. The study involves the examination of experimental conditions such as pH of the solution, concentration of the solution, contact time and adsorbent loading on the removal of heavy metals from textile industry. Finally, the maximum adsorption of 96% is achieved by adding **1.0 g** of tea waste per 100 ml of wastewater.

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Introduction:-

Water pollution is a major problem in the global context. Several industrial wastewater streams may contain heavy metals such as Cr, Cu, Pb, Zn, Ni, etc. including the waste liquids generated by metal finishing or the mineral processing industries. The toxic metals, probably existing in high concentrations, must be effectively treated/removed from the wastewaters. If the wastewaters were discharged directly into natural waters, it will constitute a great risk for the aquatic ecosystem. In recent years, the removal of toxic heavy metal ions from sewage, industrial and mining waste effluents has been widely studied. Among the many methods available to reduce heavy metal concentration from wastewater, the most common ones are chemical precipitation, ion-exchange, adsorption and reverse osmosis (Chao *et al.*, 2005). Heavy metals are the non-degradable, toxic and possess high density. Some of

the heavy metals are lead, cadmium, mercury, arsenic, chromium and thallium. Heavy metals are major pollutants and dangerous in marine, ground, industrial and even treated wastewaters because they tend to bio-accumulate. Bioaccumulation means an increase in the concentration of a chemical in a biological organism over time, compared to the chemical's concentration in the environment. The purpose of this research is to improve the method to remove high concentration of heavy metals from industrial wastewater. It is important to ensure there are no harmful heavy metals in the water stream because it can accumulate in the environment elements such as food chain and pose a significant danger to human health. The method of bio sorption offers many potential advantage in energy saving, process enhancement and processing time to remove heavy metal. In this research, tea waste was used as an adsorbent to assist bio sorption process because of its low cost, availability and its efficiency to remove heavy metal. Bio sorption is the best purification and separation techniques of heavy metals from waste water but in industries cost is very important parameter in comparison to the efficiency of adsorbent materials from waste water.

Research objective:-

The purpose of this research is to harness the potentials of tea waste by converting it to adsorbent used for adsorption of heavy metals like lead, chromium and cadmium from the textile industrial waste water collected from textile industry. To efficiently remove the metal contaminants and reuse the treated water for various agricultural and domestic purposes. To study the physio-chemical characteristics, analyze the toxicity range and their effects in environment of textile industrial waste water. To stress the advantages of bio sorption method & removal efficiency of heavy metals.

Scope of research:-

- 1) To improve current research by increasing the percentage of heavy metal removal from wastewater using tea waste as adsorbent.
- 2) To prevent the heavy metal concentration release to the water stream, reduce the cost & open up opportunities to increase the demand on wastewater treatment by using natural absorbent like tea waste.

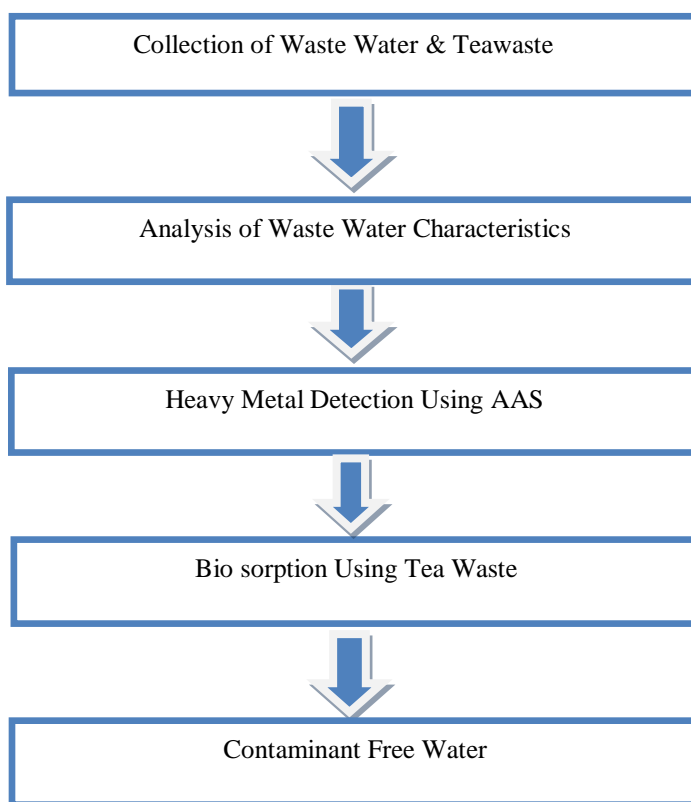
Methodology:-

Fig:- Flowchart for Experimental Investigation

Collection Of Sample:-

The textile industry which consumes very large volume of clean water for a sequence of processes produces large amount of wastewater. The use of different synthetic dyes during dyeing process contaminated the water drastically. The effluents, thus generated from textile industry, are strong in color, high in COD and BOD and less biodegradable. Textile industrial waste water is one of the most pollution sources. It can cause environmental problems related to its high organic matter, suspended solids and heavy metal concentration. The sample is collected from the textile industry in Madurai district. Initially the sample is stored in the plastic bottle and is preserved in a refrigerator in order to prevent the attack of micro-organisms.



Fig 1:- Discharge Of Textile Industrial Wastewater

Biosorbent Used:-*a) Tea waste*

Tea beverages are available as green, black or oolong tea depending on the way of manufacturing. Waste tea leaves contains lignin, cellulose & hemi-cellulose. Their high adsorption capacity is observed in solutions with pH range more than 4. The aminated bio-sorbent is used to remove metallic pollutants like lead and zinc.

b) Properties & Characteristics:

Formation of surface complexation mechanism between metal ions & ligno cellulosic adsorbent are responsible for the absorption of metal contaminants. The rough surface and widely distributed pores can offer greater surface area and more binding sites for metals.



Fig 2:- Extract of tea waste.

c) Applications of tea waste: Utilized in agricultural field to enhance the production under heavy metal stress. Abundantly available & cheap. High metal binding capacity & Helps in reducing turbidity. After adding tea waste in industrial water, the water can be reused for various agricultural and domestic purposes.

Adsorbent Preparation:-

The extract of tea powder, after it is being used as beverage is taken and is boiled till the extract gets decolorized. This tea waste is again washed with distilled water and then dried in hot air oven at 100°C. The obtained dried tea waste is screened through 150 micron IS Sieves and is used as adsorbent. The tea waste is sealed in polyethylene for

preservation so that it does not react with neighboring environment. These poly ethylene are placed in freezer so that tea waste does not get degraded. The experiments were performed using two different concentration of adsorbent dosage i.e. 0.5g and 1.0g of tea waste

Experimental Study Of Waste Water Sample Before Adding Tea waste:-

Atomic Absorption Spectroscopy:-

Atomic Absorption spectroscopy is a “spectro-analytical procedure for the quantitative determination of chemical elements using the absorption of optical radiation (light) by free atoms in the gaseous state”. As per the results obtained from atomic adsorption spectroscopy, the metals present in our sample collected from textile industry are as listed below:

Table 1:- Concentration of heavy metals in sample using AAS

S.NO	PARAMETERS IN SAMPLE	UNITS	AS PER STANDARDS (TOXICITY RANGE)	MCL	RESULTS
1.	Lead	mg/l	0.006		<0.01
2.	Chromium	mg/l	0.05		0.37
3.	Cadmium	mg/l	0.01		<0.01

Over-review of results obtained from AAS & Applications:-

From the results of atomic adsorption spectroscopy, the concentration of chromium is found to be high i.e. (0.37) compared to lead and cadmium with respect to IS Specifications. So the removal of chromium from the sample is necessary. Now, the sample containing chromium is treated by varying the dosage of tea waste and the results are compared.

Applications: It is useful in chemical analysis because of its specificity and its quantitative nature. It is widely applied in the fields of water resources because of its identity in detecting the heavy metals even in significant value.

Physio-chemical characteristics of waste water:-

Table 2:- Physio-chemical characteristics of Waste Water before adding Bio-sorbent

S.No	PARAMETERS	VALUES BEFORE ADDING BIOSORBENT
1.	Color	Brownish yellow
2.	pH value	6.68
3.	Turbidity value	12.3 NTU
4.	BOD(3days)	5.65 mg/l
5.	Total suspended solids	52900 mg/l
6.	Total dried solids	38000 mg/l
7.	Total fixed solids	1000 mg/l
8.	Total volatile solids	2000mg/l

Results and discussions:-

Effect on pH and turbidity:-

pH and turbidity variation are one of the important parameters controlling the uptake of heavy metals from waste water and aqueous solutions. pH is a term generally used to express the intensity of the acidic or alkaline condition of a solution. It is defined as the negative logarithm of reciprocal of the hydrogen ion concentration in moles per liter. The pH scale value ranges from 0 to 14; the value of 0 to less than 7 ranges indicates acidity and greater than 7 to 14 alkalinity. The effect of adsorbent on pH increases with increase in the concentration of adsorbent dosage and the turbidity in the sample decreases with increase in the concentration of tea waste.

Effect on total suspended solids:-

Total suspended solids are the dry weight of particles trapped by a filter. It is a water quality parameter used to assess the quality of wastewater after treatment plant. TSS was previously called non-filterable residue (NFR) but was changed to TSS because of ambiguity in other scientific disciplines. The amount of total suspended solids shows drastic decrease in value with increase in the concentration of adsorbent dosage of 50mg and 100 mg for an optimum contact time of 120 minutes

Effect of total fixed solids & total volatile solids:-

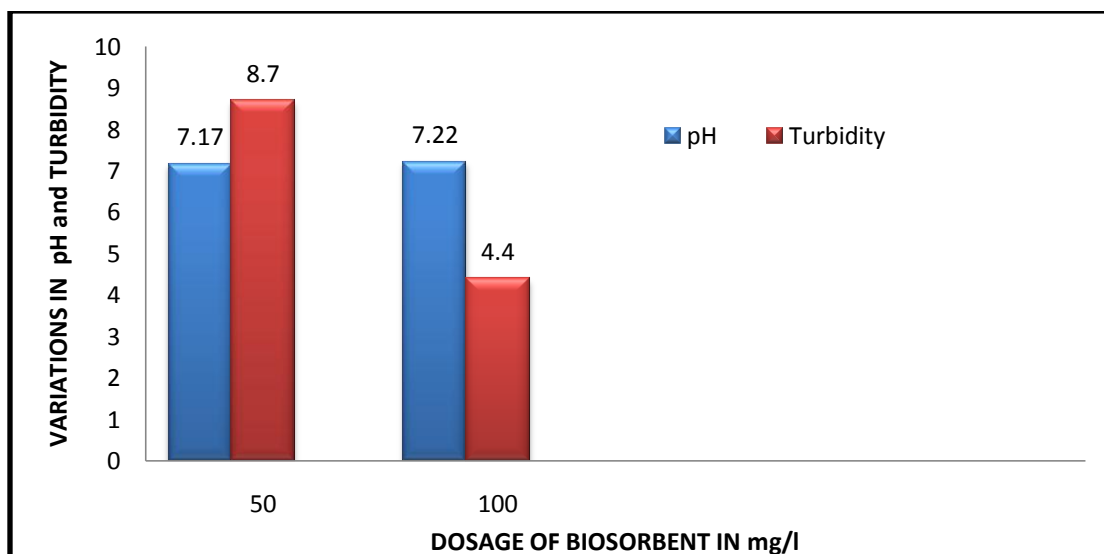
Total dissolved solids is a measure of the combined content of all inorganic and organic substances contained in a liquid in molecular, ionized or micro-granular suspended form. Although TDS is not generally considered as a primary pollutant it is used as an indication of aesthetic characteristics of drinking water. Primary source for TDS in receiving waters are agricultural and residential runoff, leaching of soil contamination and point source water pollution discharge from industrial or sewage treatment plants. In our present study to determine the amount of total dissolved solids present in the waste water a silica crucible is taken and its empty weight is taken as w₁. About 100 ml of sample is filtered using filter paper, and the silica crucible along with the filtered water is placed in the water bath maintained at 900°C till the water is evaporated to dryness. After drying, the crucible is cooled to a little above the room temperature and kept in a desiccator for 1 hour, and that crucible is weighted as w₂, the difference between w₁ and w₂ gives the value of total dissolved solids. The amount of total fixed solids decreases with increase in the concentration of adsorbent dosage. The graph shows the variations in total fixed solids values for the addition of adsorbent dosage of 50mg and 100mg. The amount of total volatile solids decreases with increase in the concentration of adsorbent dosage. The graph shows the variations in total volatile solids values for the addition of adsorbent dosage of 50mg and 100mg.

Effect on Biosorption efficiency:-

The concentration of chromium decreases with increase in concentration of chromium for the contact time of 120 minutes. The maximum percentage of adsorption is found to obtain for 100 mg of tea waste per 100 ml of sample solution.

Table 3:- Physio-chemical characteristics of Waste Water after adding Bio sorbent

S.NO	PARAMETERS	0.5g TEAWASTE	1.0g TEAWASTE	CONTACT TIME
1	COLOUR	Mild yellow	Pale yellow	120 min
2	pH	7.17	7.22	120 min
3	Turbidity	8.6	4.4	120 min
4	Total suspended solids	200mg/l	20mg/l	120 min
5	Total fixed solids	360mg/l	70 mg/l	120 min
6	Total volatile solids	340 mg/l	50mg/l	120 min
7	Adsorption efficiency	81%	96%	120 min

**Fig 3:-** Effect of biosorbent in pH and turbidity

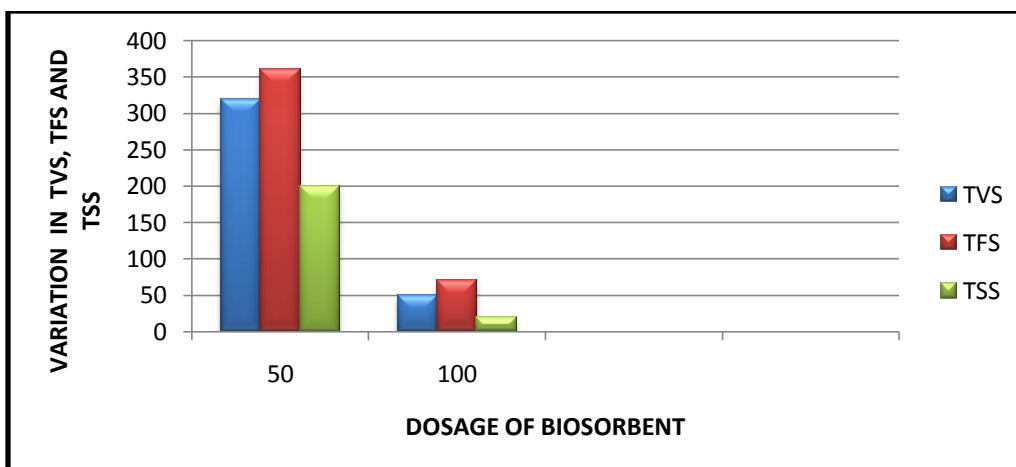


Fig 4:- Effect of biosorbent dosage in TVS, TFS and TSS

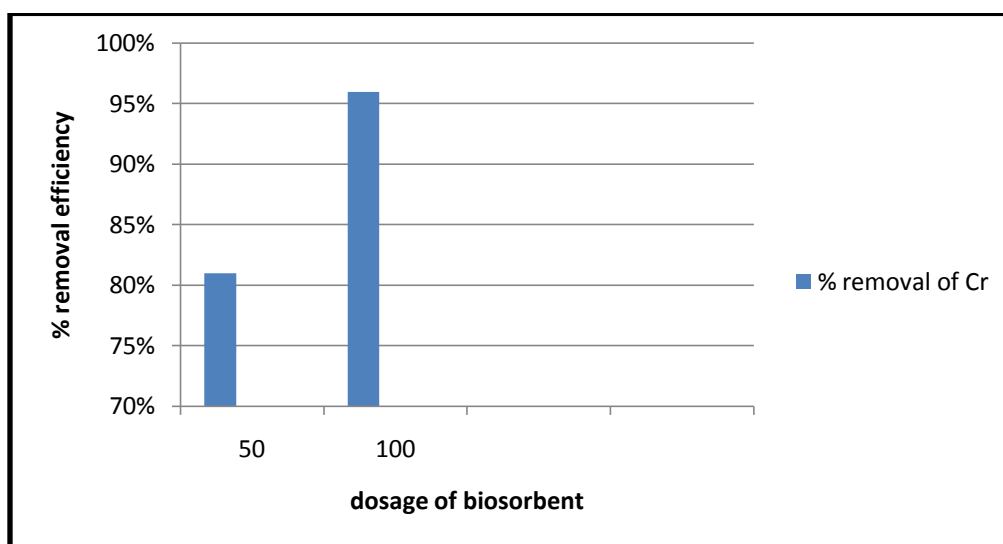


Fig 5:- Biosorption efficiency of chromium

Conclusion:-

Wastewater contamination is an ever increasing problem which the whole world is facing now. Industrialization & globalization has led to the tremendous increase in use of heavy metals over the past decades. This has inevitably resulted in increased flux of metallic wastes in marine, ground, industrial & even treated waste waters. As far as the modern treatment processes are concerned, removal of heavy metals are very expensive, require high energy and generate toxic sludge which pose serious threat to the whole ecosystem. Nowadays, the use of natural, low cost & effective biosorbents are being highly recommended. As well as the method of bio sorption can help in removing heavy metals to a greater extent. For this project, agricultural waste i.e. Tea waste is used as adsorbent for the removal of heavy metal like, chromium, cadmium & lead. With this new combination, it is possible to produce pollutant-free water & pollution-free ecosystem thereby reducing metallic pollution at low-cost, simple, efficient & by natural means. From the results and discussion, the maximum adsorption of 96% is found by adding 1.0 g of tea waste per 100 ml of wastewater.

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RESEARCH ARTICLE

DIFFERENTIAL ACTIVITY OF FOUR SELECTED ENZYMES IN THE PISTILS AND POLLEN GRAINS OF TWO VARIETIES OF *HAMELIA PATENS* JACQ. (RUBIACEAE) FOLLOWING COMPATIBLE AND INCOMPATIBLE POLLINATION.

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Abstract

Hamelia patens Jacq. commonly called fire bush, is a fast growing, semi woody and evergreen perennial shrub of the family Rubiaceae. It is a native of central and southern Florida. It is chiefly grown for the showy bunch of beautiful flowers. In tropical America, local people make use of the extract of its leaves and stem, for curing diseases such as skin rashes, sores, insect sting and various fungal diseases of the skin because of its antibacterial and antifungal properties. In India this plant is particularly cultivated on a wider scale in home gardens as an ornamental plant. In *H. patens*, sexual reproduction is difficult due to self- incompatibility, while crossing between morphologically different plants of two accessions gives viable seeds. In the present study two varieties of *H. patens*, *H. patens* variety *patens* and *H. patens* variety *glabrawas* chosen to study the activity of four enzymes, namely, peroxidase, acid phosphatase, polyphenol oxidase, esterase and total protein. Three developmental stages were identified for both the plants. Controlled pollinations were carried out in the two plants. Stigma of two varieties were pollinated with its own pollen and also pollen from other variety. Crossed stigma showed higher activity of all the enzymes than self- pollinated and stigma from flowers before anthesis. The aim of the study was to understand if pollination with self and crossed pollen grains has any influence on the activity of the four selected enzymes in post pollinated pistils. The enhanced activity of these enzymes indicates their role mainly in the defence mechanism than anything to do with stigma receptivity. The results have been discussed in the light of the available literature.

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Introduction:-

Hamelia patens Jacq. is commonly known as scarlet bush or firebush. It is a large perennial shrub or small tree. Also, the plants are used in folk medicine against a range of ailments. All parts of *H. patens* are used in natural medicine. A number of active compounds have been found in firebush, such as apigenin, ephedrine, flavanones, isomaruquine, isopteropodine, maruquine, narirutins, oxindole alkaloids, palmirine, pteropodine, rosmarinic acid, rumberine, rutin, seneciophylline, speciophylline, isopteropodine, stigmast-4-ene-3,6-dione and tannin (Cavanagh 1963, Duke 2007, Aquino et al 1990, Ahmad et al 2012 & Paniagua et al 2012) (-)-hameline, tetrahydroalstonine, aricine, uncarine F, stigmast-4-ene-3, 6-dione and 5,7,2-,5- -tetrahydroxyflavanone 7- rutiroside (Aquino 1990, Ahmad et al 2012 & and

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Paniagua et al 2012). The bark also contains significant amounts of tannins. Firebush contains crude protein and has *in vitro* digestibility (Ibrahim 1997). The plant possesses analgesic, antispetic, anti-inflammatory, febrifuge, refrigerant properties. Fruit of *H.patensis* edible (Irobi et al 1996) and the plant is used in herbal medicine to treat athlete's foot, skin lesions and rash, insect bites, nervous shock, inflammation, rheumatism, headache, asthma, and dysentery (Lachoria et 1999 and Liogier 1999). Ethanolic extract of different parts like leaf, stem and root of *H. patens* shows anthelmintic and antimicrobial activity (Sapana 2012). It has been reported to exhibit that this plant possesses anti-diarrhoeal activity (Salud 1996). It is also used for insect bites, menstrual disorders, uterine and ovarian affliction (Shrisha et al 2011). Cytostatic and cytotoxic activity against tumor cell lines (Taylor et al 2013) and wound healing activity (Sandhya et al 2011) with *H. patens*. Antifungal properties of aqueous extracts of leaves, flowers and fruits of *H.patens* was also reported (Abubacker 2013). Antibacterial properties (Camporese 2003), vasorelaxant (Reyes & Chilpa 2004) and toxicity and antinociceptive effects of *H.patens* also studied (Castro 2015).

Development of fruits with healthy seeds is the culmination of the natural process of pollination and fertilization. Seeds are important for the plant for propagation of the progeny and for a breeder to assess the results of breeding while developing new cultivars. Louis and Radhamany (2009) and Louis et al (2012) reported RNase based gametophytic incompatibility in *H.patens*. Since legitimate and illegitimate pollinations are affected by alternations in the metabolic status of stigma and style tissues (Dhaliwal 1985), it would be imperative to study the changes in the enzyme activities of stigma-style tissues following compatible and incompatible pollination. Since the enzymes control biochemical reactions, and their synthesis is under the control of specific gene(s), any change in the activity of an enzyme would reflect in the pattern of gene expression and corresponding metabolic events in the cell. Hence, the enzymes can be used as tools to study the problem of self-incompatibility at the biochemical level. In the present investigation, changes in the activities of peroxidase, polyphenol oxidase, esterase and acid phosphatase along with total amount of protein have been studied in anther lobe and pistil of two varieties of *H. Patens*; *Hamelia patens* variety *patens* and *Hamelia patens* variety *glabra*, in an attempt to understand the biochemical aspects of self-incompatibility in *Hamelia*.

Materials and methods:-

Two varieties of *Hamelia patens* Jacq. viz. *Hamelia patens* var. *patens* (Fig. 1) and *Hamelia patens* var. *glabra* (Fig. 2) maintained in the Botanic garden, Department of Botany, University of Kerala, Thiruvananthapuram, and pistils from flowers of these two plants were used in the present investigation.

Controlled pollination:-

Emasculation and controlled pollinations were carried out in both the varieties of *H. patens*. Early in the morning, a day before controlled pollination, mature flower buds were emasculated and bagged with butter paper bags at 'pre-balloon' stage, 18–24 h before pollination. For self-pollination, flower bud at the 'balloon' stage was pollinated by dusting the pollen grains from the same flower on the surface of the stigma and bagged. In the cross pollination, variety *patens* was considered as female plant and variety *glabra* considered as male plant. Following samples were collected for the test of isozymes.

1. Sample I-Self-pollinated pistil from the variety *patens*.
2. Sample II- Un-pollinated pistil from the variety *patens*.
3. Sample III-Self-pollinated pistil from the variety *glabra*.
4. Sample IV- Un-pollinated Pistil from the variety *glabra*.
5. Sample V-Pollinated pistil from the cross between variety *patens* x variety *glabra*.
6. Sample VI-Stamen from the variety *patens* after self pollination
7. Sample VII-Stamen from the variety *patens* before pollination
8. Sample VIII-Stamen from variety *glabra* after self pollination
9. Sample IX- Stamen from the variety *glabra* before pollination

Preparation of enzyme extract:-

Part of the pistil without ovary was collected and frozen in -40°C. Sample weighing about 450 mg were homogenized with 9 ml of cold 5% KCl (w/v) using a pre-chilled mortar and pestle. The homogenate was centrifuged at $0 \pm 2^\circ\text{C}$ at 10,000 rpm for 10 min. The clear supernatant was used directly for the assay of enzyme activities and estimation of protein (Jorgensen et al 1953, Ching et al 1987, Neog et al 2003 and McInnis et al 2006).

Assay of peroxidase:-

The activity of peroxidase was assayed according to Malik and Singh (1980). The assay mixture containing 2.5 ml of phosphate buffer (pH 6.5, 0.1 M), 0.2 ml of diluted enzyme extract and 0.1 ml of *o*-dianisidine (1 mg/ml methanol) was incubated at 28°C in a water bath for 2 min. The reaction was started by adding 0.2 ml of H₂O₂. The change in absorbency was recorded at 430 nm using a stopwatch at 30 s interval for 5 min. The enzyme activity was expressed in terms of the rate of increase of absorbance per hour per mg protein.

Assay of acid phosphatase:-

The reaction mixture contained 0.5 ml of substrate solution (50 mg *p*-nitrophenyl phosphate in 10 ml water + 25 ml of acetate buffer, 0.1 M, pH 4.8) and 0.1 ml of suitably diluted enzyme extract. The mixture was incubated at 35°C for 30 min. The reaction was stopped by adding 2.4 ml of 0.1 N NaOH and the absorbance was recorded at 410 nm. The enzyme activity was expressed in terms of micromoles of *p*-nitrophenol released/h/mg protein Malik and Singh (1980).

Assay of polyphenol oxidase:-

The polyphenol oxidase activity was measured as per the method of Sarvesh and Reddy (1988). The assay mixture (3 ml) contained 2.0 ml of 2 M carbonate-bicarbonate buffer (pH 10), 0.15 M of *o*-catechol and 0.2 ml of suitably diluted enzyme extract. The assay mixture was incubated for 2 min at 25°C. The reaction was stopped by adding 0.5 ml of 5% (v/v) H₂SO₄. The change in absorbency was recorded at 420 nm for 2 min. The enzyme activity was expressed in terms of micromoles of quinone formed/h/mg protein.

Assay of esterase:-

Esterase activity was assayed by naphthol-acetate method (Sawhney et al 1981). The assay mixture contained 2.0 ml of 20 mM phosphate buffer of pH 10, 0.1 M of 1-naphthol acetate and 0.2 ml of suitably diluted enzyme extract. Change in absorbance was recorded at 420 nm for 3 min and enzyme activity expressed in terms of micromoles of naphthol-acetate produced/h/mg protein.

Total protein:-

The amount of protein was determined according to the standard method (Bradford 1976 and Lowry et al 1951), using Bovine serum albumin as standard.

Statistical analysis:-

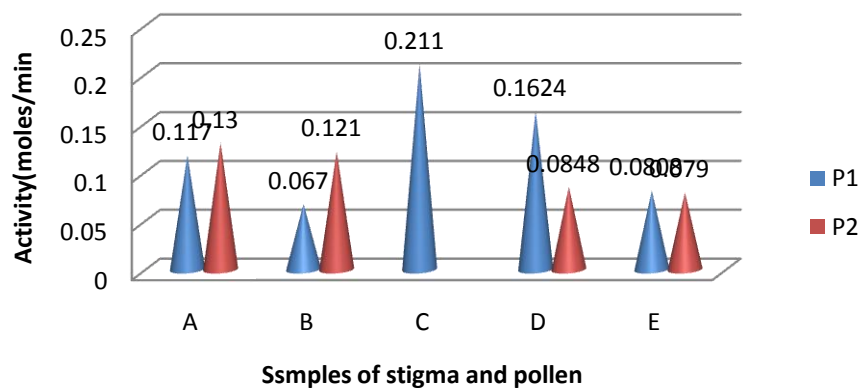
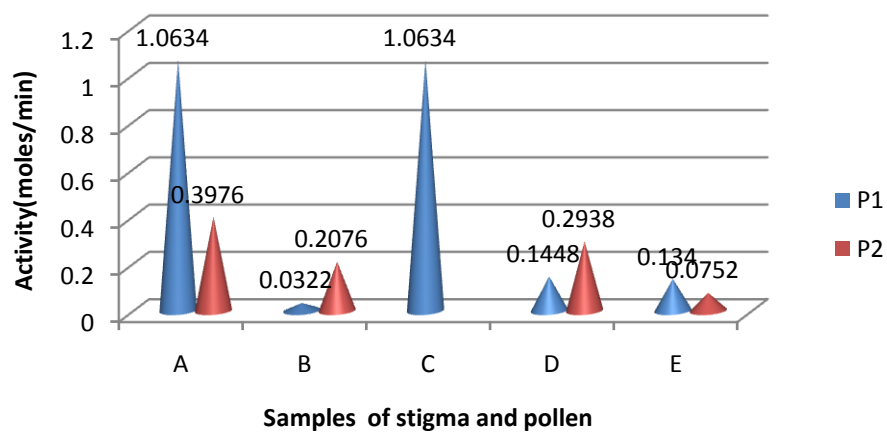
The data were collected from five sets of experiments and were subjected to analysis of standard deviation (Table-1 and Graph 1-5).

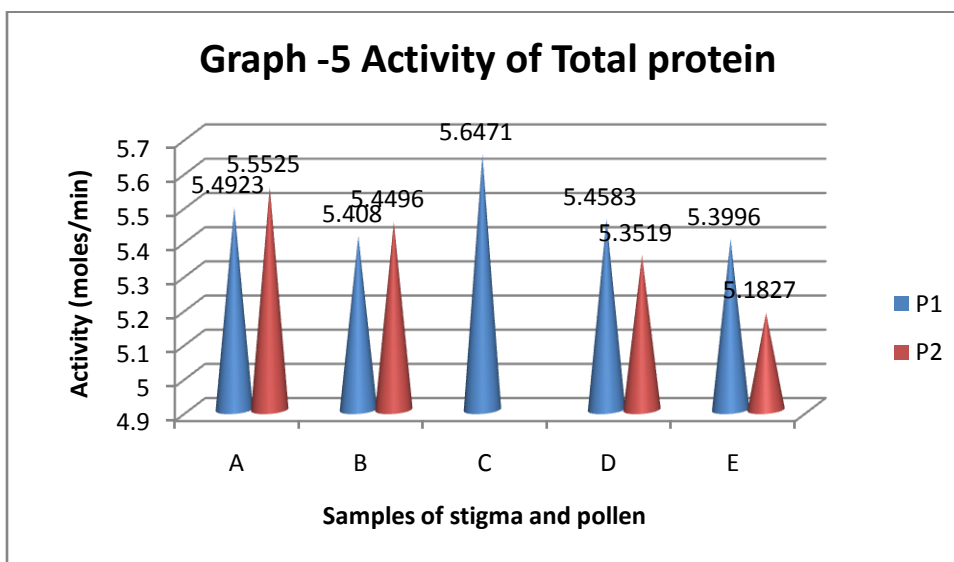
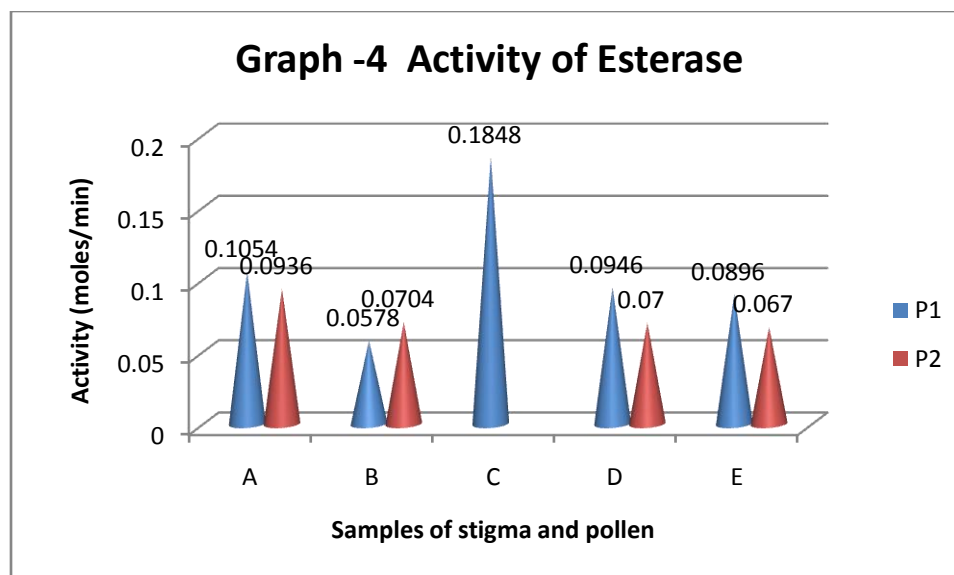
Results:-

Table I:- Level of peroxidase, acid phosphatase, polyphenol oxidase and esterase on protein content after self and cross-pollination in two accessions of *H. patens*

Sl. No	Samples	Peroxidase [h/mg(Mean±SD)]	Acid phosphatase [h/mg(Mean±SD)]	Polyphenol oxidase [h/mg(Mean±SD)]	Esterase [h/mg (Mean±SD)]	Total protein [Mean±SD]
1	Sample I	0.117±0.585	1.0634±0.0011	0.1358±0.0008	0.1054±0.0018	5.4923±0.0658
2	Sample II	0.067±0.0054	0.0322±0.00216	0.1072±0.0008	0.0578±0.0008	5.4080±0.1163
3	Sample III	0.130±0	0.3976±0.0011	0.1122±0.0008	0.0936±0.0020	5.5525±0.0903
4	Sample IV	0.121±0.0004	0.2076±0.0011	0.0952±0.0008	0.0704±0.0024	5.4496±0.0386
5	Sample V	0.211±0	1.0634±0.0011	0.1882±0.0008	0.1848±0.0008	5.6471±0.0257
6	Sample VI	0.1624 ± 0.0005	0.1448 ± 0.0012	0.1648 ± 0.0008	0.0946 ± 0.0011	5.4583 ± 0.0332
7	Sample VII	0.0808 ± 0.0008	0.134 ± 0.0012	0.105 ± 0.001	0.0896 ± 0.0011	5.3996 ± 0.0856
8	Sample VIII	0.0848 ± 0.0008	0.2938 ± 0.002	0.1528 ± 0.0008	0.07 ± 0.0075	5.3519 ± 0.0385
9	Sample IX	0.079 ± 0.00125	0.0752 ± 0.0017	0.1126 ± 0.0005	0.067 ± 0.001	5.1827 ± 0.0336

Values are mean ±sd of six sets of samples

Graph 1, Activity of Peroxidase**Graph -2, Activity of Acid phosphatase**



Graph 1-5:-Activity of the enzymes, peroxidase, acid phosphatase, polyphenol oxidase, esterase and total protein in different stages of stigma and anther(A-sample I and sample III, B-sample II and sample VI, C-sample V, D- sample VI and sample VIII, E-sample VII and sample IX)

Discussion:-

Comparative analysis of the activity of four isozymes namely peroxidase, acid phosphatase, polyphenol oxidase and esterase in *H.patens* var. *patens* and *H.patens* var. *glabra* were carried out to find out the differences in the metabolic activity of unpollinated, self-pollinated and cross-pollinated pistils of self-incompatible *H.patens* varieties in the present study. Peroxidase has been implicated as an indicator of stigma receptivity (Galen & Plowright 1987, Dupuis & Dumas 1990, Dafni & Motte-Maues 1998 and Stpiczynska 2003) and mere adherence of pollen grains to stigma has been shown to increase peroxidase activity independent of the penetration of pollen tube (Galen & Plowright 1987, Bredemeijer 1982, Bredemeijer 1984, Bredemeyer & Blaas 1975, Bredemeyer 1975, Bredemeyer 1979 and Linskens 1969). In the present study, significantly higher activity of peroxidase was observed in cross-pollinated styles were noticed when compared to self-pollinated and unpollinated stigma in both the varieties studied. The mean value was found high in variety *glabra* (0.121), followed by variety *patens* (0.117). Peroxidase

enzyme which is known to be a defence induced enzyme (McInnis et al 2006, Cheong 2002, Delannoy 2003 and Do et al 2003). It is possible that pollen grains landing on stigma set in signals which activate peroxidase as a defence strategy. The activity of this enzyme is modified by germinating pollen grains (Bredemeijer 1984). Activities of acid phosphatase showed a considerable variation between self and cross-pollinated combinations. Activity was significantly higher (1.1894) in the styles after cross-pollination when compared with the self and unpollinated stigmas. The highest enzyme activity was noticed in the variety *patens* (1.0634) than variety *glabra* (0.0322). The elevated levels of acid phosphatase in cross-pollinated stigma clearly indicate a role for this enzyme in pollen tube growth in the style. This enzyme is probably produced in response to the penetrating pollen tube for providing inorganic phosphates through hydrolysis of phosphate esters (Ching 1987). Activity of polyphenol oxidase is very low or negligible in stigmas after self-pollination and stigma before anthesis. In contrast, the crossed stigma showed increase in polyphenol oxidase activity over the selfed styles. The highest specific activity for this enzyme was recorded in the cross-pollinated samples of stigma (0.1882) and the highest enzyme activity was noticed in the self-pollinated style of variety *patens* (0.1358) than variety *glabra* (0.1122). The consistently enhanced activity of polyphenol oxidase in the two plants is in line with its possible role in the defence mechanism (Mayer & Harel 1979, Bashan 1987 and Tyagi 2000). In wet and dry stigmas of a wide range of plants the time of stigma receptivity has been correlated with esterase activity (Heslop-Harrison 1975, Heslop-Harrison 1977, Kohn & Waser 1985 and Lavithis & Bhalla 1995). A moderate level of esterase activity is maintained at all the stages of stigma. Esterase activity was higher in cross-pollinated style (0.1848) than other stages of stigma. Also it is noticed that esterase activity was very low in stigmas of two varieties. Higher activity of esterase probably could be a response to the germinating pollen and must be facilitating in the penetration of the pollen tube into the stigma by hydrolyzing the cutin layer (Heslop-Harrison 1975, Lavithis & Bhalla 1995 and Hiscock et al 2002). Estimation of total protein was found to be higher in all the stages but cross-pollinated styles shows higher amount (5.6471) than other stages of stigma.

Conclusion:-

The enzyme may directly act to prevent the growth of selfed pollen tubes or sometimes may remain absent to disturb the growth of the pollen tube (Nettancourt 1977). Higher level of peroxidase found in cross-pollinated styles than in self-pollinated ones indicates its involvement in the regulation of pollen tube growth through styles. One cannot rule out the involvement of peroxidase as a nonspecific factor of the self-incompatibility reaction, but it plays a significant part in this mechanism. There are reports that this enzyme brings about changes in structural glycoproteins, other enzymes, pectin or cellulose present in the pollen tube wall (Siegel & Galston 1955 and Stonier 1979). Higher level of acid phosphatase in self- than cross-pollination as observed in the present study is in agreement with the earlier finding of Dhaliwal and Malik (1985) in stigma-style tissues of *Brassica campestris* var. *Toria*. It can be inferred that the inhibition of pollen tube growth is due to some factor(s) that could cause the production of acid phosphatase in higher quantity. This enzyme is probably produced in response to the penetrating pollen tube for providing inorganic phosphates through hydrolysis of phosphate esters.

Elevated level of polyphenol oxidase in all the experimental samples following cross-pollination supports the complementary hypothesis and the earlier assumption of Mayer and Harel (1979) that the enzyme has the ability to produce quinone and selectively inhibit the activities of some other enzymes. Though the results of the present study do not confirm any direct relationship between this enzyme and the manifestation of self-incompatibility, it is possible that polyphenol oxidase acts as complementary factor along with other elements like environment and genetic makeup of the plant. In wet and dry stigmas of a wide range of plants the time of receptivity has been correlated with esterase activity. A moderate level of esterase activity is maintained at all the stages, to maintain the receptivity of stigma. The elevated amount of esterase in cross-pollinated stigma could be facilitating in the penetration of the pollen tube into the stigma by hydrolysing the cutin layer. Self-incompatibility mechanism in angiosperms is widespread and has received considerable attention in recent years. Self-incompatibility genes have been identified, characterized, cloned and transferred to self-compatible lines in several species (Hoopen 1998 and Brien 2002). From the present study, it may be concluded that significant changes in the activities of certain key enzymes occur in the stigma and style tissues of *Hamelia patens* following compatible and incompatible pollination. Variations in the enzyme activities as observed in the present study shows that the biochemical changes occur in the stigma style tissues affected by self-incompatibility mechanism that in turn affects the metabolic status of these tissues. As the synthesis of these enzymes is controlled by their corresponding gene(s), their quantitative changes may be due to the changes in the regulation of expression of gene(s) and hence offer a basis to study the mechanism of self-incompatibility at molecular level.

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Fig. 1:-*Hamelia patens* var. *patens*.



Fig. 2:-*Hamelia patens* var. *glabra*

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RESEARCH ARTICLE

THE APOCALYPTIC ANTHROPOCENE EPOCH AND ITS MANAGEMENT IN INDIA

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Abstract

The very existence of Holocene is debated for its tenure and transcendence by a new epoch "The Anthropocene or the age of man". The transition between two epochs is in breakthrough dialogue as human intervention to enact the hydro-meteorological, geological and biological stratigraphic changes. The homo-sapiens are the dominant forces where the earth is exponentially transforming. The exo-planet aliens, PHAs or NEOs can drive the new epoch to be apocalyptic. The era is considered as Eurocentric and launched either from Industrial revolution or from dates of nuclear explosion. The human centric activities like modernization, advancement in science, technology, hydrologic interventions, disproportionate GHG's etc. are the indicators of the new eon. The researchers have wide scope now to use ample anthropogenic, climatic, biologic and geologic data for study but which was underreported or non-reported in past. The burgeoning data available from NASA, FAO, disaster sectors, geo-engineering and bioengineering etc. have been used for the present study. The legacy of the Anthropocene is said to have propagated in spells and phases from Europe to Indian subcontinent. The shift of the epoch is geospatial in geographical time scale. The present study envisages a nonparametric approach about the influence of the earth system of Indian subcontinent by human impact. Investigation is done to ascertain the causes, impacts, effects, longevity of the human dominance over natural variability. The Anthropocene is greatly accelerated with the great oxidation and carbonization processes but the effect of the epoch can be delayed and decelerated by development in geo engineering, bio engineering, smart technology and Nano science etc. The spell, its identity, uniqueness and management of the epoch to avoid both ecologic and economic extinction are discussed in the present research.

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Introduction:-

Hominids predominate in the Holocene epoch as per the records of International Commission on Stratigraphy (ICS). But the concept of succession of the new epoch, "The Anthropocene", was claimed by a Dutch chemist Paul Crutzen, Stoermer in (2000) ^[1]. Crutzen (2002) ^[2]. The working groups of International Union of Geological Sciences (IUGS) though not officially, declared the onset of the epoch Anthropocene, but admitted about the planetary shift of the new geological age. The epoch relates to the word "anthropo" (man) and "cene" (new) where the Homosapiens have dominated the atmosphere, earth crust, ocean, and nutrient cycles.

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Geologically Mesozoic era (the age of dinosaurs), then Cenozoic era (the period of mammals) were succeeded by the Quaternary period without human activities till the tertiary era Fig 1(a). The Quaternary period consisted of the Pleistocene when Homosapiens developed their activities (2588000 to 11700 years BP) succeeded by the Holocene (11700 years BP to present). Present period is the Pleistocene interglacial age accepted by the International Geological Congress in Bologna in 1885, Steffen et. al., (2007) [3]. The Holocene age is classified as early, (11700-8000BP), mid (7999- 4500BP) and late (4500 BP to till date). It prevailed from the warmest period (hunter stone age) after a long ice age during upper Pleistocene period (<http://www.stratigraphy.org/ICSchart>). Thereafter drop in temperature was observed during the little ice age which continued till 1700 AD in northern Hemisphere. The temperature further started rising from 1700 to 2000AD but not like hunter stone age. The Holocene had many small ice ages and warm periods like Minoan, Roman and Medieval with only one prominent cold period i.e. "The little ice age" (<http://www.dandebat.dk/eng-klima7.htm>) Fig 1 (a), Fig 1 (b).

Now the researchers have wide scope for anthropogenic, climatic and geologic studies with ample data which went underreported and non-reported even upto 19th century. Present study reveals the causes, period of prevalence and felicitation of the impact and future effects of the new epoch. The period of start of the epoch, impact of the predictors, parameters, indicators, age and management in Indian subcontinent are discussed in the present research.

Quaternary Period with the Anthropocene Epoch (AWG of ICS, 2016)

Eonothem/ Eon	Erathem/ Era	System/ Period	Series/ Epoch	Stage/ Age	millions of years ago
Phanerozoic	Cenozoic	Quaternary	Anthropocene ¹		1950 ce
			Holocene		0.0117
			Pleistocene	Upper	0.126
				Middle	0.781
				Calabrian	1.806
				Gelasian	2.588

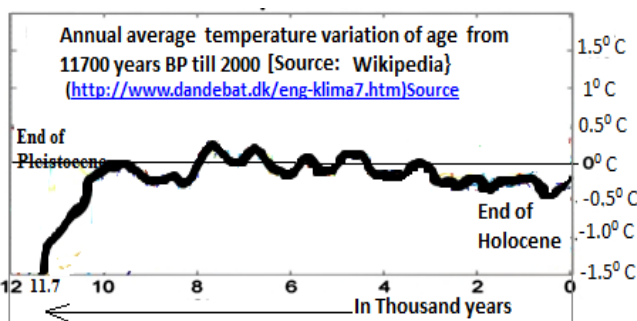


Fig 1(a):- The geological age where the Anthropocene succeeds the Holocene Fig 1(b): The temperature anomaly which shows an average temp change of 0-2^o C brings a drastic transformation in the globe.

Review of literature:-

The Anthropocene entered the geological literature by Paul Cruzen (2002) [2] and further acknowledged by different scientists and engineers like Steffen et al., (2004) [4], Syvitski et al., (2005) [5], Crossland, (2005) [6], Anderson et al., (2005) [7] informing about the end of the Holocene epoch and start of a new epoch where the environment is overruled by human commotion. Anthropogenic activities are the deciding factors in the change of climate, reported by the working group of the Intergovernmental Panel on Climate Change (IPCC). In their assessment report AR3 (2001) [8], AR4 (2007) [9], (2010) [10] and AR5 (2014) [11] have reported the human activities vs. climatic and hydrologic changes (dominant being global warming) are interrelated by 66% (certainly), 90% (very likely) and 95% (extremely likely) from 1950's respectively. Sir David Attenborough, (2006) [12] claimed record-breaking cold waves and rainfall in some parts of the globe disapproves the global warming and made a distinction between the climatic change and weather patterns. WMO have reported 1990's is the warmest century of the millennium.

Stormberg, (2013) [13] has reported that two billion years ago, vast proliferation of the oxygenated cyanobacteria in the atmosphere, forcefully made the living creatures on the earth to an extinct species. Presence of late quaternary strata with youngest Toba Tephra events (Toba super volcanic eruption) in the basins of peninsular rivers, carried valley sediments due to wetted monsoonal phase in the deltas. Hominid fossils (short and stocky pygmies) became extinct in central India during that period. Tandon et. al (2014), [14] Peter Fisher, (2014) [15] rightly stated that progenitors shall have a horrid mechanical future and current species are under mass extinction and we will generate a polluted world by inventing novice chemicals. Bruce hall, (2015) [16] have reported that the Anthropocene epoch shall devastate India as 70% of houses shall be constructed due to migration from rural to urban areas. The wells, rivers and farms shall run dry and be abandoned. India shall produce 4% of GHG gasses of the globe and 40% of population shall have inadequate energy. According to Matt Atherton, 2016 [17], identified five causes for the Anthropocene epoch are Industrialization, population, new materials, Biogenic changes and changing water ways. The Anthropocene epoch started with industrial revolution in early 16th century or as consequences of testing of the first atom bomb in 1940's or from the date of start of nuclear weapons in 1960's. Osborne claims the epoch was

initiated with geologic activities like meteorite slamming, volcanic eruptions, continents shifting, earth quake and the atomic explosions *Hannah Osborne*, (2015)^[18]. The start of his epoch has been a subject of much debate. Some have said it started during the Industrial Revolution from 1760, while others have suggested nuclear weapons testing in the 1960s heralded in a new era in the planet's history "the Anthropocene" Stanley et al (2016)^[19]. Steffen, (2007)³ reported that "The great acceleration" of human forcing on the planet have started a new age 1950's onwards. Davos, (2017)^[61], deliberated 12 parameters of anthropogenic forcing and 12 parameters of environment activity of the Earth's systems responsible for the change in the geographic period "the Anthropocene".

The Anthropocene epoch in controversy:-

To establish the new epoch, selection of a Global Boundary Strato type Section and Point (GSSP) or acceptance of its inception date need be approved by the ICS (International Commission on Stratigraphy) to find a place in GSSA (Global Standard Stratigraphic Age). We are in the Phanerozoic Eon, Cenozoic era, Quaternary period and Holocene epoch as per ICS calendar till date. The IUGS, an official unit of the ICS, is yet to approve the epoch "the Anthropocene" Zalasiewicz et al (2008)^[20], 2010^[21]. The ICS Sub-commission on Quaternary Stratigraphy established a working group in 2009 to address the strategy. Since then, discussion and deliberations on of the Anthropocene became extensive in scientific publications, the public media and in the greater academic sphere. The proposal that the confusion between the definition Chrono-stratigraphic unit as system, series, stage and Geochronologic units as period, epoch, age adopted by GSSPs, as unit, the Anthropocene was rejected in ICS workshop (2010), Zalasiewicz et al. (2014)^[22]. To have a breakthrough to the dialog on the epoch, Anthropocene, congregation of the group is going to meet at Cavallo Point, Sausalito, California from 21st June to 23rd June, 2017.

Anthropocene, Indian scenario:-

The Anthropocene is unofficially accepted with (a) priming period from the age of last warm period from 3000 years BP to 18th century (b) the margin (1850-1944) (C) the fundamental shifts (1945- 1980), (d) The great acceleration (1981 to till date). The geospatial behavior of the epoch had impacts in phases which changed the human activities from hunting and farming to the modern life. Presently it is to be accepted that we are in the Anthropocene epoch in India. The admittance of the new epoch has become more factual than less political today as reported by Zalasiewicz et al (2014)^[23 a & b]..

Establishing the shift in India:-

The Priming (3000BP-1849AD):

Early human impacts on the land use and land cover started from the great warm period, 3000 years BP (Barnosky, 2008^[24], Ellis et al 2011, 2012, 2013, 2014^[25-28] and Wilkinson et al 2014)^[29]. Many terrestrial biotic and marine micro biotic changes altered the CO₂ levels due to socio political developments, conversion from hunting to agriculture. continuous formations in the Himalayan range of mountains, undermining of the river Saraswati, desertification of Thar in India, decline of the ancient civilizations, the Mohenjo-Daro and the Harappa are the artifacts of formation of the new epoch. The pollen grain tests, carbon dating data and strand line records indicate about the reforms of land scape. The conversion of gulfs to lagoons/lakes (the Chilika Lagoon and the Pulikat Lake), marks of high MSL (mean sea level) along coast, conversion of jungles to rural settlements with the knowledge of making metals and alloys indicate about the end of the Holocene era. Archeological findings of underwater monuments at Mahabalipuram, Vet-Dawrika, Ram Setu at Rameswarm etc. relate to the eon changes in Indian subcontinent during the Paleo-Anthropocene period Foley et al. (2013)^[30].

The Margin (1850-1944):-

Sixteenth century is considered to have exhibited noteworthy changes in the world due to continental drive and collision of the meteorological asteroid (The golden Spikes) with major climate changes quoted from Mark Muslim by Rebecca Morelle, (2015)^[31]. The stratigraphic Eurocentric boundary of early Anthropocene reported by Crutzen and Stoermer (2000)^[1], Crutzen (2002)^[2], Water's et al., (2014)^[32] clearly link to the industrial revolution of 19th century with use of the coal based steam engines, shipping and boilers along with the population explosion. The anthropogenic activities shifted gradually to North America, China, India, Japan and many other countries throughout the globe. Industrialization and urbanization are the stratigraphic signals. Energy balance, CO₂ level and devastating famines in India during 19th century are the global indicators to recognize this early Anthropocene. The Q-class NEA asteroids named Apollo 1862, having dimension 1.7 km were the potentially hazardous asteroid (PHA) which along with erratic El Nino southern oscillation activities (ENSO) might have caused meteorological famines in India in the 19th century. The gigantic meteor that slammed Tunguska region of Siberia in 1908 has left the debris and carbon particles scattered in the atmosphere with very high energy which had the impact of huge mass

extinction of species. Erratic rainfall, massive recurrent floods with heavy sediment inflow and huge death due to vectors indicate the drive of the new age in India.

The fundamental shift–Anthropocene (1945-1980):-

The demography, the energy balance, GDP vs. ecological changes, amount of carbon, drastic climatic changes and globalization brought an end to the influence of the Holocene era by mid-20th century (Steffen et. al 2004^[33], 2007^[3], Barnosky, 2008^[24] and 2014^[35], Steffen et al. 2007^[312]. The human impacts which motivated the era from 1945 after the testing of Trinity A-bomb at Alamogordo, New Mexico, 1945 and Hiroshima and Nagasaki in World War II, 1945 brought the fundamental shift. More than 500 atomic tests were conducted then which increased the nuclides in the atmosphere due to the fission and fusion activities. The circulation of the nuclear debris (14C, Pu-239, Pu-240, Cs-137) received from radio activities drastically changed the atmospheric extension and metabolism, Walker, (2005)^[36]. Drilling activities in mines, petroleum wells, oil spills from marine oil tankers, hydrologic interventions, diesel and petrol engines, infrastructural developments, industrial/ agricultural-related signals, uses of plastic, techno fossils are considered triggering as the transit for the early Anthropocene in the globe and also in Indian sub-continent, Zalasiewicz et. al., (2014)^[23], Hancock et al., (2014)^[37], Wolff, (2014^[38]); Waters et al., (2015)^[39], Morrison (2015^[40]), Jain et al (2016)^[41].

The great acceleration:-

The Anthropocene epoch was focused on a small population consisting of the Organization for Economic Co-operation and Development (OECD) countries during the fundamental shift period only. The prolongation of the changes became rapid, homogeneous, consistent and widespread all over the world with the great acceleration trend from 1980 onwards - till date. With reduction of the British territory there was the fast growth of tourism, circuit engineering, science, technology, industry, urban development and economic ties all over the world. The human impact on the biosphere accelerated in the developing countries like India, China, Brazil and Indonesia and many other countries. Terrestrial mini asteroid, Chelyabinsk fire ball, 2012 DA14, orbiting at a distance of 2700 km blocked the solar radiation covered significantly resulting in meteorological catastrophe like chilled Japan during winter Dec. 2012 to Feb. 2013 (NASA). Emphasis on technology, geo-engineering, green engineering, Nano science, genome and dechronification technology and many others have changed the human decree over nature.

Substantiating the Anthropocene:-

Davos, (2014), stated 12 parameters of anthropogenic forcing are global warming, greenhouse gas (GHGs) emissions, polymerization, oceanic acidification, eutrophication, industrialization, urbanization and deforestation etc. The signatures related to Anthropocene are habitat loss, species invasions, mass extinction, predation, damming, GDP changes, and energy consumption and many physical and chemical changes, etc... He also pointed other 12 non-anthropogenic parameters of the Earth's systems which can cause devastation and shift of epoch are the Sun earth geometry, ozone depletion, volcanic eruption, tsunamis, earth quakes, oceanic disturbances, coastal erosion, PHAs, NEAs and biodiversity changes etc.

The Anthropocene epoch can be substantiated mainly in India by massive extinction of species ecologically, the global warming, rise in concentration of CO₂. Global warming has accelerated melting of ice and retreating of glaciers which have changed the ecology and biodiversity of the Himalayan glaciers.

Plastics and building wastes and its layers started forming as stratigraphic layers in soil for the future. A stable layer of airborne particulates of sediment and glacial ice shall accrue black carbon from CO₂. There shall have large impact on the nitrogen, phosphorous cycle due to excessive use of NPK fertilizers. Creation of dead zone in sea due to eutrophication shall occur for the aquatic biome. Conversion of forests to settlements, cropland, grazing zone and livelihood are the signals for the Anthropocene in India Fig 2 (a), Fig 2 (b) and Fig 2(c).



Fig 2(a):-Man pre Holocene epoch (Indian express 2nd Mar 2017), **Fig 2(b)** Man in Holocene epoch (<http://www.livescience.com/28219-holocene-epoch.html>) **Fig 2 (c):** Man in Anthropocene <http://www.smithsonianmag.com/science-nature/what-is-the-anthropocene-and-are-we-in-it-164801414/http://www.livescience.com/28219-holocene-epoch.html>

Are all monstrous disasters anthropogenic?

Table-1 depicts ten major disasters in India and in the globe. Except one, none of the disasters of the world was anthropogenic. They were either hydro-meteorological or geological which negates the fact that the new epoch is human centric. But it has been established that by proper management and adaptation, the impacts from hydro-meteorological or geologic disaster can be ameliorated and fatalities can be reduced. Examples are the great Bihar famine 1966-67, the drought in Maharashtra 1970-73 and the VSCS Phailine- 2013 where the demographic consequences were less.

Table 1:- The monstrous disasters in the globe and in India from past records available

INDIA				WORLD			
year	Name and place	Death Million	Proponent (changes)	year	Name and place	Death Million	Proponent
1770,	The Bengal Famine (Bihar/Odisha/ WB)	10.0	Climate	1887	Yellow river flood, China	0.9-2.0	climatic
1737	Calcutta Cyclone (Bangladesh/WB)	0.35	Climate	1920	Haiyuan earthquake, China	0.273	Geologic
1839	Coringa Cyclone (AP)	0.32	Climate	1931	China flood	1-4	climatic
1876	The Great Famine (India)	30	Climate	1975	Typhoon Nina (Banqiao Dam failed)	0.23	anthropogenic
1993	Latur Earthquake (Latur and Osmanabad)	0.02	Geologic	1970	Bhola Cyclone, India/Bangladesh	0.25-0.50	climatic
1999	Odisha Super Cyclone	0.01	Climate	1976	Shaanxi tremor, China	0.83	Geologic
1876	Great Backerganj Cyclone	0.2	Climate	2010	Haiti earthquake	0.16	Geologic
2001	Gujarat Earthquake (Rajasthan and Gujarat)	0.02	Geologic	2004	Indian Ocean upheaval&tsunami	2.8	Geologic
2004	Indian Ocean Tsunami, South Andaman Nicobar	0.2	Geologic	1920	Haiyuan earthquake	2.73	Geologic
2013	Uttarakhand Flash Floods, India/ West Nepal	0.05	Climate		Tangshan earthquake	2.42–6.55	Geologic

Footprints of Anthropocene in India:-

Burgeoning Demography:-

The analysis of population growth rate in the world and India have shown continuous rise from 1960-70 onwards, the annual population growth is declining from 1960-70 onwards till date. The population growth of poultry have

risen abruptly with the demographic growth of people where as growth of other live stocks is slow as against human growth. The impact of demography in the earth, India and population variations of the live stocks are in Fig 3 (a)

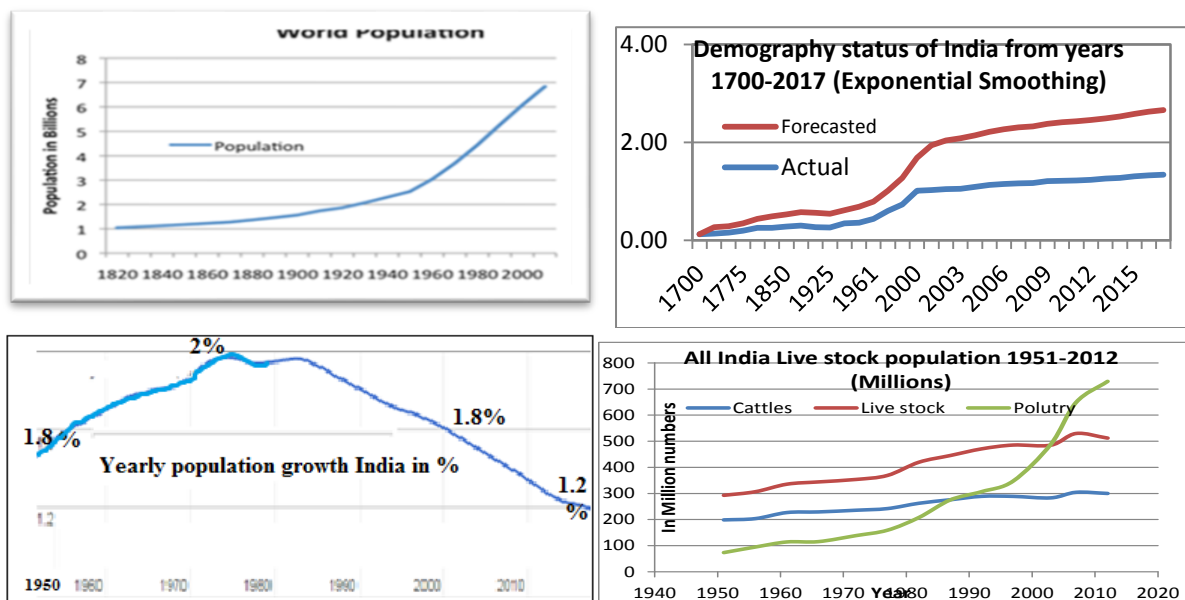


Fig 3(a):- The world, Fig 3 (b) Indian population and Fig 3 (d) livestock population, India from the year 1950 -2017

Fig 3 (c) the rate of population growth from 1980 onwards (source: <http://www.populstat.info/Asia/indiac.htm>
Source: <https://ourfinetworld.com/2012/03/12/>

Hydro-meteorological extremes:-

The Climatologic indicators for the Anthropocene are GHG's, water, Waste, Biomass, LU&LC, vectors, microorganisms, transport and energy whereas the observed events are Cold wave, fog, snow storms and avalanches, hailstorm, thunderstorm, smog, dust storms, heat waves, avalanche, lightening, heavy rain, floods, saline water intrusion, droughts and many others. The global climatic observing system (GCOS) and their essential climatic variables (ECVs) are the watchers recording about 50 atmospheric parameters (Surface, upper air and compositions of GHG's, ozone layer, aerosols and reinforced by their antecedents), Oceanic (Surface and sub-surface and terrestrial). The sectors where impacts observed are ocean, rainfall, temperature, biodiversity, health, soil, agriculture, disasters and extreme events.

Rainfall/snow fall:-

Monsoon rainfall in India from post Holocene and pre Anthropocene, from year 800 to 2200 AD (modeled) Fig 4 (b) shows the rise and fall. After 2020 during Indian South west monsoon shall decrease Fig 4(a). There shall be increase in rainy days and decrease in frequency of depression and SW monsoon annual rainfall particularly in Himalaya region Fig 4(a). The peak rainfall shall decrease in NE India which was giving maximum rainfall in India (Cherapunji). Rainfall in peninsular India shall decrease. Summer Monsoon in India may have abrupt transitions and fail due to climate change Jacob et al, (2012)^[42], Leverman A (2009)^[43]. The western disturbances originating from Caspian Sea have increased in frequency, intensity and duration during the epoch.

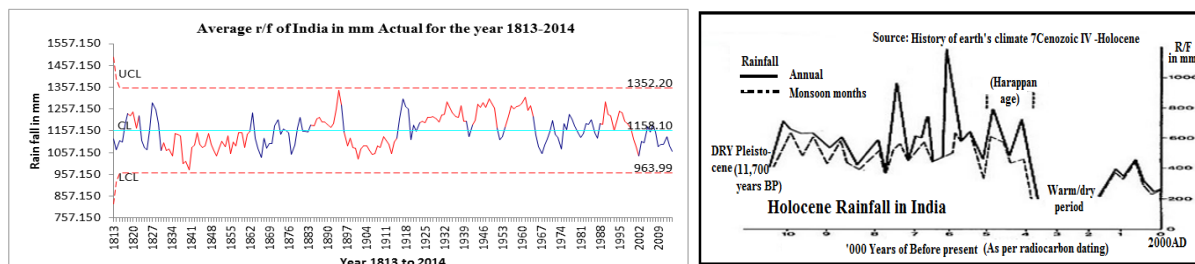


Fig 4(a):- Monsoon rainfall in India (year 1813 to 2009 AD) Fig 4 (b): The monsoon and annual rainfall of western India in Holocene period (radio carbon dating). (source: H. H. Lamb, Climate, History and the modern world)

The storms, floods and droughts:-

The storms, earth quakes, tsunamis and famines are the monstrous killers in the Indian subcontinent during Anthropocene. 30% and, 25% of cyclonic storms formed in of Bay of Bengal and Arabian Sea respectively have risk for 32crores of coastal people. The new epoch has invited urban floods which engulfed the cities like Calcutta, Bombay, Chennai and Bhubaneswar in great acceleration period in India due to impervious concrete jungles and inadequate drainage system. Poor people are sustainable to high impact due to increased floods and droughts. WG I, IPCC, AR-5^[11] reported that India shall face mortality of 86% from tropical storms. The SLR and storm impact can enhance significant vulnerability with climate changes. Droughts and famines are the signals of the end of Holocene in 19th and 20th century (fig 5 (a), 5(b) and 5(c)).



Fig 5(a):- Sup. Cyclone Odisha 1999 (b) Bhopal gas woes (1972) by S.N. Sardi (c) Extinct fishes-2016 M. Snyder

Global warming:-

World Bank Group pointed about prevalence of historical warm climate from 1970-80 and predicted that 4°C rise shall have substantial impact on agricultural yield in India. Deccan plateau shall form zones of “Heat Islands” triggering frequent floods and drought. Wet year shall be wetter and dry years shall be drier. IPCC (AR-5) estimated small average change in SAT and SST of the earth during 1906 to 2005 the change was 0.74° C. About 90-95% of global warming is transferred to sea. The National Oceanic and Atmospheric Administration (NOAA) have fixed seven increasing indicators (SST, SAT, Ocean air temperature, MSL, oceanic heat content and inland temperature) and three decreasing indicators (glaciers, snow cover and icebergs in sea). The critical high temperature shall shift to Oct in northern region and in south India, to pre-monsoon (April and June). The early paddy shall have bumper yield in south India, and devastate coral reefs by bleaching action Koronowski, (2016)^[44]. It is observed that the rise of 5°C in SST shall cause SAT rise of 2°C inland Fig 6 (a) and (b).

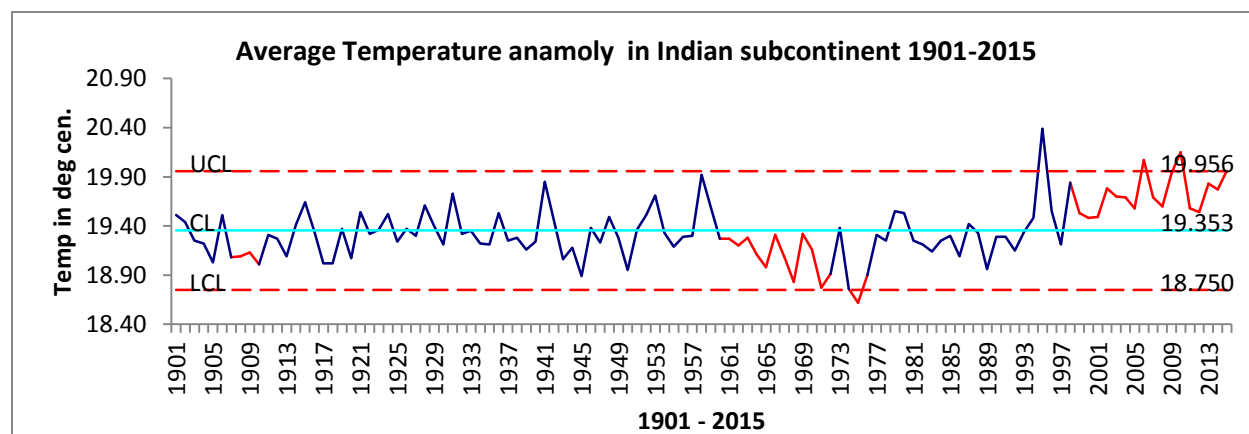


Fig 6 (a):- Average temperature anomaly of Indian subcontinent for the period 1901 to 2015

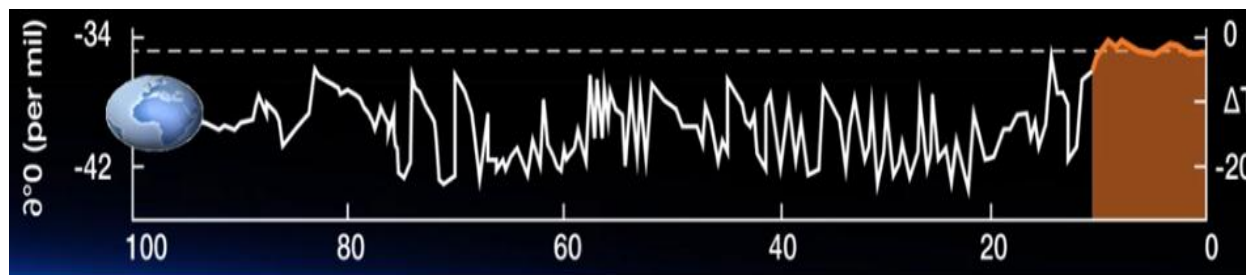


Fig 6(b):- Temperature anomaly of earth in K-years before present (Johan Rockstrom, Stokholme University)

Heat waves and Lightening:-

There is dynamic rise in temperature with a maximum in the year 2012 in 21st century. The ten top ranked Heat Waves death in the globe's history in 21st century are 71130 in Europe (2003), 55,736 in USSR (2010), 3,418 in Europe (2006), 2,541 in India (1998), 1,826 (probable) in India (2015), 1,693 in U.S. and Canada (1936), 1,260 in U.S. (1980), 1,210 in India (2003) and India 1,030 in (2002). The figure indicates that frequent heat waves have hit India in the early 21st century. However India had the hottest year, 2016 and the hottest day with highest mercury of 51°C in Rajasthan at Phalodi on 23rd May 2016. Lightening in stratosphere, electro-dynamic anomalies and thermal irregularities have increased the trend of heat waves and lightening deaths in India (Fig 7).

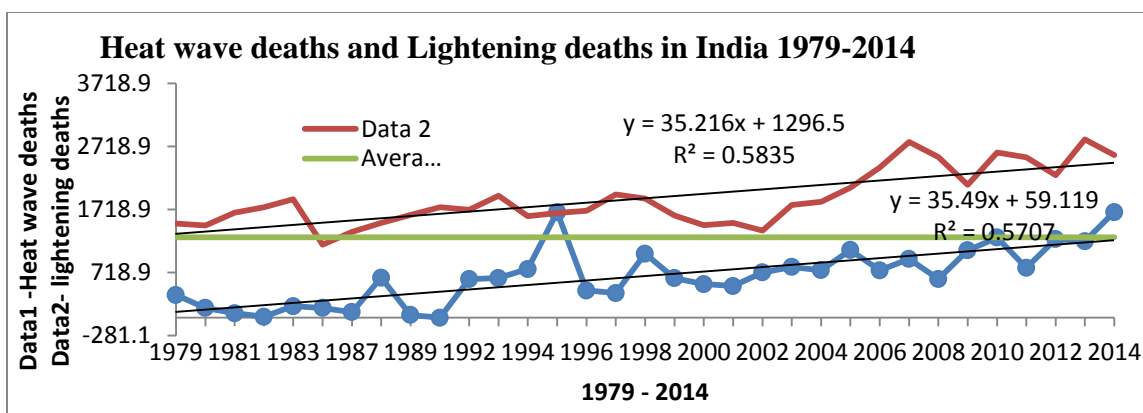


Fig 7:- the heat wave and lightening fatalities are having rising trend from year 1979 to 2014

Tornadoes and thunderstorms are common during pre-monsoon in central, north and north-east India causing loss to properties, crops and fatalities which is related to the acceleration of the new epoch. The highest recorded mortality was 500 in tornado at Dhaka in 1977 and casualties of 6000 people.

GHG gasses:-

CO₂ is one of the major greenhouse gases (GHG's) that traps heat in the atmosphere which reached maximum record in 2012. It is continuously accelerating and drifting upward which can drive hydro-meteorological changes and will bring variations in our planet in future during the new epoch (WMO report 2013). GHG's are natural and essential for humans. Methane (CH₄), carbon dioxide (CO₂), oxides of nitrogen (mainly N₂O), hydro-fluorocarbons (HFCs), per-fluoro-carbons (PFCs), trifluoro-methyl-sulphur-penta-fluoride (SF₅CF₃), sulphur hexa-fluoride (SF₆) and Water vapor are the main causes for global warming. Since 1950, India has produced roughly 4, 2 billion tons of carbon respectively. The annual generation of carbon per capita in India was 1.9 tones which was less than that in China and USA but it had increased three times since 1990 and doubled by 2009 to 2014.

Effects of CO₂:-

The natural carbon cycle is slower than the human-induced climate changes. According to Dana Nuccitelli (2016), O' Day^[45] from NOAA, CO₂ level reached maximum in the year 2015. The residence time of CO₂ is 4-15 years, which is very short (IPCC). Then reason for short residence time of CO₂ in the atmosphere and increasing trend negates the fact of its increase in the atmosphere contradicting the increase carbon level. Cawley Gavin^[46] reported that the temperature of the earth have risen to 1.5–3°C. It is estimated that the rise in temperature on earth

surface shall be warming the atmosphere over the coming decades and about 30 billion tons shall be pumped by the earth to warming over the coming decades and pumping about 30 billion tons of CO₂ shall be pumped by the earth to the air every year. Debating on CO₂ emission and climatic change, the Climate Commission, Australia, reported their climate change in 2001-2010 of 21st century as 'critical'. In India and in the globe the CO₂ emission is raising gradually Fig 8 (a) and Fig 8 (b).

GHG gases are classified as brown (industrial) Green (from inland vegetation), Blue (oceanic biomass) and black (incomplete combustion of fossil fuels) Leahy Stephen, (2007)^[47]. The status of CO₂ was @ 379ppm in 2005 whereas it was 283 ppm before the industrial revolution i.e. in 18th century. Concentrations of CO₂ in the atmosphere were 142% higher than what they were before the Industrial Revolution (WMO greenhouse gas bulletin-2013). In this new age, the CO₂ level shall be high to give many marine organisms less time to adapt. Some marine creatures are growing thinner shells or skeletons that will play a crucial role in the biodiversity and in food chain.

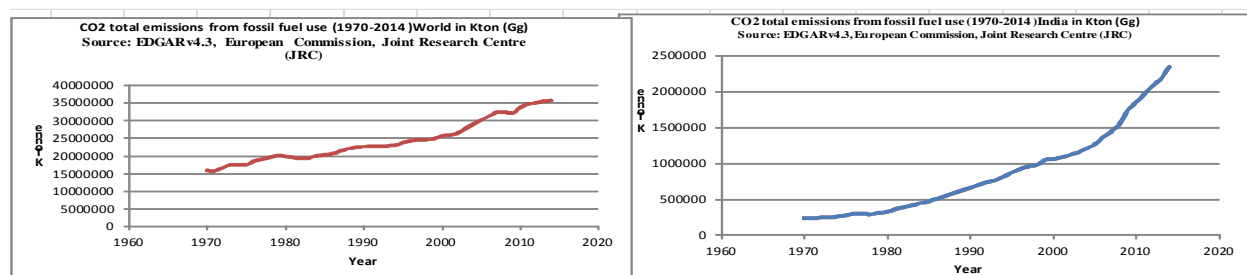


Fig 8 (a):-Global CO₂ emission global annual trend Fig 8(b) CO₂ emission in India annual trend (1970 to 2014)

Geohydrology:-

Geohydrology potential in ground water table (GWT) in India have changed due to over exploitation as 60% land is rain fed. Fifteen percent of India's groundwater resources are overexploited without retrospective recharge. This shall invoke subsidence. Growing population, comfortable life styles, modern utilities, services and industries demand shall urge to draw more GW with less recharge hence more depletion and increased salinity intrusion along coast.

De-glaciation:-

Westerly Jet dominated NW Himalayas and Karakoram ranges are stable. The NE Himalayan glaciers are retreating due to erratic SW monsoon and global warming. At 2-4⁰ rises in temperature, this shall increase discharge in Ganga, Indus and Brahmaputra. Pervasive mass losses in Himalaya and drops in snow cover in glaciers shall accelerate during the 21st century and cause water scarcity, hydropower deficit, and variability in seasonal flows from Hindu-Kush and Himalaya in India (IPCC AR -4). The rivers from Himalaya's shall enhance the frequency and intensity of floods affecting the huge coastal population of 62 million. The ice mass in poles is melting and sheet moving to warmer region since last two decades losing 215 Giga tones annually and add to mean sea level rise along the coasts (IPCC AR-5). The huge sediment reaching the deltas shall protrude and distort the Ganga-Brahmaputra delta prominently during great acceleration of Anthropocene.

Mean sea level rise:-

High emission, movement of ice sheets in a warning region, melting of ice in Himalayan glaciers and the arctic region are signs of the age. Summer urban flooding along low lying coastal areas has been aggravated. There has been change in configuration and dimensions of estuaries/lagoons and positioning of their tidal inlets. The coastal erosion, accretion and submergence have become rampant. The World Watch Institute (2000) has reported that the influences are due to land ice melting (42%), ocean warming (35%) and 12% from inland water.

Coromandel coast line in India has exhibited four to five strand lines during the period of Holocene. Ministry of Earth Sciences, India is focusing on MSL rise as there was increase of submergence, erosion and coastal flooding along the coasts of India. Saltwater intrusion and back water propagation have increased both surface and ground waters. It has impact on mangroves, salt marshes and sediment stratigraphy in lagoons. The SLR reported by Nicholas (2010) is @ 3.3 ± 0.4 mm/year in the globe. But in Indian scenario the SLR was 1.2, 1.75, 1.09 and 5.74 mm/year along coasts of Mumbai, Kochi, Visakhapatnam and Kolkata respectively (Unikrishnan et al ,2006^[48]).

Both the west and east coast of India have been worst affected by disturbances and storm surges Fig 9, Shetye et al, (1990)^[49] and Mishra et al (2014)^[50]. Submergence from flash flood and coastal erosion has made the life of poor worst in islanders of India.

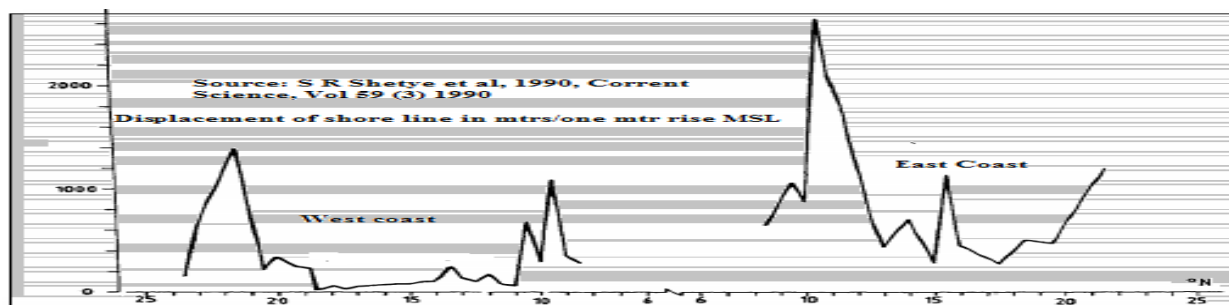


Fig 9:- Shore line shift of east coast and west coast of India per one meter rise of mean sea level

Bio-centric Anthropocene:-

Deforestation:-

Deforestation goes on unabated in tropical forests of India. But study reveals that greenness of vegetation is rising across the globe capturing about 750 MMT of CO₂ annually and reducing 24% of world's GHG emission. 33.33% of India's tropical deciduous forests shall become evergreen due to heavy rainfall in patches of high temperature zone IPCC AR-5 (2014)^[51]. Deciduous forests shall accelerate but growth shall slowdown by demographic pressure.

Pollution of water air and soil:-

Polluted and contaminated air, water and soil and climatic changes shall enhance the risk of water borne and vector borne diseases, mental disorders, post traumatic syndromes, more heat strokes and disaster prone mortality rates. Economic climate resilience actions, the storage reservoirs and integrated basin management can ameliorate the problem.

Ocean acidification:-

About (30%) CO₂ is dissolved in ocean and increases water temperature and decreases alkalinity of ocean water: $H_2O + CO_2 + CO_3A = H_2CO_3$. NOAA-2010 have reported that more acidic sea water and the carbonic acid formed shall disturb the ability of marine plants and animals to thrive, <http://www.noaa.gov/resource-collections/ocean-acidification>. Recently micro-planktons in the ocean are responsible for the sequestration of 25–50% of the carbon which is cumulating ocean acidification. The plankton plays vital role in atmospheric carbon dioxide (CO₂) fixation at lower levels. Otherwise oceanic acidification would have been a peril to environment. World Bank has predicted that 1.4°C rise from present temperatures (in 2030) shall reduce coral growth and at 2.4°C shall either dissolve or dry up them. <http://www.Worldbank.org/en/news/feature/2012/11/18/Climate-change-report>. In India risk zones have developed and shall proliferate to the Andaman, Nicobar and Lakshadweep Islands, Palk Bay and Gulf of Manner, and all lagoons along the coasts.

World Hunger and Food Security:-

Increased droughts and desertification have adversely affected agriculture and livelihoods leaving aside warm weather crops. Melting of glaciers is worsening floods in north India. Rice cultivation is less productive in Nepal forcing the farmers to change their cropping pattern. Advanced agriculture has encouraged swapping of crops by replacing genetically modified crops in last 30 years to combat the food security. Economic and technology advancement for growing population in India has increased average rice production (6% higher i.e. 75MT/Ha) whereas the wheat production in north India have reduced because of rise in temperature from the year 2000. Rise in temperature, intrusion of saline water in coastal zone and erratic monsoon have reduced crop yields and enhanced food insecurity. India shall have agricultural loss of 10% to an extent US \$7 billion if adequate climatic resilience measures are not taken. There shall be stress on food specially the wheat production IPCC AR-5, WRG II, (2014).^[51]

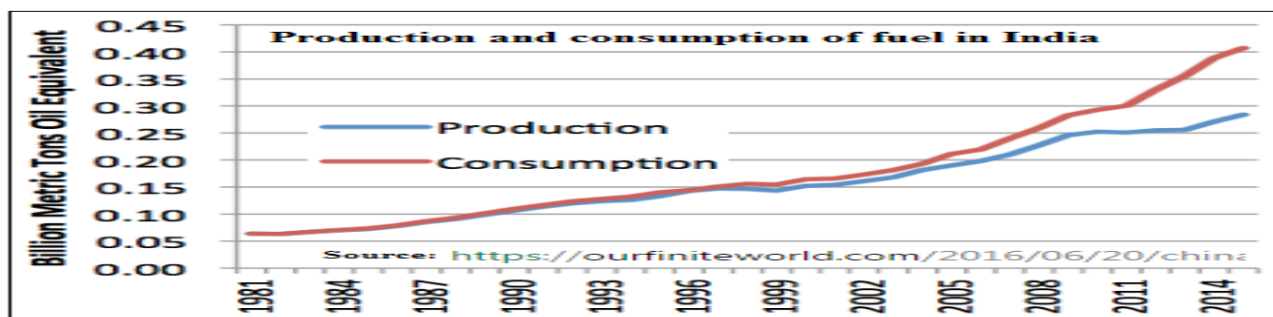


Fig 10:- Production and consumption of different fuel in India between the years 1981 to 2014

Energy and water security:-

Electrical energy of 92378.38 MW (thermal), 4560 MW (nuclear power) and 37567.40 MW (hydropower) was generated in India (CEA, 2010). Paucity of water in rivers has reduced hydropower generation. The mushroom growth of thermal power plants have increased CO₂ level and affected energy security. The commercial energy consumption in India is less in comparison to other countries. It had risen from 1965 by 9% whereas it was 19.4% in 2000 (Tata Energy Research Institute report, 2000). In the process of great acceleration there shall be diminishing supply of fossil fuels which should exacerbate production and use of biofuels. Energy demand is higher than the production, and crisis will be more in India in the Anthropocene (Fig 10)

Erratic monsoon, demographic growth, urbanization and industrialization could not satiate growing demand of water and augment water stress. Erratic SW-monsoon shall bring more floods in Sept whereas longer dry spell in the month of May shall affect the fresh water availability from peninsular rivers. Drinking water shall be so precious that apprehension of third world war may be due to water.

Synthetic Life:-

Recent advances in molecular and genetic researches could develop artificial genome; bacterial chromosomes which can replace DNA in a bacterium producing new protein sets which can bring revolution in genetic engineering as part of the great acceleration where life span shall be multifold, Venter J. C. 2010^[52]. Now the Institute of Genomics and Integrative Biology can produce genomes. The age of synthetic life has started in India.

Health hazards:-

Major health impact due to climatic disgrace, population growth and anthropogenic activities shall enhance pediatric mortality, Vector borne diseases (especially due to mosquitoes), Hearts and lungs diseases, infectious and non-communicable diseases Vaughan et al (2016)^[53]. The endemic, outbreaks and epidemics throughout the land and ocean based wildlife have become unwarranted due to disruption of natural ecosystem. Pests, Vector diseases and immunity based diseases have aggravated both among the humans and the wild life. The contaminated water and stagnated urban floods shall exacerbate diarrheal diseases but effective management shall reduce the DALYs rate Singh et al 2012^[54]. Some new genera of malicious vectors and microorganisms have come up in the epoch which shall cause unnatural deaths and many species will be endangered or extinct. Use of hormones for early high yielding grains, vegetables and meat shall make the human based Anthropocene to be apocalyptic. Revkin Andrew, (2011)^[55] told that a type of Cyanobacteria had put an end to life and species 2 billion years BP in past.

Socio-economic-political:-

Socio economic conflicts, poverty, segmented demography have forced the Indians to a continuous brain drain to western countries since 1980's. Regional cooperation on interstate and intercontinental disputes with raised demand for water have started from 1970-1980. Some crucial turns have resulted in man-made political and economic issues. Global trade policies are making the poor to be poorer. There shall be 0.2 to 2% loss to annual global income on 2°C rise in temperature. But higher increase can bring calamity IPCC WR II, AR-5, (2014).

Migration, education and skill:-

India is switching from agro-base society focused on production to a consumerist society. The impact of migration, education and skill has brought us the social mobility, both linear vertical and spatial. Migration changes life style,

GDP and the emission. The migration from rural to urban areas has been studied and the rate of migration was 314.5 million as per 2001 Census in India in quest of education and skill development which is a very high figure.

Gender based changes:-

The women in India have less resource to cope and adapt with the climate and biologic reasons than men. They are deprived, poor and unequal which undermines their social and capital stand in the orthodox family in India. There is increase in gender based violence where women are the worst vulnerable. The unprivileged women are more in rural settlements than in urban. The gender ratio in India in last few decades may be one of the reasons for decline of population.

Gross Domestic Product:-

Global energy-related changes, CO₂ emissions and the influencing factors for climate changes as per KAYA identity (1971 to 2009) are at 79% population growth increase in per capita GDP and energy emissions are 106% and 105% respectively. Increase of GDP of India (Fig 11) from year 1985-86 with higher increase in emissions of CO₂, establishes the fact that the Anthropocene is in vogue in India. (Source: IEA loc.cit.http://www.feasta.org/wp-content/uploads/2013/02/Economic_growth_Population_growth.pdf)

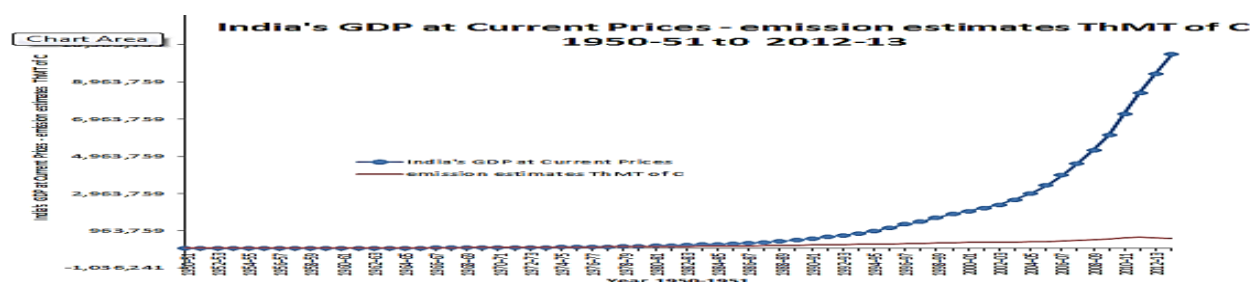


Fig 11:- GDP growth of developing country India for the period 1950-2013 and 1986 is year of increase

Increased GDP growth (Fig 12) and retardation of rural settlement shall increase urbanization and industrialization which shall augment GHG's emission. Gross domestic product in India and its nominal values and purchasing power parity have been studied and the base year for calculation of purchasing power was considered to be the year 2000 which is the point of inflection of the curve Fig 11.

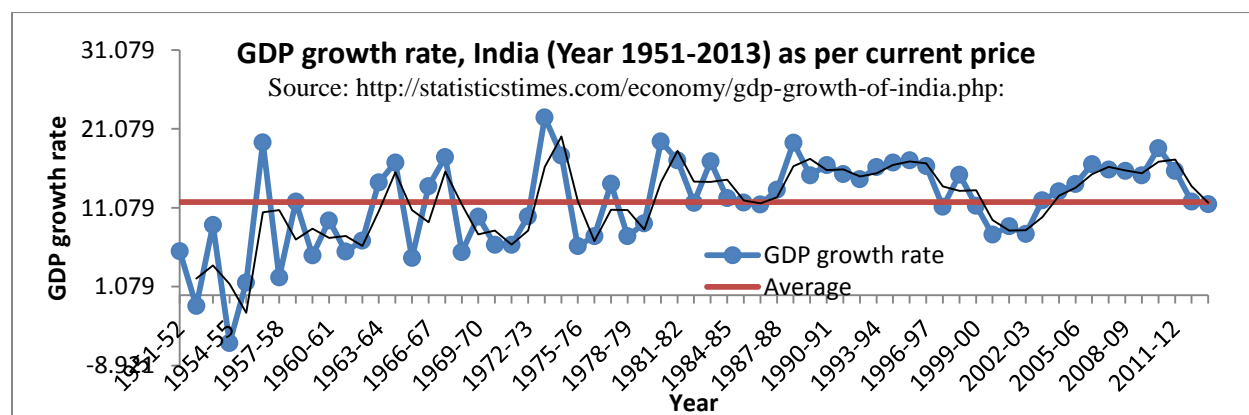


Fig 12:- The time series and moving average curves for GDP growth of India (1951-2013),

Geologic Evidences:-

Instantaneous causalities and fatalities only occur due to earthquakes, tsunamis, landslides and volcanic activities. First 2 decades of 21st century have signaled that the living kingdom have to pay huge losses due to geological extremes. India is safe towards volcanic activities as there is only one active volcano in Andaman Islands. But the impact of earth quakes, tsunamis, limnology, and lithology changes are common in Indian subcontinent.

Limnology:-

Limnology changes have disturbed the fauna and flora of water bodies, reservoirs, lakes, back waters and lagoons other than bay of Bengal and Arabian sea of India in post-Holocene, pre-Anthropocene and even during the great acceleration period. The physico-chemical parameters, rising of bed, squeeze of flood plains of rivers and sedimentation, reduced stratification and downsizing by volume of lakes/lagoons are observed. Dal lake Kashmir, Pichola lake Udaipur, Vena Lake Mahabaleswar, Koleru Lake (Andhra Pradesh), Chilika and Pulikat Lagoons were affected during the great acceleration of the Anthropocene. Construction of dams, barrages and weirs during the period of Anthropocene have retained the sediment at back and the deltas of India are in a state of sinking, shrinking and subsidence (Sivesky et al, 2009^[56], Gupta et al 2012^[57], Mishra S P, 2016^[58]).

Lithology:-

Out of 5200 types of minerals, (International Mineralogical Association), 208 minerals are of human origin formed due to great oxidation events in dump yards of mining area, smelter plant's inner walls, weathered slags, mine fire zone, walls of tunnels and debris of plastic wastes. Artificial stones, wastes from building demolitions, industrial scarps, cinder blocks and toxic radioactive byproducts have formed layers and shall be the lithological imprints of future as Anthropocene minerals. Plastic debris and trashes on beaches resulting from human activity and Tsunami activity shall form with sediment, fragments of basaltic lava and organic wastes/ shells to form a new rock. Clastic plastiglomerate which will remain as a stratum and can be a future earth's rock record, as a geographic mark.

Sea water intrusion due to decreasing inland flow has altered salinities in the deltas/estuary soils. The elite, chlorite, and Na⁺ contained montmorillonite soil have increased and Ca⁺⁺ Na⁺ mixed montmorillonite soil has decreased. Na⁺ mixed montmorillonite is major soil in swamps and elite in lagoons/ backwaters of the soil in east coast of India is major paleo changes during Anthropocene.

Seismicity and Tsunamis:-

The number of earthquakes of magnitude >7 in Richter scale has occurred in the 21st century till 10th Feb 2017 in the globe. The major earth quakes that had caused heavy fatalities and casualties in 19th, 20th and 21st century were of magnitude >7.0 in Richter scale. India was under the attack of the tsunamis from mid of 17th century in record. The intense upsurges recorded were 12m and 10m in case of Makran Tsunami 27th Nov 1945 and Sumatra tsunami 26th Dec 2004. The seismicity can alter an age or epoch because of its devastating impact as told by geologists in great dying period 200-250my BP. The extreme events during Anthropocene are in Table 2.

Table 2:- The changes and extreme events in different stages of the Anthropocene epoch in Indian Subcontinent

Decade	Changes / extreme events	Deaths/casualties	Losses	Remarks
2000-3000 BP	increased rainfall, cooler & humid climates, the beech forests	The civilization collapsed	Decline of Harappa /Mohenjo-Daro age	
1700-1850 AD				
1769-70 AD	Great Bengal Famine	10 million	Loss indigo plants for dye and opium	Manmade(Wiki pedia)
1783-84	Chalisa famine	11 million	poppies destroyed	
1791-92	Dojibara famine	11 million		
1819	Gujarat Earthquake	0.02 million		
1837-38	Agra Famine	0.8 million		
1851-1945 AD				
1860-61	Upper Doab famine	2.0 million	I Punjab/ Rajputana	https://en.wikipedia.org/wiki/Timeline_of_major_famines_
1865-67	Orissa famine	1 million	Bihar; Bellary and Ganjam	
1868-70	Rajputana famine	1.5 millions		
1881-90	Odisha famine	1.5 million		
1896-97	Indian famine	5.0 millions	Richter Scale 8.3	In British territory
1897(12 th June)	Assam earthquake	0.0015 millions		
1899-1900	Indian famine	>1.0millions		
1905 (4 th April)	Kangra earth quake	0.0019millions	Richter Scale 7.5	In British territory
1951-1980 AD				
1961-70	1970 Bhola cyclone	0.5million	\$86.4 million (1970)	Bangladesh
1971-80	Cyclone Andhra, India, 1977	10,000 people	40,000 cattle deaths	
1980-2017	The great Acceleration			
1984,19 th Oct	Bhopal Gas tragedy	20000 deaths	5lakhs affected	

1993 (29 th Sept) 1998	Latur Earthquake 1993 Super cyclone Odisha, India 1999, Heat waves	7,928 people died 0.002millions 0.0013millions	30,000 injured Max mercury reached 45 to 50 ^o C	Prominent, El Niño Transition (ENSO), 1998
2001 (26 th Jan) 2004 (26 th Dec) 2005 (24 th July) 2005 (8 th Oct) 2010 2010 2010 2010	Bhuj, Earth quake, India(M 7.6) Great Sumatra earthquake and Tsunami (M9.3) Teressa Island, Andaman (M7.3) Kashmir earthquake (Pak/ India) Asian Monsoon displaced west The snow retreat and recorded snow melt reduced Cancer death (age 30-69)	20000 deaths 280000 deaths Least deaths Well adminstered 74661 deaths 138,366 people 15,000 people 1600people 5,56,400 people	0.63million affected 10m high surges, land form distorted No reported trauma snow area by 4.9million sqkm	intra-plate area Prominent, El Niño to La Niña Transition (ENSO), heavy rainfall R. Prasad, http:// www.thehindu.com/s ci-tech/health
2011-17	Uttarakhand/ H.P., Rain/ Flood India 2013 Hiayan typhoon, Philippines,2013 But Phailin storm India, 2013	4094 people 10,000 dead but 21people(only).	> 9 million affected >9million affected	

PHA's and NEO's:-

According to NASA, exo-planet Alien asteroids of smaller size (<25m) get burnt in the Earth's hemisphere and cause inconspicuous damage. Meteors of size (>25m) slam earth at a frequency of 2,000 years or so and damaged an extensive area significantly. Asteroids of very large size are hostile to mass extinction and earths civilization (https://www.nasa.gov/mission_pages/asteroids/overview/fastfacts.html). The impact and the potential effects of potentially hazardous asteroids (PHAs) or near earth objects (NEOs) are location specific and caused damages depending upon angle of hit, velocity of slamming and the size of the alien Table 3.

The Permian-Triassic asteroid in 200-250 million years BP and the Hephaistos or SL9 of 65-66 million years BP are responsible for the mass extinction during transit period of Permian–Triassic (P-Tr) eon and Cretaceous and Tertiary (K-T) era and believed to end the Permian period and cretaceous period by destructing 90% of species and delaying the process of evolution by millions of years.

Table 3:-List of exo-planet Alien, PHAs and NEO pre Anthropocene and post Anthropocene disasters

Major alien asteroids slammed earth before 2016					
Year	Place of strike	Boundary of era	size	Effect	
200-250 my BP (great dying)	Controversy,	Paleozoic to Mesozoic Era, Triassic (P– Tr) extinction event (fullerenes/buckyballs)	9Km(Global warming, 90% mass extinct 70% -terrestrial vertebrate	G. Jeffrey Taylor, http://www.psrdr. hawaii.edu/Feb01/perm ianImpact.html
65-66 my BP	Yucatan Peninsula, Chicxulub crater, Mexico	Cretaceous–Paleocene extinction event. (Hephaistos or SL9)	10Km	Global warming, dinosaurs large Extinct	https://www.psi.edu/ep o/ktimpact/ ktimpact. html
3.3 my BP	Argentina	Cretaceous-Tertiary (KT) extinction	37–58 km	extinctions/ global cooling trend	
50000years BP	Arizona (N)	Pleistocene 180 m deep, 1.2 km diameter bowl- shaped depression	30m	impact of a 100,000-ton iron- nickel meteorite	https://astrogeology.usg s.gov/facilities/meteor
1490AD	Chiling- yang(China)	Huge number of meteorites of stones. covered 4-5 cities of China	Scatte red stones	10000 people death	http://articles.adsabs.har vard.edu/full/1994Meti c..29..864Y/0000867.00 0.html
1908	Tunguska, Siberia, Exploded 10km above ground	Breaking up of large meteorite (NEO), 50m, 2150 km ² ruin		releasing energy in the atmosphere 10- 15 megatons	http://news.nationalgeo graphic.com/news/2007 /11/071107-russia-

Major anthropogenic fatalities due to population explosion are accidents, stampedes, wars, Naxalite activities and many such. Those major fatalities were absent during Holocene epoch in India.

Present and future impacts of Anthropocene epoch in India:-

Present globe is influenced by human agents overriding the catastrophic nature and extremes of sun earth geometry. If present trend continues, 75% species will become extinct in the next few centuries on the earth Vaughan (2016)^[53]. The new age Anthropocene is still under debate as to whether it is a part of Holocene or a new one.

Melting of ice in Himalayas due to high temperature shall increase summer flow in perennial northern rivers like Sindh, Ganges and Brahmaputra and many others. Flash flood in Kedarnath (2013) was due to over topping of Chourabari lake killed 5700 people. The 427dams proposed to be constructed in Himalayas may extinct the species, increase more landslides and flash floods in Siwalik range and lower Himalayas.

Outbreak of vector, immune deficiency diseases, effect of pests and water borne diseases shall increase during Anthropocene (Ex: malaria, kala-azar, dengue and chikungunya Japanese encephalitis etc.. Some deadly diseases of Holocene epoch shall be completely eradicated in India. The disability-adjusted life year (DALY) is the years lost due to disability, ill-health, or early death shall reduce in comparison to the globe by 46.6%, Singh et al (2012)^[54]. Smoking, drugs, and alcohol shall certainly add to the DALY rate.

High yield of low paddy due to improved genetic advancement and less production rate of wheat is highly risked. The insufficiency in agricultural yield, milk and protein foods due to increased demography shall have negative impact on human health leading to malnutrition, if not addressed in time. Medication for premature growth shall be apocalyptic.

Biodiversity impacts: A large number of invasive species creep into the water bodies, lagoons and marine system causing myriad impacts on human health. Marine water snails devastate the vegetation in the coast and transmit schistosomiasis, vectors proliferate diarrheal diseases. Coastal erosion and low lying area inundation likely to affect poor people by *Anopheles sundaicus* (Diptera: Culicidae). A brackish water breeder mosquito vector of malaria shall widespread which was confined in Andaman and Nicobar Island only. MSL rise shall affect most the 7500km of Indian coast line which has started from 2004 from the east coast. Amphibians are becoming extinct and there shall be global homogenization of flora and fauna. Mammals extinct by 21st century in India are Mammals Asiatic cheetah: Extinct from India (*Acinonyx jubatus venaticus*), Shanghai, Namdapha flying squirrel (*Biswamoyopterus biswasi*), Himalayan wolf ("*Canis himalayensis*"), Elvira rat (*Cremnomys elvira*), Andaman shrew (*Crocodyra andamanensis*), Jenkins' shrew (*Crocodyra jenkinsi*) and Nicobar shrew (*Crocodyra nicobarica*).

India's flora, Fauna, avifauna and aqua-fauna: Beautiful Indian birds, either extinct or endangered species in Anthropocene era are Great Indian Bustard (India), Red Headed Vulture, Spoon Billed Sandpiper, Jerdon's Courser, Bengal Florican, White Bellied Heron, Sociable Lapwing and many others. There is also a long list of endangered species of flora, fauna, avifauna and aqua fauna in India today.

Many tribes (Homo-sapiens) in India and also in the globe are Rabari of Gujarat, Andamanies in Andaman and Nicobar islands, Nenets in Russia, Kalash in Pakistan, Batak in Philippines, Maori in New Zealand, El Molo in Kenya, Dukha in Mongolia, Goroka in Indonesia and Papua New Guinea, Piraha in Brazil and many other tribes are facing extinction.

The life of the epoch "The Anthropocene" :-

Life on our mother Earth evolved about 4 billion years BP, due to chemical action of air, water and various rocks Hazen (2006)^[60]. According to reports of NASA, three PHAS and NEOS, 2016WF9, 2015BN509 and 2017BS32 are estimated to strike earth in near future which can cause mass devastation. Boffins at the US space agency, the steroid Asteroids with lower-inclination orbits are easy for reach and likely to hit earth. (<http://www.dailymail.co.uk/sciencetech/article-2145699/New-Nasa-sky-scan-reveals>). A ~ 11-25m asteroid (2017 BS32) will burn up during its entry to our atmosphere and the other two shall have less impact. Very large number of PHA's and NEO's are on line for collision process with the earth system – but the epoch Anthropocene shall safely dominate for the next 1.35million years till collision of Gliese 710 or so Bersky et al, (2016)^[59]. But the scientists have planned to bombard and explode the earth hitting PHA's and NEO's before collision and keep the earth from the apocalyptic.

Andrew Revkin, (2011)^[55] reported that a type of unknown killer cyanobacteria oxygenated the atmosphere and extinct most of the species and can put an end to the biodiversity 2 billion years ago.

Management of Anthropocene epoch:-

Geologists and anthropologists have accepted about the fifth extinct in the globe. The sixth extinction of India and the globe is foreseen. Influences of the acceleration of climatic changes in the Anthropocene have adversely affected the underprivileged but they can have chances of survival with non-genocidal activities. The causes for main extinction are the concrete and fertilizers (adding enormous P and N to soil), sinking and shrinking of the deltas (dams and hydrologic interventions), Biomedicines (Nano-tubes and dechronification) and many others. The result of these human activities have increased the GHGs, temperature, NEOs and PHAs, meteorological extreme events etc. Some scientists have notion that the rate of extinction of species would have been much more and the modern Homosapiens would have vanished. The adverse effects of Anthropocene can be ameliorated and the extinction of species, delayed by application of modern science and technology developed.

The **geo-engineering approach** is to cool the climate and save the hot earth from GHG gas emissions by adding artificial aerosols in the stratosphere. To keep it safe from anthropogenic, forest fire, dust storms and drought actions are being taken by artificial heavy precipitations. The aerosol preferred to be efficient is the sulphate formation in the stratosphere Cruzen 2008. But the sulfate particles near earth surface are fatal and can make acid rain. The sulphur dioxide action cause death to a tune of 0.5 million annually, preventive actions are under consideration as per WMO report.

The carbon emission can be reduced by imposing heavy taxes on the countries who cannot maintain its carbon level in the developing countries in the world (as per The Kyoto Protocol, Canada, 2011). Ships and cargoes add a lot to the atmospheric carbon and sulphur pollutions from both coal and diesel engine. <http://nce. Habitats even. Work /2014/india/>. The CO₂ and energy management in India needs to be done in domestic, Industrial, Transportation, and agricultural sectors. Air pollution in India can be reduced by proactive measures projected reducing mortality rate due to reduction of PM5 and PM2.5 particulates

Hydro-meteorological extreme events: In November 2013, Haiyan, a typhoon slammed and ransacked Philippines coast causing death to 10,000 people and about 9 million became homeless. Prior another VSCS, Phailine and Hudhud, generated in east Andaman sea, in 12th Oct, 2013 and 2014 though not caused huge fatalities but devastated the Coromondol coast of India. Well preparedness and management of the cyclones were done the best and human loss was less in comparison to super cyclone at Para dip 1998 Fig 3(a).

Nano medicine: To combat human destruction and biologically process of ageing, the advances in nanotechnology, cloning and biotechnology have acted well. The Nano-medicines, with well-defined Nano-scale molecular structures, nanotubes, dendrimer-based organic devices and biologically active materials based on fullerenes can rejuvenate and slow down human ageing process. Knowledge of proteomics and genomics shall bring a revolution in medicine. Repair of damaged tissues by stem cell treatments, and biological robots created from bacteria and motile cells can deliver measured aging.

De-chronification: To continuously arrest and deplete biological ageing of human's application of intracellular respirocyte and microbivores in live cells, use of chromosome replacement therapy and repair of continuous cell damages can be done through Nano medical advances. The processes can be removal of toxins from cells, chromosome substitution therapy and cellular repair devices. It can even increase longevity from present 90 years to even 5400 years by de-chronification Freitas Jr.(1999)

From geological and hydro-metrological to biological: Mutagenic, carcinogenic or toxic microbes to reproduction shall be dominant in this epoch. The change from Holocene is not only anthropogenic but also mainly geological and hydro-metrological to biological.

The carbon pool and carbon budget: Tree cover makes major input to carbon pools on farm lands. The carbon pool adds to climatic variation mitigation and adaptation. Two third of fossil fuel is left to human which controls the carbon of the atmosphere. If from today planning, regulating the extraction, distribution and efficient use of coal, oil, electricity and atomic energy with use of maximum renewable energy not managed properly then the ambient temperature within 2^o C rise cannot be maintained, then the process of ageing of the Anthropocene shall be faster.

Conclusion:-

On stratigraphic study of recent lithology, mineralogy and limnology, the signatures of the epoch The Anthropocene during the great acceleration are found to be anthropogenic, chemo-stratigraphic, paleontological, changes in limnology and lithology, genomic and Geo-centric.

India is in the race of sixth extinction. Multi-level and polycentric efficient governance of vulnerable climatic anomalies can ameliorate and delay the effect of the great acceleration. Efficient policy makers; basin and other line managers, political leaders, economists and the teachers should stress upon effective adaptation and management which can provide better results to address the uncertainty and complexity of the epoch

Note: This paper is a prologue to the breakthrough dialogue on the Anthropocene at Cavallo point, Sausalito, California from 21st June to 23rd June, 2017.

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RESEARCH ARTICLE

PHYTOCHEMICAL SCREENING, ANTIOXIDANT & ANTIBACTERIAL ACTIVITY OF GREEN SYNTHESIZED SILVER & GOLD NANOPARTICLES USING LEAF EXTRACT OF *ZIZIPHUS NUMMULARIA* AND ITS EFFECTIVENESS AGAINST INFECTED DENTINE: AN *IN VITRO* STUDY.

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Ziziphus nummularia, Phytochemical Screening, Antioxidant activity, Antibacterial activity, Nanoparticles Synthesis.

Abstract

Green synthesis is a promising nanotechnological tool for biomedical application. The present investigation demonstrates a rapid biogenic approach for the synthesis of gold and silver nanoparticles using biologically active and medicinal important *Ziziphus nummularia* leaf extract as a reducing and stabilizing agent under ambient conditions. To our knowledge, this is the first report where *Ziziphus nummularia* leaf extract was found to be an appropriate plant source for the green synthesis. The presence of various phytochemicals viz. polyphenols, alkaloids and flavonoids, were investigated by following standard biochemical methods. The silver and gold nanoparticles were characterized by UV-visible spectroscopy (Systonic 2203) that showed Surface Plasmon Resonance (SPR) for both gold and silver nanoparticles at 551 and 438 nm. Scanning electron microscopy (SEM) revealed that gold nanoparticles were observed as nanorods in shape with 54 nm in size while the size of the silver nanoparticles was measured 200 nm with spherical shape. The antioxidant activity was determined by 2, 2- diphenylpicrylhydrazyl (DPPH) assay. Further, the antibacterial activity of synthesized silver and gold nanoparticles showed effective inhibitory activity against *Staphylococcus aureus* and *Sstrectococcus mutans*.

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Introduction:-

Nanoscience and nanotechnology has the capability to provide the most dynamic area of research in modern materials science (P.P Singh & C. Bhakat 2012). Nanoparticles are exceptional materials with exclusive features and electifying applications in diverse fields (R Geethalakshmi and DVL Sarada 2012). Metal nanoparticles, such as those containing silver (AgNp) and gold (AuNp) hold enormous application in different fields like medicine, electronics, energy saving, environment, textile, cosmetics due to their unique optical, electrical, and photothermal properties (C. Ramteke *et al.* 2013). The most important properties of silver nanoparticles are its antibacterial, antifungal and antimicrobial effects (A. Singh *et al.* 2010) while gold nanoparticles show optical and catalytic property in chemical reaction (Michael Quinten 2011). They also have strong surface plasmon resonance oscillations (L. Rivas 2001). The synthesis of nanoparticles is very quiet novel process leading to truly green chemistry in plants which supply advancement over chemical and physical method (A. Gole *et al.* 2001). As it is cost efficient and environment friendly easily scaled up for large scale synthesis and in this method there is no need to high pressure, temperature and toxic chemicals (D. Ashok Kumar. 2012). Conventionally, nanomaterials are

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synthesized by chemical or physical methods; Green synthesis of nanoparticles is the most promising method of synthesis (A. Leela & M. Vivekanand. 2008).

Dental caries is a chronic and infectious disease characterized by demineralization of the tooth due to acids formed by bacteria in biofilms produced on its surface. *Streptococcus mutans* is considered one of the most cariogenic microorganisms present in human dental biofilm (M.R. Linares *et al.* 2014). The cariogenicity of these bacteria is based on the metabolism of sucrose, which are a substrate for membrane-bound glucosyl transferases and a proficient growth substrate for *S. mutans* (V. A. Lee *et al.* 2010) (W. I. Loesche *et al.* 1986). The earlier approach to the treatment of using hand excavation was a painful and ineffective process of carries removal (A. Juntavee *et al.* 2014).

Medicinal plants play a key role in the innovation of new therapeutic agents for drug development (Elavazhgan T. Arunachalam KD 2011). *Zizyphus nummularia* belongs to the family Rhamnaceae. It is a shrub or thorny small bush or small tree with medicinal properties (M. Goyal *et al.* 2012). It is commonly known as wild jujube (English) and jharberi (Hindi). *Zizyphus nummularia* grows in great quantity in the arid and semi-arid areas of India. Especially in Rajasthan it forms 14% of the whole composition of the grassland flora (A. H. Shah *et al.* 1990). The local communities used *Zizyphus nummularia* as analgesic, anti-inflammatory, anti-colds and anti-coughs medicine (M. Goyal *et al.* 2011) and also contain numerous bioactive phytochemical materials such as polyphenols, flavonoids, pectin, saponins and alkaloids (A. F. Morel *et al.* 2009) (M. Lalitha Eswari *et al.* 2013). The leaves of *Zizyphus nummularia* have been reported to possess strong antioxidant properties (Salma Nasrullah Malik. 2015) and have also been used to treat diabetes and chronic fever in India. Fruits and roots of *Z. nummularia* are used to cure sun stroke and healing cuts. This plant is also used for removing stones (A. H. Shah *et al.* 1990) (S. Chandra *et al.* 2011).

To our knowledge, this is the first report where *Zizyphus nummularia* leaf extract was found to be an appropriate plant source for the green synthesis of silver and gold nanoparticle (D. Ashok Kumar 2012). Bioreduction of chloroauric acid (HauCl) and silver nitrate (AgNO₃) for the synthesis of silver and gold nanoparticles correspondingly with the plant extract, *Zizyphus nummularia* (Rhamnaceae) (D. Mubarak Ali *et al.* 2011). The rapid development of stable silver and gold nanoparticles is observed (S. S. Shankar *et al.* 2004). However, no studies have examined the antibacterial potential of nanoparticles using leaf extract of *Zizyphus nummularia* in an infected dentin model. The purpose of this in vitro study was to evaluate the antibacterial activity of Silver and Gold Nanoparticles against *S. mutans* and to find its efficacy as an intracanal medicament (R. Suvarna *et al.* 2014) (D. R. Herrera 2016)

Methodology:-

❖ Materials and Sample Collection:-

All chemical reagents including chloroauric acid (HAuCl₄), silver nitrate (AgNO₃) 0.135mM DPPH solution, Folin-Ciocalteu's phenol reagent, methanol and gallic acid (CDH, Central Drug House, New Delhi) were obtained and used as received. Muller Hinton agar (MHA) from obtained from (Hi-Media Pvt. Ltd. Mumbai). All the chemicals used were of the highest purity available. Ultrapure water was used for every experiment (Milli-Q System; Millipore Corp.). Fresh *Z. nummularia* leaves collected from the Maharaj Vinayak Global University, (Jaipur, Rajasthan India) were used as a plant source.

❖ Preparation of Plant Extract:-

20 gm of fresh leaves of *Z. nummularia* were washed thoroughly with double-distilled water and were then cut into small pieces. These finely cut pieces were then mixed with 100 ml double-distilled water and this mixture was kept for boiling about 20 minutes. After cooling, it was filtered through Whatman Filter paper. 1 and further use for biosynthesis of silver and gold nanoparticles.

❖ Synthesis of Silver Nanoparticles:-

10 ml aqueous extract of *Z. nummularia* leaves was added to 90 ml of silver nitrate solution (1mM). The solution was allowed to react at room temperature and after 30 minutes change in the color was observed (colorless to intense yellow).

❖ Synthesis of Gold Nanoparticles:-

5 ml aqueous extract of *Z. nummularia* leaves was added to 5 ml of chloroauric acid solution (1mM). This solution mixture was exposed to room temperature for 2 hour. Dark brown color was observed then analyzes the peak value of these particles with the help of UV Vis Spectrophotometer.

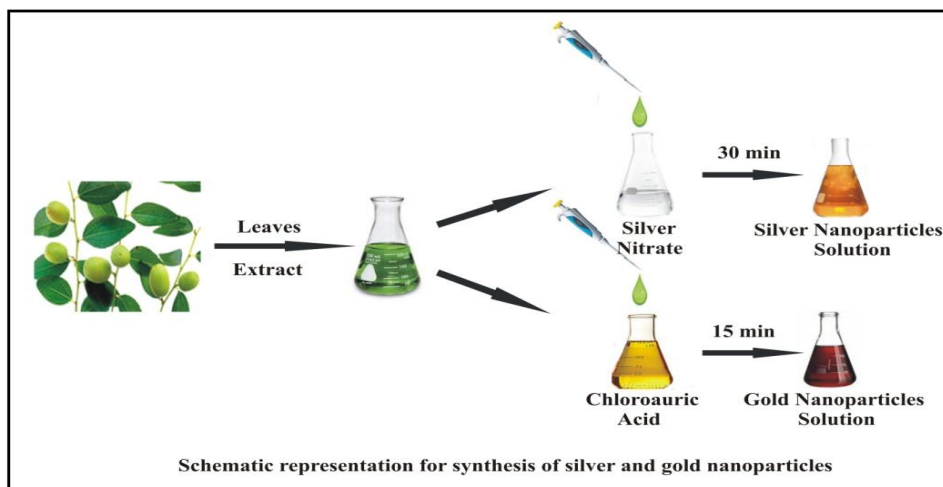


Fig. 1:- Schematic representation for synthesis Ag & AuNPs

❖ Characterization of Synthesized Nanoparticles:-

Ag and AuNPs obtained from *Z. nummularia* were characterized by recording UV-Vis absorption spectra using Double Beam UV-visible spectrophotometer 2203 through a quartz cell with 10 mm optical path that demonstrated peak value. The samples were packed in a quartz cuvet of 1 cm light- path length and the light absorption spectra were given in reference to deionized water.

The morphology of the colloidal sample was examined using Scanning electron microscopy (SEM-Zeiss) and Transmission electron microscopy (TEM-FEI Tecnai G2 S-Twin) with ultrahigh resolution (UHR) pole piece operating at an accelerating voltage of 300kV that revealed size and shape.

❖ Extraction of antioxidant:-

The leaves of *Ziziphus* were cleaned and cut into small pieces and then oven dried at 50°C for 72 hour. The dried sample was pulverized by a mechanical grinder and passed through 250 µm mesh and then stored at 4°C until use. In the extraction process about 1 gm of *Ziziphus* powder were weighed in universal bottles and 10 ml of 50% methanol was added. Then solution was homogenized using homogenizer at 24'000 rpm for 1 minute after that sample was centrifuged for 10 min. the supernatants were collected for further analysis.

❖ Radical DPPH Scavenging Activity:-

Antioxidant activity of the *Ziziphus nummularia* extract and standard was measured on the basis of the radical scavenging effect of the stable DPPH free radical. The 2, 2 diphenyl- 1- picrylhydrazyl was dissolving in methanol to prepare the DPPH solution. The DPPH solution was dilute 42 times with methanol to obtain 0.9 absorbance at 516 nm with the help of spectrophotometer. 1 ml of DPPH solution was added into 100 µl of *Ziziphus* leaf extract solution. This mixture was shaken in vortex and kept in dark room for 2 hour. After that mixture was transferred to micro plate and absorption was measured in spectrophotometer. The following equation (3.1) calculates the percentage of DPPH scavenging activity: the percentage of DPPH scavenging activity was calculated using the following equation: Radical scavenging (%) = $[(A_0 - A_1) / A_0] \times 100$ where A_0 is the absorbance of the control and A_1 is the absorbance of the sample extracts (Salma Nasrullah Malik 2015) (S. Gupta *et al.* 2016).

❖ Phytochemical Screening:-

Total Flavonoid Content:

Plant extract (0.5 ml of 1:10mg/ml) in methanol were separately mixed with 0.1 ml of 10% aluminium chloride, 1.5ml of ethanol, 0.1 ml of 1M sodium acetate and 2.8 ml of distilled water. Solution was kept for 30 min at room

temperature and absorbance of the reaction mixture was measured by double beam UV spectrophotometer at 415 nm.

Total Phenolic Content:

The total phenolic content of *Ziziphus nummularia* extract was determined by using Folin- Ciocalteu method. Standard Gallic acid (10gm) was dissolve in 100 ml of distilled water in volumetric flask (100 µg/ml of stock solution). 1.5 ml of Folin- Ciocalteu reagent and 10 ml of distilled water, diluted according to the label specification to each of the volumetric flasks. After 5 min 4ml of 1M sodium carbonate was added and made up to 25 ml with distilled water. Simultaneously leaf extract (0.5 ml of 1:10 mg/ml) in methanol were separately mixed with above reagent. After 30 minutes absorbance at 765 nm was observed.

Total Alkaloid Content:

5 gm sample of *Z. nummularia* was weighed into 250 ml and 200 ml of 10% acetic acid in methanol was added and kept for 48 hours. After filtration the extract were concentrated on a waterbath. Concentrated ammonium hydroxide was added drop by drop into the extract until the precipitation was complete. After that solution was washed with dilute ammonium hydroxide and filtered. The filtrate obtained was dried and weighed.

❖ In Vitro Study On Infected Dentine:-

Firstly infected tooth (Incisor, Premolar & Molar) collected from Maharaj Vinayak Global University in the department of Conservative and Endodontics. Healthy and infected tooth dipped into nanoparticles solution synthesized from *Ziziphus nummularia*. pH of solution should be maintained at 5.5. Nanoparticle solution kept in B.O.D. incubator (37° C) for overnight incubation. After overnight incubation results was observed.

Results:-

Silver and gold nanoparticles were synthesized using leaf extract of *Ziziphus nummularia*. Due to the reaction of the metal salt, color of the solutions changed colorless to yellow and dark brown, indicating the formation of silver and gold nanoparticles, respectively.

❖ UV Visible Spectroscopy:-

Reduction of silver ions into silver nanoparticles using leaf extracts *Z. nummularia* was proved by the change of color from colorless to intense yellow. The UV- Visible spectra show an absorption band at 438 nm indicating the presence of silver nanoparticles.

For gold nanoparticles, bioreduction of Au ions was observed by visualizing the color change from colorless to dark brown. UV- Visible spectra confirmed the presence of gold nanoparticles with absorption band at 551 nm.

❖ SEM Results:-

The scanning electron microscopy (SEM-Zeiss) has been engaged to characterization the size, shape and morphologies of formed silver and gold nanoparticles. From the images (**Fig. 2**) it is evident that the morphology of AgNP is nearly spherical and AuNP is indicating niddle shaped rod. The average particle size examined with the help of SEM images is observed to be 200 nm of silver nanoparticles while 54 nm of gold nanoparticle.

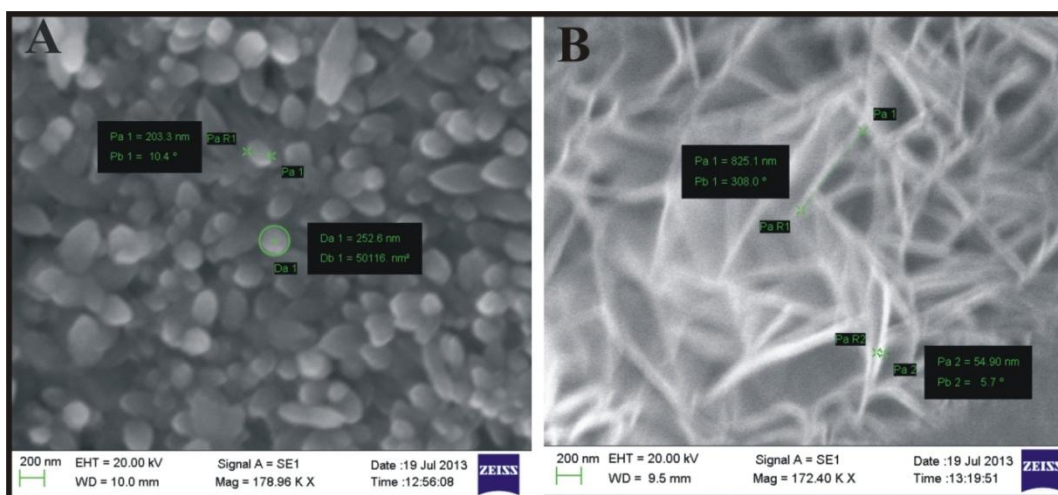


Fig. 2:- SEM Images of (A) Silver nanoparticles and (B) Gold Nanoparticles

❖ TEM Results:-

HR- TEM micrograph was examined the morphology of silver and gold nanoparticles. The data obtained from TEM images found distinct shape and size of polydisperse nanoparticles. These images suggest that the gold particles are niddle shaped rod and silver nanoparticles are mostly spherical in shape. It is evident that the size distribution of gold NPs between 50-60 nm and 200 nm for the silver NPs. The spherical and niddle shaped rod of the particle, as visible in **Figure 3**, is due to the fact that when a particle is produced, in its initial state, it tries to obtain a shape that corresponds to minimum potential energy.

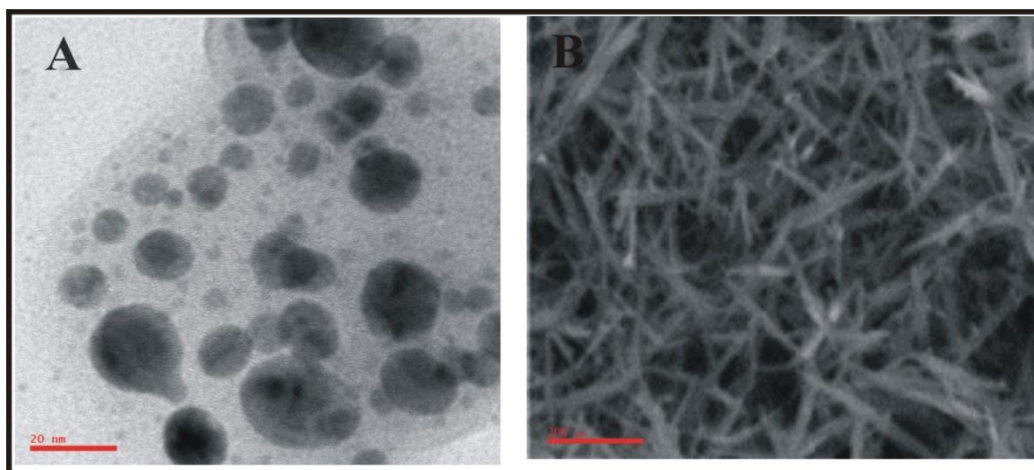


Fig. 3:- TEM Images of (A) Silver nanoparticles and (B) Gold Nanoparticles

❖ Antibacterial activity:-

The agar diffusion method was used to notice the effect of the concentrations of both silver and gold against *Streptococcus mutans* (MTCC 1890) & *Staphylococcus aureus* (MTCC 7443) bacteria. The in vitro antibacterial activity of the samples was evaluated by using Mueller–Hinton Agar (MHA). The results as shown that *Streptococcus mutans* & *Staphylococcus aureus* were susceptible to the gold and silver nanoparticle both (**Fig. 4**). The zones of inhibition created by the gold & silver particles of the *Ziziphus nummularia* compared favorably with control in this study. The activities of gold and silver nanoparticles against *S. mutans* & *S. aureus* suggested that these particles could be used to treat dental carries

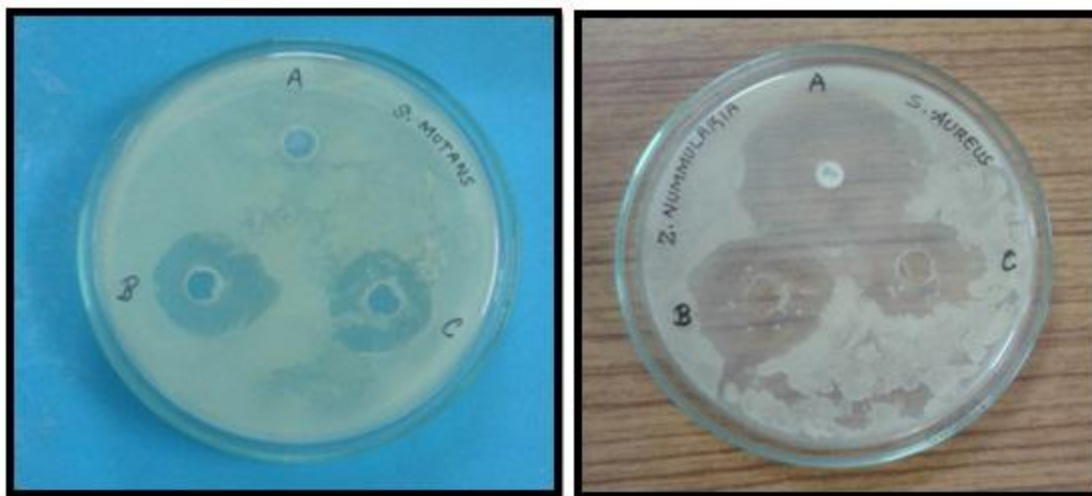


Fig. 4:- Antibacterial Activity against *Streptococcus mutans* & *Staphylococcus aureus* (a) Control (b) Silver NPs (c) Gold NPs

❖ **DPPH Assay (Radical Scavenging Activity):-**

A huge number of methods have been developed to estimate antioxidant capacity of medicinal herbs extracts. But only some of them used widely due to the complexity of measuring total antioxidant capacity owing to limitations associated with methodological issues. DPPH radical scavenging activity assay is performed to monitor the ability of compound to act as free radical scavengers and to analyze the antioxidant activity of plant extract. The methanol extracts of *Ziziphus* leaves sample was calculated for their IC₅₀ values against DPPH. IC₅₀ is the required concentration of *Ziziphus nummularia* antioxidants to scavenge 50% DPPH radicals in the reaction mixtures. Free radical scavenging property was calculated. The antioxidant activity of *Ziziphus* leaves is illustrated in Table Figure (1 and 5). These data indicated that the *Ziziphus nummularia* extract is kinetically active scavenger against DPPH under the testing conditions.

Table 1: - DPPH Free radical scavenging activity of *Ziziphus Nummularia*

Sample	Concentration (mg/mL)	% Inhibition	IC50
<i>Ziziphus nummularia</i>	0.023	31.91	0.03832
	0.046	60.48	
	0.069	84.41	
Control Absorbance = 0.00184			

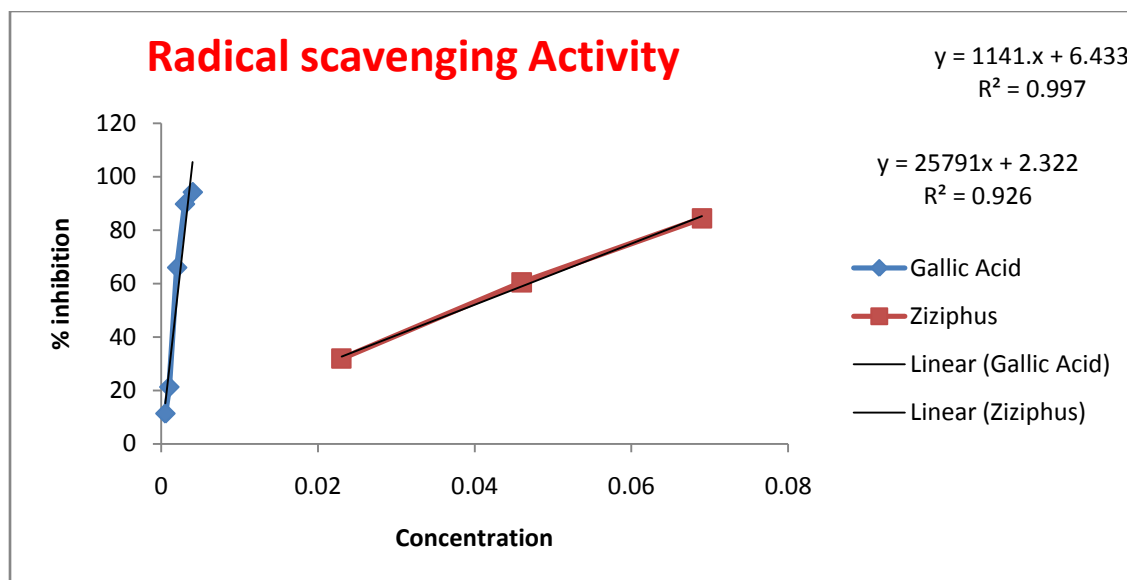


Fig. 5:- Graphical representation of DPPH scavenging activity of *Ziziphus nummularia*

❖ Phytochemical Screening:-

Phytochemicals component in the plants are identified to be biologically active complexes and accountable for various activities such as antibacterial, antimicrobial, anti inflammatory and antioxidant. In the present study, the phytochemical screening of *Ziziphus nummularia* leaf extract had been performed with methanol extract and the results shown in **Table 2** respectively. It revealed the presence of alkaloid, flavonoid and phenolic compound present in the extracts.

The total phenolic content was found to be 1.37% while total flavonoid content was found to be 0.066% in methanol extract. Total alkaloid content was found to be 4.04% in plant powder.

Table 2: - Phytochemicals detected in *Ziziphus nummularia* Extract

Parameters	Results	Method
Extractive Value (Methanol Solvent)	12.01%	API Method 2008
Total Flavonoid Content (By UV spectroscopy)	0.066%	API Method 2008
Total Phenolic Content (By UV spectroscopy)	1.37%	API Method 2008
Total Alkaloid Content	4.04%	API Method 2008

API: Ayurvedic Pharmacopeia of India

❖ In Vitro Study on infected Dentine

Silver & gold nanoparticle extracted from *Z. nummularia* provides antibacterial activity against acid producing bacteria, *Streptococcus mutans*. Healthy and infected tooth dipped into nanoparticles solution (pH 5.5). Nanoparticle solution kept in B.O.D. incubator (37° C) for overnight incubation. Dentine starts dissolve after overnight incubation in AuNPs & AgNPs solution (pH 7) was observed. The use of these NPs in the treatment of dental caries was found to be very effective.

It is proved that this project is successful in reducing infected dentine further studies must be conducted to test the carcinogenic properties either in animal model or in cell lines in order to evaluate the application of AgNPs & AuNPs as a bactericidal agent.

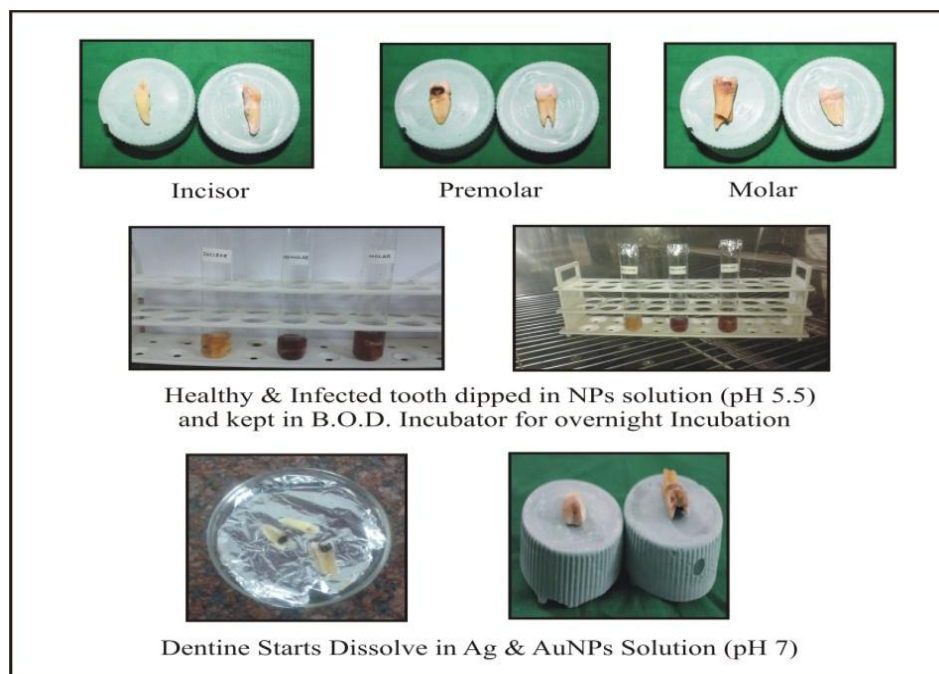


Fig. 6:- Schematic representation of Ag & AuNPs Effect on Infected Dentine

Conclusion:-

The current method eludes the use of toxic chemicals for the synthesis of gold and silver nanoparticles so it can be used for biological applications. This research has successfully employed for the development of silver & gold nanoparticles with spherical & nanorod shapes by using *Ziziphus nummularia* as reducing and stabilizing agent. This synthesis approach of gold and silver nanoparticles is rapid, cost effective and can be widely used in biological systems & medical system. Nanoparticles have been extensively researched because of their unique physical properties, chemical reactivity and potential applications in biological labeling, biosensing, drug delivery, antibacterial, antimicrobial and antiviral activity, detection of genetic disorders, gene therapy, and DNA sequencing. According to the results, *Ziziphus* leaves contain alkaloid, flavonoid and phenolic compounds, it can be considered as an excellence source of antioxidant compounds.

Silver & gold nanoparticle extracted from *Z. nummularia* provides antibacterial activity against acid producing bacteria, *Streptococcus mutans* & *Staphylococcus aureus*. The use of these NPs in the treatment of dental caries was found to be very effective. This project is successful in reducing infected dentine further studies must be conducted to test the carcinogenic properties either in animal model or in cell lines in order to evaluate the application of AgNPs & AuNPs as a bactericidal agent.

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RESEARCH ARTICLE

PREVALENCE OF OVERWEIGHT AND OBESITY AMONG RURAL PRESCHOOL SCHOOL CHILDREN CAMEROON.

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Obesity, Prevalence, Preschool children, rural Cameroon.

Abstract

Background: Pediatric obesity is becoming a public health issue across several countries. Childhood obesity has immediate and long-term impacts on physical, social, and emotional health. Limited data concerning obesity among preschool children are available in rural Cameroon.

Aim: The aim of this study was to determine the prevalence and risk factors associated with overweight and obesity among rural nursery school children in Ngog-Mapubi Cameroon.

Material and Methods: This was a cross sectional study conducted in nursery school children aged between 2-5 years during the month of March 2016 at the public rural nursery school of Ngog-Mapubi Cameroon. Obesity and overweight were defined according WHO. Data were analyzed using Stata version 11. The Chi-square test was used to assess the association between respondent characteristics and the prevalence of overweight and obesity. Logistic regression was used for determination of risk factors of childhood overweight and obesity.

Results: Out of total 100 children recruited in this study, The overall prevalence of overweight is 40% (55.10% for boys and 25.49% for girls) while the overall prevalence of obesity is 13% (8.16% for boys and 17.64% for girls).

Conclusion: Finally, the cumulative prevalence of obesity is 53% in the studied population. Parent rentals and Child watches television after school were significantly associated to overweight while Parent rentals and Child eats snacks were significant predictor of obesity.

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Introduction:-

Overweight and obesity among preschool children is becoming an emerging public health issue. It recognized that all their life, childhood obesity is associated with immediate and long-term impacts on psychological, physical, social, and emotional health troubles. Children with obesity are at higher risk for having other chronic health such as asthma, sleep apnea, bone and joint problems, type 2 diabetes, and risk factors for heart disease and diseases that impact physical health, [1, 2, 3-5]. Children with obesity are bullied and teased more than their normal weight peers, [6] and are more likely to suffer from social isolation, depression, and lower self-esteem. [7] In the long term, childhood obesity also is associated with having obesity as an adult, [8] which is linked to serious conditions and diseases such

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as heart disease, type 2 diabetes, metabolic syndrome, and several types of cancer. ^[9-10] In developed countries, many reports, have shown increasing numbers of overweight children ^[11-13]. Developing countries are facing the double burden of under nutrition over nutrition and the extent of the problem remains unknown. Projections show that prevalence among children of less than 5 years) obesity will attend the magnitude of 12.7% in Africa by 2020 ^[14] In Cameroon, scarce childhood obesity data are available ^[15]. This study was carried out to estimate the prevalence of overweight and obesity among rural preschool children of 2 to 5 years

Materials and Methods:-

Ethics:-

The study was conducted during the month of March 2016 at the public rural nursery school of Ngomapubi Cameroon. Admission to the study was based solely on voluntary authorization of the parents or the guardians who provided their verbal informed consent. The study was approved by the Education Planning Commission of the hospital of Ngomapubi.

Subjects:-

Our cross sectional study was carried out on 100 healthy preschool aged children the aged between 2 and 5 years. Data were collected during the month of March 2016 by applying a standard questionnaire to the mothers, the fathers or guardians. The parents filled the questionnaire regarding age, sex, education and occupation of the parent, family size, family history of chronic diseases, eating habits of junked food.

For each child anthropometric measurement of weight, Height, health examination and blood pressure measurement. Height and weight measurements were taken for each subject using adequate methodology of Weiner and Lourie 1981 et al [16]. The height was measured in standing position using tape meter while the shoulder was in a normal position to the nearest. Body weight were determined in participants (with very light clothing on and without shoes) using a Jocca™ scale.

The blood pressure was measured by one nurse with Labell™ electronic blood pressure on the wrist. If one child was crying, the blood pressure was not taken until he or she became calm. The determined systolic and diastolic pressures were ascertained from three readings using the mean value of the three. Blood pressure was measured 3 times on the left arm and in the sitting position, with at least 10 min of rest before the first measurement and a two-minute interval between measurements, according standardized techniques.

Definition of Overweight and Obesity:-

Body mass index (BMI) was calculated as weight in kilograms divided by square of height in metres. International criteria for BMI percentiles for age and sex (Cole et al. 2000^[17]). BMI is a measure used to determine childhood overweight and obesity. Overweight is defined as a BMI at or above the 85th percentile and below the 95th percentile for children and teens of the same age and sex. Obesity is defined as a BMI at or above the 95th percentile for children and teens of the same age and sex.

Statistical Analysis:-

The survey sample was categorized according to age and gender, All data were analyzed by STATA version 13. The data were classified into four age groups, each of one-year duration, i.e., age group 2+ included children from age 2.00-2.99 years, the age group 3+ from 3.00-3.99 years and so on. Categorical variables are presented as percentages or numbers. The chi-square test respectively, P value <0.05 was considered statistically significant.

Logistic regression was performed with adjustment for all variables for identification of risk factors for overweight and obesity

Results:-

Of the 100 children, were (49%) were males and 51% females. Their age ranged from 2-5 years. Table 1 shows the distribution of the studied population according to age. Most of the girl assessed had 4 years while the majority of the boys had five years. Table 2 reports the prevalence of overweight among boys and girls, the highest prevalence of overweight was reported among boys of 5 years while the lowest was noted among girls of 5 years. They were a significant difference among overweight prevalence of boys and girls of 5 years.

Table 3 reports the prevalence of obesity among boys and girls, the highest prevalence of obesity was reported among girls of 3years while the lowest was noted among girls of 5years and boys of 3years. They were a significant difference among obesity prevalence of boys and girls of 3years.

Table 4 show factors associated with overweight. Parent rentals and Child watches television after school were significantly associated to overweight.

Table 5 show factors associated with obesity. Parent rentals and Child eats snacks were significant predictor of obesity.

According to table 4 and 5, boys exhibit higher significant prevalence of overweight but lower prevalence of obesity comparatively to girls. The overall prevalence of overweight is 40% (55.10% for boys and 25.49% for girls) while the overall prevalence of obesity is 13% (8.16% for boys and 17.64% for girls). Finally, the cumulative prevalence of obesity is 53% in the studied population.

Table 1:- Characteristics of nursery school children

Age(years)	Boys(°/°)	Girls(°/°)	Total (°/°)
2+	7(33.33)	14(66.67)	21
3+	11(40.74)	16(59.26)	27
4+	10(37.07)	17(62.96)	27
5+	21(84.00)	4(16.00)	25
2+3+4+5+	49(49.00)	51(51.00)	100

Table 2:- Prevalence of overweight among boys and girls

Age(years)	Boys(°/°)	Girls(°/°)	P value
2+	28.57	21.42	0.717
3+	27.27	31.25	0.840
4+	10.00	29.41	0.241
5+	100	0.00	0.000*
2+3+4+5+	55.10	25.49	0.029*

Table 3:- Prevalence of obesity among boys and girls

Age(years)	Boys(°/°)	Girls(°/°)	P value
2+	14.28	7.14	0.599
3+	0.00	43.75	0.011*
4+	10.00	5.88	0.693
5+	14.28	0.00	0.420
2+3+4+5+	8.16	17.64	0.034*

Table 4:- Risk factors for overweight among nursery school

Variables	Odd ratio (95%CI)
Parent rentals	1.26(0.55-2.99)*
Parent Fatness	0.71(0.23-2.21)
Child watches television after school	1.55(0.60-4.00)*
Full meal from appetizer to dessert	0.84(0.36-1.96)
Child eats snacks regularly	0.84(0.34-2.06)

* $P < 0.05$ considered significant

Table 5:- Risk factors Obesity for among nursery school

Variables	Odd ratio (95%CI)
Parent rentals	1.77 (0.53-5.86)*
Parent Fatnesss	0.68 (0.12-3.65)
Child watches television after school	1.30 (0.35-4.87)
Full meal from appetizer to dessert	0.60 (0.18-2.04)
Child eats snacks regularly	1.64(0.49-5.48)*

Discussion:-

Childhood obesity is becoming a worldwide serious public health problem. Obesity in childhood is tend to be maintained in adulthood with series of unhealthy events such as dyslipideamia, hypertension, type 2 diabetes, metabolic syndrome and psychosocial disorders. Obesity has reached pandemic proportion in Cameroon and to limit that pandemic, prevention of adult obesity induced morbidity and mortality should be trigger as early as possible in childhood. Scarce studies have been conducted on overweight and obesity prevalence and risk factors among preschool children in rural areas of Cameroon. This study was carried out to estimate the prevalence of overweight and obesity among rural preschool children aged between 2 and 5years old of Ngog Mapubi nursery school and to contribute to find effective ways to prevent childhood obesity.

The prevalence of overweight and obesity in our study is too high and does not favour comparison with others studies. This prevalence was higher than the previous Cameroonian (8.0%) study[15] and others sub-Saharan African studies done in Ivory Coast (4.4% in 2012), in Nigeria (6.3% in 2013), in Senegal (6.8% in 2013), and Democratic Republic of Congo (5.6% in 2014) [18]. Many reason can justified this great prevalence, the small sample size, the definition used, the age of participants and the socio-economic status.

In our study, prevalence of obesity varies with gender difference, it was noted that boys were more at risk of overweight while girls were more obese. This finding is consistent with many studies [19,20,21], but in contrast with reports with girl obesity predominance[22-26] and no gender difference noticed among boys and girls [27,28]. It is well known that overweight is a result of interactions between genetic and environmental factors, metabolism, eating and physical activity behaviour, and social and individual psychology[29-32] It has been found that parent rental is a risk factor of obesity, infact our study area is a rural area where only civil servant with high incomes are transferred for professional reasons and are obliged to rent houses because they are not in their native village. This is supported by the reports show that in developing countries, a positive association is noted has between obesity and children of high parents having a high socioeconomic status, [33].

Inactivity through watching television after school and eating snacks regularly are obesity risk factors in our study. Television influence the dietary behaviors of children and reduces their daily physical activity level and consuming more energy from snack with added sugars or solid fats than the body uses for healthy functioning, growth, and physical activity, leads to obesity. [34,35,36].

This study has some major limitations, the small sample size of population recruited only in Ngog Mapubi and it cross-sectional nature prevents it to be generalized in all rural preschool children.

Conclusion:-

These findings brings out the reality of child obesity among preschoolers in rural Cameroon and support the need for practical actions to be taken at the level of schools, parents and children to bar the way to obesity epidemic.

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Declaration of Conflicting Interest:-

The authors declare that there are no conflicts of interest.

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RESEARCH ARTICLE

HEPATIC RESECTION: A CHALLENGE TO ANAESTHESIOLOGIST!!!

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Key words:-

Hepatic resection, low CVP, blood transfusion

Abstract

Background: Hepatic resection is a surgery which is associated with high risk of patient's morbidity and mortality. Thereby requiring expert skills on anaesthesia front. With the advancement in medical science there has been reduction in morbidity of patient. Starting from the assessment of the patient preoperatively for preexisting parenchymal liver disease going through the intra op period in which haemodynamic alterations can take a toll to the postoperative part, all are very crucial.

Aims: 1. To study the anaesthetic strategies during perioperative period.

2. To assess the postoperative recovery phase.

Methodology: This was a retrospective analysis of 10 patients with established liver disorders excluding patients with alcoholic hepatitis planned for hepatic resection in age range of 40-50yrs, under ASA grade 3. After induction of patient under epidural and general anaesthesia, haemodynamic parameters, blood loss, central venous pressure(CVP) were observed. CVP was kept ≤ 5 mmHg. Postoperative monitoring for vitals and complications was done.

Results: Out of the 10 patients, (Carcinoma GB, Hemangioma, Hydatid cyst). Two patients were not able to sustain low CVP, leading to acute transient renal failure in post operative period. Four (40%) patients needed blood transfusion. 20% patients had deranged coagulation profile which was managed by transfusion of fresh frozen plasma.

Conclusion: Improvement in surgical and anesthetic skills, use of risk reduction strategies, preoperative assessment, haemodynamic management can help in better patient outcome with reduction in mortality.

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Background:-

Liver is a highly vascular organ with normal hepatic blood flow being 25% to 30% of the cardiac output. Liver has eight segments; each having its own blood supply by portal vein and hepatic artery. The segmental portal venous and hepatic arterial blood supply and biliary drainage help in resecting the contiguous segments without disrupting

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the vascular supply to neighboring tissue. There are few bloodless planes of dissection and divisions are invisible on the surface of the liver. Liver has an important role in the functioning of various body systems, such as activity of gastrointestinal tract, renal function, metabolic functions including the synthesis and degradation or detoxification of essential compounds; pharmacokinetics of anesthetic and anesthesia adjuncts; and hemostasis. Hepatectomy, an important surgery, done for benign and malignant diseases of liver is associated with high risk of patient's morbidity and mortality and thus demands expert skills on the anesthesia front. Larger the liver resection, more traumatic is the surgery, greater potential for blood loss, lengthy clamping time and more chances of ischemia reperfusion liver injury. Also, hemodynamic management is the main concern during the resection surgery. Blood loss during a liver resection is associated with increased post-operative morbidity rates.^[1,2,3] Age, size of resection, elevated creatinine, bilirubin, albumin levels, deranged prothrombin time, ascites and presence of heart disease are the pre operative risk factors associated with increased post operative mortality. With the advancement in medical sciences and vigilant perioperative care there has been reduction in morbidity of patient. Eighty percent of the liver can be removed, because of the hepatocytes; which are unipotent cells and have the capacity to develop or differentiate into only one type of cell but the risks of liver failure and other complications are high. Since the surgery is complex, the morbidity and mortality rates are high.

Methodology:-

A retrospective analysis of 10 patients with established liver disorders excluding patients with alcoholic hepatitis, ascites, cirrhosis and encephalopathy planned for hepatic resection in age range of 40-50yrs under ASA grade III was done. The study was done to observe the anesthetic strategies during peri-operative period and to assess the post operative recovery phase. After a detailed pre operative assessment, investigations like complete haemogram, liver function, renal function, serum electrolytes, prothrombin time/ INR, ECG, Chest X-ray were done and optimally corrected prior to surgery. Patients were kept nil by mouth overnight prior to day of surgery. On the day of surgery after well informed high risk consent, patients were taken into the operating theatre and multi-para monitors were attached. Intravenous access with two large bore (16G /14G) cannulae was done. They were pre medicated with glycopyrolate (0.004mg/kg), ondansetron (0.08mg/kg), midazolam (0.05mg/kg) and analgesia was given with fentanyl (1-2 µg/kg). An epidural catheter for intra and post operative analgesia was inserted at L1-2 with the tip of the catheter kept at T8/T9, bupivacaine and fentanyl in calculated dose were used. After pre oxygenation with 100% oxygen for 3 minutes induction was carried out with etomidate (0.3mg/kg) and intubation was facilitated by atracurium (0.5mg/kg). Maintenance of anesthesia was carried out with O₂, N₂O, isoflurane (1-2%) and atracurium infusion (0.1mg/kg/hr) under controlled ventilation. During resection PEEP was reduced to zero to reduce the intrathoracic pressure, thereby assisting in venous return and lack of back pressure on the hepatic veins. Post induction central venous catheter for central venous pressure monitoring and drug infusions was secured. Intra arterial blood pressure monitoring and gas analysis was carried out with arterial line access. Patients were monitored with ECG, SpO₂, NIBP, EtCO₂, CVP, IABP, temperature and urine output. Haemodynamic monitoring was the mainstay during the intraoperative period. CVP was recorded and kept below ≤5mmHg in view of reducing the amount of blood loss. Fluid balance was managed meticulously with crystalloids, colloids and blood products maintaining normovolemia. Packed red blood cells were transfused to compensate the blood loss. To maintain adequate perfusion pressure, infusion of dopamine (2-4 µg/kg) and noradrenaline (0.05µg/kg/min) were used. In postoperative phase, patient were reversed with neostigmine (0.05mg/kg) and glycopyrolate (0.1mg/kg) and started on CPAP to avoid lung atelectasis. Patients were shifted to post anesthesia care unit and were extubated once fully conscious and attaining the extubation criteria. Vital monitoring including ECG, SpO₂, blood pressure and temperature was done. Post operatively complete haemogram, blood sugar, electrolytes, blood gases, input-output charting and prothrombin time were assessed and corrected if required. Post operative analgesia was maintained with epidural dose of fentanyl and other IV analgesics.

Results:-

A total of ten patients were operated of which 50% had hemangioma, 30% had Carcinoma Gall bladder and 20% had recurrent multiple hydatid cysts (Fig 1). Degree of pre operative liver dysfunction and extent of liver resection are important for maintaining coagulation profile. Two patients had deranged coagulation profile post operatively managed with transfusion of FFP and vitamin K administration. Forty percent of patients required blood transfusion intra operatively as they had profuse bleeding needing transfusion with PCV and FFP in 1:1 ratio to overcome the loss. Two patients could not tolerate low CVP. In spite of adequate volume replacement two patients had post operative transient renal dysfunction (Table1).

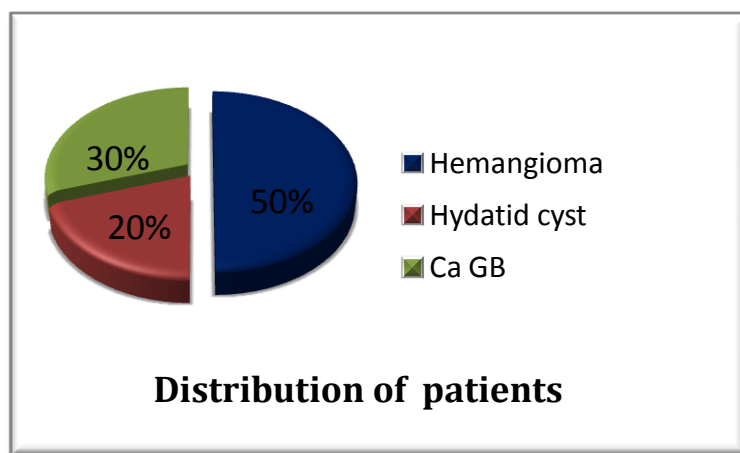


Fig 1:- A total of 10 patients: 5- Hemangioma; 3- Carcinoma Gall Bladder; 2- Hydatid cyst

Table 1:- Postoperative complications

TOTAL	10
BLOOD TRANSFUSION	04
ACUTE TRANSIENT RENAL FAILURE	02
DERANGED PROTHROMBIN TIME	02
ATELECTASIS	--
LIVER FAILURE	--
ASCITES	--
SEPTIC SHOCK	--
CARDIAC ARREST	--
MORTALITY	--

Discussion:-

Planning surgery for hepatic resection requires pre operative assessment and optimization of the physical and pathological condition of the patient. ASA grading assess the physical status, whereas Child Pugh Scoring is done for assessing the clinical status of the patient; which includes serum bilirubin, prothrombin time, presence of ascites, encephalopathy and nutritional impairment. Any pre existing liver pathology increases the risk of morbidity of the patient, thus strict perioperative monitoring and interventions are required during the procedure. Liver function tests are carried out to measure the hepatic dysfunction. Markedly increased aminotransferases reflect extensive hepatocellular injury, such as acute viral hepatitis, ischemic hepatitis and acute drug or toxin induced liver injury and hence they are contraindications to elective surgery. Raised alkaline phosphatase level indicates cholestasis due to calculi or tumors in the bile duct. Albumin, bilirubin, and prothombin time are laboratory parameters included in the Child-Pugh classification which are indicative of the biosynthetic functions and severity of cirrhosis. The MELD score (Model for End-stage Liver Disease score) is also an effective criteria for the assessment of chronic liver disease, its parameters being bilirubin, international normalized ratio (INR), creatinine and sodium levels. The coagulation profile is poor at predicting the risk of bleeding in individuals with liver disease because they reflect changes in procoagulant factors. Prolongation of the INR may reflect instability of the hemostatic balance, but not hypo or hyper coagulability in cirrhosis. Vitamin K (1mg/day) IV/ IM is administered for 3 consecutive days in patients with suspected deficiency (e.g. patients with poor nutrition, cirrhosis, cholestatic disease, with antibiotic use). Administration of antifibrinolytics such as tranexemic acid may reduce operative bleeding. As blood loss is suspected intra operatively; prior to surgery two large bore venous access is secured.

Patients with Child-Pugh scores of B or C should not undergo resection surgery because the risk of postoperative liver failure is very high. Similarly a MELD score of 10 to 15 confers increased perioperative risk, and patients with MELD scores >15 should not undergo surgery.

Alongwith anaesthesia its important to maintain organ perfusion, prevent air embolism, decrease blood loss using anaesthetic skills. Liver disease can affect the pharmacokinetics of anesthetic drugs by changing drug metabolism,

protein binding, and volume of distribution. Selection of drugs and dose adjustments depend on the severity of liver disease. Currently used volatile anesthetic agents (isoflurane, sevoflurane, desflurane) are not associated with hepatotoxicity as seen with halothane.^[4] It is because of the risk of hepatotoxicity, and reduction in cardiac output and hepatic oxygen supply, halothane is not administered to patients with liver disease.^[5] All volatile anesthetics decrease hepatic blood flow to a small extent. Isoflurane and sevoflurane result in little reduction and desflurane result in 30% reduction of hepatic blood flow. Isoflurane and desflurane, undergo less metabolism to trifluoroacetyl chloride (TFA) and sevoflurane does not undergo metabolism to TFA. There is an increase in elimination half life and free drug level of benzodiazepine leading to enhanced sedation. Metabolism is reduced in patients with liver disease, particularly for opioids like morphine and hydromorphone whereas, elimination of fentanyl is not appreciably altered in patients with liver disease, unless repeated doses are administered.^[6] Elimination and recovery from remifentanyl is unchanged by liver disease. Clearance, elimination, and duration of action of succinylcholine, rocuronium and vecuronium are prolonged in patients with liver disease and thus recovery time is delayed.^[7] Metabolism of atracurium and cis-atracurium are unaffected by liver disease because it is broken down spontaneously by Hofmann degradation.^[8] However, it is important to monitor the degree of relaxation because patients with liver disease show increased resistance to drug and shorter duration of action. Prolonged neuromuscular blockade can be reduced by decreasing the amount and frequency of neuromuscular blocking drugs. Drowsiness is related to opiates or other accumulating drugs. In case of liver failure, there is risk of hepatic encephalopathy and hypoglycemia. Hence, monitoring of ammonia and glucose levels is important.

It has also been observed that low CVP approach provides with good surgical field, decreased blood loss and requirement of transfusion. Liver has few bloodless planes of dissection therefore larger the resection more traumatic is the surgery. A study by Eid EA et al ^[9] (2005), also concluded that reduction in blood loss and blood transfusion is significantly reduced if CVP is maintained at or < 5mmHg. In 2014, meta-analysis of five randomized trials including 280 patients found that blood loss and transfusion were less with low CVP management for hepatectomy.^[10] In another meta analysis of eight randomized trials (2015), low CVP was associated with less blood loss and lower risk of transfusion (odds ratio [OR] 0.65) than control patients with higher CVP.^[11] Whereas retrospective reviews of living related donor hepatectomies have reported that CVP monitoring did not reduce blood loss, and that blood loss did not correlate with CVP level.^[12,13,14] Reduced blood loss can be attributed to the fact that hepatic venous pressure is dependent on central venous pressure. Low CVP increases negative pressure gradient with respect to right atrium and increase in air embolism which can be detected by transthoracic echocardiography. There is a sudden fall in arterial oxygen and end tidal CO₂. Positioning patient in 15° Trendelenburg position can minimize incidence of air embolism.^[15,16] Management requires infusing fluid rapidly, give 100% oxygen and aspiration of air from pulmonary artery floatation catheter in situ.

Blood loss plays an important role for predicting morbidity and mortality of the patient. Cardiac disease is ruled out as it may increase right side heart pressure, thereby increasing blood loss. In a study by Wong et al ^[17] (2003) stated that “maintaining a CVP < 5mmHg by volume restriction and intravenous infusion of nitroglycerine and a systolic blood pressure above 90 mmHg by intravenous infusion of dopamine (4–6µg/kg) had dramatically reduced bleeding and transfusion requirements.” Epidural anesthesia does not lead to changes in intravascular volume, but only promotes redistribution of blood, decreasing both venous return and portal vein pressure, thus contributing to reduced hepatic congestion and surgical blood loss.^[18] Blood loss is also reduced by clamping hepatic artery and portal vein called Pringle’s maneuver. Prolonged clamping leads to warm ischaemia. Clamping is not done for more than 15 mins and then released for 5 mins. The clamping of hepatic artery and portal vein increases the backflow through hepatic vein.

Positive end expiratory pressure (PEEP) is reduced to zero to reduce the intra-thoracic pressure, and to increase in venous return and thereby decreasing the back pressure on the hepatic veins. In case of blood loss more than expected, transfusion is required. All cases which are done under epidural anaesthesia are done after ruling out any coagulation abnormality and are observed in postop period for spinal haematoma. Patients with microvascular bleeding, or who are at risk of bleeding due to percutaneous procedures, platelets are transfused to achieve a platelet count ≥50,000/microL, and cryoprecipitate transfusion to maintain fibrinogen levels ≥200 mg/dL.^[19,20] Ho AM et al ^[21] (2007) concluded that “the high prevalence of deranged haemostatic measurements after hepatectomy peak on day 2, because of the complex haemostatic changes related to both the cancer and the surgery and it could not be determined whether these changes were associated with an increased risk of epidural hematoma”. Another study by Stamenkovic DM et al ^[22] (2011) noted that “there is no single reported case of epidural hematoma as a consequence of epidural catheter insertion or removal after elective hepatectomy”.

Enhanced Recovery After Surgery (ERAS) protocols in postoperative period are being increasingly used which help in reducing the duration of hospital stay and morbidity rates. After the surgery is over patient is reversed back and extubated after full filling the extubation criteria. Prolonged intubation and sedation may lead to retention of secretions in the basal lung leading to atelectasis. This can be prevented by early application of Continuous positive airway pressure (CPAP) after extubation. Postoperatively pain is managed with doses epidural and IV analgesics. Injection fentanyl 0.5- 1 µg/ kg in diluted form can be used for post operative analgesia through epidural catheter. IV analgesics like Nefopam which has no hepatic metabolism and can be used in combination with opioids.^[23] Paracetamol infusion is used cautiously and for a short period. Coagulation profile (PT, aPTT, INR) is monitored as there is transient insufficiency of remnant liver to synthesize new factors. Coagulation disorder if present, epidural catheter removal is delayed till coagulation profile is corrected. Intravenous fluids should be managed carefully to avoid ascites, edema, and/or heart failure.^[24] Care is taken to prevent ascites, as incidence of exudative ascites is 50% in liver resection cases.^[25] Ascites can occur because of cirrhosis, high portal pressure, exudation, splanchnic arterial vasodilatation. The management includes monitoring of fluid infusion and simultaneously taking measures to maintain renal perfusion and diuresis. The strategy includes plasma substitutes, diuretics and vasoconstrictors. Hypovolemia is corrected which can predispose to decrease renal perfusion and GFR. Preventing renal impairment is another important consideration for the anesthesiologist. Renal auto-regulation ceases below a renal perfusion pressure of 70 to 75mmHg, below which, flow becomes pressure dependent. Renal failure can be corrected by hydration. Strict input output monitoring, BUN, serum creatinine are done to detect renal dysfunction. Patients with preoperative cirrhosis tolerate a reduction in renal perfusion pressure poorly and extra care must be taken to maintain perfusion pressure in order to preserve renal function.^[26] Serum electrolytes are investigated and if hyponatremia needs to be corrected, it should be corrected and is done slowly to avoid central pontine myelinolysis. Hypokalemia and metabolic alkalosis can occur with liver disease, and may trigger or exacerbate hepatic encephalopathy. Hypothermia is taken care off with use of warming blankets, warm IV fluids and ambient room temperature. Early post operative nasogastric feeding is important. It is usually started at the rate of 10ml/hr and is gradually increased. It helps to maintain gut function.

Conclusion: Improvement in surgical and anesthetic skills, use of risk reduction strategies, preoperative assessment, and hemodynamic management can help in better patient outcome with reduction in morbidity and mortality. Anesthetic vigilance along with thorough knowledge of surgical manipulations promotes team based health care in the operative room. Keeping in mind all the above discussed strategies and planning we were able to carry out the anesthesia with reduced post operative complications and any incidence of mortality.

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RESEARCH ARTICLE

A VALUE OF ROUTINE INTRAOPERATIVE CHOLANGIOGRAPHY IN CHOLECYSTECTOMY

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Abstract

Laparoscopic procedure can be feasible in patients with acute cholecystitis though a dense adhesion is common cause of conversion to open. which is prevented by intraoperative cholangiography. In this study it is 2% cases benefited. Hence benefits of cholangiography are reduced because of pre-operative diagnosis by other investigations modalities. The present study conducted at Department of General surgery Dr. D. Y. Patil Hospital and research centre, Kolhapur and MGM College, Mumbai. To study A Value of Routine Intraoperative Cholangiography in Cholecystectomy .

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Introduction:-

The extrahepatic biliary tree was first visualized in 1918 when Reich injected bismuth and petrolatum and defined a biliary fistula, thus opening the field for further studies of the biliary tree. Intraoperative cholangiography(1) (IOC) was introduced in the 1930s. Mirizzi recorded the first series of intraoperative cholangiography in 1932 using static films(2). It was employed inconsistently in the 1940s and 1950s, but in the 1960s and the 1970s, its use became commonplace. Later, the mobile C-arm image intensifier using a TV monitor was reported in a series by Berci and colleagues in 1978. The rationale for its routine use included: demonstration of aberrant ductal anatomy; avoidance of iatrogenic ductal injury; and detection of common duct stones. Patients with signs or symptoms of choledocholithiasis or with uncertain anatomy should undergo cholangiography. There is, however, controversy over whether IOC should be performed in those patients without signs or symptoms of choledocholithiasis and with clear anatomy. Intraoperative cholangiography adds time and cost to an operation. In addition, IOC has a false positive rate of 1-3%, leading to unnecessary biliary explorations and resulting in increased morbidity and mortality. Furthermore, the IOC can result in complications itself. This study focuses on the value of IOC in cholecystectomy and would help in proving, if at all, there is a rationale for its routine use.

In 1987, 105 years later, Philippe Mouret performed the first laparoscopic cholecystectomy (LC) in Lyon, France(3). A prospective study describing 500 laparoscopic cholecystectomies, was published by C.R. Voyles. The laparoscopic procedure was completed in 95 % of the patients, There was no mortality or bile duct injury, concluded that laparoscopic cholecystectomy was a safe procedure in the treatment of gallbladder disease(4).

The Southern Surgeons Club described 1518 laparoscopic cholecystectomies in prospective analysis, With respect to mortality, complications and length of hospital stay. A slightly higher incidence of biliary tract injury in the laparoscopic procedure is probably offset by the low incidence of other complications. Furthermore it is possibly caused by the inexperience of most surgeons starting endoscopic procedures(5).

The aim of this study was to determine the necessity for intraoperative cholangiography (IOC) during pediatric laparoscopic cholecystectomy (LC).

The success rate of LC in cases of acute cholecystitis is slightly higher when IOUS is used as an aid to dissection. In the absence of definitive prospective data, we recommend routine use of IOUS when performing LC, particularly in patients with acute cholecystitis(7).

This study was to compare the results of laparoscopic extracorporeal ultrasound (LICU) to those of fluoroscopic intraoperative cholangiography (FIOC) during laparoscopic cholecystectomy (LC) after the initial learning curve for LICU.

Materials And Methods:-

The present study was conducted on patients admitted for surgery in the **General Surgery unit of Tertiary care hospital**. Total of **100 Cholecystectomy elective surgeries** in the General Surgery unit in Dr. D. Y. Patil Hospital, Kolhapur and MGM College, Mumbai from **August 2010 to December 2012** were included in the study. The study included all patients irrespective of their Age, Sex, associated co-morbidities and previous surgeries. Information was collected from the patients after a Written, Valid, Informed consent. The study was **approved by IERC**.

Inclusion Criteria:-

1. All age group
2. Male and Female
3. Cholelithiasis
4. All cholecystectomy patients
5. Biliary Pancreatitis

Exclusion Criteria:

1. Cirrhosis with Ascites
2. Deranged bleeding profile

Preoperative Evaluation of a patient with gallstones Disease:-

In addition to a careful history and physical examination, preoperative testing for 90% of patients with gallstone disease(8) is minimal. An ultrasound of the right upper quadrant usually confirms the diagnosis, and the hepatic biochemical profile usually determines the likelihood that CBD stones are present and may warrant a preoperative MRCP/ERCP or an abdominal computed tomographic scan, or both, to search for the cause.

The diagnosis of biliary colic and acute cholecystitis, when the history and physical examination are less than definitive, can be made with a nuclear hepatobiliary hepato iminodiacetic acid (HIDA) scan(9). An ejection fraction of less than 50% at 30 minutes suggests poor contractile function and incomplete emptying of the gallbladder. Intravenous cholangiography, which is rarely performed in the United States, is popular in Europe for detecting CBD stones preoperatively in patients with abnormal liver function studies.

Postoperative Management:-

With advances in anaesthetic and postoperative analgesic care, early mobilization and discharge are the norm. In those patients in whom a T-tube has been placed, a cholangiogram is obtained on the 5th to 7th postoperative day

In routine cholecystectomy, no drain or nasogastric tube is required. The patient can resume fluid intake and diet when tolerated. Closed abdominal drains should be employed in those patients in whom there are concerns for the development of a postoperative collection or when the CBD has been opened. These drains should be removed within 24 to 48 hours when the volume is less than 100mL per day and there is no bile or fresh blood evident. Mild derangement of liver function tests may arise after uneventful cholecystectomy.

Observations And Results:-

There is study of total 100 cases from a Tertiary Care Hospital. In this study the mean Age is 45.41 with standard deviation is 13.76.

Table 1:- Descriptive Statistics.

Statistic	Age
Number	100
Minimum	16.00
Maximum	76.00
Mean	45.41
Std. Deviation	13.76

According to sex there are total 64 female and 36 male. Its implies that it is common in female population.

Table 2:- Sex-wise distribution of patients

Sex	Frequency	Percent
Male	36	36.0
Female	64	64.0
Total	100	100.0

Table 3:Distribution according to Clinical Presentations

Clinical Presentation	Frequency	Percent
ACUTE	31	31
Pancreatitis	3	3
CHRONIC	66	66
Total	100	100

According to clinical presentation out of 100 patients 31 (31%) were presented as acute cholecystitis, majority of them were diagnosed as acute on chronic cholecystitis on histopathology. If patient presented with in 72 hours of pain can be taken for cholecystectomy. Out of 100 patients 66 (66%) were Chronic and were diagnosed on ultrasonography with single or multiple gall bladder calculi are straight taken for cholecystectomy. 3(3%) patients had Pancreatitis who conserve, should do CECT abdomen which is normal, operated on same admission. All patients were operated electively under general anesthesia.

Table 3:- Past abdominal surgeries:

Past Abdominal Surgery	Frequency	Percent
Yes	11	11.0
No	89	89.0

Of the 100 cases 11 patients gives past history of upper abdominal surgery because its affect the treatment modalities of the surgery. Most prefer surgery for past upper abdominal surgery patient is open cholecystectomy compare to laparoscopic cholecystectomy as there is lot of adhesion while laparoscopy is perform and high chance for conversion to open cholecystectomy.

Table 4:- Comaparison of laparoscopic or open cholecystectomy

Table 4: Laparoscopic/Open cholecystectomy & cholecystostomy		
	Frequency	Percent
Open cholecystectomy	51	51.0%
Laparoscopic cholecystectomy	42	42.0%
Laparoscopic cholecystostomy	7	7.0%
Total	100	100.0%

Of the 100 cases study, open cholecystectomy is done in 51(51%). Laparoscopic cholecystectomy in 42(42%) and 7(7%) undergone laparoscopic cholecystostomy which after 4-6 weeks undergoes cholecystectomy after intraoperative cholangiography.

Duration of Surgery:-

The mean operation time for Laparoscopic cholecystectomy was significantly longer than for Open cholecystectomy and cholecystostomy. The distribution of operation time, among these groups is depicted in Table. The mean (range) operation time for laparoscopic cholecystectomy was 84-90 min (mean = 87.02 min), 66-77 min (mean = 72 min)

for open cholecystectomy and 31-39 min (mean = 35 min) with highly significant ($p < 0.001$). The 'F' value is 37.47. During the study period operation time for laparoscopic cholecystectomy showed a tendency to become shorter. Duration increased due to intraoperative cholangiography (about 15 min) as we became more familiar with laparoscopic IOC duration decreased..

Table 5:- Duration (in Minutes) according to Type of Surgery

Type of Surgery	N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		F-Stat	Significance
					Lower Bound	Upper Bound		
Open cholecystectomy	51	71.8627	19.10395	2.67509	66.4897	77.2358	F=37.47	p < 0.001, Highly significant
Laparoscopic cholecystectomy	42	87.0238	10.59565	1.63494	83.7220	90.3256		
Laparoscopic cholecystostomy	7	35.0000	4.08248	1.54303	31.2243	38.7757		

Post operative hospital stay:

Table 6:- Hospital stay according to Type of Surgery.

Type of surgery	N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		F-stat	Significance
					Lower Bound	Upper Bound		
Open cholecystectomy	51	6.1373	3.47574	.48670	5.1597	7.1148	8.188	p < 0.001, Highly significant
Laparoscopic cholecystectomy	42	4.8333	2.17431	.33550	4.1558	5.5109		
Laparoscopic cholecystostomy	7	10.2857	7.06433	2.67007	3.7523	16.8191		

Of the 100 cases, The hospital stay for open cholecystectomy range 5-7 days(mean 6.13 days), laparoscopic cholecystectomy is range 4-6 days (mean 4.83 days) and for laparoscopic cholecystostomy is 4-17 days(mean 10.29 days) with high significance $p < 0.0001$ value. The 'F' value is 8.188.

Post operative stay in laparoscopic cholecystostomy is more because to see for drainage from the tube is proper or not. In open cholecystectomy operative pain is more compare to laparoscopic hence it takes more hospital stay.

Intra-operative Complications:-

Table 7: Intra operative Complications

Intra operative Complications	Frequency	Percent
Yes	2	2.0
No	98	98.0
Total	100	100.0

The incidence of intra-operative complications is only 2% of the study. There is difficulty in cholecystectomy and cholecystostomy. There are two interesting case are one which presented intraoperatively choledochoduodenal fistula(10) and left lobe gall bladder(11). In both cases it took little longer time but surgery done without any intraoperative complications.

Benefits of Cholangiography:

Table 8:- Benefits of cholangiography.

	Frequency	Percent
Yes	2	2.0
No	98	98.0
Total	100	100.0

In this study only 2 (2%) cases shows the benefit of the cholangiography of which both are interval cholecystectomy which presented after 72 hours to see for the position of cystic duct in relation to CBD.

Discussion:-

The present study including 100 cases was carried out in the department of surgery, on patients who underwent intraoperative cholangiography and cholecystectomy.

Our study was based on studying patients with pain in the right hypochondrium due to varied etiologies(12), both in males and females of all age group, who underwent the above procedure.

Acute:-

Patients who were admitted with severe acute pain in the right hypochondrium, were advised to do complete blood count, liver function test, serum amylase and serum lipase levels. These being normal and ultrasound findings showing cholelithiasis, patients were posted for cholecystectomy. Acute cases who presented in first 72 hours were taken for laparoscopic cholecystectomy. Patients presenting after 5 days were treated conservatively with intravenous antibiotics, intravenous fluids, discharged and operated after 6-8 weeks. If these patients did not settle with conservative management, they were taken up for surgery and more often underwent cholecystostomy due to dense adhesions. Our threshold for cholecystostomy was low because these patients were usually in sepsis and had medical co-morbidities.

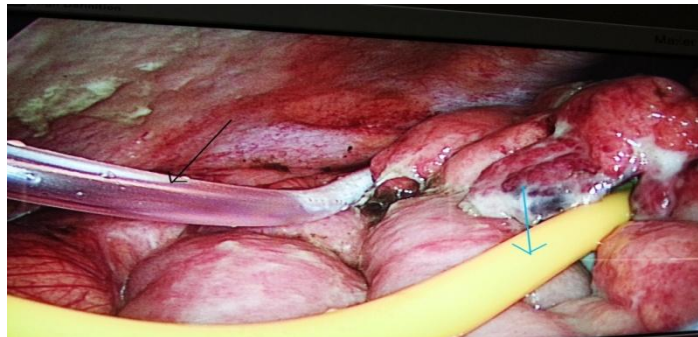


Fig 23:- Laparoscopic cholecystostomy(arrow Black- drain & Blue- cystostomy foley's drain

Biliary pancreatitis:-

In patients with cholelithiasis, pancreatitis was diagnosed with increased level of serum amylase and serum lipase(13). Patients were treated with nothing per orally, ryle's tube insertion, analgesic & somatostatin (3mg in 5% dextrose over 12 hourly for 3 days)(14)(15). If the stone was in the terminal CBD and patient was stable then endoscopic papillotomy with stenting used to be done in first 72 hours.

When pancreatic ileus(16) settled down we got CECT Abdomen to look for necrosis of any part of pancreas, fluid collection and peri-pancreatic stranding.

If only pancreatic oedema was seen, with no fluid collection or necrosis then in same admission laparoscopic cholecystectomy was advised.

Obstructive jaundice due to cholelithiasis:-

Patients who presented with obstructive jaundice due to CBD stones(17) were first taken up for ERCP with stenting after 48 hours. If the liver function test, amylase, lipase was normal then we went ahead with laparoscopic cholecystectomy in the same admission(18).

Cholangitis with choledocholithiasis:-

These patients underwent ERCP with or without naso-biliary drain and cholecystectomy was done when clinically cholangitis settled.

Symptomatic cholelithiasis(Diagnosed USG):-

Patients with recurrent right hypochondriac pain which settles without treatment

Or Murphy's sign positive were taken up for laparoscopic surgery without any further investigations apart from routine pre operative workup.

Incidental Cholelithiasis:-

Incidental cholelithiasis(19)(20) was mainly diagnosed on routine USG abdomen. Patients with less than 2.5 cm stones were advised a HIDA Scan. If it showed more than 50% Gallbladder Ejection stones were radiolucent then dissolution therapy with ursocholic acid was started.

Negative predictive ability of the set of criteria was 100% for patients up to 60 years of age and 97% for patients older than 60 years at the time of operation. No case of residual common bile duct calculi was present in the IOC and no-IOC groups at follow-up. (Our data strongly support a policy of performing IOC during cholecystectomy only when clinical criteria suggest the presence of common bile duct abnormalities or to clarify ductal anatomy)(17).

Since the introduction of laparoscopic cholecystectomy (LC), an increase in accidental common bile duct (CBD) injuries of up to 1.2-1.6% has been reported. In the present prospective study of 1,710 patients undergoing cholecystectomy we tested the predictive value of routine intraoperative cholangiography (IOC). The IOC was feasible in 92.4% of the cases in the LC group and in 83% of cases in the OC group and presented a complete depiction of the extrahepatic bile system in 98.3%. Anatomic variations of the bile duct system, which influenced the operative management, were found in 13.2% of cases. In 2.5% of the patients, preoperatively undetected CBD stones were also found. Method-specific complications did not occur in any of the patients. Additionally, in a controlled subgroup analysis of 163 patients, we evaluated preoperative intravenous cholangiography (IVC) and IOC. Intravenous cholangiography showed only 72.4% of the operation-relevant anatomic variations (vs. 100% by IOC); in 6.1% of the cases, there were reactions to the dye (vs. none in IOC), and in only 28.6% of the patients, CBD stones were detected (vs. 71.4% IOC). There were four bile duct injuries (0.29%) during LC and two (0.4%) during OC. All injuries were detected intraoperatively and fixed in the same setting without postoperative complications. In conclusion, we recommend the use of routine IOC during cholecystectomy. By this technique, anatomic variations of the bile duct system will be visualized and therefore accidental injuries will be avoided(18).

Conclusions:-

- In cholecystectomy if cystic duct is definitely identified then IOC is of no help in the safety of the procedure.
- IOC should be done in teaching hospitals for learning the procedure and also for safety as the operating surgeons are of limited experience.
- In very difficult cholecystectomies a cholecystogram should be done early in the procedure to delineate anatomy before definitive dissection of the calots triangle.
- Low threshold for cholecystostomy should be preferred rather than stubborn dissection which lands in disaster.
- Equipment for IOC should be kept ready and need based IOC should be practiced.

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RESEARCH ARTICLE

THE SECURITY VALUES THAT SHOULD BE AVAILABLE IN THE ISLAMIC EDUCATION.

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Security, values, availability, Islamic, education.

Abstract

Values are the moral or spiritual aspect of the Islamic civilization and also the soul and basis for any civilization. Therefore, they ensure the secret of human's survival and elasticity throughout the history. If values fade away one day, man will lose his/her moral warmth, which is the spirit of life and existence; mercy will abandon his/her heart; his/her ethics will not be able to play its role; he/she will no longer know the truth of his/her existence; and he/she will be bound with material restrictions. This study aimed at exploring the major domains of security values and their efficacy on the Islamic Education. Thus, the review will propose some suggestions for stakeholders, educationalist, and learners to have the proper inclusion of security values in order to maintain the coherence and integrity of Muslim societies.

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Introduction:-

The Islamic religion seeks to secure five imperatives that supports the human life; namely: the religion, brain, soul, money and dignity. So this is considered as the most comprehensive sense that necessitates building the most important pillars of the communities that must be preserved; so the soul that is honored by God must be preserved in security and safety from various hazards and disasters.

The Islamic culture curricula are one of the Islamic materials that are consistent with the other materials in the construction of Islamic thought properly; Article (12) of the education policy in Saudi Arabia calls for: "guiding the science and knowledge of various kinds and its methodology towards the Islamic doctrines and their methods of investment in order to be derived from Islam which is in consistent with the idea of Islamic governance (The Ministry of Education, 1390 H, p.9).

Hence, there was a need to consolidate the concepts and values of the Islamic culture in light of the challenges of civilization for reasons of human nature before being a Muslim. All Muslims believe in this religion and avoid its prohibitions; and it is innate and natural to every individual to belong to a family, group or a national to have his/her character represented in his/her community.

The Islamic Culture is one of the most important ways that can constitute Muslim generation which addresses these threats and challenges. The Islamic civilization can support the humanity with the values of truth, goodness, virtue, tolerance, prosperity, security, reassurance, well-being and happiness. On the basis of the educational policy in Saudi Arabia, which decided to teach the Islamic Culture in different colleges; because this is the fertile phase for the participation and dissemination of faith, and for the return of the Islamic, noble values, high ethics, and this confirms confidence in the validity of Islam to lead humanity, and to guide anytime and anywhere, and to broadcast

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the Islamic spirit which is enkindled in the soul the aspiration to high ideals, to resume the well-being, and the belonging to the nation of Islam. This shall appear as described by the Almighty by saying: *'You are now the best people brought forth for (the guidance and reform of) mankind. You enjoin what is right and forbid what is wrong and believe in Allah. Had the People of the Book believed it were better for them. Some of them are believers but most of them are transgressors.'* (Quran: Al-Imran, 110).

It is important then to adopt values of communities in the role of education by preparing curricula and textbooks properly to embrace the value of all kinds to provide learners with knowledge and action in order to maintain the interests of the individual and society hoping to show excellence and to maintain it. However, textbooks pay attention to the cognitive domains which had side effects on security in the community such as the increase in the rate of crime which poses a realistic indication that the imbalance may hit the values in society which requires competent attention to the curriculum which instill the values of security to fight crime and associate patterns of behaviour on the part of the individual and society, too (Ghanem, 1999).

Many studies such as Al-Ghanim (1994) , Al-Qaisy (1996), Al-Yahyawi (1417H), Jaroudy (1417H), and Aql (1427H), have emphasized the importance of values in the lives of individuals and groups as one of the most important inputs that govern human behavior which play an important role in both individual and group in the present and past. This is one of the components of the individual personality and the guidance of his/her conduct. Values are considered the reference framework for governing behavior, preferences and choices. On the other hand, values achieve social cohesion and integration inside the community and can build its identity and his well-being to achieve economic, industrial, social development, so the values are one means of achieving this development. However, the studies on values in school curricula and textbooks are so few compared to the studies conducted on other educational areas. Therefore, the security values have little interest, despite the suffering of Islamic and Arab countries which may adversely affect the tranquility of the groups and the individuals; as well as the negative reflections on the communities and their future.

Given the vulnerability of attention to the transplanting of security values in the hearts of young people, it was necessary for the curriculum in the kingdom of Saudi Arabia to reflect these values, besides other Islamic countries in order to help learners to cope with positive attitudes rooted in Islamic culture towards the development of noble values in the hearts of young students.

Previous Studies:-

The study of Al-Jarady (1993)

This study is entitled "Analysis of the values contained in Islamic education books for lower basic education in the Republic of Yemen" and aimed to identify the values contained in the books of Islamic education for the lower years of primary school students in the Republic of Yemen, to identify the distribution of values according to the domain, and then to find the extent of variation in the distribution of the values in the fields due to the variables of grade and subject.

The study sample consisted of all Islamic textbooks for the primary phase of Education in the Republic of Yemen. The researcher used in his study the descriptive method in accordance with the method of content analysis through a questionnaire and observation card. To achieve the goal of the study; the researcher has classified the values into two dimensions which are the content through rating values for the following areas:(the legislations, worshipping, personality, society, materialism) and the second dimension classification of the values according to (positive values, negative values).

The study concluded that the most important of which are: that the values contained in the books of Islamic education for the lowest stages of the Education need to be revisited, tested and identified. There is a need to exert more care and attention to gender-related behaviour. There is a need to take into account the balance, integration and inclusion in the distribution of values.

The study of Al-Shuhomy (2006)

The study is entitled:"The extent to which the books of Islamic education at the secondary level help in addressing peace and safety for high school students in the state of Kuwait", It aimed to identify the extent to which the books of Islamic education in the secondary phase contribute in addressing peace and safety for high school students in the

state of Kuwait, and to find out the values that help in addressing peace and safety of high school from the teachers' perspectives.

The study sample consisted of all Islamic textbooks at the secondary level in the state of Kuwait. The researcher used in his study the descriptive method through the analysis of the content addressing the teachers of Islamic education by a questionnaire in order to measure the student misconduct and identify methods of treatment. To achieve the goal of the study; the researcher described the reality of peace and safety among the students of the secondary stage from the point of view of teachers and the values that help in the treatment. The study concluded that the most important thing to highlight is the negative social behaviour. The values that help in the treatment of peace and safety are those attributed firstly to the social dimension followed by the personal dimension, followed by the religious dimension, then the scientific dimension.

Problem of the study:-

In light of the foregoing, the problem of the study is addressed in the following question:

'What are the security values to be met by the Islamic culture curricula provided for students of general education?'

Justification of the chosen problem of the study:-

A. Security values adjusts the behavior of individuals and enhances the individual, community and professional responsibility; necessitating the need to search them.

B. The Islamic education curricula maintain human culture a Muslim, as well as the past and heritage that is streamed from the Quran and the Sunnah of Prophet Mohammad (PBUH).

The objectives of the study:-

The study seeks to achieve the following points:

1. To prepare a list of the security values that should be included in the Islamic Culture curricula in general education.
2. To make some procedural recommendations necessary for the planners of curricula to improve the content of Islamic culture curricula.

The importance of the study:-

The study and its relevance to the Islamic culture themes can be highlighted as follows:

A. Scientific importance:

- Development of the content of Islamic Culture curricula in public education, and detects the extent of availability of security values that could be integrated and confirmed in the hearts of learners.
- Draw the attention of the planners of curricula and authors of educational books to security values that should be included in the books of Islamic Culture.
- Emphasize the attention to security values and the need to be addressed in the content of Islamic Culture in all institutions.

B. Practical significance:

1. Make a list of the main security values in the content material of Islamic Culture in general education.
2. Acquaint the teachers and learners with the force and effect of the reflection of the security values on their performance; to positively achieve the security values desired.
3. This study is a natural extension of studies in other areas, it is also the kernel to do similar studies in the field of security values, no one has addressed such a study in the field of Forensic Science or Humanities in general.

Definitions of terms:-

The values are all that contain integrity and constancy of human being in recognition of the lordship, the Almighty. This value is what directs the human's behavior and thoughts directly to God especially in times of distress and dismay.

Al-Sha'wan (1417H) defines the values as 'abstract concepts, metrics or standards either implicit or explicit that are used to judge something as desirable or undesirable, and guide the behavior of an individual of what is desirable by society.'

Security values:-

Security Values are a set of security standards called for by the company group to help learners guide the behavior of his/her security to sustain the community and maintain the integrity of its behavior through the content material of Islamic Culture. By studying the meanings and definitions of the values, we find that there are common features among them are summarized by Mary and Balqis (1982), Al-Jaroudi (1417H), Aqel (1427H)

- values are humane; and thus caring, pleasure, pain and thoughts are connected to the individual.
- values are subjective, anyone senses the values according to his/her criteria
- Values are relative as it varies from person to person, and from time to time, and from place to place, and from culture to culture.
- values have a hierarchy and some values have dominance over other values
- values have preferential standard that represents a reference framework and governs the actions of the human and explains his behavior.
- Values govern the human choices and his/her preferences.
- Everyone has his own value -system that is acquired through the interaction between individual knowledge and the culture in which he/she live in.

Importance of values:-

Values identify the objectives of the society and its activities, and they guide its ideology towards specific targets; no matter how scientifically and globally the nation has gone through, still it needs a system of values.

Allah (SWT) says 'What you have been given is only the fleeting enjoyment of this world. Far better and more lasting is what Allah will give to those who believe and trust in their Lord; who shun great sins and gross indecencies; who forgive when they are angry; respond to their Lord; keep up the prayer; conduct their affairs by mutual consultation; give to others out of what We have provided for them; and defend themselves when they are oppressed. The harm can be requited by an equal harm, though anyone who forgives and puts things right will have his reward from Allah — He does not like those who do wrong. (Al-Shura 42:36-40).

Therefore, Islam is the religion of faith, worship and values. Values are the ideals that we uphold and we must live by. Since we pay attention to our beliefs and worship, one should also pay attention to our values. Values give you clarity of thought and help set one's priorities. The values of Islam are considered the noblest moral principles for human development. The Prophet – peace be upon him – said, 'I am sent only to fulfill the noblest characters.' (Musnad Al-Bazzar, Hadith 8949).

The significance of values for the individual is in being the essence of the real human and without them he/she loses his humanity and becomes a beast controlled by the whims and desires. They determine the individual's paths and partnerships in life that are directed to what should be done or avoided.

Positive values are considered a fence from regression towards the lusts; and without values humans become slaves to bad habits that destroy the individual and society and threaten its security. Positive values make the individual active in his life and lead him to success and progress. They also increase the confidence of the individual and enhance his/her virtue and his/her human energy as well. They lead him/her to success and confidence. Without values, the human will be a failure.

Impact of Islamic values:-

And the Islamic values have impact on the positive behaviour of the individuals and communities. According to Al-Jallad (1430H), Al-Yemany (2009), Sallom, Taher and Jamal (2009), Islamic security values:

A- Impact on the behavior of the individual as:

- That values and standards represent the essence of the real positive human; and without them the individual loses his humanity and becomes an animal controlled by its passions and desires.
- Determine the course of individual and direct behavior in his/her life, to safety.
- Protect the individual from drifting behind the lusts and desires.
- Is the fence that saves the human from psychological, physical, social regression, and reforms him/her morally, socially and educationally.
- Provide Human with self-esteem in life and keep him full of power, away from passivity.
- Strengthen the virtue of the human and enable positive interaction with different life situations.

B. Impact on the behavior of the community as they

- Stimulate the community's survival and continuity, it depends on what values the community possesses which are the foundations and directions of positive behaviour that builds upon the progress of societies and its advancement.
- Stimulate the community's identity and distinctiveness, since they are the foundation of the culture of the society, which shape the patterns of human behavior, practice. Thus, the values are markers of distinction that distinguish the groups from each other.
- Reserve the society of negative social and moral behaviours as they are protective to the community.
- Strengthen community unity and cohesion by identifying the life's goals and ideals.
- Help the community to face the changes that occur through healthy choices.

The pioneers of Islamic education have over-emphasized the values since Imam Al-Ghazaly thinks that to achieve happiness for humanity in this world and the Hereafter is the supreme value in the moral education. Happiness in the hereafter are the main results of adherence to Islamic values. It is, therefore, necessary to take all means and ways that lead to the happiness of the humanity; which is the ultimate goal of education.

Security Values:-

In his/her life, the human is living amongst his social stratum that is not secluded from others. Although the social values shape the individual's relationship to others; the security values determine the correct path for the individual to live in stability and maintain his/her security in the community. They enhance the sense of security, especially his soul security and the rest of the members of the community generally. They help instill the values of security and dissemination in different environments or communities which prepare the individual to get health behaviour that can protect his/her nation and homeland, and secure a life free of fear, unrest and discomfort. The security values are imperative as follows:

- They achieve the nation's dignity, security and happiness .
- The absence of security values disrupt the security of all its components.
- The security values have its roots in the doctrine of the Islamic nation that determines its identity.
- In achieving the security values, then the individual, society and nation are protected from threats against their security.

The security values are so imperative in the society which desperately seeks to promote itself and direct the behaviour of the individual, society and all social groups especially the security personnel, teachers, and learners. These security values have power in adjusting the behaviour and in teaching individuals the seriousness, honesty, trust, responsibility, obedience, commitment, fight against corruption, and the development of feelings of belonging to Islamic and national levels.

Security within social security differs according to specialists depending on the proportions. Security is the society's capacity to face not only the events, facts, individual actions, but all the appearances related to the nature of things that lead to violence. Nafea (1972) explains it as 'the sense of individual and group towards the satisfaction of organic and psychological motives such as the physical, psychological security towards the contentment of the community to the demise of what threatens the manifestation of this motivation such as the permanent and stable, livelihood and the psychological motives of society's recognition of the individual and his/her role within it'. Ali (1976) sees it as 'the situation that is available when the security of the country is not breached in the form of crimes or in the image of a dangerous activity in order to take preventive measures to stop the dangerous activities.'

Therefore, security values represent the standards and principles derived from the provisions of Islam, used in guiding human behavior toward achieving security and preservation, awareness of the importance of the security, the commitment to legislations, regulations, security and respect. This is summarized in the following:

1. They are derived from divine revelation (the Qur'an and Sunnah).
2. They maintain the individual, social and national security.
3. They are moderate values.
4. They keep the legacy of the nation's cultural authenticity and identity.
5. They keep the economy of the nation locally and the globally.
6. They are connected to the individual, society and nation to the degree of nobility and innocence.

Functions of security values:-

The role of values in guiding behavior is fundamental and basic, both on the level of the individual or society because of its positive impact on the human. The system of values, if settled and assimilated by the individual, has

become the standard and authority in leading his/her positive behaviour through five basic functions as defined by Isa (1984):

1. The values provide the individual with a sense of what he is doing and geared towards to achieve its purposes.
2. They create the basis for individual and the group.
3. They are the basis for judging the behavior of others.
4. They enable the individual to know what to expect from others and their reactions.
5. They gave the human the sense of righteousness and wickedness.

Results: Domains of security values:-

The researcher has reached to a list of security values referring to the books and previous studies, and other regulations related to security matters. Six domains were identified; namely, Ideological Security, National Security, community Security, Cultural Educational Heritage Security, Military Security, Economic Security.

The first domain: ideological security:-

This value is achieved through following the Islamic faith, tolerance and commitment to the orders of the divinely orders that make the human being seek in this world and the Hereafter; and these include the following:

1. Enjoying the orders of the Islamic.
2. Cooperation in righteousness and piety.
3. Respect for Human Rights.
4. Justice in contentment and anger.
5. Promote a spirit of tolerance.
6. Refute pervert thought.
7. Link to holy shrines.
8. Achieve fidelity in the work.

Second domain: national security:-

This domain strengthens the human belonging to his/her homeland, preservation and defense of home-country and respect of its principles, and transplantation of well-being. This includes the following:

1. Boast of belonging to Islam.
2. Obeying the state representatives without disobeying Allah.
3. Good citizenship.
4. Defend issues of the nation.
5. Maintain unity and connectedness.
6. To achieve the principle of Shura (Advisory Teams).
7. Maintaining the Muslim legislations.
8. The consolidation of security and fight against crime.
9. Upgrading the security.
10. Maintaining general rules.
11. The deepening belonging to the homeland.

Third domain: societal security:-

This value binds the individual with his/her society as a whole in which they live through cooperation and commitment about being responsible towards the society without breaching the general rules in order to ensure unity among its members. This includes the following:

1. Social stability.
2. Cooperation and provoking community spirit.
3. Social integration.
4. Commitment to responsibilities.
5. Refute racism and poverty.
6. Non-violation of liberties.
7. Renunciation of defamation.
8. Avoiding rumors.
9. Maintaining legitimate rights.
10. The deployment of the principle of proportionality.

Fourth domain: cultural and educational security:-

These values tend to keep the cultural identity within positive thought in order to elevate the nation with honesty, integrity, respect, and guidance about everything that can affect that identity. This includes the following:

1. Positive Cultural openness.
2. Appreciate scientists.
3. Scientific honesty.
4. Spreading the culture of reading.
5. Respect the opinions of others.
6. Protection of the manifestations of the intellectual invasion
7. The fight against illiteracy.
8. Assist in national issues.
9. Awareness of the importance of Health prevention.
10. Instill good attitudes and conduct.
11. Awareness of the dangers of environmental pollution.
12. Awareness of the dangers of addiction (drug and alcohol).

Fifth domain: military security:-

Military values bind human beings in general with security, especially the ideals that are manifested with honesty to achieve precision and maintenance of security in which man lives safely and securely. This includes the following:

1. Respect for the principles of military.
2. Be a model of good conduct.
3. Maintaining honesty and integrity.
4. The performance of duty to the fullest.
5. Precision in the implementation regulations.
6. Well-behaved.
7. Taking into account being organized.
8. Promote self-courage.
9. Respect the rights of others.
10. The protection of the vulnerable.
11. Keeping secrets. 12. Positive leadership.

Sixth domain: economic security:-

These values tend to sustain financial stability and preservation, maintenance of public money from damage away from corrupted financial dealings in the public economy. This includes the following:

1. To instill the principle of productive work and its appreciation.
2. Provide career opportunities.
3. Maintain public facilities.
4. The preservation of public money.
5. To maintain food security.
6. The rationalization of consumption.
7. Fight goods-fraud.
8. The development of environmental resources.
9. Dealing with unemployment.
10. The optimization of wealth

Conclusion:-

Akgunduz (2016) thinks that '*religion in general and Islam in particular can play positive role in reviving common values for mankind and uniting people for justice and equality, particularly the weaker sections.*'

These days, people interact with each other much more than the past due to cultural exchanges and greater accepting of other values and norms. In each cultural and national value, there must be universal endeavor to address problems of social, educational inequalities, gender inequalities and human rights as well. Global solidarity must be maintained in order to fight injustice towards the protection of human rights. Global solidarity is vital for the protection of the environment, to respond to natural calamities and to battle poverty.

On the contrary of some alleged reasons about Islam and Muslims, Islam attaches much importance to human values, and works to establish general peace and well being. It makes firm love and brotherhood in the structure of society, and puts forward the necessity of union and harmony. It rejects determinedly movements, which obliterate the common values and genuineness between members of society, like tribalism, racialism, and negative nationalism.

Recommendations:-

There is a need to develop clear objectives and content of Islamic education books and to write them down in the introduction to these books in order to enable teachers to identify and adhere to them, and to get to achieve these values through the teaching of the content of the books. There is a necessity of building the content material of Islamic culture in accordance with the standards, principles and rules of accurate scientific targets of Islam, including Islamic security values in support of the behavioural side, the need to distribute these values appropriately to achieve sequence and integration in a more attractive, interesting with high-tech content.

The study proposes to conduct studies to identify the security values contained in the Islamic courses in the military universities and civil colleges. This will increase the sense of security and thus maintain the security of the individual, society and the nation. It is also suggested to conduct more studies on the content analysis of the decisions of the Islamic courses in general education to ensure the availability of security values. It is also proposed to conduct a study on security values included in the decisions of the police sciences that connect to young people and elevate good education which is desirable in the stages of public education.

The religion of Islam has not neglected the human security values since the human is the center of the system of creation, and everything is created for him/her. By investigating Quran and Sunna, it was concluded that Islam has paid high attention to five fields of human security including individual, social, economic, environmental and health fields paying attention to aspects such as culture, politics, society, economy, family, and judging which affect the individual, collective and international aspects. Islam considers human security values as components of the survival of human society, and as health and immunity from any danger. Therefore, realization of human security values in every society requires offering suitable solutions (Faghihi, 2015).

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RESEARCH ARTICLE

EVALUATING IMPACT OF COMMUNITY MANAGEMENT IN FOREST CONSERVATION: A STUDY ON COMMUNITY FORESTRY PROGRAM IN NEPAL.

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Abstract

Expansion of agricultural land, the growing demand for timber and the local dependency on forest firewood created long term pressure for deforestation in Nepal. This paper investigated the impact of community management on forest conservation and outlines the conversion trend of the forest outlook in Nepal in the past decades by informal interviewing 10 representative Community Forest Users Groups (CFUGs) from the middle hills of the country. The results showed notable positive changes including increased forest cover, improved forest product supply and enhanced biodiversity after implementation of the community forestry program. The main factor behind the forest restoration is sustainable management adopted by the community and control on forest fire and over exploitation. However, the adopted conservative-oriented approach hampers communities to exploit the maximum benefits. It has been recommended that market-oriented approach would help communities to get more economic benefit than the existing approach. The community led initiative in Nepal has set best example in forest conservation and this experience can be explored by other countries as well. Local people's participation is very important and necessary for common resources management. The research results provide fundamental basis for policy making process regarding common pool resources management and forest governance.

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Introduction:-

The Community forest management program in Nepal is considered as the most successful example of participatory, collaborative and sustainable forest management. This program has played a vital role in improving forest condition and conserving biodiversity and supporting the local people to enhance their livelihoods (Gurung et al., 2013; Pokharel et al., 2007; Acharya, 2004; Carter et al., 2011; Gautam et al., 2004). Over the period of two decades forest-covered area of the country has increased remarkably and condition of the forest has been enhanced after the government came up with policy and legal intervention to promote the community forestry in the country. Since management rights handover to the community from the government, 86% Community Forest Users Groups (CFUGs) reported improvements in their all forest quality (MFSC, 2013). Community forestry program is effective in increasing the biodiversity in degraded land, conserving and improving the total environmental condition and enhancing livelihood of the users by effective resource management in the middle hills of Nepal (Acharya, 2002; KC et al., 2015; Birch et al., 2014).

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By engaging one third of the country's total population, more than 18,000 CFUGs have been formed in the country and have been able to manage over 30% of the total forest in the country (MOF, 2016). Among them, 75% of community forest users groups were formed in the middle hills of Nepal. A 2013 survey report from Ministry of Forests and Soil Conservation shows that forest conditions have improved in overall since the handover to CFUGs, with 86% reporting improvements in forest conditions. 79% of the CFUGs reported an increase in tree coverage area within the community forest geographical space (MFSC, 2013). The main factor behind the forest restoration is sustainable management adopted by the community and control on forest fire and over exploitation.

Forest Resources of Nepal was mostly controlled by the government and forest administration was centralized in 1970s (Gautam et al., 2004). The idea of local involvement in forest management was conceptualized in late 1970s (Nagendra et al., 2005). As a policy innovation to address the massive forest degradation in the country, the government of Nepal endorsed Forest Rules in 1978, which allowed local governance body to manage forest land (Ojha et al., 2009). In 1982 Decentralization Act introduced the 'user group' concept and in 1988 forest master plan emphasized the concept of forest users group with legal management rights (Poudel et al., 2015). The government of Nepal enacted the Forest Act in 1993 and forest regulation in 1995 which allows to flourish the community forest users group (CFUGs) all over the country (Pokharel et al., 2007).

In the beginning, community-based management strategy was adopted in the middle hills of Nepal as an experiment, which brought successful result, afterward evolved rapidly across the country over the years gaining endorsement among the policy makers and the local forest users. In order to restore degraded forest land and cope with massive deforestation, the Nepalese government formally adopted community based management approach in 1980s, which put emphasis on the devolution of management and governance right from government to local people (Gautam et al., 2004). Handing over a forest management and governance rights to the community led to overall improvement of environment as well as brought positive transformation in the livelihood of the local people (Nepal, 2006; Acharya, 2004).

Most of the past studies (e.g., Yadav et al., 2003; Gurung et al., 2013; Kanel et al., 2004; Adhikari et al., 2007) presented the status of the community forestry in forest management program, impact on livelihood of the community, challenges and opportunities among other. Both positive and negative aspects of community forestry program have been well documented comprehensively over the past years in various research papers. However, the literature on pushing factors for reforestation and restoration are relatively insufficient in telling the latest changes of forest outlook. There is a lack of comprehensive study on influencing factors on forestry rebound in the last two decades. Latest survey on the State of Nepalese forest produced by the government of Nepal has also failed to clearly mention the community forestry program as one of the major causing factors for afforestation in the country. This paper assesses the community forestry program as a key factor behind the successful change of forest outlook in the past decades. The forest survey of different time period did not indicate and discuss the factors of deforestation and afforestation. This paper argues that the cause factor for afforestation is the adoption of the community forestry program as a main strategy in the forestry policy. The impact of community management on change in local environmental condition was assessed. Both quantitative and qualitative data were collected to evaluate the role of community management on forest conservation, qualitative data were used to interpret the conditions of the forest.

Materials And Methodology:-

Data were collected through survey and informal interviews with the members of the CFUGs and experts for this study. This study was conducted in six representative districts namely Ilam, Nuwakot, Syangja, Palpa, Surkhet and Doti covering different parts of mid-hill region of Nepal. Mid-hills account 43% of total land area of the country, with the greatest and diversified ecosystem. Additionally, about 32% of the forests in Nepal are covered in the mid-Hills. This part has extended east to west of the country between the Himalayan range in the north and the Ganges River plain in the south with an altitude range between 1000 and 3000 meters (Shrestha et al., 1992). The topography of the mid-hill zone is generally mountainous, with combination of fertile farmland, forest, shrub and grassland.

In order to examine the status of forest under the community management, 10 sample CFUGs were randomly selected from a community for one-to-one interview and survey. In the sampled areas, the majority of the populations were engaged in agriculture and dependent on forest resources such as firewood, fodder, timber and other products to sustain their local livelihood. Data were collected through informal interviews, survey among the members of CFUGs in the year 2016. It was intended to select CFUGs from the different parts of the mid-hill so that

the impacts of community management all over the country could be assessed. The forest biodiversity information was collected through the informal interviews with the members of the CFUGs.

30 interviewed members of the CF were with 100% response rate. The interview was semi-structured, with closed and open ended questions. Experts were also interviewed to examine their opinions towards the performance of the community forestry program. A survey questionnaire was designed to assess the total environmental impact of the CF program on forest conservation. To compare before and after the CF intervention, three options were presented in the survey questionnaire. The three options were 'increased or improved condition', 'same as before' and 'decreased conditions', respectively. In addition, secondary data-based discussion is also presented on environmental impact of the CF program. This discussion also provides linkage of the present results to the other studies.

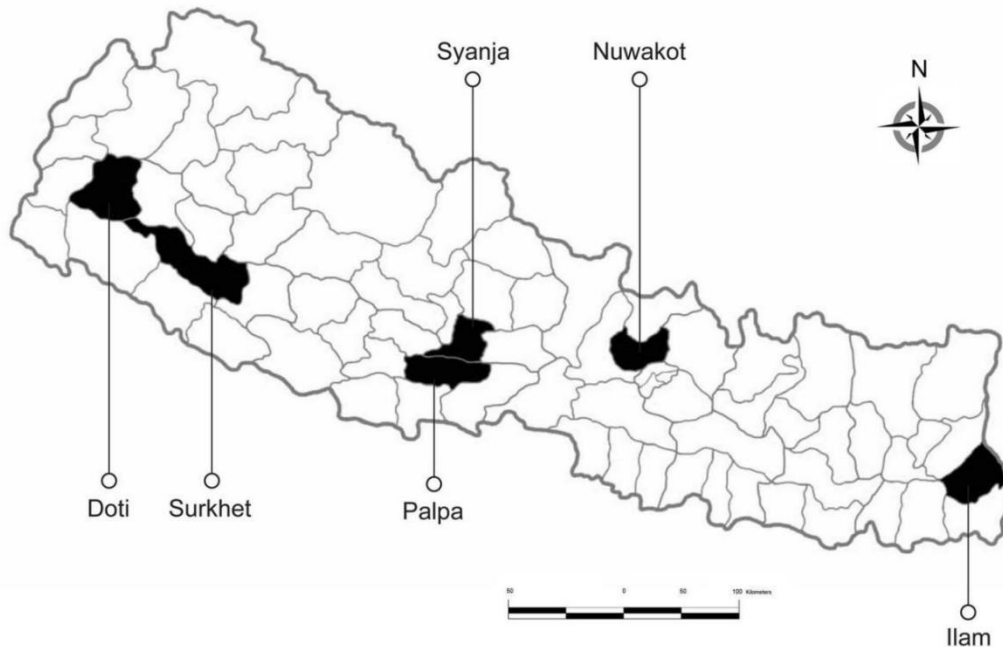


Figure 1:- The map of Nepal showing six study districts.

The main focus of the informal interview was to assess the quality of forest ecosystem before and after the implementation of community forestry program. The interview was designed to elicit forest conditions information from the users that mainly covered information on forest condition and health of biodiversity before and after CF intervention. In-depth interview is effective in collecting detailed information on significance of community management in conservation forest resources and evaluating the role in enhancing people's livelihood. The interview was designed to assess environmental impact of community management using multiple indicators. Similarly, this study also used the secondary data and reviewed literature to assess the contribution of community management. Existing literature and data related to community forestry were reviewed and analyzed to draw conclusions. The required secondary data were obtained from published reports, research papers, national data base and official records of the related agencies and stakeholders. The indicators of forest condition of this study include increasing shrub and greenery, improving biodiversity, reforestation of degraded land, availability of the resources, water resources conservation, soil erosion protection, effort to the conservation etc. The details about these conditions are presented in tables.

Limitations:-

Some of the data were collected through the informal interview and they were not quantifiable. Additionally, there is a lack of historic quantitative information on time — biodiversity for community forests. Small sample size was another limitation. However, given the objectives of the research, these limitations have not sacrificed the quality of the conclusions it has drawn. The implication of the limitations mentioned here suggests that there might be a room for further exploration or improvement on the subject matter of this study.

Results And Discussion:-

The present study revealed the improvement of forest conditions after the intervention of the Community Forestry program and CFUGs were effective in the environmental conservation. CFUGs are involved effectively in the activities of protecting the forest, managing the resources, cultivation, harvesting and benefits sharing, nursery production and planting as their own strategies and operational plan.

Deforestation in Nepal has a long history. Expansion of agricultural land, the growing demand for timber and the local dependency on forest firewood created pressure for deforestation in Nepal which lasted long (Pokharel et al., 2007). Conversion of forest to agriculture land, resource mismanagement, timber extraction for personal benefit by rulers, ineffective forest administration, unsustainable use of resources were the major causes for deforestation. The government of Nepal had already recognized alarming rate of deforestation, especially in mid-hills and Terai (lowland region in southern Nepal) in 1950s (Gautam et al., 2004). In 1964, forest and shrub covered area was 43.9% of the total land area of the country (WECS, 2010). By 1979 forest and shrub area slightly decreased to 42.7%. By the year of 1994, the total area of the forest and shrub further decreased to 39.6% of the total area of the country, while the area of forest alone was only 29% (DFRS, 1999). Over a period of 15 years (1979-1994) total forest area of the country was decreased by 23.68% or in the annual rate of 1.57% and shrub area was increased by 124.89% which was alarming rate of forest degradation over that particular period. The latest statistics show that forest and other wooded land has increased and reached 44.74%. It indicates that the forest area of the country has increased by 39.17% over a period of 20 years (1994-2014) by an annual rate of 1.95% and the area of other wooded land (shrub) decreased by 58.67% during the same period (DFRS, 2015). High decrease rate of other wooded land and increase rate of the forest area gives the clear picture of the afforestation over the period.

Table 1:- Changes in the forest and other wooded land (shrub) cover area (area in '000 ha) over time period in Nepal

	1964		1979		1994		2014		Percent change	
Type	Area (ha)	%	Area (ha)	%	Area (ha)	%	Area (ha)	%	1979-1994	1994-2014
Forest			5617	38.0	4269	29.0	5962	40.36	-23.68	+39.17
Shrub			690	4.7	1568	10.6	648	4.38	+125.53	-58.67
Total	6466	43.9	6307	42.7	5829	39.6	6610	44.74	-7.25	+12.97

Note: The total area of the Nepal is 147,181 km², percentage calculated accordingly. The data sources are WECS, LRMP, NFI and FRA.

The statistics presented in Table 1 show the changing scenario over the period of 15 years (1979-1994) when Nepal lost 1.3 million ha forest, while in the period of 1994 and 2014, the situation was reversed and 1.69 million ha forest covered area was increased.

Table 2: Number of Forest Users Group formation and Area under community forest users group

Year	Number	Area (ha)
1989	32	1281
1994	2756	112626
2005	13677	1134372
2011	15137	1340714
2016	18961	1752193

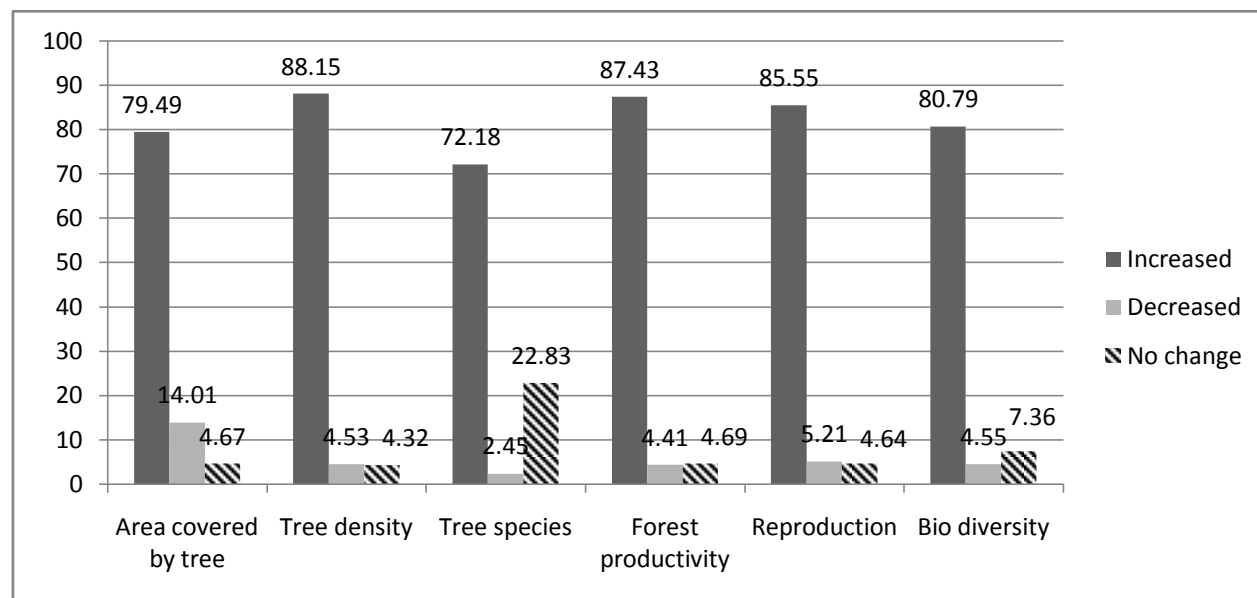
Data source: Ministry of Finance, Nepal

Table 1 and 2 depict a positive relationship of forest area rebound and CFUGs formation. Formally, community forestry program was launched in late 1980s, though practically community forest users groups were rapidly formed after 1990. In 1989, only 32 community forest users groups were established, managing only 1281 ha forest. By the year of 1994, formation of CFUGs became rapid and 2756 CF were established, managing 112626 ha by the year of 2005, the number of community forest users group extended to 13,667. By the year of 2011, the number of CFUGs increased to 15,137 managing 1,340,714 ha forest. It clearly shows that increasing number of the CFUGs across the country brings afforestation gradually. Not only forest covered area rebounded, number of stems/ha by stem diameter has also increased (Table 3), which indicate the enhancement of the forest quality. In 1960s, number of stems/ha by stem diameter was only 313, which reached 408 in 1994 and 429.93 in 2015. Similarly, carbon stock (t/ha) was 151 in 1960s which increased to 176.9 in 1994 and 176.95 in 2015.

Table 3:- Change in forest composition over time

Year	Number of Stems/ha by stem diameter	Carbon Stock (t/ha)
1964	313	151.0
1994	408	176.9
2015	429.93	176.95

Data Source: Department of Forest, Nepal .

**Figure 2:-** Changes in forest condition. Data source: MFSC, 2013

Studies have shown that there have been remarkable positive changes in community forests across the country. Changes in different dimensions of forest quality are presented in Figure 2. Figure 2 shows outstanding improvement in forest quality as a reflection of the management inputs. According to a study conducted by Ministry of Forest and Soil Conservation, since handover, 86% of CFUGs reported improvements of forest condition. Forest quality has been improved in terms of tree coverage area, tree density, species, and forest productivity, regeneration and bio-diversity. Those are the main indicators and criteria to determine the forest quality. 79.49% of CFUGs reported that tree coverage area increased after CF program intervention, only 14.01% reported that their tree coverage area has been decreased. Similarly, 87.43% CFUGs reported that forest productivity has been increased. CF program has positive effect on bio-diversity in the forests, it has helped to conserve and foster different kinds of plant species.

According to the respondents, forest was under the big threat of massive deforestation due to unsustainable use of resources in order to meet the demands for fuel wood, fodder, timber etc. before the CF program intervention. After the implementation of the program, apart from the improving forest conditions, forestry product supply and forestry income has also increased.

Table 4 describes ten community forests in the study area and provides the information regarding size, household number, forest types, forest condition and total years of forest management by communities. The investigated forests are small in size; therefore the produced forest products are not sufficient to meet the demands of local users. It was found that the condition of the forest that has been conserved for a longer period is good and has higher regeneration rate.

Table 4:- Attributes of community forest

Name of CFUG	District	Forest Area (ha)	No. of households	Years managing the forest actively	Forest Condition	Forest Type (dominant species)	Forest area per household (ha)
Thokre Firfire	Ilam	25	70	20	Good	Katus, Chilaune (<i>Castanopsis sp./schima wallichii</i>)	0.35
Choyatar	Ilam	150	170	20	Good	Pine (<i>Pinus roxburghii</i>)	0.88
Ramche Deurali	Nuwakot	17	65	21	Fair	Katus, Chilaune (<i>Castanopsis sp./schima wallichii</i>)	0.26
Salghari Silapatra	Nuwakot	37	350	16	Good	Sal (<i>Shorea robusta</i>)	0.10
Sallaghari	Syangja	27.75	27	10	Good	Katus–Chilaune (<i>Castanopsis sp./schima wallichii</i>)	1.02
Ramche	Palpa	99.7	302	24	Good	Sal (<i>Shorea robusta</i>)	0.33
Deuti Bajyei	Surkhet	749	1032	26	Fair	Sal (<i>Shorea robusta</i>)	0.72
Kalika	Surkhet	220.76	88	10	Fair	Sal (<i>Shorea robusta</i>)	2.50
Navadurga	Doti	200	73	15	Fair	Pine (<i>Pinus roxburghii</i>), Sal (<i>Shorea robusta</i>)	2.73
Ranipokhari	Doti	126.23	250	18	Good	Pine (<i>Pinus roxburghii</i>), Sal (<i>Shorea robusta</i>)	0.50

The entire sample CFUGs chosen for this study is implementing scientific forest management activities. CFUGs are promoting forest protection and plantation; prohibition of trees falling and wildlife hunting, preventing forest fire, control grazing, conserving soil and water and protecting forest from encroachment have been included in their operational management plans and programs. The primary objectives of forest managements set by all CFUGs are more or less similar to fulfilling the basic needs of forest products by using them in sustainable ways and protecting the forest resources. Income generation, conserving water sources, preventing soil erosion, maintaining greenery, protecting wildlife are the additional objectives of the CFUGs.

The activity of 'Godmel', 'Jhadifadne'¹ is an effective silvicultural practice carried out by CFUGs. This activity includes cleaning, weeding, thinning, pruning etc, which allows the favorable environment to the desired trees to establish, grow and eliminate unwanted trees and bush. Removal of shrub and unwanted wood could help to increase main crop or trees' productivity.

In six of the ten sampled community forests, the forest area per household is found to be less than national average (0.73 ha) ((Bista, Gurung, Karki, Shrestha, & Uprety, 2012). Most of the family in the sampled areas is engaged in the agriculture and dependent on forest for firewood, timber, fodder, leaf-litter etc to meet their daily requirements of cooking, infrastructure and building construction and feed to animals. Nevertheless, all studied community forests are not in the condition to supply full required forest products to the community. Generally, CFUGs distribute their forest products free of cost to the locals, therefore income of CFUGs is very low. Member household pays a very small amount as membership fee. According to them, deforestation took place exclusively before CF program intervention, and the forest condition in the studied community forests has improved since forest handover to the community from the government. The mixed forests were found in the study area, though Sal (*Shorea robusta*) is a dominant species. Most of the sampled forests are covered with Sal (*Shorea robusta*) which is commercially high valued timber species. The main aim of the community forestry program of Nepal is to improve the forest condition through the efficient management and fulfill the demand of forest products of the community as well as livelihood improvement of the poor people. The sampled communities have succeeded in improving forest condition in a sustainable way and restore the degraded forests. Reforestation is one of the major indicators to evaluate community forestry program and most of the CFUGs have achieved this goal impressively.

¹cleaning, pruning, thinning and weeding operations

The handing over management rights of forest encouraged community participation to conserve the resources and also assured the livelihood improvement through benefits generation from forest. A large number of small and medium sized trees is growing up in the sampled area because most of them were recently handed over and communities have been conserving them very well. Seven out of the 10 sampled CFUGs underwent plantation after possession of management rights of the forest in the degraded land. Since the establishment of CFUGs, no forest fire incidents have happened according to all 10 sample CFUGs which is a strong indicator of CFUGs' contribution in forest conservation and protection. Previously forest fire was the main problem. In all the CFUGs studied, the forest condition is improving and 'improving forest condition' is their most successful indicator to evaluate entire community forestry program in Nepal. Restoration of the forest from declined stage and overall indicators of the forest show that CF program is undoubtedly successful.

Data presented in Table 5 indicates that tree coverage area has been increased since the program implementation. Nine of the ten CFUGs studied clearly answered that tree coverage area has been increased and one answered that no changes have occurred. In case of regeneration of plant (seedlings and sapling), six of the 10 CFUGs have done well and four have average characteristics meaning that the forests are sustainable.

Table 5:- Indicators after intervention of community forestry program

Name of Community forest	Area covered by tree	Tree density	Tree Species	Productivity	Biodiversity	Road access to forest	Plantation	Regeneration
ThokreFirfire	Increased	Increased	No Change	Increased	Increased	Yes	No	Good
Choyatar	Increased	Increased	No Change	Increased	Increased	No	Yes (Regular)	Good
RamcheDeurali	Increased	Increased	Increased	Increased	Increased	Yes	No	Average
SalghariSilapatra	Increased	Increased	No Change	Increased	Increased	Yes	Yes	Average
Sallaghari	Increased	Increased	Increased	Increased	Increased	No	Yes(2 times)	Good
Ramche	Increased	Increased	Increased	Increased	No Change	Yes	Yes	Average
DeutiBajyei	Increased	Increased	Increased	Increased	Increased	No	Yes	Good
Kalika	No Change	Increased	Increased	Increased	Increased	No	No	Average
Navadurga	Increased	Increased	Increased	No Change	Increased	No	Yes (regular)	Good
Ranipokhari	Increased	Increased	No Change	No Change	Increased	Yes	Yes (regular)	Good

After handing over management rights to the community (as a reflection of management input), most of CFUGs regulated their forests for regeneration and adopted sustainable forest management strategies. They made effective plans and procedures themselves, implemented the plans rigorously and moved towards successful sustainable management. Amongst the CFUGs studied, tree density and productivity of the forest has been increased in all forests after adaptation of community forestry. The improvement of productivity brings many benefits to community that can be distributed for their livelihood support. As shown in Table 5 most of the CFUGs (seven among the 10) carried out plantation as a priority activity, which indicates that most of the community reforested in degraded land in a planned way. Although 3 community forests are natural forests, CFUGs conserved and managed well.

Most of the forest in Nepal regenerates very easily from seed and roots. The plantation is subjected to less priority in recent years as natural regeneration is sufficient to allow for forest restoration. According to the respondents, the harvesting system is sustainable now, which was unsustainable before the implementation of CF. After intervention of the program, tree species such as, Shorearobusta (sal), pinus sp. (sallo), Schimawallichii (chilaune),

Dalbergiasissoo (sisau) etc have been increased notably in the all CFUGs. As depicted in Table 5, about 27% respondents replied that the status of prioritized species is in good condition after intervention of the program, and 40% replied that the condition is improving. The research revealed that besides forest condition's improvement, the level of awareness of forest protection among the users has also increased after intervention of the program. Similarly, 40% of the respondents replied that the conservation efforts are good, and 23.33% responded that situation is improving whereas 23.33% responded that the conservation efforts of the CFUGs were poor.

After the program intervention, many of the CFUGs put efforts into several activities and behaviors that contributed to the forest conservation. To improve the ecosystem services, the communities focused on planning of the activities for restoration of degraded land, water resources protection, soil erosion and landslide control, biodiversity protection and greenery promotion. According to the respondents, the restoration of the degraded land was the most important issue for them. Then after, water resources conservation, biodiversity protection, soil erosion control, greenery promotion are the other factors of conservation.

Table 6:- Users perception about forest

	Good	Improving	Satisfactory	Poor
Status of prioritized species	8 (26.66%)	12(%)	7(23.33%)	3 (10%)
Conservation efforts	12 (40%)	7(23.33%)	4(13.33%)	7(23.33%)

Despite effective conservation efforts and significant improvement in the forest condition in the country, forest products are not sufficient to meet growing demands of the community. Most of the CFs studied are not in the situation to fulfill the forest product needs of the users and the revenue generation is also low. Forests do not generate enough revenue to support livelihood of the local people. CFUGs are very sensible about sustainable use of forest resources which ensures that use of resources are less than the rate of regeneration. Particularly, forests are unable to meet the demands for timber, mainly due to the immaturity of the forests to produce large amount of timber.

Most of users do not have technical knowledge about harvesting and silvicultural practices among other. They do not have technical human resource as well. Their management model somehow tends to be traditional. One of the underlying problems of the CF is most of the CFUGs adopted a conserve-oriented approach rather than taking economic benefit for the community. They harvested few forest products mainly to restore and protect the forest products. Also, the study found that most of the users are unaware of the financial opportunity from the Non timber forest products (NTFPs) to the community forest. Despite having a huge potential of commercial value, NTFPs are being ignored. Either they are selling the NTFPs in cheap price or the products are becoming useless due to no-market accessibility although, in recent years many CFUGs have shown concern towards the NTFPs and high commercial value products such as bamboo, lantusemblica (amla), cardamom, broom grass (amriso) etc. CFUGs prioritized tree species of high commercial value as required, established nursery and carried out plantation. For example, DeutiBajyei CFUGs planted Sisau (Dalbergiasissoo) Khayar (Senegalia catechu) in 47.5 ha because of their high commercial value.

There are still many challenges for community forestry program towards making it more successful. CF of Nepal is based on protection approach rather than market oriented approach. CFUGs, however, are not far from criticism over few problems regarding representation and benefit sharing.

As depicted in the Figure 3, the majority of the CFUGs member responded that forest products' availability has been increased after formation of the CF. The study examined the member's perception regarding forest condition and outcomes of the CFUGs efforts. They answered that the CF program was raising their living standards and easing the livelihood. Local people use the forest for many purposes such as fulfilling their fuel wood needs for cooking and heating. Timber and poles are highly valued as commercial forest products, which are used for house and other building construction and making agriculture tools. In recent years, seven of the 10 studied CFUGs identified some non-timber forest products as commercial products such as fruits, vegetable, herbs etc. All the studied community forests supply timber to the users. Agriculture and livestock are the integral part of the household livelihood of the rural Nepal. So, there is huge demand for grass, leaf-fodder and leaf-litter. Leaf-litter is used for animal bedding and also used for compost fertilizer making. Particularly, users responded that fire wood, grass/leaf-fodder and leaf-litter have been increased significantly, whereas herbs and non-timber forest products have not been increased in the similar ways.

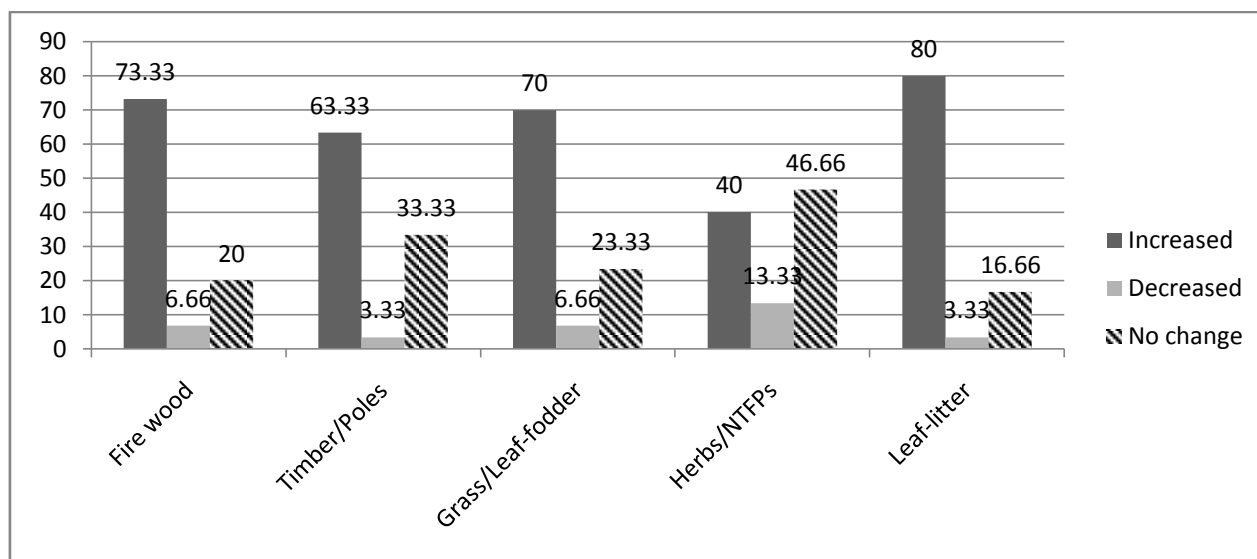


Figure 3:- Change in availability of forest products

CFUGs are legally autonomous to make decisions regarding the management action. Nepal made evident innovation in people's participation in forest governance practices (Ojha et al., 2008). Forest resources belong to the CFUGs though the property (land) right belongs to the state. Most of the CFUGs selected for this study have clear procedure for operation and benefit allocation of the forest resources. Ideally, CFUGs practice the participatory approach and involve the women, marginalized group and poor in the decision making process (Bijaya et al., 2016; Pokharel et al., 2007). However, there have been some criticisms that CFUGs are not inclusive and elite group dominate in the decision making and benefit sharing process (Yadav et al., 2003; Bista et al., 2012; Pokharel and Nurse, 2004). The study found that CFUGs practice the democratic norms to elect the executive body for operation and management of the CF. It is a strong institution for social inclusion and supporting local democracy. CF program is considered as a vehicle for wider social-economic development of the community which helps for the improvement of community development such as school, drinking water, roads, irrigation etc (Yadav et al., 2003). Most of CFUGs contribute to build social infrastructure. Apart from financial support, CFUGs contribute in raising social awareness on health, education, and forest protection in local level.

The major gain from community forest is quality improvement of forest resources and improving the livelihood of the people. Many researchers (e.g., Pandit et al., 2011; Adhikari et al., 2007; Bijaya et al., 2016; Yadav et al., 2003), CFUGs and Government officials agree that there have been significant positive changes in forest quality since community forestry was introduced. Not only ecological benefit, community forestry program has contributed to improve the livelihood of the local people. According to in-depth national survey conducted by Nepalese Ministry of Forests and Soil Conservation, households in 137 CFUGs across the country mentioned that community managed forests have increased incomes of the rural poor, women and Dalits (marginalized group) (Pandey et al., 2015).

The study found that in CFUG, the community develops the management rules and gets approval from the District Forest Office. They do occasional planting, undergo silvicultural operations on a regular basis. In community forest, harvesting of timber is restricted, harvesting of grasses, leaf litter, fallen wood are generally permitted as per rules and regulations. Time to time CFUGs extraction and distribution or sale of the timber and non-timber forest products are CFUGs' major benefit sharing activities. Local people monitor the forest as a volunteer on a rota basis. CFUGs do not have rights to sell or transfer the land itself because property rights are protected by the governments/state.

After the intervention of the community forestry program, the forest conditions have improved. Similarly, water resources, biodiversity, forest productivity, greenery have also improved (Pokharel et al., 2007). Communities are focused on creation of good atmosphere to enhance the ecosystem services. Despite low commercial benefit from the forest, livelihood of the users has improved gradually as they have access to fodder for their cattle, timber for making home and fire wood for cooking and heating.

Conclusions:-

Community forestry is an appropriate participatory resource management approach to conserving and utilizing forest in a sustainable way. This program is successful in restoration of degraded land, improving forest condition, conserving biodiversity and supporting livelihood of the local community by providing forest products. After two decades of successful management practice by local communities, Nepal gets rapid growth on forest conservation.

In Nepal most of the communities are practicing passive management system that brings few outcomes due to lack of broad knowledge and management skills. Additionally, most of the CFUGs are not aware of potential of their forests. Shift from protection-oriented to market-oriented approach is another major issue that needs to be addressed. Due to conservation approach, revenue generation of the forest is very low and livelihood support from the forest is also less than expected. Benefits for the local users are equally important as conservation of the resources. It has been reported that elite domination on decision making process has suppressed the equitable benefit sharing and social justice, this issue needs to be addressed immediately. Community forest program is likely to be beneficial to mitigation and adaptation to address the consequences of climate change. Vegetation and greenery of the forest has increased after implementing the community forestry program that contributed to the increase of carbon stock and carbon sequestration.

CFUGs are conserving their forests and using resources in a sustainable way and improving their livelihood in the mid hills of Nepal. In nutshell, this study concludes that the CF program brought significant changes to the outlook of Nepalese forestry sector by reversing the previous tendency of massive forest degradation that has resulted into preventing negative consequences. The program has improved forest condition in terms of greenery and tree coverage area and contributed to the livelihood of the people in a sustainable manner. However, it has been recommended that market-oriented approach would help community to get more economic benefit than existing approach. Community led initiative in Nepal has set best example in forest conservation and this experience can be explored by other countries as well.

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RESEARCH ARTICLE

COMPARISON OF CHEMICAL COMPOSITION, MAJOR METALS AND VITAMIN C OF CAMEL AND COW RAW MILK.

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Abstract

This study includes the determination of the contents of some of the major components of camel's milk; and to compare them with their counterparts in cow's milk. The samples were analyzed chemically to determine the percentage of Protein, Fat, Total Solids (TS), Solid Non Fat (SNF), Lactose and Ash contents. Also included in the present study, is a comparative investigation of vitamin C (ascorbic acid) concentrations in samples of both types of milk by redox titration using iodine solution. For essential metals the raw milk sample was digested by wet digestion system, and then the metals (Na and K) were determined by flame photometer in the solutions of digested samples while, (Ca and Mg) were determined directly by flame atomic absorption spectrophotometer (FAAS). The investigation showed that Protein, Fat, Total Solid, Solid Non Fat; Lactose and Ash content were low in camel milk as compared to cow milk. Also, the major metals were higher in cow milk, but ascorbic acid content is high in camel's milk.

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Introduction:-

Milk is an excellent source of most essential minerals for human. It is part of a healthy diet. The composition of the milk of various animal species differs, but in every case it has a high priority in human nutrition ⁽¹⁾. Milk also contains antibodies which protect the young mammal against infection ⁽²⁾. Cow's milk has long been considered a highly nutritious and valuable human food and is consumed by millions daily in variety of products one can also use milk from goats and camels.

Chemically milk is described as an emulsion of fat in watery solution of sugar, mineral salts with protein in a colloidal suspension ⁽³⁾.

Sudan possesses large wealth of animal livestock of which camels constitute more than three million heads raised in north of 12° N latitude ⁽⁴⁾.

There are several studies have reported the essential components in various types of animal milk.

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Ghada Z. A. Soliman⁽¹⁾ was comparison studies of chemical and mineral contents of milk from human, cow, buffalo, camel and goat in Egypt, Asif Mahmood and Sumaira Usman⁽²⁾ were researched carried out to compare the physicochemical parameters of milk samples of four different species like buffalo, cow, goat and sheep, Sabahelkhier et.al⁽³⁾ were comparative determination of chemical constituents between human, goat, cow, camel and sheep milks, Omer and Eltinay⁽⁴⁾ were evaluated the changes happen in the gross composition of camel's raw milk during storage, AbouDonia⁽⁵⁾ was determined chemical composition of raw milk, A. Enb et.al⁽⁶⁾ were researched of Whole buffalo's and cow's milk as well as dairy products manufactured from them were analyzed for chemical composition and metal contents, Rihab A. Hassan et.al⁽⁴⁾ were studied the chemical composition and microbial contents of Gariss (fermented camels milk) collected from nomadic and transhumance herders. , Z. FARAH et.al⁽⁷⁾ were reported the vitamin content of camel milk, G. Konuspayeva et.al⁽⁸⁾ were studied the variability of vitamin C content in camel milk in Kazakhstan, M. Sikiric et.al.⁽⁹⁾ were determined of metals in cow's milk by flame atomic absorption, Y.W.Park et.al⁽¹⁰⁾ were researched the physicochemical characteristics of goat and sheep milk, H.E. Mohamed et.al.⁽¹¹⁾ were studied ascorbic acid concentrations in milk from Sudanese camels was done to describe the associations between vitamin C concentrations in milk, and either breed and stage of lactation, Semaghiul Birghila et.al⁽¹²⁾ were determined of the major and minor element in milk through ICP-AES, FARID et.al.⁽¹³⁾ were determined of trace elements in cow's milk in Saudi Arabia, S.Tautkus et.al⁽¹⁴⁾ were determined of Sr micro amounts in milk by flame atomic absorption spectrometry .

The objective of this study is to compare nutritive ingredients in cow and camel's milk available for Al Hajj Yusuf area consumers, in Khartoum state (Sudan) . These include determination of some of chemical composition; the major metals include Sodium , Potassium , Magnesium and Calcium, also determination of ascorbic acid by redox titration.

Experimental Procedure:-

Sampling:-

The row milk samples were collected at milking from cows and transferred into clean bottles (250 cm³), and stored in ice tank, then transported to analysis.

Chemical Analysis:-

A chemical constituent as Fat % was determined by Gerber method and the protein % was determined by Kjeldahl method described by AOAC (1990)⁽¹⁵⁾. Similarly, total solids %, ash %, solid non fat and lactose% also described by AOAC (1990).

Fat content:-

The Gerber method was used to determine fat content in milk. In clean dry Gerber tube Sulphuric acid (10 cm³) was poured into Gerber tube and then 10.94mL of milk sample was slowly added into Gerber tube followed by 1mL of amyl alcohol. The contents of Gerber tube were thoroughly mixed till no white particles could be seen. The Gerber tube was centrifuged at 1100 rpm for 5 minutes. Gerber tube was transferred to a water bath at 65°C for at least 3 minutes and the percent fat was recorded directly from the Gerber tube scale.

Total solid:-

The total solid contents were determined according to the modified method of AOAC (1990), 5 cm³ ml of the milk samples were weighed in dry clean flat bottomed aluminum dish and heated on a steam bath for 10-15 minutes .The dishes were placed in an oven at 100° C for 3 hour. Then was cooled in desiccators and weighed quickly. Weigh was repeated until the difference between the two reading was <0.1mg.the total solid (TS) contents were calculated as fallow:

$$T.S\% = W1/W2 \times 100$$

W1=Weight of sample after drying

W2=Weight of sample before drying

Ash content:-

he ash content was determined according to the method described in the AOAC (1990); 5 cm³ of the sample were weighed in crucible and evaporated to dryness on a steam bath. The crucibles were then placed in muffle furnace at 550-600°C until ashes were carbons free (2-3hour), and then crucible were cooled in a desiccators and weighed. The ash content was calculated using the following equation:

Ash % = $W1/W \times 100$

W1=weight of ash

W=weight of sample

Protein content:-

The protein content of milk samples was determined according to Kjeldahl method as described by AOAC (1990). 10 cm³ of each milk samples were poured into clean dry Kjeldahl flask. Kjeldahl tablets of CuSO₄ and concentrated H₂SO₄ (25 cm³) were added to the flasks. The flasks were heated until clean solution were obtained (2-3hours) and left for another 30 min. the flask were then removed and allowed to cool.

The digested milk samples were poured into volumetric flask (100 cm³) and dilute with distilled water. Then 15 cm³ of 40%NaOH was added to flask and content of flask was distilled. The distillate was received in conical flask (100 cm³) containing 10 cm³ of 2% boric acid plus three drops of indicator (bromocresol green+ phenolphthalein red)

The distillation was continued until the volume in the flasks was 75 cm³ then the flasks were removed from distillatory. The distillate was then titrated with 0.1 HCL until the end point (red colour) was obtained.

The protein content was calculated as follows:

$$N\% = T \times 0.1 \times 0.014 \times 100 / W$$

$$P\% = N\% \times 6.38$$

T=reading of titration

W=weight of original sample

N=total nitrogen

P=total protein

Lactose content:-

The Lactose content was determined by subtracting the sum of protein%, fat% and ash% from the total solid %. Fat from total solids

$$\text{Lactose\%} = \text{total solid \%} - (\text{protein\%} + \text{fat\%} + \text{ash \%})$$

Solid Non Fat (SNF):

The Solid non fat was determined by subtracting Fat from total solids.

$$\text{SNF\%} = \text{T.S\%} - \text{fat\%}$$

Determination of Concentration of Vitamin C (Ascorbic acid):

The vitamin C (Ascorbic acid) was determined in milk by redox titration⁽¹⁶⁾.

0.005 mol /L Iodine and 0.5% starch indicator solutions were prepared.

Preparation of milk samples:

20 cm³ of milk solution were transferred into a 250 cm³ conical flask and then added about 150 cm³ of distilled water and 1 cm³ of starch indicator solution. Then titrated against 0.005 mol/L iodine solution.

Determination of the major metal:

Calcium and Magnesium metals were analysed by flame atomic absorption (FAAS)⁽⁹⁾, while the Sodium and Potassium measured by flame photometer in each samples of milk.

All glassware was washed with detergent and water. After being rinsed with water for several times, it was soaked in 20% HNO₃ (v/v) for 24 h, and Then were soaked again in 20% HNO₃ (v/v) for 24 h. The glassware was then rinsed several times with deionized water, and dried. Deionized water obtained from a Milli-Q system, Hydrochloric acid (37%), Nitric acid (65%), and Hydrogen peroxide (30%) analytical grade (DFCL-Hindi).

Sample Digestion and Preparation of Analytes Solution for AAS and flame photometer:

The milk sample needs to be brought into clear solution to eliminate the organic part of milk, for analysis by the Atomic Absorption Spectrometer and flame photometer. For this reason the milk sample was first digested with chemicals^(9,13) where the organic matrix of milk was destroyed and left the element into a clear solution. Wet digestion method (i.e. digestion with nitric, sulfuric and perchloric acids) has been used in the present study⁽¹³⁾.

Known volume of milk (100ml) was evaporated dryness. About 1 gm sample was transferred into a 125 ml conical flask. A 10 ml HCl-H₂O₂ [1 + 1] mixture was added to it, and the flask was covered with a watch glass. The sample

was heated on a hot plate at 100°C for about 2 h, bringing it to a gentle boil. The digested solution was filtered into a 100 ml volumetric flask through 125 mm filter paper, and diluted with deionized water⁽¹⁷⁾.

Determination of Ca and Mg by Flame- AAS:

Instrumentation:-

A Shimadzu analyst 6800 model (Japan) flame atomic absorption spectrometer equipped with hollow cathode lamps was used for the analysis. The following Instrumental conditions for the determinations of metals in raw camel and cow's milk are given in Table (1).

Table (1):- Operation condition for determination Ca and Mg by FAAS:

<i>Operation condition</i>	<i>Ca</i>	<i>Mg</i>
Lamp current flow rate [mA]	10	8
Wavelength [nm]	422.7	285.2
Slit width [nm]	0.5	0.5
Fuel gas flow rate [L/min]	2.0	1.8
Flame type	Air-C ₂ H ₂	Air-C ₂ H ₂
Burner height	7	7

Determination of Sodium and Potassium by Flame Photometry:-

Instrumentation:-

JENWAY model PFP7 (UK) flame photometer for the determination of sodium and potassium concentrations was used (table 6).

Result and discussion:-

Results of chemical composition of camel and cow's milk samples which were collected from farms in Al Hajj Yusuf city were showed in Table (2). The means of Fat, protein, lactose, T.S, SNF and ash were in camel's milk [2.9, 3.09, 2.42, 9.52, 6.62 and 0.6 %, respectively] lower than those detected with cow's milk [4, 3.89, 3.5, 12.3, 8.3 and 0.7 %, respectively].

Table (2):- chemical composition of camel and cow's milk (%):

	Fat%	Protein%	Lactose%	Total solid%	Solid non fat%	Ash%
Camel milk	2.90±0.13	3.09±0.03	2.42±0.11	9.52±0.12	6.62±0.02	0.6±0.00
Cow milk	4.00±0.07	3.89±0.04	3.50±0.01	12.3±0.05	8.3±0.03	0.7±0.02

The Table (2) indicated the chemical content of cow milk is higher than camel milk. Cow's milk seems thicker than camel's milk because it generally contained higher total solids than camel's milk; fat content of cow's milk is higher than camel's milk. Because of its high fat content, cow's milk had considerably higher energy value than camel's milk. These results are agreement with those values reported by Ghada Z. A. Soliman⁽¹⁾ and Sabahelkhier et.al.⁽³⁾. these results are comparison with other results shown in table (3).

Table (3):- comparison of milk chemical content with other reported content:

Animal milk	Fat%	Protein%	Lactose%	Total solid%	Solid non fat%	Ash%	Reference
Camel milk	3.6	2.59	4.30	11.7	-	0.75	3
	2.35	2.06	4.41	9.78	7.45	0.94	4
	3.1	4.0	5.6	-	-	0.8	7
	4.85, 3.46	2.32, 2.58	-	11.29, 9.81	-	1.30, 0.87	9
	2.9- 5.38	3.01- 4.0	3.36- 5.8	-	7.01- 10.36	0.6- 0.8	10
	2.9-5.5	2.5-4.5	2.9-5.8	-	8.9-14.3	0.35-0.95	11
Cow milk	3.44-4.96	2.98-3.87	4.08-5.00	11.23-14.26	-	0.40-0.8	2
	3.75	3.40	4.80	12.8	-	0.71	3
	3.20	3.20	5.00	12.10	-	0.65	6
	4.6	3.4	4.9	-	-	0.7	7
	3.6	3.20	4.70	-	9.00	0.7	15

Vitamin C or ascorbic acid is a water soluble antioxidant that plays a vital role in protecting the body from infection and disease. It is not synthesized by the human body.

The content of vitamin C in raw camel and cow milk was studied; it showed in Table (4), the level of ascorbic acid in camel milk is higher than the cow milk; it plays a major part in the medicinal reputation of camel milk. Ascorbic acid is highly unstable especially with temperature change. these result for camel milk are support with given by FARAH et.al⁽⁷⁾ (26.2-61.1 mg/L) and G. Konuspayeva et.al⁽⁸⁾ (15-435mg/L), but different from Mohamed H.E. et al⁽¹¹⁾ had determined according three breeds of camels and affected by the stage of lactation.

In this study vitamin C was determined in row milk by different method from other reports (redox titration) whereas the other authors as Farah et al.⁽⁷⁾ had analyzed frozen milk by HPLC (High Performance Liquid Chromatography) and G.Konuspayeva et.al⁽⁸⁾ were quantified vitamin C on fresh milk by the colorimetric method using 2,6-dichlorophenolindophenol [2,6-DIPh]. Therefore, the observed differences in vitamin C milk concentration could be partly explained by the analytical conditions, and probably also by the analytical methods used.

Table (4):- The vitamin C of camel and cow's milk

Animal milk	Vitamin C mg/100ml
Camel milk	18.4±2.11
Cow milk	5.3±1.43

The results of metals analysis of camel and cow's milk are given in Table (5) and table (6). The concentration levels of major metal in row cow milk observed in this study are higher than those observed in raw camel milk.

Table (5):- Analytical characteristic for determination Ca and Mg by FAAS:

Parameter	Ca	Mg
Regression line	$y = 0.257x + 0.030$	$y = 0.276x + 0.001$
r^2 correlation coefficient	0.99	0.99
SD Standard Deviation	0.025	0.0026
Slope ± SD	0.0045	0.0043
intercept± SD	0.022	0.0027
LOD (Limit of detection) mg/ml	0.33	0.015

Table (6):- the major metals of camel and cow's milks (ppm):

Metal	Ca	Mg	Na	K
Camel milk	73.4	6.54	13.25	135
Cow milk	128.28	12.96	51	149

The differences in the values of nutritive contents obtained in this study compared to their analogues in other studies can be attributed to differences in bred species, feeding condition, hygienic follow up, water availability, and seasons of the year. The camel milk didn't coagulate after 2 months of standing, at room temperature and that shaking was enough to bring the milk back into its original form.

Conclusion:-

There are differences in constituents of camel's milk compared to cow's milk. The most important property of camel milk is the high value of vitamin C. This high value contributes to a consideration that camel milk has a stimulating effect on the human immune system, provides sufficient vitamin C for people living in the desert, and presents normal acidity unfavorable for bacteria growth. The camel milk can be kept for longer periods than cow milk. Raw cow milk becomes sour after 2 days. Raw camel milk is richer vitamin C, so the camel milk is very much more nutritive and hygienic for human body.

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RESEARCH ARTICLE

EVALUATION OF LEFT VENTRICULAR DIASTOLIC FUNCTION IN PREGNANCY INDUCED HYPERTENSION AND ITS COMPARISON WITH NORMOTENSIVE PREGNANT ASIAN INDIAN FEMALES.

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Abstract

Introduction: Pregnancy related increased preload alters the diastolic properties of the left ventricle which have negative effects on maternal health. Scarcity of data exist on the diastolic properties of left ventricle in pregnancy induced hypertension.

Objectives: To evaluate the Left ventricular diastolic dysfunction in pregnancy induced hypertension and its comparison with normotensive pregnant females using standard 2D echocardiography and Doppler studies.

Methods: This is a prospective study done in Department of Cardiology, KLES Dr Prabhakar Kore Hospital, Belagavi, India. The study population consisted of 100 hypertensive(H) and 100 normotensive(N) pregnant females which were evaluated by 2D Echocardiography and Doppler parameters for diastolic dysfunction of Left ventricle.

Diastolic parameters studied were mitral valve E wave, A wave, E/A ratio, isovolumetric relaxation time (IVRT), E deceleration time, Deceleration slope, pulmonary vein systolic (PVs), diastolic (PVd) and its ratio, E wave velocity time integral, A wave velocity time integral and tissue Doppler study of the mitral valve annulus.

Results : Majority of the women in group N(94%) had normal diastolic function that is grade 0, compared to group H(56%).

This difference was statistically significant ($p < 0.001$).

Most significantly associated characteristics with diastolic dysfunction included MV: E/E' ($p = 0.004$), IVRT ($p < 0.001$), Deceleration Slope ($p < 0.001$), PVa(<0.001) and PVs(0.015).

Mean grade of diastolic dysfunction in hypertensives was significantly high compared to normotensives ($p < 0.001$).

Conclusions: This study concludes that there is significant incidence of diastolic dysfunction in pregnancy induced hypertension as compared to normotensive Asian Indian females.

LV inflow E/E', Deceleration slope, IVRT and Pulmonary vein flow velocities are most significant echocardiographic characteristic associated with diastolic dysfunction in pregnancy induced hypertension.

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Introduction:-

Pregnancy introduces a plethora of hemodynamic challenges to the maternal cardiovascular system which is further intensified in pregnancy induced hypertension. Pregnancy induced hypertension includes gestational hypertension, pre-eclampsia, and eclampsia. It is associated with increased maternal morbidity and mortality. The incidence of hypertensive disorders ranges from 2-8% of all pregnancies and contributes to 9% of maternal mortality in Asia and 12% in India.^{1,2}

Pre-eclampsia involves multiple systems in mother and can lead to fetal mortality and morbidity secondary to uteroplacental insufficiency.³

Gestational hypertension is diagnosed when the BP exceeds 140/90 in the absence of proteinuria or pathologic edema in a previously normotensive female at or after 20 weeks of gestation. Pre-eclampsia is considered severe when proteinuria exceeds 4gm/24 hours, blood pressure is or more than 160/110 and/or severe headache, visual disturbances or epigastric pain is noted. Eclampsia is the development of generalized seizures in a patient of pre-eclampsia.¹ The incidence in primigravidae is about 10% and in multigravidae about 5%.⁴

Pregnancy induced hypertension increases cardiopulmonary morbidity and has a higher prevalence of myocardial contraction band necrosis at autopsy suggesting that pre-eclampsia may be associated with potentially significant myocardial damage.⁵

During normal hemodynamic adaptations there is some degree of diastolic dysfunction even in normotensive pregnancy which gets further exaggerated in pre-eclampsia leading to increased cardiovascular morbidity and mortality.^{6,7,8} Normal diastolic function allows adequate filling of ventricles during rest and exercise without an abnormal increase in filling pressures. Initial diastolic dysfunction is impaired relaxation and with further progression of disease, there is mild to moderate increase in left atrial pressure and the mitral inflow velocity pattern appears similar to a normal filling pattern (pseudo normalization). However with further progression of the disease, the pattern becomes restrictive and finally irreversible. Doppler echocardiography is an excellent and routinely used non-invasive tool to assess the diastolic function of the heart.

This study is designed to find out the prevalence of diastolic dysfunction in pregnancy induced hypertension as compared to normotensive pregnant women by 2D Echocardiography and Doppler indices.

Methods:-

A case control study was performed between January 2015 and January 2016 in the Department of Cardiology, KLEs Dr Prabhakar Kore Hospital and Medical Research Centre, Belagavi. 100 singleton pregnant women with gestational hypertension, were enrolled for the study during the third trimester of pregnancy and were matched with 100 controls. Gestational hypertension was diagnosed according to Davey and MacGillivray.⁹ For each patient with gestational hypertension, a normotensive woman was matched. Approval of the University Ethics Committee was obtained, and written informed consent was collected from all patients.

Exclusion criteria included the following: undetermined gestational age, history of heart disease, undetermined development of hypertension, previously on antihypertensive treatment, and comorbidities namely diabetes, renal failure, anaemia and not in sinus rhythm. Blood pressure was measured from right arm in supine position.

Echocardiographic evaluation:-

2D Echo and Doppler data were obtained in patients in left lateral position with a 2.0 to 80MHz transducer interfaced with a commercially available echocardiographic machine (Phillips IE33). Hypertensive patients were studied prior to initiation of treatment. Severity and grading of diastolic dysfunction was done by calculating Doppler

indices namely transmitral and pulmonary venous flow patterns recorded in the apical 4-chamber view. Mitral flow velocities were detected by placing the sample volume between the tips of the mitral leaflets.

The following variables were measured:

1. Peak flow velocity in early diastole (E wave) and during atrial contraction (A wave);
2. Peak E/A ratio;
3. E- and A-wave time velocity integrals (E-VTI, A-VTI);
4. Deceleration time of the E wave (DT); and
5. Left ventricular isovolumetric relaxation time (IVRT).
6. Pulmonary venous flow velocities namely during ventricular systole (PVs) and its time-velocity integral (PVs-VTI); peak pulmonary venous flow velocity during ventricular diastole (PVd) and its time-velocity integral (PVd-VTI); peak pulmonary venous flow velocity at atrial contraction (PVa) and its time-velocity integral (PVa-VTI); and the duration of the PVa (dPVa).

Statistical Analysis:-

Data was analysed using SPSS statistical software version 20.0 The categorical data was expressed as rates, ratios and proportions and comparison was done using either Chi-square test or Fisher's exact test. The continuous data was expressed as mean \pm standard deviation (SD) and comparison was done using independent sample 't' test. A probability value ('p' value) of less than or equal to 0.05 was considered as statistically significant. Observer bias was avoided by taking in measurements by 2 different observers from random selected patient data recordings.

Results:-

The present study included a total of 200 pregnant women that is, 100 women with hypertension (Group H) and 100 women with normal pregnancy (Group N). The comparison of patient and echocardiographic characteristics of the study population is tabulated in Table 1.

Table 1:- Patient and Echocardiographic characteristics of the study population

Characteristics	Group N (n=100)		Group H (n=100)		p value
	Mean	SD	Mean	SD	
Age	23.06	2.92	23.48	2.38	0.032
Gestational age (weeks)	35.80	0.57	35.78	0.51	0.044
MV:E	5.60	1.80	8.11	9.05	0.060
MV: A	57.70	25.04	56.75	27.93	0.858
MV: E/A	1.25	1.48	1.39	1.58	0.643
MV:E'	7.23	3.01	8.53	4.55	0.096
MV:A'	9.99	5.53	9.36	3.27	0.491
MV:E/E'	33.41	54.32	10.02	25.29	0.004
IVRT	144.02	20.15	119.96	31.91	<0.001
DT	130.88	29.04	173.24	152.68	0.059
DS	294.73	166.10	487.92	208.62	<0.001
PVs	43.30	13.38	50.24	14.52	0.015
PVd	43.98	12.45	46.64	13.85	0.315
PVa	31.58	8.00	40.52	11.31	<0.001
PVs/PVd	1.12	0.43	1.16	0.38	0.591
Systolic Blood Pressure	121.88	5.51	140.16	12.47	<0.001
Diastole Blood Pressure	79.70	9.40	86.00	7.44	<0.001

It was observed that, the mean maternal age and gestational age were comparable in both the groups ($p < 0.05$). Systolic ($p < 0.001$) and diastolic ($p < 0.001$) blood pressure was significantly high in group H compared to group N. Amongst all the Doppler characteristics MV: E/E' ($p = 0.004$), IVRT ($p < 0.001$), DS ($p < 0.001$), PVs ($p = 0.015$) and PVa ($p < 0.001$) were significantly higher in Group H when compared to Group N.

On the basis of the above mentioned Doppler characteristics the grade of diastolic dysfunction was evaluated. In the present study majority of the women in group N had normal diastolic function that is grade 0 in 94% compared to 56% in group H. This difference was statistically highly significant ($p < 0.001$). (Table 2)

Table 2:- Distribution of the study population according to Grade of diastolic dysfunction

Grade of Diastolic dysfunction	Group N (n=100)		Group H (n=100)		χ^2	p value
	No.	%	No.	%		
0	94	94.00	56	56.00	19.379	<0.001
1	6	6.00	40	40.00		
2	0	0.00	4	4.00		
Total	100	100.00	100	100.00		

Discussion:-

Pregnancy induced hypertension is characterized by progressive hypertension during gestation and may be associated with pathological edema and proteinuria. It complicates 2-8% of pregnancies.^{1,2}

Pregnancy induced hypertension has a placental pathogenesis with acute onset of cardiovascular manifestations secondary to generalized endothelial dysfunction and vasospasm resulting in hypertension and multi-organ hypoperfusion. The major scientific societies provide different criteria for the diagnosis of pregnancy induced hypertension. Common to all is new-onset hypertension (≥ 140 mm Hg systolic blood pressure [SBP] or ≥ 90 mm Hg diastolic blood pressure [DBP]) arising after 20 weeks of gestation and complete resolution within 12 weeks postpartum. The term pre-eclampsia and eclampsia indicates presence of proteinuria and generalized tonic clonic seizures associated with hypertension respectively. It is one of the leading causes of maternal morbidity and mortality worldwide, and delay in the treatment of severe hypertension and diagnosis of pre-eclampsia complications contribute significantly to maternal mortality.⁵ Multiple exceptional and exclusive changes in cardiac structure and function have been described in pre-eclampsia, suggesting that these women display abnormal cardiac adaptation to pregnancy and hence towards long-term postpartum cardiovascular disease (CVD). The development of pre-eclampsia and eclampsia is now considered a risk factor for long-term CVD.⁵

The data on changes in diastolic function in pregnancy induced hypertension is limited and studies have reported varying levels of diastolic function across various stages of gestation.¹⁰

Therefore, this study was undertaken to assess changes in diastolic function in pregnancy induced hypertension and compared to normal pregnant women by 2D echocardiography and Doppler evaluation.

This was a one year prospective study, conducted in the Department of Cardiology with 200 pregnant women that is, 100 women with pregnancy induced hypertension (Group H) and 100 women with normal pregnancy (Group N). All the women underwent 2D echo for the evaluation of diastolic function.

In this study majority of subjects were between 21 to 25 year of age i.e. 66% in Group H and 62% in Group N. Also the mean age in group H was 23.48 ± 2.38 years and in group N it was 23.06 ± 2.92 years ($p=0.032$) suggesting that the age distribution of the study population in group H and group N was comparable.

In this study, 92% women in Group H and 78% in Group N had gestational age of 36 weeks. The mean gestational age in group H was 35.78 ± 0.51 years and in group N it was 35.80 ± 0.57 years ($p=0.044$). Hence the gestational age in group H and group N was comparable. These findings rule out the possible bias in study results.

In the present study 2D echocardiography revealed majority of the women in group N with normal diastolic function that is grade 0 in 94% while in group H 56% of the women have normal diastolic function. This difference was statistically highly significant ($p<0.001$).

Furthermore, Grade 1 diastolic dysfunction was present in significantly higher number of women with pre-eclampsia compared to women with normal pregnancy (40% vs 6%) and Grade II diastolic dysfunction was noted in 4% of the women with pre-eclampsia while none of the normal women was found to have Grade II diastolic dysfunction.

Hence these findings suggest that, pregnant women with pre-eclampsia are likely to have diastolic dysfunction as assessed by 2D Echocardiography.

In this study Doppler parameters including MV: E/E' ($p = 0.004$), IVRT ($p < 0.001$), DS ($p < 0.001$), PVs ($p = 0.015$) and PVa ($p < 0.001$) were significantly high in group H compared to group N.

However MV: E, MV: A, MV: E/A, MV: E', MV: A', DTI, PVd and PVs/PVd were found to be comparable but statistically insignificant ($p > 0.05$).

Hence these findings suggest that, pregnant women with hypertension are likely to have diastolic dysfunction in terms of raised MV: E/E', IVRT, DS, PVs and PVA.

These findings were comparable to other studies.

In a study by Solanki R. et al¹⁰ authors evaluated maternal cardiovascular function using echocardiography in normal and pre-eclamptic women in the third trimester of pregnancy on 20 subjects with pre-eclampsia and 20 normotensive controls with 34 weeks gestation and singleton pregnancy. Study revealed that increased afterload in pregnancy induced hypertension is linked with reduced emptying of left ventricle. Increased E wave velocity in hypertensive patients suggest elevated trans mitral gradient during early diastole reflecting reduced LV compliance. The higher peak A wave velocity suggests more important role of atrial systole in filling of the hypertrophied ventricle in these women. The prolonged IVRT in pre-eclamptic subjects suggests that longer time is required for left ventricular pressure to fall below atrial pressure. The prolongation of E wave deceleration time in pre-eclamptic subjects is consistent with increased passive filling of left ventricle during early diastole. Mean cardiac output (66.85 ± 4.56 ml/min vs. 56.1 ± 1.77 ml/min), mean LV diastolic mass (66.85 ± 4.56 ml/min vs. 56.1 ± 1.77 ml/min) and total vascular resistance (1396.85 ± 150.2 vs. 1204.5 ± 71.182 dyne, s cm⁵) was higher in the women with pre eclampsia indicating reduced LV compliance and diastolic dysfunction. This study hence concluded that there are significant structural and dynamic changes in women with pre-eclampsia particularly reduced LV compliance and diastolic dysfunction. Blood pressure monitoring alone is insufficient in identifying and managing the risk of cardiovascular complications in such patients. Including echocardiographic assessment in the management protocol could help identify women who are at higher risk of developing complications.

Melchiorre K, et al.⁸ conducted a prospective case control study comparing the diastolic function of 50 patients with pre-eclampsia and 50 normal pregnancies. It was undertaken to evaluate cardiac function and LV remodeling occurring at term in patients with pre eclampsia. Global diastolic dysfunction was seen in 40% of pre eclampsia cases as compared to 14% in normotensive pregnancies ($p=0.007$). Altered indices of LV mass indicate Left ventricular adaptation to maintain myocardial contractility against increased afterload in pre-eclampsia patients. As diastolic dysfunction precedes systolic dysfunction in other causes, its development is of prognostic value in patients with pre-eclampsia. Hence this reiterates the importance of early detection of diastolic dysfunction in women with pre- eclampsia.

Conclusion:-

Echocardiography can be used as a useful screening tool to identify diastolic dysfunction in patients with pregnancy induced hypertension. The echocardiographic characteristics including Mitral Valve: E/E', IVRT, DS, PVs and PVA were significantly higher in pregnancy induced hypertension when compared to normotensive pregnant females.

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RESEARCH ARTICLE

ASSESSMENT OF AIR POLLUTION TOLERANCE INDEX OF PLANT SPECIES GROWING NEAR METAL CASTING FOUNDRIES AROUNDPEELAMEDU, COIMBATORE.

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Air pollution tolerance index, biochemical parameters, relative water content, ascorbic acid and bio monitoring

Abstract

The study examined Air pollution tolerance index (APTI) of 35 plants growing near metal casting foundries located in and around Peelamedu, Coimbatore. Four physiological and biochemical parameters of plant leaves namely total chlorophyll content, relative water content, pH, and ascorbic acid content which can be used as predictor of air quality. It is observed that *Lantana indica*, *Nerium indicum*, *Duranta repens*, *Amaranthus viridis*, *Ricinus communis*, *Bauhinia racemosa* and *Samanea saman*, showed higher APTI values. The study indicates that plantation of tolerant species are useful for bio monitoring and to develop green belt in and around industrial areas.

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Introduction:-

Air pollution was earlier considered as a local problem around large point sources. But due to the use of tall stacks and long range transport of pollutants, it has become a regional problem. Many developing countries including India have experience a progressive degradation in air quality as a result of rapid pace of development over the last three decades. During this period newly industrialized countries underwent unparalleled economic growth, swelling urban populations and generated excessive emissions from automobiles, factories and refuse burning (Agbaire and Esiefarienrhe 2009). Much of the 20th century witnessed an increasing trend in urbanization in developing countries. While urbanization is a stimulus of development, in the process many cities in Asia, Africa, the Near East and Latin America are facing two challenges of pollution and congestion (Ashmore, 2005).

Air pollution is more complex than most other environmental challenges. No physical or chemical methods are known to ameliorate air pollution. A suitable alternative is to develop a biological by growing green plants in and around industrial and urban areas (Shannigrahi *et al.*, 2004, Ghose and Majee 2001 and Aarti *et al.*, 2012).

Plants plays a vital role in monitoring and maintaining the ecological balance by actively participating in the cycling of nutrient and gases like CO₂, O₂ and also provide enormous leaf area for impingement absorption and accumulation of air pollutants to reduce the pollution level in the air environment (Escobedo *et al.*, 2008). Several contributors agree that air pollution affects the plant growth adversely (Rao 2006, Chandawatet *et al.*, 2011 and Bhattacharya *et al.*, 2012).

Coimbatore, one of India's leading industrial centers with excellent potential for industrial growth is spread about 105.6 sq.km and has a population of 1.1 million people. The city is one of the important foundry cluster area in

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southern India. The majority of the foundry units fall under the category of small scale industry. There are about 250 small scale foundries located every 2 km in the industrial sector of Coimbatore. Air pollution due to these industries has become a serious environmental stress to plants. The emission of gases and dust particles from the industry causes serious setback to the overall physiology of plants. Among all the plant parts, leaf is the most sensitive part to air pollutants and several other such external factors. Response of plants toward air is being assessed by air pollution tolerance index. This index has been used to rank plant species in their order of tolerance to air pollution. Some plant species and varieties are so sensitive that they can be conveniently employed as biological indicator or monitors of specific pollutants. The present investigation was undertaken to study the air pollution tolerance index of plant growing near metal casting foundries located in and around Peelamedu, Coimbatore.

Materials and Methods:-

Study Area:-

Peelamedu is a residential and commercial neighborhood in the city of Coimbatore, Tamil Nadu, India, located at 11.031.N and 76.999 E. It is situated just 5 km away from the heart of the city Gandhipuram. There are many large, medium and small scale metal casting foundries are operating from here.

Sampling:-

For the present study, fresh leaves from each plant were collected from the experimental sites of urban and industrial areas of Peelamedu from July to March, 2012-2013. Three replicates of fully matured leaves of each species were taken in the morning (9:30 am to 11:30 am). The experiments were replicated three times for each biological factor. Leaves from same plant species (control) were collected from Indian Forest Genetics and Tree Breeding (IFGTB) Campus which is used as control area located far away from the industrial area which is a pollution free atmosphere. The weight of fresh leaves was taken immediately upon getting to the laboratory. Samples were preserved in refrigerator for further analysis.

Ascorbic acid content:-

Ascorbic acid content (expressed in mg/g) was measured using spectrophotometric method (Bajaj and Kaur, 1981). 1 gram of the sample was measured in to a test tube, 4 ml oxalic acid – EDTA extracting solution was added. Then 1 ml of orthophosphoric acid followed by 1 ml of 5% tetraxosulphate (vi) acid. To this 2ml ammonium molybdate was added and then 3ml of water. The solution was then allowed to stand for 15 minute, after which the absorbance at 760 nm was measured with a spectrophotometer. The concentrations of ascorbic acid in the sample were then extrapolated from a standard ascorbic acid curve.

Total chlorophyll content:-

Chlorophyll content was estimated using the method of Arnon (1949). 0.1 gram of fresh leaf material crushed with mortar and pestle. The crushed samples were collected and washed with 2 ml of 80 per cent acetone in three tubes. The contents are transformed into centrifuge tubes and the samples are centrifuged at 2,500 rpm for 10 min. The supernatant was collected in each test tube and the final volume of supernatant is made up to 10 ml by adding 80 per cent acetone using measuring cylinder and the absorbance is read at 645 nm and 663 nm for chlorophyll a and b using a spectrophotometer. The amount of total chlorophyll present in the extract (mg chlorophyll per gram tissue) was calculated by using the formula given below:

$$\text{Chlorophyll a / g tissue} = 12.7 (A_{663}) - 2.69 (A_{645}) \times \frac{V}{1000} \times W$$

$$\text{Chlorophyll b / g tissue} = 22.9 (A_{645}) - 4.68 (A_{663}) \times \frac{V}{1000} \times W$$

$$\text{Total chlorophyll / g tissue} = 20.2 (A_{645}) - 78.02 (A_{663}) \times \frac{V}{1000} \times W$$

where: A = Absorbance at specific wave length, V = Final volume of chlorophyll extract in 80 per cent acetone, W = Fresh weight of tissue extracted.

Leaf extract pH:-

For determining pH of leaf extract, 5 gram of fresh leaf material was homogenized with 50 ml of distilled water and samples of the homogenate were centrifuged at 3000 rpm for 10 min and pH value of leaf extract was determined using a calibrated digital pH meter with buffer solution of pH 4 and pH 7.

Relative water content:-

Leaf RWC is determined by using the method described by Singh (1997) and calculated with the formula $RWC (\%) = [(FW - DW) / (TW - DW)] \times 100$

where, **FW** – Fresh Weight,

DW – Dry Weight,

TW – Turgid Weight.

Fresh weight is recorded by weighting the fresh leaves. To get the dry weight, the leaves were dried in an oven at 70°C for overnight and then taken the dry weight. For getting the turgid weight, the leaves were immersed in water overnight, blotted dry and then weighed.

Air pollution tolerance index determination:-

APTI was calculated by the following formula proposed by Singh and Rao (1983). The formula of APTI is given as:

$$APTI = [A (T+P) + R] / 10$$

Where A= ascorbic acid content (mg/g)

T= total chlorophyll (mg/g)

P= pH of leaf extract

R= relative water content of leaf (%)

Results and Discussion:-

The plant being constantly exposed to the environment absorbs, accumulate and integrate pollutants impinging on their foliar surface. Consequently they show visible or subtle changes depending on their sensitivity level.

Table 1:- Ascorbic acid content, Total chlorophyll, pH, and Relative water content of the selected plant species

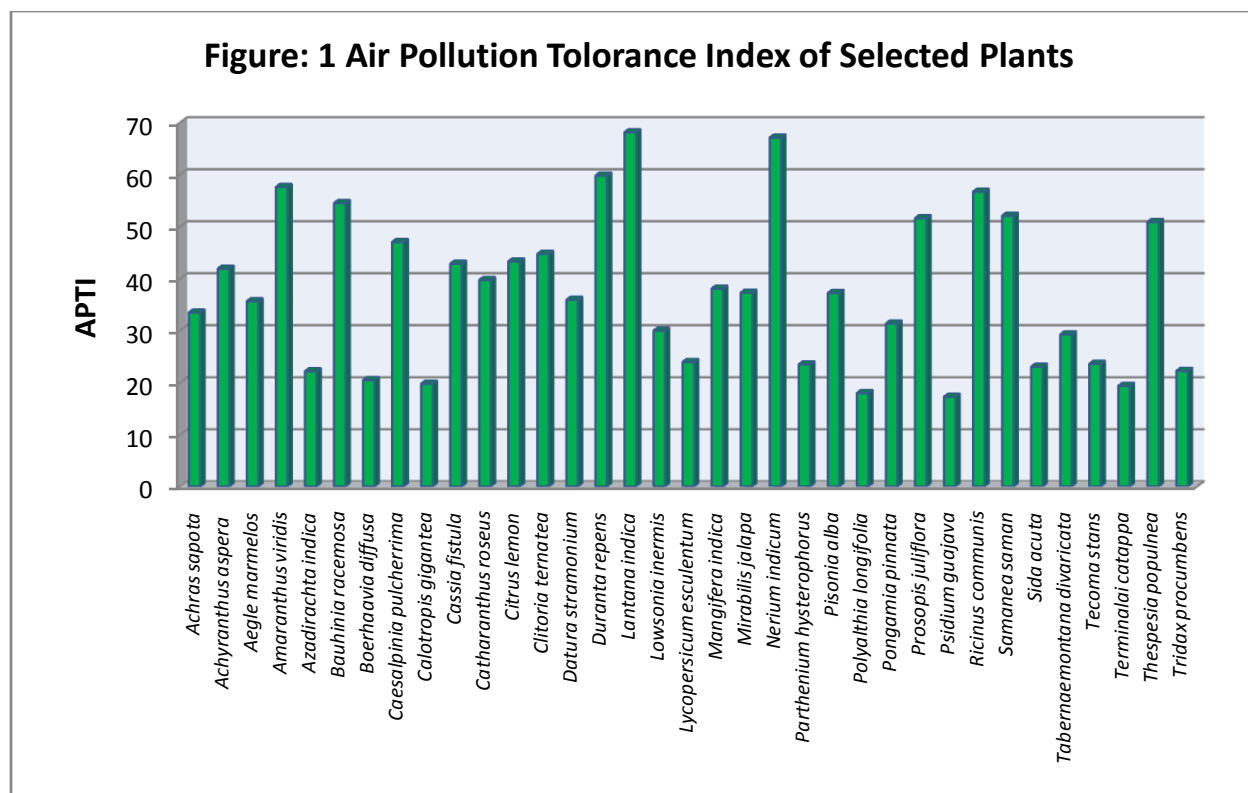
S.No	Name of the plant	Ascorbic acid content (mg/g)	Total chlorophyll content (mg/g)	pH value	Relative water content (%)
1.	<i>Achras sapota</i>	36.6± 1.22	0.49± 0.17	6.1± 0.03	73.4± 3.05
2.	<i>Achyranthus aspera</i>	37.9± 2.02	0.70± 0.07	7.2± 0.02	82.0± 2.22
3.	<i>Aegle marmelos</i>	45.7± 0.70	1.50± 0.02	6.3± 0.01	69.0± 2.01
4.	<i>Amaranthus viridis</i>	60.6± 1.44	1.85± 0.22	6.7± 0.02	62.5± 3.18
5.	<i>Azadirachta indica</i>	20.1± 1.00	1.06± 0.05	6.1± 0.02	77.9± 3.22
6.	<i>Bauhinia racemosa</i>	76.8± 1.41	0.32± 0.67	6.2± 0.02	44.8± 2.88
7.	<i>Boerhaavia diffusa</i>	21.9± 1.33	1.07± 0.34	6.3± 0.01	45.0± 2.04
8.	<i>Caesalpinia pulcherrima</i>	62.1± 1.01	0.60± 0.67	6.1± 0.02	53.8± 3.55
9.	<i>Calotropis gigantea</i>	17.8 ± 0.32	0.33± 0.04	6.4± 0.01	77.5± 1.88
10.	<i>Cassia fistula</i>	59.4± 0.06	1.46± 0.12	4.6± 0.02	78.0± 2.22
11.	<i>Catharanthus roseus</i>	47.0± 1.12	0.57± 0.09	6.1± 0.02	79.8± 3.05
12.	<i>Citrus lemon</i>	36.6± 2.23	1.06± 0.34	7.3± 0.03	66.6± 3.05
13.	<i>Clitoria ternatea</i>	56.0± 1.20	0.13± 0.07	6.8± 0.02	59.3± 1.42
14.	<i>Datura stramonium</i>	45.0± 1.22	0.83± 0.21	5.4± 0.01	78.7± 2.01
15.	<i>Duranta repens</i>	81.0± 2.45	0.29± 0.06	6.2± 0.01	72.2± 3.22
16.	<i>Lantana indica</i>	79.2± 1.22	0.48± 0.22	7.8± 0.01	50.8± 3.06
17.	<i>Lowsonia inermis</i>	32.1± 1.02	1.40± 0.01	5.4± 0.02	81.0± 2.22
18.	<i>Lycopersicum esculentum</i>	31.0± 0.82	0.17± 0.02	5.1± 0.01	74.02± 2.44
19.	<i>Mangifera indica</i>	46.1± 1.04	0.30± 0.02	5.9± 0.02	74.47± 3.05
20.	<i>Mirabilis jalapa</i>	51.5± 1.00	0.12± 0.04	5.3± 0.02	71.01± 2.22
21.	<i>Nerium indicum</i>	78.0± 1.22	1.40± 0.08	6.1± 0.01	86.03± 3.88
22.	<i>Parthenium hysterophorus</i>	27.6± 0.88	0.20± 0.04	6.1± 0.02	61.09± 4.01
23.	<i>Pisonia alba</i>	44.6± 1.05	0.32± 0.09	6.1± 0.02	83.7± 2.86
24.	<i>Polyalthia longifolia</i>	15.0± 1.22	0.17± 0.02	6.0± 0.01	84.23± 3.88
25.	<i>Pongamia pinnata</i>	37.6± 0.44	0.06± 0.12	6.3± 0.02	73.8± 2.84
26.	<i>Prosopis juliflora</i>	68.6± 1.02	0.25± 0.05	6.1± 0.02	80.0± 3.18
27.	<i>Psidium guajava</i>	17.0± 1.22	0.12± 0.34	6.1± 0.03	70.0± 2.09

28.	<i>Ricinus communis</i>	59.2± 1.33	1.60± 0.02	6.7± 0.02	75.6± 3.02
29.	<i>Samanea saman</i>	49.1± 0.88	0.34± 0.02	6.1± 0.01	46.6± 2.66
30.	<i>Sida acuta</i>	26.4± 1.08	0.46± 0.08	6.4± 0.02	49.86± 3.60
31.	<i>Tabernaemontana divaricata</i>	31.6± 1.20	0.43± 0.05	6.1± 0.02	83.07± 4.02
32.	<i>Tecoma stans</i>	25.3± 1.22	1.08± 0.08	5.6± 0.01	64.6± 3.18
33.	<i>Terminalia catappa</i>	75.4± 1.55	0.48± 0.02	4.5± 0.02	75.49± 2.02
34.	<i>Thespesia populnea</i>	63.0± 1.44	0.93± 0.02	6.0± 0.02	72.04± 1.88
35.	<i>Tridax procumbens</i>	23.6 ± 1.05	0.71± 0.02	5.9± 0.01	64.71± 3.33

The increased level of ascorbic acid in leaves will increase air pollution tolerance in plants (Chaudhury and Rao, 1977). In the present study, the plant species namely *Duranta repens*, *Lantana indica*, *Nerium indicum*, *Bauhinia racemosa*, *Terminalia catappa*, and *Prosopis juliflora* showed increased concentration of ascorbic acid (Table 1). The variation in chlorophyll content in plants is because of various environmental factors such as air, water and soil (Katiyar and Dubey 2000). Chlorophyll is the principal photoreceptor in photosynthesis. Hence its measurement is an important tool to evaluate the effects of air pollutants on plants. It plays an important role in plant metabolism and any reduction in chlorophyll content corresponds directly to plant growth (Joshi and Swami, 2009). A higher concentration of chlorophyll was recorded in *Amaranthus viridis* followed by *Ricinus communis*, *Aegle marmelos*, *Cassia fistula*, *Lowsonia inermis* and *Nerium indicum* (Table 1).

The leaf extract pH was found to be alkaline in species like *Lantana indica* (7.8), *Citrus lemon* (7.3) and *Achyranthus aspera* (7.2) whereas, it was acidic in other species. A significantly lowest pH value was shown in *Terminalia catappa* (4.5) (Table 1). Schlitz and Reck (1977) reported that in the presence of an acidic pollutant, the leaf pH is lowered and decline is greater in sensitive species. A shift in cell sap pH toward the acid side in the presence of an acidic pollutant might decrease the efficiency of conversion of hexose sugar to ascorbic acid. However, the reducing activity of ascorbic acid is pH controlled, being more at higher and less at lower pH. Hence, the leaf extract pH on the higher side gives tolerance to plants against pollution (Agarwal and Tiwari, 1997). Relative water content is associated with protoplasmic permeability in cells caused loss of water and dissolved nutrient resulting in early senescence of leaves (Masuch *et al.* 1988). It is likely that the plants species with high relative water content may be tolerant to pollutants. The relative water content was found to be higher in *Nerium indicum* (86.03), and *Polyalthia longifolia* (84.23), *Pisonia alba* (83.7), *Achyranthus aspera* (82.0), *Lowsonia inermis* (81.0) and *Prosopis juliflora* (80.0) (Table 1).

Air pollution tolerance index is an index denotes the capability of a plant to combat against air pollution. Plant which have higher index value are tolerant to air pollution and can be caused as sink to mitigate pollution, while plants with low index value show less tolerance and can be used to indicate levels of air pollution (Singh and Rao, 1983). Among the plant species studied the highest tolerance was shown *Lantana indica* (68.12), *Nerium indicum* (67.10), *Duranta repens* (59.79), *Ricinus communis* (56.70), *Amaranthus viridis* (57.60), *Bauhinia racemosa* (54.50), *Samanea saman* (52.06), *Prosopis juliflora* (51.60) and *Thespesia populnea* (50.86) (Figur.1). Plantation of these tolerant species is useful for bio monitoring and to develop the green belt among the nature and to reduce industrial air pollution. This is very essential for saving the environment for our future generation to protect the present generation.



Conclusion:-

Day by day with increasing urbanization and industrialization, the air quality is degrading. Plants play a significant role in mitigating air pollution and maintaining ecological balance. APTI determination is of utmost importance because with an increase in small scale industries the pollution load is on the rise. From the results of the present study, this tolerant plant species can be used as indicators of pollution thereby acting as a sink to all air pollutants.

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RESEARCH ARTICLE

EVALUATION OF GLOBAL LV FUNCTION AND MECHANICAL DYSSYNCHRONY IN PATIENTS WITH LEFT BUNDLE BRANCH BLOCK (LBBB) - 2D ECHOCARDIOGRAPHY, 2D SPECKLE TRACKING AND REAL TIME 3D ECHOCARDIOGRAPHY.

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Key words:-

Left bundle branch block(LBBB), Left ventricular dysfunction(LV dysfunction),mechanical dyssynchrony, Speckle tracking, Real time 3D Echocardiography(RT3DE).

Abstract

Background: LBBB affects LV function and causes LV mechanical dyssynchrony. Prevalence of LBBB increases with age about 0.05% to 5.7% between age of 30-80 years in otherwise healthy population. Aim of the study was to evaluate the LV function and mechanical dyssynchrony with 2D Echocardiography, 2D speckle tracking and real time 3D Echocardiography (RT3DE) in asymptomatic LBBB patients, non- LBBB volunteers, and with symptomatic LBBB patients.

Methods: 2D Echocardiography, 2D speckle tracking and real time 3D Echocardiography was performed in 191 patients, 65 were non- LBBB volunteers, 60 patients were asymptomatic LBBB and 66 patients were symptomatic LBBB patients. Global LV function and mechanical dyssynchrony were measured.

Results: Non LBBB volunteers mean LV ejection fraction by 2D Echo was $67.18 \pm \text{SD } 3.45$, in RT3DE mean LV ejection fraction was $61.88 \pm \text{SD } 3.12$. In asymptomatic LBBB patients LVEF by 2D was $58.43 \pm$ and RT3DE LVEF was 54.47 ± 4.08 . In heart failure (HF) with LBBB patients 2D LVEF was 43.53 ± 7.12 and 3D LVEF 39.21 ± 7.97 .

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LV dyssynchrony measurements by 2D speckle tracking analysis showed:

- A. Sdt of 6S segments by CS was 54.82 ± 11.2 , 96.43 ± 17.73 , 119.02 ± 23.17 for non-LBBB volunteers, asymptomatic LBBB patients and symptomatic LBBB patients respectively.
- B. Sdt of 12S by LS was 52.96 ± 5.1 , 90.03 ± 16.2 , 111.75 ± 34.22 for non-LBBB volunteers, asymptomatic LBBB patients and symptomatic LBBB patients respectively.

Systolic dyssynchrony (SDI) index by RT3DE was 5.07 ± 2.22 , 7.44 ± 1.45 , 15.08 ± 3.85 for non-LBBB volunteers, asymptomatic LBBB patients and symptomatic LBBB patients respectively.

Conclusion: Asymptomatic LBBB patients have more depressed LV function than non LBBB volunteers with intermediate mechanical dyssynchrony, symptomatic LBBB patients have moderate to severe LV dysfunction and most severe mechanical dyssynchrony among the 3 groups.

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Introduction:-

Left ventricular (LV) mechanical dyssynchrony is observed in left bundle branch block (LBBB). Persistent mechanical dyssynchrony contributes to progressive ventricular remodelling and impaired systolic LV function.¹ Mechanical dyssynchrony may partially be responsible for the increased morbidity and mortality in heart failure (HF) patients with conduction abnormalities as a LBBB, compared to patients without a LBBB.²⁻⁵

In patients without underlying structural cardiac disease, except the presence of a LBBB, subtle impaired systolic and diastolic LV functions are frequently found.⁶ However, impaired function in these patients is not always recognized because of lack of clinical symptoms.⁶⁻⁸

Prevalence of LBBB increases with age about 0.05% to 5.7% between age of 30-80 years in otherwise healthy population.²⁴⁻²⁶ Currently, the most widely used methods for the clinical assessment of LV function and mechanical dyssynchrony are two-dimensional (2D) echocardiography and tissue Doppler imaging.⁹⁻¹¹

Speckle tracking strain analysis method based on grayscale 2-dimensional (2D) images, and transthoracic real-time three-dimensional echocardiography (RT3DE) are relatively novel imaging technique which offers the unique opportunity to evaluate global LV function fast and accurate, without presumptions and with comparable results as 2D Echocardiography and magnetic resonance imaging.¹²⁻¹⁵ In addition, combined with specially designed software, provides detailed quantitative information of mechanical dyssynchrony.¹⁶⁻¹⁸

Therefore, we designed a prospective cross-sectional study to assess global LV function and mechanical dyssynchrony in consecutive LBBB patients. The first purpose of this study was to evaluate global LV function and the amount of mechanical dyssynchrony by Speckle tracking strain analysis and RT3DE in asymptomatic LBBB patients compared to non- LBBB volunteers and patients with symptomatic LBBB. The second purpose was to evaluate the association between LV mechanical dyssynchrony and the presence or absence of symptoms of HF.

Methods:-

LBBB was diagnosed according to standard electrocardiography criteria: A QRS duration greater than 120 ms, absence of initial septal Q waves, a broadened R wave in the left precordial leads, and an initial R wave followed by a wide, deep S wave in the right precordial leads on a 12-lead electrocardiogram. Symptoms of HF included dyspnoea, ankle oedema, fatigue and reduced exercise tolerance.

Consecutive OPD and IPD patients of LBBB attending to our department of Cardiology were included as per inclusion criteria. Ethical committee clearance was taken.

There are very few studies quantifying LV volume, ejection fraction and mechanical dyssynchrony in patients with LBBB by 2D Echocardiography, 2D speckle tracking, real time 3D echo, hence we have taken up this study. One hundred and ninety one subjects were studied, consisting of sixty five volunteers with no abnormal findings during physical examination, no conduction abnormalities on electrocardiogram; sixty consecutive asymptomatic LBBB patients, with no history of structural cardiac disease and no abnormal findings during physical examination, New York Heart Association function class (NYHA) I and sixty-six consecutive symptomatic LBBB patients, NYHA II–III, without pulmonary hypertension, were screened.

ECG gated echocardiography was performed using EPI Q 7C (Philips Medical Systems, Andover, Mass), installed with highly advanced software techniques are used for 2D echocardiography, 2D speckle tracking with strain imaging, real time 3D Echocardiography image acquisitions. A matrix array transducer with a frequency range from 2 to 5 mhz, and the transducer was maintained in a fixed orientation. A complete examination of patients with normal conduction and those with LBBB was performed

Quantification of LV function:-

Dimensional echocardiographic LV volume and LVEF by modified Simpson's method:-

Transthoracic apical 4-chamber view was performed, later Left ventricular end diastolic image and Left ventricular end systolic images were acquired, and in the same way apical 2-Chamber Left ventricular end diastolic image and Left ventricular end systolic ECG gated images were acquired with patient in left lateral decubitus position. Then LV end diastolic volume, LV end systolic volumes, LVEF was estimated in apical 4- and apical 2-chamber windows by the modified Simpson's method.

The volume of the LV is calculated from the dimension and area obtained from orthogonal apical views (four-chamber and two-chamber views). The LV volume was then calculated with the modified Simpson method or disk summation method

Three -Dimensional data acquisition and Analysis:-

Three-dimensional echocardiography, first as reconstructed images and more recently as real-time acquisition, provides reproducible assessment of LV volumes, dimensions and systolic function. Using dedicated reconstruction software, semi-automated border detection allows continuous volume calculation of all 16 myocardial segments simultaneously. Represented as regional volumetric curves during the cardiac cycle, the timing of minimal volume for each segment can be analyzed. With normal homogeneous activation, the different LV segments reach their minimal volume at the same time.

Transthoracic apical 4-chamber full volume acquisitions were made during 4 –6 s breath-hold, with the patient in left lateral position. Care was taken to include the entire LV volume within the pyramid shaped 3D scan-volume. Due to the sector width of the acquired volume, three alternate heart cycles and a constant R-R interval were obligatory or compulsory to create a full-volume acquisition of the LV.

Then apically acquired dataset was imported into an offline software package for analysis.

- Identification of three points or land marks i, e mitral valve, aortic valve and apex was done, then the volume data set was automatically sliced into eight equally rotated imaging planes, in this end systolic frame and end diastolic frames were identified.
- After identification of end systolic frame and end diastolic frames, the apex and two annulus points were determined in both end diastolic and end systolic state.
- Later endocardial borders were traced in end-diastole using an automated counter detection software system, in case the borders were not accurate, manually these endocardial borders were manipulated and adjusted, then a full volume 3-D model of the left ventricle was reconstructed.i.e a LV cast is formed.
- After a full volume 3D model of the left ventricle was reconstructed, the left ventricle was divided into 16-segments, then segmental analysis was performed, later end diastolic end systolic volumes and ejection fraction were displayed on screen along with this global and segmental volume and EF were presented.

Quantification of left ventricular mechanical dyssynchrony:-

Real time 3D Echo:-

LV cast was divided into 16 pyramid-shaped segments. For each segment, a regional time-volume curve was obtained and the systolic dyssynchrony index (SDI) was derived. From the regional time-volume curves, the systolic dyssynchrony index (SDI) was derived.²⁰ SDI is based on the standard deviation of mean time-to-minimal regional volume of 16 LV segments (six basal, six mid-segments and four apical segments) during a cardiac cycle. In other words, SDI is the dispersion of time-to-minimal regional volume for all 16 LV segments. Time is represented as percentage of the RR interval to allow comparisons of different patients which otherwise could be influenced by RR interval duration.

Speckle tracking strain Analysis:-

Standard grayscale 2D images were acquired in the 2- and 4-chamber apical views as well as the parasternal and short-axis views. From an end-systolic single frame, a region of interest was traced on the endocardial cavity interface by a point-and-click approach. Then an automated tracking algorithm followed the endocardium from this single frame throughout the cardiac cycle. Further adjustment of the region of interest was performed to ensure that all of the myocardial regions were included. Speckles, equally distributed in the region of interest, could be followed throughout the entire cardiac cycle. The distance between the speckles was measured as a function of time, and parameters of myocardial deformation could be calculated.

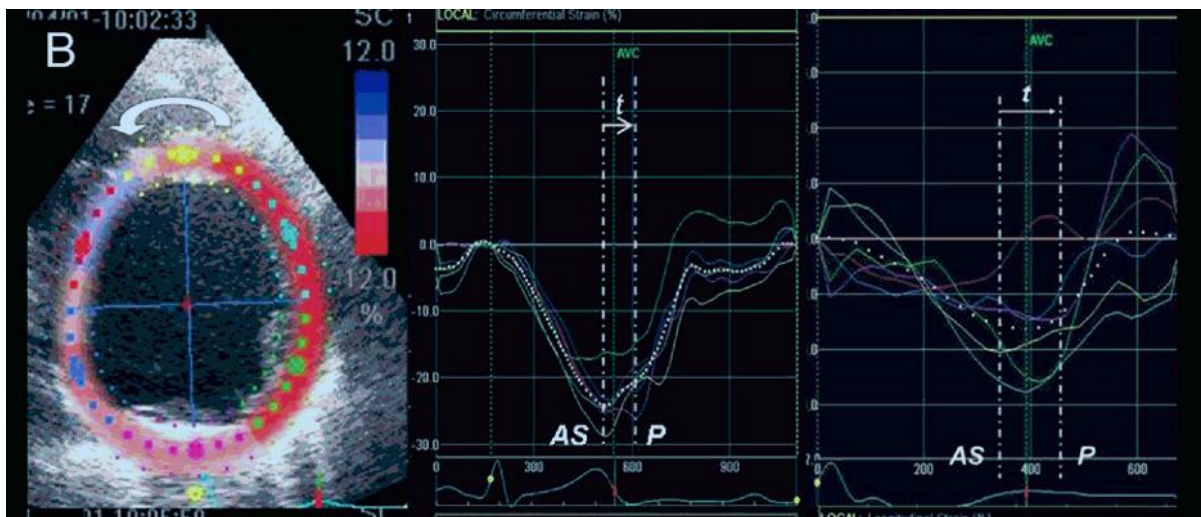
Myocardium was divided into 6 segments that were color coded and displayed into 6 segmental time-strain curves for CS, and LS.²⁷

Two different parameters for dyssynchrony were obtained:-

Circumferential Strain (CS)

- Difference between time to peak-systolic strain of the (antero) septal and posterior segments (AS-P delay).
Figure B
- The standard deviation of time to peak-systolic strain for all 6 segments (sdt6s) was measured.

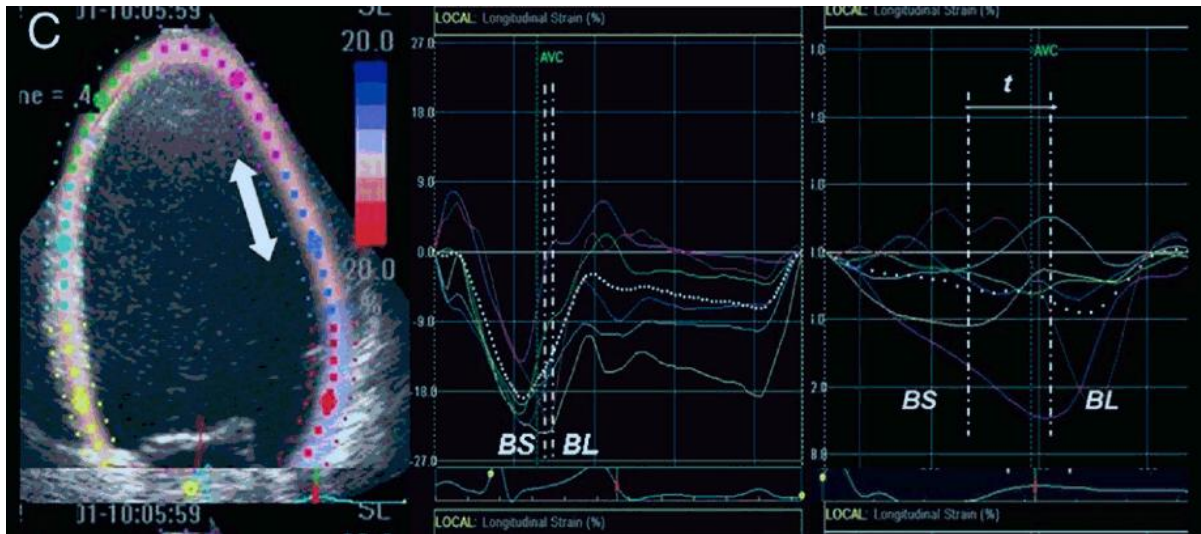
Figure B:- Difference between time to peak-systolic strain of the (antero) septal and posterior segments (AS-P delay) by circumferential strain.



Longitudinal Strain (Ls):-

- Difference between time to peak-systolic strain of the basal-septal and basal-lateral LV segment (BS-BL delay).
Figure C
- The standard deviation of time to peak-systolic strain for 12 LV segments (sdt12s)

Figure C:- Difference between time to peak-systolic strain of the basal-septal and basal-lateral LV segment (BS-BL delay) by longitudinal strain.



Statistical Analysis:-

Data are expressed as mean value \pm standard deviation(SD). For comparison of means between the three groups, a one way ANOVA test with Tukeys multiple posthoc analysis was used. A probability value of $p < 0.05$ was considered as statistically significant.

Results:-

Subjects:-

A total of 191 consecutive LBBB patients and non- LBBB volunteers were enrolled. The final study population consisted of Sixty five non LBBB volunteers (mean age 51 ± 7 years, QRS duration 96 ± 4 ms), sixty patients with an asymptomatic LBBB (mean age 60 ± 7 years, QRS duration 139 ± 2 ms) and sixty-six patients with symptomatic LBBB (mean age 61 ± 9 years, QRS duration 151 ± 10 ms).

QRS duration of both asymptomatic LBBB and symptomatic LBBB groups was significantly more compared to non- LBBB volunteers ($p = 0.0001$)

Then further comparing QRS duration of symptomatic LBBB patients and asymptomatic LBBB it was observed that symptomatic LBBB patients had more QRS duration. ($p = 0.0001$). *Table 1*

Table 1:- Patient characteristics.

	Non- LBBB volunteers	Asymptomatic LBBB	Symptomatic LBBB	P value
Number (n)	65	60	66	-
Male , n(%)	34(52.35)	17 (28.33)	48(65.15)	-
Age	51 ± 7	60 ± 7	61 ± 9	-
NYHA class		I	II-III	-
QRS duration	96.14 ± 4.22	139.67 ± 1.81 *	151.75 ± 10.09 †, #	< 0.0001
Hypertension,n	13	18	22	-
Diabetics,n	10	21	24	-
Diuretics ,n	-	6	66	-
Beta blockers,n	-	4	62	-
ACE/ ARB,n	10	15	46	-
Statin ,n	10	14	56	-
Antiplatelet agents,n	10	20	64	-

LBBB, left bundle branch block; NYHA, New York Heart Association function class; ACEI, ACE inhibitor; ARB, angiotensin receptor blockade;

* Significant difference non-LBBB volunteers versus Asymptomatic LBBB.

†Significant difference non- LBBB volunteers versus symptomatic LBBB.

Significant difference asymptomatic LBBB versus symptomatic LBBB.

Global LV function:-**2D Echocardiography by Simpsons method**

EF, ESV and EDV in both asymptomatic LBBB and symptomatic LBBB patients when compared to non- LBBB volunteers was significantly low ($p=0.0001$)

Then when EF, ESV and EDV in symptomatic LBBB compared to asymptomatic LBBB was significantly lower ($p=0.001$) *Table 2*

Real time 3D Echocardiography parameters:-

EF, ESV and EDV values showed same results of significant difference when compared among the 3 groups as in 2D Echocardiography. *Table 2*

Comparing ESV, EDV, EF values of 2D and 3D Echocardiography:-

Comparison of EDV, EF values on 2D and 3D Echocardiography of all the 3 groups showed that real time 3D echocardiographic values were lesser, which was statistically significant. ($p=0.0001$)

Comparison of ESV values on 2D and 3D Echo was also statistically significant and lesser by real time 3D echocardiography in non-LBBB volunteers and symptomatic LBBB groups ($p=0.0001$)

But comparison of ESV values on 2D and 3D Echo in asymptomatic LBBB was not significant (0.254). *Table 2*.

Table 2:- Global LV function.

Groups		2D Echo	RT3D E	P value
Non-LBBB	LVEF	67.18±11.05	61.88±3.13*†‡#	<0.0001
	LVEDV	58.75±7.79	47.77±7.92*†‡#	<0.0001
	LVESV	25.25±4.47	21.69±3.84*†‡#	<0.0001
Asymomatic LBBB	LVEF	58.43±2.63	54.47±4.08*†‡#	<0.0001
	LVEDV	76.02±11.58	70.52±7.71*†‡#	<0.0001
	LVESV	30.68±5.33	31.52±3.50 ‡‡#	0.254, 0.0001†‡#
Symptomatic LBBB	LVEF	43.53±7.12	39.21±7.97*†‡#	<0.0001
	LVEDV	97.44±37.84	84.53±30.74*†‡#	<0.0001
	LVESV	58.42±29.49	49.17±25.39*†‡#	<0.0001

LBBB- Left bundle branch block; Lv- Left ventricle; EF- Ejection fraction; EDV- end-diastolic volume; ESV- end-systolic volume; 2D- 2-dimensional Echocardiography; RT3DE- Real time 3-dimensional Echocardiography.

‡Significant difference non-LBBB volunteers versus Asymptomatic LBBB.

†Significant difference non- LBBB volunteers versus symptomatic LBBB.

Significant difference asymptomatic LBBB versus symptomatic LBBB

* Significant difference 2D Echo versus RT3D Echo.

Quantification of LV mechanical dyssynchrony:-**Real time 3D Echo:-**

Mean SDI of both groups asymptomatic LBBB ($7.44±1.45$) and symptomatic LBBB ($15.08±3.85$) was statistically significant and more when compared to non- LBBB volunteers ($5.07±2.22$) ($p=0.0001$) and mean SDI of symptomatic LBBB was more and statistically significant than asymptomatic LBBB ($p=0.0001$). *Table 3, Figure A, Figure 1.*

Figure A:- Regional time-volume curve and systolic dyssynchrony index by RT3DE

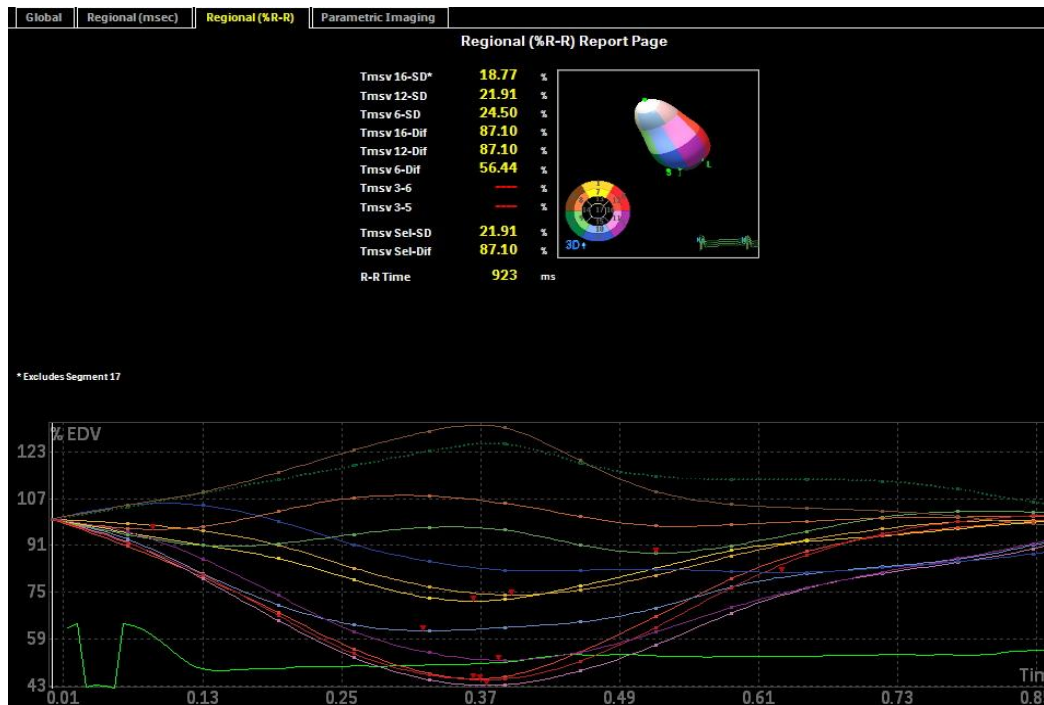
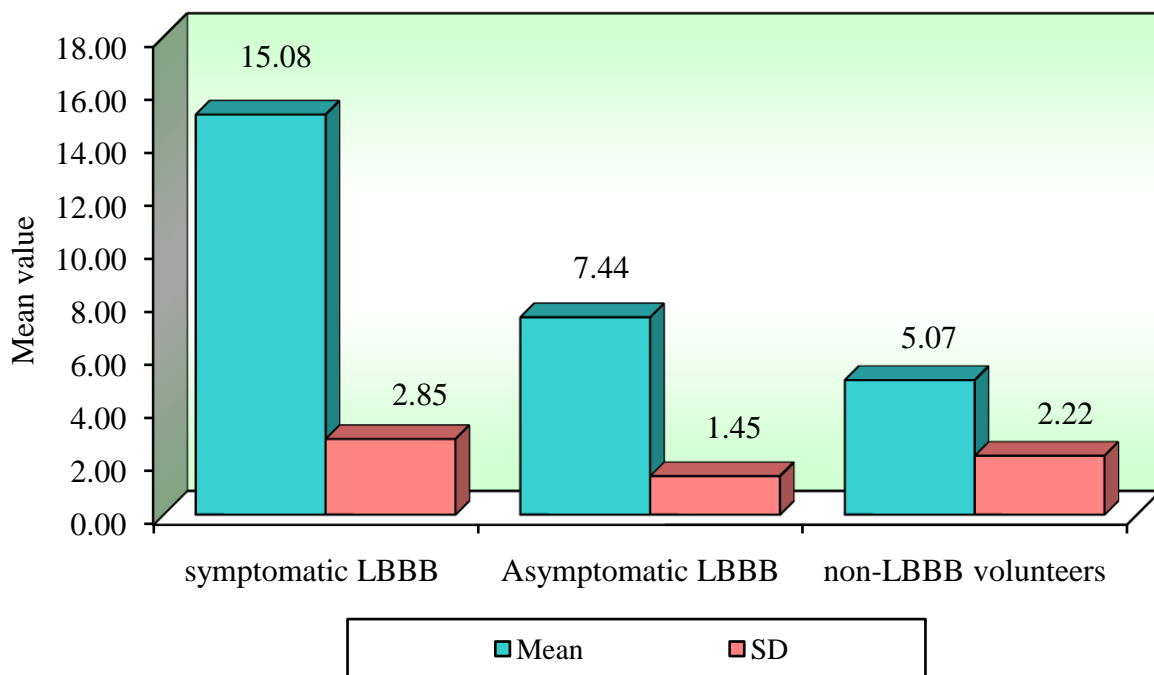


Figure 1: Comparison of three groups (symptomatic LBBB, asymptomatic LBBB and non-LBBB volunteers) with mean SDI scores



Speckle tracking strain Analysis:-

AS- P delay and sdt6s as assessed by CS and BS- BL delay and sdt12s assessed by LS were significantly more in asymptomatic LBBB and symptomatic LBBB patients when compared to non- LBBB volunteers ($p=0.0001$).

Further when asymptomatic LBBB patients and symptomatic LBBB patients were compared for these parameters, symptomatic LBBB patients had significantly more LV dyssynchrony ($p=0.0001$). Table 3, Figure D, Figure 2,3.

Table 3:- Mechanical dyssynchrony.

	Non- LBBB volunteers	Asymptomatic LBBB	Symptomatic LBBB	P value
SDI(%)	5.07±2.22	7.44±1.45 *	15.08±3.85†,#	<0.0001
AS-PS delay by CS	46.2±11.30	73.65±23.01*	133.80±40.95†,#	<0.0001
Sdt 6s by CS	54.88±11.29	96.43±17.73*	119.02±23.17†,#	<0.0001
BS -BL delay by LS	47.03±9.31	102.67±21.62*	113.88±40.69†,#	<0.0001
Sdt 12s by LS	52.96±5.15	90.03±16.29*	111.75±34.22†,#	<0.0001

LBBB, left bundle branch block; SDI, systolic dyssynchrony index; AS-P delay - difference between time to peak systolic strain of the anteroseptal and posterior segments; BS-BL delay - difference between time to peak systolic strain of the basal-septal and basal lateral segments; CS - circumferential strain; LS - longitudinal strain; sdt6s - standard deviation of the time to peak systolic strain of 6 segments; sdt12s - standard deviation of the time to peak systolic strain of 12 segments;

* Significant difference non-LBBB volunteers versus Asymptomatic LBBB.

†Significant difference non- LBBB volunteers versus symptomatic LBBB.

Significant difference asymptomatic LBBB versus symptomatic LBBB.

Figure D:- Standard deviation of time to peak-systolic strain for all 6 segments (sdt6s) by speckle tracking.

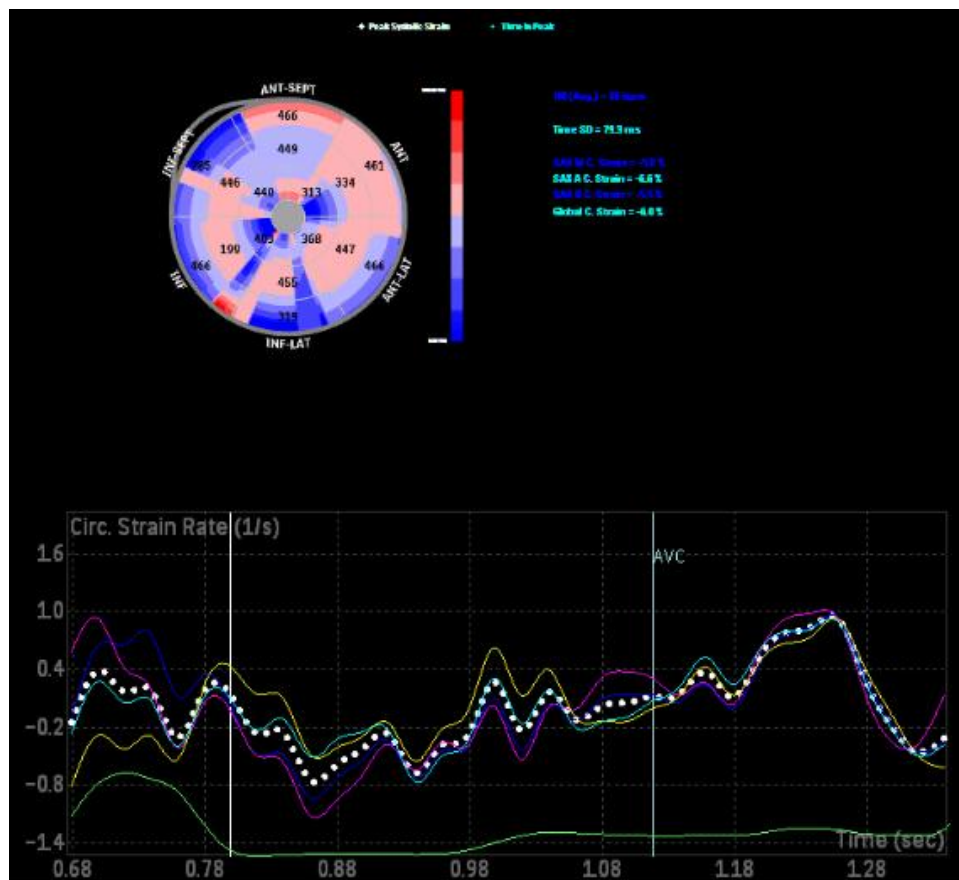


Figure 2: Comparison of three groups (Symptomatic LBBB, Asymptomatic LBBB, Non- LBBB volunteers) with mean SD+6S by CS scores

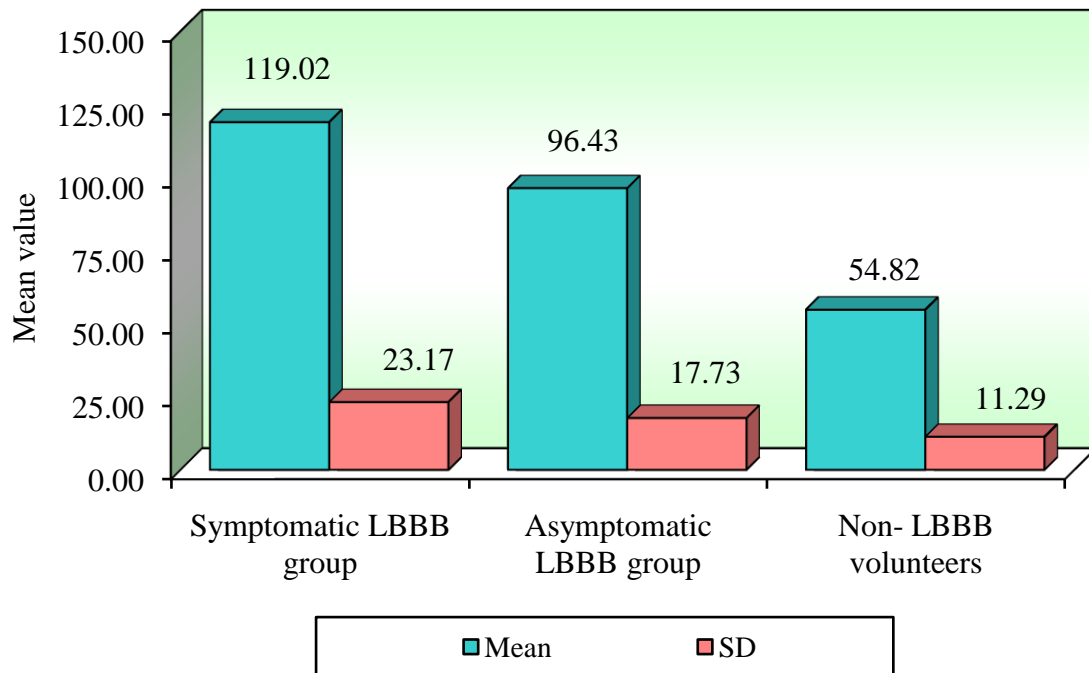
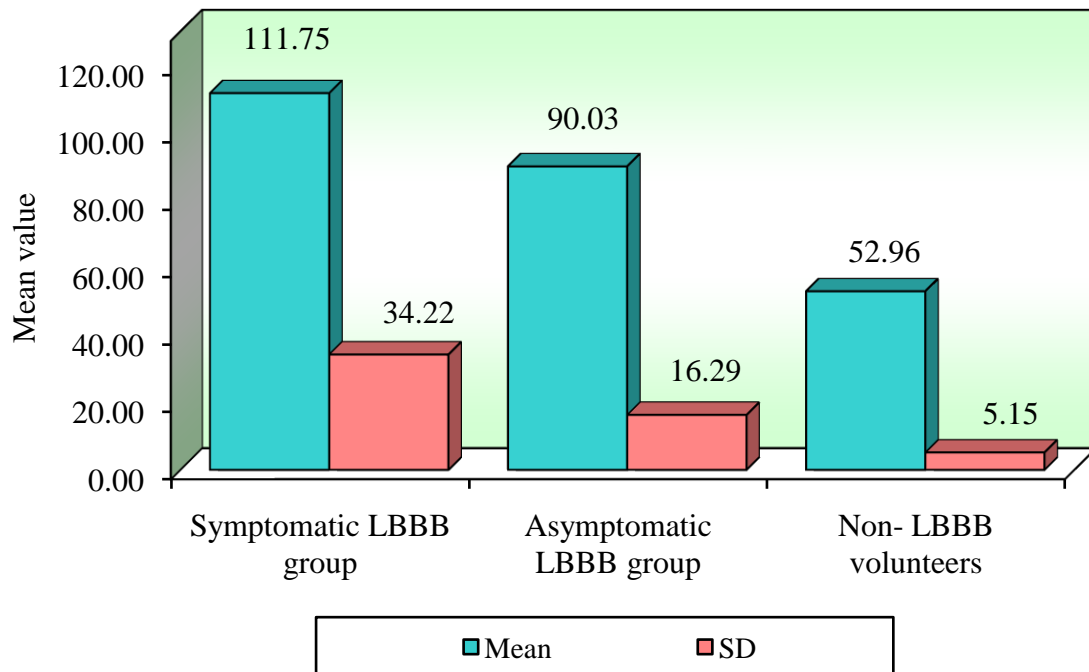


Figure 3: Comparison of three groups (symptomatic LBBB, asymptomatic LBBB, Non- LBBB volunteers) with mean SD= 12s by LS scores



Discussion:-

Important finding of this study is that, 2D Echocardiography, 2D speckle tracking, RT 3D Echo are useful for detecting differences in global LV function and mechanical LV dyssynchrony in LBBB patients with or without symptoms compared to non- LBBB volunteers.

Assessment of global LV function demonstrated that patients with asymptomatic LBBB and patients with symptomatic LBBB had a mild and moderate to severe reduction in EF respectively.

The EF, EDV, ESV measured with RT 3D Echo was lesser than one would expect when conventional 2D Echocardiography is used.

Mechanical dyssynchrony assessed by RT 3D Echo and *speckle* tracking strain analysis showed that non-LBBB volunteers had mild mechanical dyssynchrony. In asymptomatic LBBB patients intermediate mechanical dyssynchrony was observed and in symptomatic LBBB patients severe mechanical dyssynchrony was noted, which differed significantly among these 3 groups.

As observed previously in other studies, the EF and the amount of mechanical dyssynchrony showed an inverse correlation.²⁰ In accordance with results of a study by Vernooij et al.,¹ the trend to increased mechanical dyssynchrony in asymptomatic LBBB patients and the significant increased mechanical dyssynchrony in symptomatic LBBB patients in the present study, might be held responsible for the observed mild global LV dysfunction in asymptomatic LBBB patients and moderate to severe global LV dysfunction in symptomatic LBBB patients. Thus, demonstrating that mechanical dyssynchrony might negatively affect LV function and the resulting symptomatic status.

A study conducted by Kapetanakis et al.²⁰ showed HF with symptomatic LBBB patients with severe LV dysfunction had SDI of 15.7 ± 6.7 which was in accordance with SDI of 15.08 ± 3.85 in our symptomatic LBBB patients.

Another study conducted by Jeroen van dijk et al.²⁸ found SDI of 12.8 ± 4.8 in HF patients with LBBB which was in accordance with SDI of 15.08 ± 3.85 in our study.

In our study, number of LBBB patients enrolled is larger when compared to other older studies. It is also one of the first of its kind where LBBB patients are evaluated to assess global LV function and mechanical dyssynchrony in comparison to non- LBBB volunteers by means of 2D Echocardiography, 2D Speckle tracking strain analysis and 3D Echocardiography.

Clinical implications:-

With evaluation of LBBB patients with and without symptoms, differences in global LV function and mechanical dyssynchrony was observed which suggested that patients with a LBBB, who achieve a certain amount of mechanical dyssynchrony, may develop symptomatic HF. Therapeutic interventions that will decrease mechanical dyssynchrony may lead to a reduction of symptoms and improvement in LV function.

Study limitations:-

In patients with a LBBB, it is often unknown for how long a LBBB has been present. The process of remodelling of the LV that might occur after the formation of a LBBB, may be more distinct in a patient with a long lasting LBBB, than in a patient with a new onset LBBB.

In RT3DE, frame rates are relatively low (20 –25 Hz) compared to tissue Doppler imaging (100 –150 Hz). For evaluation of mechanical dyssynchrony, low frame rates can be a limitation in detecting very subtle dyssynchrony differences between the segments. However, RT3DE acquisitions have high spatial resolution and post-processing analysis can be done fast with good reproducibility, compared to other imaging techniques.²¹⁻²³

Conclusion:-

2D Echocardiography, 2D Speckle tracking strain analysis and 3D Echocardiographic evaluation of LBBB patients with or without symptoms and non-LBBB volunteers showed that global LV function of asymptomatic LBBB patients already slightly impaired compared to non-LBBB volunteers. However it was observed that patients with symptomatic LBBB have most severely impaired LV function.

Based on global LV function analysis in all 3 groups by 2D Echocardiography and real time 3D Echocardiography found that the values observed by real time 3D Echocardiography were significantly lesser when compared to 2D Echocardiography as 2D Echo considers LV in the form of a cylinder. Whereas, real time 3D Echocardiography during measurements take endocardial contours which are semi-automatically detected during the complete cardiac cycle creating a LV cast which is used to measure the LV volumes and EF. Hence real time 3D Echocardiography is more accurate and standardised method when compared to 2D Echocardiography.

Mechanical LV dyssynchrony in asymptomatic LBBB patients is already increased and patients with symptomatic LBBB have the more severe mechanical dyssynchrony.

Quantification of mechanical dyssynchrony with 2D speckle tracking strain imaging and RT3DE is useful, as increased mechanical dyssynchrony might have an effect on global LV dysfunction and consequential symptoms. EF and the amount of mechanical dyssynchrony showed an inverse correlation in our study indicating that when a substantial level of LV mechanical dyssynchrony is reached, it might be suggestive for the presence of symptoms.

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RESEARCH ARTICLE

EFFECT OF DIVALENT DOPING ON YOUNG'S MODULUS OF ZNO NANOWIRES.

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Nanowires, Semiconductor, Doping, Stress, Young's Modulus.

Abstract

The present work brings to light the effect of divalent (viz. Mg) doping on the Young's Modulus (YM) of ZnO nanowires (NWs), as a function of diameters considering surface stress effect. It is seen that the YM exhibits contrasting behavior for different doping concentrations of Mg. The variation in the YM is seen to be of semiconductive type, which then is seen to change to depict conductive type behavior on the immersion of Mg as a dopant. The doping along with surface stress play as deciding factors in determining the mechanical properties; especially the YM of the ZnO nanowires.

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Introduction:-

Thin Films and Nanowires are emerging as one of the most enticing fragments offered by the recent developments in the field of Nanotechnology in current times. Also, along with the Thin Films and NWs developed from pure undoped materials, a new horizon has been put forth by the concept of doping and the variation that this doping can bring in the material. More recent studies show that this idea of doping has proved to be an extremely valuable aspect as it can be used to unlock the hidden potentials and frontiers that the material has to offer. So, decent understanding of the outcomes of doping on semiconductor materials is essential. ZnO is one of the most commonly used semiconductor materials with a direct bandgap of ~3.37 eV at room temperature which leads to its abundant applications in the field of Photonics and Optoelectronics [1-4]. Previous Studies [5-7] of size dependence of YM of ZnO NWs has further lead us to explore out their mechanical properties for the practical applications. Along with diameter, the concentration of doping usually exerts a great influence on its overall mechanical properties; especially the YM which is believed to be determined by the contribution of both bulk elastic modulus and surface elastic modulus. So, it is of great interest to explore how the YM of the ZnO NWs varies as a function of their diameters and concentrations of doping.

Theoretical Approach:-

In order to estimate the YM of the ZnO nanowires the effects due to surface stress as discussed in the ref. [5,6] and can be written as,

$$E_{\text{nanowire}} = \frac{8}{5} g(1 - \nu) \frac{L^2}{D^3} (1)$$

Where D is the diameter of the nanowire, L is the length of the nanowire, ν is the Poisson's ratio and g is the surface stress [5,6,10]. As, the diameter of the nanowires increases, the equilibrium strain approaches to zero [5,7]. Further,

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it is the surface stress that plays as an important parameter and changes with the change in the concentration of doping [5,6,9].

Results and Discussion:-

In this work, we bring forth our results on the investigation of the YM of Mg doped ZnO nanowires. Here, the length of the nanowire is assumed to be 1000 nm [5,6], the typical length suspended when the nanowire is suspended using three point's clamped bending condition. The concentrations of Mg doping in ZnO is taken to be 0%, 2%, 4%, 6%, 8% and 10%. We have seen that the ZnO shows different behavior for undoped (i.e. 0%) and the doped case (i.e. 0%, 2%, 4%, 6%, 8% and 10%. respectively) [9]. We have taken into consideration (100) and (101) directions of growth of the NWs at growth temperature and annealed at 500°C and 700°C with the data acquired from other studies regarding the same [9]. From Fig 1, we can see that the YM increases for the as grown state of the Mg doped ZnO NWs but we see that when we anneal the same at temperatures above 450°C, the YM decreases with decreasing diameters showing semiconductive behavior [5-7]. Previous studies [10] suggest that the crystallization in ZnO occurs after crossing the 300°C temperature which is believed to be one of the reasons that the following trends and behavior are seen in the undoped ZnO case. Also, it is seen that the increase in temperature of annealing leads to further decrease in the YM of undoped ZnO NWs.

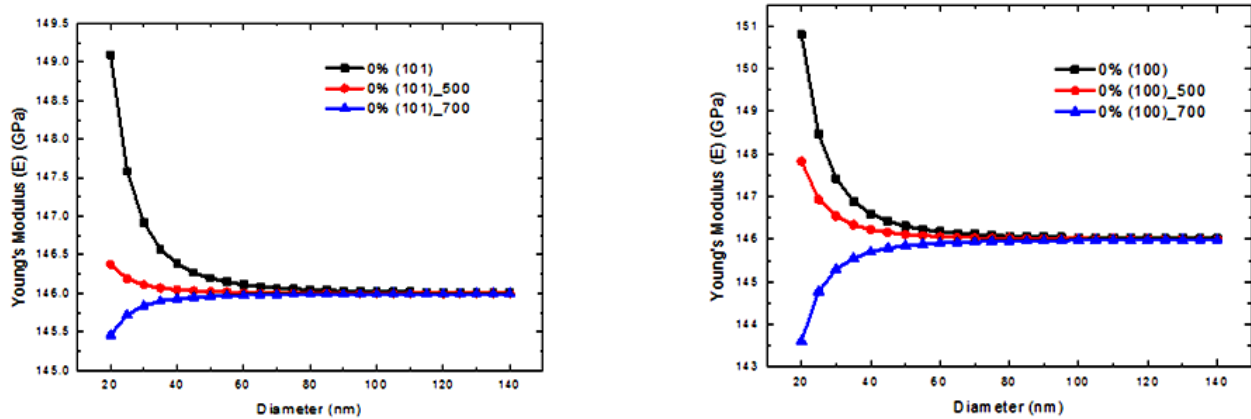


Figure 1:- YM vs. diameter for undoped ZnO NWs for (100) and (101) orientations at different temperatures.

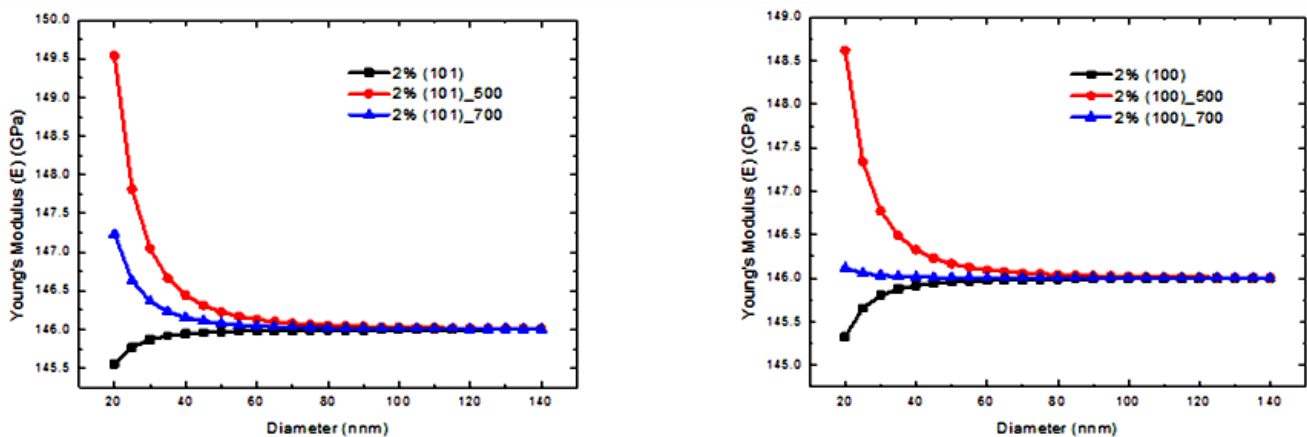


Figure 2:- YM vs. diameter for 2% Mg doped ZnO NWs for (100) and (101) orientations at different temperatures.

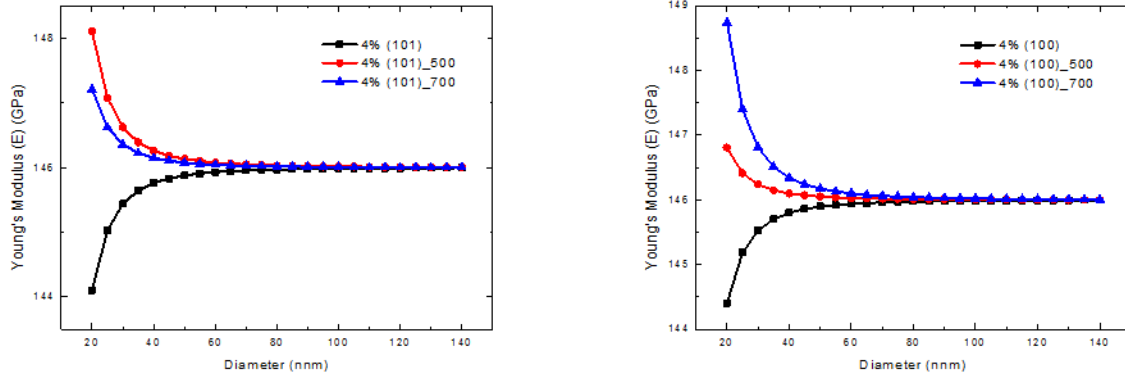


Figure 3:- YM vs. diameter for 4% Mg doped ZnO NWs for (100) and (101) orientations at different temperatures.

A similar approach was used to demonstrate the effect of Mg doping on the ZnO nanowires and it was observed that the results are quite exciting. It was seen that unlike the undoped case, the doping of Mg in various concentrations lead to an increase in the YM of the Mg doped ZnO NWs. The YM is found to increase with the decreasing diameters and hence show the conducting behavior [5] which is supported by the fact that the doping of Mg in ZnO leads to a change in bandgap of the material. Fig 2-6, show the effect of various doping concentrations on the YM of the ZnO NWs. An evident point is seen that as the doping concentrations increase the maximum value of YM of the NWs decreases. It may be because the effect of Mg doping on ZnO is substitutional. The peak value of YM in both orientations of growth is seen at 500°C for 2% doping and at 700°C for 4% doping of Mg showing the effect of doping on the crystallization temperature of the resultant material.

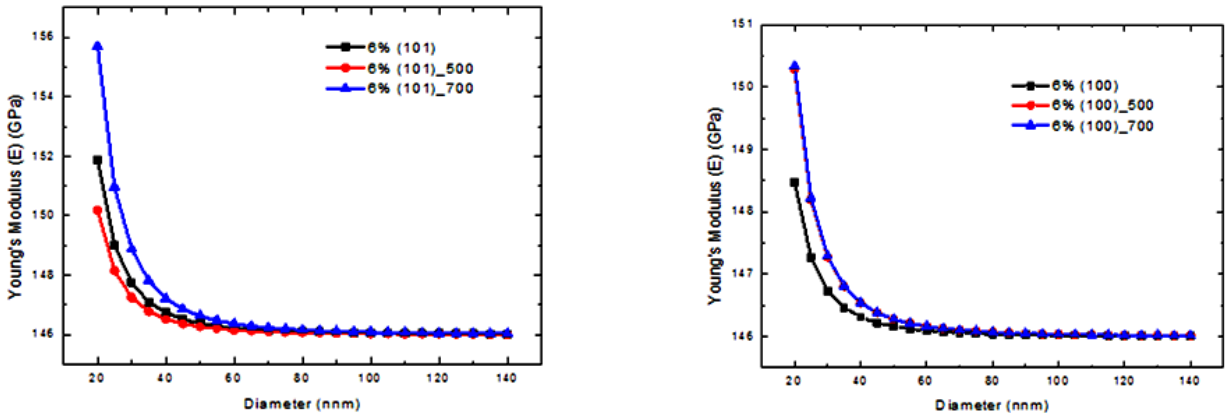


Figure 4:- YM vs. diameter for 6% Mg doped ZnO NWs for (100) and (101) orientations at different temperatures.

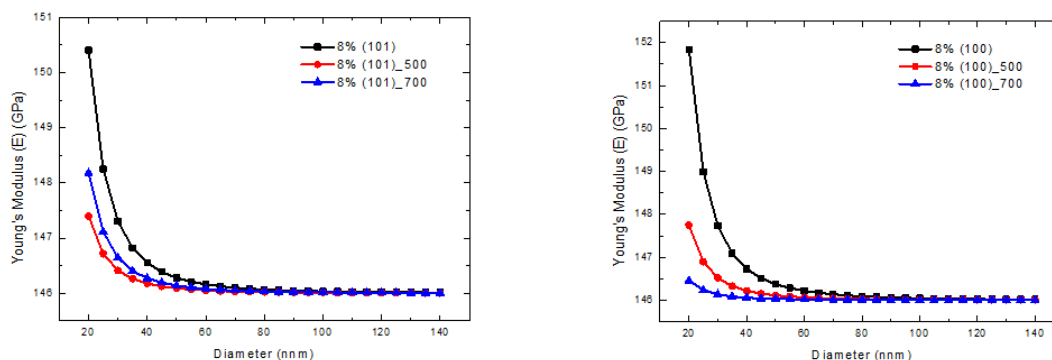


Figure 5:- YM vs. diameter for 8% Mg doped ZnO NWs for (100) and (101) orientations at different temperatures.

A major change in the behavior of the YM is seen from Fig. 4 and 5. It is seen that the trend of the YM for the as grown temperature is changed from the behavior exhibited in the previous two doping concentrations. The maximum value of YM for the 6% doping case was found to be at 600°C temperature which is in good agreement with the 4% result. But, to our surprise the maximum value of the YM unlike the above two cases was seen to be that of the sample at the growth temperature for both the orientations. This has opened a new window for us to explore as to how the above change has taken place.

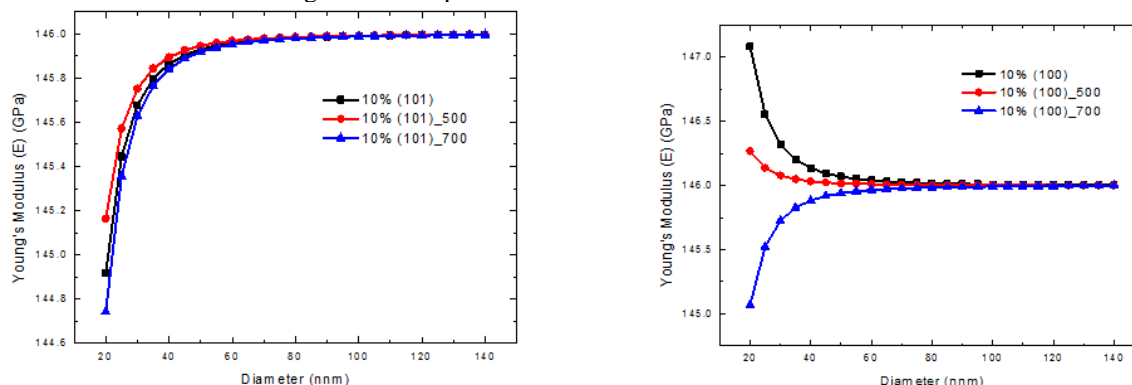


Figure 6:- YM vs. diameter for 10% Mg doped ZnO NWs for (100) and (101) orientations at different temperatures.

An even unexpected turn of behavior was seen in the 10% doping sample because it shows a decrease in the YM of the NWs with decreasing diameters in all the three temperatures at the (101) orientation, but at (100) orientation the behavior was seen to strike an unusual resemblance to that of the undoped case. This is yet to be seen as to what is the reason behind such a behavior seen in the NWs. All of the above discussed results have been computed from the values of the surface stress obtained from ref. 9 and we can see that the surface stress plays the deciding factor in the considered model for the estimation of behavior of Mg doped ZnO NWs. The variation in the surface stress with the corresponding change in the doping concentration of the material is shown in Fig7. This change in the surface stress is one of the pivotal reasons to the behavior observed in 8% and 10% doped samples.

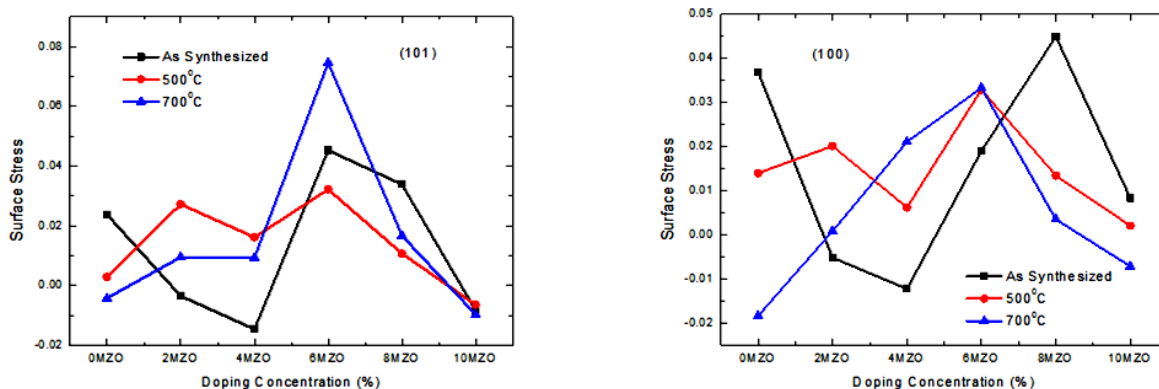


Figure 7:- Surface Stress vs. Doping Concentrations of Mg doped ZnO NWs for (100) and (101) orientations.

Conclusion:-

The results suggest that ZnO NWs can give different natures with the same diameter if the concentrations of doping are altered which would result in the respective change in the surface stress as well. We have mainly focused on the variation that occurs when we consider doping at different temperatures and orientations, and its effect on the YM of the NWs. It is clear that a surface with positive (tensile) surface stress will lead to an increase in the YM whereas the surface with negative (compressive) surface stress will lead to a decrease in the YM of the NWs[5-7]. Thus, as the surface stress plays an important role we can see that it might be the possible key factor in engineering the desired mechanical properties.

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RESEARCH ARTICLE

PHYTOCHEMICAL SCREENING AND TOXICOLOGICAL STUDY OF THE AQUEOUS EXTRACT OF STEM BARK OF *MITRAGYNA INERMIS* (WILD) O. KUNDZE (RUBIACEAE), A TRADITIONAL MEDICINE PLANT.

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Abstract

Background: The use of plants gained widespread as well in developing countries as in developed countries for different purposes and both as mixture or single formula. That widespread use does not guarantee their safety and efficiency. The aim of the present study is to assess the phytochemistry and subacute toxicity of *Mitragynainermis*, an alleged nephroprotective properties plant acclimated in Benin.

Methods: By complexation and/ or precipitation reactions, the different groups of secondary metabolites in these plants were highlighted. The crude aqueous extract of *M. inermis* has been force-fed to rats for seven days. They were observed for toxicity signs and some blood biochemical parameters have been evaluated before and after treatment. We used Microsoft excel and Minitab 1.6 to process data.

Result: Different phytochemicals were found and crude aqueous extract of *M. inermis* doesn't induce toxicity signs on rats after seven-day force-feeding up to 2500 km/kg of body weight using oral route. Biochemical analysis shows a significant decrease in blood level of urea and creatinine up to 2500 km/kg of body weight. The significant increase in ALAT by D2 (1000 km/kg dose) and both in ALAT and ASAT by D3 (2500 km/kg dose) after 7 days is decreased and normalized during the observation period.

Conclusion: Crude aqueous extract of *M. inermis* stem bark is less toxic up to 2500 km/kg of body weight and should be taken during consecutively less than 7 days.

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Introduction:-

The use of plant for nutritional and therapeutic purposes dates back to the 'old world' where trial and error methods were employed in the folklore means to treat ailments and diseases. Traditional medicine has proved to be a key element among the rural communities in developing countries for the provision of primary health care especially

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where there are inadequate primary health care systems (Tijani *et al.*, 2012). The existence of traditional medicine depends on plant species diversity and related knowledge of their use as herbal medicine (Ssegawa *et al.*, 2007). Nowadays, many plants have been studied and some of their attributed empirical properties in healing proved to be true. Most of these plants often display a wide range of biological and medicinal properties such as anti-inflammatory, antihemorrhagic, fertility enhancer, laxative, sexual enhancer, anti-microbial, anti-fungal (Tijani *et al.*, 2012). Thus, the use of these plants gained widespread as well in developing countries as in developed countries. Today, due to the side effects of western medicines (i.e. chemical drugs), drug tolerance, rise of the complications of the disease (despite careful management), and high financial implications imposed on society and individuals; the use of medicinal herbs and traditional medicine has become a priority globally (Zareiet *et al.*, 2015). Conversely, the widespread use and increasing popularity of herbal medicines does not guarantee their efficacy and safety (Shafaei, 2011).

Mitragynainermis is a savannas, forest and forest galleries shrub of 3-14 m in height and it is widely distributed in West to Central Africa (Senegal, Cameroon, Ghana, Ivory Coast, Mali, Chad, DR Congo and Sudan), where it is termed *Nkiwu*, *lèninFonorNago*, *kabe* in *dendi* (Akouegninouet *et al.*, 2006). Well known for its ornamental and medicinal purposes, *Mitragynainermis* medicinal applications however still depends as practiced by inhabitants of different geographical areas. In Benin, the aqueous decoction of root barks and leaves is used orally against anorexia, constipation and in steam bath against leprosy (Akouegninouet *et al.*, 2006). *Mitragynainermis* is reported to be used for the treatment of diabetes, ulcer, piles, dysentery and bone pain among the Hausa/Fulani tribes of northern Nigeria (Adoumet *et al.*, 2012). The hypotensive, cardiothoracic and vasodilatory properties of the aqueous extracts of the plant has also been reported (Ouedragoet *et al.*, 2004). Beyond its medicinal use, crude extract mixtures of leaves and stem from *M. inermis* showed good biopesticide activity against *Lepidopterous caterpillars* infesting plants of cotton (Ndiaye *et al.*, 2008).

The effects on haematological and biochemical parameters should be assessed in order to determine the safe and unsafe use of medicinal plants. Change from normal physiological levels of these parameters after administration of a chemical agent to an experimental animal is an indication of adverse effects of such agent on living organisms (Kolawole *et al.*, 2011). The present study is a preliminary work which contributes to a better knowledge on properties of medicinal plants used in traditional medicine in Benin for their alleged hepatoprotective and nephroprotective properties, especially *Mitragynainermis* of which phytochemical and toxicity are reported here.

Material and Methods:-

Preparation of plant extract:-

For the purpose of this study, the plant material used is composed of stem bark *Mitragynainermis*. Plant material was air-dried to constant weight at room temperature afterwards it was grounded into powder using an electric blender. The powder was then dissolved in distilled water for three days. After every 24 hours, the mixture was filtered through a Whatman filter paper and water was then added for the following 24 hours. Crude extract was collected by removal of water in oven at 40°C. The concentrated extract obtained was used for the trials. Phytochemical examinations of the extracts were carried out for detection of alkaloids, phenols, flavonoids, tannins, saponins, phytosterols, terpenes, glycosides and steroids following the method reported by Koudoro *et al.* (2014).

Experimental Animals:-

Male and female Wistar strain albino rats weighing between 140-200g were used for the purpose of this experiment. The animals were obtained from the Institute of Applied biomedical sciences (ISBA-FSS/FAST, Cotonou, Bénin, from Prof.Laleye) and kept in well ventilated and hygienic animal house of Animal Physiology and experimental pharmacology laboratory under constant environmental and nutritional conditions (18 to 22°C). All the rats were kept in cages and given food and water *ad libitum*.

Experimental Design:-

Animals were randomly divided into four groups of three rats each after body weight measurement to ensure homogeneity of the batches. The experimental groups (labelled group D1, D2, and D3) were administered 100 mg/kg of body weight, 1000 mg/kg of body weight and 2500 mg/kg of body weight of the extract respectively each day. The control group received distilled water. The administration was carried out for a period of 7 days and data was collected up to the 37th day.

Biochemical essays:-

The biochemical assays were performed using blood from the eyes of the rats and samples were collected three times. The first batch of samples were collected five days before drug administration (D0) from the rats was started and the second batch was done on the eighth day after the start of drug administration (D+7); with the third and last batch of samples collected on 37th day from the start of drug administration (D+30). Blood samples were collected into a clean dry test tube without an anticoagulant. The blood then was centrifuged at 3000 rpm for 10 minutes and the collected serum was used for estimation of biochemical parameters. Estimation of biomarkers for kidney (alanine aminotransferase (ALAT) and aspartate aminotransferase (ASAT)) and liver (urea and creatinine) functions were done using spectrophotometer KENZA MAX BioChemistry Photometer according to standard procedures provided along with the kits supplied by Biolab (France).

Data Analysis:-

The values of parameters were compared between groups using one-way analysis of variance (ANOVA) followed by Tukey multiple comparison tests. All the data analysis was performed using Minitab 16. Data was presented as mean \pm SD and significance was taken at $p < 0.05$

Results:-**Phytochemical group:-**

Table 1 showing phytochemical group identified into aqueous crude extract of *Mitragynainermis* reveal the presence of catechic tannins, alkaloids, flavonoids, saponosides, terpenes and sterols. However, compounds such as gallic tannins, anthraquinones and coumarins were absent.

Toxicity study of crude extracts of *M. inermis*:-

Along this part of our paper D0 means Starting day (Fives days before first gavage), D+ 7 means the day after the last gavage, D+ 30 means 30 days after the last force-feeding.

We recorded no particular signs of toxicity on animal during 24 hours following first drug administration. Recorded Signs are piloerection, convulsions, lethargy, diarrhea and sleep. The alive animals number was also recorded. From the following table 2, no death was recorded during the experimental period.

Change in body weight of rats administered with the aqueous extract of *M. inermis* are shown in figure 1.

As per figure 1, there was no significant variation of animals body weight during the experiment ($P > 0.05$) in all groups.

Table 1:- Phytochemical groupements in *M. inermis* stem bark.

		<i>Mitragynainermis</i>
Tanins	catechic	+
	gallic	-
Anthraquinones		-
Alcaloides		+
Flavonoides		+
Mucilages		+
Saponosides		+
Terpens et sterols		+
Coumarines		-

Table 2:- Animals group.

	Number of animal			
	Control group	D1 (100mg/Kg BW)	D2 (1000mg/Kg BW)	D3 (2500mg/Kg BW)
D0	3	3	3	3
D+7	3	3	3	3
D+30	3	3	3	3

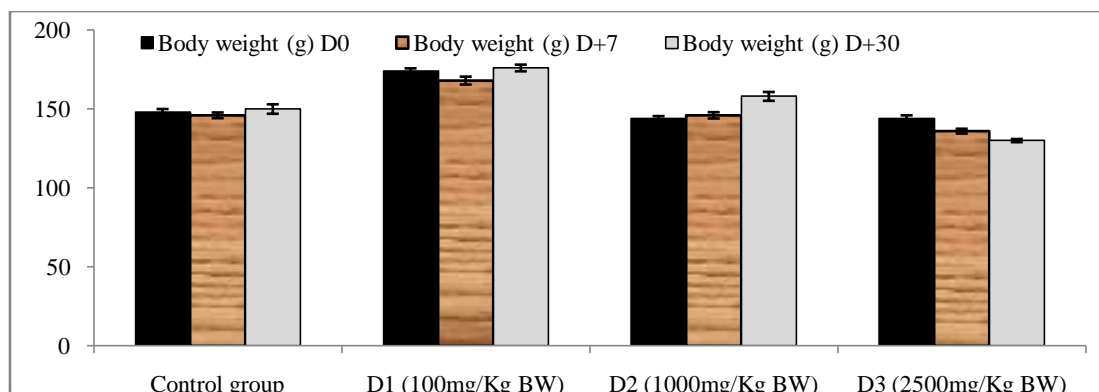
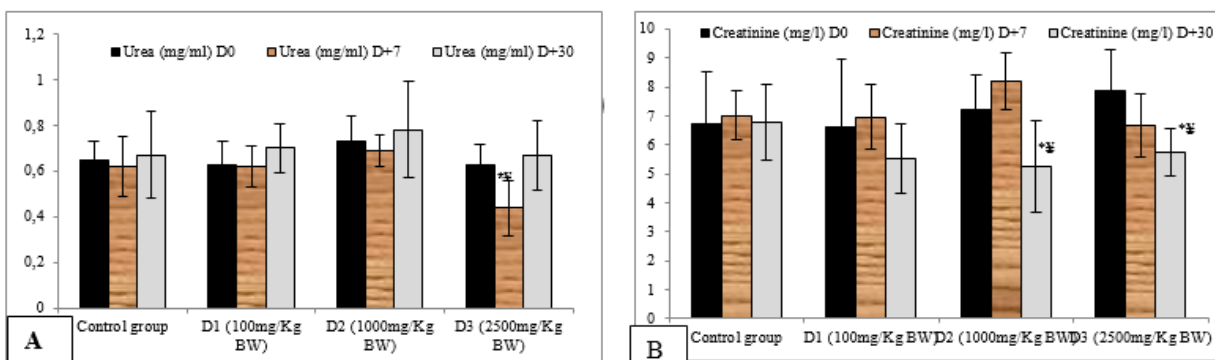
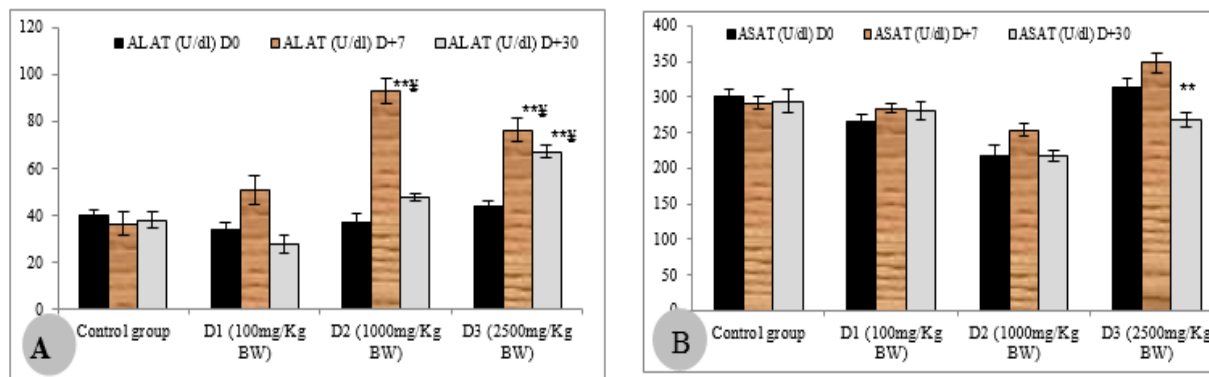


Figure 1:-Body weight of rats.



Significantly different ($p < 0.05$) *: compared to control group ;‡: Compared to D0 in the same group

Figure 2:- Effect of extract on kidney function.



Significantly different ($p < 0.05$) **: compared to control group ;‡: Compared to D0 in the same group

Figure 3:- Effect of extract on Liver function.

Effect of the drug administration on biochemical parameters:-

Effect on kidney function:-

Drug administration to rats at 100 mg/Kg BW and 1000 mg/Kg BW did not show any effect on the urea level in blood during the first 7 days and also after 30 days of observation. But at 2500 mg/Kg BW, the extract reduced significantly the urea level in blood (Fig.2 A). At 1000 mg/Kg BW, the extract significantly reduced the blood creatinine level that had been increased by the seven-day administration (Fig.2 B). The 2500 mg/Kg (BW) dose also reduced blood creatinine level after 30 days from the drug administration stop day.

At 100 mg/Kg (BW), the extract did not change the ALAT level in blood, but at 1000 mg/Kg (BW) and at 2500 mg/Kg (BW), the extract increased ALAT level significantly after seven consecutive days of drug administration (Fig.3 A). After 30 days, the normalization of ALAT level was noticed in group D2, which was significantly higher in comparison to the starting level and the control group (Fig.3 A). Blood level in ASAT was affected by seven-day administration of the extract at 2500 mg/Kg (BW) but normalized after 30 days of observation (Fig.3 A). Other doses (D1 and D2) didn't affect ASAT levels. 100 mg/Kg (BW) and 1000 mg/Kg (BW) doses did not induce significant changes, although there was a small variation after seven-day drug administration (Fig.3 A).

Discussion:-

In the current study, the phytochemicals found presented results similar to those of Konkonet *et al.* (2008) and those of Wakirwaet *et al.* (2013) in *Mitragynainermis* leaves extracts (infusion and decoction); however, contrary to the above mentioned studies saponosides was also present. Similarly to the current study, Ndiayeet *et al.* (2008) also reported finding saponosides besides other phytochemicals such as flavonoids, alkaloids and tannins. The main differences in the findings could be attributed to the difference in plant parts or solvent systems used in different studies. Indeed, these two research groups used *Mitragynainermis* leaves extracts (Konkonet *et al.*, 2008 and Wakirwaet *et al.*, 2013). Ndiayeet *et al.* (2008) used both leaves and stem bark whilst the present study used only stem bark extract for the phytochemical screening. This difference can also be attributed to climate and soil conditions of the harvesting areas (Olaleye, 2007).

The present study showed no particular signs such piloerection, convulsions, lethargy, diarrhoea and sleep. Moreover, after 37 days experimental period, no death within animals was recorded. Such results suggested that the crude aqueous extract of *M. inermis* do not possess acute toxicity up to 2500mg/kg of body weight in rats. Several research groups came to the same conclusion. Konkonet *et al.* (2008) using *M. inermis* leaves extracts estimated LD50 over 5000 mg/kg in mice and concluded that leaves freeze-dried aqueous extract of *M. inermis* was less toxic. Timothy *et al.* (2015) estimated the LD50 of the ethanolic extract of *M. inermis* stem bark at 1587.45 mg/kg for intraperitoneal route and at 4298.84 mg/kg for oral route. Atingaet *et al.* (2015) also concluded that methanol leaf extract *M.inermis* in mice is relatively less toxic after finding intraperitoneal LD50 to be more than 2000mg/kg of body weight. Furthermore, in the current study, the observation of less toxicity was reinforced by other research groups' conclusions using different solvents (water, methanol); in different animal (mice, rats) and different route (oral and intraperitoneal) (Timothy *et al.*, 2015, Uthmanet *et al.*, 2013, Adoumet *et al.*, 2012, Atingaet *et al.*, 2015, and Konkonet *et al.*, 2008).

The experimentation period did not affect animal's body weight suggesting that the crude aqueous extract of *M. inermis* doesn't affect animal appetite. Because kidney and liver toxicity has been reported following the use of phytotherapeutic products (Saadet *et al.*, 2006), biochemical parameters evaluation was important. In this study, it was determined the effect of a regular administration during one week of aqueous extract of two medicinal plants acclimated in Benin on rat serum creatinine, urea, ASAT and ALAT levels as bio-markers of kidney and liver activities. Since plasma urea concentration reflects the balance between urea production in the liver and urea elimination by the kidneys, its increase in plasma can be caused by increased urea production, decreased urea elimination, or a combination of the two. Urea is the dominant urinary osmole in most mammals and may be concentrated a 100-fold above its plasma level in humans and even more in rodents. The present study results showed a significant decrease in blood urea at 2500 mg/kg of body weight. This finding suggested that extracts could increase urea elimination by kidney acting on glomerular filtration rate (Higgins, 2016). Creatinine, which is a waste product in the blood that came from muscle activity and it, is normally removed from the blood by kidneys. Its levels rising when kidney function slows down. Generally, increased urea levels are associated with nephritis, renal ischemia and urinary tract obstruction (Guyton, 2012). Similarly, to urea, creatinine level is reduced significantly by drug administration at 1000 mg/kg and 2500 mg/kg of body weight. That observation supposed a nephroprotective effect of extract.

Transaminases are enzymes having significant metabolic activity within cells and their increase suggested a cellular lesion particularly in the liver, heart, kidney or muscular (Sanogo, 2008). Seven-day administration of the extract increased both ALAT and ASAT levels at 1000 mg/kg of body weight (ALAT) and at 2500 mg/kg of body weight (ALAT and ASAT). Interestingly, the levels of those parameters decreased and normalized after 30 days from drug administration date suggesting that the first seven days of drug intake can cause damage to either liver, heart, kidney or muscular cells.

Conclusion:-

The phytochemical screening in crude aqueous extracts from stem bark of *Mitragynainermis* revealed different phytochemical compound such as tannins, alkaloids, flavonoids, terpenes and sterols but saponosides was absent in leave *M. inermis*. The solvent and plant part used have been shown to be the main causes of the difference. Toxicological study allowed confirming the less toxicity of crude aqueous extracts from stem bark of *M. inermis* using oral route in rat wistar. Biochemical essay also revealed and confirmed less toxicity, suggesting the beneficial effect of the extract up to 2500 mg/kg of body weight for the liver; but the safety duration of the extract intake must be determined for the further pharmacological study of crude aqueous extract from stem bark of *M. inermis*. However, seven-day drug administration affect ALAT and ASAT levels negatively at 2500 mg/kg of body weight.

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RESEARCH ARTICLE

IP-10 AS A PREDICTOR OF TREATMENT RESPONSE IN CHRONIC HEPATITIS C PATIENTS.

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Abstract

Background & Aim: Hepatitis C virus (HCV) is a globally prevalent pathogen and a leading cause of death and morbidity. IFN-gamma inducible protein 10 kDa (IP-10, or CXCL10), predict a more pronounced first phase decline of HCV RNA during anti-viral therapy. We assessed pretreatment serum IP-10 as a predictor of SVR in treated chronic HCV.

Subjects and methods: Seventy adult chronic HCV patients were included in the prospective controlled study and 10 healthy subjects as a control group. All patients were treated with combined pegylated interferon and ribavirin therapy: pegylated IFN- α 2b, 1.5 μ g/kg/week or pegylated IFN- α 2a, 180 μ g/week SC plus ribavirin (15mg/kg/day orally) for 48 weeks and were followed up for 6 months later.

Results: There was statistically significant difference between the Non-SVR group and SVR group as regards IP10 level (395 ± 167.9 vs. 159.9 ± 55.2) pg/ml; AFP level (7.2 ± 1.4 vs. 3.8 ± 1.8) ng/ml and Liver Stiffness measurement by transient elastography (10.9 ± 6.3 vs. 7.4 ± 2.5) kpa, respectively. On multivariate regression analysis for predictors of post-treatment SVR: IP10 was the only independent predictor of achieving SVR (Odds=0.97; C.I 95%= 0.95-0.99; P=0.002). At cutoff point for IP10 more than 216.6 pg/ml has a sensitivity of 90%, specificity 87.5%, PPV 84.4% and NPV 92.1% at P = 0.001; C.I 95% = 0.900-1.000 to define patients with SVR.

Conclusion: Pretreatment IP-10 level should be included as a predictor of SVR. Attempts to neutralize IP-10 or block IP-10 receptor should be considered as additional strategies to improve the outcome of the antiviral therapy.

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Introduction:-

Hepatitis C virus (HCV) is a global health problem with >185 million (2.8%) infections worldwide (Mohd et al, 2013). There is substantial regional variation where Egypt is estimated to have the highest HCV prevalence in the world (14.7%) (Cuadros et al., 2014).

As a consequence of asymptomatic progression of chronic HCV infection, almost 75% of patients first present with complications of cirrhosis, portal hypertension and later develop hepatocellular carcinoma (HCC) (Mitchell et al,

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2010). Some estimations suggest that death related to HCV infection (due to liver cell failure or hepatocellular carcinoma) will increase over the next two decades (**Ghany et al., 2009**).

Cellular immune response induced to HCV may affect the severity of liver injury. Inflammatory responses are regulated by complex mechanisms and probably depend on genetic determinants such as HLA expression and chemokines such as interferon-gamma-inducible protein-10 (IP-10) (**Hraber, 2007 and Larrubia, 2008**).

The treatment regimen with pegylated interferon (PEG-IFN) alfa-2a or alfa-2b together with ribavirin (RBV) was the standard of care (SOC) for HCV patients (**Alexopoulou & Karayiannis, 2015**). With improved understanding of the HCV genome, key viral enzymes and life cycle, direct-acting drugs (DAAs) have developed, that show good promise for the future (**Alexopoulou & Karayiannis, 2015**).

Response to treatment is affected by various viral and host factors. Among the identified predictors of response, the host genetic polymorphisms and the pre-treatment activation of IFN-stimulated genes (ISG) (**Kau et al, 2008, Fattovich et al, 2011**).

Single nucleotide polymorphisms (SNPs) near IL28B, which encodes the type III interferon IFN- λ 3, can be used in routine clinical practice for informing treatment decisions. IFN- λ 3 are strongly associated with the response to treatment of chronic hepatitis C, it up regulates IFN-stimulated genes and affects the adaptive immune response (**Rauch et al, 2010; Thompson et al, 2010**). However, the IL28B genotype alone is not a perfect predictor of treatment outcome (**Ogawa et al., 2012**).

On the other hand, it has been shown that low levels of intrahepatic and systemic chemokine CXCL10 (IP-10, or CXCL10), a valid surrogate marker of ISG activation, predict a more pronounced first phase decline of HCV RNA during anti-viral therapy (**Askarieh et al, 2010**) and increased SVR rates (**Romero et al, 2006**).

HCV itself is suggested to be responsible for elevated serum IP-10 levels found in HCV-infected patients. HCV proteins such as NS5A and core, alone or in combination with proinflammatory cytokines, can induce IP-10 gene expression and secretion (**Apolinario et al., 2005**). Pretreatment IP-10 levels correlated with HCV viral load, ALT levels, hepatic inflammation and fibrosis. Potent DAA therapy is associated with a rapid reduction in plasma IP-10 level that parallels the reduction in HCV RNA level, both in treatment-naïve patients and treatment-experienced patients (**Chatterjee et al., 2012**).

Aim:-

The aim of this work was to assess pretreatment serum IP-10 as a predictor of SVR in treated chronic HCV Egyptian patients

Subjects and Methods:-

This prospective controlled study was conducted on 70 patients with chronic hepatitis C virus infection. They were recruited from the outpatient clinic of Hepato-Gastroenterology department, National Liver Institute, Menoufia University. All patients were ≥ 18 years old, and eligible for pegylated interferon plus ribavirin therapy. Diagnosis of chronic HCV was confirmed by real-time PCR. Patients underwent antiviral treatment and follow-up protocol according to the standard clinical practice for 18 months. In addition, 10 apparently healthy subjects of matched age and gender were enrolled as a control group. The study was approved by the ethics committee, and enrollment in the study was conditioned by a written informed consent from all participants in the study.

Exclusion criteria:-

1- Previously treated, or not eligible for treatment. 2- Patients with HBV or HIV coinfection. 3- Patients with evidence of HCC or other liver diseases. 4- Patients having moderate to severe uncorrected anemia, neutropenia and thrombocytopenia. 5- Patients with a history of cardiovascular and neuro-psychiatric diseases.

All patients were treated with combined pegylated interferon and ribavirin therapy: pegylated IFN- α 2b 1.5 μ g/kg/week or pegylated IFN- α 2a 180 μ g/week SC per week, plus ribavirin 15mg/kg/day orally. Patients received treatment for 48 weeks and were followed up for 6 months after the end of treatment.

A rapid virological response (RVR) was defined as undetectable HCV RNA in serum at week 4 of therapy. Early virological response (EVR) was defined as serum HCV RNA negativity or any $>2 \log_{10}$ decline in HCV RNA levels at week 12 of therapy compared with baseline. Patients with sustained virological response (SVR) were those with undetectable HCV RNA in serum 24 weeks after stopping therapy. Treatment failures included patients who had a $< 2 \log_{10}$ drop in viral load at week 12 as compared to baseline, those whose HCV RNA was still detectable at week 24 (i.e. nonresponders), and those who had undetectable HCV RNA at the end of therapy but detectable HCV RNA at 24 weeks after cessation of therapy (i.e. relapsers) (Ghany et al., 2009).

Laboratory Assessment:-

Base line investigation; blood samples were taken before starting treatment. They were tested for: HCV RNA using the COBAS® TaqMan® HCV Quantitative Test (Roche Molecular Diagnostics, CA, USA) with lower limit of quantitation of 15 IU/ml; complete blood count (CBC) using Sysmex K-21 (Sysmex Corporation, Japan); blood sugar, liver & kidney function tests using fully automated Beckman Coulter chemistry analyzer Au 480 (Beckman Coulter Inc., CA, USA); alpha fetoprotein (AFP) & TSH hormone using (Advia Centaur CP immunoassay system, Siemens Healthcare Diagnostics Inc, USA)

Repeated measurements of liver function tests, complete blood count and HCV RNA were done on weeks (12, 24, and 48) during treatment and at 6 months after stopping therapy.

End Points:-

SVR response was considered as primary end point. Non-response and/or relapse were considered as the secondary end point.

Quantification of serum level of IP-10:-

Interferon- γ Inducible Protein -10 (IP- 10) levels were measured in serum samples (separated and stored at -80°C) using a solid phase sandwich ELISA immunoassay (Quantikine human CXCL10 / IP10 immunoassay, R&D System, Minneapolis, USA) according to the manufacturer's instructions.

Liver biopsy:-

Ultrasound-guided percutaneous liver biopsy was performed to determine the degree of liver fibrosis at baseline. The histopathological assessment of necro-inflammatory grade and fibrosis stage was recorded using Metavir scoring system (Fibrosis was staged on a 0-4 scale. Activity was graded as: A0, none; A1, mild; A2, moderate; and A3, severe. Fibrosis stages \geq F2 were considered significant) before commencing treatment (Bedossa et al., 1996).

Radiological investigations:-

All patient had abdominal ultrasonography; liver stiffness measurement (LSM) by transient Elastography using fibroscan apparatus ((Echosens, Paris, France) done initially within a week of liver biopsy and at the end of follow-up period (Andersen et al., 2009).

Statistical analysis:-

Data was statistically analyzed using IBM® SPSS® Statistics® version 21 for Windows. Data are expressed as mean \pm standard deviation, number with column percentage and the median \pm range or 95% confidence interval for non-parametric data. All p-values are 2 tailed, with values <0.05 considered statistically significant, $p = 0.01$ is highly significant and $p = 0.001$ is very highly significant. Comparisons between two groups were performed using the Student's t-test for parametric data "normally distributed", and Mann-Whitney test for nonparametric data "not normally distributed". CHI-squared test (χ^2) and Fisher exact test for categorical data analysis. Regression analysis is used to find how one set of data relates to another. Univariate and multivariate binary logistic regression were done for detecting the predictors of the event.

The area under the receiver operating characteristic (AUROC) curve analysis was used for detection of the cutoff value of the proposed tests. An AUROC value of 0.90-1.0 indicated excellent, 0.80-0.89 good, 0.70-0.79 fair, 0.60-0.69 poor and 0.50-0.59 no useful performance for discrimination of the outcome under assessment. **Sensitivity:** the proportion of patients with disease who are correctly identified. **Specificity:** the proportion of patients without disease who are correctly identified. **Positive predictive value:** the proportion of patients with positive test results who are correctly diagnosed. **Negative predictive value:** the proportion of patients with negative test results who are correctly diagnosed.

Results:-

This was a prospective randomized controlled study which was conducted on 70 chronic hepatitis C patients and 10 healthy persons enrolled as a control group. They were recruited from the outpatient clinic of Hepatology department, National Liver Institute, Menoufia University.

Comparison between control group and treated group:-

There was statistically significant difference between both groups regarding **Albumin** (4.4 ± 0.4 vs. 4 ± 0.2) g/dl, **AST** (21.3 ± 4 vs. 49.6 ± 38.2) U/L, **ALT** (29.9 ± 4.2 vs. 53.7 ± 39.2) U/L, **Creatinine** (0.7 ± 0.1 vs. 0.8 ± 0.2) mg/dl, **Platelets** (253900 ± 56398.3 vs. 6734.4 ± 2165.5) /mm³ and **IP10** (102.2 ± 51 vs. 260.7 ± 165.2) pg/ml in the control group vs. treated group respectively.

However, there was no statistical difference between both groups regarding **age, TSH, RBS, AFP, bilirubin, WBCs, Hb, Prothrombin conc. And INR**

The mean \pm SD of transient elastography was (8.9 ± 4.8) kpa in the treated group patients before starting treatment. The base-line HCV RNA level was (1548683.5 ± 1955482.9) IU/ml in treated group (table 1, Figure 1)

Comparison between sustained vs. non sustained virus responders:-

Sustained virus response (SVR) comprise 57.1% of chronic hepatitis C patients as 40 patients achieved SVR while 30 patients were Non-SVR achievers, who were further subdivided to 20 non responders who did not achieve the end of treatment (EOT) and 10 relapsers (achieved EOT but not SVR).

There was statistically significant difference between the Non-SVR group and SVR group as regards **IP10 level** (395 ± 167.9 vs. 159.9 ± 55.2) pg/ml (table and figure 2); **AFP level** (7.2 ± 11.4 vs. 3.8 ± 1.8) ng/ml and **Liver Stiffness measurement by transient Elastography** (10.9 ± 6.3 vs. 7.4 ± 2.5) kpa, respectively (table 2, figure 3). On the other hand, there was no statistically significant difference between the two groups as regarding **age, bilirubin, albumin, AST, ALT, creatinine, hemoglobin, WBCs, platelets, prothrombin conc., INR and base line HCV RNA level** (table 2)

However, there was a significant difference between the two groups regarding the presence of significant fibrosis ($F \geq 2$) ($P = 0.009$) (table 3) and activity ($P = 0.023$) by Metavir scoring (table 4). Furthermore, there was a significant difference between both groups regarding the achievement of RVR (rapid viral response), EVR (early viral response) and EOT (end of treatment response). As 37 (92.5 %) RVR achievers ($P = 0.023$) (table 5, figure 4), 39 (97.5 %) EVR achievers ($P = 0.001$) and 40 (100%) patients who achieved EOT were SVR achievers ($P = 0.001$) (table 6, figure 4).

Comparison between patients with non-significant fibrosis and patients with significant fibrosis:-

There was statistically significant difference between both groups as regarding **age** which was significantly higher in patients with significant fibrosis than in those with non-significant fibrosis (47.5 ± 7.8 vs. 39.7 ± 10.6 years old). **Prothrombin conc.** was significantly lower in patients with significant fibrosis (83.7 ± 10.2 vs. 89.3 ± 8.6) and **INR** was significantly **higher** in patients with significant fibrosis than in those with non-significant fibrosis (1.2 ± 0.1 vs. 1.1 ± 0.1).

RBS and Transient Elastography were **higher** in patients with significant fibrosis than in those with non-significant fibrosis with values (121.6 ± 37.7 vs. 100.8 ± 17.7 mg/dl) and (15.4 ± 6.5 vs. 7.1 ± 1.9 kpa) respectively.

On the other hands, there was no statistical significance between two groups concerning **IP10, Bilirubin, Albumin, AST, ALT, Creatinine, Hb, WBCs, Platelets, AFP, TSH and base-line HCV RNA level** (table 7).

However, there was a significant difference between the two groups regarding **SVR** as 11 out of 15 patients (73.3%) with significant fibrosis were Non-SVR achievers vs. 19 out of 55 patients (34.5 %) with non-significant fibrosis were Non-SVR achievers ($P = 0.009$) (table 8).

Predictors of treatment:-

On multivariate regression analysis for Predictors of post-treatment SVR **IP10** was the only independent predictor of achieving SVR (Odds=0.97; C.I 95%= 0.95-0.99; $P=0.002$) (table 10).

At cutoff point for IP10 more than 216.6 pg/ml has a sensitivity of 90% and specificity 87.5% and PPV 84.4% and NPV 92.1% at P = 0.001; C.I 95% = 0.900-1.000 to define patients with SVR (figure 6).

Table 1:- Baseline characteristics of treated group vs. control group

	Groups		P-value	Treated Group		P-value
	PegINF/RBV N=70 M \pm SD	Control N=10 M \pm SD		SVR N=40 M \pm SD	Non-SVR N=30 M \pm SD	
Age (years old)	41.4 \pm 10.6	41.4 \pm 10.6	0.5	40.1 \pm 9.4	43.2 \pm 11.9	0.2
Bilirubin (mg/dl)	0.7 \pm 0.3	0.7 \pm 0.3	0.96	0.7 \pm 0.3	0.7 \pm 0.3	0.3
Albumin (g/dl)	4 \pm 0.2	4.4 \pm 0.4	0.002*	4.4 \pm 0.4	4.4 \pm 0.3	0.6
AST (U/L)	49.6 \pm 38.2	21.3 \pm 4	0.001*	45.6 \pm 34.2	54.8 \pm 43.1	0.3
ALT (U/L)	53.7 \pm 39.2	29.9 \pm 4.2	0.02*	48.6 \pm 28.3	60.5 \pm 49.9	0.2
Creatinine (mg/dl)	0.8 \pm 0.2	0.7 \pm 0.1	0.01	0.9 \pm 0.2	0.8 \pm 0.2	0.3
Hemoglobin (g/dl)	14.4 \pm 1.5	15.2 \pm 0.6	0.07*	14.6 \pm 1.4	14.2 \pm 1.6	0.3
WBCs (/mm ³)	6734.4 \pm 2166	7690 \pm 1495.5	0.2	6507.8 \pm 2282.7	7036.7 \pm 1996.1	0.3
Platelets (/mm ³)	213700 \pm 58226	253900 \pm 56398	0.04	213275 \pm 57493.4	214266.7 \pm 60172.1	0.9
Prothrombin conc. (%)	88.1 \pm 9.2	91.6 \pm 5	0.2	89.6 \pm 8.7	86.1 \pm 9.6	0.1
INR	1.1 \pm 0.1	1.1 \pm 0.1	0.5	1.1 \pm 0.1	1.1 \pm 0.1	0.1
RBS (mg/dl)	105.3 \pm 24.6	94.9 \pm 8.3	0.2	106.1 \pm 24.6	104.1 \pm 25	0.7
AFP (ng/ml)	5.2 \pm 7.7	3.2 \pm 0.9	0.4	3.7 \pm 1.8	7.2 \pm 11.4	0.03*
TSH (uIU/ml)	1.8 \pm 0.8	2.1 \pm 0.8	0.3	1.9 \pm 0.8	1.7 \pm 0.6	0.2
IP10 (pg/ml)	260.7 \pm 165.2	102.2 \pm 51	0.001*	159.9 \pm 55.2	395 \pm 167.9	0.001*
Transient Elastography (Kpa)				7.4 \pm 2.5	10.9 \pm 6.3	0.02*
Baseline HCV RNA level (IU/ml)				1210637.6 \pm 1422299.5	1999411.2 \pm 2451529.2	0.4*

*Mann-Whitney test

*AST: Aspartate Amino Transferase

*ALT: Alanine Amino Transferase

*WBCs: White Blood Cells

*INR: International Normalized Ratio

*RBS: Random Blood Sugar

*AFP: Alpha Feto Protein

*TSH: Thyroid Stimulating Hormone

*IP10: Interferon γ Inducible Protein 10

*SVR: Sustained Virological Response

Table 2:- Significant fibrosis and activity by Metavir scoring system in SVR group and Non-SVR group

Metavir scoring system			Non-SVR		SVR		P value
Fibrosis	Non-significant	F < 2	N	%	N	%	
	Significant	F \geq 2	19	63.3 %	36	90 %	0.009
Total			30	100%	40	100%	
Activity	Activity grade	A1	15	50%	31	77.5%	0.023
	Activity grade	A2	15	50%	9	22.5%	
Total			30	100%	40	100%	

Table 3:- RVR, EVR and EOT in SVR group and Non-SVR group.

		Non-SVR		SVR		P value
RVR	Non-RVR	N	%	N	%	
	RVR	21	70 %	37	92.5 %	0.023
Total		30	100%	40	100%	
EVR	Non-EVR	11	36.7%	1	2.5%	0.001
	EVR	19	63.3%	39	97.5%	
Total		30	100%	40	100%	
EOT	Non-EOT	20	66.7%	0	0%	0.001
	EOT	10	33.3%	40	100%	
Total		30	100%	40	100%	

*RVR: Rapid Virological Response

*EVR: Early Virological Response

*EOT: End of Treatment Response

Table 4:- Comparison between patients with non-significant fibrosis and patients with significant fibrosis:

	Significant Fibrosis	Non-significant Fibrosis	P
	N=15	N=55	
	M ±SD	M ±SD	
Age (years old)	47.5 ±7.8	39.7 ±10.6	0.02*
Bilirubin (mg/dl)	0.6 ±0.3	0.7 ±0.3	0.4
Albumin (g/dl)	4.3 ±0.4	4.4 ±0.4	0.6
AST (U/L)	60.5 ±39	46.6 ±37.8	0.2
ALT (U/L)	71.1 ±58.5	48.9 ±31.1	0.2*
Creatinine (mg/dl)	0.8 ±0.1	0.8 ±0.2	0.4*
Hemoglobin (g/dl)	14.3 ±1.5	14.4 ±1.5	0.8
WBCs (/mm ³)	7073.3 ±2316.8	6642 ±2135.4	0.5
Platelets (/mm ³)	194866.7 ±59567.3	218836.4 ±57330	0.6
Prothrombin (%)	83.7 ±10.2	89.3 ±8.6	0.04
INR	1.2 ±0.1	1.1 ±0.1	0.01
RBS (mg/dl)	121.6 ±37.7	100.8 ±17.7	0.03*
AFP (ng/ml)	5.2 ±2.5	5.3 ±8.6	0.9
TSH (uIU/ml)	1.8 ±0.8	1.8 ±0.7	0.9
IP10 (pg/ml)	286.7 ±155.6	253.6 ±168.4	0.5
Transient Elastography (kpa)	15.4 ±6.5	7.1 ±1.9	0.001*
Baseline HCV RNA level (IU/ml)	1741870.5 ±2492666.4	1495996.1 ±1806111.7	0.2

*Mann-Whitney test

*AST: Aspartate Amino Transferase

*ALT: Alanine Amino Transferase

*WBCs: White Blood Cells

*INR: International Normalized Ratio

*RBS: Random Blood Sugar

*AFP: Alpha Feto Protein

*TSH: Thyroid Stimulating Hormone

*IP10: Interferon γ Inducible Protein 10

Table 5:- Comparison between patients with non-significant fibrosis and patients with significant fibrosis regarding SVR achievement.

			Fibrosis Stage		Total	p
			Non-significant Fibrosis	Significant Fibrosis		
SVR	Non-SVR	N	19	11	30	0.009
		%	34.5 %	73.3 %	42.9 %	
	SVR	N	36	4	40	
		%	65.5 %	26.7 %	57.1 %	
	Total	N	55	15	70	
		%	100 %	100 %	100 %	

*SVR: Sustained Virological Response

There was statistically significant difference between the two groups regarding **SVR** as 11 patients (73.3%) with significant fibrosis were Non-SVR achievers vs. 19 patients (34.5 %) with non-significant fibrosis were Non-SVR achievers (P = 0.009)(table8).

Table 6:- Multivariate regression analysis for Predictors of post-treatment SVR Predictors of SVR [multivariate analysis]

	B	Wald	P	Odds	95% C.I
IP10 (pg/ml)	-0.03	9.73	0.002	0.97	0.95 -0.99
Transient Elastography (kpa)	-0.26	1.11	0.291	0.77	0.47 – 1.25
Positive RVR	-0.10	0.00	0.980	0.90	0.001 – 2549.78
Positive EVR	-4.13	0.70	0.404	0.02	0.01-260.38
Non-significant Fibrosis	1.49	0.95	0.329	4.42	0.22 – 87.46
Constant	8.64	4.39	0.036	5667.04	

*IP10: Interferon γ Inducible Protein 10

On multivariate regression analysis for Predictors of post-treatment SVR:-

IP10 was the only independent predictor of achieving SVR (Odds=0.97; C.I 95%= 0.95-0.99; P=0.002)(table 10).

Table 7: Receiver operating characteristic (ROC) curve analysis for IP10 to define patients with SVR:

Cutoff \geq	AUC	P	95% C.I
216.6	0.950	0.001	0.900 – 1.000
Sensitivity		90.0	
Specificity		87.5	
Positive predictive value		84.4	
Negative predictive value		92.1	

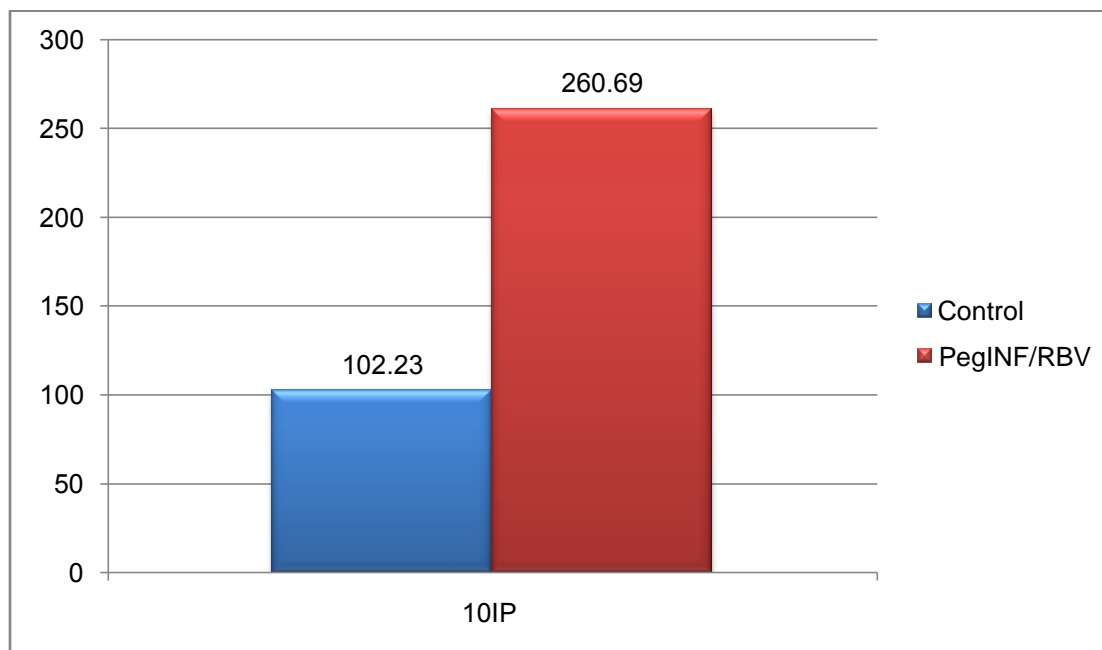
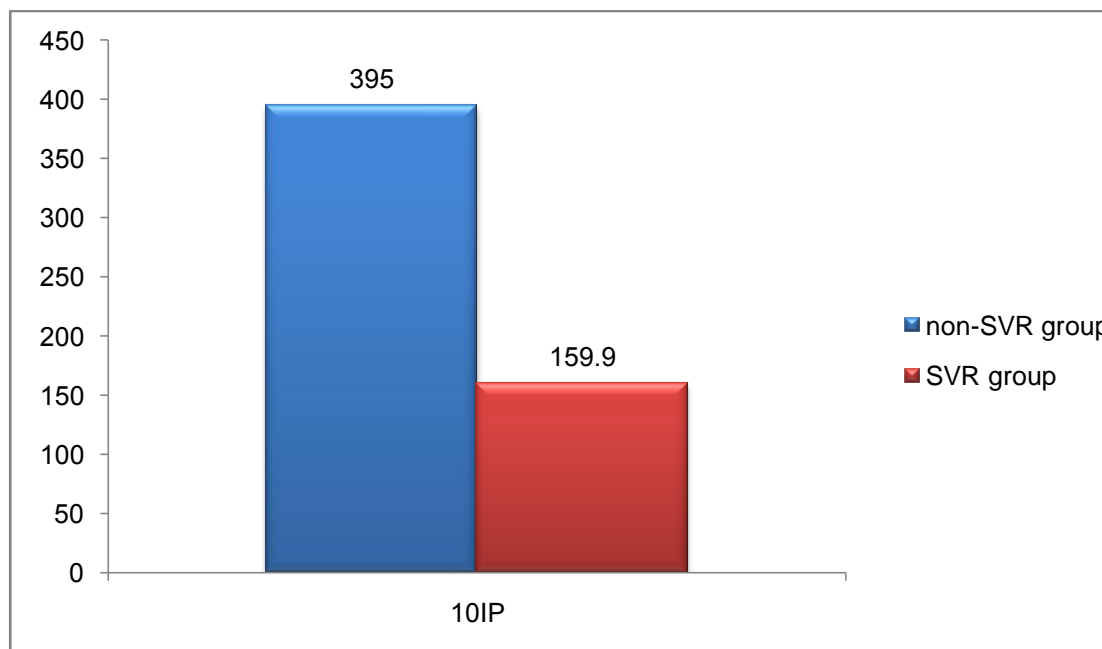
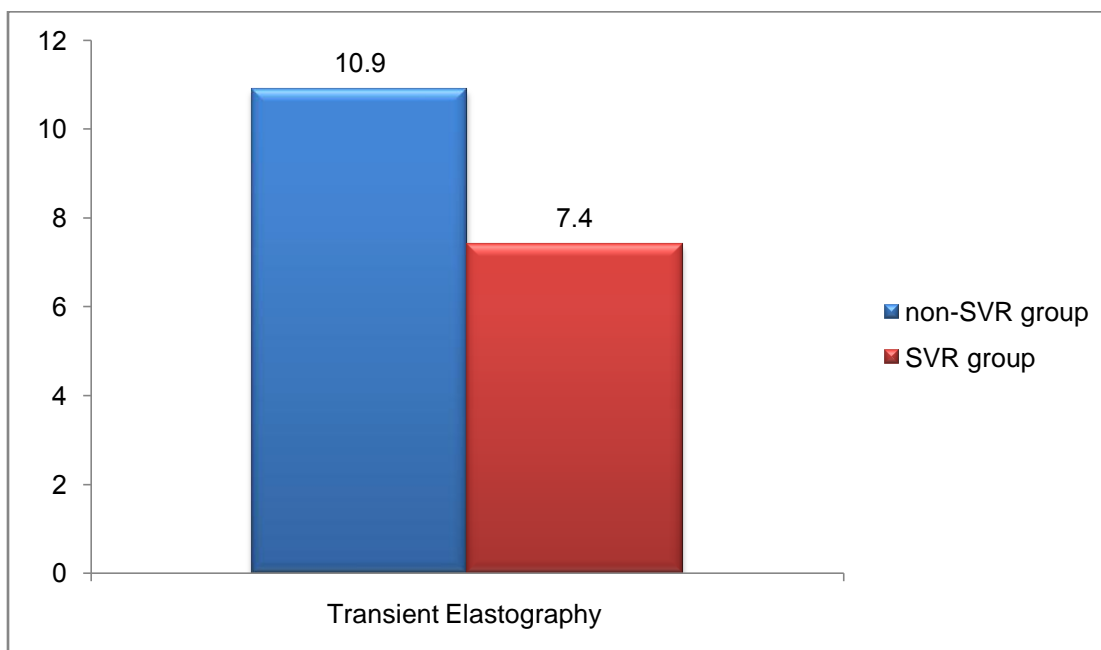
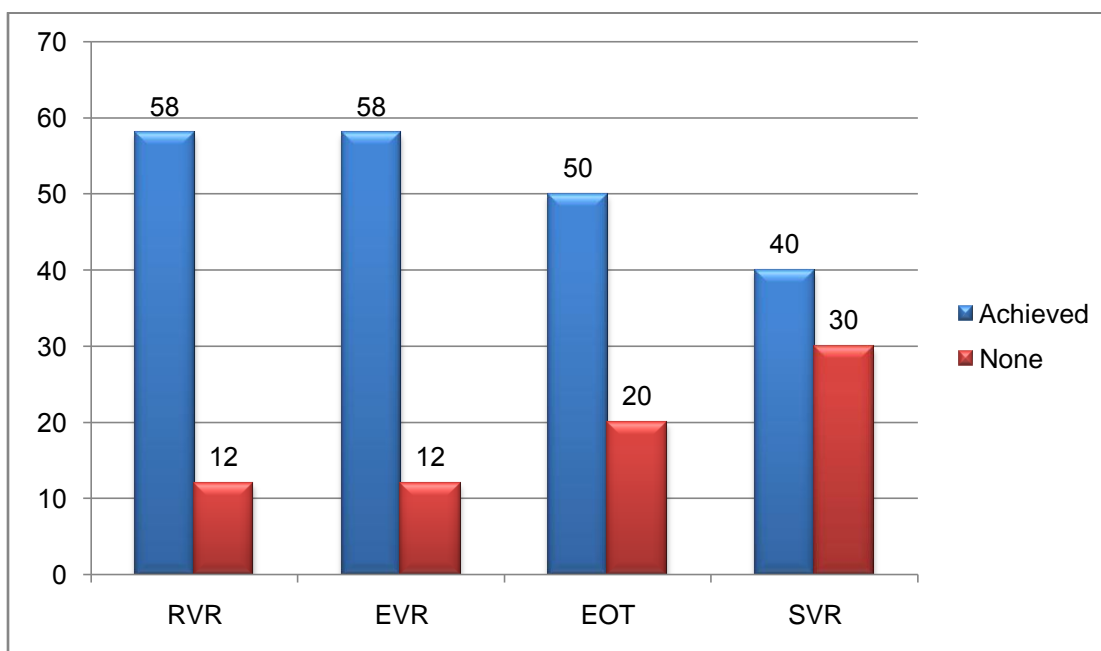
**Figure 1:- IP10 level in control and treated groups**

Figure 2:- IP10 level in SVR group and Non-SVR group.**Figure 3:-** Transient Elastography in SVR group and Non-SVR group.**Figure 4:-** Achievement of (RVR, EVR, EOT and SVR) among the studied patients.

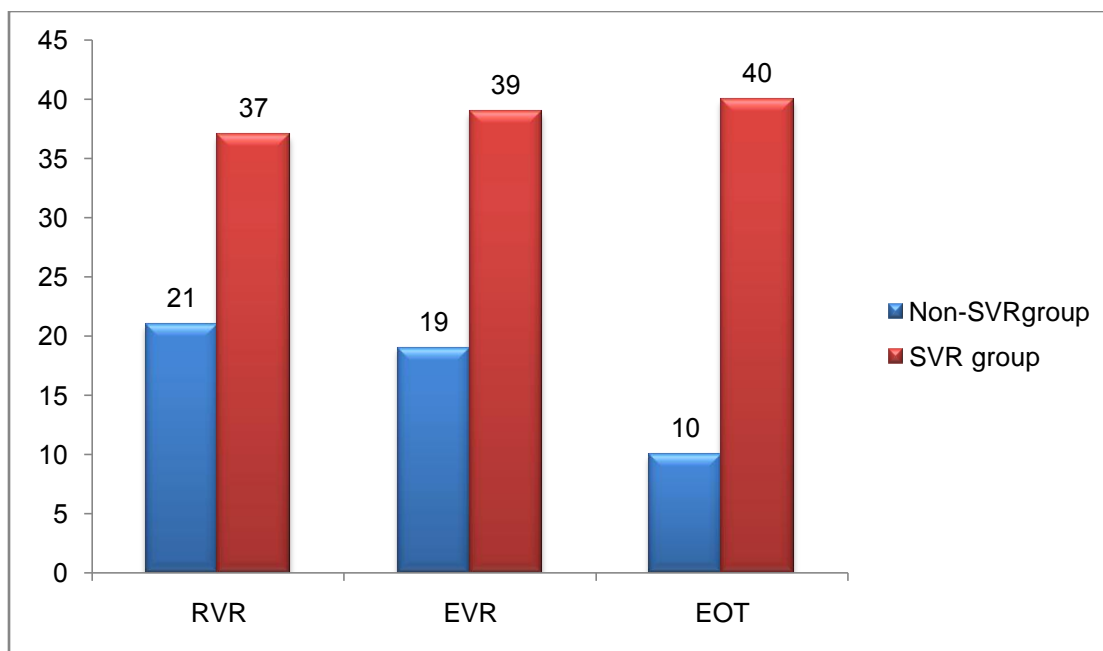


Figure 5:- Achievement of (RVR, EVR and EOT) between SVR and Non-SVR patients.

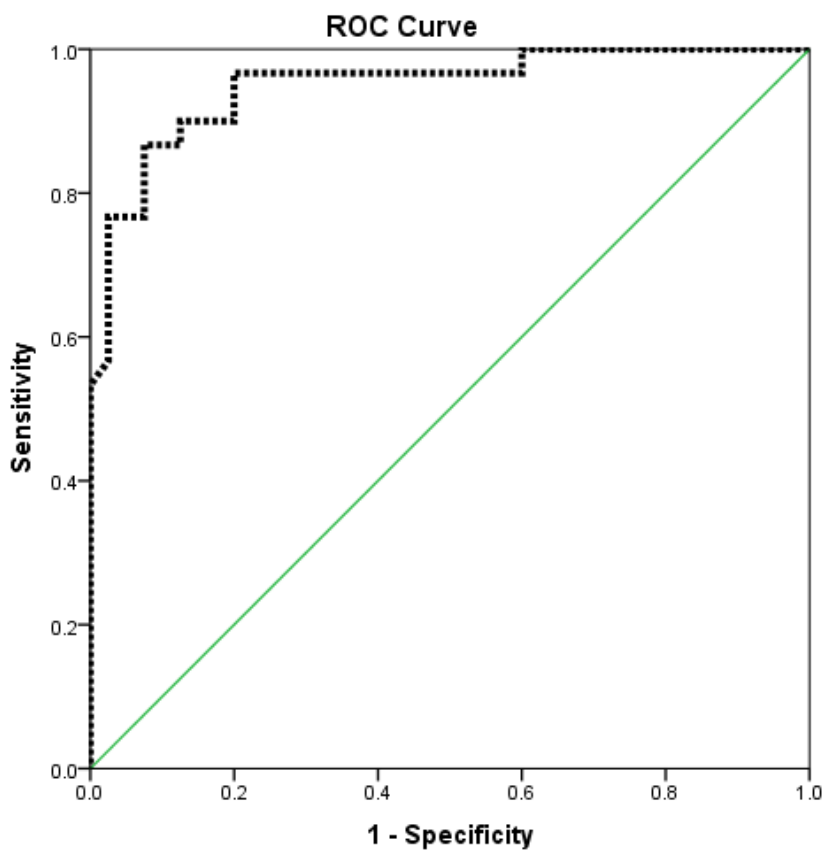


Figure 6:- Receiver operating characteristic (ROC) curve analysis for IP10 to define patients with SVR

Discussion:-

There are many estimates of the number of people in Egypt that are infected. Many publications suggest that nearly 14 % of the people in Egypt were infected. This is ten times greater than in any other country in the world (Sievert et al., 2011). The genotype 4 is predominant in 91% of these patients (Guerra et al., 2012).

The combination therapy with interferon- α and ribavirin, neutralize the virus after 6 months in 40-50% of the infection cases with genotype 1 and in 80% of the infection cases with genotype 2 and 3 (Schuppan et al., 2003). Infections of HCV genotype 4 is, as genotype 1, relatively resistant to the interferon- α /ribavirin combination therapy medical treatment (El Makhzangy et al., 2009). A major paradigm shift happened in HCV treatment, with the advent of highly effective, simplified and short duration (12 weeks) oral DAA-based regimens (Dore et al., 2012). However, the effect of these new therapies, even in developed countries, will be modest without expanded access to treatment (Thomas et al., 2010).

Host genetic factors may predict the outcome and treatment response in HCV infection. IFN- γ -inducible protein 10 (IP-10; also known as CXCL-10) is a member of the CXC subfamily of chemokine. It is induced in monocytes, fibroblasts, and endothelial cells by IFN- γ . IP-10 stimulates monocytes, natural killer cell, and T-cell migration and acts as a chemoattractant for CXCR3 +T cells. Baseline plasma IP-10 levels reflect intrahepatic levels and are a prognostic marker of treatment outcome for patients treated with pegylated interferon and ribavirin (Askarieh et al., 2010).

IP-10 decrease to baseline in those who spontaneously clear infection but remain elevated in those who develop chronic infection suggesting that HCV infection itself is driving the elevated IP-10 levels (Chattergoonn et al., 2011).

The aim of this study was to assess the efficacy of serum IP-10 as a predictor of SVR in chronic hepatitis C virus Egyptian patients receiving Peg-IFN and RBV therapy. This was a prospective study which was conducted on 70 patients with chronic hepatitis C. In addition, 10 healthy persons with matched age and sex were enrolled as a control group.

Patients were categorized into two groups according to their sustained virological response (SVR). **Group (1):** patients who achieved SVR. **Group (2):** patients who didn't achieve SVR including non-responders and relapsers.

SVR was achieved in 40 patients (57.1%) while 30 patients were Non-SVR achievers (42.9%); 20 of them were non-responders and 10 were relapsers. These results coincide with Gad et al. (2008) who reported that SVR rate was 54.8% in genotype-4. Another study conducted on 88 Egyptian patients with chronic HCV infection naive to treatment with PEGIFN/RBV reported SVR rate of 39.8% (El Razikyet al., 2015).

As regards **age and gender distribution** we revealed that no statistically significant difference was found between SVR and Non-SVR groups ($p=0.2$, 0.13 respectively). Previous studies showed that younger age of patients was a significant predictor of treatment response (Codes et al., 2007; Antonucci et al., 2007; Assad et al., 2014). However, the significance of age has not been observed concordantly with other reports. Shiffman et al. (2007) showed that older age, was not an unfavorable marker for IFN α treatment. In addition, Adnan et al. (2013) reported a non-significant relationship between age and response to IFN therapy. This discrepancy may come from weak power of significance and/or differences in sample size.

Several reports (Codes et al., 2007; Assad et al., 2014) showed that female sex was a significant predictor of treatment response. In addition, Di Marco et al. (2013) reported that menopause is related to an increased severity of liver fibrosis, and a lower likelihood of response to therapy with peg-IFN and ribavirin.

The decreased rate of a complete response to IFN α treatment may correspond to decrease in estrogen levels. Interleukin 1, associated with an inflammatory response, is stimulated by low concentrations of estrogen and progesterone. A low concentration of estrogen allows peripheral blood monocytes to secrete more interleukin 1. The spontaneous production of interleukin 1 by peripheral mononuclear cells has been shown to be significantly higher in patients with CHC than in healthy control individuals, and decreased in those with a complete response after the administration of IFN α . This cytokine production may alter the effectiveness of IFN α treatment in perimenopausal and menopausal women with CHC infection (Di Marco et al., 2013).

It is well known that advanced fibrosis or cirrhosis is a poor prognostic factor for response to antiviral therapy compared with no or minimal fibrosis (**Poyfriedard et al., 2000; Poynard et al., 2002; Fried et al., 2002; Hadziyannis et al., 2004; Everson et al., 2006**). In agreement with these studies; we found that SVR was significantly higher in patients with non-significant fibrosis than in patients with significant fibrosis as (73.3% vs. 34.5 %; $P = 0.009$).

In addition, we found that the mean of Transient Elastography was significantly higher in Non-SVR achievers (10.9 ± 6.3) KPa than in the SVR achievers (7.4 ± 2.5) KPa. This was matched with **Stasi et al. (2015)** who showed that liver stiffness (LS) >12 KPa prior to the initiation of dual therapy was significantly associated with poor response to therapy ($p < 0.025$). LS >12 KPa should be considered a strong prognostic indicator of non-response to anti HCV treatment (**Martinez et al., 2012**).

In our work, we found that RVR was a significant predictor of SVR. Thirty seven patients (92.5 %) of forty SVR achievers were RVR while twenty one patients (70%) of thirty Non-SVR achievers were RVR ($P = 0.023$). This was coincided with a study conducted by **Ferenci et al. (2004)**, he observed that patients infected with HCV genotypes 2 or 3 achieved RVR in higher proportions than patients infected with genotype 1. However, regardless of the HCV genotype, patients who reached RVR have the highest rates of SVR. In the study by (**Fried et al., 2011**) RVR was achieved by 16% of patients with genotype 1, 71% of genotype 2 and 60% of genotype 3. Among individuals who reached RVR, the SVR rate was high across all HCV-genotypes and ranged from 88% to 100% (genotypes 1-4) (**Mangia et al., 2010**).

Also in our study, we found that there was statistically significant difference between SVR achievers and Non-SVR achievers as regarding achievement of EVR as 39 (97.5 %) patients who achieved EVR were SVR achievers ($P = 0.001$). This was matched with a study from China which showed that patients without EVR rarely achieve SVR (**Reddy et al., 2005**).

In our work we found that, **AFP** level, was **higher** in Non-SVR achievers than in SVR achievers (7.2 ± 11.4 vs. 3.8 ± 1.8 ; $p = .03$). This was matched with **Males and colleagues in 2007** who confirmed the predictive role of AFP concerning treatment response to chronic hepatitis C. In a previous study, serum AFP remained independently associated with SVR after controlling other factors associated with SVR, including liver fibrosis (**Dabeva et al., 1993**).

Previous studies have shown that AFP can be considered as one of the serological predictors of response to antiviral therapy (**Abdoul et al., 2008**) as SVR to Peg-IFN α /RBV was associated with a decrease in serum AFP (**Kasztelan et al., 2003**).

In our study, **IP10** level was 260.7 ± 165.2 pg/ml in the treated group. High serum IP-10 levels were associated with a failure to achieve SVR. IP-10 levels were significantly lower in patients who achieved SVR pg/ml than in Non-SVR achievers (159.9 ± 55.2 vs. 395 ± 167.9 pg/ml; $p = 0.001$). While there was no statistically significant difference between patients with significant and non-significant fibrosis regarding IP -10 ($P = 0.5$). This was matched with a study done in **Cairo University, Egypt** which included 80 treatment naïve HCV patients. Pretreatment serum IP-10 levels were significantly lower in patients who achieved SVR than in non-responders and they concluded that low pretreatment serum IP-10 is a favorable predictor of response to antiviral HCV therapy in Egyptian patients (**Omran et al., 2014**).

Also our results were matched with another study done on 104 Japanese, genotype 1 CHC individuals treated with PEG-IFN/RBV and 45 treated with PEG-IFN/RBV/telaprevir, and evaluated the impact of pretreatment serum IP-10 concentrations on their virological responses. They concluded that pretreatment serum IP-10 concentrations are associated with treatment efficacy in PEG-IFN/RBV and with early viral kinetics of HCV in PEG-IFN/RBV/telaprevir therapy (**Matsuura et al., 2014**).

Diago et al. (2006) reported the association of serum IP-10 levels with SVR to (PEG-IFN/RBV) therapy in patients with genotype 1 chronic HCV. They showed that the levels of IP-10 were lower in SVR patients. Noteworthy, after successful antiviral therapy, serum IP-10 concentrations decreased to levels lower than baseline, whereas they were unchanged in non-responders, suggesting that HCV itself may be responsible for elevated serum IP-10

concentrations found in HCV-infected patients, and thus, pretreatment serum IP-10 is an independent predictive factor of SVR in HCV patients infected by genotype 1.

On contrary; **El Razikyet al (2015)** found that the pretreatment serum IP-10 levels were not significantly different in relation to different grades of necro-inflammatory activity and fibrosis stages. Verifying the predictive value of pretreatment serum IP-10 levels, their study did not find a significant relation to response at week 12, 24, 48, and 72. Concentrations lower than 594.1 pg/mL had a positive predictive value of 86.8% in their study population. Also, **Reiberger et al in 2008** did not find a clear association between IP-10 levels before or during treatment and SVR. Also, **Yoneda et al in 2011** did not confirm the association between a low baseline IP-10 level and SVR. Other reports confirmed these findings; no difference was seen in baseline IP-10 levels between CHC patients with or without RVR (**Falconer et al., 2010**) or with or without SVR (**Wan et al., 2009**).

In our study, we found that a cutoff point for IP10 more than 216.6 had a sensitivity of 90% and specificity 87.5% and PPV 84.4% and NPV 92.1% ($P=0.001$; C.I 95%= 0.900-1.000).

IP-10 has a chemotactic function on different cell types following binding to its receptor. IP-10 recruits T lymphocyte subsets expressing the CXCR3 receptor, including activated T lymphocytes of the T helper type 1 phenotype. Also, cytotoxic T cells and NK cells express CXCR3 and are targeted by the IP-10 chemotactic effect (**Zeremski et al., 2008**).

IP-10 is upregulated in the liver cells infected by HCV since strong IP-10 staining was found in the cytoplasm of the hepatocytes but not in other liver cells. It has been suggested that HCV itself may be responsible for elevated serum IP-10 levels found in HCV infected patients. HCV proteins such as NS5A and core, alone or in combination with pro-inflammatory cytokines, can induce IP-10 gene expression and secretion in human hepatocyte derived cells, leading to the accumulation of CXCR3 expressing T lymphocytes in the liver. IP-10 expression may also provide important retention signals, resulting in the observed accumulation of T lymphocytes (**Apolinario et al., 2005**).

Thus, the mechanism by which IP-10 induces treatment failure may be explained by its role in the recruitment of effector Th1 lymphocytes into the liver cells of chronic HCV patients and its potential in contributing to the host immune response against the virus as well as to the disease progression. Also, IP-10 modulates the efficacy of IFN- α plus ribavirin therapy by enhancing the expression of the HCV NS5A protein which induces IL-8 (CXC chemokine) secretion. IL-8 was also found to be associated with the inhibition of the antiviral effects of IFN- α (**Zeremski et al., 2008**).

We can assume that HCV and IP-10 have synergistic effects towards each other; HCV induces IP-10 secretion by the hepatocytes, and IP-10 stimulates viral replication and inhibits the antiviral effects of IFN- α (**Apolinario et al., 2005**).

It was found that circulating CXCL10 is indeed processed into the shorter form and turns it into a CXCR3 antagonist. It was demonstrated that the short, antagonist form of CXCL10 predominates in the plasma of chronically infected patients who are destined to fail anti-HCV therapy and thus is responsible for the lack of desired antiviral effects of IP-10 and could play an important role in pathology (**Casrouge et al., 2011**).

Thus, IP-10 is a strong negative predictor of response to Peg-IFN/Ribavirin therapy in HCV-4 mono-infected patients. This highlights the need to consider this factor in the individualization of treatment, and augments the level of predictiveness of IL28B polymorphisms for final treatment outcome (**Charles and Dustin, 2011**). As IP-10 may play a role in the mechanism of failure of antiviral therapy, interventions neutralizing endogenous IP-10 or blocking the function of its receptor, CXCR3, may provide new strategies to improve the treatment outcome of these difficult-to-cure patients (**Charles and Dustin, 2011**).

Our study has limitations as it was conducted on a small number of patients and this population contained small percentage of patients with significant viremia and significant fibrosis. Data may not be generalizable to other study populations. Also, we did not analyze the IL-28 B polymorphism in our cohort which may interact with our results.

In conclusion, we found a strong association between low pretreatment IP-10 levels and SVR in HCV infected Egyptian patients. Pretreatment serum IP-10 level should be included as one of the predictor of response to therapy.

Attempts to neutralize IP-10 or block IP-10 receptor should be considered as additional strategies to further improve the outcome of the antiviral therapy.

Further studies with larger samples are needed to determine the association of IP-10 and HCV treatment response especially with DAA therapy.

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RESEARCH ARTICLE

Translational Research; From Bench to Bedside and Community–Brief Review.

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Abstract

Science does not flourish in vacuum. There is a connection between science and its application. Scientific discoveries need to be translated into practical applications in order to improve health and well-being of mankind. Many discoveries typically began at “the bench” with basic research in which scientists studied disease at a molecular or cellular level then progressed to the clinical level, or the patient's “bedside. It is important that basic research and applied research need to go hand in hand to provide complete answers for many research questions. Translational research in nursing is an emerging area of interest for many. Translational research conveys different meaning to different people, but it seems it is important to everyone. Hence this review is intended to highlight the concept of translational research, its need and applicability in the field of nursing.

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Introduction:-

Translational research has become an extremely popular buzzword in the world of biomedical research lately and was designed for the medical world. It emerged in response to concern over the long time lag between scientific discoveries and changes in treatments, practices, and health policies that incorporate the new discoveries(1).

Translational research is broader than the traditional term “applied research.” Applied research is any research that may possibly be useful for enhancing health or well-being. It does not necessarily have to have any effort connected with it to take the research to a practical level. For example, an applied research study might analyse longitudinal data that tracks participants’ health and social relationships. But in translational research, the same researchers would partner with a community and ask for ideas about how their findings might apply there. Together, they would come up with an intervention plan that would also include scientific evaluation of its effectiveness (2).

Definition:-

Research that translates scientific discoveries and advances from the bench or laboratory into a clinically germane application (1).

“Translational research in nursing is scientific investigations of methods, interventions and variables that influence adoption of evidenced based practice by individuals and organizations to improve clinical and organizational decision making in health care” (3).

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Need for translational Research:-**Knowledge - Practice Gap:-**

The 'knowledge driven model' was used within the medical field assumed that basic research would progress to applied research and would eventually lead to development stages, such as new medicine or technology and then application in the realm of practice. Example it took 200 years for convincing cure for scurvy (4).

Cultural Differences:-

Early knowledge-driven and problem solving models reduced interaction to one-way due to cultural and value differences between academicians and decision makers (4).

Research and Practice Gap:-

Expanding research literature being difficult to access in a timely fashion, gaps in the current understanding of health and disease processes, and the difficulty of interpreting the results of research publications, there remains also the tension of conjoining a knowledge base from a scientific research community with that of clinical communities (2).

Evidence Based Movement:-

Focused the two-way nature of knowledge flows and the need for active engagement, rather than passive diffusion. Rather than assessing knowledge flow solely by the efficiency of outcomes, the conceptual focus shifted on the process of interaction and collaboration. Also, while EBM reconciled the volitional variations involved in the problem selection and analysis around the logic of efficiency, the emerging conceptualizations of knowledge exchange considered and reconciled cultural differences in a symmetrical and reciprocal interaction of researchers and practitioners (5,6).

Emergence of the research translation Models:-

Emphasize interactions between researchers and decision-makers in developing and implementing research in practice (2).

Components of Translational Research:-

There are two areas of translation.

- Type One Translation (T1): It is the process of applying discoveries generated during research in the laboratory, and in preclinical studies, to the development of trials and studies in humans. Usually called "bench to bedside," (i.e., services, programs, practices and products etc.).
- Type two Translation (T2): this area concerns research aimed at enhancing the adoption of best practices in the community. Cost-effectiveness of prevention and treatment strategies is also an important part of translational science. (e.g., health care settings, community-based organizations, schools etc.) (2,5).

Translational Research - A Collaborative Approach:-

Enhancing team science to improve its ability to work across disciplines and professions is the major hallmark of 21st century science and is now commonly seen as the key for advancing translational research. Team science may make use of a range of modalities:

- ★ Multidisciplinary: Coordination of research among scientists of different backgrounds and disciplines.
- ★ Interdisciplinary: Cooperative effort of scientists on issues that fall between disciplinary boundaries.
- ★ Transdisciplinary: Collaborative projects where information exchange, the modification of scientific approaches, and the integration of scientists and approaches from different disciplines advances research toward a common scientific goal (7,8).

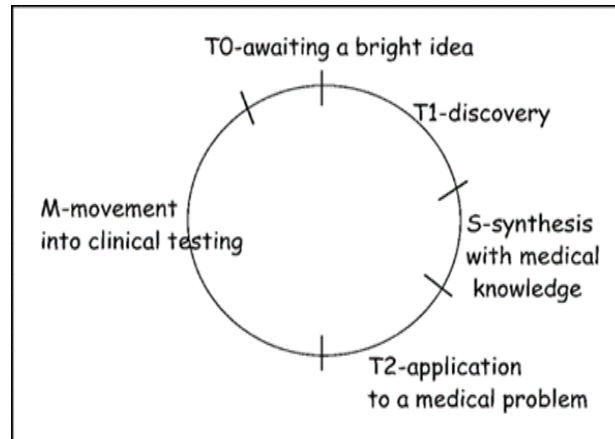
Translational Research Cycle:-

"Translational research cycle", includes several identifiable stages and clearly defined checkpoints. Each phase has its unique features. For example, the resting phase (T0) requires the generation of a great idea and the formulation of that idea into a testable hypothesis.

The testing phase (T1) is when the great idea is subjected to interrogation through scientific inquiry. This hypothesis testing requires the assembly of a research team that includes students, postdoctoral fellows, and more established scientists. The principal investigator or team leader must acquire funding for the project and show that progress is

being made. With a combination of luck, skill, and tenacity, a discovery will occur that may become the basis for translation.

The synthesis phase (S) requires the ability to identify the potential importance of a discovery to a clinical problem. This is often dependent on mechanisms to place fundamental information in front of people who understand the scientific basis of medicine and are familiar with medical problems.



Movement into the application phase (T2) requires the ability of researchers to determine the importance of a discovery to human biology. Here, the team of investigators must have access to human tissue and body fluids, predictive animal models, as well as pathologists, pharmacologists, surgeons, and biostatisticians.

The movement phase (M) is where preclinical studies enter the clinic. Now the research team must expand to include practicing physicians, nurses, research coordinators, toxicologists, informaticians, regulatory experts, lawyers, and a variety of other seemingly unrelated species. Ironically, as the discovery moves closer to human application, the science tends to get less sophisticated, yet the funding gets more difficult (9).

Translational Research in Nursing:-

In recent decades, the initiatives to develop individual, collaborative, or multicentric nursing research have multiplied throughout the world. The objective of such initiative has been to improve the quality of nursing care given, offering greater security to patients and influencing more effective health care policies. Research based on evidence, clinical research, systematic literature reviews, convergent care research, phenomenological studies, and social representations all share the same objective: to respond to professional practice questions. However, despite all the knowledge such efforts have produced, there is a vacuum among the studies and the use of the results from research carried out in public or hospital health care services. Translational research, with its mantra "bench to bedside and back," emerged as a top priority in the field of nursing has arisen in an attempt to fill in the vacuum and approximate the researcher to fields of practice (10).

Clinical and translational Research Center Nursing: nurses skilfully combine both the clinical and research aspects of patient care. With patient safety a priority, maintaining the highest standards of research conduct, utilizing expert clinical judgment, critical judgment, critical thinking skills, and a patient-centred approach are the key components which are emphasised (8).

Nursing Leadership in Translational Research:-

Nurses should assume leadership for the majority of translational science projects in hospitals. Nurses need to identify optimal mechanisms for enriching the care provided in the more remote communities. Also, nursing has had long-established historical efforts at studying broad range outcomes in health care, which positions them very well to lead, including establishing institutes focusing on translation within hospitals and health systems. Nursing homes and other long term care services and home care should be very high on the priority list. Given nursing's commitments to address health disparities, they must assume more leadership responsibilities for translational research that illuminates how the communities at greatest risk can benefit from the very expensive work they are all doing. It is not good enough to continue to generate studies that show how one can change health and health care in

communities that have access to high quality health care as well as adequate housing, nutritionally adequate diets, and good health education. Nurses should be devoting more of their efforts to the census tracts in inner city and rural areas (4).

Future of Translational Research in Nursing:-

Nurses must engage immediately and assertively in setting an agenda for translation of their research to practice in health care systems and to public. In order to achieve this, nurses need to focus on three sectors: traditional, mainstream health services including complementary and alternative health services and community health services, and the general public.

- ✦ Nurses must be visible and vocal on focusing on the areas in which nurses have expertise. Nurses should expect that they exist to embrace, advance, and translate nursing science, given that nurses have been contributing to clinical sciences
- ✦ Nurses should make their translational science models visible, and their efforts should be supported by federal agencies.
- ✦ Contemporary communication Medias have a great reach and nurses should use them more effectively to promote health and health-care. Nurses need to author articles and books for the public and contribute to internet sources designed to translate research findings to the public for their benefit.
- ✦ If nurses are truly committed to translation of research in practice, they will resolve to educate clinicians who are capable of the translational work necessary to truly achieve this goal (11).

Conclusion:-

Although the science of translating research into practice is fairly new, there is some guiding evidence of what implementation interventions to use in promoting patient safety practices. However, there is no magic bullet for translating what is known from research into practice. To move evidence-based interventions into practice, several strategies may be needed. Nurse scientists must reevaluate how proposed research fits within and contributes not only to the development of new knowledge, but to the translation of that knowledge to the care of diverse communities and populations.

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RESEARCH ARTICLE

TOXICITY OF SELECTED BOTANICALS TO THE COTTON LEAF WORM, *SPODOPTERA LITTORALIS* (BOISD.)

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Key words:-

Spodoptera littoralis, lavender oil, mint oil, *Catharanthus* extract, *Artemisia* extract, toxicity, feeding deterency.

Abstract

The sensitivity of *Spodoptera littoralis* 2nd and 4th instar larvae towards the essential oils (mint and lavender oils) and aqueous plant extracts (*Artemisia herba-alba* and *Catharanthus roseus*) were investigated under laboratory conditions and the effect of sublethal concentrations on the feeding deterrence were evaluated on the insect. Results revealed that 2nd and 4th instar larvae of *S. littoralis* were more susceptible to lavender oil than mint oil as it has higher LC₅₀ values. In addition, the results showed that the mean feeding deterrence (FDI%) of essential oils and botanicals extracts was concentration-dependent. Therefore, these botanicals could be important as eco-friendly accessible pest control alternatives against *S. littoralis* and other closely related species.

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Introduction:-

The Egyptian cotton leafworm, *Spodoptera littoralis* (Boisd.) (Lepidoptera: Noctuidae) is one of the most destructive pests of several crops such as cotton, corn, peanut, clover, vegetables and various fruits in Egypt as well as in Mediterranean and Middle East countries (El-Sinayet *et al.* 2008 and El-Zoghby *et al.* 2011). The continuous and unwise use of insecticides to control agricultural pests usually lead to development of resistance, adverse effects on beneficial insects and residues in foods (Rizket *et al.* 2010 and Ehab 2012). The essential oils and other plant extracts, as a new class of natural products for controlling insect pests environmentally friendly have begun to play an increasing prominent role as alternatives to synthetic insecticides (El-Sinayet *et al.* 2008; Tripathi *et al.* 2009 and Ragaei and Sabry 2011). The selected essential oils and the botanical extracts used in this study are among those compounds under investigation as potential biopesticides.

Materials and Methods:-

Insect rearing:-

The colony of cotton leafworm, *Spodoptera littoralis* (Boisd.), was obtained from the division of the cotton leafworm, Plant Protection Research Institute, Dokki, Egypt. Larval stages were reared on castor bean leaves at 27 ± 2°C and 65 ± 5% R.H. and photoperiod of 16:8 hr (L:D) as described by El-Dafrawiet *et al.* 1964.

Commercial essential oils:-

Lavender oil and mint oil (containing carvone, menthol, menthone, sinod, kadenin and limonene as major constituents) were obtained from El Captain Company, Cairo, Egypt.

Preparations of aqueous botanical extracts:-

Healthy plants *Catharanthus roseus* (family: Apocynaceae) and *Artemesia herba-alba* (family: Asteraceae) were collected in the morning hours from the medicinal plant garden, Faculty of Pharmacy, Cairo university and after separating the leaves to test their insecticidal properties against *S. littoralis*, they were washed with distilled water and left to dry in the shade. Finally, they were transferred to an oven (70°C) for 24 hour and the dried leaves were blendered to make fine powder. Fifty gram of dried powder were stirred with 1 Liter distilled water for 1 hour and incubated for 48 hour at 4°C and then stirred for additional 1 h and filtered twice through whatman No.1 filter paper. The volume was made up to 500 ml and it was considered as stock solution of the extract. This stock extract was maintained in a refrigerator until being used and the diluted concentration of the extract were made up.

Insect Bioassay:-

Leaf-dip method as described by **Tabashnik et al. 1991** was followed using castor leaves. Fresh castor leaves, of almost the same size, were dipped in different concentrations (0.1, 0.5, 0.75, 1, 1.5 and 2%) in case of treatment each of *S. littoralis* 2nd and 4th instars with essential oils but dipped in (1, 5, 10, 15, 20 and 25%) concentrations for botanical extracts treatments. The dipping lasted for ca. 5-10 seconds and left to dry in air from excess solution. The treated leaves were transferred singly in plastic cups where 10 individuals of 2nd and 4th instar larvae were allowed to feed on these treated leaves. Treated leaves were offered to larvae for 48 hrs. Three replicates of each concentration were performed. The untreated castor bean leaves (control) were dipped in distilled water for the same period of time as treated ones. Insect mortality were recorded daily starting after 24h from treatment. The experiment was conducted at laboratory temperature of 27 ± 2 °C, $70 \pm 5\%$ R.H. with photoperiod of 16:8 (L: D) hr.

The mortality % was corrected according to Abbott's formula (**Abbott, 1925**) as follows:

$$\text{Corrected mortality \%} = \frac{\text{Observed mortality \%} - \text{control mortality \%}}{100 - \text{control mortality \%}} \times 100$$

Probit analysis was determined to calculate the median lethal concentration values (LC₅₀) and related parameters, according to **Finney (1971)**.

Feeding deterrent activity (non-choice method):-

Feeding deterrent activity of the botanical solutions was assayed against *S. littoralis* 2nd and 4th instar larvae using a leaf-dip bioassay in no-choice situations. For this purpose the concentrations (LC₂₅ and LC₅₀) of essential oils and botanical extracts were prepared for each instar. The Leaf discs of (Ø= 8 cm) were impregnated for 5-10 seconds in each concentrations and the control leaf discs were impregnated in distilled water for the same time. In each plastic Petri dish (1.5 cm x 9 cm) wet filter paper was placed to avoid early drying of the leaf discs and ten larvae per replicate of either 2nd or 4th instar were introduced. Progressive consumption of leaf weight by the larvae after 24 hrs was recorded in control and treated discs. Amount of leaf eaten by the larva in essential oils and botanical extracts treatments was corrected from control. Three replicates were maintained for each treatment with 10 larvae per replicate (total, n= 30). Feeding deterrent activity was assessed by calculating the Feeding Deterrence Index by the formula of **Saleh et al. 1986**:

Feeding Deterrence Index (FDI);

$$= \left(1 - \frac{\text{Percentage of treated consumed leaf}}{\text{Percentage of untreated consumed leaf}} \right) \times 100$$

Statistical Analyses:-

Using the computed percentage of mortalities versus corresponding concentrations, Probit analysis was adopted according to **Finney (1971)** using a software computer program (**SAS, 2002**). This yields determination of the toxicity indices (LC₂₅ and LC₅₀) as well as the related parameters (95% confidence intervals, slope and Chi-square, χ^2) for established toxicity regression lines.

Obtained data were statistically analyzed using one-way analysis of variance (**ANOVA**) supported by Duncan's multiple range test (**Duncan, 1955**) running on **Costat statistical software, 1990**. Means were compared using L.S.D. (5% significance level).

Results and Discussion:-

Toxicity of tested botanicals to *S. littoralis*:-

Table (1 & 2) revealed that the LC₂₅ and LC₅₀ values were 0.179 and 0.379 %, respectively for lavender compared with 0.296 and 0.417 %, respectively for mint against 2nd instar larvae. The LC₂₅ and LC₅₀ values were 0.293 and 0.504 %, respectively for lavender compared with 0.39 and 0.839 %, respectively for mint against 4th instar larvae. The essential oils act at multiple levels in the insects, so the possibility of generating resistance is little probable (Gutierrez *et al.* 2009). The main components of lavender oil were linalool acetate and linalool while the mint oil consisted of menthol in high percentage, menthone (*iso*), β -pinene, and menthyl acetate (Karamaouna *et al.* 2013).

In addition, the LC₂₅ and LC₅₀ values were 2.297 and 5.016 %, respectively for *Catharanthus* compared with 2.633 and 6.527 %, respectively for *Artemisia* extract against 2nd instar larvae. While the LC₂₅ and LC₅₀ values were 3.456 and 6.56 %, respectively for *Catharanthus* compared with 3.818 and 8.332 %, respectively for *Artemisia* against 4th instar larvae. The slope values indicated that the insect population was relatively heterogeneous in their susceptibility toward tested essential oils and botanical extracts by leaf-dip method. Our results showed LC₅₀ values, the range of toxicity was in the decreasing order of lavender > mint > *Catharanthus* > *Artemisia* against both *S. littoralis* 2nd and 4th instars.

The *Artemisia* sp. belonging to the important family Asteraceae (Compositae) has known to possess several important biological properties, such as insecticidal activity (Saleh 1984). Hifnawy *et al.* (2001) reported larvicidal activity of *A. herba-alba* against cotton leafworm, *S. littoralis* (Biosd.) larvae.

Among the plants found to contain insecticidal or growth regulatory effects of insects, plants from the genus *Ageratum* and *Artemisia* were reported to have insecticidal activity (Anjoo and Ajay 2008). *Artemisia herba-alba*, is rich in terpenoids like monoterpene hydrocarbons (Behtari *et al.* 2012), oxygenated monoterpenes (Hudaib and Aburjai 2006) and sesquiterpenes (Laid *et al.* 2008 and Paolini *et al.* 2010). Ramya *et al.* (2008) and Alaguchamy and Jayakumararaj (2015) studied the effect of leaf aqueous extract of *C. roseus* and they recommended that it can potentially be used as ecofriendly bio-pesticide to control the devastating damage caused by larvae of *Helicoverpa armigera*.

Kumar and Yadav (2013) showed that screened phytochemical constituents of *Catharanthus roseus* (family: Apocynaceae) possesses carbohydrates, anthraquinone glycosides, flavanoids, saponins, and alkaloids. Also, the work on the isolation of a possible insect growth regulator (IGR) from *C. roseus* is in progress (Summarwar and Pandey 2015).

Table (1): Toxicity indices (LC₂₅ and LC₅₀) of the essential oils (Mint and Lavender) and the botanical extracts (*Artemisia herba-alba* and *Catharanthus roseus*) against *Spodoptera littoralis* 2nd instar larvae.

Phytochemicals	LC ₂₅ (Conc. %) 95% confidence interval	LC ₅₀ (Conc. %) 95% confidence interval	Slope \pm SE	χ^2
Mint	0.296 (0.12 - 0.52)	0.417 (0.34 - 0.67)	2.44 \pm 0.31	4.17
Lavender	0.179 (0.12 - 0.37)	0.379 (0.26 - 0.77)	2.41 \pm 0.35	2.13
<i>Artemisia</i>	2.633 (1.96 - 3.27)	6.527 (4.81 - 7.96)	1.71 \pm 0.24	4.83
<i>Catharanthus</i>	2.297 (1.11 - 3.27)	5.016 (4.81 - 7.96)	1.98 \pm 0.26	3.42

* LC₂₅ and LC₅₀ values are significant (p < 0.05) whenever confidence intervals do not overlap.

Table (2): Toxicity indices (LC₂₅ and LC₅₀) of the essential oils (Mint and Lavender) and the botanical extracts (*Artemisia herba-alba* and *Catharanthus roseus*) against *Spodoptera littoralis* 4th instar larvae.

Phytochemicals	LC ₂₅ (Conc. %) 95% confidence interval	LC ₅₀ (Conc. %) 95% confidence interval	Slope \pm SE	χ^2
Mint	0.390 (0.13 - 0.49)	0.839 (0.53 - 0.99)	2.96 \pm 0.30	3.42
Lavender	0.293 (0.19 - 0.58)	0.504 (0.33 - 0.78)	3.02 \pm 0.35	3.67
<i>Artemisia</i>	3.818 (1.75 - 6.29)	8.332 (6.00 - 11.17)	1.12 \pm 0.24	1.52
<i>Catharanthus</i>	3.456 (1.97 - 5.78)	6.56 (4.40 - 7.71)	1.15 \pm 0.23	3.41

* LC₂₅ and LC₅₀ values are significant (p < 0.05) whenever confidence intervals do not overlap.

Feeding deterrence activity:-

Data presented in table (3) showed that the mean feeding deterrence activity (based on feeding deterrence index values) was significantly different (P < 0.05) between lavender and mint oil treatments on 2nd instar larvae at both

LC₂₅ and LC₅₀ where mean feeding deterrent values at LC₂₅ were higher in case of mint oil (68.369 %) than that in case of lavender oil (65.833 %) for four days after treatment. while for the same instar at LC₅₀ levels, the mean feeding deterrent values were higher in case of lavender oil (79.151 %) compared to that in case of mint oil (75.272%). In connection with the 4th instar, also the mean feeding deterrent values were significantly different between lavender and mint oil treatments either at LC₂₅ or LC₅₀ where mean feeding deterrent values at LC₂₅ were higher in case of mint oil (63.561%) than that in case of lavender oil (60.408 %) for four days after treatment. Also, at LC₅₀ levels, the mean feeding deterrent values were still higher in case of mint oil (73.413 %) compared to lavender oil (70.837%) (table 3).

Depending on the data, the mint oil exhibited relatively more feeding deterrent effect than lavender oil treatments. The higher feeding deterrence index normally indicates decreased rate of feeding. Also, the *Mentha pulegium* oil significantly inhibits the feeding of fall armyworm, *Spodoptera frugiperda* (Zalkowet *et al.* 1979). Any substance that reduces food consumption by an insect can be considered as antifeedant or feeding deterrent (Isman 2002).

Abd El-Galeil and Nakatani (2003) indicated that the antifeedant activity was dose-dependent in some of the isolated compounds. Elumalai *et al.* (2010) reported that all tested essential oils are showed moderate antifeedant activity against 4th instar larvae of *S. litura*; however, the highest antifeedant activity was observed in the essential oils of *Cuminum cyminu*, *Mentha pipertia*, *Rosmarinus officinalis*, *Thymus vulgaris*. *Coriandrum sativum* exhibited (100%) complete antifeedant activity at 6 mg/cm².

Table (3): Percentage feeding deterrent indices (mean \pm SE) of *S. littoralis* 2nd and 4th instars larvae treated with LC₂₅ and LC₅₀ of essential oils (Mint and Lavender).

Treatment	LC ₂₅		LC ₅₀	
	2 nd instar	4 th instar	2 nd instar	4 th instar
Mint	68.369 \pm 4.195 ^a	63.561 \pm 3.895 ^a	75.272 \pm 4.613 ^b	73.413 \pm 4.504 ^a
Lavender	65.833 \pm 4.116 ^b	60.408 \pm 3.706 ^b	79.151 \pm 4.850 ^a	70.837 \pm 4.342 ^b
L.S.D.5%	2.173	2.199	2.217	2.249

*Within the same column, means followed by the same letter are not significantly different (P > 0.05).

While table (4) showed that the mean feeding deterrent activity (based on antifeedant index values) was significantly different (P < 0.05) between *Artemisia* and *Catharanthus* treatments on 2nd instar larvae at both LC₂₅ and LC₅₀ where mean antifeedant values at LC₂₅ were higher in case of *Catharanthus* (63.503 %) than that in case of *Artemisia* (56.46 %) for four days after treatment. Also, for the same instar at LC₅₀ levels, the mean antifeedant values were still higher in case of *Catharanthus* (70.903 %) compared to *Artemisia* (63.617 %).

In connection with the 4th instar, also the mean feeding deterrence values were significant different between *Catharanthus* and *Artemisia* treatments either at LC₂₅ or LC₅₀ where mean antifeedant values at LC₂₅ were higher in case of *Catharanthus* (57.65%) than that in case of *Artemisia* (54.342%) for four days after treatment. Also, at LC₅₀ levels, the mean feeding deterrent values were still higher in case of *Catharanthus* (67.75 %) compared to *Artemisia* (63.871 %) (table 4).

It is obvious from data that the *Catharanthus* extract exhibited more feeding deterrent effect than *Artemisia* extract. In addition, the data indicates that feeding deterrence of both botanical extracts has increasing trend till the 4th day after treatments. Also, it is interesting to notice that the feeding deterrent activity of both botanical extracts was higher in 2nd instar than 4th instar larvae.

The extracts of *Artemisia monosperma*, *Calotropia procera* and *Tagetes patula* were the powerful antifeeding effect against *S. littoralis* larvae (Ahmed 1985). In general, the antifeeding effect of plant extracts depend mainly on insect species, however, the plant structure-activity relationship associated with its components on insect feeding is complex and no clear trends emerge (Bruno *et al.* 2002).

Erturk (2006) reported that the extracts derived from different plants *Artemisia absinthum*, *Aesculus hippocastanum*, *Viscum album*, *Sambucus nigra*, *Buxus sempervirens*, *Diospyros kaki*, *Alnus glutinosagoertn*, *Origanum vulgare*, *Hypericum androsaemum* and *Ocimum basilicum* had antifeeding effect against the 3rd – 4th instar larvae of the *Thaumetopoea solitaria* (Lepidoptera).

While **Summarwar and Pandey (2015)** observed that at 5% of leaf extract of *C. roseus* the percent feeding of *S. litura* 4th instar larvae was reduced to 47.77 compared to 82.47 % in control. Also, the antifeedant activity caused a reduction in food consumption and chronic toxicity leading to delayed growth, development and increased mortality (**Vattikonda et al. 2015**).

Table (4): Percentage feeding deterrent indices (mean \pm SE) of *S. littoralis* 2nd and 4th instars larvae treated with LC₂₅ and LC₅₀ of botanical extracts (*Artemisia herba-alba* and *Catharanthus roseus*).

Treatment	LC ₂₅		LC ₅₀	
	2 nd instar	4 th instar	2 nd instar	4 th instar
<i>Artemisia</i>	56.460 \pm 3.462 ^b	54.342 \pm 3.332 ^b	63.617 \pm 3.902 ^b	63.871 \pm 3.920 ^b
<i>Catharanthus</i>	63.503 \pm 3.896 ^a	57.650 \pm 3.535 ^a	70.903 \pm 4.349 ^a	67.750 \pm 4.157 ^a
L.S.D.5%	2.203	2.162	2.203	2.361

*Within the same column, means followed by the same letter are not significantly different (P > 0.05).

Conclusion:-

Our results confirmed that the tested botanicals either oils or extracts resulted in increased mortality, reduced food consumption via their feeding deterrent effect and exert a adverse impact on *S. littoralis* growth and development. These effects were dose-dependent. The findings may be helpful and effective for studying the efficacy of such botanicals as a part of the Integrated Pest Management (IPM) against this pest and closely related ones.

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RESEARCH ARTICLE

CHARACTERIZATION AND ANTIMICROBIAL SUSCEPTIBILITY PATTERN OF CLINICAL ISOLATES OF ENTEROCOCCI AT A TERTIARY CARE HOSPITAL IN BHAVNAGAR, GUJARAT.

Dr. Dhvani D. Parmar.

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Enterococci, Catalase test, Bile esculin agar, Sugar fermentation test.

Abstract

Enterococcal infection is a life threatening systemic infection prevalent in developing countries like India and is a major public health problem. Enterococci are important nosocomial pathogen of human disease; second most common cause of nosocomial urinary tract and wound infections; third most common cause of nosocomial bacteremias. Because of their resistance to different antimicrobial agents these bacteria are often involved in serious super-infections among patients receiving broad-spectrum anti-microbial therapy. Correct diagnosis is crucial in proper management of Enterococcal infection, so laboratory tests are essential to establish identification of Enterococcal species. The study was conducted to isolate and characterize *Enterococcus* species from different clinical samples and to perform antimicrobial susceptibility testing in all age groups and both genders. Out of 200 Enterococcal species isolated, 164 (82%) were *E. faecalis* and 34 (17%) were *E. faecium*; majority were isolated from urine (60%) and blood (35.5%); they showed more resistance to penicillin (92%), less resistance to vancomycin (1%) and no resistance to linezolid (0%).

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Introduction:-

Enterococci are Gram-positive cocci which are normal residents of the gastrointestinal tract, biliary tract and, in lower numbers, of the vagina and male urethra, in saliva. They are becoming increasingly important nosocomial pathogen of human disease, largely because of their resistance to antimicrobial agents to which other streptococci are generally susceptible.

Enterococci are the second most common cause of nosocomial urinary tract and wound infections and the third most common cause of nosocomial bacteremias. *Enterococcus* species also cause endocarditis, intraabdominal and pelvic infections, soft-tissue infections, neonatal sepsis and rarely meningitis¹.

Because of their resistance to penicillin and cephalosporin group, the acquisition of high-level resistance to aminoglycosides, and now the emergence of vancomycin resistance, these bacteria are often involved in serious super-infections among patients receiving broad-spectrum anti-microbial chemotherapy. Newer agents such as linezolid and daptomycin may also be effective; other agents such as chloramphenicol may be used when multidrug-resistant strains are encountered. For urinary tract isolates, nitrofurantoin, tetracycline, or quinolone may be effective^{1,2}.

In mid-1980s, *Enterococci* were distinguished from *streptococci* and related taxa by their ability to grow at 10°C and 45°C, growth in the presence of 6.5% NaCl, growth at pH 9.6, grow in 0.1% methylene blue milk, ability to hydrolyze esculin in the presence of 40% bile, and production of pyrrolidonyl arylamidase (PYR)^{1,3}.

Enterococcus faecalis is the most common isolate found in human clinical specimens, followed by *Enterococcus faecium*. Other Enterococcal species, including *E. avium*, *E. cassiliflavus*, *E. cecorum*, *E. durans*, *E. gallinarum*, *E. hirae*, *E. raffinosus*, *E. malodoratus*, *E. dispar* and *E. mundtii* are infrequently isolated from human infections.

Risk factors for the development of bacteremias include advanced age, immunosuppression, underlying disease and conditions (e.g.; prematurity, diabetes, malignancy, congestive heart failure, renal insufficiency, deep-seated infections), prior gastrointestinal, genitourinary, or respiratory tract instrumentation, long-term hospitalization, and the use of broad-spectrum antibiotics having little or no antienterococcal activity (e.g., cephalosporin)¹.

Isolation measures to prevent person-to-person transmission according to CDC's Guidelines includes⁴:

- Hand washing with antimicrobial soaps and hand rubs with alcohol-based solutions,
- Wear gloves, Wear a gown,
- Dedicate the use of items such as stethoscopes, blood pressure cuffs, and thermometers to a single patient.

Aims and Objectives:-

- ▶ To isolate Enterococci from different clinical samples and to speciate Enterococci by different tests.
- ▶ To determine antibiotic susceptibility pattern of *Enterococcus* species for proper treatment of patient by specific antimicrobial drugs.

Materials and Methods:-

The study was conducted from May 2014 to August 2015 in Sir Takhtsinhji General Hospital, Bhavnagar, Gujarat, India.

In this study 200 Enterococci species isolated from different clinical samples- Urine, blood culture, pus, catheter tips and body fluids collected in sterile container from all patients attending to Hospital. Samples were collected from patients having risk factors like advanced age, immunosuppression, prematurity, diabetes, malignancy, congestive heart failure, renal insufficiency, deep-seated infections, prior gastrointestinal, genitourinary, or respiratory tract instrumentation, long-term hospitalization, and long- term use of broad-spectrum antibiotics.

Identification and Characterization:-

Urine samples were inoculated on CLED agar, other mentioned samples were inoculated on Blood agar. After overnight incubation at 37°C colonies were identified. Further identification was done by inoculation on Bile-Esculin agar, PT agar, by doing Gram stain, Catalase test, Growth in 6.5% NaCl and Carbohydrate Fermentation Test using six sugars- Mannitol, Sorbitol, Sucrose, Raffinose, Lactose, Arabinose.

Antimicrobial Susceptibility Testing:-

Antibiotic susceptibility testing of the isolates was performed on Muller Hinton agar by Kirby-Bauer disc diffusion method⁵. The antibiotic concentration of disc used and zone size interpretation was accordance to Clinical Laboratory Standards Institute (CLSI)⁶. These antimicrobial drugs are Ampicillin, Ampicillin-Sulbactam, Penicillin, Piperacillin-Tazobactam, Linezolid, Ciprofloxacin, Tetracycline, Roxithromycin, Vancomycin, High level Gentamycin (120µg), High level Streptomycin (300µg), and Nitrofurantoin. Commercially available six mm disks (Himedia Laboratories, Mumbai) were used.

Quality Controls:-

Standard operative procedures were followed during processing of each sample. *E.faecalis* VRE ATCC 51299 and *E.faecalis* VSE ATCC 29212 were used as a reference strains^{7,8}.

Results:-

Out of 1850 culture positive samples proceeded, 200 Enterococci were isolated from urine, blood, pus, catheter tip (hospital acquired) & body fluid (peritoneal fluid), suggesting 11% prevalence rate of Enterococci.

Enterococci are Gram positive oval cocci, arranged in pairs at an angle to each other; catalase negative; can grow in 6.5% NaCl. After overnight incubation small yellow or pink colonies were formed on CLED medium; small, cream or white or grayish-white, smooth entire, non-hemolytic colonies were formed on Blood agar. As Enterococci hydrolases esculin in presence of 40% bile, blackening on BEA media seen. *E.faecalis* could grow on PT agar producing black colonies, but *E.faecium* could not^{1,3,9}.

Isolated Enterococci species distribution, sample wise, age and sex wise distribution of Enterococci species shown in Table - 1, 2, 3 and 4 respectively.

Table 1:- Species of Enterococci isolated

Enterococci Species	Number	Percentage
<i>E.faecalis</i>	164	82%
<i>E.faecium</i>	34	17%
Other	2	1%

Other Enterococcal species might be *E.avium*, *E.gallinarum*, *E.cassiliflavus*, *E.durans*, *E.raffinosis* or *E.mundtii*.

Table 2:-Samples showing Enterococcal species isolation

Samples	Number	Percentage
Urine	120	60%
Blood	71	35.5%
Pus	3	1.5%
Peritoneal fluid	2	1%
Catheter tip	4	2%

Table 3:- Age-wise distribution of Enterococci Species.

Age Group	Number	Percentage
<1 year	44	22%
1-15 year	32	16%
16-30 year	28	14%
31-45 year	40	20%
46-60 year	34	17%
>60 year	22	11%

Table 4:- Sex-wise distribution of Enterococci Species.

Sex	Number	Percentage
Female	113	56.50%
Male	87	43.50%

The antimicrobial susceptibility pattern of Enterococci species has been shown in Table-5. In spite of predominance of *E.faecalis*, it was observed that *E.faecium* was more resistant than *E.faecalis*.

Table 5:-Antimicrobial drug resistance in Enterococci.

Antimicrobial Drugs	Isolates Resistant to Antimicrobial Drugs (200 species)	Percentage of Resistance
Ampicillin	106	53%
Ampicillin-Sulbactam	83	41.5%
Penicillin	184	92%
Piperacillin-Tazobactam	108	54%
Linezolid	0	0%
Ciprofloxacin	115	57.50%
Tetracycline	136	68%
Roxithromycin	125	62.50%
Vancomycin	2	1%
High level Gentamycin	85	42.50%
High level Streptomycin	118	59%
Nitrofurantoin	76	38%

Results of Sugar Fermentation Test done on Enterococci species has been shown in Table-6. Acid production occurs from sugar fermentation & yellowish color develop.

Table 6:- Sugar Fermentation by Species of Enterococci isolated.

Sugars used for fermentation test	<i>E.faecalis</i>	<i>E.faecium</i>
Mannitol	+	+
Sucrose	+	+
Sorbitol	+	-
Lactose	+	+
Raffinose ¹⁰	-	+/-
Arabinose	-	+

Discussion:-

Enterococci colonize the bowels of more than 90% of healthy humans. *E. faecalis* accounts for more than 80% of the Enterococcal isolates in clinical samples, but in recent years *E. faecium* has become more common, probably because of its greater antibiotic resistance to multiple antibiotics. For the last two decades, *Enterococcus* have been the 3rd most common cause of hospital-acquired infections (HAI) after *E.coli* and *Staphylococcus aureus* and ahead of *Pseudomonas aeruginosa*. The most common nosocomial infections caused by Enterococci are urinary tract infections (associated with instrumentation and antimicrobial administration) followed by intra-abdominal and pelvic infections. They also cause endocarditis, neonatal sepsis and rarely meningitis.

In last two decades infections due to Enterococci have increased. Now, it is among the leading causes of bacteremias. Enterococci is the second most common cause of surgical infections and the third most frequently reported cause of HAIs. Gastrointestinal colonization, length of stay in hospital, ICU stay, older age, neutropenia, hematological malignancies and transplantation are risk factors identified for Enterococcal colonization and infection¹¹.

The emergence of high-level resistance to aminoglycosides in Enterococci, particularly *E.faecalis* and *E.faecium*, seriously affected the therapeutic approach; vancomycin become a first-line drug effective against Enterococci¹².

Until the early 1980s, the susceptibility of Enterococci to ampicillin and vancomycin remained fairly predictable. Since that time, *Enterococcus* species resistant to Ampicillin and vancomycin have been reported with increasing frequency. Linezolid seemed to be an appropriate therapeutic option for VRE in our study¹³.

In present study isolated *E.faecalis* and *E.faecium* were 82% and 17%, which is comparable to studies conducted by Bose et al¹⁴ (82% and 18%) and Gupta et al¹⁵ (80% and 20%); In present study only two other species (1%) isolated which might be *E. durans*, *E. raffinosus*, *E. avium*, *E. gallinarum* comparable to study done by Grey et al¹⁶ (1.2%).

In present study, the most common clinical sample from which Enterococci were isolated was urine (60%), comparable to studies conducted by Agrawal et al¹⁷ (62.79%) and Bhatt et al¹⁸ (57%); followed by blood (35.50%), pus (1.50%), catheter tips (2%), peritoneal fluid (1%).

Present study showed that isolation of Enterococci from all age groups, ranging from newborn baby to an old age of 86 years and highest percentage (22%) of Enterococcal isolates was obtained from <1 year age group, similar to observed by Nepal et al¹⁹ (20%).

In present study female predominance (56.5%) over males seen corresponds to the Bhatt et al¹⁸ (54%). Greater vulnerability to acquire urinary tract infection and pelvic infection due to anatomic position of urethra and genital tract might have played a role for predominant female infection by this organism^{11,18}.

In present study, beta-lactam drugs, flouroquinolones, high level aminoglycosides and tetracycline were found to be highly resistant; penicillin was most resistant (92%), comparable to study done by Bhatt et al¹⁸ (97.5%). In present study linezolid has shown 0% resistance, followed by vancomycin shown 1% resistance, comparable to studies conducted by Bose et al¹⁴ (linezolid 0% resistant) and Saraswathy et al²⁰ (vancomycin 1% resistant).

Conclusion:-

- In developing country like INDIA Enterococci are important nosocomial pathogens of human disease; because of their resistance to antimicrobial agents like penicillins, cephalosporins group, the acquisition of high-level resistance to aminoglycosides and now the emergence of vancomycin resistance, these bacteria are often involved in serious super-infections among patients receiving broad-spectrum anti-microbial chemotherapy. Prompt diagnosis is essential for the patient and the community.
- Identification of Enterococci done by urine culture, blood culture, Gram stain and also by other methods like catalase test negative, its ability to grow at 10°C and 45°C, growth in the presence of 6.5% NaCl, ability to hydrolyze esculin in the presence of 40% bile and sugar fermentation test.
- Mainly two species of Enterococci were isolated, *E. faecalis* (82%) & *E. faecium* (17%).
- Out of 200 Enterococcal species 60% were isolated from urine samples, followed by 35.5% were from blood samples.
- As far as antibiotic susceptibility was concerned, resistance to penicillin (92%), suggest that resistance to β -lactam group was more among *Enterococcus* species. Resistance to vancomycin shown in 1% Enterococcal species, linezolid is more effective drug as all isolates are sensitive to it.

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RESEARCH ARTICLE

SYNTHESIS AND HERBICIDAL ACTIVITIES OF PYRAZOLE- 4-CARBOHYDRAZIDE DERIVATIVES.

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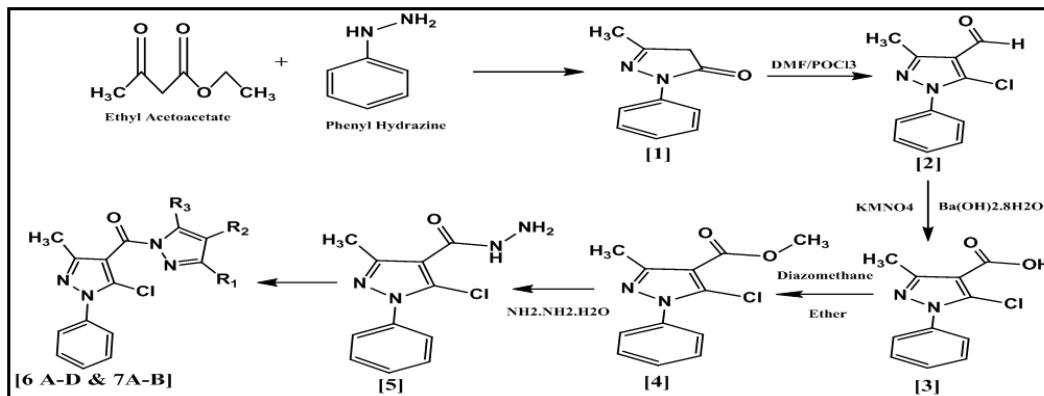
Pyrazole Carbohydrazide,
Chloro(aminopyrazoly) Ketones
derivatives, Herbicidal Activity.

Abstract

In order to obtain new chloro(aminopyrazoly)ketone compounds with good herbicidal activity, a series of 5-Chloro-3-methyl-1-phenyl-1H pyrazole-4-carbohydrazide derivatives were designed and synthesized using a series of relational synthons. Their structures were determined by IR, ¹H NMR, and elemental analyses. These compounds were screened for Herbicidal activities against Barnyard grass and Rape seed.

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Scheme 1:-



Where R₁= CH₃, H; R₂= COOC₂H₅, CONHNH₂, CN, H; R₃=NH₂, OH

Introduction:-

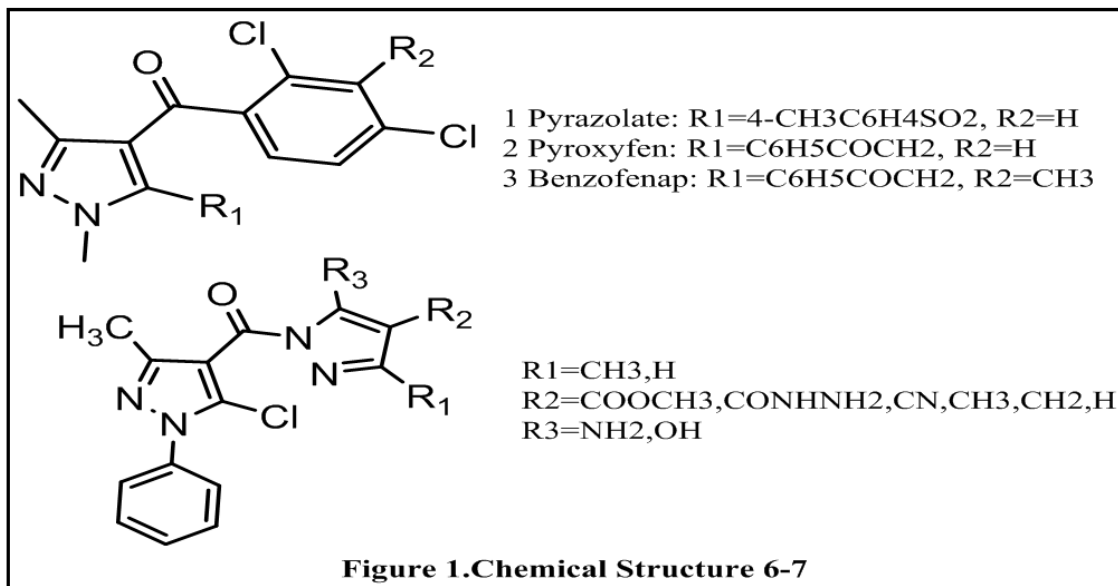
Pyrazole type herbicidal are those compounds which having a low phytotoxicity, exhibiting excellent herbicidal activity at lower dosages than known analogous herbicidal. As reported, many 4-carbonyl pyrazole derivatives classified as the inhibitors of 4-hydroxyphenyl-pyruvate dioxygenase (4-HPPD) displayed excellent herbicidal activities, such as pyrazolynate 1, pyrazoxyfen 2 and benzofenap 3 (Figure 1). 4-HPPD inhibitor are a class of herbicides that prevent plants by blocking 4-Hydroxyphenyl pyruvate dioxygenase, an enzyme in plants that breaks down the amino acid, tyrosine into components that are used by plants to create other molecules that plants need. This process of breakdown, or catabolism, and making new molecules from the results, or biosynthesis, is something all living things do¹.

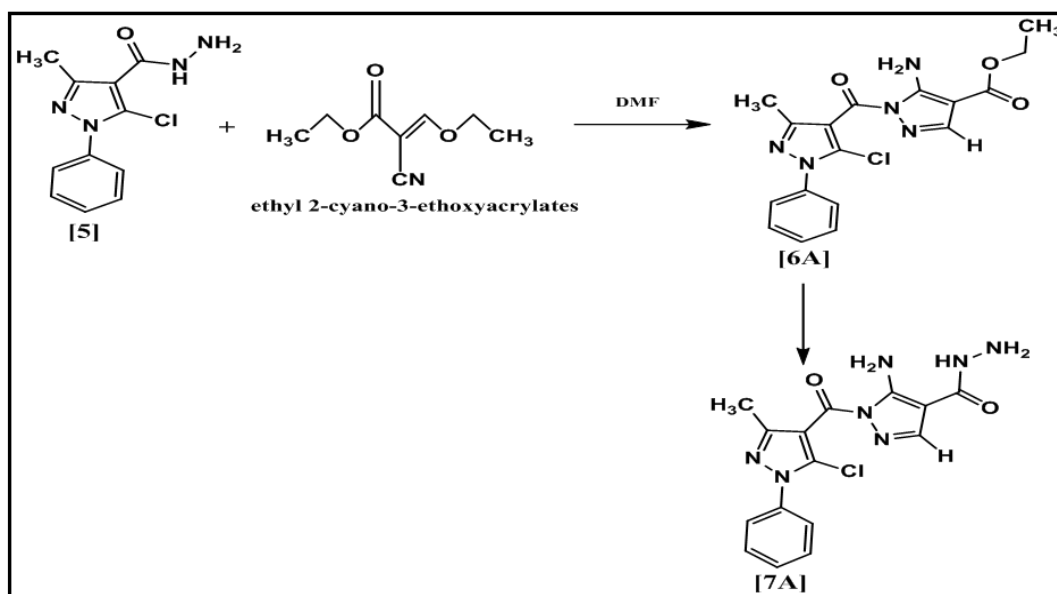
Corresponding Author:- Shailesh Prabhakar Ghag.

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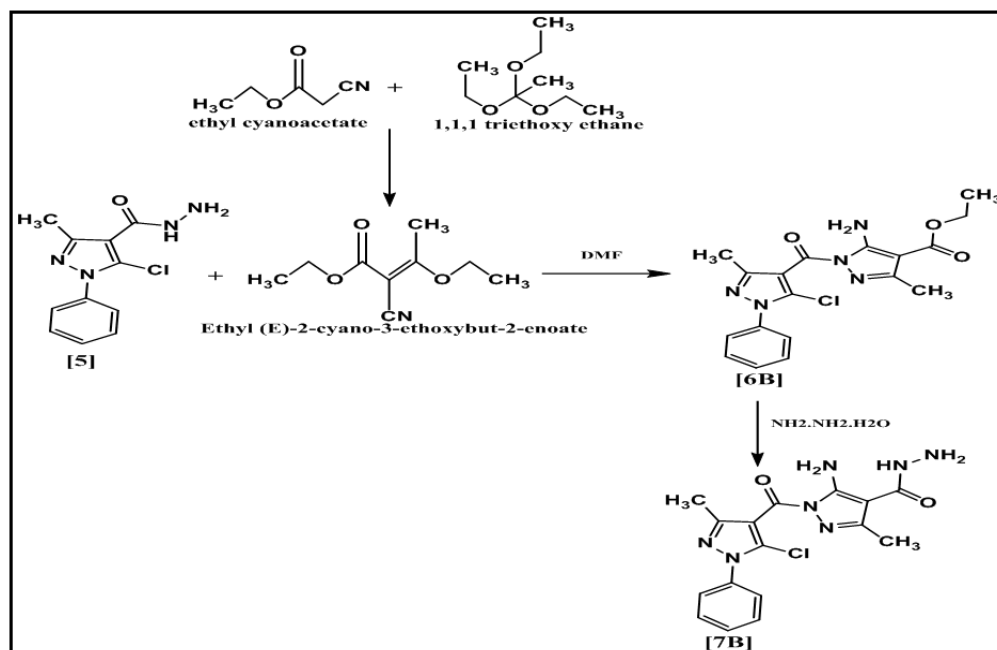
HPPD inhibitors were first brought to market in 1980, although their mechanism of action was not understood until the late 1990s. They were originally used primarily in Japan in rice production, but since the late 1990s have been used in Europe and North America for corn, soybeans, and cereals, and since the 2000s have become more important as weeds have become resistant to glyphosate and other herbicides. Among these Benzofenap is a Pyrazole herbicide first synthesized in 1981 by Mitsubishi Petrochemical Company². It is a translocated, selective, root and shoots absorbed herbicide used mainly for broad-leaved water weed control in Rice. Pyrazolynate, an herbicide belonging to the group unclassified is used for controlling weeds. Several Pyrazolynate also has a high activity on rice weeds³. Pyrazoxyfen is a benzoyl pyrazole herbicides, it is used pre or post emergence after transplanting in paddy rice to control annul and perennial weeds⁴. It can be used to direct seeded rice.

By comparisons with other 4-hydroxyphenol-pyruvate dioxygenase (4-HPPD) inhibitors, it was concluded that their main pharmacophore was 5-hydroxyl-4-benzoyl pyrazole. It was also noticed that amino group possessed a similar capability of forming complex compounds with hydroxyl group, and according to the bioisosterism theory, both pyrazole ring and benzene ring were ring equivalents. Also it had chloro group at 5th position in main pyrazole moiety. However, the pyrazole derivatives containing two pyrazole rings used as herbicide have been rarely reported. In order to find valuable lead compounds with high herbicidal activity, a series of the title compounds 6-7 (Figure 1) were designed and synthesized.

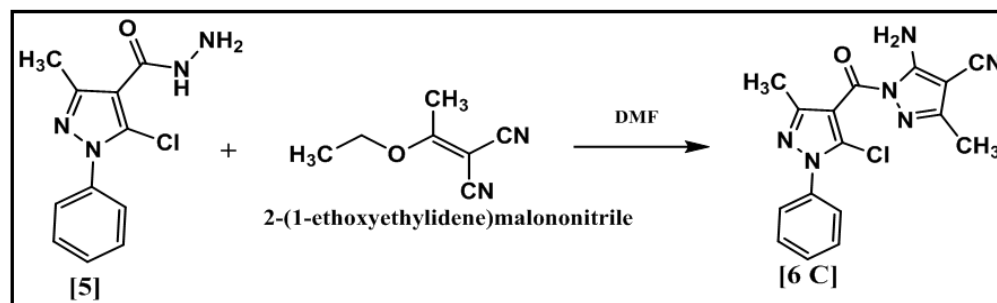




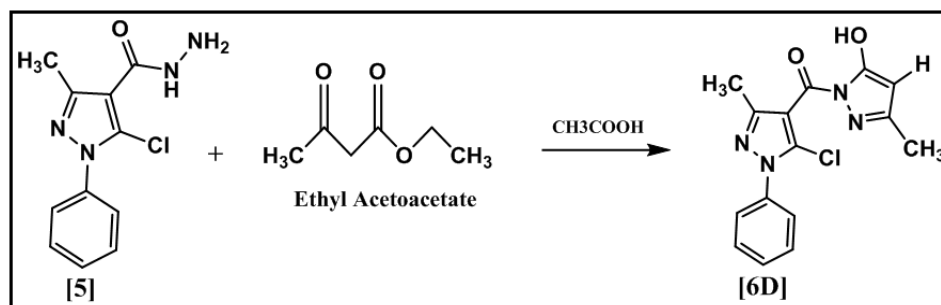
Schemes 1.



Schemes 2.



Scheme 3.



Scheme 4.

Material and Method:-

Synthesis:-

5-Chloro-3-methyl-1-phenyl-1*H*-pyrazole-4-carbohydrazide was prepared as per the procedure described in our research paper⁵

The title compounds were synthesized according to Scheme 1-4 by in a parallel synthesis fashion, by which a number of 5-Amino compounds with various substitutions and 5-Hydroxyl compound are produced. In First, ethyl 2-cyano-3-ethoxyacrylates were reacted with 5-Chloro-3-methyl-1-phenyl-1*H*-pyrazole-4-carbohydrazide [5] to obtained Ethyl-5-amino-1-(5-chloro-3-methyl-1-phenyl-1*H*-pyrazole-4-carbonyl)-1*H*-pyrazole-4-carboxylate[6A], then [6A] was transformed to its hydrazine derivatives [6B].

In second scheme, ethyl cyanoacetate reacted with 1,1,1 triethoxy ethane to obtained Ethyl (E)-2-cyano-3-ethoxybut-2-enoate. This is reacted with 5-Chloro-3-methyl-1-phenyl-1*H*-pyrazole-4-carbohydrazide [5] to obtain Ethyl-5-amino-1-(5-chloro-3-methyl-1-phenyl-1*H*-pyrazole-4-carbonyl)-3-methyl-1*H*-pyrazole-4-carboxylate [7A]. Then [7A] was transformed to its hydrazine derivatives [7B].

In third scheme 2-(1-ethoxyethylidene)malononitrile was reacted with 5-Chloro-3-methyl-1-phenyl-1*H*-pyrazole-4-carbohydrazide [5] to obtained 5-Amino-1-(5-chloro-3-methyl-1-phenyl-1*H*-pyrazole-4-carbonyl)-3-methyl-1*H*-pyrazole-4-carbonitrile [6C].

In forth scheme Ethyl acetoacetate was reacted with 5-Chloro-3-methyl-1-phenyl-1*H*-pyrazole-4-carbohydrazide [5] to obtained (5-Chloro-3-methyl-1-phenyl-1*H*-pyrazol-4-yl)(5-hydroxy-3,4-dimethyl-1*H*-pyrazol-1-yl) methanone [6D]. All compounds are characterized by ¹H NMR and FTIR analysis.

Herbicidal activity:-

These compounds were screened for herbicidal activities against rape and Barnyard grass (Table 1). The results showed that, when R₂ was 4-carbohydrazide group [6B & 7B], compounds always possessed better herbicidal activities against barnyard grass rather than ester, cyano and hydroxyl group at 100µg/mL; All the synthesized compounds showing non-inhibitory activity against Rape root.

Table 1:- Structure and herbicidal activity of targeted compounds.

Sample No.	Comp.	R ₁	R ₂	R ₃	MP (°C)	Yield %	Rape root Test (cm) 100 µg/mL	Barnyard grass cup test (cm) 100 µg/mL
1	Water						6	5.7
2	Blank						5.8	5.0
A	6A	-H	-COOC ₂ H ₅	-NH ₂	236-238	54	3.75	1.25
B	6B	-H	-CONHNH ₂	-NH ₂	289-292	60	4.0	0.75
C	7A	-CH ₃	-COOC ₂ H ₅	-NH ₂	225-228	58	3.80	2.25
D	7B	-CH ₃	-CONHNH ₂	-NH ₂	282-285	60	4.0	0.45
E	6C	-CH ₃	-CN	-NH ₂	285-288	50	3.65	2.0
F	6D	-CH ₃	-H	-OH	250-254	45	3.77	1.25

Figure 2:- Herbicidal Activity of compounds containing carbohydrazide group at 4th position [6B & 7B] on Rape seed & Barnyard grass.



Experimental:-

General Methods:-

Melting points were determined in open capillary on an electrically heated block are uncorrected. IR (KBr) spectra were recorded on Perkin Elmer FTIR and only noteworthy absorption levels (cm^{-1}) are listed. PMR spectra on Varian 300 MHz and AMX-400 Bruker spectrophotometer using TMS as an internal standard and chemical shifts measured in δ (ppm). The required 3-Methyl-1-phenyl-2-pyrazolin-5-one was prepared according to Vogel's procedure⁶. Diazomethane required for the methylation of acid was prepared according to Organic synthesis method⁷.

5-Chloro-3-methyl-1-phenyl-1*H*-pyrazole-4-carbaldehyde [2] was prepared under Vilsmeier conditions^{8,9} from 3-Methyl-1-phenyl-2-pyrazolin-5-one [1] in which chlorination of C-5 occurs in addition to the expected formylation. This 4-carboxaldehyde is then oxidized with Potassium permanganate¹⁰ to give 5-Chloro-3-methyl-1-phenyl-1*H*-pyrazole-4-carboxylic Acid [3]. This acid is then methylated by diazomethane in dry ether to give 5-Chloro-3-methyl-1-phenyl-1*H*-pyrazole-4-carboxylic acid methyl ester [4]. The methyl ester is then treated with hydrazine hydrate to give 5-Chloro-3-methyl-1-phenyl-1*H*-pyrazole-4-hydrazide [5].

TLC was used to assess the reactions and the purity of the synthesized compounds. All the compounds gave satisfactory elemental analysis.

3-Methyl-1-phenyl-2-pyrazolin-5-one [1]:-

50 gm (49 mL, 0.384 mole) of redistilled ethyl acetoacetate and 40 gm (36.5 mL, 0.37 mole) of phenyl hydrazine were mixed together in a large evaporating dish. The mixture was heated on a boiling water bath in the fume cupboard for about 2 hours and stirred from time to time with glass rod. The heavy reddish syrup was allowed to cool somewhat, about 100 mL of ether was added to it and the mixture was stirred vigorously. The syrup, which was insoluble in ether, solidified within 15 minutes. The solid was filtered at the pump and washed thoroughly with ether to remove colored impurities. It was recrystallized from hot water to give 3-methyl-1-phenyl-2-pyrazolin-5-one.

Chloro-3-methyl-1-phenyl-1*H*-pyrazole-4-carbaldehyde [2]:-

For the preparation of 5-Chloro-3-methyl-1-phenyl-1*H*-pyrazole-4-carbaldehyde, POCl_3 (0.35 mole, 32.0 mL) was added to ice-cold dimethyl formamide (0.16 mole, 12.0 mL). To this mixture 3-Methyl-1-phenyl-2-pyrazolin-5-one [1] (0.05 mole, 8.1 gm) was added and the mixture was heated under reflux for 1.0 hrs. After cooling the reaction mixture was poured into ice-cold water (300 mL). The solid precipitated was collected by filtration, washed with cold water, dried and recrystallized from ethanol to give pale yellow crystal of 5-Chloro-3-methyl-1-phenyl-1*H*-pyrazole-4-carbaldehyde. Yield (90 %), mp: 145-148 °C.

Chloro-3-methyl-1-phenyl-1*H*-pyrazole-4-carboxylic Acid [3]:-

In three 500 mL flask, 5-Chloro-3-methyl-1-phenyl-1*H*-pyrazole-4-carbaldehyde [2] 0.05 mol and 100 mL of water was slowly added dropwise 0.075 mol KMnO_4 in 200 mL aqueous oxidation. Dropping was completed, the

temperature was raised to 70 -80 ° C reactor 8 h. Adjusting the pH of the reaction solution was made alkaline with 10% Barium Hydroxide solution was slowly cooled, insoluble's were removed by filtration, and the filtrate was acidified with concentrated hydrochloric acid, the precipitated white solid was suction filtered, washing and drying, a white solid; Yield: Yield- (70.0 %) mp: 230 °C.

Chloro-3-methyl-1-phenyl-1H-pyrazole-4-carboxylic Acid Methyl ester [4]:-

2.365 gm (0.01 mole) of 5-Chloro-3-methyl-1-phenyl-1 H-pyrazole-4-carboxylic acid [3] was dissolved in 25 mL of ether. The solution was cooled to 0°C, with proper precaution excess diazomethane solution was added. As the reaction continued nitrogen generated in the reaction solution bubbled out. The reaction mass was kept in refrigerator overnight. Excess diazomethane was distilled out. The solid precipitate of 5-Chloro-3-methyl-1-phenyl-1 H-pyrazole-4-carboxylic acid methyl ester was recrystallized in ethanol. Yield- 75 % mp: 68 °C.

Chloro-3-methyl-1-phenyl-1 H-pyrazole-4-carboxylic acid hydrazide [5]:-

2.0 gm of 5-Chloro-3-methyl-1-phenyl-1 H-pyrazole-4-carboxylic acid methyl ester [4] was kept overnight under reflux in 10.0 mL of hydrazine hydrate. 25.0 mL of chloroform was added to the reaction mass and the hydrazine hydrate layer was separated. The chloroform layer was washed three times with water. The chloroform layer was dried on anhydrous sodium sulfate. The chloroform was evaporated to get solid precipitate of 5-Chloro-3-methyl-1-phenyl-1 H-pyrazole-4-carboxylic acid hydrazide. It was recrystallized in ethanol. Yield- (60.0 %) mp: 172-175 °C.

General procedure for the synthesis of title compounds [6A, 7A & 6C]:-

To a solution of intermediate [5] (5.0 mmol) in 15mL DMF is added Cyano compound (5.5 mmol). The mixture is refluxed for 8 h and cooled to room temperature, then poured into 30 mL water. The precipitate is collected by filtration and a white solid is obtained. The solid is purified by Column Purification. Desired products [6 A, 7 A & 6C] are obtained.

Procedure for the synthesis of title compounds [6D]:-

To a solution of intermediate [5] (5.0 mmol) in 15mL Acetic Acid is added Ethyl acetoacetate (5.5 mmol). The mixture is refluxed for 10 h and cooled to room temperature, then poured into 30 mL water. The precipitate is collected by filtration and a white solid is obtained. The solid is purified by Column Purification. Desired product [6D] is obtained.

General Procedure for the Reaction of 5-Amino-1H-pyrazole-4-carbohydrazide [6B & 7B]:-

A suspension of pyrazole derivatives [6A or 7A] (0.05mol) in 14.7 mL 85% hydrazine hydrate is heated at 105° for 6 h. Then the solution is evaporated under vacuum and cooled to room temperature. The residue is filtrated; washed with 25 mL diethyl ether three times. White crystalline solids [6B or 7B] are obtained.

Ethyl-5-amino-1-(5-chloro-3-methyl-1-phenyl-1H-pyrazole-4-carbonyl)-1H-pyrazole-4-carboxylate [6A]:-

This compound is obtained as colorless white crystals (alcohol), yield 54 %, mp 236~238 °C;

¹H NMR (400 MHz, δ ppm, CDCl₃): δ 0.85 ppm (t, 3H, COOCH₂CH₃); δ 2.1 ppm (d, 3H, CH₃); δ 3.85 ppm (q, 2H, COOCH₂); δ 5.45 ppm (s, 2H, NH₂); δ 7.0-7.1 ppm (m, 5H, ArH); δ 7.39 ppm (s, 1H, -CH)

IR (potassium bromide): 900-675 — C—H stretching; 1097— Cl stretching; 1252.79— C=C=O—O stretching; 1375-1450— CH₃ bending; 1400-1450 — CH₂ bending; 1300-1600— Ring stretching vibration; 1590.34 — C=C stretching; 1719.57 —C=O stretching; 3211.53-3415.03 — Heteroaromatic N-H stretching

Anal. Calculated for C₁₇H₁₆ClN₅O₃: C=54.63%; H=4.31%; N=18.74%

Found: C=53.51%; H=5.21%; N=18.00%

5-Amino-1-(5-chloro-3-methyl-1-phenyl-1H-pyrazole-4-carbonyl)-1H-pyrazole-4-carbohydrazide [6B]:-

This compound is obtained as colorless solid, yield 60 %; mp 292 °C.

¹H NMR (400 MHz, δ ppm, DMSO): δ 2.5 ppm (s, 3H, CH₃); δ 4.5 ppm (s, 2H, CONHNH₂); δ 5.5 ppm (s, 2H, NH₂); δ 7.5-7.7 ppm (m, 5H, ArH); δ 8.15 ppm (s, 1H, -CH); δ 9.6 ppm (s, 1H, -NH, D₂O Ex.)

IR (potassium bromide): 706.92 — C-Cl stretching; 1622.16 — C=C stretching in ring; 1655.92, 1696.42 — C=O stretching; 3211.53, 3285.79 — NH₂ stretching; 3432.39 — NH stretching

Anal. Calculated For C₁₅H₁₄ClN₇O₂: C=54.41%; H=3.61%; N= 15.86%

Found: C=55.10%; H=4.01%; N=16.50%

Ethyl-5-amino-1-(5-chloro-3-methyl-1-phenyl-1H-pyrazole-4-carbonyl)-3-methyl-1H-pyrazole-4-carboxylate [7A]:-

This compound is obtained as colorless solid (alcohol), yield 58%; mp 228 °C.

¹H NMR (400 MHz, δ ppm, CDCl₃): δ 1.4 ppm (t, 3H, COOCH₂CH₃); δ 2.4 ppm (s, 3H, CH₃); δ 2.65 ppm (s, 3H, CH₃); δ 4.4 ppm (q, 2H, COOCH₂); δ 5.5 ppm (b, 2H, NH₂); δ 7.5-7.7 ppm (m, 5H, Ar H)

IR (potassium bromide): 1096— Cl group stretching; 1261.47— C—C=O—O stretching; 1591.30— C=C stretching; 1710— C=O stretching; 3217.32-3422.74 — Heteroaromatic N-H stretching

Anal. Calculated for C₁₈H₁₈ClN₅O₃: C=55.75%; H=4.68%; N=18.06%

Found: C=55.10%; H=3.99%; N=17.65%

5-Amino-1-(5-chloro-3-methyl-1-phenyl-1H-pyrazole-4-carbonyl)-3-methyl-1H-pyrazole-4-carbohydrazide [7B]:-

This compound is obtained as colorless solid, yield 60 %; mp 285 °C.

¹H NMR (400 MHz, δ ppm, CDCl₃): δ 2.55 ppm (s, 3H, CH₃); δ 2.7 ppm (s, 3H, CH₃); δ 4.1 ppm (s, 2H, CONHNH₂); δ 5.5 ppm (s, 2H, NH₂); δ 7.5 ppm (m, 5H, ArH); δ 9.55 ppm (s, 1H, -NH, D₂O Ex.)

IR (potassium bromide): 1074.37— Cl group stretching; 1309.69 & 1552.72 — C-H Stretching; 1605.77 & 1639.52 — C=O stretching; 3062.05 — Aromatic C-H stretching; 3280.01— NH₂ stretching

Anal. Calculated for C₁₆H₁₆ClN₇O₂: C=51.41%; H=4.31%; N=26.23%

Found: C=52.20%; H=4.55%; N=25.85%

5-Amino-1-(5-chloro-3-methyl-1-phenyl-1H-pyrazole-4-carbonyl)-3-methyl-1H-pyrazole-4-carbonitrile [6C]:-

This compound is obtained as colorless solid, yield 50 %; mp 288 °C,

¹H NMR (400 MHz, δ ppm, CDCl₃): δ 2.33 ppm (s, 3H, CH₃); δ 2.69 ppm (s, 3H, CH₃); δ 7.50-7.61 ppm (m, 5H, ArH); δ 10.98 ppm (s, 2 H, NH₂)

IR (potassium bromide): 1598 — C=C (in Ring) (3 bond conjugated); 1635— C=O stretching; 2222 —CN stretching; 2924.13 — aliphatic C-H stretching; 3431.42 — NH₂ stretching

Anal. Calculated for C₁₆H₁₃ClN₆O: C=56.39%; H=3.85%; N=24.66%

Found: C=56.15%; H=3.21%; N=24.11%

(5-Chloro-3-methyl-1-phenyl-1H-pyrazol-4-yl)(5-hydroxy-3-methyl-1H-pyrazol-1-yl) methanone [6D]:-

This compound is obtained as colorless solid, yield 45%; mp 254 °C

¹H NMR (400 MHz, δ ppm, CDCl₃): δ 2.27 ppm (s, 3H, CH₃); δ 2.6 ppm (s, 3H, CH₃); δ 5.75 ppm (s, 1H, -CH); δ 7.5-7.7 ppm (m, 5H, ArH); δ 8.25 ppm (s, 1H, OH)

IR (potassium bromide): 1658— C=O stretching; 1591.30 — C=C (in Ring); 3320.51— OH— stretching

Anal. Calculated for C₁₅H₁₃ClN₄O₂: C=56.88%; H=4.14%; N=17.69%

Found: C=55.92%; H=3.89%; N=18.13%

Herbicidal Activity Tests:-

Inhibition of the root-growth of rape (*Brassicacampestris* L):-

The compounds to be tested are made into emulsions to aid dissolution. Rape seeds are soaked in distilled water for 4 h before being placed on a filter paper in a 6-cm Petri plate, to which 2 ml of inhibitor solution had been added in advance. Usually, 15 seeds are used on each plate. The plate is placed in a dark room and allowed to germinate for 65 hat 28° (±1). The lengths of 10 rape roots selected from each plate are measured and the means are calculated. The percentage inhibition is calculated relative to controls using distilled water instead of the inhibitor solution.

Inhibition of the seedling growth of barnyard grass (*Echinochloacrus-galli* (L) Beauv):-

The compounds to be evaluated are made into emulsions to aid dissolution. Ten barnyard grass seeds are placed into a 50-ml cup covered with a layer of glass beads and a piece of filter paper at the bottom, to which 5 ml of inhibitor solution had been added in advance. The cup is placed in a bright room and the seeds allowed germinating for 65 h at 28° (±1). The height of the aboveground parts of the seedlings in each cup is measured and the means calculated. The percentage inhibition is calculated relative to controls using distilled water instead of the inhibitor solution. The results of herbicidal activity listed in Table 2.

Conclusion:-

In conclusion, a 5 number of Chloro(aminopyrazoly) ketone compounds and 1 number of Chloro(hydroxypyrazoly) ketone compound were synthesized and tested for herbicidal activity. However, only a few compounds exhibited moderate activity against rape or barnyard grass. In general, the results indicated that all the compounds possessed a certain extent inhibiting activities against the rape root growth and exhibited activities against barnyard grass, but their activities are not as good as the diketone herbicides.

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RESEARCH ARTICLE

PERFORMANCE OF GROWTH TRAITS IN BANDUR SHEEP UNDER FIELD CONDITION.

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Abstract

Data pertaining to the body weight and body measurements of Bandur Sheep were collected on 737 animals from March 2015 to October 2015 of different age groups reared by farmers in the home tract village Bandur and its surrounding villages of Malavalli Taluk in Mandya district of Karnataka. The least squares mean body weight, height at withers, body length, chest girth and paunch girth were recorded to be 27.46 ± 0.27 kg, 53.48 ± 0.26 , 61.64 ± 0.15 , 72.62 ± 0.20 and 74.99 ± 0.23 cm, respectively. All these traits were significantly affected by age and sex.

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Introduction:-

Karnataka possesses well defined breeds of sheep such as Bandur, Decanni, Hassan, Bellary, Chitradurga, Yellaga and other local strain that are prevalent in certain parts of the state as adjunct to cropping pattern. Mandya sheep is the best mutton breed of sheep in India and is in great demand for upgrading of other native breeds for Mutton within the country and abroad (Arora and Acharaya, 1976, Rai *et al.*, 1988). Mandya sheep is also called Bandur sheep because this breed is known to have originated from Bandur Village of Mallavalli taluk in Mandya district of Karnataka. Bandur sheep is the best mutton breed of the country and is being slaughtered regularly at high rate for mutton production leading to depletion in its number and genetic erosion over time lag. Hence, this warrants conservation and proliferation of this breed. Data on production traits of Bandur sheep at the field condition is scanty. No systematic attempt has been made to study the production parameters of Bandur sheep under field conditions at farmer's flocks. In this context the present study was undertaken in Bandur sheep under field conditions with the objectives to quantify the performance traits such as body weights and body measurements and to study the effect of non-genetic factors on body weight and body measurements.

Materials and Methods:-

Data pertaining to the body weight and body measurements were recorded on 737 sheep reared by the farmers at different ages based on dentition. The above data was collected from March 2015 to October 2015 in the home tract area of Bandur sheep, Bandur village and its surrounding eighteen villages of Malavalli Taluk of Mandya district in Karnataka. The home tract of this breed is located at an altitude of 609.60m above the mean sea level between the latitude 12.33° and longitude 70.04° . The average rainfall of this area ranges from 518.9 mm to 897.4 mm over the last five years. The mean maximum daily temperature recorded was 35.2° during April and a minimum was 17.2° c in December. The farmer's flocks were grazed from 10.00 am to 6.00 pm on uncultivated waste land and other

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harvested fields. No supplemental feed is given to sheep maintained by the farmers. Lambs were allowed to suckle and remain with their mothers up to three months of age and weaned thereafter. Breeding rams were maintained under stall feeding conditions. Selective breeding was practiced in a limited way followed by flock mating system. Breeding is carried out throughout the year.

The animals were weighed using sensitive dial type (Salter) weighing balance before allowing for grazing and watering. Body measurements were recorded nearest to centimeter. All the measurements were taken using measuring tape after making the animal to stand squarely on an even surface. The animal was secured by an attendant with its head held a little high in a normal position as suggested by Turner *et al.*, (1953) and measurements were taken. The whole operation was carried out by a single operator to minimize possible errors. The data on body weight and body measurements were classified into five age groups such as, milk teeth, two teeth, four teeth, six teeth and eight teeth groups. The pooled data again classified into two sex groups i.e. male and female.

The least squares mean and the standard error of various traits of growth were computed by adopting Harvey's least squares method of fitting constants using the mixed model least squares and maximum likelihood computer programme (Harvey, 1990). Data used in the present study comprised of unequal subclass frequencies, thus leading to non-orthogonality. To overcome non-orthogonality of data, the least square analysis of variance technique was adopted to detect the significant sources of non-genetic variation if any (Harvey, 1987). The following fixed model was used for analysis of body weight and body measurements,

$$Y_{ijk} = \mu + A_i + S_j + e_{ijk}$$

Where Y_{ijk} = the record of the k^{th} individual belonging to the i^{th} age group and j^{th} sex group.

μ = population mean

A_i = fixed effect of the i^{th} age group ($i = 1, 2, 3, 4, 5$)

S_j = Fixed effect of j^{th} sex group ($j = 1, 2$)

e_{ijk} = Random error associated with Y_{ijk} and assumed to be identically, independently and normally distributed with mean zero and the unit variance

Result and Discussion:-

The least squares mean along with their standard errors of body weight and body measurements at different ages and sex of Bandur sheep are presented in table 1. The overall least squares mean body weight of Bandur sheep was 27.46 ± 0.27 kg (table 1). The mean body weight at different ages revealed that a faster growth rate from milk teeth to six teeth and the growth rate declined thereafter. This observation tallied closely with the reports of Acharaya and Bhat (1984) in Gaddi, Nagaraja *et al.* (1996), Siddalinga Murthy (2001) and Vasundradevi (2013) in Bandur Sheep and Taye *et al.* (2010) in Washera Sheep. The effect of age on body weight was found to be significant. Significant difference in body weights due to age were also reported by Taneja *et al.* (1993) in Magra sheep and Swarnkar and Arora (1996) in Malpura sheep, Dayananda (1998) in Tumkur strain and Siddalinga Murthy (2001) in Bandur Sheep. The increase in body weight is being noticed with advancement of age might be due to physiological phenomena of growth.

The body weight recorded for males (34.66 ± 0.49 kg) was higher compared to the females (25.63 ± 0.24 kg) indicating sexual dimorphism with respect to growth. This observation confirms the reports of Singh and Mathur (1971) in Coimbatore breed, Nimbkar (1993) in Decanni breed, Taneja *et al.* (1993) in Magra breed, Taye *et al.* (2010) in Washera Sheep and Dayananda (1998) in Tumkur strain, Nayak *et al.* (2008) in Ganjam Sheep and Siddalinga Murthy (2001) in Bandur sheep. However, no significant difference in sex was reported by Singh and Singh (1974) in Bikaneri breed.

Body measurements:-

The least squares mean body measurements at different ages in Bandur sheep is shown in table.1.

Height at withers:-

The least squares mean of height at wither recorded 53.48 ± 0.24 cm (Fig.3, table. 1). The present result was closely related with the values reported by Mahajan and Bahra (1977) in Gaddi, Taneja *et al.* (1993) in Magra, Dayananda (1998) in Tumkur strain, Taye *et al.* (2010) in Washera Sheep and Siddalinga Murthy (2001) and Vasundradevi (2013) in Bandur sheep. The height at withers of milk teeth group sheep differed significantly from all other age groups. Age had a Significant differences in height at withers as reported by Mahajan and Bahra (1977) in Gaddi,

Taneja *et al.* (1993) in Magra, Dayananda (1998) in Tumkur stain and Siddalinga Murthy (2001) in Bandur sheep. In contrast, Basu Thakur and Negi (1967) in Rampur Bushair reported non-significant effect of age on height at wither. The variation in height at withers between age groups appears to be a normal physiological function. The height at withers recorded for males (56.96 ± 0.24 cm) was higher compared to that of females (52.51 ± 0.11 cm) and the difference was significant ($P \leq 0.01$). Similar observations were made by Nimbkar (1993) in Decanni, Taneja *et al.* (1993) in Magra, Dayananda (1998) in Tumkur strain, Nayak *et al.* (2008) in Ganjam Sheep, Taye *et al.* (2010) in Washera Sheep and Siddalinga Murthy (2001) and Vasundradevi (2013) in Bandur sheep.

Body length:-

The least squares mean of body length measured was 61.64 ± 0.15 cm (Fig.2, table 1). The present result was similar to the reports of Nagaraja *et al.* (1996) in the same breed. The present observation was lower than the values reported by Basu thakur *et al.* (1967) in Rampur Bushair and Kalra *et al.* (1987) in Nali breed. However, body length recorded in the present study was higher than the values reported by Taneja *et al.* (1993) in Magra and Dayananda (1998) in Tumkur strain. Body length in milk teeth and two teeth age groups had significantly lower than all other age groups. The effect of age on body length was significant ($P \leq 0.01$). Significant differences in body length due to age were also reported by Taneja *et al.* (1993) in Magra, Dayananda (1998) in Tumkur strain and Taye *et al.* (2010) in Washera Sheep. The mean body length recorded for males was more (63.70 ± 0.27 cm) compared to females (59.54 ± 0.13 cm) and the difference was found to be significant. Similar reports were also made by Taneja *et al.* (1993) in Magra, Nayak *et al.* (2008) in Ganjam Sheep, Taye *et al.* (2010) in Washera Sheep and Siddalinga Murthy (2001) and Vasundradevi (2013) in Bandur Sheep. In contrast, Sharma *et al.* (1977) in Muzaffarnagari breed reported no significant effect of sex on body length.

Chest girth:-

The least squares mean chest girth recorded was 72.62 ± 0.20 cm (Fig.1, table.2), which closely tallied with the values reported by Basu Thakur *et al.* (1967) in Gaddi, Kalra *et al.* (1987) in Nali and Siddalinga Murthy (2001) in Bandur sheep. The chest girth was lower than the values reported by Taneja *et al.* (1993) in Magra (80.03). However, the chest girth was higher than the values reported by Dayananda (1998) in Tumkur strain and Siddalinga Murthy (2001) in Bandur sheep. The chest girth of milk teeth and two teeth age groups varied significantly from other age groups. Significant differences in chest girth due to age were also reported by Taneja *et al.* (1993) in Magra, Dayananda (1998) in Tumkur strain, Taye *et al.* (2010) in Washera Sheep and Siddalinga Murthy (2001) in Bandur sheep. In contrast, Basu Thakar and Negi (1967) reported non-significant effect of age on chest girth in Rampur Bushair breed. The chest girth recorded for males (76.42 ± 0.41 cm) was significantly ($P \leq 0.01$) higher than females (71.43 ± 0.19 cm). Similar observations were made by Nimbkar (1993) in Deccani, Taneja *et al.* (1993) in Magra, Nayak *et al.* (2008) in Ganjam Sheep, Taye *et al.* (2010) in Washera Sheep and Siddalinga Murthy (2001) and Vasundradevi (2013) in Bandur sheep. In contrast, Dayanda (1998) in Tumkur strain, reported non-significant effect of sex on chest girth.

Paunch girth:-

The least squares mean paunch girth measured was 74.99 ± 0.23 cm (Fig.4, table .1) which was tallied with the reports of Nagaraja *et al.* (1996) in Bandur breed. This value is lower than the values reported by Taneja *et al.* (1993) in Magra. However, it was higher than the value reported by Bhadula *et al.* (1979) in Muzzarffarnagari breed. The paunch girth of milk teeth and two teeth groups were lowest and differed significantly from those of remaining 4, 6, and 8 teeth age groups. Age had a significant effect on paunch girth which was in agreement with Taneja *et al.* (1993) in Magra, Dayananda (1998) in Tumkur strain and Siddalinga Murthy (2001) in Bandur sheep. The paunch girth recorded for males was more (78.99 ± 0.48 cm) compared to the females (73.78 ± 0.23 cm) and the difference was found to be highly significant ($P \leq 0.01$). Significant effect on paunch girth due to sex was also reported by Nimbkar (1993) in Decanni, Taneja *et al.* (1993) in Magra, Nayak *et al.* (2008) in Ganjam Sheep, Taye *et al.* (2010) in Washera Sheep and Siddalinga Murthy (2001) and Vasundradevi (2013) in Bandur sheep. However, Dayananda (1998) reported non-significant effect of sex on paunch girth in Tumkur strain.

Conclusion:-

The growth performance of Bandur sheep is quantified under field conditions in the home tract area. The study concludes that the Bandur sheep breed maintained by farmers under grazing system were equally efficient as those maintained at organized farms. Further the body weights and body measurements recorded in Bandur sheep were found similar to other recognized breeds found in southern peninsular India.

Table.1:- Least squares means and standard error of body weight and body measurements at different ages of Bandur sheep.

Particulars	No. of observations	Body weight(kg)	Height at withers (cm)	Body length (cm)	Chest girth (cm)	Paunch girth (cm)
Overall	737	27.46±0.27	53.48±0.24	61.64±0.15	72.62±0.20	74.99±0.23
Age groups		0.01	0.01	0.01	0.01	0.01
Milk teeth (1year)	116	23.76±0.57 ^a	51.79±0.27 ^a	56.91±0.29 ^a	68.73±0.45 ^a	70.66±0.55 ^a
Two teeth (1½ year)	169	27.99±0.46 ^b	54.14±0.21 ^b	60.18±0.23 ^b	72.15±0.33 ^b	74.52±0.47 ^b
Four teeth (2½ year)	120	32.97±0.49 ^c	54.21±0.24 ^c	62.85±0.27 ^c	75.74±0.43 ^c	78.60±0.51 ^c
Six teeth (3½ year)	144	33.46±0.51 ^c	56.19±0.22 ^d	64.47±0.26 ^c	76.49±0.41 ^c	79.23±0.50 ^d
Eight teeth (4½ year)	198	31.64±0.46 ^d	56.39±0.21 ^d	63.63±0.24 ^c	76.57±0.36 ^c	78.94±0.43 ^d
Sex		0.01	0.01	0.01	0.01	0.01
Male	120	34.66±0.49 ^a	56.96±0.24 ^a	63.70±0.27 ^a	76.42±0.41 ^a	78.99±0.48 ^a
Female	617	25.51±0.24 ^b	52.51±0.11 ^b	59.54±0.13 ^b	71.43±0.19 ^b	73.78±0.23 ^b

Within the column means with at least one common superscript do not differ significantly from each other.

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RESEARCH ARTICLE

AN EFFICIENT DESIGN OF DIGITAL DOWN CONVERTER FOR SOFTWARE DEFINED RADIO APPLICATION

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ABSTRACT

Adaptive wave pipelining is the methodology used for improving the overall performance of Software Defined Radio (SDR). This method is a functional combination of wave pipelining and hybrid technique. Wave pipelining is the methodology used for improving the performance without using the intermediate latches, at the same time it performs the same operation as pipelining. Hybrid technique is the mechanism that is used for introducing registers for achieving the timing constrain. This technique is implemented in the Direct Digital Synthesizer (DDS) which is the integral part of a Digital Down Converter (DDC) in the Digital Front End (DFE)/ Intermediate Frequency (IF) of Software Defined Radio (SDR). This paper presents the principles of wave pipelining and the method for executing the computer algorithm named Coordinate Digital Rotation Digital Computer (CORDIC) using wave pipelining. Xilinx ISE 14.75 design suite is used as the software for simulation of the proposed system.

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Introduction:-

A SDR is defined as the functional system that is implemented in software and whose physical layer behavior can be changed by changing the software function. In SDR waveform signal processing is done digitally. The SDR was developed to obtain transmission link inside different bands of spectrum with a single device. SDR is a radio that solves the gap between different Link-layer protocols and provide an ideal solution for the different functional and performance problem by building a generic platform that switches the functionalities using software control. Fig. 1 shows the basic architecture of SDR.

Fig. 2 shows SDR modules in the two-axis graph Processing Intensity vs. Flexibility that determines some of spectrum signal processing associated with SDR system [1]. The upper left area indicates dedicated functions like ADC & DDC that functions with hardware. Flexibility defines the range of ease to complete the function. The lower area determines the functions like analysis and decision making which are functional parameters.

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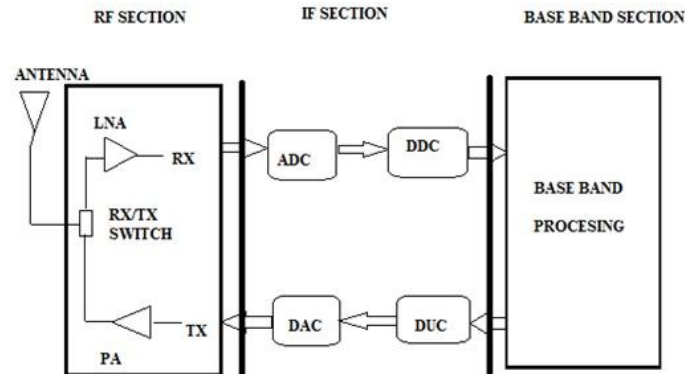


Fig. 1:- Basic architecture of SDR.

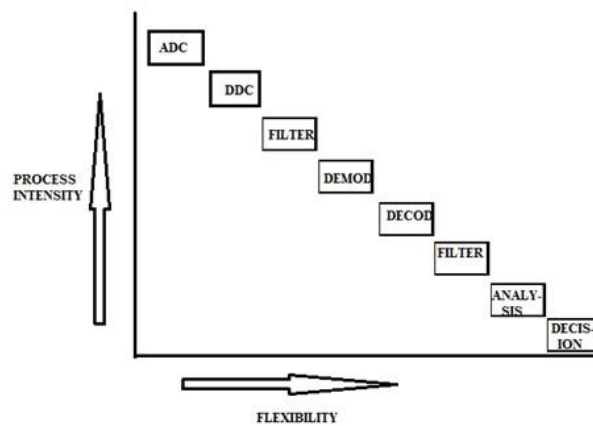


Fig. 2:- SDR modules.

Theory of ddc:-

DDC are implemented in communication processors for converting the sample frequencies. Digital down conversion occurs whenever a signal is of any particular frequency band to baseband [6]. The Fig. 3 (a) & (b) shows the functional block diagram, block diagram of DDC. DDC include frequency shifting operation with chain of mixers, decimators and filters in addition to sampling rate conversion.

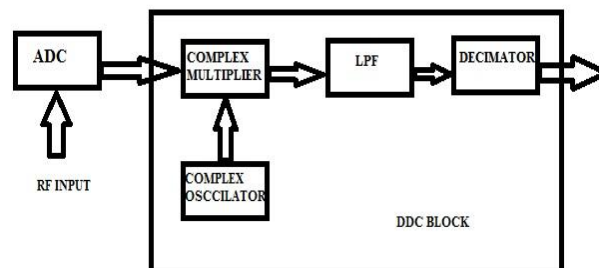


Fig. 3:- (a). Functional Block Diagram of DDC.

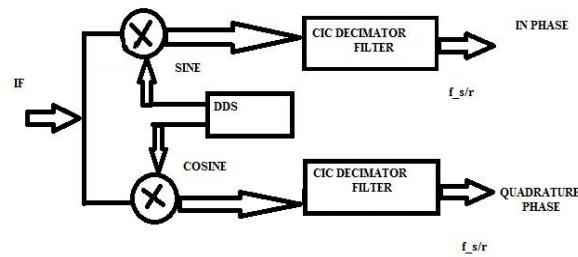


Fig. 3:- (b). Digital Down Converter Block Diagram

DDC is a complex mixer that shifts the frequency of particular range to baseband frequency. The functionality of DDC is mixing and multiplying the retrieved samples with the available digitized stream of data that produces corresponding sine and cosine phase and quadrature channels. DDC's decimate to a lower sampling frequency rate by using different stages of decimation filters. Filtering is performed to limit the bandwidth with the help of linear phase filters. The signals with low data rates are easy to be processed on a low speed functional DSP processor.

DDS/NCO:-

Direct Digital Synthesizer (DDS) is also termed Numerically Controlled Oscillator (NCO). When brought together with a DAC to create an analog output waveform, the system is called a (DDS).

NCO is digital signal generators that generate synchronous, discrete time, discrete value representation of a waveform. In NCO, digital accumulator is used to generate the address into a lookup table. Fig. 4 shows the NCO/DDS functional block diagram.

The system is common, both in hardware and in software. It allows instantaneous changes in the instantaneous frequency or phase of the generated waveform, while maintaining a continuous phase property in the output.

The NCO has 2 parts: [1] Phase Accumulator
[2] Phase To Amplitude Converter

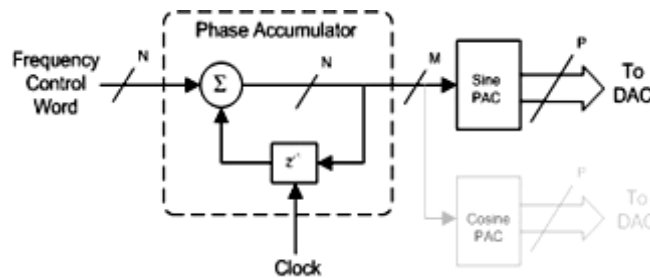


Fig. 4:- NCO/DDS functional block diagram

Theory of cordic algorithm:-

Jack. E. Volder in 1959 coined the Coordinate rotation digital computer (CORDIC). The methodology is applicable for functional execution of digital communication system. It has the characteristics of reading and analyzing the values of any functions that include trigonometric, hyperbolic, arithmetic, vector rotation, and logarithmic [3]. It computes the rotation of vectors by addition and shift operation. They are executed either in rotation mode or vectoring mode [4]. Table 1 shows the output for each mode of operation in different coordinate system. There are three different systems in which the CORDIC can be operated and they are circular coordinate system or in linear coordinate system or in hyperbolic coordinate system.

Fig. 5 shows ith stage for CORDIC from which the basic operating functions of CORDIC for ith iteration can be fetched.

$$\begin{aligned} X_{i+1} &= [X_i - C_i * Y_i * 2^{-i}] * K_i \\ Y_{i+1} &= [Y_i - C_i * X_i * 2^{-i}] * K_i \\ Z_{i+1} &= Z_i - C_i * \arctan(2^{-i}) \end{aligned}$$

C_i Denotes direction of rotation that can have value of 1 or -1. This is determined by the rotational direction whether positive rotation or a negative rotation. K_i Denotes scaling factor of ith iteration that can be computed at the end stage as it is determined by combination of all stages.

The K value is given theoretical as

$$K = \pi_i * K_i = .60725$$

Table 1:- Output for each mode of operation in different coordinate system

COORDINATE	ROTATION ($Z_n \rightarrow 0$)	VECTORIZING ($Y_n \rightarrow 0$)
CIRCULAR(m=1)	$X_n = \frac{1}{K_m} (X \cos Z - Y \sin Z)$ $Y_n = \frac{1}{K_m} (Y \cos Z + X \sin Z)$	$X_n = \frac{1}{K_m} \sqrt{(X^2 + Y^2)}$ $Z_n = Z + \tan^{-1} \left(\frac{Y}{X} \right)$
LINEAR(m=1)	$X_n = X$ $Y_n = Y + X * Z$	$X_n = X$ $Z_n = Z + \left(\frac{Y}{X} \right)$
HYPERBOLIC(m= 1)	$X_n = \frac{1}{K_m} (X * \cosh Z + Y * \sinh Z)$ $Y_n = \frac{1}{K_m} (Y * \cosh Z + X \sinh Z)$	$X_n = \frac{1}{K_m} \sqrt{(X^2 - Y^2)}$ $Z_n = Z + \tanh^{-1} \left(\frac{Y}{X} \right)$

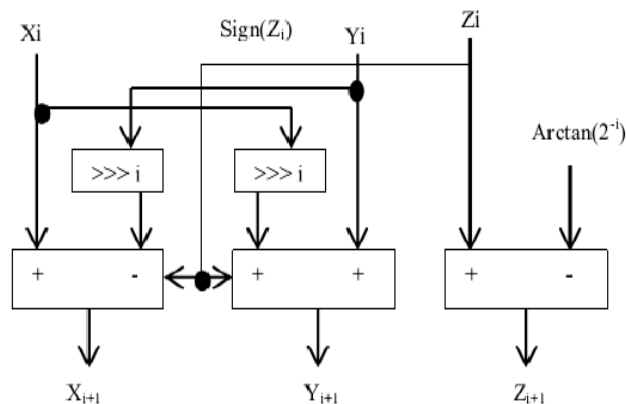


Fig. 5:- ith stage for CORDIC.

Pipeline CORDIC:-

The entire existing system is designed based on pipeline CORDIC. In the pipelined CORDIC architecture, each module is responsible for each elementary rotation occurring in the different modules selected [8] [9]. The modules are combined through immediate latches as shown in Fig. 6. Every stage of CORDIC includes addition, subtraction and shifting operation.

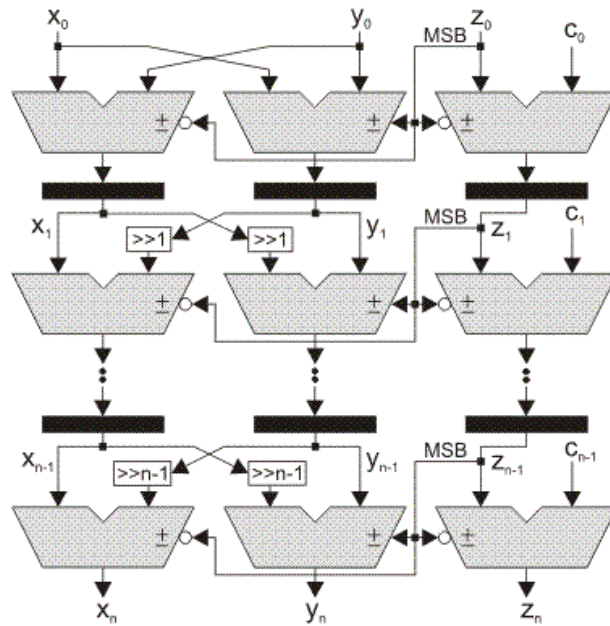


Fig. 6:- Cascading through immediate latches.

Fig. 7:-represent pipelined CORDIC. For a dedicated pipelined architecture, either IN_REGS or OUT_REGS is eliminated there by to improve latency of the design.

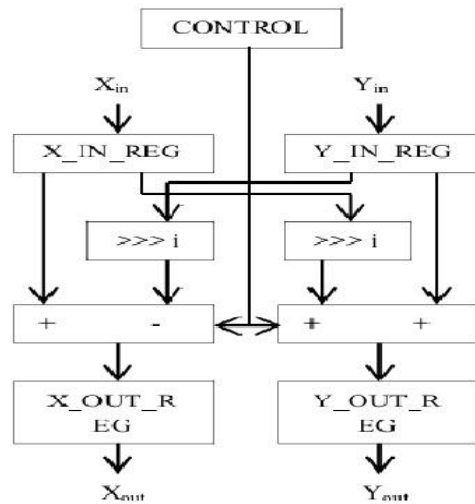


Fig. 7:- i^{th} stage for CORDIC

Table 2 shows the pre computed values of the angle α_i needed for the i^{th} iteration that will be stored in ROM memory location.

Table 2:- Pre-computed angles.

I	$2^{-i} = \tan \alpha_i$	$\alpha_i = \arctan(2^{-i})$	α_i in radians
0	1	45^0	0.7854
1	0.5	26.565^0	0.4636
2	0.25	14.063^0	0.245
3	0.125	7.125^0	0.1244
4	0.0625	3.576^0	0.0624
5	0.03125	1.7876^0	0.0312
6	0.015625	0.8938^0	0.0156
7	0.0078125	0.4469^0	0.0078
..

Overflow is parameter that occurs whenever a rotational angle crosses from a positive right angle to negative one [5] [9]. To avoid this there is a functional overflow system control is added. This analyses the nature of operand involved in the operation. If an overflow occurs it retains its previous value.

Advantages of CORDIC Pipeline Over Other Architecture:-

1. Less area consumption
2. Low power consumption
3. Lesser delay
4. Greater throughput

On comparison with the performance

Disadvantage:-

1. Area can be reduced
2. Power consumption can be reduced
3. Reducing delay increasing the speed
4. Throughput can be reduced

Proposed system:-

The proposed system is to design further better architecture that can improve the overall performance of SDR in its applications. There by through comparative and analytical study Adaptive Wave Pipelining (AWP) is the technique that can further improve overall performance of SDR in all aspects comparative of area, speed, throughput, delay.

Adaptive wave pipelining (AWP) is the technique that is implemented in the proposed system. It is combinational logic of wave pipelining and hybrid technique. Wave pipelining is the methodology which is similar to pipelining with the difference that intermediate registers are not used for the wave pipelining execution.

Wave pipelining is an alternative pipelining technique that reduces the clock loads, area, power and latency at the same it retains the external functionality and timing of the circuit [12] [13]. Wave pipelining was coined by Cotton [8] and initially named it as maximum rate pipelining. Synchronization of signals in wave pipelining is achieved by manipulating the timing of signals due to lack of intermediate registers. Fig. 8 represents the block diagram of wave pipelining.

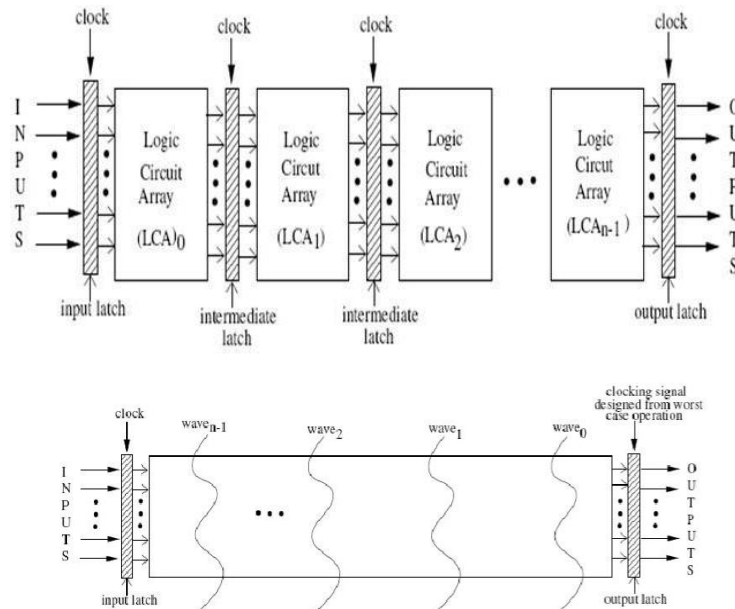


Fig.8:- Block diagram of wave pipelining.

Since there is manipulation of the signals on a periodic basis registers are introduced for the clocking function. This method of introducing the registers in wave pipelining leads to the design of proposed system AWP.

Timing Function:-

Timing requirements for a wave pipeline circuit is done by attaching the registers at the input and output at the periodic basis [14]. The clocking function is derived using certain parameters that help in deriving the timing constrain for the clocking function.

Fig. 9 (a) represents the logic of adaptive wave pipelining and (b) represents the spatial/temporal diagram of wave pipelining.

The timing function is illustrated through the following formulas:-

$$T_{\max} > D_{\max} + T_{\text{set}} + S_{\text{clk}}$$

$$T_{\text{capture}} < T_{\text{clk}} + D_{\min} - (T_{\text{hold}} + S_{\text{clk}})$$

$$T_{\text{clk}} > (D_{\max} - D_{\min}) + (T_{\text{set}} + T_{\text{hold}} + S_{\text{clk}})$$

Where:-

D_{\max} – Is difference between longest path

D_{\min} – Is the difference between shortest path

T_{set} – Set time

T_{clk} – Clock time

T_{hold} – Hold time

S_{clk} – Clock skew

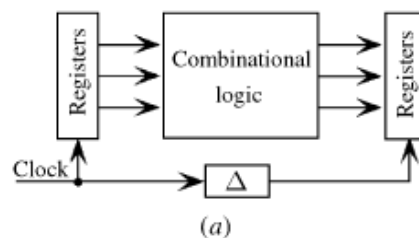


Fig. 9:- (a) Data flow through combinational logic.

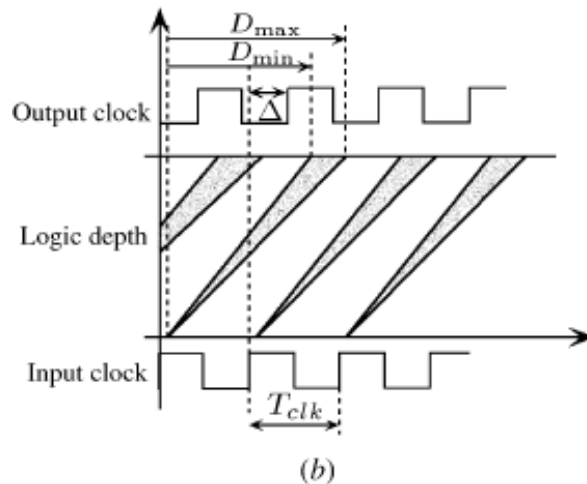


Fig. 9:- (b). Temporal/Spatial representation of wave pipelining.

Fig. 10 shows the functional flow diagram, of AWP. In AWP the entire wave pipelining process is executed by introducing the clocking function for registers. These intermediate registers re used for timing constrain.

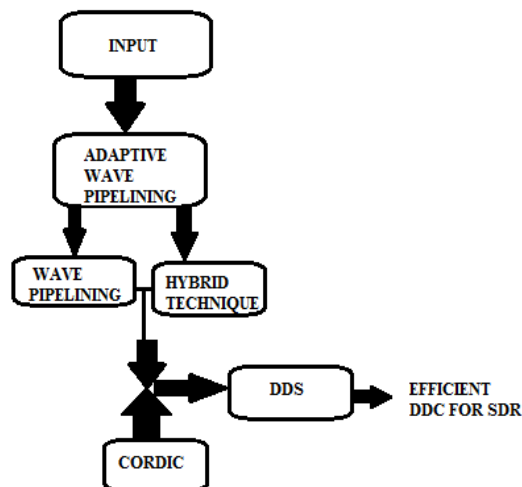


Fig.10:- Flow diagram of AWP

Advantages of Proposed System:-

1. Reduction in area
2. Reduction in power consumption
3. Increase in speed
4. Increase in throughput

Outputs and case study:-

Table 3:- Theoretical comparison of existing and proposed.

FACTORS	CORDIC PIPELINE	ADAPTIPE WAVE PIPELINING WITH CORDIC
SPEED	LESS	MORE
POWER CONSUMPTION	LESS	MORE
MEMORY UTILISATION	LESS	MORE
THROUGHPUT	LESS	MORE

Outputs obtained for the CORDIC with AWP using Xilinx synthesis.

Fig. 11:- And Fig. 12. Represent the output wave for AWP for the specified inputs. They are analyzed through the case studies.

Clk: - 50 duty cycles force constant 1
Enb: - 1
Xi: - 2
Yi: - 3
Zi: - 4



Clk: - 50 duty cycles force constant 1
Enb: - 1
Xi: - 3
Yi: - 4
Zi: - 5

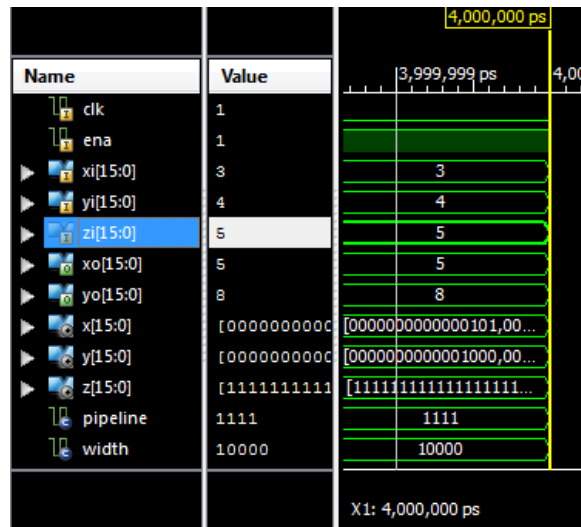


Fig. 14:- Output for AWP

Xo: - 5

Yo: - 8

Inference from the above wave forms fetched is that, whenever each time the value for inputs is forced in the ISim simulator there is corresponding change in the outputs of the wave. The number of register values is clearly available from the output window for further clarification synthesis reports are taken and compared.

Fig. 14. And Fig. 15. Is the utilization summary report that determines the number of registers and LUT's used along with which the delay and maximum frequency, is specified from the synthesis report. These utilization summary of both CORDIC pipeline and AWP are used for comparison.

Device Utilization Summary (estimated values)				
Logic Utilization	Used	Available	Utilization	
Number of Slice Registers	1000	126800	0%	
Number of Slice LUTs	1129	63400	1%	
Number of fully used LUT-FF pairs	968	1161	83%	
Number of bonded IOBs	74	170	43%	
Number of BUFG/BUFGCTRLs	1	32	3%	

Minimum period: 2.398ns (Maximum Frequency: 416.963MHz)

Fig. 14:- Utilization summary for CORDIC pipeline.

Device Utilization Summary (estimated values)				
Logic Utilization	Used	Available	Utilization	
Number of Slice Registers	680	126800	0%	
Number of Slice LUTs	723	63400	1%	
Number of fully used LUT-FF pairs	652	751	86%	
Number of bonded IOBs	50	170	29%	
Number of BUFG/BUFGCTRLs	1	32	3%	

Minimum period: 2.036ns (Maximum Frequency: 491.087MHz)

Fig.15:- Utilization summary for AWP

Comparison and analytical study:-

The entire proposed system stated as AWP, is executed using the Xilinx ISE 14.75 and the outputs are fetched. The outputs are compared with the help of utilization summary and synthesis reports.

Graph representations are made for comparison and displaying the comparative area, delay and frequency (throughput). Table 4 shows the practical comparison of the both systems using the physical values fetched after running the codes in Xilinx ISE 14.75 design suite. Fig. 16,17,18,19 are the graph representation done from the synthesis report and utilization summary.

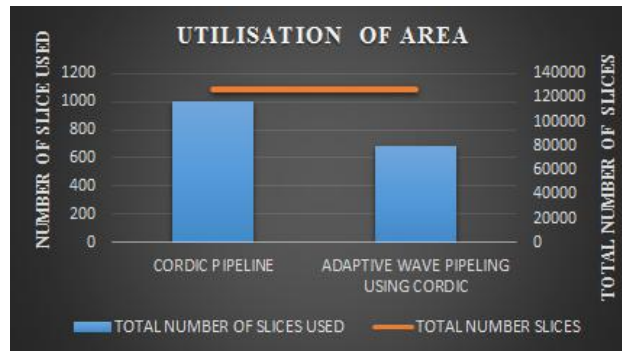


Fig. 16:- Graph representation of area utilization

Inference: From the above graph it is clear to knowledge that the number of slices used by AWP is lesser than the number of slices used by CORDIC pipeline. Hence the area occupied by AWP is less compared to other method.

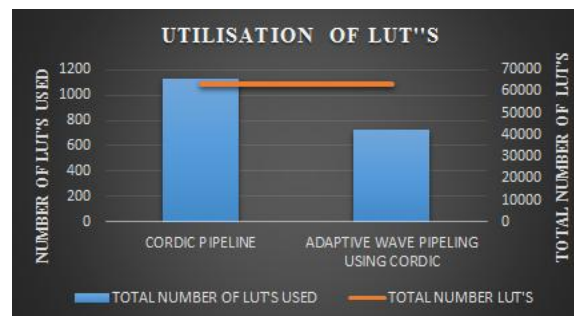


Fig. 17:- Graph representation of utilization of lut's

Inference: the above graph denoted the number of LUT's used by the two different processes. From the graph it is clear that the number of LUT's used by AWP is lesser than the comparative method.

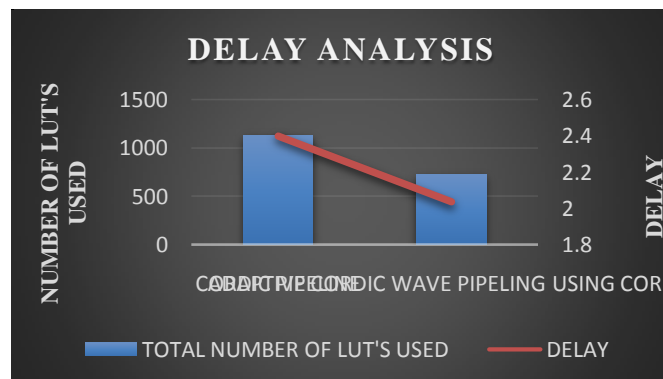


Fig. 18:- Delay analysis

Inference: the above graph is plotted between number of slice LUT's used vs delay. From the previous graph representation it is clear that CORDIC pipeline uses more LUT's than AWP. Hence the delay acquired for CORDIC pipeline is more than the proposed system that is AWP.

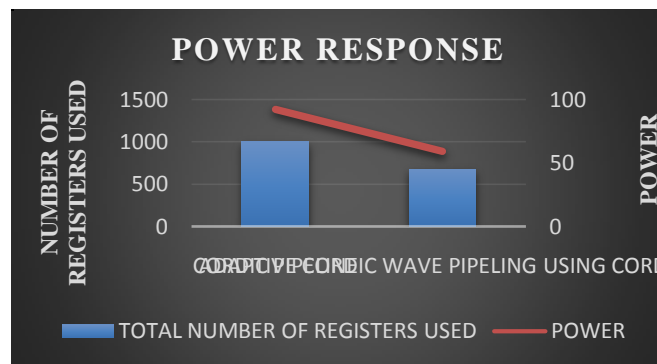


Fig. 19:- Power response.

Inference: the above graph is plotted between number of registers used vs power. Since the number of registers used is less in AWP the power consumed in this method is less. This is been analyzed by plotting the graph

Table 4:- Physical value comparison.

FACTORS	CORDIC PIPELINE	ADAPTIVE WAVE PIPELINING WITH CORDIC
NUMBER OF SLICE REGISTERS USED	1000/126800 UTILISATION-0%	680/126800 UTILISATION-0%
NUMBER OF SLICE LUT'S USED	1129/63400 UTILISATION-1%	723/63400 UTILISATION-1%
FREQUENCY	416.98Mhz	491.087Mhz
DELAY	2.398ns	2.036ns
POWER	$1129 \times .082 = 92.578W$	$723 \times .082 = 59.286W$

Conclusion:-

The entire proposed system is designed and executed using Xilinx. The function is designed in such a manner the end users can modify them according their requirements and application. Since FPGA is chosen as target technology the design has resulted in consumption of low area, power with increased speed and throughput.

The entire project work is focused on designing of efficient DDS to improve the efficiency of DDC and improve the overall performance of SDR. In future the entire DDC structure can be designed with the developed AWP technique and better efficient SDR structure can be evolved.

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RESEARCH ARTICLE

A STUDY ON RELATIONSHIP BETWEEN ERP IMPLEMENTATION AND MANAGEMENT ACCOUNTING PRACTICES.

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Abstract

With ever changing global scenario in business sector, uses of ERP (Enterprise resource Planning) to cut down on accounting cost have gained a rapid growth. Not only large enterprises, but medium as well as small enterprises have gone on the path of ERP implementations. Using an ERP in your accounting, not only cuts down the time consumption in maintaining accounting records, but also helps in managerial accounting and decision making with ready to use real time data on click of a button. This paper has been analyzed on impact of ERP implementations on the management accounting process on different kinds of industries. Previous study talks about many references to factors underlining successful ERP implementations with reference to plummeting management accounting decision making burden. Multiple regression analysis has been done on the basis of factors and methods of management accounting extracted from review of literature. The focus of this paper is to give a scenario for further research with respect to impact on management accounting as a result of ERP implementations.

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Introduction:-

With the rapid advancement of ERP systems in LE's and SME's the management has got sheer benefits of quick decision making process with the help of readily available data on click of a button. But does implementation of an ERP into a company's environment actually affects the process of Management Accounting.

ERP is a planning philosophy enabled with software that attempts to integrate all the business processes of all the departments and functions across a company on to a single computer system that can serve particular needs of the different departments. ERP combines all the business requirements of the company into a single, integrated software program that runs of a single database so that the various departments can more easily share information and communicate with each other. Fig.1 shows ERP integrated system. (Altekar, 2009)

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Figure 1:- ERP Integrated Systems.

Management Accounting is a process of identifying, measuring, analyzing, interpreting and communicating information in pursuit of an organization's goals. Management accounting is an integral part of management process, and managerial accountants are important strategic partners in the organization's management team. In pursuing its goals, an organization acquires resources, hires people and then engages in an organized set of activities. It is up to management team to make the best use of organizations resources, activities and people in achieving the organization's goals. The management accounting has four activities involved:

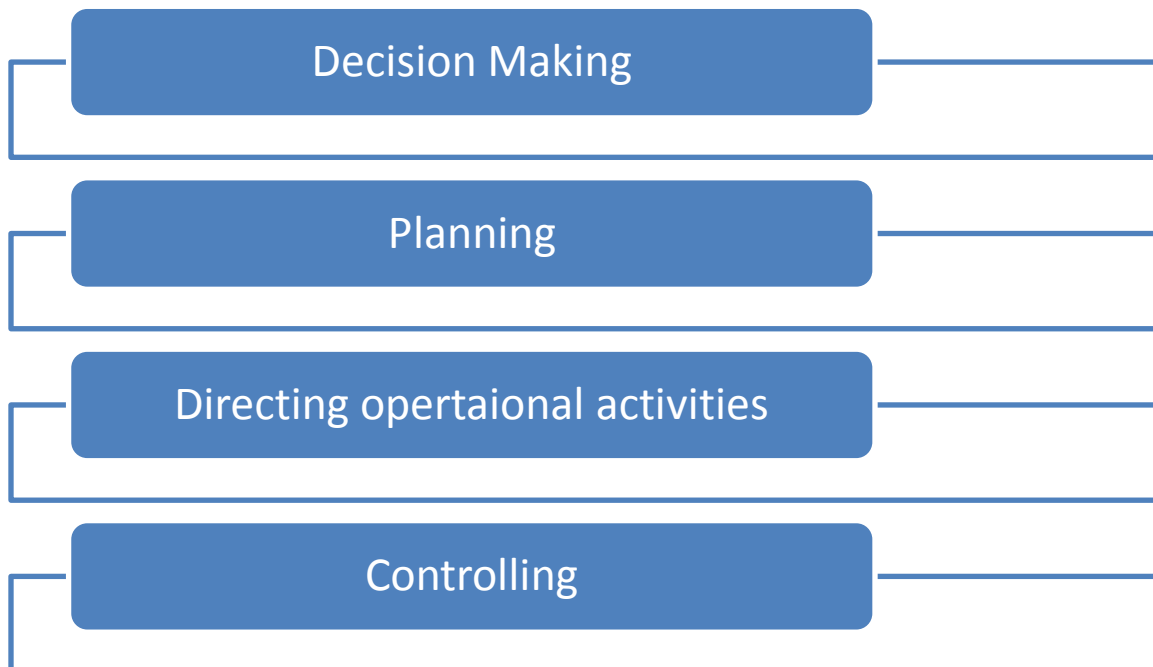


Figure 2:- (Ronald W Hilton, 2008)

Management Accounting techniques which are usually followed in any LE or SME are:-

1. Activity based costing
2. The balanced score card
3. Customer Satisfaction surveys
4. Target Costing
5. Life cycle costing
6. Benchmarking
7. Non-Financial key performance Indicators
8. Others

The combination of ERP and Management accounting does brings changes in a company's day to day activities. Management accounting relies on combined data of all the departments combined together to attain a common decision making goal. With the help of ERP this can be done easily as the data is readily available. Now the hypothesis arise as to whether implementing an ERP can be beneficial to a company or it can bring heavy costs in implementation as compared to easy data availability to the management.

Objective of Research and Hypothesis:-

The research has been done with an objective of analyzing the post ERP implementation effect of management accounting processes. The major objectives of research are listed below:

1. To find out the impact of ERP implementation on the organizations overall management accounting process.
2. To find whether management accounting processes are related to size of organization in an ERP environment.
3. To find the impact of Management processes on different factors of better performance in an ERP environment.

The hypothesis of research is as follows:**. Objective 1:- Hypothesis**

H_{a0}: ERP implementation has a positive impact on management accounting Processes.

H_{a1}: ERP implementation does not bring a positive impact on management accounting processes.

Objective 2:- Hypothesis

H_{b0}: To find whether management accounting processes are related to size of organization in an ERP environment.

H_{b1}: There is no relation between management accounting processes to size of organization in an ERP environment.

Objective 3:- Hypothesis

H_{c0}: All management process has similar impact in an ERP environment

H_{c1}: All management process has different impact in an ERP environment.

Review Of Literature:-

The deployment of ERP systems generally results in significant reductions in the routine tasks of management accountants [(Lowe, 2004); (Arnold, 2006); (Drury, 2008)] and the subsequent a transition in their role from one of information gatherer or transaction-orientated role, to one of information analyst or business orientated role [(Granlund M. a., 1998); Hunton, 2002] or more simply from the back office to the front office (Holtzman, 2004). Furthermore, management accounting practices, whether traditional or modern, may become more efficient and effective when supported with ERP systems (see, for example, Edwards, 2001; Baxendale and Jama, 2003; Lea and Min, 2003; Scapens and Jazayeri, 2003; He, 2007; Lea, 2007). This is possible through greater information integration, greater flexibility in information access, and superior functionality (Booth et al., 2000). More recently, the enrichment of ERP systems with new ES, such as business intelligence (BI) systems, appears to have had a significant boost to the role of management accounting. BI systems normally sit on top of ERP systems, and are intended to bridge transactional efficiency with strategic planning. BI comprise a set of tools used for data analysis, query and reporting (Elbashir et al., 2008) and supporting strategic decision-making (Fahy, 2001; Bucher et al., 2009; Mikroyannidis and Theodoulidis, 2010) by providing managers with insights into their business operations (Seah et al., 2010). BI systems are composed of three complementary and interrelated tools, namely data warehousing, online analytical processing (OLAP), and data mining. As Teorey et al. (2006) describe, data warehousing deals with the storage, maintenance and retrieval of historical data; OLAP provides quick answers to ad hoc queries; and data mining is a collection of algorithms which find patterns in the data and return valuable user information. (Nikolaos Vakalfotis, July 2011)

(Gullkvist, Sept, 2013) Says empirical findings demonstrate a statistically significant influence of a successful ERP implementation as having a direct effect on changes in MA practices. While Sangster et al. (2009) identified increased data quality, improved decision-making and changes in accountants tasks under successful ERP implementations, this study adds to those findings by emphasizing change in MA techniques as associated with successful ERP implementation. Consistent with reasoning in prior research (Granlund and Malmi, 2002), the results of this study indicate that time since ERP adoption is a significant determinant of changes in MA techniques, but also that the majority of the changes occur within eight years after ERP implementation. Further, late ERP adopters perceive greater changes in MA techniques than early adopters. One possible explanation could be that late adopters perceive the changes as greater because of the short time since ERP implementation. Another explanation could be that newer software versions include features and functionality that were missing from their predecessors, and which could be driving the changes already at the ERP implementation stage. Some respondents maintained in an open question in the questionnaire that change in MA techniques occurred simultaneously with the ERP implementation, not afterwards. Furthermore, the findings indicate that the scope of installation, local or global, is a significant determinant of changes in MA techniques. This is an important finding, as many organizations in recent years have pursued global ERP implementation. As many previous studies on the attributes of MA change have been conducted at an early stage of ERP implementation and early in the first decade of the century, this study offers an update, and also indicates that change may occur over time.

Where (Hyvonen, 2003) gave a comparison of ERP: Enterprise Resource Planning and BoB: Best of breed with respect to management accounting systems. He collected data through 300 postal questionnaires from large and middle sized industrial units in Finland. The questionnaire addressed questions concerning IS implementation (why, how and by what the IS project was introduced), management accounting function, and the use of advanced management accounting techniques. The results obtained indicate that financial departments have been more interested in traditional BoB systems, while other departments have concentrated more on ERP solutions. Further, as the articulated motives behind the IS project were strategic, and moreover technical in nature, the solution in most cases was ERP, while in the cases where motives were either strategic or technical, the choice was BoB. Otherwise, there were no statistically significant differences between the groups of BoB or ERP adopters, and the problems perceived in management accounting or the adoption of advanced management accounting techniques (e.g. ABC, ABM and BSC).

Two seminal studies are (Granlund M. a., 2002) and (Scapens, 2003). Granlund and Malmi (2002) undertook exploratory field studies at ten firms. They recognized that ERP systems provided easier and faster access to standardized operational data, enhanced forecasting, emphasized the accounting department as the "nerve center", reduced the need for accountants to handle routine tasks, and gave accountants more time for sophisticated analyses. Nevertheless, they concluded (Granlund and Malmi, 2002, p. 309) "that there has been no major direct or indirect impact so far on management accounting and management control systems". Granlund and Malmi (2002) appear to be assessing the impact of ERP systems on management accounting from the third or information level, rather than the physical and transactional levels, in recognizing the greater ease and speed in accessing standardized data and in recognizing that the work of management accountants had shifted towards more analysis and fewer routine tasks. In recognizing better information, they did not mention that significant changes to transaction processing were necessary for better information. Moreover, they did not mention the standardization of the state-of-the-art physical processes that were necessary to support improved transaction processing.

(Rohde, 2006) Had done a paper to contribute to the body of knowledge about to what extent integrated information systems, such as ERP and SEM systems, affect the ability to solve different management accounting tasks. They made a questionnaire survey with 349 responses and data so collected was analyzed using linear regression models. Analyses indicate that ERP systems support the data collection and the organizational breadth of management accounting better than SEM systems. SEM systems, on the other hand, seem to be better at supporting reporting and analysis. In addition, modern management accounting techniques involving the use of non-financial data are better supported by an SEM system. This indicates that different management accounting tasks are supported by different parts of the IIS. On the basis of the findings, there is a need to consider the potential of closer integration of ERP and SEM systems in order to solve management accounting tasks.

(Sánchez-Rodríguez & Spraakman, 2012) Studied eight international scientific journals published between 1990 and 2010 to relationship between ERP and Management Accounting. They found on the basis of conceptual study that

not much work has been done in ERP and MA as such. They also concluded that SAP has a major role when it comes to ERP in Management Accounting decision making. They recommended area for further research in this field as not much work has been done on following:

- What are the critical context factors for implementation of ERP system?
- Analysis and interaction between ERP and organizational structure.
- Impact of ERP on management accounting and management accountant.
- The integration of BI, Cloud computing and ERP.

(Malinic & Todorovic, 2012) Talks about the effect of SAP on management accounting in Serbian industrial enterprises. They studied 9 enterprises from different industry to explore the relationship between SAP, as only one of the ERP software and management accounting changes. They concluded the impact of SAP on MA is modest in relation to expectation "Granlund and Malmi (2002)". Also they concluded that there is no causal relationship between ERP implementation and management accounting procedures. They suggested certain remarkable points to be considered while comparing MA and ERP:

- SAP implementation is complex at initial stage.
- SAP implementation is time consuming and it takes a lot of time to implement in all the sectors of the organizations.
- Time gap between implementation and go live is another important factor which influences the decision.
- Wrong decision in ERP tie-ups are also a reason of failure.
- SAP brings in time effectiveness in managerial decision making.

Again (Sangster, Leech, & Grabski, 2009) also found out by floating 700 questionnaires in different IT professionals that SAP was the dominate vendor of ERP implementations. On the basis of their research it has been identified that ERP implementations results in changes in the tasks of the management accountants, with the quality of many operational factors, such as inventory control, and the overall quality of data and information improving irrespective of the success or failure of the implementation. When an ERP implementation is successful, management accountants have time for other, less mundane activities; and their role becomes more enriching as a result.

Table 1:- Table showing Comparison of factors for performance measure of ERP and Management Accounting Relationship

Authors	(Gaertner & Feldbauer-Durstmüller, 2012)	Hyvonen (2003)	(Malinic & Todorovic, 2012)	(Sangster, Leech, & Grabski, 2009)	(Sangster, Leech, & Grabski, 2009)	(Malinic & Todorovic, 2012)	(Nikolaos Vakalofotis, July 2011)	(Gullkvist, Sept, 2013)	(Rohde, 2006)	Summary
Factors										
corporate size	GS		GS	GS						LS
corporate strategy,	GS	GS			GS		LS			LS
Organizational Structure	GS		GS	GS	GS	GS	GS		GS	GS
Management Accounting & Management Accountants	GS	GS	GS		GS	GS	GS	GS	GS	GS
Integration of business intelligence	GS	GS	GS	GS	GS	GS	GS	GS	GS	GS
Cloud Computing	LS									LS
Performance Measurement	LS	GS	GS	GS	GS	GS				GS
Cost Accounting	LS	GS				GS	GS		GS	GS
Data Analysis	LS	GS		GS		GS	GS	GS	GS	GS
Reporting	GS	GS		GS		GS			GS	GS
Budgeting	LS	GS	GS	GS		GS	GS		GS	GS

Note:-

GS= Good Support for inclusion as a performance measures in ERP and Management Accounting Relationship. (Performance measure referenced in 50% or more of studies in the table).

LS= Low support for inclusion as a performance measures in ERP and Management Accounting Relationship. (Performance measure referenced in less than 50% of studies in the table).

Table 2:- Table Showing Methods of Management Accounting Implemented / Accepted in studies under review

Authors	(Gaertner & Feldbaue r-Durstmueller, 2012)	Hyvonen (2003)	(Malini c & Todovic, 2012)	(Sangster, Leech, & Grabski, 2009)	(Sangster, Leech, & Grabski, 2009)	(Malini c & Todovic, 2012)	(Nikolaos Vakalof, July 2011)	(Gullkvist, Sept, 2013)	(Rohde, 2006)	Summary
Methods										
Activity Based Costing	GS	GS	GS	LS	GS	GS	LS	GS	GS	GS
Balance Score Card	GS	GS	LS	LS	GS	GS	LS	GS	GS	GS
Target Costing	GS	GS	LS	LS	GS	GS	LS	GS	GS	GS
Lifecycle Costing	GS	GS	LS	LS	GS	GS	LS	GS	GS	GS

Note:-

GS= Good Support for inclusion as a performance measures in ERP and Management Accounting Relationship. (Performance measure referenced in 50% or more of studies in the table).

LS= Low support for inclusion as a performance measures in ERP and Management Accounting Relationship. (Performance measure referenced in less than 50% of studies in the table).

Based on the review of literature it can be ascertained that Organizational Structure, Management accounting and accountants, integration of business intelligence, Performance measurement of management decisions, cost accounting, data analysis, reporting, budgeting etc. are good support for measuring the relationship between ERP and Management Accounting while factors like corporate size, strategy, cloud computing etc. are low support factors with respect to it. As far as methods of management accounting are concerned, most of the papers under review suggested that all the methods so recognized under table 2, are equally been taken under post ERP systems for management accounting and control.

How Does Enterprise Resource Planning And Management Accounting Are Interrelated:-

Enterprise resource planning is one of the software tools which make an organization's data management easy and handy. An organization with too many departments each more or less linked to finance and accounting department makes it difficult to extract data for managerial decision making in manual accounting systems. With introduction of ERP large industries like Big Organizations like Reliance, Essar, Tata, L & T, Coca Cola, Adani, Bombay Dyeing, Amul Birla etc. which are widely spread all over the globe and has offices/ plants everywhere has got a benefit of easy data access with click of a button. ERP helps a management accountant to make decision on the basis of MIS data which is available on his table through software popularly known as ERP.

The relationship between management accounting and ERP can be summaries on the following basis:-

- ERP implementations are huge and are time consuming. If an ERP implementation takes more time than its estimated one, it becomes costly and has a negative impact on management decision making.
- ERP can give all the past data but cannot give future data to make a 100% accurate decision on the basis of it. Budgets prepared on ERP are based on historical data. This cannot be considered accurate when it comes to non-accounting factors influencing managerial decision.
- For Activity based costing, customer satisfaction survey, non-financial key performance indicators etc. kinds of management accounting processes only ERP cannot suffice the requirement of decision making. It needs human intervention (That of management accountant) and outside data (That of customers or competitors act) to make decisions.

- ERP like SAP which is very user's friendly and can be implemented with minimal customization is widely accepted by LE's. But for SME's, an ERP like SAP is more costly and implementing any other cheaper ERP makes it very difficult to extract managerial accounting data for the purpose of decision making.
- First time adaptability of any ERP into a manual accounting firm is very difficult. Getting a user's acceptance on ERP is an issue for vendors. At the same time from implementation to first positive use, there is a time lag which makes it difficult for management accountant to make decision and use an ERP.
- With global competitive environment ERP has become a fashion rather than a necessity. Companies often implement ERP just because their competitors are doing so, without acknowledging their budgets and requirements. These kinds of implementations also hampers the quality of decision making in management accounting when an inappropriate software is implemented within organization to hamper day to day working.

Research Methodology:-

The purpose of this paper is to investigate the research done in the field of ERP and management accounting. For this, the database of established scientific management accounting journals and information system journals were investigated. The list of journals under the study was as follows:

Accounting Journals	Information system journals
<ul style="list-style-type: none"> • Management Accounting Journal (MAJ) • Journal of Management (JM) • European Accounting Review (EAR) • International Journal of Economic Science and applied research • Auditing and Accountability Journal. 	<ul style="list-style-type: none"> • International journal of accounting information systems. • Journal of Enterprise Information Management • Journal of Information System & Technology Management

These journals are chosen for the purpose of extracting the review of literature also the methodologies and extract on account of inter-relation between ERP and Management accounting.

The method considered under the research was to recognize the most reviewed or analyzed methods for the purpose of determining relationship in ERP and Management accounting. Based on the literature review factors of testing the inter-relationship of ERP and management accounting were identified as Organizational structure, Management accounting and management accountant, business intelligence, performance of firm, cost accounting procedures, data analysis, reporting and budgeting.

Also, four management accounting techniques viz., Activity Based Costing (ABC), Balance Score Card (BSC), Target Costing (TC) and Life cycle costing (LC) were also identified to find implications of these management accounting practices in an ERP environment.

Research design and Data Collection:-

A five point likert scale questionnaire was developed to find users view on management accounting process in ERP environment. The questionnaire was sent to 70 managers / users of ERP in different type of manufacturing organizations have been given these questionnaires to be filled for the purpose of data analysis. 40 users /managers responded and on the basis of the data so collected multiple regression analysis was to analyses the data.

Data Analysis and findings:-

The questionnaire so received from the sources only 30 were considered for ANOVA .10 responses were rejected on the grounds of either not completely filled in or filled by a non-user. The data analysis was done with the help of ANOVA using excel from data analysis option.

The data collected was segregated and drafted in excel and the ANOVA was generated as follows:

Objective 1:- To find out the impact of ERP implementation on the organizations overall management accounting process

Multiple regression analysis was done for to check all the Management Accounting techniques relations to factors of performance evaluation in an ERP environment. The data gave following results.

Regression Statistics	
Multiple R	0.76
R Square	0.57
Adjusted R Square	0.50
Standard Error	0.88
Observations	29.00

ANOVA	df	SS	MS	F	Significance F
Regression	4	25.07	6.27	8.03	0.00
Residual	24	18.73	0.78		
Total	28	43.79			

	Coefficients	Standard Error	t Stat	P-value	Lower 95%	Upper 95%	Lower 95.0%	Upper 95.0%
Intercept	0.618	0.495	1.248	0.224	-0.404	1.641	-0.404	1.641
ABC	3.109	1.108	2.805	0.010	0.822	5.396	0.822	5.396
BSC	2.000	0.883	2.264	0.033	0.177	3.823	0.177	3.823
TC	-3.301	1.284	-2.571	0.017	-5.950	-0.651	-5.950	-0.651
LC	-1.192	0.629	-1.896	0.070	-2.489	0.106	-2.489	0.106

The P-value when more than 0.05 needs to be excluded from the list of independent variables (ABC, BSC etc.). As P value 0.05 or greater says that the predicted value of the variable is as such it is insignificant for the analysis of the outcome. As lifecycle costing is the independent variable whose P- value is more than 0.05, it means that the variable is insignificant for the purpose of analysis.

Checking on Significance of f which is coming to 0.000294188 shows that the variables so considered in the analysis is although below 0.05 but is significant for the analysis.

Looking at coefficient we can see again LC (-1.191881919) is totally insignificant along with TC (3.300738007) which is in negative. This show that on the basis of data so collected the only significant management accounting practice post ERP adoption are ABC and BSC. This analysis can also change as the number of users are too less as compare to world ERP using organization.

Here on the basis of above analysis we partially accept H_{a0} : ERP implementation has a positive impact on management accounting Processes.

Objective 3:- To find the impact of Management processes on different factors of better performance in an ERP environment.

Where, analysis of each management practice under study like ABC, BSC, TC and LC individually w.r.t the factors of performance evaluation gave another picture as:

Multi regression analysis of Activity costing (Dependent Variable) to Factors of performance evaluation (Independent variables)

Regression Statistics	
Multiple R	0.774
R Square	0.598
Adjusted R Square	0.438
Standard Error	1.068
Observations	29.000

ANOVA					
	<i>df</i>	<i>SS</i>	<i>MS</i>	<i>F</i>	<i>Significance F</i>
Regression	8	34.01	4.25	3.73	0.01
Residual	20	22.82	1.14		
Total	28	56.83			

	<i>Coefficients</i>	<i>Standard Error</i>	<i>t Stat</i>	<i>P-value</i>	<i>Lower 95%</i>	<i>Upper 95%</i>	<i>Lower 95.0%</i>	<i>Upper 95.0%</i>
Intercept	-0.61	1.00	-0.61	0.55	-2.69	1.47	-2.69	1.47
Organizational Structure	0.20	0.19	1.05	0.30	-0.20	0.60	-0.20	0.60
MA	0.14	0.21	0.68	0.50	-0.30	0.59	-0.30	0.59
BI	0.13	0.22	0.59	0.56	-0.33	0.60	-0.33	0.60
Perf. Man	0.36	0.18	1.99	0.06	-0.02	0.73	-0.02	0.73
Cost Acc	0.07	0.16	0.41	0.69	-0.27	0.40	-0.27	0.40
Data Analysis	-0.26	0.18	-1.42	0.17	-0.64	0.12	-0.64	0.12
Reporting	0.06	0.16	0.38	0.70	-0.27	0.39	-0.27	0.39
Budgeting	0.69	0.21	3.36	0.00	0.26	1.12	0.26	1.12

Again when we check the analysis data, P-value of most of the variables except data analysis and performance management are more than 0.05 i.e., totally insignificant. It shows that as far as ABC analysis is concerned, the performance of management accounting in post ERP companies is mostly likely to be depending on whether the organization is able to do better data analysis of the records in ERP.

A significance *f* on the contrary shows that independent variables are significant and ABC in an ERP environment can be a significant measure for the analysis.

Multi regression analysis of Balance Score card (Dependent Variable) to Factors of performance evaluation (Independent variables)

<i>Regression Statistics</i>	
Multiple R	0.678
R Square	0.460
Adjusted R Square	0.244
Standard Error	1.217
Observations	29

ANOVA					
	<i>df</i>	<i>SS</i>	<i>MS</i>	<i>F</i>	<i>Significance F</i>
Regression	8	25.22	3.15	2.13	0.08
Residual	20	29.60	1.48		
Total	28	54.83			

	<i>Coefficients</i>	<i>Standard Error</i>	<i>t Stat</i>	<i>P-value</i>	<i>Lower 95%</i>	<i>Upper 95%</i>	<i>Lower 95.0%</i>	<i>Upper 95.0%</i>
Intercept	-0.07	1.14	-0.06	0.95	-2.44	2.30	-2.44	2.30
Organizational Structure	0.15	0.22	0.69	0.50	-0.31	0.61	-0.31	0.61
MA	0.20	0.24	0.83	0.42	-0.30	0.70	-0.30	0.70
BI	0.11	0.25	0.42	0.68	-0.42	0.64	-0.42	0.64
Perf. Man	0.25	0.20	1.22	0.24	-0.18	0.68	-0.18	0.68
Cost Acc	0.33	0.18	1.79	0.09	-0.05	0.71	-0.05	0.71
Data Analysis	-0.26	0.21	-1.25	0.22	-0.69	0.17	-0.69	0.17
Reporting	0.05	0.18	0.27	0.79	-0.33	0.42	-0.33	0.42
Budgeting	0.42	0.23	1.78	0.09	-0.07	0.91	-0.07	0.91

Looking at the P value many factors like Organizational structure, management accounting & accountant, performance management, Cost accounting, data analysis and budgeting are significant factors for analysis of balance score card as a good practice in firms using ERP.

Multi regression analysis of Target Costing (Dependent Variable) to Factors of performance evaluation (Independent variables)

<i>Regression Statistics</i>						
Multiple R	0.745746942					
R Square	0.556138501					
Adjusted R Square	0.378593901					
Standard Error	1.023929427					
Observations	29					
ANOVA						
	<i>df</i>	<i>SS</i>	<i>MS</i>	<i>F</i>	<i>Significance F</i>	
Regression	8	26.27274987	3.284093733	3.13238759	0.018228166	
Residual	20	20.96862945	1.048431472			
Total	28	47.24137931				

	<i>Coefficients</i>	<i>Standard Error</i>	<i>t Stat</i>	<i>P-value</i>	<i>Lower 95%</i>	<i>Upper 95%</i>	<i>Lower 95.0%</i>	<i>Upper 95.0%</i>
Intercept	-0.34	0.96	-0.36	0.72	-2.34	1.65	-2.34	1.65
Organizational Structure	0.14	0.18	0.75	0.46	-0.25	0.52	-0.25	0.52
MA	0.12	0.20	0.58	0.57	-0.30	0.54	-0.30	0.54
BI	0.15	0.21	0.70	0.49	-0.30	0.59	-0.30	0.59
Perf. Man	0.18	0.17	1.04	0.31	-0.18	0.54	-0.18	0.54
Cost Acc	0.22	0.15	1.46	0.16	-0.10	0.55	-0.10	0.55
Data Analysis	-0.06	0.17	-0.37	0.71	-0.43	0.30	-0.43	0.30
Reporting	0.07	0.15	0.48	0.63	-0.24	0.39	-0.24	0.39
Budgeting	0.53	0.20	2.70	0.01	0.12	0.94	0.12	0.94

Accept for budgeting no other factors shows a significant P value for the purpose of analyzing target costing as a significant dependent variable on the independent variables for the purpose of analysis of variance. A 0.018228166 of "Significance f" make the analysis significant for the purpose of performance evaluation.

Multi regression analysis of Lifecycle Costing (Dependent Variable) to Factors of performance evaluation (Independent variables)

Regression Statistics								
Multiple R	0.683918904							
R Square	0.467745067							
Adjusted R Square	0.254843094							
Standard Error	1.229771586							
Observations	29							
ANOVA								
	<i>df</i>	<i>SS</i>	<i>MS</i>	<i>F</i>	<i>Significance F</i>			
Regression	8	26.58082313	3.322602892	2.196997334	0.073398625			
Residual	20	30.24676307	1.512338154					
Total	28	56.82758621						
	<i>Coefficients</i>	<i>Standard Error</i>	<i>t Stat</i>	<i>P-value</i>	<i>Lower 95%</i>	<i>Upper 95%</i>	<i>Lower 95.0%</i>	<i>Upper 95.0%</i>
Intercept	-0.71	1.15	-0.62	0.54	-3.11	1.68	-3.11	1.68
Organizational Structure	0.36	0.22	1.63	0.12	-0.10	0.82	-0.10	0.82
MA	0.41	0.24	1.69	0.11	-0.10	0.92	-0.10	0.92
BI	0.15	0.26	0.60	0.56	-0.38	0.69	-0.38	0.69
Perf. Man	0.35	0.21	1.67	0.11	-0.09	0.78	-0.09	0.78
Cost Acc	0.28	0.18	1.51	0.15	-0.11	0.67	-0.11	0.67
Data Analysis	-0.15	0.21	-0.73	0.47	-0.59	0.28	-0.59	0.28
Reporting	-0.09	0.18	-0.49	0.63	-0.47	0.29	-0.47	0.29
Budgeting	0.35	0.24	1.46	0.16	-0.15	0.84	-0.15	0.84

A Significance F 0.073398625 shows that again the dependent variable has a significant relationship with the independent variables. Although most of the factors again have a P-value which is below 0.05 recommending that target costing is being taken care in ERP environment as a management accounting practice.

On the basis of above we can again accept H_{b0} : As out of four three of the Management processes can be considered as significant in ERP environment on the basis of factors of performance.

Objective 2:- To find whether management accounting processes are related to size of organization in an ERP environment.

Regression Statistics						
Multiple R	0.2816007					
R Square	0.079298954					
Adjusted R Square	-0.07415122					
Standard Error	1.475603287					
Observations	29					
ANOVA						
	<i>df</i>	<i>SS</i>	<i>MS</i>	<i>F</i>	<i>Significance F</i>	
Regression	4	4.500899256	1.125224814	0.516773307	0.724121854	
Residual	24	52.25772143	2.17740506			
Total	28	56.75862069				

	Coefficients	Standard Error	t Stat	P-value	Lower 95%	Upper 95%	Lower 95.0%	Upper 95.0%
Intercept	3.72	0.83	4.49	0.00	2.01	5.44	2.01	5.44
ABC	-0.09	0.33	-0.28	0.78	-0.78	0.59	-0.78	0.59
BSC	0.14	0.35	0.39	0.70	-0.59	0.87	-0.59	0.87
TC	0.01	0.41	0.02	0.98	-0.85	0.87	-0.85	0.87
LC	-0.31	0.37	-0.83	0.41	-1.08	0.46	-1.08	0.46

Looking at the regression analysis of the data on the basis of four practice of management accounting a standard error of 1.47 and a Significance “f” of 0.724121854 (which is above 0.05) shows that the size of firm is not being significantly related to management practice. Also the P-value of all the independent variables is more than 0.05 which signifies that none of the factor has any significant relationship with type of organization and hence we reject the null hypothesis and select the alternative hypothesis.

H_{b1}: There is no relation between management accounting processes to size of organization in an ERP environment.

Conclusion and Recommendations:-

This small empirical study on ERP and management accounting has given an area for further research as to how implementation of ERP has an impact on management decision making. So far from the empirical study it can be concluded that ERP when implemented gives a very positive impact on management accounting as to decision making in the field of production, make or buy decisions, BOM, supply chain management, outsourcing, etc. ERP's like SAP, PeopleSoft, MSD and others needs to be studied in detail industry wise to comment upon the acceptance as to management accounting. Although ERP has gain rapid advancement in last decade, it is still an unread book for researchers with the prospective of end users eyes. **Thus, although we accept out hypothesis as to “there is a positive relationship between ERP implementations and Management accounting”**, but at the same time we also had restricted this paper to a small group of users/ managers and the research analysis and outcomes can come to total contradiction when it comes to the huge sample in different types of companies using ERP. As this research is primary investigation opening up the doors for a broader frontier, it is highly recommended that before coming to any conclusion factors like type of ERP for MA processes, many other kind of management practices and large pool of end users view should be considered for the higher end research.

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Abbreviations:-

ABC : Activity Based Costing

BSC: Balance Score Card

LC: Lifecycle Costing

TC: Target Costing

ERP: Enterprise Resource planning

MA: Management Accounting.



RESEARCH ARTICLE

COMPARATIVE EVALUATION OF EFFICACY OF TWO COMMERCIALLY AVAILABLE HERBAL AND ALLOPATHIC DENTIFRICE ON DENTINAL HYPERSENSITIVITY AND THEIR EFFECT ON PLAQUE AND GINGIVITIS.

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Abstract

Background: Dentinal hypersensitivity is a transient condition that often resolves with the natural sclerotic obturation of dentinal tubules. A potent topically applied in-office desensitizing treatment is indicated as the choice of treatment when dentinal hypersensitivity is localized to one or two teeth.

Aim: The present study aimed to evaluate and compare the clinical efficiency of two commercially available herbal and allopathic dentifrices that were used in treating dentinal hypersensitivity, additionally this study evaluates their effectiveness in reducing plaque and gingivitis on both the groups.

Materials and Methods: 30 patients aged 20-60 years reporting with dentinal hypersensitivity were randomly assigned to 2 groups of 15 patients each. Plaque index scores and gingival bleeding index scores were evaluated and response to air stimuli and thermal stimuli were measured using visual analogue scale at baseline, 7th, and 30th day.

Statistical Analysis: Statistical analysis was carried out using Mann Whitney u test, Friedman test and Wilcoxon sign rank test.

Results: Allopathic dentifrice group showed better results in reducing dentinal hypersensitivity during 7 days assessment whereas at 30 days of evaluation herbal dentifrice group showed significant reduction in dentinal hypersensitivity. It also showed that there was no statistical significant difference between both the groups but herbal dentifrice showed better results in gingival index scores and plaque index scores.

Conclusion: Allopathic dentifrices is fast acting whereas herbal dentifrice has long term effect and has better anti plaque and anti gingivitis effect compared to allopathic dentifrice.

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Introduction:-

Dentinal hypersensitivity (DH) is a characteristic pain arising from exposed dentine, typically in response to various stimuli such as thermal, evaporative, tactile, osmotic or chemical, which cannot be attributed to any other form of dental defect or pathology.¹

When constantly present over a long time, DH can provoke chronic discomfort and emotional distress.² In addition; it can make dental plaque control difficult. According to hydrodynamic theory by Braennstroem and Astroem,

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enamel or cementum loss in cervical areas and the consequent opening of dentinal tubules to the oral environment, under certain stimuli, allows the movement of dentinal fluid inside the tubules, indirectly stimulating the extremities of the pulp nerves, causing pain.³ It was also found that open dentinal tubules serve as pathways for the diffusive transport of bacterial elements in the oral cavity to the pulp, which may cause a localized inflammatory pulpal response.⁴ Microscopic examination reveals that patent dentinal tubules are more numerous and wider in hypersensitive dentine than in non-sensitive dentine.⁵

Various strategies have been used in the treatment of DH which include lasers, iontophoresis, dentine sealers and soft tissue grafting.⁶ Toothpastes are the most widely used dentifrices for delivering over-the-counter desensitizing agents.⁷ Agents for topical relief of dentine hypersensitivity either block the exposed dentine tubules or have a direct desensitizing effect on the pulpal nerve fibres.^{8,9} The use of potassium nitrate as an effective desensitizing agent dates back to the testimonial report of Hodosh in 1974.¹⁰ Since then, many pastes containing potassium chloride or potassium citrate have been made available.¹¹

Recently there has been a growing interest in natural products, and herbal based toothpastes have been found as effective as the conventionally formulated dentifrice in the control of plaque and gingivitis.¹²

This was the first study which was conducted to assess and compare the efficacy of a commercially available novel herbal dentifrice containing potassium nitrate and *Spinacia oleracea* to an allopathic dentifrice which contains potassium nitrate on DH and also to evaluate anti plaque and anti gingivitis effects of both the tooth pastes over a period of 4 weeks.

Methodology:-

A total of 30 patients were selected from department of periodontics and oral implantology, A.M.E'S Dental college and hospital, Raichur, Karnataka, India with dentinal hypersensitivity. The individuals selected at baseline had a history of DH caused by gingival recession, cervical erosion, and attrition. Subjects age ranging from 20-60 years were required to have 20 natural permanent teeth and at least two teeth with a VAS score of ≥ 4 . Allergic to ingredients, teeth with occlusal overload, teeth with caries, defective restorations, and chipped teeth were excluded.

After obtaining patient informed consent, oral prophylaxis with oral hygiene instructions was given. Randomisation was done to allocate patients in two groups, group A-Herbal dentifrice -HiOra K, (The Himalaya Drug Company Research and Development, Makali, Bangalore) and group B-Allopathic dentifrice-Sensodent K (Warren pharma). Each gram of Hi Ora K dentifrice contained 2.5 mg *Cinnamomum zeylanicum*, 2.5 mg *Syzygium aromaticum*, 10.0 mg *Spinacia oleracea*, 6.0 mg Triphala, 4.0 mg Trikatu and 30.0 mg Suryakshara (Potassium nitrate) and 10.0 mg Yashada bhasma (zinc oxide), sodium benzoate, calcium carbonate with sorbitol, glycerine and xanthan gum. Each gram of Sensodent-K contains 5% of potassium nitrate. Plaque index and gingival index were recorded and hypersensitivity response to air stimuli using air jet from triple syringe & thermal stimuli using cold water was evaluated on the VAS, at baseline. After recording sensitivity scores and plaque index and gingival index scores at baseline, subjects were randomly given dentifrice and advised to use it with a soft bristle toothbrush twice daily for 3 weeks. Patients were recalled after 1 week and 4 weeks for clinical assessment.

Statistical analysis:-

Statistical analysis was carried out using Mann Whitney u test and Friedman test to compare two individual independent groups at baseline, 1 week and 4 weeks. Wilcoxon sign rank test was used as post hoc test for intergroup comparison.

Results:-**Table I-** Percentage change in mean sensitivity scores for two groups at all time points for both measures of plaque scores, gingivitis scores and sensitivity score

Groups	Clinical assessment	Change in mean sensitivity score (%)		
		Baseline to 1week	Baseline to 4 week	1 to 4weeks
Group-A	Plaque scores	-88.63 ± 42.9	-139.26 ± 69.3	-51.9± 65.5
	Gingival scores	-97.93 ± 41.9	-174.33 ± 58.56	-76.4 ± 44.3
	Thermal stimulus	-100.2 ± 49.8	-267.2 ± 82.4	-167 ± 66.18
	Air stimulus	-111.2 ± 32.4	-255.8 ± 59.2	-144.6 ± 54.8
Group-B	Plaque scores	-78.16 ± 34.02	-130.1 ± 54.3	-50.63 ± 78.5
	Gingival scores	-83.26 ± 51.05	-147.73 ± 56.6	-88.73 ± 42.03
	Thermal stimulus	-115.53 ± 46.43	-231.81 ± 56.76	-116.26 ± 40
	Air stimulus	-118.2 ± 55.70	-229.13 ± 65.54	-110.9 ± 34.2

Table II:- Comparison of two groups of percentage change in plaque, gingivitis, air and thermal sensitivity scores.

		Baseline to 1 week	Baseline to 4 weeks	1 week to 4 weeks
Plaque scores	t value	0.740	0.402	-0.050
	P value	0.466	0.691	0.960
Gingival scores	t value	-0.86	-1.264	0.498
	P value	0.397	0.217	0.441
Evaporative Stimulus	t value	0.871	-1.369	-2.001
	P value	0.391	0.182	0.017*
Air Stimulus	t value	0.421	-1.169	-2.016
	P value	0.677	0.252	0.05*

*Statistically significant

Discussion:-

HiOra K dentifrice contains *Cinnamomum zeylanicum* that has been found to have bactericidal properties and is found to be equally effective against both Gram-positive and Gram-negative organisms. Cinnamaldehyde, the predominant active compound in cinnamon oil, has been found to be a natural antioxidant.¹³ Extract of *Syzygium aromaticum* (clove) has been found to exhibit growth-inhibitory activity against Gram-negative anaerobic periodontal oral pathogens, including *Porphyromonas gingivalis* and *Prevotella intermedia*.¹⁴ Triphala has been found to have similar effects on dental plaque and gingival inflammation compared to chlorhexidine.¹⁵ Trikatu has anti-inflammatory properties.¹⁶ Yashada bhasma contains zinc and has been formulated into oral health products to control plaque, reduce malodor and inhibit calculus formation.^{17,18}

In the present study dentinal hypersensitivity was reduced in allopathic dentifrice group during 7 days assessment whereas after 30 days herbal dentifrice group showed significant reduction in dentinal hypersensitivity. This means that desensitizing affect was maintained even 7 days after discontinuing the product in herbal dentifrice group, this might be due to the component *Spinacia oleracea* present in HiOra K group. *Spinacia oleracea* may have a possible mechanism of having a synergistic effect along with potassium nitrate in reducing DH by its dentinal tubule obliterating property, as *Spinacia oleracea* was absent in the group B there was not statistical difference at 30 days. It has been found that potassium nitrate does not diminish dentine hydraulic conductivity, or promote obstruction of dentinal tubules by the deposition of crystals. According to Wilchgers¹⁹ and Ermer²⁰ potassium nitrate has an effective desensitizing action. The increase in the concentration of extracellular potassium around the nerve fibers causes their depolarization, avoids repolarization and blocks the axonic action and passage of nerve stimulus, thus inactivating the action potential.^{19, 20} Hence it was shown that group B which contains 5 % of potassium nitrate showed rapid relief in hypersensitivity during 7th day of assessment. In another study which compared HiOra K with that of placebo, Sensitivity scores for controlled air stimulus and cold water were recorded at baseline, 6 weeks and 12 weeks and concluded that test group was found to be significantly better compared to the placebo group at the end of 6 and 12 weeks in reduction of dentinal hypersensitivity.²¹

As per our knowledge this is the first study which also compared the effectiveness of desensitizing agents on plaque and gingivitis and it has been shown that there was no statistical significant difference between both the groups, but HiOra K showed better results in gingival index scores and plaque index scores which might be due to the presence of triphala which has effect on dental plaque and gingival inflammation.

Conclusion:-

Allopathic dentifrices had shown significant reduction in dentinal hypersensitivity at 7 days but herbal dentifrice had shown better results at 1 month follow up, which may be due to presence of *Spinacia oleracea*. Effect of antiplaque and gingivitis was also shown better results in herbal dentifrice group. Therefore according to our study, allopathic dentifrices is fast acting whereas herbal dentifrice has long term effect and has better antiplaque and antigingivitis effect compared to allopathic dentifrice.

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RESEARCH ARTICLE

A STUDY ON EFFECT OF SELECTED ANTENATAL EXERCISES ON OUTCOME OF LABOUR AMONG PRIMIGRAVIDA MOTHERS - A LITERATURE REVIEW.

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Effect, Antenatal exercises, Outcome of labour, Primigravida mothers, Maternity services

Abstract

The researcher aims to assess the effect of selected antenatal exercises on outcome of labour among primigravida mothers. It was observed that most of the caesarean section and assisted deliveries occurred due to rigid cervix, prolonged labour and inability of the mother to cope up with the labour process and that affects both maternal and fetal outcome and further inquiry showed that the antenatal mothers lacked even basic knowledge about the importance of antenatal exercises. So it was concluded that there is association between antenatal exercises and labour outcome.

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Introduction:-

According to Global Health Observatory data given by World Health Organization, every day in 2015, about 830 women died due to complications of pregnancy and child birth. The risk of a woman in a developing country dying from a maternal-related cause during her lifetime is about 33 times higher compared to a woman living in a developed country.¹

According to UNICEF, globally about 800 women die every day of preventable causes related to pregnancy and childbirth, 20% of these women are from India. Annually, it is estimated that 55,000 women die due to preventable pregnancy related causes in India.²

To study the same, the researcher reviewed many literature and it was obtained through various database includes CINHALL (Cumulative index TO Nursing & Allied Health Literature), MEDLINE (Medical Literature Analysis & Retrieval System Online), PubMed, Science Direct, SpringerLink, ProQuest & Google scholar.

Material Methods & Findings:-

The study is headed mainly on the effect of antenatal exercises on outcome of labour among primigravida mothers attending maternity services.

A RCT was done to examine the efficacy of antenatal exercises on maternal and neonatal outcomes in elderly primigravida at the Physical Therapy Department of Bolak Abu-Ela General Hospital, Cairo, Egypt in 2013-14. 60 elderly primigravida at 14 weeks' gestation within the age range of 35-40 years were divided randomly into equal experimental and control group. Present Pain Intensity scale, partograph and APGAR score were the main tools of data collection. Participants of experimental group showed a statistically significant difference in the intensity of labor pain when cervical dilatation was between 7 and 8 cm ($P = 0.000$), duration of first stage of labor ($P = 0.026$),

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and neonates' APGAR scores at the first and the fifth minute of life ($P = 0.000$) compared with participants of control group. The study concluded that antenatal exercises are very effective in decreasing adverse effects and labor complications in elderly primigravida and their neonates.³

A cross-sectional study was conducted to find out effectiveness of antenatal exercise in facilitating normal labor and also other benefits associated with antenatal exercise during pregnancy in antenatal exercise facility of private clinic and government institution of Indore district in 2013-14. 200 females of age group of 20- 35 years who had recently delivered and did not have any medical/surgical/obstetric complications were selected through sequential sampling technique and equally divided in experimental and control group. A semi structured questionnaire was used as study tool for interview. The data was analyzed using appropriate statistical software (MS excel and SPSS version 20). Chi square test was applied for significance association between variable. The study showed that only 36% of control group females were delivered normally, as compared to 74% experimental group females. In experimental group only 9% females had urinary incontinence after delivery. Main reason for not doing antenatal exercise was not having enough time (42%) followed by not having enough knowledge (31%). Only 30% females of experimental group had the problem of backache, as compared to 63% control group females who suffered from same problem. ($X^2=21.887$, $p=0.001$, $RR=0.493$, $CI\ 0.356-0.683$). Thus the study concluded that females practiced antenatal exercise had less chances of caesarean section, back ache and urinary incontinence.⁴

A quasi-experimental control study was conducted to assess the effect of pelvic floor muscle exercise training protocol for pregnant woman during 3rd trimester on labor duration at El-Manial University Hospital, Cairo University, Egypt in 2013-14. 100 pregnant women were recruited by convenient sampling technique and equally divided in to experimental and control group. Experimental group received pelvic floor muscle training exercise whereas control group received only standard routine care. Structured interviewing tools, pelvic floor muscles strength assessment, antenatal follow up checklist, partograph were used as tools of data collection. The study showed that there was a highly significant statistical difference between the study and control groups in relation to duration of 1st, 2nd and 3rd stage of labor. There was a high statistically significant difference found for the mean cervical dilatation and effacement among the two groups. The study also showed that mothers in the study group had better labour progress than those in the control group. Thus the study concluded that pelvic floor muscle training exercise can be useful to decrease duration of labor.⁵

A cross-sectional study to analyze the knowledge, attitude and practice of exercise during pregnancy among antenatal mothers was conducted at Indira Gandhi Medical College and Research Institute, Puducherry, India in 2015. A total of 200 antenatal mothers who aged between 18-35 years were included in the study. Data were collected using a self-administered pretested close ended questionnaire. Data were analyzed using Statistical Package for Social Sciences (SPSS). The study showed that total mean knowledge score was 20.53 ± 2.08 . 51% felt it was necessary to do exercise during pregnancy. Only 18% were practicing exercise in pregnancy. The study concluded that the knowledge of women on exercise during pregnancy was less than average, and their attitude was favorable. However a very few were actually practicing exercises in pregnancy.⁶

A quasi-experimental study to assess the effect of antenatal exercises on labour outcome among primigravida mothers was conducted at Seethalakshmi Corporation Maternity Centre for experimental group and CTM Centre for control group in 2009-10. Simple random sampling technique was used to select 300 low risk primigravida mothers who were equally divided in each experimental and control group. Observation tools used for labour process included: Observation record on progress of labour (Partograph), Intrapartum variable proforma, Numerical Pain scale, Primigravida's behavioral responses during labour, neonatal variable proforma (APGAR score & presence of birth injuries). The overall mean knowledge score for control group was 5.37 ± 1.21 whereas in experimental group it was 25.11 ± 0.92 . There was a significant statistical difference found between control and experimental group in all the areas of knowledge on antenatal exercise among primigravid mothers ($t = 173.04$, $p<0.001$ level). Comparison of antenatal exercises on labour process showed that 10 (14%) had poor outcome, 46 (62%) had fair outcome and 18 (24%) had good outcome in control group whereas in experimental group 16 (17%) had fair outcome and 79 (83%) had good outcome. The overall mean value for control and experimental group was 12.45 ± 2.51 and 16.42 ± 1.698 respectively and the obtained 't' value is 12.33. at $p<0.001$ level, so there was a significant difference in the outcome of labour process among control & experimental group. The study was concluded that the regular practice of antenatal exercises along with knowledge had enhanced the labour process with good labour outcome which can be adopted by the nurses in their clinical practice.⁷

A descriptive study to assess the knowledge of primigravida mothers regarding antenatal care was conducted at Kadusonnaphalli PHC for rural and Narayanapuram PHC for urban area in 2014. 100 primigravida mothers were selected using convenient sampling technique which was from rural and urban areas. Self prepared a structured interview questionnaire was used for data collection. Majority of 82% primigravida mothers had inadequate knowledge and 18% had moderately adequate knowledge in rural area. In urban area 54% had moderately adequate knowledge and only 10% had adequate knowledge of antenatal care. There was no significant association found between knowledge of primigravida mothers with selected demographic variable. The overall aspects mean knowledge score was 40% with standard deviation 12.78 in rural and in urban 51.90% with standard deviation 14.14. Highest mean knowledge score was found on general information 48% in rural and 61.50% in urban regarding antenatal care. Thus the study concluded that more stress should be given to increase the knowledge of primigravida mothers regarding antenatal care.⁸

A randomized controlled trial was conducted to evaluate the effect of antenatal exercises on pulmonary functions and labour outcome in uncomplicated primigravida women at UCMS & GTB Hospital, Delhi, India in 2015. 122 uncomplicated primigravida in age group of 18-35 years at 24th - 28th week of gestation were randomly allocated into study and control groups. Baseline pulmonary function tests and parameters related to labour outcome were used as tools for data collection. Pulmonary function tests (PFTs) were done for all subjects at 24th week of gestation and after 36th week of gestation. A significant improvement in FEV1, FVC, FEF25-75%, MEF50%, MIF50% and MVV between 24 & 36 weeks was observed in the study group after exercises. In the study group 41.67% patients did not require augmentation compared to 22.95% in the control group. In study group 13.11% patients had meconium stained liquor which was significantly less in comparison to 41% patients in the control group. Mean duration of first, second and third stage of labour was shorter in study group than control group. In study group 96.72% had normal and none had instrumental vaginal delivery compared to 88.52% and 6.6% from control group respectively. Caesarean section rate was 3.3% in study group and 4.9% in control group. Fetal weight gain, birth weight, birth length and head circumference was significantly higher and NICU stay was significantly lesser in study group compared to control group. The study was concluded that antenatal exercises lead to improvement of pulmonary functions and hence labour and neonatal outcomes.⁹

A critical review of the literature examining the effect of exercise on maternal outcome was done in 2006. Literature reviews were done from PubMed (all available years), personal communications, and personal searches of files. Results indicated that exercise during pregnancy is safe and perhaps even reduces the risk of preeclampsia and gestational diabetes. The evidence for weight gain and labor and birth (rates of cesarean sections, duration of labor) was found to be mixed. Unfortunately, much of the researches examining exercise during pregnancy were observational, and the few randomized controlled trials that existed were small and inadequately powered. The study concluded that practitioners should encourage their healthy pregnant patients to exercise. Practical guidelines for recommending exercise to pregnant women were also presented.¹⁰

A quasi experimental study was conducted to evaluate the effectiveness of selected nursing measures on labour outcome among primigravid mothers at Nirmala Hospital, Suryapet in 2015. 40 primigravida mothers were selected using non – probability convenient sampling technique and equally divided into experimental and control group. A structured observational record sheet was used for assess labour outcome. This study revealed that there was high significant difference found in pain at $p < 0.001$ level between study and control group. The study concluded that, clinical implementation of nursing measures usage during antenatal period could be an effective non pharmacological intervention in improving labour outcome.¹¹

A quasi-experimental, clinical trial study aimed to determine the effect of participating in the labor preparation classes on vitality and positive affect during pregnancy and in postpartum period in the pregnant women referred to the Isfahan public health centers was done in 2010. 120 eligible pregnant women since 20 weeks of pregnancy who referred to these centers were selected simply and randomized in two groups (60 in the case and 60 in the control group). Questionnaires of vitality and positive affect toward the labor were used for data collection. There was a significant difference in vitality and positive affect regarding type of delivery before and after intervention and after delivery in case group ($p < 0.0001$). There was no significant difference in vitality and positive affect scores after delivery and after intervention ($p < 0.083$, $p < 0.545$). There was significant difference in vitality and positive affect scores between case and control groups after the intervention and after the delivery ($p < 0.001$, $p < 0.0001$). This study concluded that it could be recommended that all pregnant women should contribute in delivery preparation classes to improve their mood, confidence, vitality toward labor.¹²

Conclusion:-

Various searched studies suggest that there is association between antenatal exercises and labour outcome. Hence, Researcher is interested to assess the effectiveness of antenatal exercises on labour outcome among primi gravid mothers attending maternity services.

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RESEARCH ARTICLE

EFFECTS OF CONFIDENCE AND SOCIAL PRESSURE ON DECISION MAKING PROCESSES.

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Opinion Dynamics, Consensus, Majority Rule, confidence, agent base simulations

Abstract

A new opinion dynamics model in which the individuals are identified by two variables, namely parameterized with their binary opinion and confidence levels is introduced. In this model the exchange of opinions are realized through pair wise interactions. The dynamics is governed by the pressure of the neighboring sites and personal convictions of the individuals living on a regular lattice. The model successfully to describe opinion exchange mechanisms of the societies where individuals with various levels of confidence exchange opinion through pair wise interactions under the influence of nearest neighbors.

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Introduction:-

Societies consist of individuals who try to optimize their welfare. Each individual make some local changes with an expectation to improve its welfare state which leads to underlying dynamics of the global changes of the whole society. In this sense social systems and physical systems, such as spins systems, exhibit close similarities: Both want to optimize their statue in the system. These similarities are basic motivation for using statistical physics in many situations that occur in social systems. In applying statistical physics to the social systems the essential assumption is that, the individual preferences do not play crucial role in the global changes of the societies. Simulation techniques play an important role in statistics which are successfully applied in various areas of the social sciences. Rumour propagation, epidemics, opinion dynamics are the major areas of social sciences that simulation techniques have been applied with considerable success [1].

To modeling social systems, it is necessary to quantify the outcome of the individual behavior of the by suitable variables. These variables can take continuous or discrete values. Together with determining the variables, also the local the interactions between the individuals must be formulated. Basic difference between the physical systems and the social systems appear at this point. In physical systems the constituents have well defined interaction rules based on the energy minimization, while for the social systems different rules for dynamics are proposed for the evolution of the system. These rules are based on which behavior of the individual is observed.

One of the major issues in simulation studies of the social behavior is how opinions evolve in a social system, namely opinion dynamics. In opinion dynamics variables are opinions of the individuals which may take continuous [2]-[4] or discrete [5]-[6] values. Starting from randomly distributed initial opinions, dynamics may leads to consensus or fraction. Consensus is unanimously agreement on one of the opinions. While fraction is more than one opinion are supported by various group of the society. In magnetic systems all spins can be aligned (consensus) or sum of the spins may point one direction while the others point opposite (fraction).

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There exist a great number of opinion dynamics models which describe the formation of opinions in a social system. Schelling model [7] is known as the first one of the simulation model. Since then various models and their modifications are improved [8]. Some of these models introduce detailed opinion exchange mechanisms such as bounded confidence model [9] and they are applied to various social events successfully [10]-[12]. Bounded confidence model assumes continuous opinion and pair wise interaction. In this model the interacting individual may change opinion only if two interacting individuals have opinion differences less than a threshold value. In Hegselmann and Krause [13] model a randomly selected individuals accepts an average opinion of the neighbors which is different from his opinion by a confidence parameter ε . Some others take simple opinion exchange rules. In these simple models exchange of opinions can simply be accepting opinion of randomly chosen neighbor [10]-[12] or the individual may be influence by its neighbors and take the opinion of majority [14]-[15]. Sznajd model among these simple models is an exception. This model assumes that every individual try to influence its neighbors. If two neighbors share the same opinion neighbors of the pair accept their opinion.

As a sociological fact, opinion formation is strongly related to social pressure and confidence among many other effects. Some people easily accept the opinion of the neighbors while some others are reluctant to accept opinions coming from the others. Hence, even simple model of opinion formation requires taking into consideration social pressure on the individuals. The majority rule, in which agents agree with the local majority opinion at every time step is widely used in opinion dynamics studies [17]-[18]. This type of behaviour called herding behaviour, it is assumed that the minority definitely follows the majority. It is obvious that together with the effects of the neighbors, some personal merits plays important role in persuasion process.

Recently a variation the bounded confidence model has been introduced [19]. In this model agents are placed at the vertices of a regular lattice. Interactions are possible within a certain range the agents to interact with those which are located beyond their first neighbors. Each agent is identified by two parameters, its opinion and degree of conviction. The degree of conviction of an agent is affected during the interaction. If the interaction is with a neighbors of higher conviction, the agent lose some of his/her conviction. If the opponent has lower conviction in the light of the above discussion gain conviction. When one of the agents reaches a certain lower bound of conviction then its opinion changes to that of the opponent. Therefore, in this work, the difficulty in persuading someone who has a strong conviction was taken into consideration.

In this work, a new model which aims to study the relation between the social pressure and the confidence is proposed. In this model the individuals interact only with their nearest neighbors and it is assumed that they may exchange opinion through pair wise interactions. The model is essentially a variation of the majority rule model: two randomly selected individuals exchange opinion under the pressure of the neighboring sites and their own personal convictions which is added to the model as a new parameter for each individual. If an individual interact with one of its neighbors, a) may convince the neighbor to change opinion, b) may change its opinion. If an individual is in local agreement with its surrounding neighbors and have high confidence may hardly change opinion. In this model a function which measures the satisfaction level of the individuals is introduced. This function sets numerical value for the local agreement with the surrounding neighbors and the confidence of the individual living at site i . When two neighbors exchange opinion the relative values of their satisfaction functions play the decisive role and exchanging opinion can be realized according to the rules give in Section II.

The main aim of this study is to determine the role of confidence of the individuals and the pressure created by the neighboring agents in opinion exchange mechanisms. This work differs from the existing literature by the opinion exchange dynamics.

This work is organized as follows. Section II is devoted to a detailed description of the model. The results and discussions on the results are presented in Section III whereas concluding remarks are drawn at Section IV.

Computer Simulation Model:-

The society consists of N interacting individuals living on a regular 2-dimensional lattice with only nearest neighbor interactions. The dynamics of the opinion exchange is assumed to take place through discrete time steps. The agents are identified by two parameters: their opinion and confidence level. Opinion can only take discrete values while the confidences of individuals are parameterized by real numbers.

Agents possess binary opinions: ± 1 for supporting and for opposition the proposition. Initially, the opinions are set randomly ± 1 with probabilities p and $q = 1 - p$. The confidence parameters are set according to the initial distribution of the opinion of the individuals. In this work, all individuals may carry i) the same confidence level, ii) two different confidence levels for $+1$ and -1 opinion holders, iii) randomly distributed confidence levels for each individual. Since both, confidence levels and the opinions depend on the probability, p is a crucial control parameters of the simulation. During this work the probability is kept as $p = 1/2$.

Opinion dynamics models such as Sznajd Model [19] assume interaction between the agents sharing the same opinion. When such a pair interacts, they influence all of their nearest neighbors. The Sznajd model is an exception in this sense. There exist also some other models where the majority opinion is the decisive factor for changing opinion. The proposed model is based on mutual interaction of pair of individuals occupying two neighboring sites. In this sense, the interaction type is similar to Sznajd model with the difference that the Sznajd model considers only two sites with the same opinion whereas in this model pairs can carry any opinion. The selected pair can influence each other according to their level of satisfaction. In such a societies satisfaction can be described as the level of cooperation with the neighbors: if the majority of the neighbors share the same opinion the individual feels comfortable and satisfied with its position. The satisfaction, in this model, has another component: the confidence. An individual with high confidence level feels satisfied even when some neighbors are not in accord. If the majority of the neighbors carry the opposite opinion then this individual either have to change its opinion to reduce the pressure exerted by the neighbors or has to move to another place. This is common in physical systems where under pressure constituents of the system change state to reduce their energy or exerted pressure. Hence in a society, satisfied members are the highly confident individuals which feel comfortable in interactions with the like individuals.

The satisfaction of the individual living at site i can be formulated as,

$$E_i = O_i * \left[\sum_{j \in NN} O_j + C_i \right] \quad (1)$$

where E_i , O_i and C_i are the satisfaction, opinion and the confidence parameters of the individual living at site i . j and O_j indicate the location and opinion of the neighboring sites.

The model is based on interaction of two neighboring individuals. One of the individual may convert the other through their mutual interaction. This interaction and the opinion changing mechanism depends on the satisfaction parameters of the individuals.

As far as the satisfaction parameters are concerned three different cases must be considered.

1. Both neighbors are unsatisfied
 $E_i < 0, E_j < 0$.
2. Either one of the neighbors is satisfied while the other is unsatisfied
 $E_i < 0, E_j > 0$ or $E_i > 0, E_j < 0$.
3. Both neighbors are satisfied
 $E_i > 0, E_j > 0$.

These three cases can be used to formulate the local dynamic of the opinion changing mechanism.

1. Case 1: if both parties are unsatisfied, changing opinion for both will create satisfied individuals. Hence in this situation both interacting individuals change opinion.

$$E_i < 0, E_j < 0 \Rightarrow O_i = O_j = O_i.$$

2. Case 2: In the case one of the neighbors are unsatisfied while the other is satisfied.

$$E_i < 0, E_j > 0 \Rightarrow O_i = O_j = O_j.$$

$$E_i > 0, E_j < 0 \Rightarrow O_j = O_i = O_i.$$

Unsatisfied individual of the pair will accept opinion of the other.

3. Case 3: If both neighbors are satisfied, $E_i > 0$ $E_j > 0$, and they both share the same opinion. Opinion change is not possible. But if the opinions are different, $O_i \neq O_j$ relative satisfaction determines which one of the interacting neighbors will change opinion:

$$\begin{aligned} E_i < E_j \quad O_i &= O_j \quad O_j = O_j \\ E_i > E_j \quad O_j &= O_i \quad O_i = O_i \end{aligned}$$

Here in this opinion updating process opinion changes are realized according to the satisfaction level of the individuals.

These updating rules are the local opinion exchange moves. After setting the opinion distribution and confidence levels, the observation of the dynamic changes of the opinions of the individuals or the society as the whole constitute the basic steps of the simulation.

A configuration is considered as a snapshot of the society at a given time t . Initial configuration is the distribution of opinions at the time t_0 , the beginning of the debate. Debate continues with pair wise interactions of the individuals until the society reaches the final distribution of opinions. Reaching this final state takes some time. The final state can be a consensus or polarization. In order to observe the final state of the society it is best to analyze the configuration average of the opinions distributed in the society at any given time t . Configuration average is calculated as follows,

$$\bar{O}(t) = \frac{1}{N} \sum_{i=1}^N O_i(t) \quad (2)$$

Where $O_i(t)$ and $\bar{O}(t)$ are the number of individuals, the opinion of the i^{th} individual at time and $\bar{O}(t)$ is the configuration average of the opinion held by the society respectively. $\bar{O}(t) = \pm 1$ indicate consensus on one or the other opinion.

Since the initial configuration is an arbitrary distribution of opinions with a given fixed value of probabilities p and q , one must take average over many statistically independent configurations. The number of statistically independent configurations are $N_{samples}$. The simulation should be repeated and averages must be taken over the statistically independent configurations.

$$\langle \bar{O} \rangle(t) = \frac{1}{N_{samples}} \sum_{i=1}^{N_{samples}} \bar{O}_i(t) \quad (3)$$

Given any starting configuration the dynamics determined by the defined interactions change the average opinion held by the society to the most stable configuration. This process requires the transition time, $t_{Transition}$. After this transition period the society reaches a final state which may fluctuate around an average value or reaches to consensus over one of the states. The time duration from a given initial state, to the state where stationary state are observed $t_{max} \gg t_{Transition}$ varies with the values of free parameters and the interactions between the individuals. For a given set of parameters, configuration averages calculated at t_{max} fluctuate around an average $\langle O \rangle$.

$$\langle \bar{O} \rangle = \frac{1}{N_{sample}} \sum_{i=1}^{N_{samples}} \bar{O}_i(t_{max}) \quad (4)$$

where, N_{sample} is the number of different statistically independent initial configurations which contribute to the average.

If both opinions live together, the average opinion represents the opinion of the society. In some cases, in the long term, the societies end up with consensus on one or the other opinion. Depending on the initial configuration either one of the opinions can win. In the case of equal distribution of opinions consensus may be reached on one or the other opinion values. Hence, taking the average gives misleading result. Together with the average one also must check whether this average is truly representing the opinion of the society. Best method is to calculate the distribution of the configuration averages. If distribution is normalized to the number of configurations one can obtain the percentages of the configurations which end with consensus on the pro or con opinions.

Results:-

The simulations are performed on (50×50) square lattice. Initial configurations are prepared by setting two parameters which identify the opinion and confidence level of each individual. A configuration is created by filling the sites randomly with opinion values ∓ 1 with equal probability. Confidence parameters are set according to two scenarios: i) All individuals have fixed confidence values, ii) Supporters of one of the opinion have higher confidence level. After preparing the initial configuration the time development of the opinion change of the society is observed for 10000 discrete time steps. During the simulation a new configuration is created by updating randomly chosen lattice sites. Updating each site is repeated volume times (50×50) . Since pairwise interactions are considered, this updating procedure corresponds to approximately updating each lattice site twice. Averages are calculated over 500 statistically independent configurations.

During the simulations, opinion averages are calculated by taking the configuration averages, Eq. (2) at each time step. The average of the configuration averages are calculated for each time slice over the 500 statistically independent runs, Eq. (3). The configuration averages are also used to calculate configuration distribution at every time step. Distributions are very illuminating since opinion averages may be misleading for the situations where consensus may be reached on either one of the opinions.

Fig. 1 shows the time evolution of the average opinion formation for various confidence parameters. From bottom to top, first two curves corresponds to equal confidence parameter values which are $C_1 = C_2 = 0$ and $C_1 = C_2 = 0.1$. For the case where vanishing confidence parameter is considered, all configurations exhibit very low average opinion value. This is an indication that in an equally divided society, with individuals ready to change opinion under any pressure, both opinions will always survive. Even small confidence parameters change this situation. In the case where supporters of both opinions have equal, small confidences, an immediate change in the behavior appears. This changing behavior can be summarized in two stages. The first stage corresponds to a very rapid opinion changing regime. After this rapid change the system gets into a more stable phase where more and more individuals share the same opinion. The effect of the unbalanced confidences is shown in top curve. Considering supporters of either one of the pinions with slightly larger confidence, $C_1 = 0.1$, $C_2 = 0.2$ change the picture completely. The initial rapid opinion changes are much faster in this case. System reaches consensus after a very rapid persuasion process.

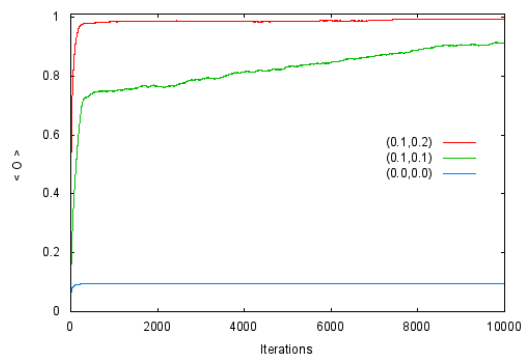


Fig. 1:- Time evolution of the average opinion of an initially equally divided society.

Fig. 2-5 show the time evolution of the distribution of configuration averages of the opinion formation. Each figure exhibits the effects of a different set of confidence parameter. In Fig. 2 vanishing confidence level is considered for supporters of either one of the opinions. As it is seen from the time development of average opinion formation Fig. 1 the distribution also exhibit that with vanishing confidence, individuals change opinion under the influence of

neighbors only. This influence is a random effect. Hence the configuration averages are distributed around the vanishing average value.

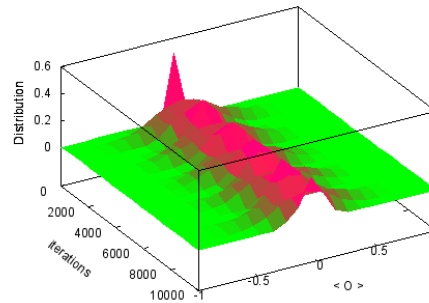


Fig. 2:- Distribution of average opinion for a society with vanishing confidence $C_1 = 0, C_2 = 0$.

Fig. 3, also shows a similar behavior to that of Fig. 2: distributions are centered around the vanishing average opinion value. Similarity between the Fig. 2 and Fig. 3 ends here. The reason of having distributions centered around vanishing average opinion value, in this case is, very different. The strong confidence does not allow the individuals to change opinion under the influence of the neighbors. Since the original initial configuration consist of equal number of both pro and con individuals, configuration average remains very small under the influence of the high and equal confidence levels.

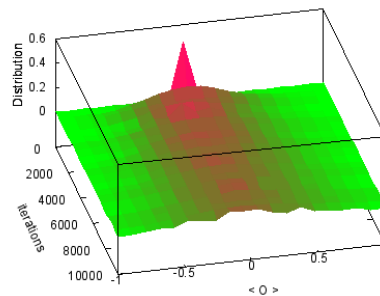


Fig. 3:- Distribution of average opinion for a society with high and equal confidence $C_1 = 2.0, C_2 = 2.0$.

Fig. 4 shows time development of the distribution of configuration average of opinions. Here both opinions are supported by the individuals with non-vanishing and equal confidence. Supporters of both opinions try to persuade the individuals with opposite opinion. Depending on the initial configurations, one or the other opinion succeed to reach consensus. The peaks at the extreme ends, $\langle \bar{O} \rangle = \pm 1$ indicate that consensus is reached considerable times. In the case where both parties have equal confidence, it is almost impossible to predict which initial configurations will lead to consensus. There are also many configurations with average opinion values lies in between -1 and $+1$ which give a distribution around 0. This behavior can be better understood by comparing the Fig. 1 and Fig. 4.

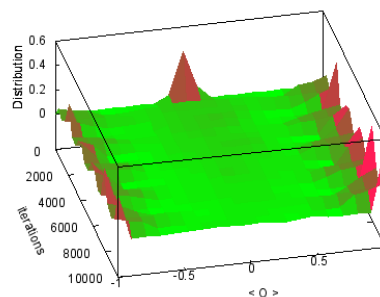


Fig. 4:- Distribution of average opinion for a society with non-vanishing and equal confidence $C_1 = 0.1, C_2 = 0.1$

This situation completely different when even a small difference appear in confidences. Fig. 5 is devoted to the discussions of the effects of difference in the consensus parameter. Individuals with strong confidence are difficult to persuade. Moreover they have advantage that they are more satisfied in their local environment.

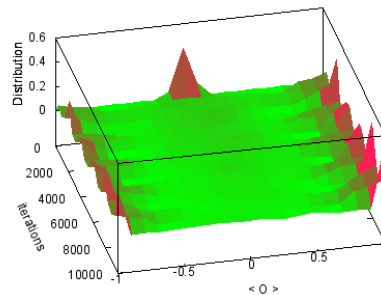


Fig. 5:-Distribution of average opinion for a society with high and equal confidence $C_1 = 2.0, C_2 = 2.0$

This high satisfaction enables them to persuade the opponents in interactions. This is clearly seen in Fig. 1, and Fig. 3. In Fig. 5 distribution of the configuration averaged opinion values peak around zero at the initial state of the simulation. But immediately after few hundred iterations every configuration ends with consensus on the opinion which is supported by high confidence supporters. Here in this simulation work the consensus is reached on the opinion with the value -1 , whose supporters carry confidence level of 0.2 .

Conclusions:-

The proposed model is a simple, deterministic model of opinion formation with confidence parameter which is similar to the external field in the physical systems. In this simple form, the model is capable of predicting the conditions for partition or consensus. Numerical results give the indication that even a small non vanishing confidence parameter is essential for reaching consensus. If supporters of both opinions have equal confidence, the consensus is just coincidental.

The model in this simple form can be considered as a laboratory to discuss the decision making mechanisms. Moreover it is open to extensions. The simple extension can be that the confidences may change in time with pairwise interactions.

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RESEARCH ARTICLE

LE VOYAGE DOCUMENTAIRE DE MICHEL LEIRIS EN AFRIQUE NOIRE.

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Abstract

This paper tries to find out the implications of trade liberalization on international trade tax revenues and its macroeconomic implication on Pakistan economy. The theoretical approach of trade reforms and its impact on the direction of change of revenue is ambiguous because of its dependence on the productivity of 'trade tax revenues'. By using the data of twenty five years, the paper first establishes the productivity of 'trade tax revenues' in Pakistan, then by an econometric analysis exploration of the relationship between trade revenues and trade liberalization in Pakistan is done. It was observed through our analysis that there is high productivity of trade tax revenues, further, our results show a positive and significant relationship between trade liberalization and collection of trade tax revenue. To support this relationship, a second test of Granger's causality is also performed which confirm the causal relationship between trade tax revenues and trade liberalization. It was suggested that supportive macroeconomic policies are prerequisite for successful trade liberalization.

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Plus que la plupart des participants au monde anthropologique, Michel Leiris échappe à la définition des catégories traditionnelles de l'activité intellectuelle. Avec un pied dans Anthropologie et littérature, sa vie a également été centrée sur des relations personnelles étroites avec un réseau diversifié d'artistes et de penseurs créateurs, de Picasso à Sartre. Dans le domaine de l'anthropologie, tant sa reconnaissance de l'importance de l'ethnologue, la subjectivité et sa fascination pour les situations sociales et culturelles «hybrides» (particulièrement en milieu colonial) placent son œuvre en 1930 un demi-siècle en avance de son temps. Son vrai nom était **Julien Michel Leiris**, il est né le 20 avril 1901 à Paris et il est décédé le 30 septembre 1990 à Saint-Hilaire, Essonne, France. Il était un écrivain surréaliste et aussi un ethnographe.

Michel Leiris, en 1931, prit la fuite pour l'Afrique, un homme frustré souffrant de phobies sexuelles, d'inhibitions écrivains et d'un penchant pour l'abus d'alcool (Roudinesco 1992: 45-48). Suivant les conseils de son ami Georges Bataille, Leiris a commencé la psychanalyse en 1929 pour ces problèmes interdépendants qu'il a dû interrompre pour entreprendre son voyage à travers le continent africain. Avec le consentement de son analyste, Leiris accepte l'invitation à se joindre à la mission Dakar-Djibouti en tant qu'archiviste et secrétaire, ce qui l'éloignera de sa vie à Paris pendant une bonne partie de deux ans. Il n'est donc pas surprenant que les notes de terrain du jeune auteur reflètent sa santé mentale fragile où les obsessions personnelles, l'auto-reproche, et l'âme angoissante l'accompagnent de village en village et même sur le bateau de retour à la maison. Cette information biographique, considérée dans ses contextes historiques et culturels, jette une lumière précieuse sur L'Afrique fantôme de Leiris à

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cause de son filtre remarquablement subjectif en tant qu'observateur d'Afrique (Leiris 1981). En fait, le lecteur apprend autant, sinon plus, sur le monde intérieur de Leiris dans *L'Afrique fantôme* que sur les peuples et les cultures de l'Afrique qu'il a rencontrés le long du chemin. Même dans les circonstances ordinaires, la localisation de l'écrivain comme sujet de cette manière est utile à l'étude de la littérature de voyage et de l'ethnographie comme discipline, mais cette approche prend de plus en plus d'importance avec *L'Afrique fantôme*, étant donné l'état d'esprit troublé et la mission personnelle de l'auteur : Leiris est allé en Afrique à la recherche d'une nouvelle peau. Ses attentes culturelles de l'Afrique ont tenu l'espoir pour sa propre transformation personnelle en se reconnectant avec l'enfant sauvage à l'intérieur.

Bien que n'étant plus officiellement membre du groupe surréaliste lorsqu'il est parti pour l'Afrique, des traces de la participation de Leiris à cette expérience révolutionnaire sont présentes tout au long de son journal de voyage. Parmi tous ceux qui ont participé au surréalisme français, Michel Leiris fournit le meilleur exemple de la manière dont son milieu intellectuel d'avant-garde à Paris a combiné une fascination africaniste avec des notions de protestation politique. Dans cet esprit, je vais explorer à la fois les limites épistémologiques et institutionnelles de l'engagement de Leiris avec l'Afrique dans *L'Afrique fantôme*. L'attention portée à l'épistémologie de Leiris se focalisera sur sa perception de lui-même, de l'Afrique et des autres ethnographes de l'équipe avec lui par rapport aux constructions dominantes de «l'Afrique» avec des catégories «primitives» et «civilisées» - l'opposition structurant le primitivisme d'avant-garde. La description de Leiris de leur travail en tant qu'ethnographes (Collection d'objets, utilisation de traducteurs) et la justification de leurs actions fournit des preuves troublantes de la relation problématique de sa génération avec l'Afrique. Le rôle de l'archiviste et secrétaire de la mission Dakar-Djibouti, première expédition ethnographique française en Afrique, l'a placé dans une position privilégiée pour enregistrer à la fois les domaines où il et sa cohorte ouvrent de nouveaux horizons ainsi que les lacunes de leurs pratiques.

En raison de la nature collective de cette mission ethnographique, il sera utile de considérer la dimension institutionnelle de l'engagement de ces Français avec l'Afrique en plus des concepts et des catégories organisant le discours de l'auteur. Leiris fait partie d'une équipe d'ethnographes, dirigée par Marcel Griaule, qui a traversé le continent africain d'ouest en est, commençant à Dakar à l'automne 1931 et finissant à Djibouti au printemps 1933. Bien que l'inspiration pour l'expédition ait été multiforme, l'entreprise était avant tout une entreprise de collecte de musées financée en partie par le gouvernement français. Au cours de l'année et demie du groupe passé en Afrique, ils ont rassemblé plus de 3500 œuvres d'art et artefacts qui sont maintenant logés au Musée de l'Homme. La manière dont la Mission Dakar-Djibouti, en tant qu'initiative institutionnelle parrainée par l'Etat, structurerait et circonscrirait les actions des participants sera considérée comme faisant partie d'un processus collectif d'incorporation d'informations sur les colonies comme moyen de faire face aux impériales expansions. Ces deux dimensions de *L'Afrique fantôme* - l'épistémologique et l'institutionnel - mettent en évidence la nature paradoxale du rejet de Leiris de la culture française bourgeoise en fuyant en Afrique en quête d'un nouveau moi tout en restant redevables à sa nation et à sa classe, La préservation de leurs intérêts.

Il y a une longue tradition d'écrivains français qui se préparent à échapper à la fatigue de leurs occupations intellectuelles à Paris; Deux prédécesseurs mentionnés par Leiris sont Arthur Rimbaud et André Gide. Le cas de Rimbaud est le plus extrême des deux: il abandonne entièrement la poésie d'écriture et se met à la voile pour l'Afrique, seulement pour revenir des années plus tard sur une civière avec des histoires sauvages à raconter (Nicholl 1997). Pour Gide, c'est juste après avoir terminé *Les Faux-monnayeurs* (1925) qu'il entreprend un long voyage à travers l'Afrique équatoriale française. À son retour, Gide publia ses journaux de voyage '*Voyage au Congo*' (1927) et '*Retour du Tchad*' (1928), qui vendit immédiatement des milliers d'exemplaires. Non seulement les impressions de Leiris sur l'expédition africaine de Gide l'accompagnent sur la route, Leiris nourrit les espoirs de rencontrer réellement Rimbaud en Afrique - la quintessence des rebelles devenus natifs. Le début de *L'Afrique fantôme* révèle l'état d'esprit initial de Leiris et comment le voyage a été entrepris avec le désir d'échapper à la monotonie de la vie métropolitaine.

"En somme, très peu de différence entre la vie du fonctionnaire à Paris et sa vie à la colonie (j'entends : dans les grands centres) ; il a chaud et il vit au soleil au lieu d'être enfermé ; en dehors de cela, même existence mesquine, même vulgarité, même monotonie, et même destruction systématique de la beauté. J'ai grand 'hâte d'être en brousse. Cafard" (Leiris 1981 : 28).

La quête de Leiris n'était pas seulement de découvrir l'Autre, mais de se trouver en Autre. Loin de ses devoirs conjugaux, de la société française et des querelles de la Centrale surréaliste, il trouve son chemin non pas vers un

être sauvage et sauvage, mais vers l'écriture. La distance dans cette équation était décisive, libératrice; Il a créé un sentiment de sécurité et de stimulation. Il a été confronté à une abondance continue d'informations à interpréter et sa tâche officielle était de prendre des notes. C'était une recette parfaite pour guérir le bloc de l'écrivain mais pas nécessairement celui qui ferait la carrière d'un écrivain. Leiris écrit *L'Afrique fantôme*, l'œuvre qui le rendait célèbre, sans aucune idée qu'il deviendrait un classique, alors que ses publications ultérieures - souvent le fruit d'un travail laborieux - n'avaient jamais obtenu la même reconnaissance. La combinaison de l'ethnographie et de l'autobiographie - une recherche de soi dans l'Autre - qu'il a trouvée lors de son premier voyage en Afrique est devenue un aspect durable de sa vie d'écrivain. Dès le début, le jeune poète fait des observations approfondies sur son environnement, ce qui prête une qualité d'écrivain aux notes de terrain de l'ethnographe en difficulté. Il n'épargne ni les Européens ni les indigènes son ironie humoristique rendue avec un usage habile de détails descriptifs:

“Allé le soir avec les B... et Griaule à *L'Oasis*, dansant nègre de Dakar. On y voit : des négresses — femmes ou amies de sous-officiers de tirailleurs — habillées à l'européenne ; des putains noires, métisses ou arabes ; quelques grosses négresses en costume local ; des pédérastes nègres qui dansent ensemble en petit veston cintré ; un pédéraste blanc à l'allure d'employé de bureau dansant, une fleur à la bouche, avec un marin nègre à pompon rouge ; deux sous-offs de la coloniale dansant en couple ; trois types de la marine marchande ou de la marine de transport, dont l'un (à casquette blanche à visière, petite moustache en fil, et cigare) a aussi merveilleuse allure que les plus beaux aventuriers des films américains“ (*ibid.*: 33).

Ce passage illustre le ton d'une grande partie de *L'Afrique fantôme*: sa qualité hybride combine la pratique noteuse de l'archiviste-secrétaire ("On y voit: ..."), une sensibilité littéraire à l'amoureux à la recherche de sensations fortes, comme Ainsi que l'œil d'un ethnographe pour documenter l'Autre. Le style conscient de Leiris, l'utilisation libérale des dates et des noms de lieux permettent au lecteur de situer le sujet narratif dans le temps, l'espace et la culture. En termes plus généraux, l'intersection entre autobiographie, ethnographie et vision surréaliste de la culture fait de *L'Afrique fantôme* un document particulièrement précieux de l'engagement de l'auteur dans les tendances culturelles qui ont façonné les valeurs et les perspectives de sa génération, notamment le primitivisme d'avant-garde.

Compte tenu de son âge et de son état d'esprit - Michel Leiris n'avait que trente ans quand il partit pour l'Afrique en 1931 - il démontre parfois un admirable scepticisme et une maîtrise de soi dans ses perceptions. Il exprime des doutes sur la crédibilité de ses sources d'information et même suspecte que certaines des performances.

Leiris exprime ouvertement des réserves quant à l'objectivité de l'ethnographe et se permet plutôt de communiquer ses propres contradictions personnelles - joies, peines et frustrations - à la fois dans la sélection et le commentaire des scènes, qui donne une touche d'humanité unique à *L'Afrique fantôme*.

Leiris commence par un rejet viscéral de tout ce qui est européen et laisse échapper des occasions de railler les indigènes qu'il rencontre - pour être sauvage, analphabète, prélogique - contrairement à d'autres témoins comme Gide. Mais la perspective française de Leiris est néanmoins opérationnelle et nulle ne part plus évidente que dans son rapport paradoxal à la culture métropolitaine bourgeoise, si typique du primitivisme d'avant-garde. Il remarque, par exemple: "Voici enfin que j'aime l'Afrique. Les enfants donnent une impression de gaieté et de vie que je n'ai rencontrée nulle part ailleurs. Cela me touche infiniment " (*ibid.*, 34).

Cette idéalisation de l'Afrique en tant que royaume des enfants, libre des contraintes du monde civilisé, représentait un pays des merveilles inaccessible pour Leiris. Bien que ce mythe primitiviste persistait pour la plus grande partie de son voyage, il finit par reconnaître l'impossibilité de devenir l'enfant ou l'indigène dont il rêvait. Après sept mois de route, Leiris regrette que sa vie en Afrique n'ait pas livré le paradis d'enfance qu'il avait espéré et note: "Avec mon casque, ma chemise kaki, ma culotte de trappeur, je reste le même homme d'angoisse que Certains considèrent comme un bon type, à la fois tranquille et pittoresque, Une sorte d'artiste bourgeois " (*ibid.*, P. 162). Cette frustration résultant de son échec à la quête de l'auto-transformation le suivit tout le chemin du retour à Paris. Plutôt que la moyenne tabula rasa du discours colonial, «l'Afrique» devient une vaste étendue de territoire qui permet à l'écrivain d'explorer lui-même le continent et ses fantasmes, qu'il décrit en termes libidinaux qui ressemblent au désir d'un homme Femme (*ibid.*, 182). Beaucoup d'éléments de primitivisme d'avant-garde structurent la représentation de Leiris de son expérience de l'Afrique: une idéalisation du buisson primitif en contraste avec un mode de vie civilisé métropolitain, le désir persistant de se reconnecter à un être sauvage et indompté, un penchant pour l'exotique et tropical à La dépense d'évaluations plus précises et réalistes, et ainsi de suite.

Leiris note régulièrement leurs acquisitions et les circonstances entourant ces activités, y compris l'utilisation de traducteurs, les négociations prolongées et les scandales intermittents. Ainsi, le processus de collecte des objets apparaît comme un thème constant de la revue. Les remarques initiales de Leiris sur ce sujet reflètent sa participation volontaire au groupe, sa reconnaissance d'une incompréhension entre les Français et les villageois ainsi que la preuve de ce que l'auteur a par la suite appelé son arrogance européenne:

«L'enquête et la collecte d'objets commencent et se poursuivent dans une ambiance parfaitement idyllique. Les gens s'amusent beaucoup de nos questions, qui leur semblent invraisemblables de futilité. Il en est de même de nos achats, puisque tous ces éléments sont susceptibles d'être très frustrés (ibid., P. 41).

Ses réactions, ici celle d'un amusement détaché mélangé à un sentiment d'autosatisfaction, subissent de nombreuses transformations le long du chemin allant de la frustration profonde à l'émerveillement comme Leiris relie les interactions du groupe avec les populations indigènes. Cependant, ce renversement non autorisé des valeurs, des perspectives et de l'intégrité culturelle des populations autochtones caractérise les descriptions de Leiris.

En plus de rassembler des objets pour exposer dans les musées parisiens, l'équipe d'ethnographes sous la direction de Griaule a documenté et étudié divers aspects des cultures africaines qu'ils ont rencontrées y compris les arts traditionnels, le rituel, la religion, la langue et l'histoire. Les notes de terrain qu'ils ont acquises, cependant, reflètent de brèves rencontres puisque l'équipe a passé très peu de temps dans chaque endroit. Ils se sont également appuyés sur des traducteurs et n'ont pas été en mesure de connaître les personnes ou leurs pratiques. En ce sens, la dimension collecte d'objets de l'entreprise et la documentation subséquente qu'ils ont inspirée a contribué plus longtemps à l'étude ethnographique des cultures africaines France. Griaule (1933: 7-12) décrit leur approche ethnographique comme méthodologiquement expérimentale; Forgeant une combinaison originale d'esthétique, de sociologie et de psychologie. Un groupe autour des anciens surréalistes a commencé à développer un intérêt ethnographique dans les cultures non occidentales des colonies françaises d'outre-mer; Des intellectuels tels que Bataille et Leiris ont franchi une étape au-delà des surréalistes et ont pris les cultures dites «primitives» comme objet d'étude ethnographique. Cette synthèse interdisciplinaire dont l'ethnographie française a émergé reflète l'atmosphère et la sagesse de l'entre-deux-guerres, marquant la première étape d'un processus continu qui donnerait lieu à la combinaison de l'anthropologie et du marxisme par les écrivains qui ont contribué aux Temps modernes. Tour ont raffiné leurs outils analytiques en accord avec leur propre ère et ses impératifs. Le point ici est que les engagements français avec la culture coloniale sont dynamiques, mis à jour à mesure que chaque nouvelle génération redéfinit ses méthodes et ne peut être homogénéisée en une seule épistémologie colonialiste.

Si son intention était d'exploiter le côté sauvage et indompté de l'individu dont Breton et les surréalistes écrivaient, l'expérience de Leiris en Afrique produisit presque l'effet inverse. Dans sa quête de quitter lui-même et sa culture - pour devenir Autre - ses quelques moments d'accomplissement viennent d'accomplir ses devoirs bureaucratiques comme archiviste et secrétaire. Le lecteur est témoin d'un double vol dans L'Afrique fantôme: incapable de perdre son moi européen dans le buisson africain tant attendu, Leiris s'enfuit de son angoisse par écrit. La prise de notes devient son refuge ultime:

"Au dîner, je reçois plusieurs lettres. Comme toujours, elles me comblent de joie d'abord puis me plongent dans un abîme de tristesse, en me faisant sentir plus durement ma séparation. Je me couche et je dors à peine, réveillé d'abord par une petite pluie qui me force à regagner mon compartiment, puis par les moustiques, la voiture n'ayant pas de lampe électrique je n'ai pas pu installer ma moustiquaire dans le wagon. 12 juillet. Cafard terrible le matin, à pleurer. Puis salut, en me plongeant dans les travaux bureaucratiques et dans la rédaction de ce journal, depuis quelques jours abandonnés "(ibid., 62).

Un autre coup de cafard! De toute évidence, la réalité de cette expédition n'était pas aussi remplie d'aventure que l'ancien surréaliste l'avait espéré. Quand les émotions deviennent accablantes et les raisons pour lesquelles il quitte la France, il se tourne vers l'écriture, qui devient le véritable processus de découverte.

Après son retour d'Afrique, Leiris a travaillé comme ethnographe, documentant un grand nombre des objets recueillis sur cette mission dans son bureau du Musée de l'Homme. Ces développements marquent le début d'un processus continu d'incorporation des éléments des territoires français d'outre-mer aux pratiques et institutions culturelles françaises. Tout cela faisait partie d'une réorganisation et d'une rationalisation plus profonde de l'identité nationale française dans le contexte de l'héritage impérial de la nation.

Une des interprétations les plus influentes de ce processus d'ajustement conceptuel de l'intelligentsia française à la suite des nouveaux termes de leur participation aux cultures et aux peuples colonisés par la France a été l'argument avancé par James Clifford dans 'The Predicament of Culture (1988)'. Pour rendre compte des affinités conceptuelles entre le surréalisme et l'ethnographie, Clifford propose que les deux activités impliquent un processus de défamiliarisation culturelle. Il compare l'effort du surréalisme pour rendre les aspects familiers de la culture française étranges avec les ethnographes dont le but est de rendre étranges cultures familières. Le processus élémentaire de repenser les valeurs culturelles et de pousser les limites conceptuelles se reflète dans la manière dont Clifford a lié les dimensions d'avant-garde de l'ethnographie et du surréalisme. Il associe l'assaut aux perceptions classiques des valeurs culturelles qui avaient été dirigées contre l'establishment bourgeois par le dadaïsme et le surréalisme avec une relativisation de la supériorité culturelle européenne par rapport aux colonies. Il fait ressortir les similitudes entre l'art d'avant-garde et l'ethnographie et affirme qu'ils ont tous deux servi à décentrer l'autorité culturelle européenne pendant les années 1930.

Pour illustrer sa comparaison du surréalisme et de l'ethnographie, Clifford cite le Collège de sociologie comme son exemple unique de surréalisme ethnographique et L'Afrique fantôme de Leiris comme son seul exemple d'ethnographie surréaliste, mais il admet que les deux sont des constructions utopiques (Clifford 1988: 147). La portée limitée de ses exemples est moins un problème que leur statut utopique; Cela soulève des questions sur ce que c'est en cette période de l'histoire intellectuelle française que Clifford cherche à s'approprier pour faire avancer la critique contemporaine des pratiques culturelles européennes. Si l'ethnographie a contribué à rendre plus visibles les cultures lointaines des populations colonisées par la France à un public métropolitain, la nature du processus conservait encore la centralité d'une perspective française. Bien que Clifford considère la collection et l'exposition d'objets d'art africains dans les musées parisiens comme rédempteurs et alimentés par une idéalisation nostalgique de l'art africain comme quelque chose de «pur» à récupérer par l'Occident, il ne reconnaît pas les implications de cette idéalisation française de l'Afrique Est inscrite dans une épistémologie colonialiste.

L'attention persistante de l'auteur aux détails ici est remarquable. La façon dont il observe ses propres gestes théâtraux suggère un plaisir pervers dans leur démonstration de volonté et de puissance. De même, la description minutieuse de détails tels que la fuite des villageois, les larmes de la jeune femme et la place déserte du village témoigne d'un manque total de compassion pour la perturbation du bien-être de la communauté. La façon dont les hommes d'oreilles roses venaient et violaient l'intégrité spirituelle de cette communauté était sans doute regrettée pour les années à venir. De plus, Leiris semble apprécier le statut de démon, de voleur et de coupable. Il décrit leur départ:

"Les 10 francs sont donnés au chef et nous partons en hâte, au milieu de l'ébahissement général et parés d'un auréole de démons ou de salauds particulièrement puissants ou osés.

À peine arrivée à l'étape (Dyabougou), nous déballons notre butin: c'est un énorme masque à la forme vaguement animale, malheureusement détérioré, mais entièrement recouvert d'une croûte de sang coagulé qui lui confère la majesté que le sang confère à Toutes choses "(ibid., P. 104).

Curieusement, Leiris considère les discussions sur la littérature et l'esthétique comme discontinues avec leur entreprise ethnographique en Afrique. Cet aveu révèle à la fois les attentes de Leiris sur les voyages en Afrique et la disparition éventuelle de ces illusions après plusieurs mois sur la route. Cette admission clarifie encore comment les informations recueillies au cours de leur voyage à travers l'Afrique a été filtré à travers l'esprit des intellectuels qui ont trouvé la conversation sur les grandes œuvres de la tradition littéraire européenne irrésistible, même une fois qu'ils sont arrivés dans le buisson longtemps attendue d'Afrique. Mais le fait qu'un filtre esthétique soit présent n'est pas du tout incongru; Au contraire, l'esthétique était à l'origine de l'engagement des intellectuels français avec les cultures non occidentales en termes ethnographiques. Ce que cette affirmation dément, c'est la façon dont leur centre de gravité est resté européen et que le sien a été la cause de la consternation.

Une autre explication influente des engagements français avec l'Afrique est une version recyclée de l'hypothèse originale d'Edward Said que le geste européen d'atteindre vers les cultures étrangères met le visage intellectuel face à son propre désir. C'est aussi une idée puissante et convaincante, mais elle exclut la possibilité de processus dynamiques et de changements progressifs au fil du temps, contrairement à l'approche de Clifford, qui prétend trop optimiste sur l'avenir pour les avant-gardes européennes. Bien que le filtre perceptif de Leiris ait été nuancé par des

préoccupations personnelles et un primitivisme d'avant-garde alors qu'en Afrique, au cours des années 1930, des semences ont été plantées qui ont permis des expériences ultérieures - son amitié avec Aimé Césaire, plus de voyages en Afrique et dans les Caraïbes - . En ce qui concerne les épistémologies européennes et les perceptions de l'altérité, Marc Augé a observé de façon provocante que les ethnographes du type décrit dans *L'Afrique fantôme* étaient principalement engagés dans le dialogue avec eux-mêmes et que cela tend généralement à être le cas (Augé 1992: 36-40).

Il semble difficile de contester la vision euro centrique de l'art africain et son parrainage par l'establishment artistique en France pendant les années de l'entre-deux-guerres. La mission Dakar-Djibouti, par exemple, a été financée en partie par le gouvernement français et par des soirées de cravate noire organisées en tant que fonds de levée, auxquels ont assisté des hommes blancs à chaussures brillantes. La tendance de «l'art nègre», comme on l'appelait, pendant les années d'entre-deux-guerres attirait à la fois l'establishment et ses dissidents; Il s'adressait aux marchands d'art riches ainsi qu'aux artistes d'avant-garde et aux ethnographes. Le parrainage et l'intérêt divers de la mission Dakar-Djibouti montrent comment le primitivisme français était un phénomène culturel moins lié à la décentralisation de l'autorité culturelle française qu'à l'adaptation de certains aspects de l'identité française à la consolidation de l'empire colonial français. En tant qu'événement, la mission Dakar-Djibouti suggère un processus élaboré d'interprétation et de tri des différences culturelles. La première ethnographie française a pris forme ainsi dans le cadre d'une tentative de dériver un vocabulaire conceptuel permettant d'articuler les mutations internes de la culture française résultant d'un engagement sans précédent avec les cultures et les populations des territoires d'outre-mer français. Il a fallu du temps, cependant, pour que les pratiques culturelles qui ont été inspirées par le primitivisme d'avant-garde se développent en des formes plus réfléchies d'enquête culturelle. A ce stade, le primitivisme d'avant-garde partagé par les surréalistes et les surréalistes dissidents devenus ethnographes provient de leur propre désillusion avec la société européenne en tant qu'intellectuels français. Au cours de l'entre-deux-guerres, l'appropriation conceptuelle de cette lointaine altérité par l'avant-garde française a été motivée par un désir occidental d'une existence «primitive» idéalisée, comme celle qui est venue avant la vie cosmopolite décadente.

La pratique de rejoindre l'Afrique et de ramener les artefacts culturels de la manière dont Leiris décrit inéluctablement la réinscription des relations de force fondamentales du colonialisme, qui permet à ces «matières premières» d'être façonné en produits pour la consommation métropolitaine. Les relations de pouvoir qui définissent cette forme d'«échange» culturel sont plus claires pour ceux qui sont désabusés par la transaction. En fait, la position de privilège inavoué que les artistes et les intellectuels d'avant-garde jouissaient pendant les années d'entre-deux-guerres a créé une cécité critique dans leur perception de tous les aspects des cultures «primitives» en question. Dans les fantasmes occidentaux, l'art «primitif» tend à se situer dans le passé, en association avec une existence antérieure idéalisée, et cette vision s'avère incompatible avec la vision des artistes africains comme contributeurs actifs à la redéfinition de la culture contemporaine.

Avec le temps, Leiris reconnaît les limites de son filtre subjectif dans la préface qu'il écrivit à la deuxième édition de *L'Afrique fantôme* en 1950:

"Je ne puis nier, cependant, que l'Afrique du début de l'avant-dernière décade était elle aussi bien réelle et que ce n'est pas fait à elle mais à moi il faut que je m'en prenne si Les problèmes humains qui s'y posent déjà ne m'ont frappé que les enlées revêtent l'aspect d'abus absolument criants, sans m'arracher pour autant à mon subjectivisme de rêveur "(Leiris 1981: 12).

Le subjectivisme de rêveur de Leiris ne lui permettait pas de s'engager sérieusement dans les réalités sociales et politiques qu'il rencontrait, bien qu'il reconnaisse rétrospectivement qu'elles étaient là pour lui voir. Le vocabulaire qu'il utilise pour décrire les limites de sa perspective en 1931 évoque explicitement le surréalisme et sa conception du sujet. Comme Leiris décrit son changement de perspective, il se réfère, comme Breton, à son expérience de la Seconde Guerre mondiale:

"C'est un livre bien dépassé par la situation - et pour moi bien vieilli - que cette Afrique fantôme réimprimée aujourd'hui quelques années après la mise au pilon, pendant l'occupation allemande, de presque tout le reliquat de sa première édition" (Ibid., P. 11).

Leiris discute de la censure de *L'Afrique fantôme* sous Vichy France, ainsi que d'un changement d'attitude face aux questions de race, de nation et de formes de protestation politique. La Seconde Guerre mondiale a aidé l'intelligentsia de gauche en France à voir l'exploitation coloniale et la réaction de la Négritude à une nouvelle

lumière. En fin de compte, Leiris allait plus loin que Breton en mettant à jour ses idées et ses valeurs avec les temps. Au cours de la période d'après-guerre, Leiris devient rédacteur en chef de la revue *Les Temps modernes* de Jean-Paul Sartre, où il présente la poésie d'Aimé Césaire.

Essentiellement, il l'a écrit pour lui-même. Il croit qu'il a déjà dit que c'était un livre expérimental, documentaire. Il avait plein de littérature, surtout de surréalisme; Il avait eu plus que lui pourrait prendre de la civilisation occidentale. Il voulait voir ce qui se produirait quand il s'est forcé à enregistrer pratiquement tout ce qui s'est passé autour de lui et tout ce qui a traversé dans sa tête. C'était essentiellement l'idée de son écriture.

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RESEARCH ARTICLE

UPDATE ON THE BIOMARKERS FOR TARGETED THERAPY IN TRIPLE-NEGATIVE BREAST CANCER.

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Abstract

Breast cancers that demonstrate the absence of estrogen receptor and progesterone receptor and no overexpression of human epidermal growth factor receptor 2 (HER2) are referred to as triple-negative breast cancer (TNBC). Globally 1 million cases of breast cancer are diagnosed annually and Triple-negative breast cancer comprises 15%-20% of all breast cancers. The last decade has seen a revolution in the understanding of breast cancer, with new classifications proposed that have significant prognostic value and provide guides to treatment options. There are many biomarkers in TNBC used for its sub-classification. Clinically-practical assay/biomarkers that can reliably identify TNBC are necessary. Biomarkers may be useful as prognostic or predictive indicators as well as suggest possible targets for novel therapies.

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Introduction:-

Triple-negative breast cancers (TNBCs) are defined by the absence of estrogen and progesterone receptors and the absence of HER2 overexpression. These cancers represent a heterogeneous breast cancer subtype with a poor prognosis. In 2012 about 1.7 million women worldwide were diagnosed with breast cancer (BC), and 521,900 women died from it. (1-2) TNBC represents 10%–20% of invasive breast cancers and has been associated with African-American American race, deprivation status, younger age at diagnosis, more advanced disease stage, higher grade, high mitotic indices, family history of breast cancer and *BRCA1* mutations. TNBC is regularly reported to be three times more common in women of African descent and in pre-menopausal women, and carries a poorer prognosis than other forms of breast cancer. (3) These statistics include all subtypes of BC, but it is well known that BC is not a homogeneous disease. Tumors in the breast have long been classified according to their morphologic features, histologic type, and grade (severity). Four major intrinsic subtypes have been identified by genomic studies: the luminal subtypes A and B, which express hormone receptor-related genes, basal-like (BL) BC, and HER2-positive BC. (4) More recently, gene expression analysis using DNA microarray technology has identified additional breast tumor subtypes that were not apparent using traditional histopathologic methods. Based on gene expression profiles, breast cancer can be classified into 5 main groups Luminal A, Luminal B, Basal-like, HER2, Normal breast-like. (5-6) Most breast cancers originate from the inner ("luminal") cells that line the mammary ducts. Luminal A and luminal B tumors are similar in that both are typically ER+ or PgR+, or both. However, they are dissimilar in that the A type is usually HER2- and the B type is more likely to be HER2+ and lymph node-positive. Women with luminal A tumors are often diagnosed at a younger age. They tend to have the best prognosis, with relatively high rates of overall survival and relatively low rates of recurrence. Those with luminal B tumors tend

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to have a higher tumour grade and a poorer prognosis (7-8). TNBC is a distinct breast cancer. It is classified into six groups based upon the GEP and DNA microarray. This sub-classification is not only useful in understanding the disease better but also to find molecular targets for its treatment (9).

BL-1 and BL-2:-

The BL-1 subtype was found to be composed rapidly dividing cells associated with increased proliferation and cell cycle checkpoint loss consistent with the increased expression of DNA damage response genes. Due to its high proliferation rate it has increased Ki67 mRNA expression and it is more responsiveness to antimetabolic agents targeting cell cycle. The BL-2 subtype on the other hand displayed unique gene ontologies involving epidermal growth factor signaling as well as glycolysis and gluconeogenesis pathway. On microarray it showed a higher expression of epidermal growth factor receptor (EGFR), TP63, MET, *etc.*

Immunomodulatory subtype:-

Immunomodulatory (IM) is composed of immune cell responses such as immune cell and cytokine signaling, antigen presentation and processing and signaling of immune transduction pathways. Its GEP substantially overlaps with the medullary breast cancer, histologically a rare distinct form of TNBC which carry favorable prognosis despite its high grade.

Mesenchymal and mesenchymal stem like subtype:-

On GEP these subtypes consist of epithelial-mesenchymal (M) transition and growth factor pathways. The mesenchymal stem like subtype is also expressed by genes involved in angiogenesis including VEGFR2 and was found to be highly responsive to dasatinib [tyrosine kinase (TK) inhibitor], and mTOR inhibitors.

Luminal androgen receptor subtype:-

This subtype is characterized by androgen receptor (AR) signaling. It is ER negative but gene ontologies were heavily composed of hormonally regulated pathways such as steroid synthesis, porphyrin metabolism and androgen/estrogen metabolism. AR mRNA expression was nine times higher than other subtypes therefore, these lines were found to be highly sensitive to AR antagonists enzalutamide. Patients with this subtype had decreased DFS and OS.

Biomarkers in TNBC:-

Identification of molecular markers such as expression of the estrogen (ER) and progesterone receptors (PgR) and the human epidermal growth factor receptor 2 (HER2) has offered additional predictive value for the therapeutic assessment of women diagnosed with breast cancer. TNBC is characterized by the marked expression of certain biomarkers. The presence of these molecules though is not restricted to TNBC but somehow show increased prevalence in this subgroup. The following are the important biomarkers in TNBC.

Epidermal growth factor receptor (EGFR):-

The four receptors being EGFR (or ErbB-1), HER-2/neu (ErbB-2), HER-3 (ErbB-3), and HER-4 (ErbB-4). The inactive monomer receptor dimerizes after ligand activation followed by TK, the intracellular domain of the receptor is activated by autophosphorylation, leading to a cascade of intracellular events (10). Epidermal growth factor receptor (EGFR) is a transmembrane tyrosine kinase receptor that plays a central role in regulating cell division and death. It belongs to the HER family of receptors, which includes EGFR (HER1/ErbB1), ERBB2 (HER2/neu), ERBB3 (HER3), and ERBB4 (HER4). These receptors are characterized by an extracellular ligand-binding domain, a transmembrane domain, and a cytoplasmic domain containing the tyrosine kinase region followed by a carboxy-terminal tail with tyrosine autophosphorylation sites as depicted in figure 1 (Honey V et al: 1/10/2013 AACR). EGFR signal cascade is important for cell proliferation, angiogenesis, metastatic spread, and the inhibition of apoptosis. Most of the TNBCs express EGFR, and poses a strong therapeutic challenge (11).

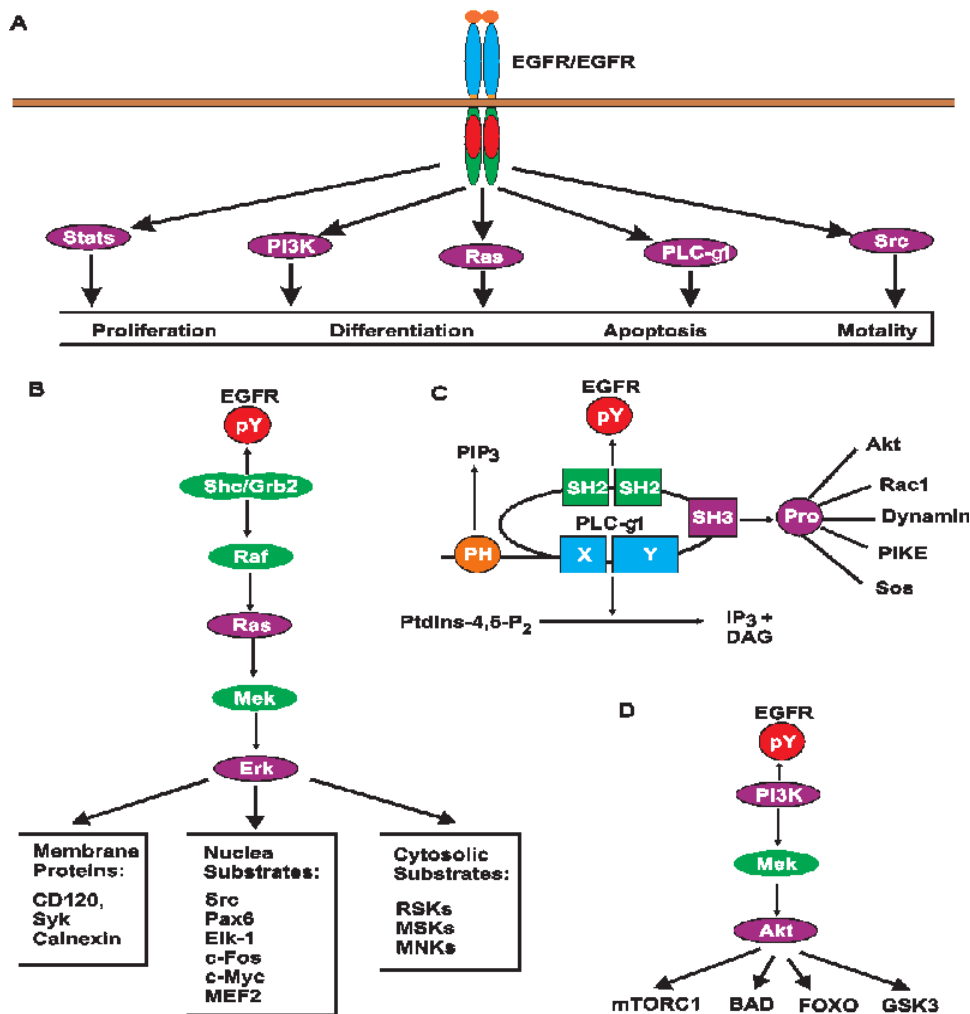


Figure 1:-Honey V et al: 1/10/2013 AACR.

EGFR expression is found in 40%-50% of patients with breast cancer and in 80% of TNBC; and is estimated to substitute major proliferation pathways of breast cancer induced by activation of HER-2, ER, PR proteins which are thereby absent in TNBC(12). EGFR expression in TNBC is associated with poor response to chemotherapy (13).Nogi et al (14),observed that EGFR was expressed in 24% of the TNBC patients and was related to less favorable response to chemotherapy and poorer survival and on the contrary the luminal groups where EGFR expression showed a good response to chemotherapy and better survival. Recently EGFR has been defined with other markers to differentiate BL subtype from TNBC (15).**Epidermal growth factor receptor (EGFR) is more frequently expressed in TNBCs than in non-TNBCs. Therefore, the EGFR is considered a candidate treatment target for TNBCs.TNBCs have an aggressive clinical course, and EGFR might be candidate therapeutic targets in this disease. This aids in segregating TNBC into subtypes and thus defining the prognostic difference and molecular target specification between the two. So EGFR is a biomarker in TNBC and a target for cetuximab, a TK inhibitor.**

Vascular endothelial growth factor:-

High VEGFR2 expression, EGFR expression, and EGFR gene copy number were significantly correlated to TNB, supporting their role as putative candidate biomarkers for selection of targeted therapy in TNB. Vascular endothelial growth factor (VEGF-A) is considered a prognostic indicator for clinical outcome in breast cancer.Angiogenesis is important for tumor growth and spread especially beyond a diameter of 2 mm as oxygen and nutrients cannot diffuse beyond this distance. Angiogenic signals are mediated by vascular endothelial growth factor (VEGF) to aid neovascularisation. VEGF protein is found in 4 isoforms because of alternative splicing of its mRNA(16). Among the different isoforms VEGF165, the 165-amino acid molecule is more common (17). Its gene

expression is controlled by many of stimuli such as hypoxia, nitric oxide, growth factors, oncogenes, tumor suppressor genes and HER-2 (18). Vascular endothelial growth factor A (VEGF-A) and vascular endothelial growth factor receptor 2 (VEGFR2) are often coexpressed in breast cancer, and potentially affect cellular pathways and key proteins such as the estrogen receptor (ER) targeted by endocrine treatment. In TNBC higher VEGF levels are associated with shorter DFS, OS, and DDFS. Also, VEGF levels have been significantly related to the size of the tumor, grade and metastatic sites. In patients with higher VEGF levels disease progressed despite of therapy and such patients were associated with significantly lower progression free survival as compared to patients with lower levels. In TNBC patients it was found that VEGF level elevated from baseline to middle of the therapy significantly but showed a non significant increase from the middle of the therapy to its end when patients were administered FAC (19). VEGF is a target for bevacizumab in TNBC patients. High VEGFR2 expression was significantly correlated to decreased BCSS in TNB patients. TNB was associated with decreased BCSS and clinicopathological characteristics of an aggressive tumor type. High VEGFR2 expression was significantly correlated to TNB, supporting their role as putative candidate biomarkers for selection of targeted therapy in TNB (20).

Poly(ADP-ribose) polymerase (PARP):-

The discovery of poly(ADP-ribose) >50 years ago opened a new field, leading the way for the discovery of the poly(ADP-ribose) polymerase (PARP) family of enzymes and the ADP-ribosylation reactions that they catalyze. Although the field was initially focused primarily on the biochemistry and molecular biology of PARP-1 in DNA damage detection and repair, the mechanistic and functional understanding of the role of PARPs in different biological processes has grown considerably of late. *PARPs* are a family of cell signaling enzymes present in eukaryotes, which catalyzes the poly(ADP-ribosylation) of DNA binding proteins (21-22). Till now eighteen enzymes of *PARPs* has been detected, but *PARP1* the most common isoform. *PARP1* is responsible for the majority of its functions. The main function of *PARP1* is as DNA damage nick sensor. It forms polymers of ADP-ribose and nicotinamide with the use of NAD^+ . Activation of *PARP1* is important in tumors because of three interesting biological reasons: First, it plays a vital role in DNA repair through base excision repair pathway; second, it is capable of depleting cellular energetic pools, which results in cell dysfunction and necrosis; and third, its ability to promote the transcription of proinflammatory genes. *PARP* enzymes are involved in cellular response to inflammation, ischemia and oxidative stress. *PARP1* are involved in all these cellular processes, indicating the possible link between *PARP1* function and carcinogenesis (23). *BRCA2* operates through excision repair pathway like *BRCA1*, mutation of this gene make the cells susceptible to *PARP* inhibitors as well (24,25). *PARP* also plays a vital role in DNA repair as *BRCA*. Unlike *BRCA* it recognizes SSBs and repairs by base excision repair pathway. *PARP* inhibitors are effective in TNBC because damage to one of the arms of the DNA could not be repaired by homologous recombination due to *BRCA* mutation and *PARP* inhibition in synergism will create a state of "synthetic lethality" - a process that occurs when inactivation of individual genes have no effect but mutations in both the genes lead to death of cancer cells (26). So *BRCA* mutation is responsible for the action of many chemotherapeutic agents in TNBC. The inhibition of *PARP1* is also known to potentiate the effect of ionizing radiation and many drugs such as DNA methylating agents, topoisomerase inhibitors, and platinum compounds. Studies in mouse models have shown that the addition of *PARP* inhibitors with platinum compounds increases RFS and OS (27) while many of other studies on cell lines reveal that the activity of *PARP* inhibitors was increased in presence of *BRCA* mutations or dysfunction (28). *PARP1* has been targeted as therapeutic option in TNBC with drugs like iniparib, olaparib etc though not found to be independently helpful but their addition to cytotoxic agents have surely brought synergism to their activity and improvement in treatment response in TNBC patients

MicroRNAs Biomarkers:-

To date, the treatment strategies for TNBC are limited to surgery, chemotherapy and radiation, owing to the lack of effective therapeutic targets. Therefore, it is important to identify specific targets for TNBCs. MicroRNAs (miRNAs), a family of small non-coding RNAs regulating gene expression, are an emerging class of regulators of various biological processes, including cell proliferation, invasion, epithelial-mesenchymal transition (EMT) and drug resistance. Actually, miRNAs may serve as a novel therapeutic target in TNBC. In a more recent study, 34 miRNAs were observed to be significantly differentially expressed among the luminal A, HER2-amplified and triple negative cell lines (29), and the biologic profiling of miRNA signatures for different subtypes were also extensively explored in breast cancer tissue samples (30,31). It was functionally demonstrated that miR-498 inhibits *BRCA1* in breast cancer cell lines, and showed that inhibition of miR-498 led to reduced proliferation in the triple negative cell line Hs578T and results indicate that miR-498 regulates *BRCA1* expression in breast cancer and its overexpression could contribute to the pathogenesis of sporadic TNBC via *BRCA1* downregulation.

MiR-21 (oncomiR)-miR-21 an oncomiR was proven to regulate *PTEN* by targeting the mRNA 3'UTR, leading to an anti-apoptosis effect in TNBC (32). Specifically, miR-21 exhibits a higher expression level in TNBC than non-TNBC, and it is positively correlated with a poor clinical outcome.

miR-203 (oncosuppressor)-In contrast to the miRNAs afore-mentioned, miR-203 is an oncosuppressormiRNA which plays a specific metastatic suppressor role by targeting *LASPI* in TNBC, it can significantly inhibit cell proliferation by regulating *BIRC5* (33). Thus, the miRNAs controlling the expression of quite a few key genes are likely to affect tumor behavior and progression in TNBC. With the improvement of tumor biology understanding, an increasing number of known miRNAs have been identified to be prognostic in TNBC. Inverse correlation between miR-27b and distant relapse-free survival (DRFS) was found by performing miRNA expression profiles, while miR-342 and miR-150 were correlated with better prognosis (34).

There are some miRNAs, such as miR-210 (35), miR-155 (36), miR-27b-3p (37), miR-34b (38) and miR-21 (39), that could be poor prognosis indicators in TNBC. It is worth mentioning that the protective miR-497, was identified downregulated in TNBC vs. both normal tissue and non-TNBC cells (40), indicating its putative positive effect against aggressive malignant phenotype in TNBC. The miR-210, was up-modulated in TNBC vs. both normal and non-TNBC tissues, thereby, providing a potential therapeutic target for TNBC.

C-kit and basal cytokeratins:-

C-kit is a cytokine receptor present on the surface of hematopoietic stem cells and also in other cells. C-kit binds to stem cell factor and is a growth factor receptor that stimulates major cellular functions such as cell survival, proliferation, differentiation, adhesion and chemotaxis. It induces apoptosis and also increases the invasiveness of the cancer cells (41). KIT plays a critical role in cell proliferation and differentiation and represents a logical therapeutic target in GIST and CML. The study of the CK profile by IHC techniques is very important for tumor pathologic classification (42). CKs were earlier used to distinguish malignant breast lesions from benign ones (43), but later their prognostic value was ascertained and it was seen that expression of CK-5, CK-14 and CK-17 was related to poor prognosis, high grade tumors, ER negativity, short DFS and OS (44). It is expressed in BLBCs. Since BLBC and TNBC show overlapping features therefore C-kit and basal CKs along with other markers and pathological features are used for the differentiating BLBCs from TNBC. Many studies have revealed that presence of CKs is higher in TNBC than non-TNBC and also among TNBC subgroup it is higher in the BL subclass. BL subclass of TNBC was identified on the basis of CK and EGFR expression and when the clinicopathological features were compared between the basal and non-BL it was seen that BL subclass of TNBC was more aggressive (45).

The Ki-67 protein (also known as MKI67):-

Antigen Ki-67 is a nuclear protein that is associated with and may be necessary for cellular proliferation. Furthermore, it is associated with ribosomal RNA transcription. Inactivation of antigen Ki-67 leads to inhibition of ribosomal RNA synthesis. Ki67 is a cellular marker for proliferation. Ki67 antigen is present inside the cell nucleus during interphase and during mitosis it is relocated to the surface of the chromosomes. Since it is a marker of proliferation it is found in all cells when they are in dividing phases of the cell cycle (G_1 , S, G_2 , and mitosis) and it is absent from cells during their resting phase (G_0). Its absence in resting cells and generalized presence in dividing cells had made it a marker of cell proliferation (46). Proliferation is a salient feature for the spread of cancer and can be assessed by the IHC measurement of the nuclear antigen Ki67. Its over expression also correlates with levels of bromodeoxyuridine uptake and S-phase fraction, other markers of proliferation. Ki67 expression is less in normal breast tissue (< 3%). Recently it has been reported in many studies that Ki67 antigen and steroid-receptor are expressed in different cells in normal human breast epithelium. Ki67 was over expressed particularly in ER-negative cells and its expression in carcinoma cells was much higher (47-48). In TNBC, it was found that Ki67 levels were significantly increased in ductal TNBC compared to other histologic types (80% in TNBC vs 10%-30% in other types). Its expression also represented a direct co relation with tumor size and grade in TNBC patients and higher levels (> 35% staining) were linked with an increased risk of death (49-51). In TNBC patients Ki67 accumulation was associated with a higher pCR to chemotherapy but poor RFS and OS. Its expression was also used for subdivision of TNBC into two subtypes where only 26.7% of TNBC patients showed lower Ki67 expression (52).

DNA topoisomerase 2-alpha:-

Amplification or deletion of the topoisomerase II alpha (*TOP2A*) gene in breast cancers has been postulated to be more closely associated with responsiveness to anthracycline-containing chemotherapy than amplification of the

human epidermal growth factor receptor type 2 (*HER2*) gene. Patients with *TOP2A* overexpression have a worse prognosis compared with those with *TOP2A* nonexpression, and *TOP2A* may be a useful biomarker in patients receiving adjuvant taxane-platinum regimens with moderate- to high-risk endometrial cancer (53-54). DNA topoisomerase 2- α is an enzyme that in humans are encoded by the *TOP2A* gene. This gene encodes a DNA topoisomerase, an enzyme that controls and alters the topologic states of DNA during transcription. This nuclear enzyme is involved in processes such as chromosome condensation, chromatid separation, and the relief of torsional stress that occurs during DNA transcription and replication. It catalyzes the transient breaking and rejoining of two strands of duplex DNA which allow the strands to pass through one another, thus altering the topology of DNA. Two forms of this enzyme exist as likely products of a gene duplication event. The gene encoding this form, α , is localized to chromosome 17 and the β gene is localized to chromosome 3. The gene encoding this enzyme functions as the target for several anticancer agents and a variety of mutations in this gene have been associated with the development of drug resistance. Reduced activity of this enzyme may also play a role in ataxia-telangiectasia (55). In TNBC or breast carcinoma the gene acts as a target for anthracycline therapy which is a topoisomerase II inhibitor (56). So it is a marker for the evaluation of resistance to the anthracycline therapy. A study revealed a higher expression of *TOP-2A* in 2.7% to 8.8% of TNBC patients (57). Its over expression in TNBC leads to the decreased sensitivity towards the anthracyclines and thus decreased response. *TOP2A* expression is a marker of the tumor's proliferation rate and sensitivity to anthracycline-based chemotherapy, and does not depend on the amplification of its gene.

MAP kinase pathway:-

Mitogen-activated protein kinase (MAPK) pathways are evolutionarily conserved kinase modules that link extracellular signals to the machinery that controls fundamental cellular processes such as growth, proliferation, differentiation, migration and apoptosis (58). MAPK pathways are comprised of a three-tier kinase module in which a MAPK is activated upon phosphorylation by a mitogen-activated protein kinase kinase (MAPKK), which in turn is activated when phosphorylated by a MAPKKK. To date six distinct groups of MAPKs have been characterized in mammals; extracellular signal-regulated kinase (ERK)1/2, ERK3/4, ERK5, ERK7/8, Jun N-terminal kinase (JNK)1/2/3 and the p38 isoforms $\alpha/\beta/\gamma$ (ERK6)/ δ (59). The mitogen-activated protein (MAP) kinase cell signaling pathway promotes cell growth, proliferation, and survival. The MAP kinase pathway may be activated via several different routes, including upstream growth factor receptor tyrosine kinases and downstream mutations in pathway gene components (60). The MAP kinase pathway has also been evaluated as a potential target in TNBC, with corresponding biomarker studies performed. Preclinical evaluation of 21 breast cancer cell lines treated with MEK1/2 inhibitor trametinib demonstrated the greatest sensitivity in the 11 TNBC cell lines evaluated. The authors ultimately identified DUSP6, a phosphatase that decreases pERK2 activity upon MAPK pathway activation, as a potential marker of sensitivity to the drug. Expression of this gene was associated with greater sensitivity to trametinib in multiple solid tumor cell lines, though was not evaluated in the breast cancer models (59). Mutations that serve as targets for currently available agents, including *EGFR*, *HER2*, *KRAS*, and *BRAF* mutation as well as *MLL4-ALK* fusion and *EGFR* copy gain have also been studied in TNBC. Of 65 TNBC specimens, the only abnormalities identified were a *HER2* mutation in one patient and *EGFR* gene amplification in a second patient (61). This suggests little utility for use of these mutations as biomarkers or as potential targets for therapy. These numerous studies are encouraging in their efforts to identify biomarkers for TNBC in the setting of novel targeted agent development. However, the best data for any of these biomarkers has still been developed in only small preclinical or retrospective studies. There is hope that these promising findings will be further evaluated in large, prospective trials, and that coordinated drug and biomarker development may be central to such trials in the future.

Conclusion:-

Evaluation of biomarkers for currently available chemotherapeutic agents should focus on the mechanism and characteristics of each agent separately, rather than on chemotherapy in general. With these changes in the development of predictive biomarkers for TNBC comes the possibility for exciting advances in this field for the future. TNBC is the most poorly understood and is refractory to current targeted therapies. It is a cause of significant breast cancer mortality because of very few treatment options. A biomarker may be useful as prognostic or predictive indicators as well as suggest possible targets for novel therapies.

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RESEARCH ARTICLE

THE BENEFITS OF PROBLEM POSING IN THE LEARNING OF MATHEMATICS: A SYSTEMATIC REVIEW.

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Problem posing; Mathematics learning;
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Abstract

Problem posing has received considerable attention recently by researchers in mathematics education. Researchers in mathematics education have reported that problem posing is an effective strategy for teaching and learning mathematics. This study was carried out to bring to limelight the benefits of problem posing intervention in learning mathematics. The study was a systematic review of empirical studies on the effects of problem posing instructional strategy in the learning of mathematics. Search for the studies was both electronically and manually, and many studies were found, but only 16 studies met the requirements for inclusion, and the participants involved in these studies were 1871. The studies that met the requirements were examined in depth, especially in terms of the title, study design, participants and findings of each study. It was revealed that all the studies reported significant improvement in the learning outcomes studied. The learning outcomes related to all the 3 learning domains of Bloom's taxonomy (cognitive, psychomotor, and affective). Problem posing instructional strategy was reported to have impacted positively on the students' learning of mathematics. Problem posing instruction was shown to support knowledge-based, skill-based, ability-based and attitude/belief-based learning. It was advocated that problem posing be made an integral part of the mathematics curriculum.

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Introduction:-

Researchers in mathematics education view problem posing as a significant mental activity in mathematics (e.g. Rosli, Capraro & Capraro, 2014; Sengul & Katranci, 2012; Lavy & Shriki, 2007; Pittali, Christou, Mousoulides & Pitta-Pantazi, 2004). In fact, some researchers referred to it as being at the heart of mathematics (Kojima, Miwa & Matsui, 2015). Problem posing is a research topic in mathematics that is currently receiving the attention of many mathematics educators. Researchers such as Akay and Boz (2008) and Lavy and Shriki (2007) pointed to problem posing as an important strategy for teaching and learning mathematics, while others consider it an important skill that helps in solving problems (Kojima, Miwa & Matsui, 2015; Guvercin & Verbovskiy, 2014; Silver, 1994). Akay and Boz (2008) pointed a close relation between problem posing as an approach and academic success.

There is a general consensus of researchers in Mathematics education that problem posing is the creation of new problems and the recreation of given problems. For instance, Stoyanova and Ellerton (1996) see posing problem as

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the process by which students create personal explanations of concrete situations and formulate such situations into sensible mathematical problems. Similarly, Pittalis, Christou, Mousoulides and Pitta-Pantazi (2004) define posing problems as creation of new problems and reformulation of given problems. This study considers problem posing as the creation of new problems and changing certain parameters of existing problems.

Posing problems in the learning of mathematics:-

The benefits of posing problems have been pointed out by many researchers in mathematics education. For instance, Pittalis et al (2004) pointed out that students could solve mathematical word problems as a result of using problem posing as an instructional strategy. Similarly, English (1997) found that problem posing afforded teachers the opportunity to comprehend students' thinking about concepts and processes in mathematics. Research has also indicated that problem posing improves students' thinking and problem solving skills (Guvercin & Verbovskiy, 2014), attitudes and confidence in mathematics (Guvercin, Cilavdaroglu & Savas, 2014). Similarly, according to Lavy and Shriki (2007) and Stoyanova (1999) posing problem fosters flexible thinking, and enhances students' problem solving skills, reasoning and reflection. Problem posing is a necessary part of learning mathematics (Sengul & Katranci, 2012; Lavy & Shriki, 2007). Kojima, Miwa and Matsui (2015) say it is an important activity in mathematics education, and lies at the heart of mathematical activity. They assert that problem posing is a necessary skill for solving problems. Singer, Ellerton and Cai (2013) assert that posing problem raises creativity in students and adds to mathematics talent and independent learning. Posing problem is viewed as an investigative-oriented instruction, and also as a distinct feature of creative activity (Silver, 1994). In fact, Ticha and Hospesova (2009) point out the potential benefit of problem posing as the diagnosis of teacher-students' subject teaching knowledge. In other words, problem posing can detect the nature of the student's mathematics learning problem.

Similarly, Ticha and Hospesova (2006) assert that problem posing is one of the potential options for pre-service primary mathematics teachers to acquire professional aptitude in mathematics. According to Ticha and Hospesova (2009), experience has revealed that pre-service primary mathematics teachers' subject teaching competence is dependent on their ingenuity to create mathematical problems. This, therefore, underscores the importance of engaging in activities that are targeted at problem posing as an instructional strategy in the teaching and learning of mathematics. This importance has been pointed to by many mathematics education researchers such as English (1997), Pittalis et al (2004), Akay and Boz (2008), and Cankoy and Darbaz (2010). These mathematics educators and a host of others have stressed the desire and importance for students to acquire the ability to pose problems. Ticha and Hospesova (2009) opine that problem posing should not be seen only as a goal of teaching, but also as a means of teaching. Problem posing relates to competence in mathematics and, this is the reason for emphasizing that students be given the opportunity to create their own problems in the learning of mathematics. There is a consensus among mathematics educators that problem posing enriches student's learning of mathematics, and guides the students to a deeper understanding of mathematical concepts (e.g. Abu-Elwan, 2002; Guvercin & Verbovskiy, 2014; Haghverdi & Gholami, 2015).

This study focused on searching the literature to highlight the benefits of problem posing in the learning of mathematics. It is a systematic review of experimental studies that have been conducted on the effect of problem posing as an instructional strategy in the teaching and learning of mathematics. Many experimental studies have been carried out in mathematics with problem posing as an instructional strategy (e.g. Abu-Elwan, 2002; Akay & Boz, 2008; Priest, 2009; Kesa, Kaya & Guvercin, 2010; Guvercin & Verbovskiy, 2014; Kojima, Miwa & Matsui, 2015). And a variety of outcomes have resulted from such studies. There is therefore the need for a systematic review of the literature to highlight these results, especially that the studies are conducted at various locations on the continent.

Purpose of the study:-

Research has shown that there are many studies in mathematics education that used problem posing as an instructional strategy to determine students' achievement in mathematics, and so the desire to investigate the benefits of problem posing is timely. Research evidence has indicated improvements on students' learning of mathematics through problem posing intervention. The purpose of this study was to answer the question: do students benefit more when problem posing is used as an instructional strategy in the teaching and learning of mathematics? The study employed a systematic review of empirical studies to investigate the effect of problem posing on students' learning of mathematics. This was done by considering experimental studies only.

Method:-**Criteria for the selection of studies to be included:-**

The first step was to determine the keywords to be used for searching on the website. These words or phrases included, *problem posing in mathematics*, *effect of problem posing in mathematics*, *problem solving and problem posing in mathematics*, *experimental studies in mathematical problem posing*, *investigation of problem posing in mathematics* and *effectiveness of problem posing*. The next step was a manual search in the Journals of Mathematics Education and other Multidisciplinary Journals. The search was restricted to only journals written in English Language. Only experimental studies that indicated clearly the research design, respondents/population, and the results were considered as meeting the requirements for inclusion. However, the literature review in the background was not restricted to review of experimental studies only.

Upon these searches, many studies relating to problem posing in mathematics were found. In fact 3,815 studies resulted from these multiple searches. But most of the studies found did not meet the requirements for inclusion. Majority not experimental, some were duplicates, others inaccessible or not traceable, and a few others not published in English Language. Therefore, all those studies that did not meet the requirements for inclusion were discarded. The experimental studies were examined in depth with respect to the eligibility criteria for inclusion. Applying these criteria only 16 studies were retained, and the analysis is shown in Table 1.

Table 1:- Experimental Studies on Effects of Problem Posing.

Author and date	Title of study	Participants	Results/findings
Abu-Elwan (2002)	Effectiveness of problem posing strategies on prospective mathematics teachers' problem performance	Student-teachers 50	1.Problem posing significantly improved the mathematics achievement of the experimental group 2. Experimental group exhibited significantly higher level of problem posing performance compared to the control group.
Akay & Boz (2008)	The effect of problem posing oriented Calculus-II instruction on academic success	Pre-service teachers 79	1.Problem posing approach increased the academic success of the experimental group 2. A close and positive relation between academic success and problem posing approach was found.
Akay & Boz (2010)	The effect of problem posing oriented analysis II course on the attitudes toward mathematics and mathematics self-efficacy	Elementary pre-service teachers 82	1. Significant differences were found in the participants' attitudes toward mathematics and mathematics self-efficacy between the experimental and control groups. 2. The experimental group showed positive attitude toward mathematics.
Cankoy & Darbaz (2010)	Effect of a problem posing based problem solving instruction of understanding problem (UP)	3 rd grade students 53	1. Experimental group students were better than the control group students in terms of UP scores in all dimensions (rephrasing, visualization and qualitative reasoning). 2. Experimental group was better than control group in finding missing information in a given problem and the contradictions of a problem.

Demir (2005)	The effect of instruction with problem posing on tenth grade students' probability achievement and attitudes toward probability	Tenth Grade 82	<ol style="list-style-type: none"> 1. The students taught by problem posing instruction had significantly greater probability achievement than students taught by traditional method. 2. There was a significant difference between the mean scores of the students who received instruction with problem posing and those who were taught without in terms of attitudes toward probability.
English (1997)	The development of fifth grade children's problem posing abilities	Fifth Grade children 27	<ol style="list-style-type: none"> 1. Substantial development was exhibited by children who participated in the program
English (1998)	Children's problem posing writing formal and informal contexts.	Year 3 in elementary school 54	<ol style="list-style-type: none"> 1. Children's participation in the problem posing program led to greater diversity in the problems posed.
Fetterly (2010)	An exploratory study of the use of a problem posing of pre-service elementary education teachers' mathematical creativity, beliefs, and anxiety.	Elementary Pre-service teachers 32	<ol style="list-style-type: none"> 1. It was found that mathematical creativity can be fostered and sustained by problem posing. 2. Problem posing impacted mathematics beliefs positively.
Guvercin & Verbovskiy (2014)	The effect of problem posing tasks used in mathematics to mathematics academic achievement and attitudes toward mathematics	High school students 54	<ol style="list-style-type: none"> 1. Problem posing instruction significantly increased students' mathematical academic achievement. 2. Gave the students more confidence and positive attitudes toward mathematics. 3. Problem posing encouraged students to be creative, divergent and flexible in their thinking.
Guvercin, Cilavdaroglu & Savas (2014)	The effect of problem posing instruction on ninth grade students' mathematics academic achievement and retention	Grade 9 students 60	<ol style="list-style-type: none"> 1. Problem posing instruction increased students' academic achievement. 2. Higher positive attitudes toward mathematics. 3. More confidence and positive attitudes through active involvement
Haghverdi & Gholami (2015)	A study of the effect of using "What if Not" strategy in posing geometry problems	High school students 29	<ol style="list-style-type: none"> 1. Problem posing led to deeper understanding of geometrical concepts 2. Problem posing strengthened the interconnections between relevant geometrical concepts 3. Improved students' skills
Kesan, Kaya & Guvercin (2010)	The effect of problem posing approach to the gifted student's mathematical abilities	Grade eight students 64	<ol style="list-style-type: none"> 1. Increased and fostered mathematical abilities. 2. Enhanced interaction between teacher

Kojima, Miwa & Matsui (2015)	Experimental study of learning support through examples in mathematical problem posing	Undergraduates 219	and student, and makes student active in the lesson. 1. Problem posing benefited the experimental group more than the control group (i.e. experimental group posed problems with novel solutions). 2. Found that learning by posing problems was more effective.
Priest (2009)	A problem posing intervention in the development of problem solving competence of underachieving middle-year students	Year seven students 31	1.Participation in problem posing activities facilitated the re-engagement of disengaged middle-year mathematics students 2. Participation in problem posing resulted in improved problem solving competence. 3. There was increased integration of prior knowledge with new knowledge.
Walsh (2016)	Pre-service primary teachers' understanding of mathematical problem posing and problem solving: Exploring the impact of a study intervention	Pre-service primary teachers 415	1.Improved students' conception of what constituted a mathematical problem
Xia , Lu & Wang (2008)	Research on mathematics instruction experiment based on problem posing	Junior high school and grade nine students 540	1. Found significant effects on students' interest in learning mathematics. 2. Problem posing improved students' ability to pose problems. 3. Enhanced students' mathematics learning ability.

Results and Discussion:-

Description of the selected studies:-

A total of 16 experimental studies that made the criteria for inclusion in this study were reviewed as shown in Table 1. Nine studies were published between 2010 and 2016. Five studies published between 2000 and 2009, one study published in 1997 and one in 1998. These studies involved 1871 participants, made up of pre-service teachers, high school students, middle and lower grade students. All the studies employed experimental designs. Three studies used matching-only pretest-posttest control group design. Two used posttest only control group design, and the remaining eleven studies used quasi-experimental non-equivalent groups, and all were pretests-posttests control group design. The instruments used were either researcher constructed or adoption/adaption from the literature. The research targets of these studies were either on performance in mathematics, mathematics achievement in academics or attitudes and beliefs toward mathematics. The researchers were interested in determining the impact of problem posing as an instructional strategy on the performance of students, the achievements of students and attitudes of students toward mathematics. The summary of the outcomes or results of their findings is presented in Table 1. It can be seen from Table 1 that all the studies presented showed positive improvements in either performance in mathematics, achievement in mathematics or attitudes toward mathematics using problem posing as an intervention in the teaching and learning of mathematics.

Eight studies focused on effect of problem posing on mathematics performance and achievement (e.g. Abu-Elwan, 2002; Akay & Boz, 2008; Demir, 2005;Guvercin, Cilavdaroglu &Savas, 2014), and findings from these studies showed significant improvements in students' performance and achievement in mathematics. Two studies showed

that problem posing could foster and sustain mathematical creativity, enhance divergent and flexible thinking in students. In a study titled, 'Problem posing as a means for developing mathematical knowledge of prospective teachers', Lavy and Shriki (2007) pointed out that problem posing enhanced mathematical creativity, and this was corroborated by Singer, Ellerton and Cai (2013). They said that problem posing enhances creativity, adds to mathematical ability and learning independence.

Regarding attitudes and beliefs toward mathematics, six of the studies reviewed indicated that problem posing as an intervention impacted positively on the attitudes and beliefs of students toward mathematics (e.g. Xia, Lu & Wang, 2008; Akay & Boz, 2010; Demir, 2005; Fetterly, 2010; Guvercin & Verbovskiy, 2014). Pittalis, et al (2004) had pointed out that problem posing increased students' attitudes positively toward mathematics. Furthermore, the review revealed that problem posing helped students to understand the concept of what a 'problem' is, and to find missing information in the given problems. A study by Cankoy & Darbaz (2010) revealed that the participants in the treatment condition were better off than those in the control condition in terms of problem understanding. And also, the experimental group was better in identifying missing information in given problems than the control group. These authors' focus was determining the effect of a problem posing based problem solving instruction of understanding problem. The need for a problem solver to understand the problem he/she is to solve is imperative; and in fact, it is the first step toward a successful solution. The students were not only able to find missing information in a given problem, but also could detect contradictions of a problem. Walsh (2016) focus was on understanding mathematical problem posing and problem solving, using problem posing intervention. The study findings revealed that the problem posing intervention improved students' conception of what constituted a mathematical problem.

The study of Haghverdi and Gholami (2015) focused on skills in mathematics. The findings of their studies showed that problem posing improved students' skills in mathematics significantly. In fact, besides the improvement of skills, problem posing instruction strengthened interconnections between relevant geometrical concepts, and led to a deeper understanding of geometrical concepts (Haghverdi & Gholami, 2015). Another benefit of problem posing according to Kesan, Kaya & Guvercin (2010), is that it enhanced interaction between teachers and students, and this led to students being active in the lesson. Furthermore, an increase in the integration of prior knowledge with new knowledge was what Priest (2009) found. He also reported that problem posing activities facilitated the re-engagement of disengaged middle-year mathematics students. His study was on problem posing intervention in the development of problem solving competence of underachieving middle-year students. The intervention was reported to have improved the students' problem solving competence.

Conclusion:-

The findings of the empirical studies examined in this study can be grouped or categorized into the 3 Bloom's taxonomy of learning domains (cognitive, psychomotor and affective domains). The studies revealed that problem posing impacted positively on the following learning outcomes viz- knowledge-based, ability-based, skill-based and attitude/interest and belief-based. This shows clearly that every aspect of learning mathematics can be supported by problem posing instruction. For instance, the studies by Abu-Elwan (2002), Akay and Boz (2008), Cankoy and Darbaz (2010), Guvercin and Verbovskiy (2014), and Walsh (2016) indicated that problem posing intervention supported knowledge-based learning outcome. Similarly, studies by English (1997 & 1998), Abu-Elwan (2002), Xia, Lu, and Wang (2008), Fetterly (2010), and Kojima, Miwa and Matsui (2015) revealed that problem posing instruction impacted positively on ability-based learning outcome. Skill-based learning outcome was seen to have improved significantly through the use of problem posing (Abu-Elwan, 2002; English, 1997; Priest, 2009). Problem posing instructional strategy also improved participants attitudes/interest and beliefs toward mathematics (Demir, 2005; Akay & Boz, 2010; Guvercin, Verbovskiy, 2014 Guvercin, Cilavdaroglu & Savas, 2014).

It could be said with every confidence that, if properly handled, problem posing instructional strategy can be one of the most effective strategies in the teaching and learning of mathematics. This is because not only students benefit from problem posing; but teachers also do. Problem posing affords teachers the opportunity to get an insight into students' thinking processes. Problem posing revealed that mathematics anxiety and fears could be reduced significantly in students. It encourages the use of diverse representations, building of a knowledge network, development of creativity, enhancement of attitude toward mathematics and increment in self-confidence. It can safely be advocated that problem posing be an integral part of the mathematics curriculum, considering the role it can play in the teaching and learning of the subject.

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RESEARCH ARTICLE

Oral findings in Zinsser-Engman-Cole syndrome: a case report.

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Abstract

Zinsser Cole Engmansyndrome also known as dyskératosiscongenita (DC) is a very rare genetic affection characterized by the association of reticular hyper pigmentation, nail dystrophy and leucoplakia of the mucous membranes. We report the case of a 14 years old child already diagnosed with DC and which was complaining about extensive caries, and teeth loss. Clinical examination revealed a poor oral hygiene, extensive caries and periodontal disease. DC is a inherited bone marrow failure characterized by excessively short telomeres in highly proliferative tissues. Its prevalence is estimated at one in a million. DC has many clinical features and oral manifestations. It has a poor prognosis and its current treatment remains non satisfactory. DC needs a multidisciplinary approach and a regular follow-up.

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Introduction:-

Zinsser Cole Engmansyndrome also known as dyskératosiscongenita (DC) is a very rare type of ectodermal dysplasia. It's a multi-system disorder whose symptoms appear in childhood and leads to death between 16 and 50 years.

DC occurs mainly in young male with a male to female ratio of 13/1 and mostly appears between 5 and 12 years [1-2].

This genetic affection is typically characterized by the association of a traditional dermatological triad: Reticular hyper pigmentation, nail dystrophy and leucoplakia of the mucous membranes. The medullary insufficiency associated to this disease in 85, 5% is responsible for opportunistic infections [3].

Case report:-

A 14 years old male teenager was admitted to both departments of pediatric of Casablanca University following extensive caries, teeth loss and toothache.

Medical history revealed that he's diagnosed with dyskeratosis congenital after genetic and telomere length testing provided the identification of telomere disorder.

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He was lately admitted for chronic mucocutaneous candidiasis and an abscess of the right buttock. The patient suffers also from growth retardation. His skin was thin, dry and wrinkled in some areas and he showed nail dystrophy of the hands and feet. (Fig 1, 2 and 3).



Figure 1: Growth retardation and thin skin



Figure 2 and 3 : Nail dystrophy of the hand and feet

His obstetrical antecedents were without particularities and his birth weight was 4 kg without signs of suffering of any neonatal infection. His vaccination was carried out correctly according to the national immunization program, without any complication.

Family history revealed that he was born from a consanguineous marriage of second degree and that his brother which was also diagnosed with DC died 3 years ago from a lethal infection.

Extra oral examination showed facial deformity with reticular hyperpigmentation of his face and neck. The patient was pale but vital signs were within normal limits (Fig 4).



Figure 4 : Facial deformity with reticular skin hyperpigmentation

Intra oral examination revealed poor oral hygiene, leukoplakia on the tongue and the mandible gingival mucosa, extensive caries of all his first permanent molars. The patient suffered from periodontal disease which led to many early teeth loss (Fig 5).



Figure 5: Extensive caries of the first permanent and leukoplakia in the mandible gingiva

Radiographic examination, showed a bone radiolucency image around the permanent molars and the two central incisors (Fig 6).

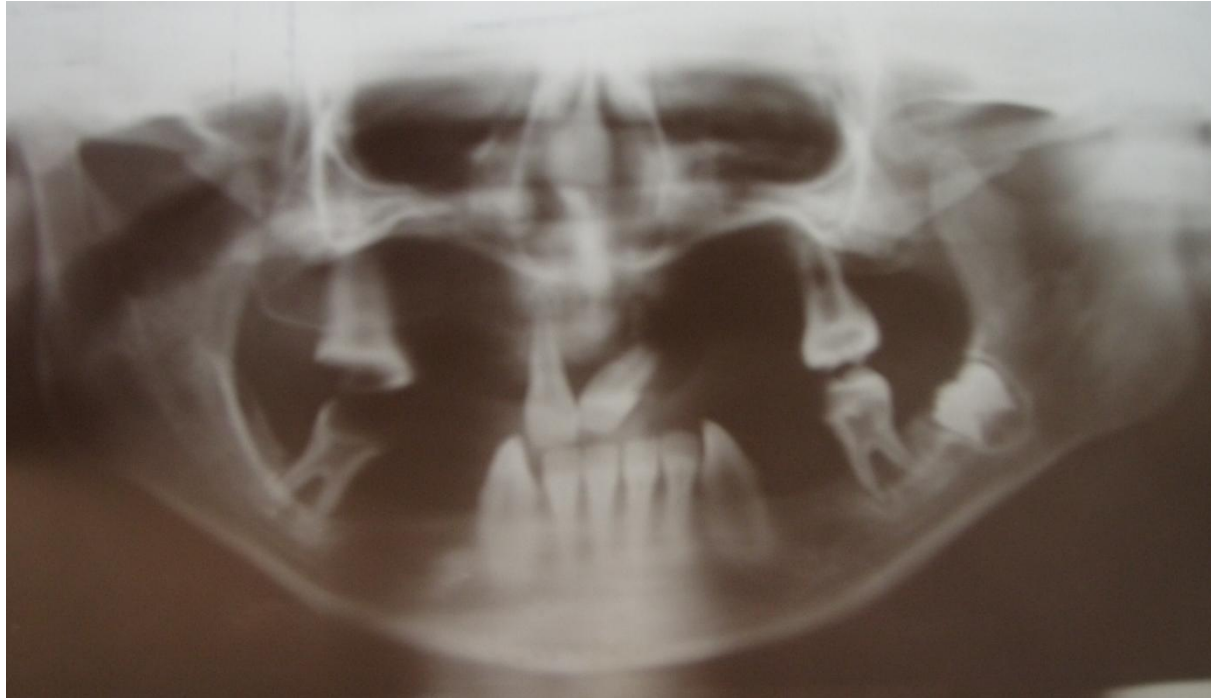


Figure 6: Bone radiolucency image around the permanent molars and the two central incisors

Periodontal treatment was performed and the first molars with the two incisors were removed. A dental prosthesis has been made and the child was very happy to find his lost smile.

Discussion:-

DC is a rare inherited bone marrow failure characterized by excessively short telomeres in highly proliferative tissues. Its prevalence is estimated at one in a million [2-3].

Five genes have been identified, all encoding for components of telomerase and involved in maintaining telomere length: DKC1 (locus Xq28, sex-linked transmission), TERC (locus 3q21-q28, autosomal dominant), TERT (locus 5p15.33, autosomal dominant), NOP10 (locus 15q14-q15, autosomal recessive) and TIN2 (locus 14q12, autosomal dominant)[4].

DC is a very polymorphous disease at the clinical level, with several modes of inheritance. Several clinical symptoms of the disease can appear after a latency period. These features make DC particularly difficult to diagnose.

The main clinical features of this multi-system bone marrow failure disease are mucocutaneous abnormalities (cutaneous hyperpigmentation, dystrophy of the nails, leukoplakia of the oral mucosa), haematopathies (pancytopenia 50%) and an increase predisposition to cancer.

Other abnormalities in other systems may also be associated: pulmonary disease fibrosis, abnormalities of pulmonary vasculature, eye defects like conjunctivitis, blepharitis, pterygium and epiphora, mental retardation, short stature, hair loss/grey hair/sparse eyelashes, eye defects and deafness [5].

The oral manifestations described in the literature are: Hypodontia, Short blunted roots, Extensive caries lesions, Delayed eruption, Taurodontism, Gingival inflammation with oedema, Alveolar bone loss, Periodontitis, Thin enamel gingival recession, Smooth atrophic tongue mucosa, Dry mouth, Angular cheilitis and Leukoplakia [3–6–7]. General hygiene recommendations include brushing teeth two to three times a day with fluoridated toothpaste and flossing once a day at a minimum to help prevent tooth decay. Some dentists recommend using prescription strength fluoridated toothpaste or antibacterial mouth rinse to aid in reducing oral disease.

Biannual dental checkups and cleanings are recommended to monitor for the presence of oral pathology and prevent the development of significant dental decay and gum disease.

Precautions during routine dental treatment may be necessary in the presence of low platelet counts and white blood cell levels.

Access to quality dental care for medically compromised patients may be challenging outside of large urban centers.

The risk of squamous cell carcinoma is high in individual with DC estimated approximately 35%, with a peak in the third decade. Many other malignant tumors and hemopathies have also been reported like bronchial, colon, larynx, esophagus, stomach and pancreas carcinomas, Hodgkin's disease and leukemia [3].

Malignant degeneration of these leukoplakia is not uncommon and it's expected after 10 to 15 years of evolution, hence the need for periodic reviews to detect signs of transformation.

DC has a poor prognosis, the current treatments remain non satisfactory.

Mortality is often associated to severe pancytopenia, development of malignancy and other multisystem complications of the disease [2].

Conclusion:-

DC needs a multidisciplinary approach: genetic, pediatric, hematologic and a special dental care. Regular follow-up of the patient is essential especially with the high possibility of malignant changes within oral and other mucosal sites.

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RESEARCH ARTICLE

POSTNATAL DEVELOPMENT OF BRUNNER'S GLAND IN RABBIT

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Abstract

Introduction- Brunner's glands are located in the submucosa of the duodenum in all mammals. The principal function of the glands is thought to be protection of the duodenal mucosa against the erosive effects of the gastric juice by virtue of the mucoid nature of its secretion, its alkalinity, and possibly by the buffering capacity of its bicarbonate content. In the present study postnatal development of Brunner's glands in the rabbit was observed in 21 rabbits.

Material & methods- This research work was done during the period between 1977-1979 as a part of thesis work. The present work, postnatal development of the duodenal submucosal glands in the four parts of the duodenum was studied in rabbit. Method-For postnatal developmental study of Brunner's glands in rabbit animals were used either the littermates or the young ones of the same parents. Rabbits of 0hrs, 1 week, 2 weeks, 3 weeks, 4 weeks, 5 weeks, and 6 weeks age formed one series. Three such series were examined. Four pieces were taken from first, second, third and fourth part of duodenum and placed in fixative 10% formal saline. Tissues were processed routinely and stained with: haematoxylin and eosin for light microscopy.

Results- The origin and development of Brunner's glands in the rabbit have been studied in postnatal animals. Brunner's glands arise principally from the bases of crypts of Lieberkuhn. The glands commence as buds which proliferate deep into the submucosa and branch to give rise to tubules and to a duct system. The Brunner's glands in rabbit extended up to the second part of duodenum at birth and their extent gradually increased distally. By the end of second week the glandular tissue reached the fourth part of duodenum. The glands were more in the proximal part of duodenum and decreased from the proximal to the distal part. Compactness of glands when traced at weekly intervals in the identical parts of duodenum showed marked increase up to sixth week. The serous cells were differentiated in the terminal portions of tubules at birth but mucous acini started differentiated only after two weeks.

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Introduction:-

The duodenal glands or the glands of Brunner are present in all mammals and are believed to secrete an alkaline fluid containing mucin. The duodenal glands were first observed in 1679 by Wepfer¹ who described them during the autopsy. Wepfer¹ described their arrangement in the duodenum and observed that when they were macerated in water, the duodenal glands discharged copious amounts of mucus. Nine years later, Brunner^{2,3} who was Wepfer's son-in-law, described the duodenal glands in the horse, ox, sheep, deer, dog, and beaver and presented a brief description in man. Brunner considered these glands to be accessory pancreatic tissue and termed them "glandulae duodeni" or "pancreas secundarium." Middeldorpf⁽⁵⁾ realized that the secretion of the duodenal glands differed from that of the pancreas and suggested that these glands be designated "the glands of Brunner." Florey and Harding^(6,7) found that the region of the duodenum containing the duodenal glands is more resistant to acid erosion than other regions of the small intestine. Grossman⁽⁸⁾ in 1958 has reviewed the literature with regard to these glands and recent studies Moe, 1960;⁽⁹⁾, Cochrane, Davies, Palfrey & Stockwell, 1964;⁽¹⁰⁾ Friend, 1965;⁽¹¹⁾ Leeson & Leeson, 1966⁽¹²⁾ have reported upon the fine structure of the glands in cat, guinea-pig, mouse and rat respectively. The latter studies have emphasized species differences. In the cat and in the mouse, gland cells possess features intermediate between those of mucous and serous cells. Component cells of the glands in the guinea-pig are typically mucous in type and in the rat cells appear to be more mucous than serous with regard to their fine structural appearance. Little work has been reported on the development of Brunner's glands. Barth(1868)⁽¹³⁾ reported that in the rabbit the duodenal glands sprout off from the crypts of Lieberkuhn as double outgrowths. In man, the glands first appear at approximately 78 mm (3 months) (Brand, 1877;⁽¹⁴⁾ Johnson, 1910⁽¹⁵⁾) and arise from the bottoms of the crypts of Lieberkuhn. The glands develop in a progressive fashion starting in the upper one-third of the duodenum and extending downward so that in the 120 mm stage the greater part of the duodenum contains glands of Brunner. As the glands develop they branch several times, each branch becoming arranged into a lobule. The present paper reports upon the postnatal development of the Brunner's glands in the rabbit.

Material and methods:-

This research work was done during the period (1977--1979) as a part of thesis work. Twenty one rabbits divided into seven specific age groups were used in this study. For postnatal developmental study of Brunner's gland in rabbit, animals were used either the littermates or the young ones of the same parents. Rabbits of 0 hrs, 1 week, 2 weeks, 3 weeks, 4 weeks, 5 weeks, and 6 weeks age formed one series. Three such series were examined. The animals were anesthetized by giving ether. Entire duodenum was removed from pyloro duodenal junction to duodenojejunal junction in each rabbit. Four pieces were taken from first, second, third and fourth part of duodenum and placed in fixative 10% formal saline. Tissues were processed routinely and stained with Haematoxylin and Eosin for light microscopy.

Results:-

Postnatal development of Brunner's gland in four parts of duodenum was studied in rabbits from birth (zero hour) upto six weeks of life.

Zero Hour-Tubular structures lined with stratified cuboidal epithelium having two cell layers were scattered in the sub-mucosa throughout its circumference in the first part of duodenum. Branching of tubules and their continuity with deeper parts of intestinal glands was seen at places (Fig.1). Related with the tubules, ill defined serous acinar cells were evident. Similar tubular structures but in reduced number were also seen in the submucosa of second part of duodenum and were not seen in the submucosa of third and fourth part of duodenum at all. Muscularis mucosae was seen as a fine interrupted streak like structure.

First Week-Tubular structures were seen in the greater number than that at zero hours in the submucosa of first and second part of duodenum. Few tubular structures were also seen in the submucosa of third part of duodenum but were not seen in the submucosa of fourth part of duodenum. Muscularis mucosae was seen as a thin interrupted line.

Second week-Tubular structures were seen in the increased number in the submucosa of first, second and third part of duodenum than that at first week. Few tubular structures were also seen in the submucosa of fourth part of duodenum also. (Fig.2)

Third Week-Glandular tissue in increased number with well differentiated acini were seen in the submucosa of all

parts of duodenum. In the first part well differentiated mucous acini were more in number (Fig 3) while in second and third part serous acini were more than mucous acini and only serous acini were seen in fourth part (Fig.4). In the first part glandular tissue was more dense. Ducts of the gland lined with simple cuboidal epithelium and opening into the deeper parts of intestinal glands were also seen. Well developed muscularis mucosae was observed in all parts.

Fourth Week-Well differentiated glandular tissue with increased quantity were seen in the submucosa of all parts in comparison to that at third week (Fig 5). It was most dense in first part and very less in fourth part. There was predomination of mucous acini in the first part and of serous acini in the second, third and fourth part. In the fourth part mucous acini were rarely seen.

Fifth week-Glandular tissue in the submucosa was more in comparison to that at four weeks in all parts. Predomination of mucous acini was observed in the first part and of serous acini in the third part and fourth part while of both mucous and serous acini in second part.

Sixth week-Glandular tissue in the submucosa was more in comparison to that at fifth week in all parts. Glandular lobules were seen in the submucosa as well as in the lamina propria of first part of duodenum (Fig 6). Gradual decrease in density was observed in distal parts of duodenum. Predomination of mucous acini in the first and second part of duodenum (Fig.7) and of serous acini in the fourth part (Fig 8) was seen. While in the third part mucous and serous acini were almost in equal proportion.

Discussion:-

In the present work postnatal development of Brunner's glands were studied in rabbits from birth to six weeks of age. The Brunner's gland in the rabbit at birth was represented by two cell layered tubular growth from the deeper parts of intestinal gland, branching into the submucosa in the present study (fig.1). The Brunner's glands in rabbit extended upto the second part of duodenum at birth and their extent gradually increased distally. By the end of second week the glandular tissue reached the fourth part of duodenum (fig.2). Obouforibo and Martin⁽¹⁶⁾ (1977) also described similar stage of glandular tissue in the newborn mouse. Krause and Leeson⁽¹⁷⁾ (1967), however mentioned that rat possessed all the mucosal constituents of duodenum at birth and it only differed in the extent and degree of differentiation of various structures in adults. The duodenal glands in the golden hamster developed on the first day after birth (Mann et al. 1967)⁽¹⁸⁾ and reached functional maturity by the fifteenth postnatal day. Woff⁽²²⁾ (1961) found that glands first appeared near the pylorus and developed pari passu with the duodenum. Krause and Leeson (1967)⁽¹⁷⁾ working on rat, also mentioned that the development and differentiation of Brunner's gland followed a progressive pattern, slowly extending down the duodenum till the final distribution was reached. Adult stage was achieved by third week in mouse (Obouforibo and Martin, 1977)⁽¹⁶⁾. With the increasing downward extent, the compactness of glands in the proximal part was found to be increased in the present work. Similar observations were found by Obouforibo and Martin (1977)⁽¹⁷⁾ in the mouse. Mohammadpour, A. A., (19) also found in the guinea pig the glands were well developed in the cranial part of duodenum. There was a significant difference in the thickness of duodenal submucosal glands in all parts of the duodenum that decreased from the cranial to the ascending part. In the present study compactness of glands when traced at weekly intervals in the identical parts of duodenum showed marked increase upto sixth week (fig.4 & 6). Obouforibo and Martin (1977)⁽¹⁷⁾ mentioned that the growth rate of glands in mouse was found to be diminished after third week. Cooke (1967)⁽²⁰⁾ mentioned that the serous cells differentiated a day before birth and mucous cells were four to five days after birth. In the present study also the serous cells were differentiated in the terminal portions of tubules at birth (fig.1) but mucous acini started differentiated only after two weeks. Gabe (1956)⁽²¹⁾ showed that all the cells of submaxillary glands were serous at birth and five days after changed to mucous type. In the present study no such things were found.

Florey and Harding (1933)⁽⁶⁾ and Cooke (1967)⁽²⁰⁾ mentioned that the secretion of the Brunner's glands being alkaline in nature protected the duodenum from ulceration by acid pepsin. In the present study at sixth week predomination of mucous acini in the first (Fig.6) and second part of duodenum (Fig.7) and of serous acini in the fourth part (Fig 8) was seen. While in the third part mucous and serous acini were almost in equal proportion. So the proximal part of duodenum showed large proportion of mucous acini as compared to the distal part. This could be correlated to the greater need of alkalinity in the vicinity of pyloric region which is the first to receive the acidic content of the stomach.

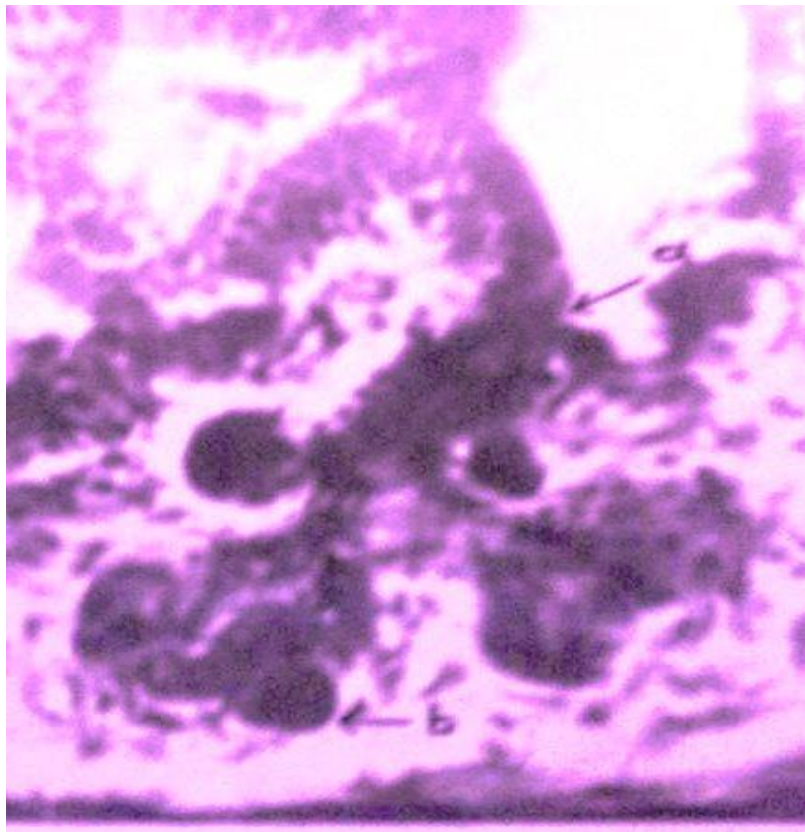


Fig. 1 Rabbit -Zero hour-H&E stain x500 Photomicrograph of T.S, of First part of duodenum showing -
a) branching of tubules and their continuity with deeper parts of intestinal glands
b) muscularis mucosae .

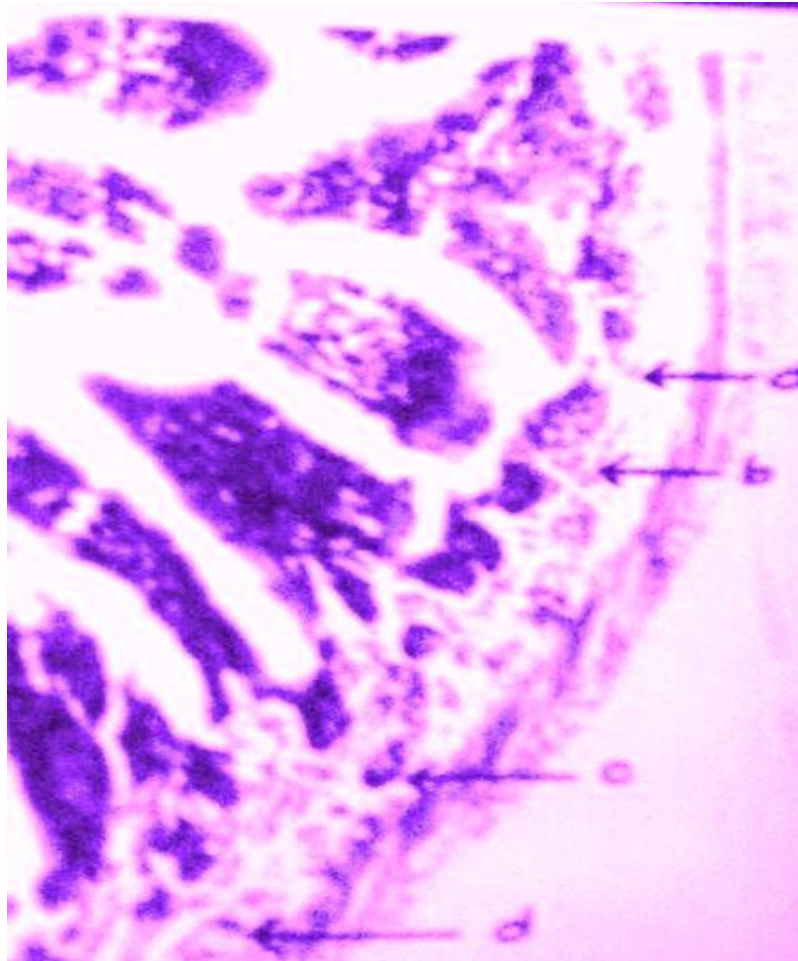


Fig. 2 Rabbit-Second week(H &E stain x80) Photomicrograph of T.S, of fourth part of duodenum showing

- a) submucosa
- b) muscularis mucosae
- c) ill defined seros acini
- d) tubular structure

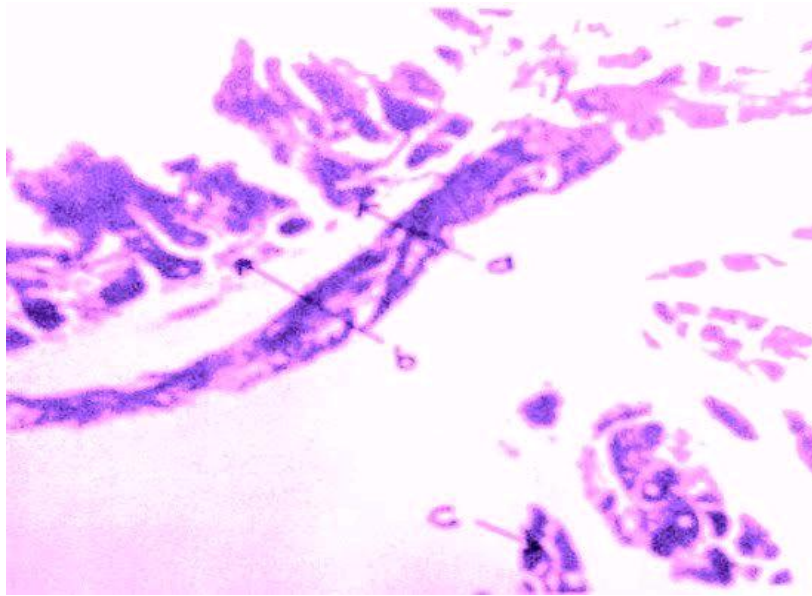


Fig.3 Rabbit-Third week(H &E stainx80) Photomicrograph of T.S, of fourth part of duodenum showing
a) submucsa having few serous acini
b) muscularis mucosae
c) pancreatic acini

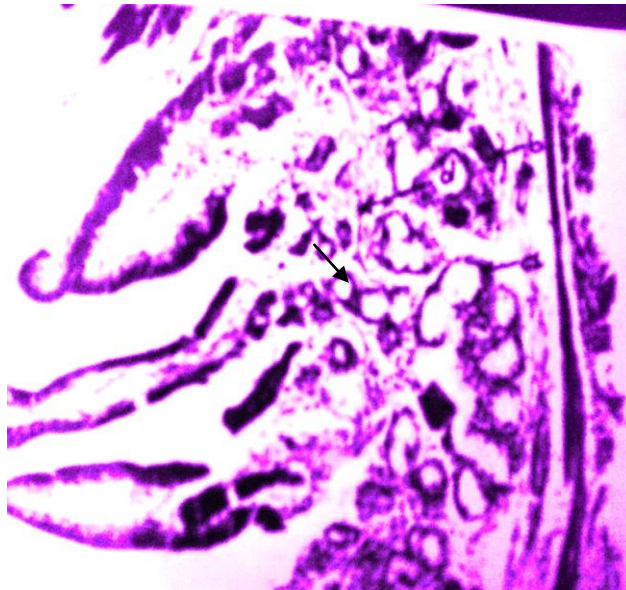


Fig. 4 Rabbit-Third week(H &E stain x80) Photomicrograph of T.S, of first part of duodenum showing-
a) well differentiated mucous acini in the submucsa
b) serous acini
c) mucous acini in the lamina propria(arrow)
d) muscularis mucosae

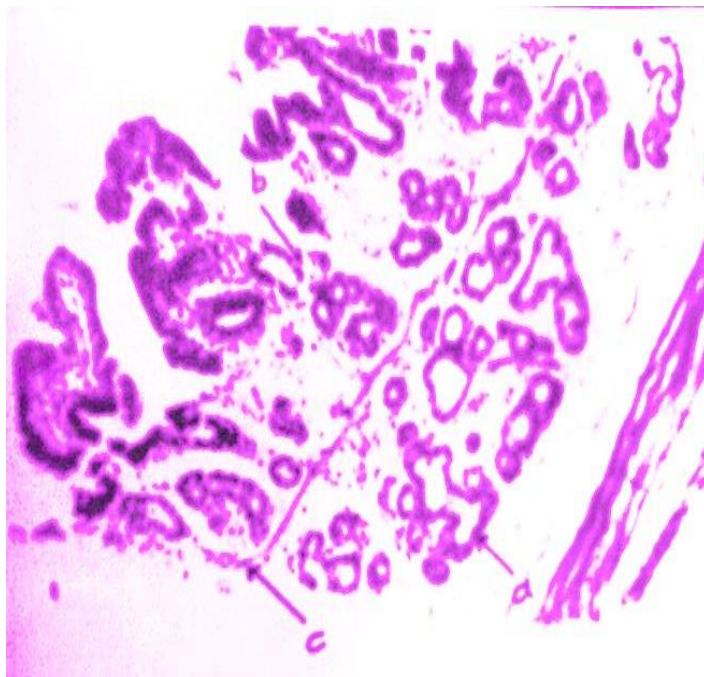


Fig. 5 Rabbit Fourth week (H &E stain) Photomicrograph T.S, of first part of duodenum showing

- a) glandular lobules in the submucosa
- b) glandular lobules in the lamina propria
- c) muscularis mucosae

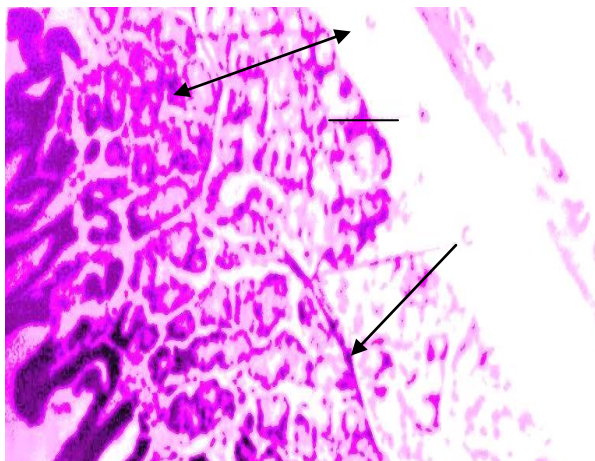


Fig.6 Rabbit-Six weeks(H &E stain x80) Photomicrograph T.S, of first part of duodenum showing

- a) glanduar lobules in the lamina propria (double arrow)
- b) glanduar lobules in the submucsa (line)
- c) muscularis mucosae branching and forming the network occupied by glandular lobules(arrow)

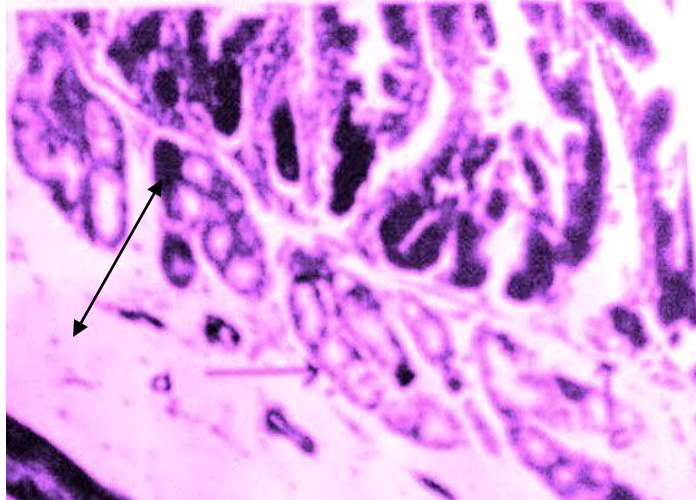


Fig.7 Rabbit-Six weeks(H &E stainx80) Photomicrograph of T.S, of second part of duodenum showing-

- a) mucous acini forming major part of glandular tissue in the submucosa
- b) occasional serous acini (double arrow)
- c) muscularis mucosae

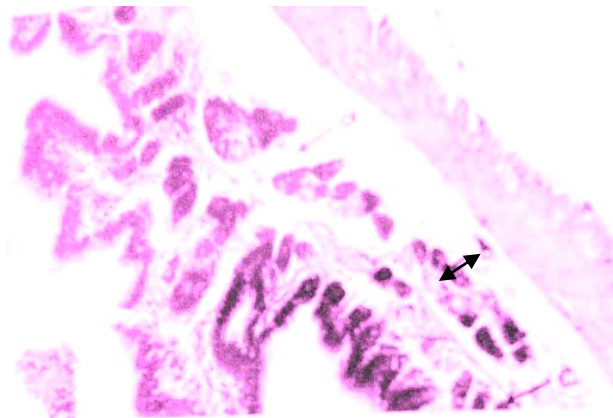


Fig. 8 Rabbit-Six weeks(H &E stain x 80) Photomicrograph of T.S, of fourth part of duodenum showing

- a) submucosa showing small patches of serous acini
- b) less number of mucous acini (double arrow)
- c) muscularis mucosae

Conclusion:-

Postnatal development of Brunner's glands in the rabbit were studied Total twenty one rabbits were sacrificed. The Brunner's gland in the rabbit at birth was represented by two cell layered tubular growth from the deeper parts of intestinal gland, branching into the submucosa of first and second of duodenum By the end of second week the glandular tissue extended to the fourth part of duodenum. Compactness of glands when traced at weekly intervals in the identical parts of duodenum showed marked increase upto sixth week The serous cells were differentiated in the terminal portions of tubules at birth but mucous acini started differentiated only after two weeks .At sixth week predomination of mucous acini in the first and second part of duodenum and of serous acini in the fourth part was seen while in the third part mucous and serous acini were almost in equal proportion This could be correlated to the greater need of alkanity in the vicinity of pyloric region which is the first to receive the acidic content of the stomach.

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RESEARCH ARTICLE

CORPORATE SOCIAL RESPONSIBILITY: A STRATEGIC DECISION AND GOVERNANCE

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Abstract

Corporate sector being one of the many investors in a corporate structure, it has been surveyed that all the companies possess Policies and practices of Corporate Social Responsibility (CSR). In the process of design and implementation of CSR by taking initiatives in the area of work, covers the entire community. Moreover, it has been unearthed that many companies carry out the promotion and implementation through Human Resource Department, in collaboration with NGOs but a full fledged CSR department. Nevertheless, CSR enterprises undertaken by the Indian companies for rural development created a positive impact on the inclusive development of business and society at large.

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The vast majority of the poor in India has been gripped in the state of neglect and underdevelopment. The problems of illiteracy, mortality rate, ignorance, poor health and hunger are more serious issues in rural areas than in urban. The reason being merely, lack of equipment resources and defective planning investment process and model. India does have a potential to meet these tests in rural areas provided government efforts are sufficient to provide basic services to its citizens. It is alarmingly recognized that the progress and welfare of society is not just the responsibility of government rather other stakeholders also must give hand to achieve the goal of development.

The concept of CSR took birth basically in western world out of the public retort for the new stages of power and freedom that was acquired by transnational sectors due to continuous neo-liberal economic policies which were in practice in 1980s and 1990s initiated by the governments, then. Subsequently, the citizens of these nations felt discomfort with the mounting power of corporations when the media started exposing a few negative environmental and social impacts that the large chunk of Western owned corporations had in developing countries. Consequently, some academics, government agencies and development organizations thrust the responsibility on the part of corporations with a solution to lessen few of the negative impacts which would cater to contribute more to socio-economic development in its largest sense.

Thus, the corporate sector does have an important role to play in ensuring that the private investment must reach these rural areas which have been neglected and remained out of development endeavour till now and looking forward to sustained development in rural areas, too. Surprisingly, many companies resulted in supporting the concept of CSR with the title of business of profit with due social responsibility. Very soon later the companies voluntarily decided to join hands with society to make it a cleaner and better environment (European Commission, 2001). Therefore, making CSR as "The commitment of businesses to contribute to sustainable development as well as to economic development by working with employers, their families, local community and society as a whole and

improve their quality of life, so that it is good for business and for development. A widely-cited definition by the World Business Council for Sustainable Development the state of development that "Corporate Social Responsibility is the continuing commitment by business to behave ethically and contribute to economic development while improving the quality of life of workers and their families and the local community and society at large." (WBCSD, 1999)

Therefore, to define the CSR two folded, it could be stated that firstly, it offers ethics to the organization to practice for its inner and outer employees. Furthermore, it also makes the organization realize its responsibility towards the environment and the society in which it functions. Companies consider CSR as a means of giving the society its due returns. Undoubtedly, it is quite important to study and understand businesses and how they initiate CSR by creating their impact of activities in the domain of socio-economic development of the rural population. It is a mission to contribute innovative solutions to the environmental and social challenges. In order to identify the priorities and the CSR inventions in regard to rural development sector by the professional community, certain parameters must be drawn:

1. The objectives of CSR must be aligned with the Millennium Development Goals (MDGs).
2. The companies must create divisions assigned for CSR by comprising them with well qualified, well trained and well experienced professionals coming from social science background, Rural Development and Development Studies for better planning, application and evaluation.
3. CSR programmes which are to be put to action by the Indian companies must be founded on the felt needs of existing population through which their participation can be enhanced. Moreover, it should also be implied that the jobs are created by way of economic activities in the rural community to better their lives.
4. The performance of CSR of a company must come under social audits by external source agencies.
5. It would be unfair on the part of government to act just like a director of CSR. Rather, the government must take charge of being a facilitator, promotor and a catalyst to encourage the big players of the business world asking them to take into account the ethical, environmental and social issues of the country.
6. In order to highlight the company commitment to sustained development of the community, an annual report must be prepared on social responsibility.

It has been observed that companies have designed the policies of practicing CSR. A wide range of CSR initiatives which are designed and implemented ranges from agriculture development, drinking water management, development of natural resources, income generation schemes, health check-up and camps, health services on wheels, general and adult education and infrastructure services established by such companies. Although the manner of working is changing from philanthropic development to sustainable welfare, there doesn't seem to be any relation between company CSR plan and the Millennium Development Goals. It has also come to light that the companies carry out CSR initiatives through Human Resource Department, in collaboration with NGOs. There is an absence of full-fledged Corporate Social Responsibility department.

Thus, to conclude, CSR is surely considered as a vital issue of Indian companies no matter what its size, sector and objective or location is. Moreover, the Indian companies have realized that stability and sustainability of economic activities to compete with global market is not possible without socio-economic development of local communities. It has definitely resulted in the positive effect and impact on the overall development of society and business.

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RESEARCH ARTICLE

UTILIZATION OF FLOURS OF FOX NUTS AND WATER CHESTNUTS FOR PREPARATION OF PUA.

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Key words:-

Water chestnuts, Fox nuts, Product development, Nine point hedonic scale.

Abstract

Background: The idea of incorporating of flour being nutritionally rich fulfills the requirement of healthy snacks. As protein and vitamin deficiencies are very common in India, efforts are underway to enrich cereals, in supplements with, vegetable used in effective manner.

Aim: The objective of the study is to evaluate organoleptic attributes, nutritive value and cost of prepared food product.

Methods: The products were organoleptically evaluated for the colour and appearance, body and texture, taste and flavour and over all acceptability using Nine point Hedonic scale. Nutritional composition was determined using the standardized AOAC (2005) methods.

Statistical analysis: The data were statistically analyzed by using analysis of variance technique, critical difference test.

Results: On the basis of findings, it was observed that in context with organoleptic attributes, treatment T₂ was best as compared to control and other treatments, however all the treatments were found acceptable. Nutrient composition of *Pua* treatment T₂ was rich in Protein (7g/100g), Carbohydrate (66g/100g) and Iron content (2mg/100g). Average cost of the *Pua* per 100g of raw ingredients was Rs. 13.25 for T₀ (control), Rs.17.25 for T₁, Rs. 19.25 for T₂ and Rs. 21.25 for T₃.

Conclusion: Incorporation of flours of Fox nuts and Water Chestnuts in *Pua* is well acceptable and can be used for the development of food product.

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Introduction:-

The idea of incorporating of flour being nutritionally rich fulfills the requirement of healthy snacks. As protein and vitamin deficiencies are very common in India, efforts are underway to enrich cereals, in supplements with, vegetable used in effective manner (Pragati and Paul, 2010).

Fox nut or Gorgon nut, commonly known as *Makhana*, Popped *Makhana* is used in preparation of a number of delicious and rich sweet dishes, pudding and milk based sweets. Fox nuts are low in calories, fat (saturated fats) and sodium and high in magnesium, potassium, phosphorus and fiber. They have antioxidants properties and good food for diabetics, arthritis, kidney diseases, constipation, diarrhea, cancer, anemia, heart problems. It improves appetite, moisture level in body tissues and prevents the inflammations in the body. It helps the body to remove the waste and thus prevent the accumulation of toxins. It also contains useful medicinal properties (Das *et al.*, 2006).

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Water chestnut (*Trapa Bisinosa Roxburg*) commonly known as “Singhara”, is an annual aquatic warm season crop. They are almost fatless and are therefore, a healthy food option. Ground water chestnut powder mixed with water can relieve cough. Boiling water chestnuts in water makes the best drink for measles patients and is appropriate for all measles patients from the third day till the ninth day of the cycle. It helps to speed up the measles cycle. They are good sources of calorie, carbohydrate, dietary fiber, vitamin B6 and also contain fair amount of calcium, potassium, iron and zinc. Water chestnut used for many therapeutic purposes e.g. for jaundice, measles, cough and summer heat etc (*Masrizal et al., 1997*).

The study was carried out to develop pua by incorporation of flour of fox nuts and water chestnuts, to evaluate the organoleptic acceptability of the prepared product and to determine the nutrient composition of the prepared product.

Methodology:-

The present study was conducted in Sam Higginbottom University of Agriculture Technology and Sciences, Allahabad.

Procurement of Raw Materials:-

The required materials i.e. Fox nuts and water chestnuts, Bengal gram flour, sugar, cardamom, coconut powder, coco powder, refined oil and vegetable etc were collected from local market of Allahabad city.

Preparation of fox nuts and water chestnuts flour:-

Fox nuts and water chestnut was Cleaned, after that Sun drying was done for one day, then fox nuts and water chestnuts was Grinded, then Fox nuts and water chestnuts flour was Packed (Airtight tin containers or polythene bags) and Storage (At ambient temperature in dry place) **Srivastava and Kumar (2009)**.

Control and three experimental treatments were prepared as follows:-

T₀ (Control): In this, the standardized recipe was followed to prepare the products without any incorporation fox nuts flour.

T₁ (90%, 10%): In this treatment, 90 percent Water chestnut flour and 10 percent Fox nut flour to prepare products.

T₂ (85%, 15%): In this treatment, 85 percent Water chestnut flour and 15 percent Fox nut flour to prepare products.

T₃ (80%, 20%): In this treatment, 80% Water chestnut flour and 20 percent Fox nut flour to prepare products.

Replications: All the treatments of the four products respectively were replicated four times to get average values.

Organoleptic Evaluation:-

Sensory evaluation of the Pua for their acceptability was done by a panel of judges. Colour, Texture, Flavor, Taste and Overall acceptability were evaluated using the nine point hedonic scale based score card. (**Srilakshmi, 2003**).

Calculation of Nutritive Value of Prepared Products:-

The nutrient composition of the value added product were determined by **AOAC method (2005)** to estimate the moisture, total ash, protein, fat, carbohydrate, iron and calcium content.

Cost of prepared products:-

The cost of the products was calculated on the basis of price of raw ingredients at rupees/kg.

Nutrient/100gm of product:-
$$\frac{\text{Ingredient used (g.)} \times \text{nutritive value of ingredient}}{100}$$

Statistical analysis of the Products:-

Data is ascertained from the experiment was statistical analyzed using analysis of variance technique and critical difference test (**Fisher, 1995**).

Results and Discussion:-

Average Sensory Attributes Of Value Added Pua.

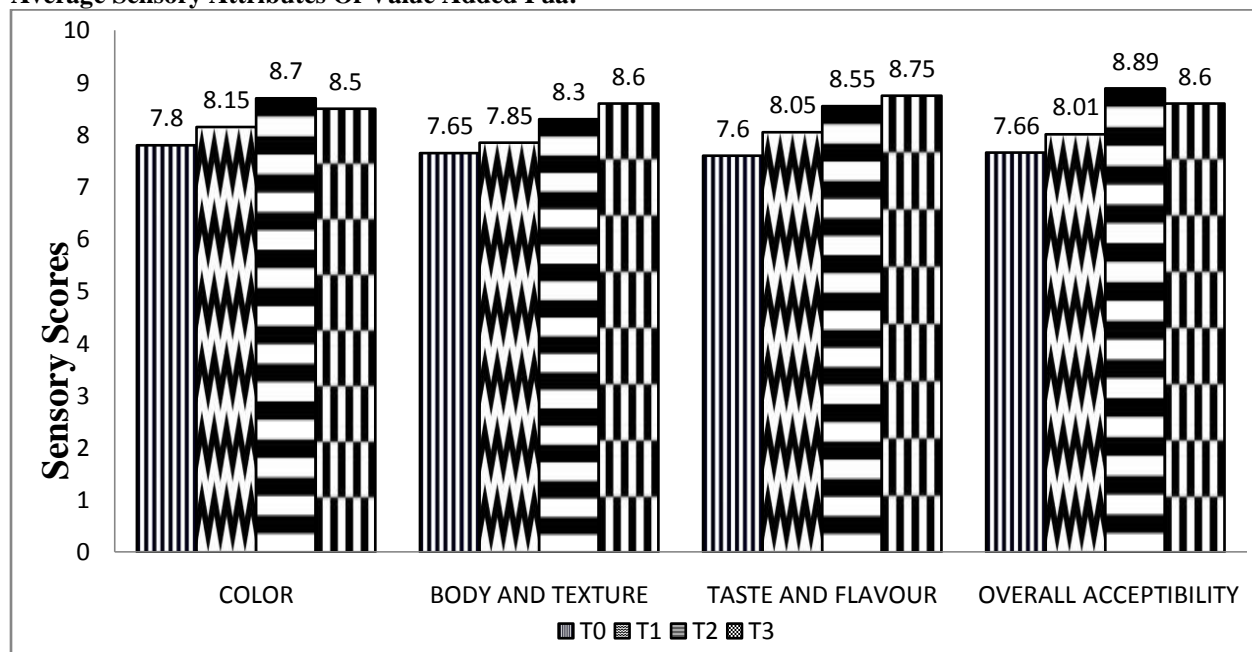


Figure 1:- Average Sensory Score for Different Treatments of Pua

Figure 1 shows the mean scores of *Pua* in relation to overall acceptability indicates that the treatment T_2 (8.89) has the highest score followed by T_3 (8.60), T_1 (8.01) and T_0 (7.66) respectively, making quite obvious that the incorporation of 85 percent water chestnut flour and 15 percent fox nut flour showed the best result for overall acceptability. Therefore, it can be calculated that there was significant difference between the treatments for overall acceptability of *Pua*. All treatments were acceptable on 9 point Hedonic scale and liked very much by the panel of judges.

Kumari and Paul (2012) studied the incorporation of coarse grain bajra flour and chick pea flour into the traditional recipe *Pua*. The result shown that the treatments T_3 were most acceptable among all the treatment they obtained a mean score of 6.92, 7.6, 7.68 and 7.42 for colour, body and texture, taste and flavour and overall acceptability.

Nutrient Composition Of The Product:-

Table 1:- Mean nutrient composition of the *Pua* developed by incorporation of flours of water chestnuts and fox nuts (per 100 g).

Product/ Treatments	Moisture (g)	Ash (g)	Fat (g)	Protein (g)	CHO(g)	Calcium (mg)	Iron (mg)
Pua T_0	12.64±1.12	22.07±0.43	15.85±0.92	4.12±0.08	60.23±0.34	30.28±3.24	1.16±0.03
T_2	8.20±0.28	22.36±0.39	14.37±0.51	7±0.19	66.10±0.87	28.11±2.19	1.64±.43

A wide range of variation was observed in the nutrient content of the product and treatment. T_2 was found to be the best treatment among all the treatments. Therefore along with T_0 , nutrient composition of T_2 was determined. Results from the table 2 shows that the moisture content was highest in pua T_0 (13g/100g), Ash content was highest in T_2 (23g/100g), Fat content was T_0 (16g/100g), protein content was T_2 (7g/100g), in carbohydrate content was T_2 (66/100g), calcium content was T_0 (30mg/100mg) and iron content was highest in T_2 (1.64)

The result is supported by the findings of **Singh et al., (2009)**. The *Pua* is prepared with the incorporation of bajra flour, maize flour in whole wheat flour. Protein content increased as the incorporation level of bajra and maize flour increased. Fat content was lowest in T_0 (51.07 g), followed by T_1, T_2, T_3 . Calcium content was highest in T_2 (112.45 mg), followed by T_3, T_2, T_1 and T_0 and carbohydrate content as the amount of bajra and maize flour increased. Iron

content was found highest in T₀ (12 mg), followed by T₁, T₂, T₃. Moisture content was high in T₃ (13.45), followed by T₂, T₁ and T₀.

Cost Of Raw Ingredients:-

The cost has been calculated on the basis of prevailing price of raw materials. It is evident that the cost of *Pua* T₁ was lowest i.e. (17.25 Rs/100g) as compared to experimental *pua* samples, T₂ (19.25 Rs/100g) and T₃ (21.25 Rs/100g). The cost of the developed product is increasing due to incorporation of water chestnut and fox nut flour at different levels.

Conclusion:-

It is concluded that incorporation of flour of water chestnuts and fox nuts in product like *Pua* is well acceptable. In *pua* T₂ (85percent water chestnuts flour + 15percent fox nuts flour) was the best on all sensory parameter. For *Pua* treatment T₂ (85percent water chestnut and 15percent fox nut flour) was rich in protein (7g/100g) and carbohydrate content (66g/100g). The lowest cost of the developed food product per 100g of raw ingredients was Rs-13.25 for *Pua* of the treatment T₁.

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RESEARCH ARTICLE

USE OF CACTUS OPUNTIA AS A NATURAL COAGULANT: WATER TREATMENT IN DEVELOPING COUNTRIES.

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Abstract

Observing the need for clean water and recycling of wastewater through effective treatment to help counter the continued water scarcity facing Kenya, the study sought to explore the possibility of using cactus in the coagulation-flocculation process. The study employed an analytic literature review model, sourcing recent and authoritative studies to investigate the effectiveness of cactus plants as coagulants, and the involved pH implications. The findings indicate that plant-based coagulants such as cactus are being considered as alternatives to conventional synthetic chemical coagulants in aspects such as costs, health effects, non-biodegradability, altered pH in post-treatment water and corrosion and transmission problems. In addition, cactus appears to have no significant effects on the pH of treated water. However, the findings demonstrate a lack of consensus regarding the optimal pH at which cactus is best effective in coagulation.

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Acronyms:-

NTU Nephelometric Turbidity Meter
UPC Platinum- Cobalt Unit
STD Total Dissolved Solids
COD Chemical Oxygen demand
PAC Polyaluminium chloride

Introduction:-

Today's rapidly growing populations outstrip available water resources, which is especially problematic in developing countries that may be on the verge of water scarcity. In the context of limited water resources and water scarcity, water treatment and recycling procedures constitute one of the alternatives to obtain potable water in future. It is therefore necessary to develop suitable, rapid, and inexpensive water and wastewater treatment methods to remove turbidity, coloration, and heavy metals among other contaminants (Gupta et al. 2012). Such an approach to water treatment should also take into consideration contributions to environment and sustainable development. Wastewater treatment primarily entails the coagulation-flocculation process, which is a widely employed

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physicochemical treatment approach. Traditionally, synthetic inorganic chemicals such as alum and ferric chloride have found wide use as coagulants and flocculants (Nilsen *et al.*, 2005). However, the use of such conventional synthetic chemical coagulants raises environmental, safety, and sustainability concerns, surrounding aspects such as high procurement costs, possible human health risk concerns, their effects on the final pH of the treated water, and non-biodegradability which have detrimental environmental concerns (Yin 2010). These issues have led to the pursuit of alternative coagulants in wastewater treatment, with plant-based coagulants emerging as potential replacements. Such natural coagulants bear much promise in delivering effective wastewater treatment while responding to environmental and sustainability considerations.

In the Kenyan context, wastewater treatment is pertinent because the country has a water scarcity problem, with over 80% of its land considered arid and semi-arid (Patel, 2013). The country has a freshwater per capita of only 647m³, which is well below the United Nations' recommended minimum (1,000m³), with projections indicating a possible decline to 235m³ by 2025. As a case example, water supply to Nairobi stands at 580,000 m³/day compared to a demand of 750,000 m³/day, underscoring the necessity of sustained huge investments to expand water supply. In a scenario in which 20 % of water supplied to the city ends up as wastewater, the wastewater volume may increase from 116,000 m³/day to 172,000 m³/day by the year 2017, and 600,000 m³/day by 2035, an amount which is equivalent to the total water supplied in 2013 (Patel, 2013). These observations demonstrate that wastewater treatment constitutes an important potential source of water to relieve the water scarcity in Kenya. However, urban municipalities currently rely heavily on conventional treatment plants, which not only have the earlier mentioned demerits, but are also inadequate, non-functional at times due to technical hitches, or entirely lacking owing to associated operational costs and capital investments (Opaa&Omondi, 2012). Such observations indicate the need for alternative coagulants in Kenya to address the weaknesses of conventional wastewater treatment to meet the water supply needs of the country.

In terms of environment and sustainability considerations, Kenya seeks to make progress on the 17 Sustainable Development Goals contained in the United Nations' 2030 Agenda (United Nations Development Programme, 2017). Pursuing alternative, natural, and effective coagulants contributes directly to one of these sustainable development goals and indirectly to a number of the other goals. In this case, goal 7 of the 2030 Agenda is to "ensure availability and sustainable management of water and sanitation for all" (Kimani, 2016). As a result, securing a locally available and inexpensive plant-based coagulant would help Kenya take significant steps towards the 2030 Agenda in the aspect of sustainable management of water and sanitation. Other goals to which alternative natural coagulants are relevant include goal 3, concerning healthy lives and wellbeing of all, goal 1 (poverty), goal 3 (improved nutrition and sustainable agriculture, and goal 10 (addressing inequality).

The possibility of using plant-based coagulants in treating wastewater would prove useful for the country in terms of such environmental, health, and sustainable development objectives, as these coagulants are generally considered to be safe to human health, inexpensive and locally available therefore affordable, biodegradable and result in low-level sludge production (Vijayaraghavan *et al.*, 2011). One such plant-based coagulant under research entails the use of cactus species, with multiple studies seeking to establish the plant's effectiveness as a coagulant. Cactus is native to Kenya making it readily available, which indicates that it bears significant promise as a natural coagulant to address the problem faced in potable water supply and sustainable wastewater treatment in the country. However, its use as a coagulant first requires an overview of its effectiveness levels and optimum conditions especially in relation to pH, which would then guide further research and use in local contexts in Kenya.

The objective of this paper is to review available scholarly work on the possible use of the cactus as an alternative coagulant through consolidating the available published literature concerning cactus' effectiveness in water treatment. The review is expected to avail information that can be useful to develop infrastructure for affordable and sustainable potable water provision in rural areas and informal sector communities of Kenya. The review will also offer technical know-how to informal settlement dwellers on the use of cactus to treat their wastewater for re-use.

Literature Review:-

The review of literature involved an analysis of published studies regarding the use of cactus-based coagulants published within the last 10 years to ensure that the insights are current and up-to-date. The review commences with studies that took some interest in the relevance of natural, plant-based coagulants such as cactus in light of the merits of such bio-coagulants and demerits of conventional chemical treatment. The next section covers the use of cactus derivatives as coagulants in water treatment, exploring their effectiveness when compared to other bio-coagulants

and conventional ones. The review then narrows to focus on studies that included investigations on the effects of pH on the effectiveness of treatment including the pH of the final treated water when using cactus-derived coagulants. The literature review concludes with an overview analysis of the findings from the scholarly.

Opportunity for Use of Cactus as a Natural Coagulant:-

According to Yin (2010), lack of proper and effective water treatment systems in rural areas of underdeveloped regions of the world necessitates the use of cost-effective point-of-use approaches. The study demonstrates how the opportunity for use of natural plant-based coagulants arises. Among the various point-of-use approaches, coagulation has found wide use in treating both surface water and industrial wastewater. Such wide application in the removal of dissolved chemicals and turbidity from water entails the use of conventional chemical-based coagulants, including alum ($\text{Al}_2(\text{SO}_4)_3$), poly-aluminium chloride (PAC) and ferric chloride (FeCl_3). However, Yin (2010) notes that these conventional coagulants are associated with demerits such as ineffectiveness in low-temperature water, high procurement costs, deleterious impacts on human health, production of large sludge volumes and significant effect on the pH of the treated water. Meanwhile, the study establishes that natural plant-based alternatives avert these shortcomings of conventional coagulants through their low cost as they are locally available, the unlikely effect on extreme pH changes and high biodegradability. This study demonstrates that cactus may be advantageous because it is not only effective, but also helps avoid the environmental impacts of synthetic coagulants, besides also providing a sustainable option towards wastewater treatment.

Feria-Díaz *et al.* (2016) make similar observations regarding the use of conventional chemical coagulants in treating surface water and wastewater. The scholars noted that such coagulants are based on inorganic salts or synthetic organic polymers, which do have various disadvantages including risk to human health, high cost, high sludge volume production, water temperature dependent and significant alteration of the treated water. Further, chemical coagulants lead to high aluminum concentration in water after treatment, which interferes with water disinfection, which poses some distribution problems, decreases transmission capacity, and generates corrosion problems. Meanwhile, the scholars noted that alternative coagulants from natural sources have advantages in their safety for human health, biodegradability, low acquisition costs, low toxicity, and low-level sludge production. These merits underscore the environmental and sustainability gains that would be made upon embracing natural coagulants in treating wastewater. The study concludes that natural coagulants are equally effective in treating water and are also unlikely to alter the pH of treated water.

Vijayaraghavan *et al.* (2011) reviewed the use of plant-based coagulants in the treatment of water and wastewater. Among the factors contributing to the interest in the use of plant-based coagulants, the scholars highlight the presence of residual monomers during the use of synthetic polymers, which are undesirable owing to their neurotoxicity and carcinogenic properties. This led to researchers getting interested in plant-based coagulants, which could solve the problem of residual monomers. The plant-based coagulants are designed as point-of-use technologies in less-developed societies, as they are relatively cost-effective when compared to their chemical counterparts. In addition, the scholars note that plant-based coagulants are also advantageous owing to the ease of processing to useable form alongside their biodegradability, translating to sustainable wastewater treatment. These bio-coagulants achieve comparable treatment efficiency to chemical coagulants when employed in treating waters that have a low-to-medium turbidity range of 50 –500 NTU. Although the scholars observed that application of the natural coagulants in industrial wastewater treatment is still in infancy, they noted that such coagulants hold much technical promise in water treatment. Cactus, Nirmali seeds (*Strychnos potatorum*), tannin, and *Moringa oleifera* are among the most studied plant-based coagulants, representing important progress in sustainable water treatment technologies.

Cactus Use in Coagulation:-

Several studies demonstrate that cactus is potentially an effective natural coagulant that is also responsive to environmental and sustainability concerns. Betatachee *et al.* (2014) compared north Algerian prickly pear cactus (*Opuntia ficus-indica*) juice with polyelectrolytes (Chimfloc C4346, Sedipur NF 102, a cationic polymer, and Sedipu AF 400) and inorganic conditioners (FeCl_3 and $\text{Al}_2(\text{SO}_4)_3$) in coagulating wastewater sludge. The study entailed a chemical characterization of the cactus juice alongside conditioning of the sludge with the juice and comparison chemicals, with analysis involving Fourier Transform Infrared spectral analysis and photometry. The findings demonstrated that the juice from the cactus promoted the coagulation of almost all of sludge, enabling liquid and solid separating and easing filtration process. Further, residual turbidity, the specific resistance of filtration and dryness obtained were comparable to those of Chimfloc C4346, and significantly better than those of Sedipur AF400

and the inorganic chemicals analyzed, with the optimum dosage for the cactus being 0.4 g kg⁻¹ of dry matter. These findings led these researchers to conclude that the juice from the prickly pear cactus could find use as a natural conditioner in treating sewage sludge.



Fig. 1:- Cactus *Opuntia* (*ficusindica*).

Rodiño-Arguello *et al.* (2015) compared five natural coagulants, *Hylocereus cf. trigonus* (cactus) stems, Campano (*Albizia saman*), exudate gum, the bark of Guácimo (*Guazuma ulmifolia*), and bark and seed from Moringa (*Moringa oleifera*) in treating raw water samples taken from Sinú river, Colombia. The scholars measured initial turbidity levels and turbidity removal efficiencies as a percent activity function for coagulant dosages at 5 mg/L to 200 mg/L. Although the Moringa extract was found to be most effective in turbidity removal efficiency and coagulation activity, the cactus also had attractive results in removing turbidity (98% turbidity removal). The aforementioned observations underscore the turbidity removal prospects of cactus extracts, demonstrating why it may find crucial use in wastewater treatment in the third world.

In another study, Fedala *et al.* (2015) evaluated the performance of *Opuntia ficus-indica* as a flocculating agent. Collecting the cactus from Tipaza, Algeria, the scholars evaluated the coagulation-flocculation performance of the cactus and the quality of the treated water through measuring residual turbidity. After measuring the effectiveness of the cactus in coagulation at different solutions, the findings demonstrated that 0.2 mg/L cactus concentration was effective for the most turbid water, achieving best removal to 0.5 NTU. The residual turbidity levels were between 0.5 and 1.2 NTU, with the results being dependent on the initial turbidity of the water. These findings led the scholars to conclude that mucilage from *O. ficus-indica* is attractive as a natural material for water treatment technologies.

Nougbodé *et al.* (2013) also evaluated the properties of cactus as a natural coagulant for water clarification, using an aqueous *Opuntia dillenii* solution to clarify highly turbid surface water (turbidity varying from 418 NTU to 186 NTU). After undertaking flocculation and coagulation tests, the scholars found that *Opuntia dillenii* was useful for

the treatment of highly turbid water. The solution had a removal efficiency varying from 89% to 93% for turbidity and suspended solids, as well as efficiency varying from 4% to 15% for obvious color using optimum values 1 mL to 10 mL. Successive addition of the natural coagulant and lime resulted in an improved elimination of turbidity and suspended solids in the water alongside the improvement of color. Removal efficiency for turbidity and suspended solids reached over 95% while that for color was between 67% and 94%. Such observations underscored the value of *Opuntia dillenii* in water clarification, with the scholars recommending that studies move beyond laboratory-scale settings.

Sellamiet *et al.* (2014) compared the use of cactus juice as a bio-flocculant with conventional polyacrylamide in industrial wastewater treatment. The scholars conducted experiments in which they substituted polyacrylamide with cactus juice, noting the need to reduce the use of chemical products in coagulation. From the data collected, the scholars found that substituting polyacrylamide with cactus juice in coagulation–flocculation process was highly effective. The bio-flocculant had removal efficiencies of 59.1%–69.1% in removing chemical oxygen demand and 83.3%–88.7% in removing suspended solids. Upon addition of lime to the process, the removal efficiencies with cactus juice for both suspended solids and chemical oxygen demand improved to over 90%. Based on these findings, they concluded that using cactus juice as flocculants meets sustainability requirements and is highly appropriate for regions in which cactuses grow naturally.

Pichleret *et al.* (2012) studied the natural coagulants also through a comparison model, using mucilage from *Opuntia ficusindica* obtained from Tucson, Arizona against aluminum sulfate ($\text{Al}_2(\text{SO}_4)_3$), the commonly used synthetic flocculant. After undertaking jar and cylinder experiments, the results demonstrated a high efficiency of the cactus mucilage in eliminating turbidity. At dosage concentrations of 3 mg/L, the extract heightened particulate settling by 330% in comparison to aluminum sulfate. Meanwhile, its effectiveness was similar to that of the synthetic flocculants at 0.3% dosage of the necessary aluminum sulfate concentration. These findings led the scholars to conclude that cactus bears the potential of a ‘green’ coagulant that has the advantages of being environmentally friendly and inexpensive.

Effectiveness and pH:-

Yang *et al.* (2007) investigated the effectiveness of cactus *Opuntia* as a natural macromolecular coagulant in removing turbidity. The study involved a jar test technique, with the coagulant’s effectiveness being reviewed on surface water (river and estuarine, 499 NTU and 547 NTU respectively) and landfill leachate. The scholars found that the coagulant reduced turbidity by as much as 98% in estuarine surface water and 70% in river water, but was less effective for leachate water (31.6 – 41.9%). In terms of impacts on water pH, the results indicated that increasing cactus dosage from 13 to 853 mg/L resulted in the marginal effect on the water’s final pH (ranging from 7.25 to 7.69 for estuarine water and 7.83 to 8.49 for river water). These observations implied that substantial increases in dosage did not affect the final pH of the water compared to the effect of chemical-based coagulants.

Noting that natural coagulants provide benefits in local availability, affordability, and lesser health hazards, Mukhtaet *et al.* (2015) evaluated the efficiency of turbidity removal of *Opuntia stricta* from Lahore, Punjab-Pakistan, as well as the effect of the water’s pH on such efficiency. They assessed the effectiveness of this cactus species through measuring turbidity removal at various pH levels and coagulant doses. The findings demonstrated that *Opuntia stricta*, even when used at low doses, achieved residual turbidity levels of below 20 NTU. Unlike other coagulants that are highly pH dependent, the turbidity removal efficiency of cactus remained consistent across a wide range of pH levels (5 to 10). In addition, the scholars found that the pH of the water remained constant during the coagulation process, which means that pH adjustment may be unnecessary in treatment processes. Such a merit is crucial because post-treatment pH adjustments are often necessary when using metal coagulants.

Lozano *et al.* (2015) explored the efficiency of mucilage extracted from *Opuntia ficus-indica* (nopal) as a natural coagulant complementing aluminum sulfate in clarification of water obtained from river Magdalena, Colombia. The scholars employed the jar test method, considering factors such as proportion and concentration of the coagulants, pH, and agitation speed in evaluating turbidity (NTU), color (UPC), and total dissolved solids. The study established that a maximum of 20 % nopal mucilage coagulant proportion alongside agitation speeds of 200 rpm reduced turbidity values to below 2 NTU, an efficiency corresponding to over 50% (96-95% highest). In addition, the combination reduced color to zero UPC and STD content to values under 200 mg/L without affecting the pH of the treated water significantly. These findings demonstrate that cactus mucilage can be used in combination with conventional coagulants to increase the efficiency of water treatment.

Noting that natural coagulants promise advantages such as abundance, inexpensiveness, multifunction, and biodegradation, Shilpa *et al.* (2012) evaluated *Opuntia ficusindica* and hyacinth bean peels from Mysore, southern India in water treatment. After treating surface water with the two natural coagulants, they studied parameters such as turbidity, pH, alkalinity, and bacterial contamination. The findings demonstrated that the turbidity removal effectiveness for the cactus was 89.03% at an optimum dosage of 20 mg/L while the percentage reduction in bacterial count was 20.15%. A pH of 9 was also discovered to be the optimum level for effective water treatment when using this cactus species. These findings underscore the promise of cactus as a natural coagulant and alternative to synthetic chemical coagulants such as aluminum and iron salts.

Nharingo and Moyo (2016) explored the use of cactus-derived biomaterials in decontamination of wastewater, observing that the past decade had witnessed much interest into the biological material in environmental remediation. The two scholars undertook an analysis of literature covering aspects such as characterization, physico-chemical compositions, application in bio-sorption and flocculation of pesticides, dyes, and metallic species. The studies reviewed provided findings based on different parts of the plant, including cladodes, fruit pulp, mucilage, and electrolytes. Their findings demonstrated that all these parts of the cactus had very high pollutant maximum sorption capacities, as well as removal ranging between 125.4 1000 mg/g and 0.31 2251.56 mg/g for bio-sorption of dyes and metallic species. Cactus also had removal percentage ranges of 98.7% for turbidity, 93.62% of chemical oxygen and 100% for heavy metals. In addition, cactus biomaterials proved to have wide pH ranges (3.27 for fresh cladodes, 6.03 for mucilage powder, and 5.3-7.1 for the fruit pulp) in which they work efficiently. Ultimately, the cactus biomaterials proved effective in removing pollutants, leading the two scholars to recommend scaling up from laboratory settings to community pilot plants and at industrial levels.

Mouniret *et al.* (2014) explored the flocculating activity of pectin and mucilage extracted from *Opuntia ficusindica* obtained from Pettit, Morocco. The tests were done on sanmix clay suspension, which served as synthetic turbid water. The findings demonstrated that the bio-flocculant had 98% flocculating activity in the sanmix clay suspension. In addition, the scholars found that the optimum dosage for mucilage was 2-8 mL while that of pectin was 2-6 mL by one-liter turbid water. In terms of pH, the study established that flocculation activity was poor in strong alkali conditions than in acidic conditions, as demonstrated by less than 60% flocculation in pH of 10 and above. Flocculation activity was however higher in acidic conditions, peaking at 98.8% for pectin and 97.2% for mucilage at pH 3.0. As a result, the scholars noted that pH played a significant role in influencing the effectiveness of cactus-derived bio-flocculants.

Miller *et al.* (2008) quantitatively evaluated the coagulant properties of *Opuntia spp.*, exploring its use as a coagulant for water treatment. The scholars undertook tests to evaluate the plant for turbidity removal in synthetic water samples (turbidity caused by kaolin clay particles), besides exploring some possible underlying coagulation mechanism for the plant. The findings indicated that the cactus species reduced turbidity in the water by 98% for a range of various turbidity levels, with such turbidity removal occurring at a pH of 10. The scholars observed that such turbidity removal levels were similar to those established for *Moringa oleifera*, but noted that the cactus' turbidity removal occurred through a bridging coagulation mechanism. The scholars concluded that *Opuntia spp.* could find valuable use as part of point-of-use water treatment technology, offering a practical and affordable strategy to producing potable water in developing communities.

Noting the promise of *Opuntia ficusindica* as a cheap, locally abundant, and eco-friendly bio-flocculant, Nharingo *et al.* (2015) explored the effectiveness of cactus powder in bio-coagulation-flocculation of heavy metal ions from wastewater drawn from Mukuvisi River, Harare, Zimbabwe. At natural ionic strength and dosage of 8 mg/L alongside a floc settling time of 180 min, the cactus powder achieved heavy metal removal of 84.16% (Cd), 85.74% (Zn), 93.02% (Cu) and 100% (Pb). The scholars also noted that the mechanism of the coagulation-flocculation process could have involved adsorption-bridging and adsorption-charge neutralization mechanisms. In terms of pH, the scholars found that the process of coagulation-flocculation was optimum at pH 5, with the percentage removal increasing gradually as pH increases up to pH 5 and then decreasing beyond a pH of 8. Based on these findings, the scholars concluded that the use of cactus powder in optimum conditions to remove heavy metal ions such as lead was highly promising and could be tested at commercial and industrial scales.

Another study by Bouatay and Mhenni (2014) involved effectiveness of cactus as an eco-friendly flocculant in the treatment of textile wastewater. Noting that *Opuntia ficusindica* mucilage bore promise owing to its abundance and inexpensiveness, the scholars reviewed its performance as a flocculant across parameters such as decolorization,

turbidity abatement and COD removal. The findings demonstrated that a combination of cactus mucilage with aluminum sulfate was highly effective, with the best conditions for the flocculation process occurring at pH 7.25, flocculant dosage of 40 mg/L, mixing speed of 30 rpm and a mixing time of 11 minutes. Based on these observed optimum conditions, the cactus mucilage yielded a decolorization of 99.84%, turbidity abatement of 91.66% and COD removal of 88.76%. Upon comparing flocculation performance of the cactus bio-flocculants agent and the commercial flocculants, the scholars found that the former exhibited the highest levels of pollution removal from wastewater.

Taa *et al.* (2016) explored the use of a cactus bio-flocculants extracted from *Opuntia ficusindica* in treating wastewater, basing their study on the need to replace chemical products and eliminate their negative effects on water disposal processes. The study entailed experiments using cactus juice on two types of wastewater, namely, synthetic chromium sulfate solution (100 ppm or 500 ppm) and real effluent obtained from a tannery (90 ppm or 900 ppm). From the findings, the scholars established that the cactus extracts facilitated rapid flocculation at a pH range of 7-9. In addition, the findings indicated that metal charge values were greatly reduced when cactus juice flocculation followed neutralization and lime coagulation. In this case, an initial charge of 176 mg/l reduced by 99.9% after 500 ml of 90 ppm effluent was treated with 4ml cactus juice. In the same fashion, cactus juice treatment led to significant drop in chemical oxygen demand (94%) and biological oxygen demand (95%). In terms of the tannery effluent, at a concentration of 900 ppm, the cactus juice achieved a 98.6% turbidity reduction. Ultimately, the flocculation results from the two solutions were satisfying when compared to results from DKFLOC, the chemical flocculants.

Overview of Literature Reviewed:-

The literature reviewed demonstrates that opportunities for use of bio-coagulants are high because these coagulants solve some of the drawbacks of synthetic chemical-based coagulants. Here, bio-coagulants such as cactus have shown promise in averting the health effects caused by treatment of water and wastewater with chemical products. Besides being highly effective, they are inexpensive, environmentally benign and abundantly available. Such merits make bio-coagulants especially appealing in the context of the developing world regions where wastewater treatment is a significant challenge. Such effectiveness is evidenced in aspects such as reduction of turbidity, coloration, chemical oxygen demand and heavy metals among other parameters. However, the studies reviewed reveal diverse findings regarding aspects such as effectiveness levels and optimum conditions. For example, the studies provide varied results in aspects such as optimum dosage and concentration, mixing times and speeds and optimum pH. Some studies demonstrate effectiveness at specific pH level while others demonstrate effectiveness over a wide range of pH levels. Most of the studies indicate that treatment with cactus-based bio-coagulants may not affect the final pH of the water. This diversity of insights from the literature and the lack of local studies in the context of Kenya underscore the need for localized research into the effectiveness of cactus-based bio-coagulants in treating wastewater in aspects such as turbidity, pH, and COD before making recommendations.

Methodology:-

This study employed an extensive literature review to collect data on the need, use, and effectiveness of cactus as a coagulant. The studies included in the review were restricted to those published after 2005 to ensure up-to-date findings. Publications from peer-reviewed journals only were also targeted to ensure academic rigor. Identification and selection of relevant articles for use in the study entailed a search on Google Scholar with the search terms informed by the main areas of focus of the present study. The slashes indicate the use of related phrases or keywords to expand the literature search.

- i. Use of natural/plant-based materials as coagulants in water and wastewater treatment
- ii. The effectiveness of cactus/*Opuntia ficusindica*/*Opuntia spp.* as a coagulant in water/wastewater treatment
- iii. Impacts on pH when using cactus/*Opuntia ficusindica* as a coagulant in water/wastewater treatment

Upon restricting the date, the initial search yielded 42 items published on the topic since 2005. Upon undertaking further screening to ensure the literature was restricted to those studies addressing strongly and specifically the topics of need, use, and effectiveness of cactus as a coagulant, the articles were reduced to 19. More than 19 articles are however cited in the present study owing to the use of other articles in the background section to help contextualize the research and provide a justification. Efforts were made to ensure rigor in these additional articles, restricting them to studies obtained from academic journals and important workshops or conferences on the wider and Kenyan context of wastewater treatment in particular.

Results and Discussion:-

After an analysis of the literature, a number of emerging themes regarding the three main areas of focus that informed the literature search were established. Table 1 provides a summary of the main findings.

Table 1:- Summary of Key Findings on Use of Cactus as a Coagulant.

Opportunities for use of natural/plant-based coagulants in water/wastewater treatment	Cited References
Lack of proper, effective, and sustainable water treatment systems in rural or underdeveloped areas	Yin (2010)
Demerits of chemical coagulants: - Ineffectiveness in low-temperature water, prohibitive procurement costs - Significant alteration of water pH, decreased transmission capacity, and corrosion problems - Environmental and sustainability issues: Negative effects on human health, large sludge volumes, costs, non-biodegradability	Yin (2010); Feria-Díaz <i>et al.</i> (2016); Vijayaraghavan <i>et al.</i> (2011)
Merits of plant-based coagulants: - Effectiveness aspects, no effect on pH - Environmental and sustainability aspects: Safe for human health, biodegradability, inexpensive, low-level sludge production	Yin (2010); Feria-Díaz <i>et al.</i> (2016); Vijayaraghavan <i>et al.</i> (2011)
Effectiveness of cactus/<i>Opuntia ficusindica</i> as a coagulant	
Removal of turbidity	Lozano <i>et al.</i> (2015); Betatachee <i>et al.</i> (2014); Rodiño-Arguello <i>et al.</i> (2015); Fedala <i>et al.</i> (2015); NougboDé <i>et al.</i> (2013); Pichler, Young, and Alcantar's (2012); Shilpa <i>et al.</i> (2012); Yang <i>et al.</i> (2007); Mukhtar <i>et al.</i> (2015); Nharingo and Moyo (2016); Mouniret <i>et al.</i> (2014); Miller <i>et al.</i> (2008)
Removal of suspended solids	NougboDé <i>et al.</i> (2013); Sellamiet <i>et al.</i> (2014)
Removal of dissolved solids	Lozano <i>et al.</i> (2015)
Removal of chemical oxygen demand	Sellamiet <i>et al.</i> (2014); Taa <i>et al.</i> (2016); Bouatay and Mhenni (2014); Nharingo and Moyo (2016)
Removal of biological oxygen demand	Taa <i>et al.</i> (2016)
Removal of color	Lozano <i>et al.</i> (2015); NougboDé <i>et al.</i> (2013); Bouatay and Mhenni (2014); Nharingo and Moyo (2016)
Removal of heavy metals	Nharingo <i>et al.</i> (2015); Nharingo and Moyo (2016)
pH Implications	
Little/marginal/no effect on final pH	Yang <i>et al.</i> (2007); Mukhtar <i>et al.</i> (2015); Lozano <i>et al.</i> (2015)
Efficient removal over wide pH range	Mukhtar <i>et al.</i> (2015); Nharingo and Moyo (2016); Taa <i>et al.</i> (2016)
Specific pH optimum for removal	Shilpa <i>et al.</i> (2012); Mouniret <i>et al.</i> (2014); Miller <i>et al.</i> (2008); Nharingo <i>et al.</i> (2015); Bouatay and Mhenni (2014)

Use of Plant-Based Coagulants:-

The literature reviewed reveals two main drivers behind the emergence of natural plant-based coagulants for water treatment, surrounding effectiveness, environmental, and sustainability considerations. The first driver entails the demerits associated with conventional chemical coagulants, which create an opportunity for consideration of their plant-based counterparts. Summing the demerits across the studies reviewed indicates that chemical coagulants are ineffective at low water temperatures, significantly alter water pH, decrease transmission capacity, and cause corrosion problems, besides causing environmental and sustainable development concerns such as costliness, detriments to human health, non-biodegradability, and production of large sludge volumes (Yin, 2010; Feria-Diaz *et al.*, 2016; Vijayaraghavan *et al.*, 2011). The number and significance of disadvantages associated with synthetic chemical coagulants, especially considering the need for environmentally conscious and sustainable water treatment approaches in today's world, therefore have been a driving factor behind the emerging consideration of plant-based coagulants for use in water treatment. One study by Yin (2010) contextualizes the problem to developing world

settings, where conventional water treatment may be insufficient or absent while at the same time proving too costly and environmentally undesirable.

The next significant driver for the use of plant-based coagulants is being necessitated by the merits presented by such coagulants, which are significant when juxtaposed against the drawbacks of chemical coagulants. The studies reviewed altogether indicate that plant-based coagulants are advantageous in aspects such as safety for human health, biodegradability, low cost acquisition, low-level sludge production, effectiveness in treating water, and no significant alteration pH of treated water (Yin, 2010; Ferial-Diaz *et al.*, 2016; Vijayaraghavan *et al.*, 2011). Notably, most of the aforementioned merits indicate that plant-based coagulants such as cactus align with local, national, and global objectives in environmental protection and sustainable development. Such observations indicate that plant-based coagulants provide promising alternatives to synthetic chemicals commonly used in coagulation. In addition, the local availability noted in literature such as Yin (2010) also suggests that plant-based coagulants contribute to sustainable development of potable and wastewater treatment in underdeveloped regions of the world where there are challenges currently on treatment capacity. Such findings demonstrate the opportunities for rural and urban places in Kenya to explore the use of locally available plant-based coagulants.

Effectiveness of Cactus as a Coagulant:-

Besides its merits in promoting environmental and sustainable development concerns, the prospects of cactus in wastewater treatment in Kenya also centrally depends on its effectiveness as a coagulant. Narrowing down to the use of cactus in coagulation, evidence for removal of various contaminants and undesirable elements in wastewater varies. From the literature reviewed, there is overwhelming evidence regarding the effectiveness of the plant parts and extracts in the removal of turbidity, as demonstrated in the summary table (Table 2) indicating that twelve of the nineteen studied articles established such effectiveness. There is also some evidence for the effectiveness of cactus in removing chemical oxygen demand and coloration in wastewater from four studies for each of these parameters, which suggests that cactus may be useful in this respect. Meanwhile, removal of heavy metals (two studies), suspended solids (two studies), and dissolved solids (one study) indicates the need for more inquiry to establish sufficient evidence for the effectiveness of cactus in such respects. From these observations, the inference is that cactus has proven value in removing turbidity and significant evidence for removal of color and chemical oxygen demand, while removal of heavy metals, suspended solids, and dissolved solids still call for further research. Notably, the effectiveness of cactus in the aforementioned areas was influenced by the use of varying conditions in aspects such as pH, optimum dosage, agitation speeds, and temperature. As a result, application of cactus in wastewater treatment in the Kenya context also necessitates studies to establish the optimal conditions within which the plant can be effective.

Table 2:- Evidence for the Effectiveness of Cactus in Wastewater Treatment.

Contaminant Removed	Number of Supporting Studies
Turbidity	12
Suspended solids	2
Dissolved solids	1
Chemical oxygen demand	4
Biological oxygen demand	1
Color	4

Implications for pH:-

One determinant of the effectiveness of cactus as a coagulant is pH implications, where the pH of the treated water determines the value of cactus for coagulation. Besides, pH of water or wastewater being treated also constitutes a crucial consideration for optimal coagulation. In terms of effects on the final pH, three studies reviewed reveal that cactus has little to no effect, which makes the treated water safe for use (Yang *et al.* 2007; Mukhtar *et al.*, 2015; Lozano *et al.*, 2015). These findings underscore the promise of cactus as a coagulant, especially considering that significant alteration of the pH of treated water is one of the demerits noted among synthetic chemicals commonly employed as coagulants.

The studies regarding the pH at which cactus is effective in the coagulation process provide diverging observations. On one hand, three studies reveal that the effectiveness of cactus in treating wastewater occurs across a wide range of pH values (Mukhtar *et al.*, 2015; Nharingo&Moyo, 2016; Taa *et al.*, 2016). Meanwhile, five other studies indicate that the effectiveness of cactus in the coagulation process is determined by pH, where the plant is effective within a

narrow pH range (Shilpa *et al.*, 2012; Mouniret *al*, 2014; Miller *et al.*, 2008; Nharingoet *al.*, 2015; Bouatay & Mhenni, 2014). In addition, the values of pH given as optimal for effective coagulation vary among the studies. Shilpa *et al.* (2012) find cactus effective at an optimal pH of 9 for turbidity removal, Mouniret *al.* (2014) at pH 3.0 for turbidity removal, Miller *et al.* (2008) at pH 10, Nharingoet *al.* (2015) at pH 5 for heavy metal removal, and Bouatay and Mhenni (2014) at pH 7.25 for removal of turbidity, color, and chemical oxygen demand. Such observations indicate that optimal pH may vary according to the contaminant being removed. At the same time, optimal pH may be varying because of the source and properties of the water, as well as the cactus species used in coagulation. Based on these observations, use of cactus in the Kenyan context may first require studies to establish effectiveness under various acidic or alkaline conditions using the Cactus specie that is native to the country.

Conclusion:-

The study sought to explore the promise of cactus as a natural plant-based coagulant for the environmentally friendly and sustainable treatment of water and wastewater, which would benefit developing world contexts such as Kenya. Following a review of the literature, the study establishes that plant-based coagulants offset the demerits associated with conventional chemical coagulants, while also being locally available. In this case, the pursuit of plant-based coagulants such as cactus is happening because such coagulants have the potential of not only being effective, but also allowing wastewater treatment that is sustainably manageable and environmentally appropriate. Such merits over traditional synthetic coagulants would be highly appropriate for the Kenyan context, helping to not only address water stress problems, but also deliver an approach aligned with Kenya's pursuit of sustainable development goals as set in the UN's 2030 Agenda. Cactus has been found to be effective in removal of turbidity, chemical oxygen demand, and coloration, with other possible uses being in the removal of heavy metals, suspended solids, and dissolved solids. The studies reviewed also showed that cactus has no significant effect on the pH of the treated water, unlike most synthetic chemical coagulants. However, there is no consensus regarding whether cactus is effective over a wide range of pH values or across a narrow range of pH, as well as specific optimal pH for highest efficacy. Considering such effectiveness alongside the advantages in biodegradability, local availability, and lower costs, cactus potentially represents an environmentally friendly and sustainable solution for wastewater treatment in the Kenyan context. Further scholarship into cactus as a coagulant in the Kenyan context is necessary before a recommendation is made, where experimental models will help establish its effectiveness, optimal conditions, and cost-effectiveness for its successful use.

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RESEARCH ARTICLE

ASSESSMENT OF BREAST CANCER RISK AMONG WOMEN USING THE GAIL MODEL, EGYPT.

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Breast Cancer, Risk Assessment, Gail model, Prediction.

Abstract

The rationale of this study: breast cancer is an increasing global public health problem due to its high incidence and mortality. Lots of debates over the actual value of screening program at population level continue justifying, enthusiasm for risk-stratified screening is gaining momentum and can be useful for guiding public health strategies of breast cancer prevention. The Gail model is considered the best available means to quantify an individual woman's risk of developing BC and is crucial to provide risk-benefit analysis before deciding interventions designed to lower breast cancer risk.

Objectives: to identify the mean five years and lifetime risk development of breast cancer among Egyptian women.

Methods: cross sectional study conducted on a sample of 156 women attending family planning clinics using systematic random sampling technique. The instrument used is based on the information required for analysis by the Gail model.

Results: after analyzing the collected data according to the Gail Model, about 30% of the sampled females had high risk for developing BC >1.66% with their mean five-year breast cancer risk was 1.53 ± 1.49 , and their mean lifetime breast cancer risk up to age 90 years was 20.54 ± 11.86 .

Conclusions: our findings surges the need for BC screening services for early detection directed mainly for women with multiple risk factors making informed decisions about the screening methods best suited to their individual situations.

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Introduction:-

Although advances in early detection, diagnosis, and treatment, led to significant declines in breast cancer (BC) deaths yet it remains the most prevalent and the second leading cause of cancer deaths in females worldwide based on the findings from the WHO Global Health Estimates 2013⁽¹⁾. BC among Egyptian women, accounts for 18.9% of total cancer cases, according to findings from the Egyptian National Cancer Institute's (NCI),⁽²⁾ Incidence rates are steadily rising in the world. Between 2002 and 2020, breast cancer incidence and mortality are expected to rise 50%, with the greatest rise occurring in developing countries 19,20. With the steadily rising incidence rates, it is imperative to continue research, increase resource opportunities and raise global awareness of BC⁽³⁾.

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Egyptian BC patients showed a shift towards a younger age distribution affecting mainly 30–60 years of age. The mortality rate of BC is decreasing in developed countries, compared to the increasing mortality in the developing countries as people are diagnosed with BC in late advanced stages⁽⁴⁾. According to WHO, early detection is the cornerstone for BC control and it can improve its survival and is extremely important to reduce the burden on the health care system. There is growing interest in trying to estimate individual BC risk women and follow-up screening for “high-risk” individuals. This not only to improve early detection, but also to avoid many unnecessary inspections and reduce the burden on the economy if the women subgroup with higher risk can be adequately identified (5). Consequently, the accuracy of a woman's perception of her risk of developing BC has gained importance as a priority for primary prevention. “High-risk” individuals in BC can be identified by the risk assessment tool for women aged 35 and older⁽⁶⁾.

BC is one of the diseases predominantly influenced by risk factors, according to American cancer society, the reported risk factors of BC are being a female, ageism, race, ethnicity, younger age at menarche (before 12 years old), older age at menopause (55 years old), mutations in BC genes namely BRCA1 or BRCA2, history of BC in first degree relatives, having history of previous BC, some benign breast diseases, hyperplasia or lobular carcinoma in situ, exposure to huge dose of radiation at a young age, HRT, smoking, postmenopausal women, overweight or obesity, didn't breast feed their babies, and being physically inactivity⁽⁷⁾.

Several models were developed to assess the additive effect of multiple risk factors to estimate the overall risk. Gail model (GM) is commonly used BC risk assessment model and is used to assess the individual risk of BC. And estimating the probability of a currently healthy women having given risk factors will develop BC within 5 years and lifetime by developing a mathematical model that provides individualized risk estimates of developing BC with varying risk factors, including age, age at menarche, number of prior breast biopsies, age at first live birth, and number of first degree relatives affected with BC. Relative risk was calculated for each of these risk factors; then were used to calculate the absolute five years risk from the time of assessment and a lifetime risk up to 90 years old⁽⁸⁾.

The results of risk assessment can be used to develop an individualized plan to assist patients in decision-making regarding the implementation of frequent surveillance, chemoprevention, or prophylactic surgery⁽⁹⁾. In women aged ≥ 35 years with a 5-year risk of $\geq 1.67\%$, it is recommended to perform CBE biannually and mammography annually. It is crucial that among a population with increasing incidence and mortality, to monitor the risk of developing BC and take precautionary public health interventions. The purpose of this study is to utilize quantitative assessment to. Despite the fact that, there are a lot of up-to-date investigation equipment and screening projects directed for females, most of them are very expensive. Therefore, if the subgroup of specific high-risk females can be identified, unnecessary inspections avoided and burden on the economy reduced. It can also achieve the purpose of early detection and early treatment of BC. Those who are at high risk for BC (Gail score $\geq 1.67\%$) will benefit more from invasive approaches, while women who are categorized as low risk are followed-up through the routine screening program to avoid complications and high costs⁽¹⁰⁻¹¹⁾. When women are more knowledgeable about their personal risk, they can make informed decisions about prevention methods and screening options to detect cancer early. Women at higher risk should be educated on their need for higher screening methods such as magnetic resonance imaging, and initiating screening at an earlier age and at more frequently should be taken into consideration. Ultimately, planning and implementing early detection and screening programs of BC can be more successful⁽¹²⁾.

Hypothesis:- using the Gail model helps predict BC risk.

Objectives:-

- 1- To estimate the risk of developing breast cancer over next five years
- 2- To estimate the risk of developing breast cancer over lifetime periods

Methodology:-

Study design and setting: to fulfill the objectives of the study a cross sectional study conducted over 4-month period (from May to end of September 2016) on women attending the family planning outpatient clinic at Zagazig University Hospital.

Sampling technique and sample size:-

The participants considered eligible for the study if they met the following inclusion criteria: being able to read and write women, without a history of BC and being 35 years or older (as in the GM) as GM provides the five-year and

lifetime (up to age 90) BC potential risk for women ≥ 35 years. The exclusion criteria included women younger than 35 years, refused to participate, and those suffering of acute medical conditions. A sample size of 156 women was calculated using open epi program ⁽¹³⁾, based on the following parameters: 80% power of the test, 95% confidence interval, and 19%¹³, 450 monthly flow to the clinic, and prevalence from published literature ⁽²⁾. For data collection (three days per week) using a systematic random technique, where every third (450/156), attending the clinics included if she met the inclusion criteria. If did not meet the eligibility criteria, then the next (4th) was selected and so on till the required sample size was achieved.

The tool for data Collection:-

The assessment made using a self-administered questionnaire, the instrument used for data collection is the BC risk assessment tool using the model developed by Gail ⁽⁸⁾. The questionnaire formed of 2 parts, the first part about the socio-demographic characteristics: educational level, occupation status, place of residence and, marital status.

The second part included information about women's characteristic and risk factors related to breast cancer: age, age at first menstrual cycle whether (responses were: at least 14 years, 12 to 13 years, or fewer than 12 years), age at first childbirth (responses were: nulliparous, fewer than 20 years, 20 to 24 years, 25 to 29 years, or at least 30 years), and having a family history of BC (i.e., mother, sister) and their number. History of previous breast biopsy, those women had had any previous biopsy were asked about the number of biopsies (1, or 2), presence of atypical hyperplasia in a biopsy specimen (yes or no).

Questionnaire translation and Pilot testing:-

Using forwards- backwards translation process by two bilingual expert translators, then comparing the agreement (between the original English version and the back translation version) was done. The translated Arabic version of the questionnaire pilot tested on a sample of females before the actual commencement of the study to assess its face validity, its clarity and the time required for filling it up.

Statistical Methods:-

The collected data were analyzed by SPSS 24 ⁽¹⁴⁾ using descriptive statistics including the mean, standard deviation, frequency, and percentage. To estimate each woman's 5-year and lifetime risk for developing BC, calculated using the software program available from the NCI online ⁽⁸⁾. We used 1.66% in Gail score as a cut-off value to define low and high risk groups for developing BC according to the estimated BC five years risk assessment as women with the BC risk of $\geq 1.67\%$ were considered as high-risk ⁽¹⁵⁾.

Ethical considerations:-

Before collecting data, the entire required official permissions were obtained before carrying out the study. Participants were informed that their participation is voluntary and were informed about the objectives of the study and importance of the research and those who agree to participate signed an informed consent. The confidentiality of the collected information is assured through anonymity of the participant, the questionnaires were numerically coded and the study results would be used only for the purpose of research.

Results:-

Table 1: - Personal characteristics of the sampled females (n=156).

Variables	Categories	No.	Percent
Marital status			
	Not married	31	19.9
	Married	125	80.1
Educational level			
	Primary	12	7.7
	Secondary	56	35.9
	University	88	56.4
Employment state			
	Not Working	40	25.6
	Working	116	74.4
Family income			
	High	19	12.2
	Middle	107	68.6
	Low	30	19.2
Total		156	100.0

Our finding revealed the personal characteristics of the sample in table (1) as the majority of them were married, working, having middle income and are university graduates (80.1, 74.4, 68.6 and 56.4%) respectively.

Table 2:- Distribution of Risk Factors among Participants according to the Gail model.

Variables	Category	No	Percent
Age groups (y):			
	36- 45	92	59.0
	46-55	29	18.6
	≥55	35	22.4
Age at 1st menstrual period (y):			
	Unknown	13	8.4
	≥14	19	12.2
	12-13	84	53.8
	7- 11	40	25.6
Age at 1st child birth (y):			
	No children	8	5.1
	≤ 20	16	10.2
	20-24	93	59.7
	25-29	29	18.6
	≥30	11	6.4
Number of 1st degree relatives with breast cancer			
	Unknown	6	3.8
	Zero relative	33	21.2
	1 relative	72	46.2
	More than 1 relative	45	28.8
Biopsy			
	Unknown	0	0.0
	Yes	19	12.2
	No	137	87.8
TOTAL		156	100.0

On assessing women's risk factors among the study subjects the results showed that the majority of them (59.0%) aged from 36-45. 53.8% experienced their first menses at age 12 to 13; 59.7% of the sample women had first live birth in age group (20 - 24) years old, 46.2% of them had first degree relatives suffered from BC. Only 12.2 % of them performed breast biopsies. Table 2

Table 3:- Breast Cancer five-year Risk and life time Risk Based on the Gail Model.

Risk	X±SD	Minimum risk	Maximum risk
Mean five-year risk	1.53 ±1.49	0.3	10.80
Mean five-year risk for women of the same age without risk factors	0.91±0.44	0.3	1.80
Mean risk of participants up to age 90 years	20.54±11.86	6.8	91.6
Mean risk up to age 90 years for women of the same age without risk factors	12.65±9.14	9.10	95.0

Table 3 assessed the mean five-year BCR was 1.53 ±1.49 according to the GM, and The mean lifetime BCR up to age 90 years was 20.54±11.86. In comparison with women of the same age and average risk factors, 46 (29.6%) had a higher five years risk and 29 (18.7%) had higher lifetime risk (figure 1 and Table 3).

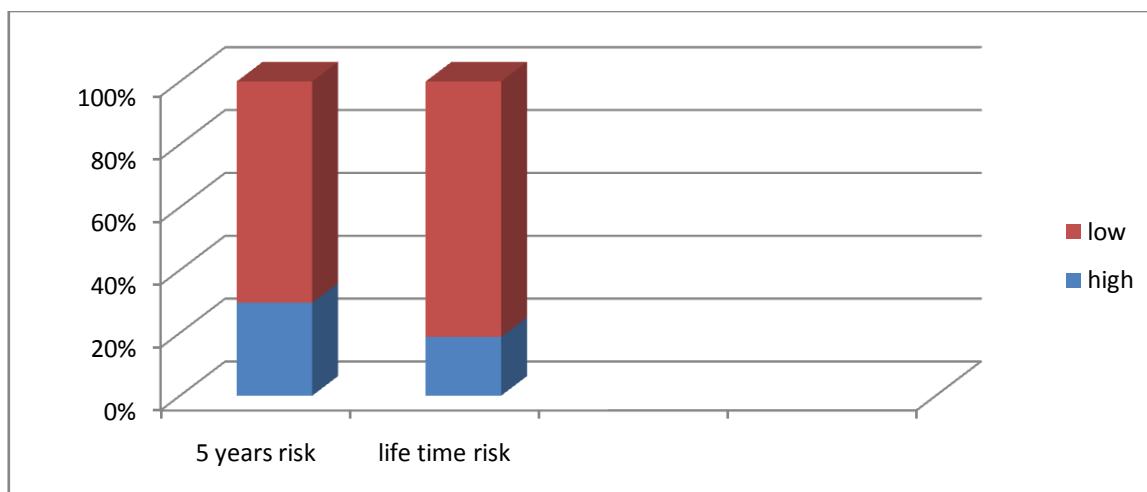


Figure 1:- comparing high and low 5 years and lifetime risk among the sampled female.

Discussion:-

Breast Cancer Risk Assessment (BCRA) is crucial when making decisions about screening for individual women with recommended intervals for screening mammography. Breast cancer risk was calculated using the National Cancer Institute's on-line BCRA Tool or the Gail Risk Assessment Tool developed by Gail, based on data from the BC Detection and Demonstration Project, a mammography screening project conducted on 280,000 women between 1973 and 1980. By applying the GM for the current study, the findings showed that about (30%) of the sample had a higher five years risk and about (19%) had a high lifetime risk. On the other hand, our findings are higher than other previously published researches⁽¹⁶⁻¹⁸⁾.

BC risk factors using the GM are as follows: current age, age of menarche, previous breast biopsies, including the number and presence of atypical hyperplasia, age of first live birth, family history of BC in first-degrees, and ethnicity⁽¹⁹⁾. The findings of the current study support the growing body of evidence from female breast cancer incidence suggesting that age is a well-known risk factor for BC susceptibility²⁴, as the findings revealed that the majority the sampled women (59%) were in age group 35-45 years old. The finding contradict with another 2 studies indicated the highest occurrence of female BC is shown in the age category of 45-50 years⁽²⁰⁾. A much higher risk for BC was found in women who had experienced early menarche, a previous breast biopsy, and a first live birth after 30 years of age⁽²¹⁾.

Regarding the age of the sample at first childbirth we found that the majority of them (59.7%) have their birth were in the age group from 20-24 years, this supported the long-standing hypothesis that the earliest age at first pregnancy and longer duration breastfeeding has a protective effect lower incidence of BC⁽²²⁾. Nulliparity and first live birth at older than 30 years of age are associated with an increased risk of subsequent breast cancer. Women who have never given birth or who have given birth to their first child after age 30 are at high risk for BC⁽²³⁾. Pregnancy at a young age is associated with a markedly reduced risk for BC⁽²⁴⁾.

In a previous study, after controlling for age, the greatest increase in risk has been associated with a family history of BC but the number, type, and age at onset in their relatives are important in estimating the magnitude of risk⁽²⁵⁾. Together with having family history in first-degree relative increasing the susceptibility to develop BC, where one first-degree relative could double the risk and having two first degree relatives increases the risk approximately 3-times. The findings in present study illustrated that 46.2% have one first degree relative this is even higher than findings of a study conducted in Alexandria, where the 20% of them had first degree cousins and nearly two thirds of the cancers which will appear in the next 25 years will occur due to unawareness of the public⁽²⁶⁾. Which surges the need for BC screening for early detection prior to the onset of symptoms through various methods as frequent monthly breast self examination, periodic clinical examination and mammogram and management of breast cancer to decrease the burden of morbidity and mortality associated with the disease⁽²⁷⁾.

Conclusion and Recommendations:-

The current study showed that about thirty percent of the sample had a higher five years risk and nineteen percent had higher lifetime risk.

1. Our findings surges the need for BC screening services for early detection directed mainly for women with multiple risk factors with health education on proper performance of monthly BSE, stress on the importance periodic clinical examination and mammography and making individualized informed decisions about the screening methods best suit them through individual counseling approach.
2. For clinicians , health providers and practitioners, incorporating risk assessment as part of routine clinical visits using the GM can supply women with accurate information about BC risk factors, consequently the appropriate prevention strategies and screening modalities can be tailored to each individual woman's risk profile to lower their risks.
3. For policy makers, they need to put an organized screening program on their agenda and develop national breast cancer screening guidelines.
4. Systematic screening and Early detection of breast cancer is improved by a follow-up screening for "high-risk" individuals. Therefore, it is important to define what is meant by "high-risk" individuals.
5. Women at higher risk should be educated on the their need for additional screening methods also consider initiating screening at an earlier age and at more frequently. When women are more knowledgeable about their personal risk, they can make informed decisions about prevention methods and screening options to detect cancer early.

Conflict of interest:- the author declared no conflict of interest.

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Ethical approval:-

"All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards."

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RESEARCH ARTICLE

THE RELATIONSHIP BETWEEN LEARNING PROCESS IN PESANTREN AND ENERGY CONSERVATION LAW (STUDY OF THE INTERNALIZATION OF RELIGION VALUES RELATED WITH LIFE ENVIRONMENT).

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 punishment.

Abstract

Research attempts to review the understandings and concepts of life environment learned in Moslem Boarding House (Pesantren), to examine how is the learning of religion values related with life environment, and to acknowledge how this learning is implemented. This research was located at Pesantren Al Kahfi Sidoarjo, and exploration was done through several ways. First, primary data are collected from respondents of santri (students in Pesantren), either boys and girls. Second, in-depth interview is conducted with the managing boards of Pesantren. The findings of research were described as following: (1) The learning is given to produce a change in students on the directions made by Pesantren. This change concerns with environmental issue, meaning that the understanding of students about environmental issue would change. Most students (90.2%) have understood life environment concept and agreed if the review of life environment must be delivered every day or every week. Others (98.7%) have agreed with proverb that cleanliness is a part of the faith. (2) Few students reject such life environment concepts and its reviews. The reason behind this reject is that there is still a strong patriarchy culture which frames the way the students think. The condition where students live still influences their behavior. (3) The learning is emphasized on the importance of environmental cleanliness. It is done by reciting books about this issue, by organizing the outing class or the contests of making poster with contents of environmental cleanliness and learning comfort, and by conducting homage service around Pesantren. (4) Some students (78.1%) agree that homage service could be held once a week, and some others (75.6%) declare that punishment should be given for the negligent. (5) Any violation to the agreement (homage service) will be subjected to fine or other kind of punishment.

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Introduction:-

Pesantren is an organization of preaching or education. It has some functions. First, Pesantren prepares Moslem youths with education in order to produce Islamic generation with good moral quality (khaira ummah), with an understanding on mission of amar ma'ruf nahi munkar and also with a sense of kindness. Second, Pesantren is also a

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place to educate the cadres of Moslem scholars, the agent of excellence, and the developer of science, especially theology. It can be said that the tasks of Pesantren are to prepare and educate thâ'ifah mutafaqqihah fid-dîn, respectively the cadres of Moslem scholars and the caretaker of Pesantren who always respect personality of Prophet Mohammed and have a willingness to implement the duty of indzârul qawm (Abdurrahman, 2008).

A certain effort may determine the force that moves a certain thing. But, this effort does not influence the length of time when the force subjects to a thing to move. Sometimes, the effort is fast, or even slow at other times. For instance, Siti push the cupboard to move it to the other corner at distance of 3 m. Siti needs 5 minutes to move the cupboard there. Budi can move the same cupboard only by 3 minutes. Siti and Budi do same effort, but time needed for each effort is different. The effort per unit of time is called energy. It is then said that Budi has bigger energy than Siti. Energy is defined as the rate of effort in unit of time. Energy represens a capacity to use the effort. Energy is indestructible but it only changes its shape.

Method of Research:-

Research uses a mixed method comprising of quantitative and qualitative measures. Both are used simultaneously in a way suggested by Creswell (2012), and it is called as concurrent mixed methods.

Quantitative method is represented by survey, whereas qualitative method involves case study. Survey is used to collect quantitative data, especially those concerning with undertandings of life environment in Pesantren and the effect the life environment may have on the learning in Pesantren. Case study is useful to collect data on the description of "unique cases" related with learning process.

The object of research is Pondok Pesantren Dakwah Al Kahfi, Tarik-Sidoarjo. This Pesantren also opens formal school, called Integated Islam Junior High School. It tries to prepare Moslem youths with Islamic personalities, knowledge of Islam religion, Arab language, knowledge of preaching, and general knowledge.

To enrich information about these unique cases in learning process, then in-depth interview is conducted with some selected informants, which in this case, involving teachers and religious elders or Kyai. Questionnaire is given randomly to Pesantren students.

All data are subjected to analysis. First, the analysis of quantitative data involves arranging the data into frequency table to describe the reality observed. Second, qualitative data are analyzed by displaying data into narrative textual explanations to describe the reality observed.

Result and Discussion:-

The Learning Process in Pesantren Al Kahfi:-

Religion values represent the guidance to produce good, fair and proper behavioral standard. These values are derived from religious norms. These norms usually bind the believers into several rules or prohibitions. Believers must obey the rules to obtain happiness in the world and the hereafter. Violating these rules will lead to the act of sin.

Educational environment in Pesantren is very suitable to be a secondary learning medium to create good personality. Formal education does not match with this personality goal. It is also incompatible for personality building because teaching at formal school is not wisely given and teaching style is too much monotonous. Learning structure and infrastructure are not feasible to develop good personality in students.

The list of incompatibility factors still grows. Students may be assigned into less proper learning groups. Improper learning habit, poor learning achievement, bad learning behavior among friends, and too strict school regulation, are only factors causing difficulty, anxiety, disappointment and dissatisfaction among students. Healthy and convenient school environment is therefore needed. Education at school is a secondary learning place, and it is where formal learning is done. At school, children not only learn how to read, write and count, but also understand the importance of self-dependence, achievement, universalism, and specificity.

Learning Process of Religion Values. Learning the religion values related with life environment in Pesantren Al Kahfi is an action directly done by students with their consciences. Students are aware with the importance of

working together with friends and of making interaction with other individuals in Pesantren. Rules in Pesantren must be understood to keep students to realize that certain norms and values have prevailed in Pesantren.

After accepting these rules, students are considered as able to take a certain position into the community. In other words, students indeed have a capacity of tolerance and interaction not only with others at same age, but also with the community.

Curriculum To Support Religion Values. Learning environment in Pesantren Al Kahfi is a classroom type but religion values and character education are set into learning materials. Religion material (diniyah) is also given and it is supported by reviewing some religion books, such as (1) Book of Bulughul Maram (containing rules of devotion and rules of behavior); (2) Book of Taklim Muta'alim (explaining politeness and morality); (3) Book of Fiqh Sayid Tsabit (concerning with rules of devotion, issues of holiness and cleanliness, others); (4) Book of Siroh Nabawiyah (history of Prophet Mohammed and his struggle on Islam mission); and (5) Conflict Management.

Change Process in Pesantren. Social change has many definitions. According to Ranjabar (2001;17), social change is a process where structural change of the community goes along with the change of the culture and function of a social system. In the words of Gillin and Gillin (in Poloma, 1979), social change is a variation of life styles that are accepted as good because it is compatible with the change of geographical condition, material culture, geographical composition, or ideology. The change may be desired because it is diffused fast or there is new inventions in the community. In short, Samuel Koenig (1957; 279) said that social change refers to modifications occurring in the living pattern of the human. These modifications occur with internal and external causes.

Students as one stakeholder in Pesantren are subjected to the rules prevailed in Pesantren. These rules will organize their activity in 24 hours a day from wake-up in the morning to the sleep at night. Learning-teaching activity is also conducted on these rules.

Pesantren is unique if compared to other educational institutions. Students or santri in Pesantren are living together with their teacher or kyai in one certain complex of self-dependence settlement. The following is general characteristics of Pesantren.

- a. There is intimate relationship between santri and kyai.
- b. Santri is always obedient and submissive to kyai.
- c. Santri lives at simple and self-dependent ways.
- d. There is mutual-share with sense of kinship.
- e. Santri is trained with discipline and asceticism.

In the history of Indonesia, Pesantren plays significant role in enforcing faith, improving piety, fostering morality, developing self-dependence, and enlightening the nation through informal, non-formal and also formal educations provided by Pesantren. Being informal institution, Pesantren acts as a big family that helps enshaping the personality of santri. Pesantren also teaches skills that may help santri to develop their self-dependent capacity.

Energy and Effort. Energy plays very important role for the living of human and also for the progress of a nation. All human activities are done with energy. From pre-history to history periods, woods are the only source of energy for cooking and heating. In early part of thirteenth century, the coal was introduced. Steam engine was invented and using coal as its energy source until at eighteenth century, when new development came to the human life. Steam engine could produce quite enormous energies to mobilize industrial machines. It shot the fire of industrial revolution in European countries where energy was used in huge quantity.

In the early of nineteenth century, the emergence of fossil-oil as energy source has replaced coal as the source of heating and lighting. This oil is then displaced by electric in the end of nineteenth century. Electricity is produced from the rotating components in generator. However, energy source used to activate generator is coming from fossil-oil, coal, and natural gas. Electrical energy plays important role in human living. In twentieth century, some alternatives of energy are ready to be used by human, including native heat, nuclear, and solar energies.

Energy Conservation Law concerns with the change of energy. Some energies are considered, such as radiation energy, potential energy, and chemical energy. Radiation means as disseminating toward the affordable spots. Radiation energy can be found in the beams of the sun that subject directly to the earth, or the sense of warm from

the bonfire. Potential energy is an energy that moves to produce other energy. The silent propeller does not produce energy, but when the propeller moves with the wind, it creates electricity. Therefore, the propeller has changed its potential energy into kinetic energy, and finally, producing electric energy.

It is said that energy is a capacity needed for doing an effort. Someone may need energy to walk, run, work, and do other activities. Foods and beverages provide chemical energy to be burnt in human body, and this energy is used for many efforts or activities. Someone is said as doing an effort when pushing a certain thing. The rate of effort depends on the force used to push the thing and the distance of displacement of this thing.

The definition of effort in physic involves two requirements. First, the force subjected to a thing must move the thing to displace at certain distance. Second, to ensure that a force does an effort on the thing, then the force must have a direction in parallel with the direction of displacement. Effort in daily life is activity done by human. For instance, kyai tries to improve the progress of Pesantren with various strengths and strategies. Teachers apply learning method on students' needs. Santri learns physic for preparing daily exam and others.

The Relationship Between Learning Process and Energy Conservation Law:-

Pesantren Al Kahfi is full-day school at the level of Junior High School. Learning schedule ends in ashar (15.00 pm). Teachers control the process in shifts. In Teacher Office, male and female teachers are separated for convenience. This separation is aimed to reduce unnecessary talk and interaction, and the interaction ends in ashar. The exception of separation is only made during the meeting.

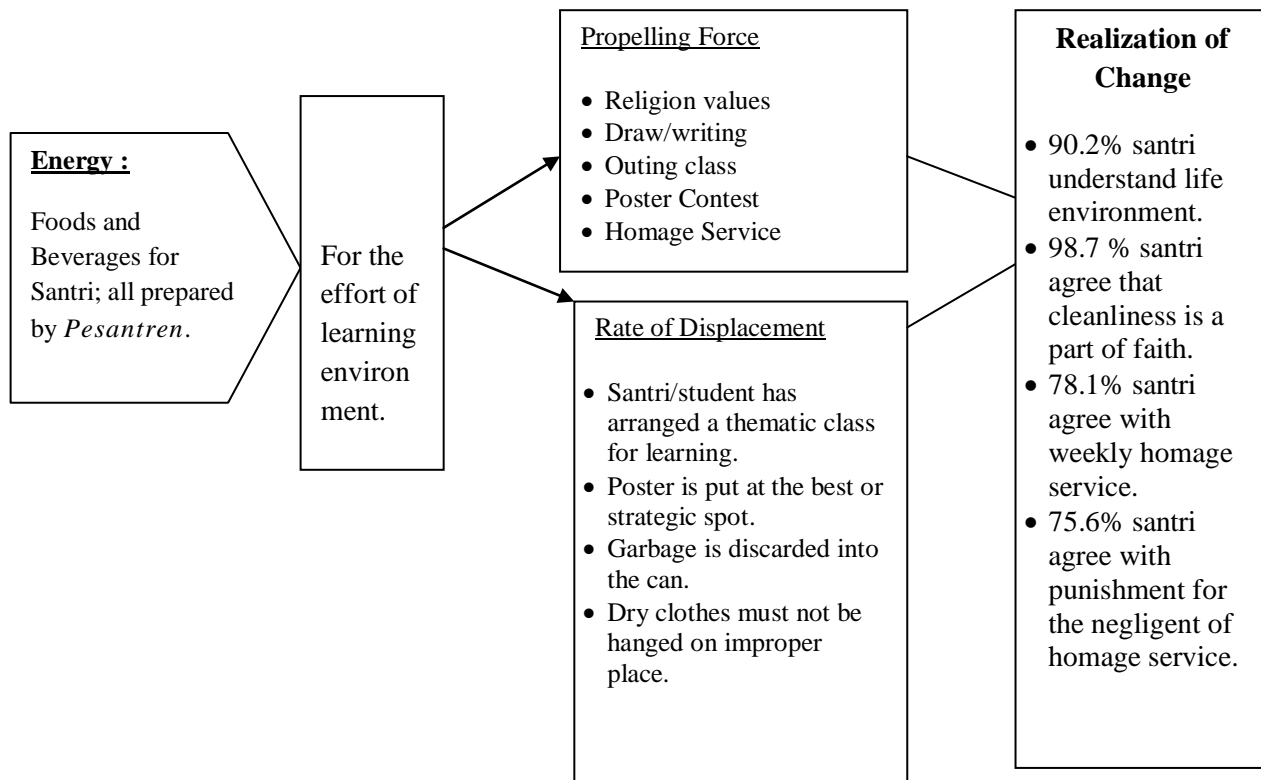
Learning activity begins from 03.15 am in early morning. Santri is waken up and prepared for sholat yaumul lail (night prayer), and after that, it continues with waiting the time for sholat Subuh. After sholat shubuh, santri recites the Pray of Al-Ma'tsurat and follows it with dzikir. Recitation of Al Qur'an is done until 6.30 am. When this ends, santri takes shower and does a breakfast. At 7.20 am sharp, school begins with diniyah (religion) and other materials relevant in Junior High School.

Pesantren integrates three materials into one learning package. These materials are: school internal (curriculum from National Education), diniyah (religion, related with interpretation and riyadus sholihin) and Al-Quran recitation expertise. It becomes the distinctive marker of Pesantren Al Kahfi that distinguishes it from other Integrated Islamic School. Students at other schools may memorize 2 juz or 3 juz of Al-Quran. In Pesantren Al Kahfi, students at Second Grade have memorized 30 juz of Al-Quran, and one of them is Rani.

The integration of learning materials is communicated early to parent or student guardian. At Pesantren Al Kahfi, 100% items in the National Education Curriculum are implemented, but it is added with Religion and AL-Quran Recitation. It may answer the question of parent about what kind of school is provided by Pesantren. During the preparation of national exam, the materials of exam are drilled to expect students for obtaining high academic achievement.

The internalization of religion values related with life environment is conducted during learning process at school or Pesantren. These religion values are delivered by reciting religion books, by organizing the outing class or the contests of making poster with contents of environmental cleanliness and learning comfort, and by conducting homage service around Pesantren. Some students suggest punishment for the negligent, such as buying cleaning tools or cleaning two rest rooms in two days in a row. Santri also expects that teachers must intensify the message that damanging the environment is a sin.

If related to energy conservation law, learning at Pesantren Al Kahfi has brought a change consistent with the plan of Pesantren. The fact that learning process has given good change is described as following:



Learning process needs energy, and this energy is contained within foods and beverages that must be prepared by Pesantren. The rate of effort depends on the rate of force given to the effort and also on the rate of displacement (result of learning). All these rates are needed to ensure the success of learning.

Conclusion:-

Pursuant to data and discussion on the learning of religion values related with life environment in Pesantren, some conclusions are made.

1. Learning process about life environment in Pesantren is performed by internalizing religion values. It aligns with energy conservation law that expects a change from learning result.
2. The learning has brought a change in the direction planned by Pesantren. This direction concerns with environmental issue or with the understanding of students about the issue. Most students (90.2%) have understood that life environment must be maintained by keeping environment cleaned, discarding garbages into can, refraining from spitting at public space, and helping the people with difficulty. Few students reject this understanding, and the rejecter is dominated by male. However, there is agreement that the review of life environment must be given every day or every week. Only few students reveal a refute to daily or weekly review of life environment. Male students are dominant.
3. Most students (98.7%) have agreed with the relationship between cleanliness and faith. Learning process at school/Pesantren is related with the importance of environmental cleanliness. It is done by reciting books about environmental cleanliness, by organizing the outing class or the contests of making poster with contents of environmental cleanliness and learning comfort, and also by conducting homage service around Pesantren. Some students agree these measures despite few male students reject it.
4. Some students (78.1%) agree with once a week homage service. Other students (75.6%) agree to give punishment for the negligent. The suggested punishment is buying cleaning tools or cleaning two rest rooms in two days in a row.
5. Few male students reject life environment concepts and its reviews. This rejection derive from a strong patriarchy culture which frames the way the students think. The condition where students live still influences their behavior.

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RESEARCH ARTICLE

DIFFUSION OF PHYSICOCHEMICAL WATER QUALITY PARAMETERS IN A LAB SCALE WATER DISTRIBUTION NETWORK.

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Incomplete mixing, water quality, experimental network, diffusion, physico-chemical parameters

Abstract

This paper investigates the impact of incomplete mixing at cross junctions on the variation of physicochemical water quality parameters in a lab scale water distribution network. The experimental setup was constructed in the Hydraulic Laboratory of the Engineering Institute at the National Autonomous University of Mexico where a series of experiments with varying inflows, chlorine and natural organic matter concentrations were carried out. Water samples were collected at various sampling points for water quality analysis. The results show how water quality parameters can vary in water distribution network due to the impact of incomplete mixing at cross junctions.

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Introduction:-

The mission of a water distribution network is to provide safe drinking water to the costumers. The Safe Drinking Water Act (SDWA) is the federal law that protects public drinking water. Under the SDWA, the Environmental Protection Agency (EPA) sets standards for drinking water quality and implements various technical and financial programs to ensure drinking water safety. Although most treatment plants are able to achieve these standards, the water distribution systems can be a point where issues regarding pathogens can occur. Variations in hydraulic conditions at junctions can vary the mixing conditions and modify physicochemical parameters of water. Therefore, the objective of this research was to investigate the impact of mixing phenomenon at cross junctions on the diffusion of water quality parameters in the distribution network using a lab scale water distribution network.

Materials and Methods:-

Experimental setup and preparation:-

In order to study the impact of incomplete mixing at cross junctions on the diffusion of physicochemical water quality parameters in the network, a series of experiments were carried out in a laboratory network system. The experimental setup was divided into three sectors:

1. Sector A consisted of a cross junction pipe system with two inlets (North and West) and two outlets (South and East), Fig 1.
2. Sector B, supplied from East outlet of sector A and;
3. Sector C, supplied from South outlet of sector A.

The experimental setup was constructed with polyvinyl chloride (PVC). The network used pipes of diameter 32 and 38 mm (Pipe diameters of each segment are shown in the schematic diagrams).

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The system also included.

- a water reservoir (2.12 m³ of capacity);
- two storage tanks (450 L of capacity each) at elevated position for gravity flow;
- four flow meters (CZ300s model Contazara S.A, Spain) located at each inlet and outlet of the cross junction (sector A) to measure instantaneous flow rate in the system. To ensure the precision of the measurement, a relative error was calculated for each test, a margin of error of 5% was considered acceptable.
- ten flow control valves (labelled 1-10), one valve at each inlet and outlet of sector A and three at the entrance of sectors B and C;
- ten water valves (labelled a-j), one at each inlet and outlet of sector A and three at the exits of sector B and C;
- two dosing pumps (BL3-12, HANNA instrument , Mexico) were used to control the chlorine concentration released from the storage tank;
- an advanced system (CL763, B&C electronics, Italy) that detects chlorine concentration in the range from 0.1 to 20 mg/L was installed in sector A (at inlets and outlets). This equipment is designed for inflow continuous measurement of residual chlorine in solution. A different chlorine measurement method was used in sectors B and C and;
- a data logger (EI-USB-4, Lascar electronics, USA) was connected to each controller. Schematic diagram of the experimental distribution network is shown in Fig 1.

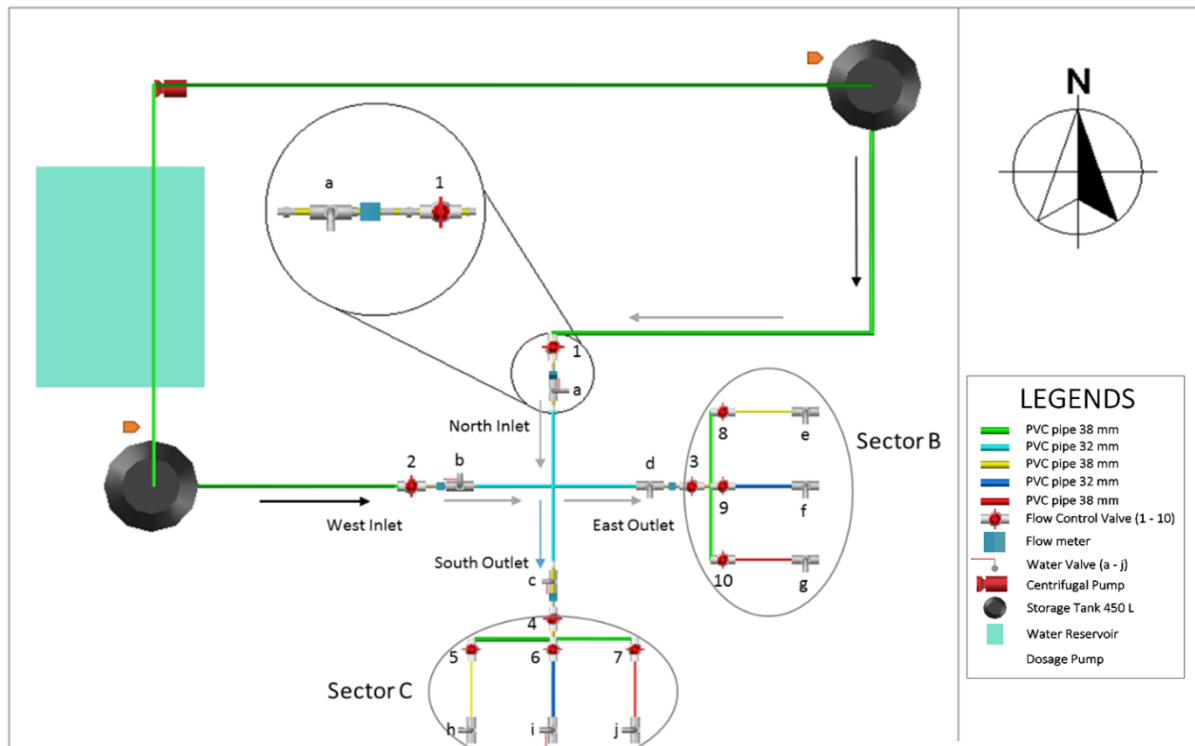


Fig. 1:- Schematic description of the experimental distribution network [sector A inflows at 90°]

In order to get a homogeneous mixture of tap water and chemicals (natural organic matter and chlorine), a manual mixer was installed at each storage tank (see Figure 2)

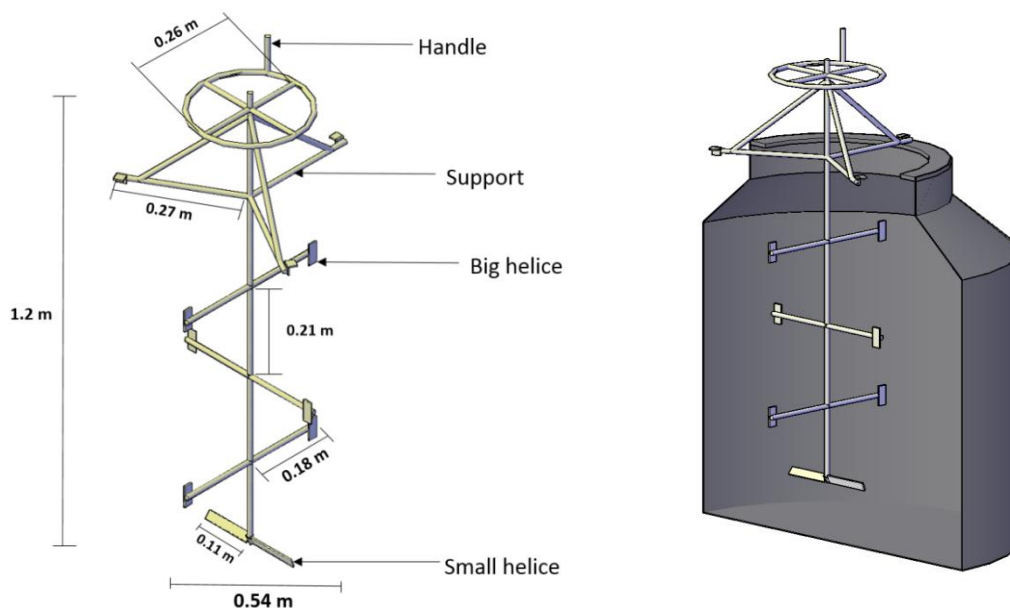


Fig. 2:-Schematic drawing of a manual mixer

Experimental procedure:-

To carry out the experiments, tap water from the reservoir was pumped to the elevated storage tanks using a 4HME200 centrifugal pump. K-Tonic solution (1.15 g/mL) which is a mixing of six compounds such as Total Nitrogen, Urea Nitrogen, K₂O water-soluble Potassium, Extract of Total Humic Carbon, Humic Acid Carbon and Fulvic Acid Carbon was used as contaminant agent. Then, 2.73 mL of K-Tonic solution were added to the North storage tank, which resulted in Total Organic Carbon (TOC) of 0.76 mg/L and 3.13 mL of the same solution (K-tonic solution) were added to the West storage tank to get TOC concentration of 0.87 mg/L. About 4.66 mL and 6.13 mL of sodium hypochlorite solution (at 13%) were added to the West and North storage tanks, respectively, which resulted in corresponding chlorine concentrations of approximately 1.35 mg/L and 1.77 mg/L. The solutions in each tank were mixed using a manual mixer.

The experiment provides varying inflows, varying chlorine and NOM concentrations. Prior to that, flow control valves 1 and 2 (of sector A) were maintained partially opened (approximately 50% and 75% respectively) to provide a flow rate of 0.50 L/s and 1.14 L/s at North and West inlets, respectively (Table 1). Flow control valves (from 6 to 10 of sectors B and C) were totally opened.

Instantaneous flow rates were measured at each inlet and outlet of sector A using the flow meters. Free chlorine concentrations were also measured at each inlet and outlet of sector A. Triplicate samples were collected at each sector for water quality analysis. Five minutes later, flow control valves 5, 6 and 7 as well as water valves e, f, and g of sector B were closed. Flow control valves (8, 9, 10) and water valves (h, i, j) of sector C were also closed at the same time. Thus, water was stagnated in sectors B and C. Once completed a period of time of eighteen hours, three samples were collected from pipe segments of sectors B and C for analysis of water quality. The experiment extended for about twenty-four hours.

Analytical Methods:-

Turbidity was measured using a turbidimeter (2100A, HACH, Mexico). Total organic carbon (TOC), Total Carbon (TC) and Inorganic Carbon (IC) analysis were carried out based on catalytic combustion at 720°C using a TOC-L CSH (Shimadzu Corporation, Germany). A 350 mercury thermometer was used to measure water temperature and an M530P (Pinnacle series, USA) unit was used for pH measurement. A DR 6000 UV-VIS Spectrophotometer with RFID Technology (HACH) was used for absorbance measurement, Total solids and Biochemical Oxygen Demand (BOD) were measured based on the methods for the examination of water and wastewater (APHA 1992), chlorine concentration of samples from sectors B and C was measured using the Iodometric titration method based on Na₂S₂O₃ titration. In this case, chlorine concentration was calculated using Eq. 1.

$$mgCl = \frac{(A \pm B) \times N \times 35450}{mL.sample} \quad [1]$$

where:

A mL titration for sample

B mL titration for blank

N normality of $Na_2S_2O_3$

Results and Discussion:-

Sector A:-

In the experiment, the flow at North inlet was blocked by the incoming water from the West inlet, which had greater flow (Table 1). This forced some of the flow (from North inlet) to get pushed across the junction and exit through the East outlet. This resulted in only slight mixing and had an impact on water quality characteristics at outlets. This results corroborated with those obtained by other researchers such as Chávez et al. 2005, Ho et al. 2007; Ho and O'Rear 2009 and Austin et al. 2008. Water quality parameters of sector A (cross junctions) are presented in Table 1.

Table 1:- Water quality parameters for samples from sector A

Sampling points	Flow rate [L/s]	Chlorine [mg/L]	pH	Abs UV	BOD [mg/L]	Total Solids [mg/L]	Turbidity [NTU]	TOC [mg/L]
North inlet	0.50	0.70	7	0.030	13.0	0.49	1.2	0.87
West inlet	1.14	0.35	7	0.026	10.5	0.36	0.9	0.76
South outlet	0.72	0.37	7	0.025	9.30	0.39	1.0	0.40
East outlet	0.92	0.53	7	0.020	13.0	0.43	1.1	0.69

Sectors B and C:-

Effects of incomplete mixing:-

Because of the incomplete mixing occurred at the cross junctions, physicochemical characteristics of the water varied in the same experimental setup. In the case of sector A, the level of disinfectant at East and South outlets was different. In general, water in the East outlet had higher values of free chlorine, Total Organic Carbon (TOC) levels, total solids, Biochemical Oxygen Demand (BOD), Turbidity that corresponded to the higher level of these parameters in the North storage tank. This is due to the fact that water in East outlet contained mostly water from North inlet and only a slight mix from the West inlet. Samples from sector B (which comes from East outlet of sector A) and sector C (which comes from South outlet of sector A) were collected after an incubation period of eighteen hours and taken to a laboratory for analysis of water quality. Results are presented in Tables 2 and 3 respectively.

Table 2:- Water quality parameters in sector B.

Sampling points	Chlorine [mg/L]	pH	Temperature [°C]	Turbidity [NTU]	TOC [mg/L]	TC [mg/L]	IC [mg/L]	BOD [mg/L]	Total solids [mg/L]	Abs UV
Initial water quality parameters of sector B										
	0.53	7	18.6	1.1	0.69	27.81	27.41	13	0.43	0.020
Final water quality characteristic of sector B										
Sector B	0.00	7	18.3	1.1	0.98	27.69	26.71	6.8	0.43	0.027

Table 3:- Water quality parameters in sector C

Sampling point	Chlorine [mg/L]	pH	Temperature [°C]	Turbidity [NTU]	TOC [mg/L]	TC [mg/L]	IC [mg/L]	BDO [mg/L]	Total solid [mg/L]	Abs UV
Initial water quality parameters of sector B										
Initial data	0.37	7	18.6	1.0	0.40	27.81	27.41	9.3	0.39	0.025
Final water quality characteristic of sector B										
Sector C	0.00	7	18.3	1.1	0.62	27.94	27.72	6.9	0.39	0.020

Results obtained in sectors B and C were aimed at understanding the effect of mixing phenomenon on the diffusion of physicochemical parameters in the network. As it can be seen water quality parameters in sectors B and C such as total organic carbon (TOC), total carbon (TC), inorganic carbon (IC), Biochemical Oxygen Demand, Total solids, absorbance UV, Turbidity were different.

As it can be seen, water quality characteristic measured in both sectors were different due to the incomplete mixing occurred in sector A. This phenomenon is perhaps the most important controlling factor influencing microbial diffusion in water distribution systems.

Conclusions:-

This study investigated water quality parameters at cross junctions that had incoming flows from different directions (90°). The study showed that incomplete mixing at cross junctions could lead to a heterogeneous diffusion of physicochemical water quality parameters. Results from this study are useful in understanding the effect of pipe configuration in distribution systems and help develop better management systems to address issues related to the variation of water quality parameters in the water distribution system.

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RESEARCH ARTICLE

ASSESSMENT OF DEMONETISATION, IT'S MATHEMATICAL ANALYSIS AND IMPACT ON THE WHOLE FINANCIAL SYSTEM, TAKING A STEP ON THE ROAD TO DIGITAL TREASURE HUNT IN INDIA.

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Transaction,Corruption,Black money.

Abstract

Taking the nation by surprise, Indian Government on 8th November,2017 announced demonetisation of Rs1000 and Rs 500 notes with effect from the midnight, making these notes invalid and the move was aimed at cracking down on the shadow economy.The immediate effect of the move brought the entire financial system to halt, hitting the large sections of the economy hard.The noble move by the government was indeed to control the counterfeit currency.This paper is based on the secondary data which was collected from various sources like published journals,articles,magazines and it elucidates the impact of such a move on the different sectors of the financial system.

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Introduction and History:-

Demonetisation means the withdrawal of the currency from circulation.The concept of demonetisation was not new.The first was when Rs 1000,Rs 5000 and Rs 10000 notes were taken out of the circulation in January 1946,a year and a half before the country won independence from the British.The Rs 10000 notes were the largest currency denomination ever printed by RBI,introduced for the first time in 1938.All three notes were reintroduced in 1954.In the early '70s,the Wanchoo Committee,a direct tax enquiry committee set up by the government,suggested demonetisation as a measure to unearth and counter the spread of Black money.However the public nature of the recommendation sparked black money hoarders to act fast and rid themselves of high denominations before the government was able to clamp down on them.Then in 1977 the Janata Party Coalition govt came into power.A year into the government's term,party leader Moraji Desai was more bullish about cracking down on counterfeits and black money.The high denomination bank notes(Demonetisation)act,instated by the ruling party on Jan 16,1978,deemed the Rs1000,Rs 5000 and Rs10000 notes illegal for the second time.The Rs 1000 note made a comeback in November 2000 and Rs 500 note was brought in to circulation in October 1987.But on November 8,2016 Rs 1000 and Rs 500 notes were demonetised with an aim to curb the black economy.The new series of Rs 500 and Rs 2000 banknotes are in circulation since 10 November,2016.

Why was the bold step taken by the Government? The government's stated objectives behind the demonetisation policy are as follows:

- First it is an attempt to make India Corruption free.
- Second it is done to curb the undisclosed money
- Third to control the escalating price rise
- Fourth to eliminate fake currency and dodgy funds which have been used by the terror groups to fund terrorism in India

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- Fifth to make people accountable for every rupee they possess and pay income tax return
- Finally it is an attempt to make a cashless society and create a Digital India

Though the Government pulled off a major coup today to check black money, it has laid its foundation much before. Though today's unprecedented financial measure may have come as a rude shock to many, Narendra Modi Government gave enough opportunities and threw enough hints in this regard. The following are the possible measures:

- In the very first Cabinet meeting, the first such initiative constituted a Supreme Court Monitored Special Investigation team (SIT) on black money.
 - The second such move was the launch of Pradhan Mantri Jan Dhan Yojana (PMJDY) on August 28, 2014. People were asked to deposit all their money in the Jan Dhan accounts and do their future transactions in banking methods only. Now that the 500 and 1000 currencies are banned and transactions from banks will acquire importance. Opening of accounts even in the remote areas will help the rural villagers. They will not feel the pinch of the banning of the currency notes. Had the bank accounts not been opened, the people would have faced immense problems. Till February 22, 2017 the total 27.77Cr accounts have been opened so far and Rs 64720.94 Cr has been deposited in these accounts. 22.22Cr accounts (Rural 12.18Cr+ Urban 10.04Cr) under Public Sector Banks, 4.65Cr accounts (Rural 4Cr+Urban 0.65Cr) under Regional Rural Banks, 0.90Cr accounts (Rural 0.54Cr+Urban 0.36Cr) under Private Banks have been opened.
 - The government renegotiated the Double Taxation Avoidance Agreement (DTAA) with Mauritius to impose capital gains tax if such capital asset is situated in India. The govt also negotiated an Automatic Information Exchange Agreement with Switzerland. Agreements are also being negotiated with other Tax Havens. From 2017, Organisation of Economic Cooperation and development (OECD) countries have agreed to share information on foreign account holders with their home countries.
 - The Undisclosed Foreign Income and assets (imposition of tax) Bill, 2015 will apply to Indian Residents and seeks to replace the income tax act 1961 for the taxation of foreign income. It penalizes the concealment of foreign income and provides for criminal liability for attempting to evade tax in relation to foreign income.
 - Income Declaration Scheme 2016 came in to effect from June 1, 2016. It's an opportunity for those who didn't disclose income or pay taxes thereon in the past. Under this scheme, such persons can declare the undisclosed income and pay tax, surcharge and penalty on the declared undisclosed income. The person making a declaration under the scheme will have to pay income tax at the rate of 30% of the value of undisclosed income plus a surcharge of 25% thereon. Additionally the person will be liable to pay a penalty at the rate of 25% of the tax, which would make the total payments 45% of the undisclosed income.
 - The govt also imposed a penalty of 20% on all cash transaction exceeding 20000 in Real estate to curb black money.
- However there were many who still hoarded the black money and in order to tackle them the govt announced the demonetisation of 500 and 1000 currency notes.

Mathematical calculation of the Process:-

Let us understand the process of demonetisation through some mathematical logic to predict the impact of demonetisation in the Indian economy.

Indian GDP is supposed to be 125-130 lakh crore

Cash circulation before demonetisation was 17.54 lakh crore = approximately 14% of cash to GDP ratio

Size of the black economy = 20% of GDP

= 20% of 125-130 Lakh Crore (approx)

= 25-28 lakh crore (approx)

Black cash in the system = 4.5 Lakh crore

Some prior studies kept the black cash = 3.35 lakh crore (instead of 4.5 lakh crore)

Total cash circulation in the system = 17.5 lakh crore

Total cash in 500 and 1000 Rs denomination = 86% of 17.5 Lakh crore (approx)
= 15 lakh crore

Black cash is worth = 4.5 lakh crore

Legitimate cash = (17.5 - 4.5) lakh crore
= 13 lakh crore

Assuming a figure of 5% of the cash to GDP ratio in line with other similar economies, it would like to see no more than 9 lakh crore cash in circulation of which around 2.5 lakh crore of cash was in small denominations in the system already before the exercise started. This essentially means that (9 - 2.5) = 6.5 lakh crore of cash is what the

Govt will replace in lieu of the Rs 15 lakh crore (in 500 and 1000 denominations) it will take out. Now if the logic stated above is correct we will see that Rs 15 lakh crore worth of high denomination notes will be replaced by 6.5 lakh crore of cash. But we cannot expect that all replacement will be in similar high denomination notes. It may be noted that, even before the new high denominations(500 and 2000) were introduced around 50000 crore cash of new cash was injected in small denomination notes. Now say the total new high denomination notes in cash is 4 lakh crore. Therefore apart from the 4.5 lakh of black money being burnt, we can expect that 6.5 lakh of high denomination notes(old 500+1000) notes will never come back in to the system.

Whether the process will work or will fail like 1978,1946? After the failure of First and second demonetisation that took place in 1946,1978 it is a question that now our third demonetisation will be a success or not. The major difference is in the quantum however. The first and second demonetisation affected really high value notes which formed a small part of the notes in circulation. If we consider per capita GDP of India now and earlier then we can arrive at a conclusion. In 1978 per capita GDP of India was approximately Rs 1700 whereas today it is approximately Rs 1 lakh. At that time it was really a great thing when people earned four digit salaries. The common middle class people did not even use much of higher denomination notes. Also circulation of fake notes were also very less. Probably a very large % of the population did not even know the move as there was no effective social media and even televisions were only found in few houses. No of Banks and branches were also very less compared to the present scenario. Black money at that time was very less as of now. So only about 15% of the currency got exchanged. Above reasons are the major reasons for failure in 1946 and 1978.

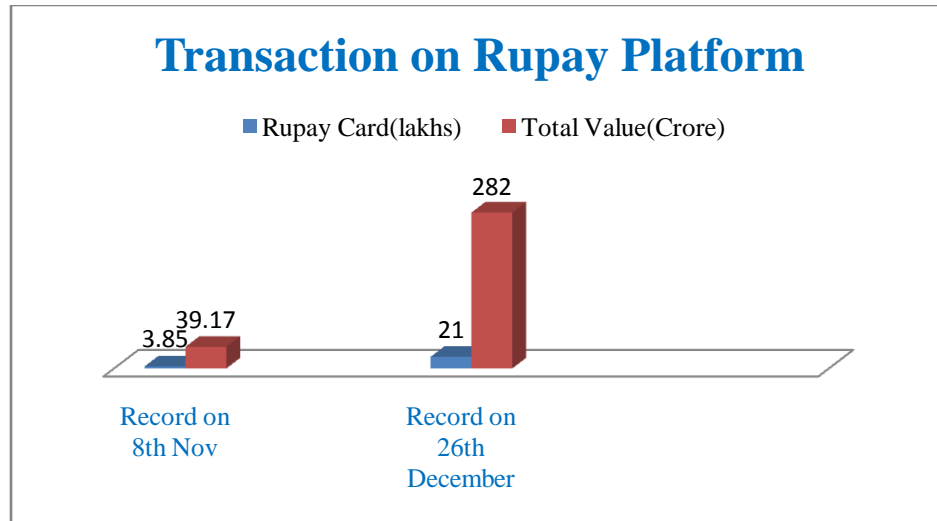
But now the situation is a lot different. The black money has increased drastically with no leaps and bounds. It was held mainly in form of Jewellery, Currency notes in denomination of 500 and 1000 which accounts for 85% of total currency and in real estate market etc and in the recent times the circulation of fake notes has also increased rapidly. Also IDS ,2016 was declared prior to the demonetisation which was a strong move against the black money. Most important thing is that now cashless move is adopted and so illegal way of transmitting money will be reduced. The govt is just in the beginning of effort against black money but it will surely be a success in the long run.

Impact of Demonetisation : Demonetisation effected various sectors of economy. Few are discussed as

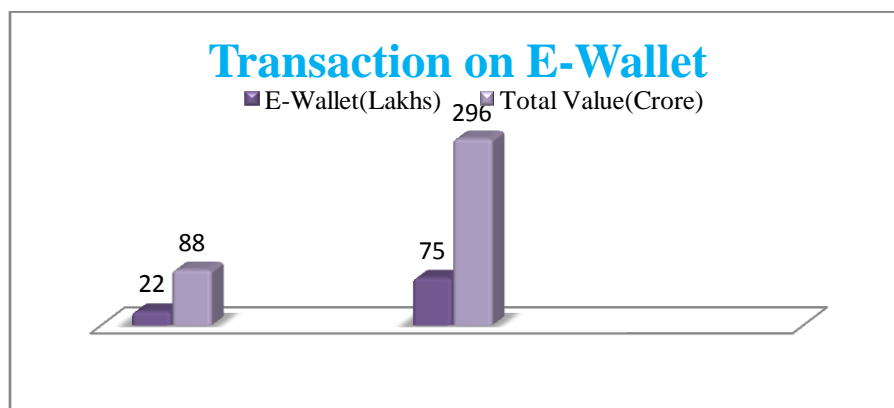
- **Effect on Agriculture:** The move hit badly the farmers as people in cities could queue-up to exchange or deposit their old notes at banks but people staying at villages, miles away from cities are not in a position to carry out exchange or deposit properly. The entire agricultural system runs on cash and the poor farmer might become the worst casualty in this drive. Not only are farmers likely to find it hard to pay off their debts, they may not get any further credit for processing seeds and other inputs for the next sowing season. The situation is exacerbated by the fact that poor sections are reportedly being used as middle man to exchange old notes for new by the black money hoarders.
- **Effect on Real Estate:** The real estate sector, especially the secondary market, has virtually come to a standstill. Black money has been to the Indian real estate sector, what foreign institutional investors have been to the stock markets. Much of the boom of the Real Estate sector has been due to the proliferation of the black money, which accounts for anywhere from 30% to 40% of the secondary market. With black money being sucked out of the market, real estate prices in bigger cities should see a correction of 30%.
- **Effect on Gold and other precious metals:** After demonetisation the demand for gold and other precious metals has increased greatly because people with black money trying to invest in gold to make it white. But the govt has said that it is aware of this and is cracking down with tax raids on jewellers across the country. Moreover jewellers will be required to produce details of any cash transactions. In the long run though with cash being sucked out of the system, sales will take a hit, as most jewellery purchases are done using cash.
- **Effect on anti social funding:** Money laundering is one of the important financial mechanism for terrorists. The antimoney laundering investigation fails when the money trail leads to cash. In India the terror finance trail starts and ends with cash making it impossible to get early alerts of terrorist active in the country. Demonetisation will upset the financing chain for the terrorists as all the money will get back to the banks and from there it is easy to identify the fake currency.
- **Effect on Political funding and donations:** Demonetisation has badly affected the political parties. After the note ban, the govt also put some restrictions regarding the party fund and donations. Anonymous donations are going to get identified now and upcoming 2017 elections will be transparent to some extent.

India's Way to the Cashless: The demonetisation drive has forced millions of Indians to fall in line with Narendra Modi Govt's Cashless move. The immediate impact of demonetisation is seen across the country where even small

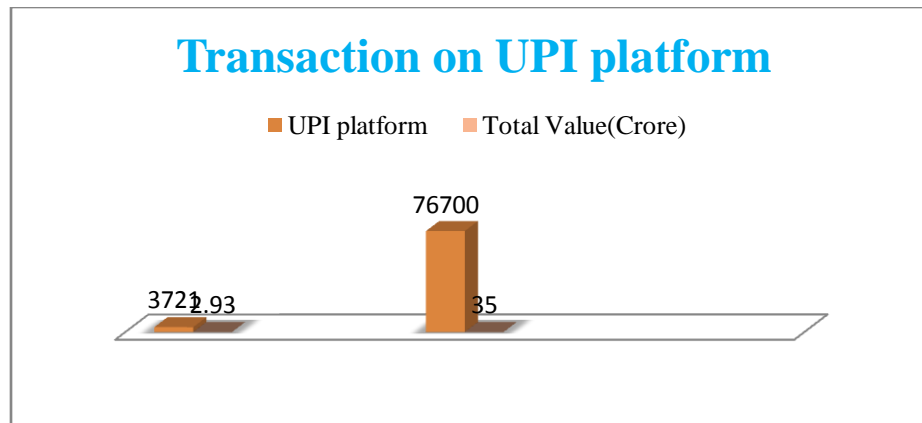
traders took to digital modes of transactions including E-wallets ,POS terminals etc.The consumer class also found prices of commodities like pulses,edible oil,fruits and vegetables coming down in retail market.Perhaps the biggest beneficiaries are Paytm,Freecharge and Mobikwik. Digital payments have increased in the range of 400 to 1000 percent since November 8th.Transactions on Rupay cards have increased from 3.85 lakh per day to 21 lakh per day and the value of transactions have increased from 39.17crore to 282 crore.Net percentage change is 445% in terms of subscribers and 620% in terms of transactions.



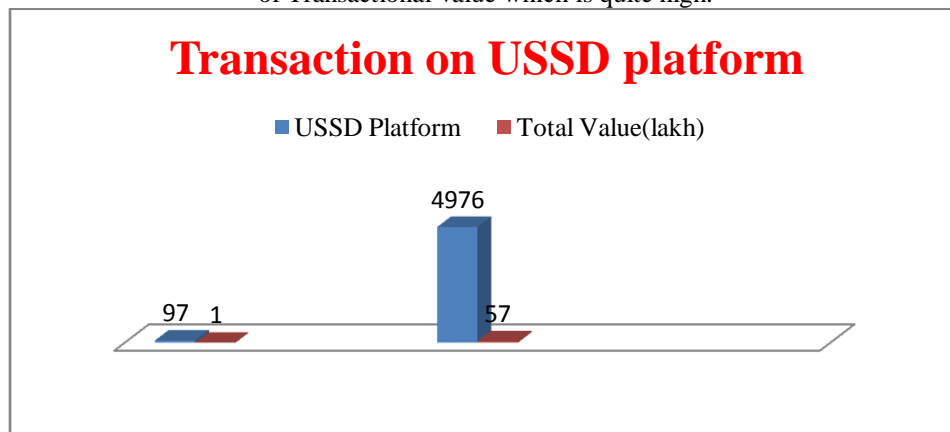
E-wallets have increased from 22 lakhs per day to 75 lakh per day and the value of transactions have increased from 88 crore to 296 crore.The net percentage change is 240% in terms of subscribers and 236% in terms of Transactional value.



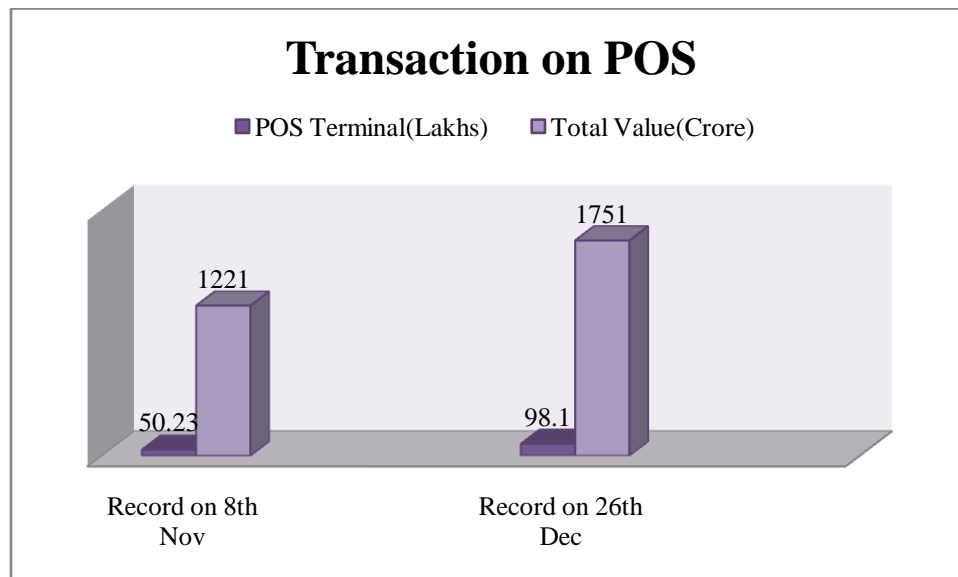
Transactions on UPI have increased from 3721 per day to 76700 per day and the value of transactions have increased from 2.93 crore to 35 crore.The net percentage change is 1961 % in terms of subscribers and 1095 % in terms of Transactional value.



Transactions on USSD platform have increased from 97 per day to 4976 per day and the value of transactions have increased from 1 lakh to 57 lakh. The net percentage change is 5000% in terms of subscribers and 1095 % in terms of Transactional value which is quite high.



Transactions on POS Terminals have increased from 50.23 lakhs per day to 98.1 lakhs per day and the value of transactions have increased from 1221 crore to 1751 crore. The net percentage change is 95% in terms of subscribers and 43% in terms of Transactional value.



Detailed Analysis of Digital Transaction:-

Demonetisation Impact on Cashless Transaction						
Mode of Transaction	Record on 8th Nov		Record on 26th December		Total % Change	
Rupay Card(lakhs)	3.85		21		445.4545	
Total Value(Crore)	39.17		282		619.9387	
E-Wallet(Lakhs)	22		75		240.9091	
Total Value(Crore)	88		296		236.3636	
UPI platform	3721		76700		1961.274	
Total Value(Crore)	2.93		35		1094.539	
USSD Platform	97		4976		5029.897	
Total Value(lakh)	1		57		5600	
POS Terminal(Lakhs)	50.23		98.1		95.30161	
Total Value(Crore)	1221		1751		43.40704	

Conclusion:-

It can not be told now clearly that whether the ban on 500 and 1000 notes will completely eradicate the black money or not but it is just one move of one piece in the chess board of black money. To checkmate the black money king, have to win the board. The effort of the govt was supported by majority of common people. Although the process did not prove that much success but a good sign is the move to the Cashless. Since demonetisation was carried out to curb the black money and soon after the move to digital transaction will surely portray some good result. Since with digital transactions money will come into the system, it will be accountable, generate tax and help develop the country's economy. When money will come in to the banks the govt will be able to come up with better welfare schemes. The govt will significantly benefit from the additional cash that is pushed in to the system. There will also be a much higher collection of income taxes and other taxes by the govt. More than short term benefits, there is an angle of long term GDP growth which will positively impact our economy by around 2018. To sum up a little patience will go a long way in making this demonetisation growth a success. The inconveniences or difficulties of time consuming, currency exchange and reduced withdrawal limits at ATMs and Banks will gradually pave the way for a more resilient economy. The entire process in turn will not only benefit us individually but as a nation proceeding towards a glorious future.

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RESEARCH ARTICLE

DRAFTING AND REVISING STRATEGIES TO DEVELOP ENGLISH ESSAY BY INDONESIAN EFL LEARNERS.

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drafting, revising, writing process, proofreading.

Abstract

This qualitative study reports features of drafting, revising and strategies to write an English essay. The study was done in State Islamic Institute (IAIN) Surakarta, Indonesia on January to April 2015, assigning 30 students as the research subject. The study revealed that in drafting and revising process in writing were differently produced by lower, middle and upper groups. Lower group did not write outline and produced first draft without planning. Thesis statement, topic sentence and concluding paragraph did not appear. The middle group was able to produce thesis statement, body paragraphs and conclusion, but they focused less on revising and proofreading. The upper group exhibited by performing all aspects of an essay in each rhetorical patterns. Drafting and revising were used appropriately. All groups asserted literal translation to develop ideas and overcome problems on diction.

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Introduction:-

Good writing is essential for success in school and the 21st Century workplace. Writing is a complex combination of skills which is best taught by breaking down the process. The writing process involves a series of steps to follow in producing a finished piece of writing. Focusing on the process of writing, almost everyone learns to write successfully. By breaking down writing step-by-step, the mystery is removed and writer's block is reduced. Most importantly, students discover the benefits of constructive feedback on their writing, and progressively master, and even enjoy, writing (Time4 Writing, 2016).

The writing process—prewriting, drafting, revising and editing, rewriting, publishing—mirrors the way proficient writers write. Studies show that students who learn the writing process score better on writing tests than those who receive only specific instruction in the skills assessed on the test. Success in writing greatly depends on a student's attitude, motivation, and engagement. The writing process takes these elements into account by allowing students to plan their writing and create a publishable, final draft of their work of which they can be proud (ILA/NCTA, 2017:1).

When teaching expository essay in Indonesian contexts, empirical evidences appear pertaining to quality of rhetorical patterns. There is still a debate if process of writing is emphasized, attentions in the language revision will decline and improvements on grammar are limited. Conversely, if language corrections take priority, writing process is delayed.

For many students, writing can be intimidating, upsetting and mystifying but writing process answers this problem. Every writer follows his or her own writing process (Pouladian, et. al. 2017). Therefore, process writing has been

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considered as writing technique that suffices benefits in the teaching process. According to Krashen (1984:21) convention as a model is required in process of writing. To maintain how to write for newspaper, read newspapers; textbooks about them will not suffice. For magazines, browse through magazines rather than through correspondence courses on magazine writing. To write poetry, read it. For the conventional style of memoranda in a school, consult school file.

Research Questions:-

- 1) How is the feature of revising and drafting the English essay by Indonesian EFL learners?
- 2) How are elements of introductory paragraph, body paragraphs and concluding paragraph in an essay by Indonesian EFL learners?
- 3) What strategies are used by Indonesian EFL learners to develop an essay?

Review of Literature:-

Process Writing:-

Historically, educators have struggled how to write well, focusing on the finished product. Since the 1970s, teachers no longer emphasize the finished product; instead, they teach the writing process. Researchers discovered that writing was a highly complex process, made up of various sub-processes that had a strict linear sequence and varying patterns (Caudery, 1995). Good writers differ from poor writers in their composing process, that is, they have better and more sound procedures for getting their ideas down on paper. Specifically, good writers differ in three ways: in planning, rescanning, and revising (Krashen, 1984:12).

Writing is recursive and any of the steps can be repeated. The term *recursive* refers that writers can engage in any act of composing--finding ideas, thinking about ways of organizing them, imagining ways of expressing them--at any time during writing and often perform these acts many times while writing (Wingersky, Noerner, & Holguin-Balogh, 1992; Langan, 1992). In addition, Krashen (1984:17) asserts writers employ recursive: writing a draft may be interrupted by more planning, and revision may lead to reformation, with a great deal of recycling to earlier stages. According to Caudery (1995) students can start the process from brainstorming, prewriting outline or collection of ideas in their head. Students also create a topical outline to help organize their ideas, and to develop a working thesis statement.

Weaknesses and benefits of writing process exist, however. Because the emphasis is on the process of writing, much of the sensitivity about receiving constructive criticism is eliminated. The strength is its usefulness for a wide range of diverse learners. Students are taught a variety of styles to structure their thinking, ranging from analytical outlines to highly visual graphic organizers. Students explore ways for organizing and visualizing their ideas that is the most effective for them.

Steps of Process Writing:-

Simply, process writing consists of five steps to write a piece of writing product. Often the process is a routine that comes naturally and is not a step-by-step guide to writer. Here are five steps of the writing process.

Pre-writing:-

Prewriting is a planning phase in writing process. Here students brainstorm, research, gather and outline ideas in terms of diagrams for mapping out their thoughts. Students also address audience and purpose, and formulate a thesis statement for the writing. Prewriting includes: (1) Deciding on a purpose, or reason, for writing. (2) Thinking about the audience's needs and interests. (3) Choosing a subject for writing. (4) Limiting the subject to a suitable topic. (5) Gathering information on the topic. (6) Organizing the information (Langan, 1996).

Drafting:-

The second process of writing is composing a first draft or rough draft. A *draft* is the rough form of a piece of writing. Definitely, drafting is writing the first version of ideas on a paper without thinking right or wrong the ideas are. Writing the first draft is expressing ideas in sentences and paragraphs. Drafting involves taking information the writer has generated and organized in an essay. The writer consciously starts with main ideas and adds supporting ideas that flow smoothly (Wingersky, Noerner, & Holguin-Balogh, 1992). Students create initial composition by writing down all their ideas in an organized way to convey a particular idea or present an argument. Audience and purpose are finalized later.

Revising:-

Revising means to see again. Revising is a big change like wording, adding sentences, and moving paragraphs. In writing, revising is making changes to improve the first draft by moving around words, phrases, and ideas. Writers review, modify, and reorganize their work by rearranging, adding, or deleting content, and by making the tone, style, and content appropriate for the intended audience (Warriner, 1988; Langan, 1998; Nordquist, 2015). When revising, a writer is changing the meaning or way in which the reader perceives, experiences and interprets it.

Prior to revising, a writer does an evaluation on the paper, that is, re-examining ideas, organization, and word choice in the first draft (Langan, 1996). A space for improvement appears in the first draft and to improve this, the writer evaluates or makes judgements on strengths and weakness of the draft to see intended purpose of the paper, organization, clarity, and appropriateness to its audience (ILA-NCTE, 2017). See Table 1 for the guideline of evaluating a paper.

In addition, revision focuses on the whole paper. It may change several points: point of view, tone of paper, order of paragraphs, eliminating one of the paragraphs completely, adding a whole new idea, and changing conclusion (Nordquist, 2015). Revising naturally adopts the ARRR approach:

Add: number of words or details required, e.g. 350 or 3,500 words

Rearrange: organization or contents of the paper appropriately

Remove: making changes, organization, rearrange ideas, eliminate passages

Replace: enough details, any contradicted facts

Table 1:- Guidelines for Evaluating Writing (Warriner, 1988:32-33)

Contents		
1	Purpose	1. Do all the ideas help to achieve a main purpose (explain, tell a story, describe, or persuade)?
2	Topic	2. Is the topic suitably limited for the form of writing?
3	Audience	3. Are the topic and details suitable for the audience?
4	Development	4. Is enough information given to understand the topic? Is too much given—does it all “belong?”
Organization		
1	Coherence	1. Does the order of ideas fit the purpose? Will this order make sense to the reader?
2	Transition	2. Are ideas clearly connected, with words such as <i>also</i> , <i>first</i> , <i>these</i> , <i>however</i> , and <i>therefore</i> ?
Style		
	Words Choice	1. Are exact and vivid words used?
		2. Does the vocabulary fit the audience and purpose?
	Sentence Variety	3. Do the sentences vary in length and how they begin?

Table 2 is presented to show how revising activities and its focus should work in a small scope.

Table 2:- Revising focus on a paper (Adapted from Monica (1996), Student Learning Center, University of California, Berkeley).

No	Focus of Revision
1	A paper as a whole: strengths, weaknesses, arguments, focus, organization, support, voice, and mechanical issues
2	Dialogue-based, the purpose or revision is to ask questions, expanding ideas and challenging arguments which require discussion between the writer and the reader
3	Non-hierarchical, offering questions and making observations, allow the writer and reader to hold separate and valid opinions. The purpose is to expand and clarify ideas rather than correct them
4	focuses on the writer in the process of writing and increasing the writer's understanding of the paper's strengths and weaknesses
5	Clarifies and focuses the writer's arguments by defining terms, making concessions and counter-arguments, and using evidence; moving or removing entire paragraphs, extending or narrowing ideas, rewriting vague or confusing text, and adding to existing paragraphs

More complete version of revising activities, Table 3 is presented as a guidance of revising for a paper in a broader context.

Table 3:- Revising a Draft (adopted from Warriner, 1988: 35-36)

No	Problem	Techniques	Revision
1	Purpose is not clear	Add/Cut	Make sure you know your purpose. Add details that fit it. Remove comments that do not fit it.
2	The ideas are too general	Replace	Make general ideas more specific. Do more research, if needed. Be sure the topic is not too broad for the form of writing.
3	Some information is "extra" or not related to the main idea.	Cut	Remove "padding" and unrelated ideas. Keep only the background needed by the particular audience.
4	It is hard to follow the ideas.	Reorder/Add	Move sentences or paragraphs to fit what a reader needs to know first, second, and so on. Add linking words such as <i>this</i> , <i>when</i> , <i>then</i> , <i>these</i> .
5	Some terms are too complex or too simple for the audience.	Add/Replace	Add definitions to explained unfamiliar terms. Change the wording to fit the audience's age and background.
6	The beginning is dull	Add	Add examples, incidents, or other details that will interest the audiences. Be sure the topic will interest the audience.
7	Some words are vague or overused.	Replace	Replace words that stand for a group of things with more exact words. Use exact, vivid adjectives, adverbs, and verbs.
8	Some wording seems very informal.	Cut/Replace	When light, informal expressions do not fit the purpose and audience, remove slang, contractions, and so on or replace them with standard formal words.
9	The sentences are monotonous.	Replace/Reorder	Combine choppy sentences. Split rambling sentences. Change the word order so sentences begin in different ways.

Editing:-

Editing begins as soon as a writer finished the first draft. Editing is rereading draft to see whether paper is well-organized, transitions between paragraphs are smooth, and evidences really fit the argument (Langan, 1996). Editing takes place in two ways: *First*, editing focuses on errors on punctuation, grammar, and spelling. In this stage, editing a manuscript focuses only on the structure of writing that is to be corrected. These are minor changes that include: spelling, grammar, punctuation, sentence structure. The overall text remains exactly the same because an editor simply corrects mistakes in the paper, such as typos, grammar errors, and spelling mistakes (Time4 Writing, 2016). *Second*, editing undergoes with linkages of developments and connections of ideas in the whole paper. This way, level of editing includes: contents (claim, argument, evidence, relevant goal), overall structure (thesis statement, introduction, body, conclusion, logical sequence, transitions between paragraphs), and structure within paragraph (clarity, style, citation) (The Writing Center at University of North Caroline. 2014). Table 4, shows activities how an editing a paper should work.

Table 4:- Focus of Editing (Adapted from Monica (1996), Student Learning Center, University of California, Berkeley).

No	Focus of Editing
1	A sentence level, addressing problems with spelling, grammar, punctuation, or word choice
2	One-sided: The editor writes comments and corrections on the paper and returns the paper to the writer
3	Hierarchical: An editor looks for "mistakes" and "fixes" them. An editor places value on writing (such as a grade)
4	Focuses on the paper as a product

After editing, the editor's job is proofreading. Errors in spelling, grammar, usage, or mechanics can distract the reader from the ideas in the paper. At this point, writers proofread and correct errors in grammar and mechanics, and improve style and clarity. It's time to fine tune the manuscript line by line by checking for repetition, clarity, grammar, spelling and punctuation. Proofreading consists of checking for errors in grammar, usage, mechanics, and correcting errors. The result of conscientious proofreading is a final draft—the finished product (Time4 Writing, 2015; Nordquist, 2015).

Proofreading is different from revision. Proofreading is the final stage of the editing process, focusing on surface errors such as misspellings and mistakes in grammar and punctuation. Proofreading is done only after all editing revisions are finished. *Proofreading* is finding and correcting mistakes in grammar, usage, and mechanics. *Usage* means using words correctly according to the rules for standard written English. *Mechanics* includes capitalization, punctuation, and spelling (Langan, 1996). See Table 5 for the guidelines of proofreading.

Table 5:- Guidelines for Proofreading (adopted from Warriner, 1988: 39)

No	Focus to proofread
1	Do plural verbs have plural subjects? Do singular verbs have singular subjects?
2	Are verbs in the right form and tense?
3	Are the verbs <i>sit/set</i> , <i>rise/raise</i> , and <i>lie/lay</i> used correctly?
4	Are subject and object forms of pronouns used correctly?
5	Are adjective and adverb forms used correctly in comparison?
6	Are double negatives avoided?
7	Do all sentences and proper nouns and adjectives begin with a capital letter?
8	Are all words spelled correctly? Have spellings been checked in a dictionary?
9	Is the punctuation correct at the ends of sentences and within sentences?
10	Is every sentence a complete sentence?

Publishing:-

Publishing is the last step of the writing process. Publishing is making the final copy of the paper into a correct form. Publishing for English teachers and students means the completion of the cyclical writing process for a specific product. The writer prepares a document to be distributed to an audience. This may involve printing the work or duplicating it in print or online.

Writing Process in This Study:-

Overall writing process is presented in Figure 1. Parts of this process cannot operate at the same time, even if the writer is skilled and the writing task is easy. Altogether they represent what is called the writing process that is recursive, which means going back and forth. Writing process in this study includes three steps and two activities of writing: drafting and revising.

Drafting

- a. Stage One : Exploring-Experimenting-Gathering information
- b. Stage Two : Writing the controlling idea/Organizing and developing support

Revising

- c. Stage Three : Writing-Revising-Editing.

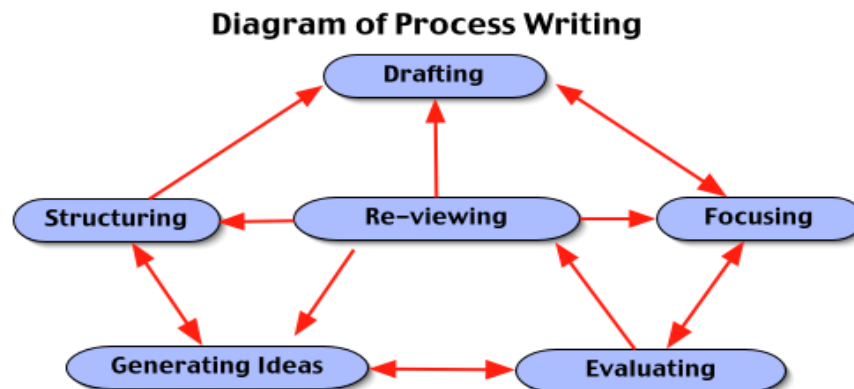


Figure 1:- The Process of Writing (from: *Process Writing* by Ron White and Valerie Arndt, 1991.)

English Essay:-

An essay is a piece of writing several paragraphs long. An essay is usually divided into several paragraphs, tying together an introduction, body and a conclusion (Oshima and Hogue, 2006). An essay is a short piece of non-fiction writing that often requires a number of skills such as close reading, analysis, comparison and contrast, persuasion, conciseness, clarity, and exposition (Langan, 2009). The purpose is to develop ideas and concepts more formal, concise, clarity in purpose and direction.

Standard short essay is between 350 and 500 words long; usually has one paragraph of introduction, a three-paragraph body, and one paragraph of conclusion. Some essays have two to three paragraphs, others have twenty or thirty. The essays written in class contain from four to six paragraphs, with the most common number being five: one introductory paragraph, three developmental paragraphs, and one concluding paragraph (Smalley and Ruetten, 1982:139). A-500 essay is equal to one page A4 paper for single space. A long essay such as paper or journal article may consist of 3,500 to 6,000 words long or 8-10 pages typed in a single space format. Organization of the paper normally includes: title, author name, abstract, introduction, review of literature, methodology, findings, discussion, conclusion and reference (Solikhah, 2017).

The introduction, the first section of an essay, is usually one paragraph that introduces topic to be discussed and central idea (the thesis statement) of the essay. The purpose is to arouse interest and introduce the controlling idea of an essay and states the main idea and thesis statement. The thesis statement is called the controlling idea that brings an essay into focus, giving it direction and drawing its ideas together (Kirsznier and Mandell, 1978:5).

The body paragraphs are the longest section of an essay. In a short essay, there are usually three body paragraphs, each one considering in detail one aspect of the essay's controlling idea. This is called a three-point essay. These paragraphs develop various aspects of the topic and the central idea. They may discuss causes, effects, reasons, examples, process, classification, or points of comparison and contrast. They may also describe or narrate (Smalley and Ruetten, 1982:139).

The concluding paragraph is the end of an essay that concludes the thought developed in the essay as the closing word. The ideas in the conclusion must be consistent with the rest of the essay. In the conclusion, the writer should draw together all that has come before by restating the controlling idea of the essay. This statement is usually most effective when it is located at the beginning of the conclusion (Kirsznier and Mandell, 1978:7).

Methodology:-

Research Design:-

This study used descriptive design with qualitative approach aiming to explain writing process on the accomplishment of expository essay. Writing process in this study was restricted into two kinds: drafting and revising. Descriptively, the study delineated drafting and revising in terms of frequency and rate percentage; in addition, elaboration of the rate percentage was done through explanation on the statements of writing comments. This study took place in English department at State Islamic Institute (IAIN) at Surakarta, Indonesia and conducted from January to April 2015.

Research Subject:-

Subjects of this study were senior 30 students learning in the sixth semester of English Language Education program of IAIN Surakarta joining in Writing Class. As this research ended, there were 5 students absent so the final number of participants were 25 students, 17 female and 8 male. Subjects were all at about to have similar competence in writing as all have received courses on Paragraph Writing, Essay Writing and Academic Writing at respective previous semester prior to this research was conducted. To this end, their knowledge on writing convention, academic writing, process of writing, and strategies of writing was equal among students. Subjects were all aware that they were incurred in the research class of writing the researcher conducted and they received treatments on the same materials.

Research Instruments:-

Instruments of this study were Writing Prompt on Expository Essay, Checklist to Assess Expository Writing, and Interview Guide. Writing prompt was used to see writing students worked. The expository test asked students to write an English essay of 5-10 paragraph with 350 to 750 words length. The mode of the essay was expository that explained a topic the researcher determined in the writing prompt. The essay checklist was used to evaluate the availability of the components of the essay After each essay was evaluated by raters and scores had been provided in each essay, the researcher classified the essay into three categories: low, middle and high. Based on the quality, the researcher interviewed to the subjects to see the strategies, process, and the works during preparations on drafting and revising process in developing the essay.

Data and Sources of Data:-

Sources of data in this study was Expository Essay obtained from the test of writing done by the research subjects. The essays were assessed and scored its quality. Therefore, data of the study comprised of quality of the introductory paragraph, the body paragraphs, and the concluding paragraph. Data on elements to support good introductory paragraph (e.g. thesis statement), the body (e.g. transition words, details, coherence) and the concluding paragraph (e.g. restatement of topic) were also identified.

Data Analysis Techniques:-

Data of this study consisted of two main concerns: frequency account and narrative accounts. First of all, the essays were scored and were identified their rhetorical patterns. The presence of account on each rhetorical patterns and its components were identified. Frequency data were used for two main concerns of analysis; First, it indicated frequency used by low, middle, and upper students during drafting and revising process to include in the rhetorical patterns; *Second*, it showed an account of how each group of the subject used elements of rhetorical patterns to build good introductory paragraph, the body and concluding paragraph.

In addition, exploration on comments and opinions of the subjects obtained through interview was used to collect narrative data. The interview described subjects' testimony on how they worked during the process of writing. Data of this view were categorized into strategies, problems, challenge and attitude during the writing expository essay in this study.

Results:-**Drafting and Revising the Essay:-**

Table 5 summarizes frequency of account on drafting and revising to develop an essay. Analysis was focused on frequency of correct form in drafting and revising to develop rhetorical patterns of an essay.

Table 5:- Drafting & Revising to Develop Rhetorical Patterns

No	Process	Rhetorical patterns	Lower (5)		Middle (14)		Upper (6)	
			F	%	F	%	F	%
1	Drafting	1. Introductory paragraph	1	20	7	50	6	100
		2. Body paragraph	3	60	12	86	4	67
		3. Concluding paragraph	1	20	13	93	5	83
2	Revising	1. Introductory paragraph	2	40	12	86	6	100
		2. Body paragraph	4	80	10	71	5	83
		3. Concluding paragraph	3	60	12	86	5	83

As table 5 shows attainment of lower, middle and upper groups in developing rhetorical patterns increased in the frequency of subjects. Lower group showed low empowerment in drafting but improved in revising. Similarly, middle and upper groups also improved their accomplishment in developing essay in the revising process.

Characteristics of rhetorical patterns in drafting and revising process of lower, middle and upper groups are as follows:

1. Lower group
 - a. Drafting process: low introductory paragraph (20%), fair body paragraphs (60%) and low concluding paragraph (20%).
 - b. Revising process: low introductory paragraph (40%), very good body paragraph (80%), and fair concluding paragraph (60%).
2. Middle group
 - a. Drafting process: low introductory paragraph (50%), very good body paragraphs (86%) and very good concluding paragraph (93%).
 - b. Revising process: very good introductory paragraph (86%), good body paragraph (71%), and very good concluding paragraph (86%).
3. Upper group
 - a. Drafting process: excellent introductory paragraph (100%), fair body paragraphs (67%) and very good concluding paragraph (20%).
 - b. Revising process: excellent introductory paragraph (100%), very good body paragraph (80%), and very good concluding paragraph (83%).

Rhetorical Patterns of the Essay:-

Elaboration of rhetorical patterns was done in this section. Components of introductory paragraph, the body and concluding paragraph were rated. The findings on Table 6 showed that lower group achieved lower attainment to induce rhetorical components to make quality of the introduction better. In addition, middle and upper groups exhibited from the lower group. Upper group even 100% of the subject succeeded to develop parts of one of the rhetorical patterns.

Table 6:- Description on Rhetorical Patterns

No	Rhetorical elements	Lower (5)		Middle (14)		Upper (6)	
		F	%	F	%	F	%
1	Introductory paragraph						
	1. Introductory sentence	3	60	10	71	6	100
	2. Thesis statement	2	40	8	57	5	83
	3. Topic sentence	1	20	8	57	5	83
	4. Controlling ideas	-	-	8	57	5	83
2	Body paragraph						
	1. Transition words within and between paragraphs	3	60	10	71	6	100
	2. Has a topic sentence	1	20	9	64	6	100
	3. Using appropriate details or synthesis	1	20	7	50	4	67
	4. Only one main idea in each paragraph	-	-	4	57	5	83
	5. Supporting sentences directly relate to main idea	-	-	3	21	5	83
3	Concluding paragraph						
	1. Restate the controlling idea	1	20	9	64	6	100
	2. Have a final closing statement	-	-	7	50	5	83

Characteristics of rhetorical patterns in the the essay made by the the lower, middle, and upper groups are as follows:

1. Lower group
 - a. Introductory paragraph: introductory sentence is fair (60%) but thesis statement and controlling idea are low (20%)
 - b. The body paragraphs: transition words is fair (60%), topic sentence and details are low (20%). Other components did not appear.

- c. Concluding paragraph: restatement is low (20%) and closing statement is absence.
- d. Middle group
- e. Introductory paragraph: introductory sentence is fair (60%) but thesis statement and controlling idea are low (20%)
- f. The body paragraphs: transition words is fair (60%), topic sentence and details are low (20%). Other components did not appear.
- g. Concluding paragraph: restatement is low (20%) and closing statement is absence.
- h. Upper group
- i. Introductory paragraph: introductory sentence is excellent (100%), thesis statement, topic sentence, and controlling ideas are good (83% each).
- j. The body paragraphs: transition words and topic sentence are excellent (100% each), detail is fair (67%), one main idea in one paragraph and supporting sentences relate to main idea are good (83% each).
- k. Concluding paragraph: restatement is excellent (100%) and closing statement is good (83%).

Writing Strategies:-

Results on writing strategies were obtained from structured interview. The focus of interview was to explore students' problems in writing an essay and in developing parts of the essay, e.g. introductory paragraph, the body and concluding paragraph. Strategies to overcome the problems were reported on the basis of level of attainment of the groups: lower, middle, and upper. The results of analysis were presented into drafting process that included: generating ideas, outlining, and writing the first draft; and revising that consisted of editing and proofreading.

Interviews with research subjects showed that students' problems in essay writing included: convention, rhetorical patterns, word-for-word translation, lack of content knowledge, grammatical errors, cohesion errors, lack of proofreading, and lack of academic style. Word-for-word translation is the most common problem to any students. A large number of the students literally translated Bahasa Indonesia into English when writing the essays.

The following are writing problems and solutions the students undergo that is ranked based on the results of interview.

Problems to write:-

1. Arranging ideas and structuring in plan to write
2. Outlining
3. Writing the first draft
4. Limited vocabulary
5. Lack of model
6. Grammar and sentence construction
7. Thesis statement in the introductory paragraph
8. Confused between thesis statement and topic sentence in a paragraph
9. Developing details in the body paragraphs
10. Citation or quotation
11. Making conclusion in a sentence and in a paragraph
12. Revising, editing and proofreading

Strategies to overcome:-

1. Translating
2. Consulting with bilingual dictionary
3. Referring to essay model
4. Browsing from internet and adopting the model
5. No outline, no revision, no editing, no proofreading
6. Stating outline, revising, editing and proofreading individually and with peer
7. Revisiting to achieve final copy for publishing or submission
8. Consulting with references online or in prints
9. Revising drafts several times to improve organization, parts of essay and substance
10. Controlling appropriateness among introductory paragraph, body paragraph, concluding

paragraph; thesis statement, topic sentence in the body paragraph, developing details, transition words and conclusion.

During drafting process, some students worked with outline and generated ideas through discussion or reading sources. Some worked in mind and some wrote on paper. There were different strategies used by lower, middle and higher students to develop the essay and its specific parts: the introductory paragraph, body paragraphs, and concluding paragraph.

Lower Group:-

Planning in this study means the first process students used to generating ideas, outlining and drafting. The results show that lower students caught ideas by recalling any experiences and did not write an outline on paper. Writing an outline was very difficult and time consuming for the lower students.

Excerpt (1) testifies how lower group planned writing and wrote an outline in the drafting an essay.

I did not write an outline before writing the essay. I tracked ideas from mind and wrote whatever I remember and knew on paper. I don't think about appropriate topic but the most important is I write down the ideas on paper. (L-1-DRAF)

Students of lower group also reported that planning was annoyance and took too formal and long time. Some focused too much on the outline and stared at the blank format of the outline. To write the first draft of the essay based on the outline was other difficult matters for lower students. *First*, stating thesis statement in the outline was complicated. *Second*, rhetorical patterns of an essay that consists of introductory paragraph, body paragraphs and concluding paragraph became obstacles to express ideas on the paper as seen on quote (2).

I write an outline but I cannot use as guideline of ideas on my paper. I just dropped my thought on paper, read at glance and submitted to the lecturer. I did not reread the text nor revised the contents. If I found any grammar or sentence errors, I corrected on the items that were wrong, not on the sentence or on the context. (L-2-REV).

More problems for lower group pertain to revising, editing, and proofreading. For them, revising is hard to do and they are not fully aware that revising gives benefits to improve the quality of an essay. Errors on grammars, sentences, organization, and structure of the essay are ignored by the low group.

Middle Group:-

Middle group exhibited from lower group in terms of drafting and revising strategies. *First*, drafting process was considered important for the students and they planned through outline. Thesis statement of the essay was developed and links to support the thesis in each body paragraph were incorporated. *Second*, in revising process, students made editing and proofreading with peers or by themselves. Though focuses of revision were limited, middle group students proceeded the steps of revision as the writing process suggested. Excerpt (3) testifies how middle group student maintains his thought on planning.

I am fully aware that drafting is very initial writing. Many errors in grammar, sentences, and ideas are found. Sometimes, it is very difficult to write correct sentences in drafting and I have to stop so long time to find the best words or sentence. It is bored. But, I have to do with the outline and follow ideas that I have planned. (M-3-DRF).

In addition to revising process, middle group students claimed that they improved quality of their essay from revising process. Editing is helpful to identify errors and correct the substance of the essay. They edited grammars, dictions, sentence variation, and mechanics as the first priority. The next, they revised rhetorical patterns and their contents. See quote (4) as for the example.

Revising is very important for me. I can correct many errors on sentences, for example, tenses, passive voice, plural. I can also revise the patterns, that are introduction, developing paragraphs, and conclusion. If we don't check the contents of introduction, body paragraphs, and conclusion, the results may be bad. Thesis statement does not match to the body and conclusion. (M-1-REV).

Upper Group:-

Accomplishment on essay of higher group is better than middle and lower group. Indication was shown from the quality and strategies used by the higher group in the drafting and revising process in writing the essay.

In the drafting process, most students in higher group were aware of the presence of outlining. They asserted that prior to writing the outline, they browsed and generated ideas to focus the topic. They formulated thesis statement as a claim of the essay and tried to find evidences to support the claim. Relying on the outline, they formally drafted the essay as the outline suggested. See excerpt (5).

To me, selecting certain topic is very important. I usually browse through internets or discuss with friends about the topic I am going to write. Initially, I jot down the ideas on a paper as an outline then I revise if I think new ideas are better. I will write confidently if I consider that my outline is complete. (H-1-DRAF).

Actions with care on revising process were stated by the higher group students. Surprisingly, higher group students did all aspects in the revising process: evaluating the paper, editing, revising and proofreading. In the evaluation process, students focused to see rhetorical patterns. Identification on thesis statement, topic sentence, developing details in introductory paragraph, body paragraphs, and concluding paragraph was done. The next, students checked any errors in the whole texts. The last step, students asked their peers and teachers to proofread the paper.

Overall, results of interview revealed that majority of students literally translated from L1 to English when writing English essays. The students seemed to be bothered by lack of vocabulary knowledge. Literal translation was done for two main causes: use of bilingual dictionaries and lack of exposure to English materials. Lack of exposure resulted word-to-word translation and writing deficiencies including grammar errors, cohesion errors, and lack of academic style.

Discussion:-

Writing quality is affected by strategy use. Processes for writing differ from one writer to another and from one situation to the next (Reid, 1992). However, three basic stages of writing process are common: pre-writing, drafting, and post-writing through which writers practice a number of strategies. Pre-writing includes generating ideas relevant to the subject (Dupont, 2004). Drafting involves writing the first version and composing thoughts in sentences and paragraphs (Winterowd & Murray, 1988). Post-writing stage cleaned up errors such as spelling, omission, extra spaces, formatting, and punctuation errors (Dupont, 2004).

Writing is a multidimensional communicative process, concerning “the generation, analysis, and synthesis of ideas; the organization of discourse; the control of sentence structure; the vocabulary, spelling, and mechanics” (Williams, 2007:12). Writing is related to emotion, cognition, motivation, logical reasoning, linguistic skills and contextual adaptation; it is also based on sufficient acquisition of English, so some people say writing can best reflect an individual’s education.

EFL writing practice has two goals: language acquisition and writing skill practice. The first goal focused on linguistic accuracy, discourse pattern, and made students write from sentence to paragraph to essay. This way, students were equipped to write with a clear and rigid format: an introduction with thesis statement, supporting paragraphs, a concluding paragraph, sometimes some rhetorical devices like compare-and-contrast or cause-and-effect were used. The latter goal focused on writing process, express meaning and content (Budiharso, 2006; Yiu, 2012; Solikhah, 2017).

Majority of students produced low quality essays because they were not serious or critical enough when producing drafts or revising. The prominent problems related to writing attitude and habits. When composing, some students did not use certain strategies like to think critically about writing process, plan and develop contents, edit or even revise writing outputs (Ismail, et.al., 2010). They simply wrote without much concern to their writing steps or process, e.g. outline and brief draft.

Regarding translation process in writing, students used more bilingual dictionary than monolingual because bilingual dictionaries may lead to errors. In writing process, students consulted on both the dictionaries. To overcome this problem, students are advised to use both bilingual and monolingual dictionaries.

The final finding drawn in the present study was that most students finished writing process without proofreading. The students were not aware the function of proofreading to qualify an essay. Proofreading means examining the essay carefully for errors in grammar, spelling, punctuation and style. Proofreading can make writing more professional and clearer.

Conclusion:-

This study provides insights that writing characteristics EFL learners to write an essay are about identical. In the process writing, drafting, revising and strategies to write an essay in this study showed similarities with other studies. Lower students were absence of some aspects in the drafting and revising process. Lower group did not write outline and produced first draft without planning. Thesis statement, topic sentence and concluding paragraph did not appear. The middle group was able to produce thesis statement, body paragraphs and conclusion, but they focused less on revising and proofreading. The higher group exhibited by performing all aspects of an essay in each rhetorical patterns. Drafting and revising were used appropriately. Overall, all groups asserted literal translation to develop ideas and overcome problems on diction.

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RESEARCH ARTICLE

A CONTROL STRATEGY FOR THE IMPROVEMENT OF POWER QUALITY IN MICROGRID.

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Abstract

Model Predictive Controller (MPC) is described in this paper for the improvement of power quality in Microgrids. Microgrid is a low voltage grid which is subjected to disturbances. The distributed energy resources are connected to the LV bus by means of power electronic converters. The power quality issues are analyzed with MPC control algorithm in both- Grid connected & Islanded modes. But the Islanded mode is analyzed separately. Predictive control based Distribution static compensator (DSTATCOM) injects voltages at the point of common coupling (PCC) in the Microgrid. Reactive power compensation is done with two linear & one non linear load in both modes of operation. The principles and the proposed algorithm are verified by MATLAB/SIMULINK simulation. Simulation results suggest that this control method can decrease the power quality issues and increase the microgrid stability.

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Introduction:-

In traditional power systems, a few large centralized power generation plants produce most of the power and emissions. The generated electric power is then transferred towards large load centers over long distance transmission lines. The system control centers monitor and controls the system continuously to ensure the quality of the power, namely the frequency and the voltage. However, due to the rapid increase in global energy consumption and the diminishing fossil fuels, the demand for new generation capacities and efficient energy production, delivery and utilization is rising. Utilizing renewable energy [1], distributed generation, and energy storage in large scale can potentially solve such problems as energy shortage and global climate change.

A promising structure to interconnect these distributed Energy resources are the microgrid paradigm. A microgrid is a low-voltage distribution system which is subjected to lot of disturbances when non linear loads such as electric arc furnaces, electric arc welders, adjustable speed drives and switch mode power supplies are connected [3]. These loads may generate harmonics and increase the demand of reactive power flow from the renewable energy resources. Microgrids have to be operated isolated from the main grid in case of faults, which are controlled by the Microgrid central controller. The elimination of these power quality issues in the connection with the distributed energy resources can be done by the usage of Series active filters or the Custom power devices (CPD) like DSTACOM, Dynamic voltage restorer(DVR) and Unified power conditioner(UPQC).

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Traditionally, inverters used in Microgrids behave as current sources when they are connected to the grid, and as voltage sources when they work autonomously [2]. Currently, most inverters in Microgrids adopt the voltage-source inverter (VSI) topology with a current controller to regulate the current injected into the grid. Current-controlled inverters have the advantages of the high-accuracy control of an instantaneous current, peak-current protection, overload rejection, and very good dynamics. The performance of the VSI depends on the quality of the applied current-control strategy and, in order to meet power quality requirements, inverters in Microgrids should have very good capability in harmonic rejection.

There has been considerable research in the field of power quality issues. The various techniques used in the control of custom power devices for the solution to the active & reactive power compensations are Instantaneous reactive power theory, Model Predictive control, Modified power balance theory, Enhanced PLL technique, Artificial Neural network.[5].

Predictive control technique has been chosen for the mitigation of issues in the distributed energy resources based microgrid. Three distributed energy resources are being modeled and the DSTATCOM with MPC controller has been injected into the point of common coupling to compensate the active & reactive power. MPC is an advanced method of process control that allows the time slot to be optimized keeping the future timeslots into account. It has the advantage to anticipate the future events and take control actions accordingly. MPC predicts the change in dependent variables of the modeled system that will be caused by changes in independent variables e.g. reactive power and Harmonics.

2. EXISTING METHODS

This section describes the methods that were already implemented in the mitigation of power quality issues in the distribution systems. Different strategies like proportional–integral (PI), proportional–resonant (PR), predictive deadbeat (DB), or hysteresis controllers have been proposed in the control of active and reactive power compensation. The PI control scheme in the synchronously rotating (d, q) reference frame is commonly used and can work well with balanced systems, but it cannot cope with unbalanced disturbance currents, which are common in Microgrids. The PR control scheme in the stationary (α, β) reference frame is popular due to the capability of eliminating the steady-state error, while regulating sinusoidal signals, and the possible extension to compensate multiple harmonics. However, the resonant frequency in the controller has to be identical to the varying grid frequency in order to maintain good performance. Hysteresis control is simple and brings fast responses, but it results in high and variable sampling frequencies, which leads to high current ripples, poor current quality, and difficulties in the output filter design. In order to obtain a fixed switching frequency, the complexity of the controller will be increased, if an adaptive band hysteresis controller is used.

Compensation and control strategies to eliminate the influence of non-linear and unbalanced loads on Microgrid and the utility grid that connected based on instantaneous power in d-q reference frame has been proposed [3]. In the paper a reference current is generated for the distributed sources (DS) with the difference between the rated power of DS and the power demand of the local non-ideal loads. The power sharing of the DS are not considered in this paper. Compensation algorithm proposed in this paper was based on instantaneous power theory in abc reference frame. It takes no consideration about the unbalanced loads at PCC which are long distances from any DS in the Microgrid. There were no related studies in the field of Microgrid power quality improvement using this approach. Another paper proposes an algorithm and control, based on instantaneous power in d-q reference frame by considering the non-linear and unbalanced loads in the Microgrid [5]. The reference currents converters are generated with respect to the difference between the instantaneous currents at the distribution network and the load sides. Inverse droop control was proposed to achieve the switching of the operating modes. The paper does not tell anything related to fault analysis mode of Microgrid with the existing method.

The work described in another paper regards the evaluation through numerical simulation, of the inverter-fed MG dynamic behavior under islanded operation for different load conditions and using two different control strategies [11]. Control scheme based on droop concepts to operate inverters feeding a standalone ac system is presented. Two kinds of controlling was used to operate an inverter. PQ inverter control and voltage source inverter control (VSI). In the latter control, inverter was controlled to feed the load with pre-defined values for voltage and frequency. This paper describes and evaluates the feasibility of the control strategies needed for Microgrid (MG) islanded operation, where no directly grid-connected synchronous generators are used.

A control algorithm based on correlation function approach in DSTATCOM was done in another paper [5]. In this approach, an extraction of fundamental active and reactive power components of load currents is based on correlation and cross correlation functions in time domain. For estimation of fundamental active and reactive power components of load currents, a numerical integration is applied in correlation and cross correlation function. The improvement of the transient response of parallel connected inverters without using the communication signals were proposed in ac distribution system [6]. A wireless controller was developed, by adding supplemental transient droop characteristics to the conventional static droop approach, which improves the paralleled-system dynamics. Several intrinsic problems led to its limited transient response, as the system dynamics depends on the power calculation characteristics. Another new adaptive neuro-fuzzy control method is utilized to achieve smooth bidirectional power flow and nonlinear unbalanced load compensation simultaneously [8]. In this case, the traditional PI controller might be insufficient because of the instantaneous changes in the dynamics of the system. An integrated diagnostic system was developed for islanding detection using a neuro-fuzzy model for grid-tied inverter-based DGs. In a paper, an adaptive neuro-fuzzy inference system was used for islanding detection.

In reference[8], a current control method for inverters based on the sigma delta modulation algorithm called the sigma delta-based current controlled voltage source inverter interfaced with DG generators was studied. A particle swarm optimization method was used for the optimum tuning of the controllers as a result of the existent number of PI controllers. $\Sigma\Delta_CC_VSI$ minimizes the harmonics of the unfiltered voltage. Thus, it is powerful in minimizing electromagnetic interference, which is critical for sensitive loads. The development of new control strategies and execution of multifunctional compensation capability are the main research trends related to both active power flow control and mitigation of various PQ disturbances using MFIs.

A previous study analyses the limitations of the standard resonant current control operating under abnormal grid conditions and then introduces a control scheme that improves the current harmonic distortion in such adverse conditions without increasing the computational load of the standard current control[12]. The resonant current control has been extensively employed to reduce the current harmonic distortion in a wide range of DG applications. This control uses a proportional-resonant (PRES) compensator to track the fundamental component of the current reference signal and a resonant harmonic (RESH) compensator to attenuate the most important current harmonics. The main problem of the resonant current control is the performance deterioration given by abnormal conditions in the utility grid. Power quality improvement in micro grids by using Active power conditioner was proposed in another literature [13]. The Active Power Conditioner (APC) acts as an interface between renewable energy sources and the AC bus of a micro grid and uses an APC control strategy, which makes it possible to inject energy in the micro grid, compensate the current harmonics and correct the power factor. Proposed control algorithm is compensation method that makes the APC compensate the current of a non-linear load by forcing the Microgrid side current to become sinusoidal and balanced. Compared with linear controllers, the non-linear ones based on hysteresis strategies allow faster dynamic response and better robustness with respect to the variation of the non-linear load. A drawback of the hysteresis strategies is the switching frequency which is not constant and can generate a large side harmonics band around the switching frequency. To avoid this drawback, the switching frequency can be fixed using different solutions like variable hysteresis bandwidth or module bandwidth.

Proposed Method:-

In this section a model of a typical Microgrid is explained that was used to carry out the analysis of the power quality issues. A typical Microgrid is shown in Figure 3.1.

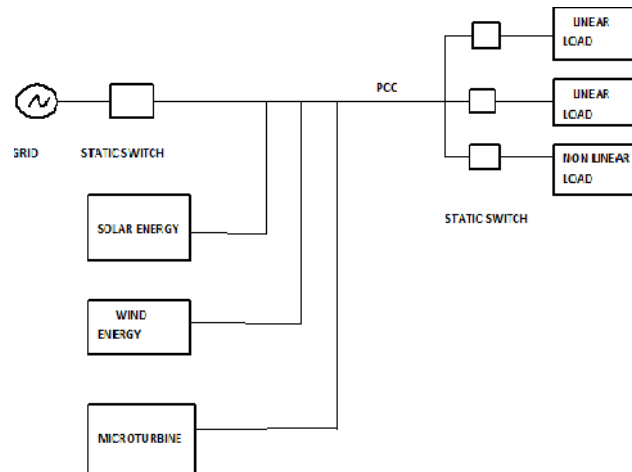


Fig.3.1:- Proposed Microgrid.

In the proposed Microgrid diagram, three distributed energy resources blocks like Solar Energy, Wind Energy and Microturbine are modeled in both Grid connected and Isolated modes of operation. It is then connected to two linear and non-linear loads via a static switch. Linear loads like Incandescent lamps, Heaters etc and the Non-linear loads are SMPS, refrigerators, Television etc. The static switch is closed in grid connected and opened in islanded mode. The compensation of reactive power and harmonics is done by Model Predictive controller (MPC). Each Distributed resources are modeled with the Predictive algorithm separately and added to the point of common coupling.

Model Predictive controller (MPC) concept is most widely used in all modern control techniques. The MPC formulation is based on giving a reasonably accurate model for current system, possible consequences of the current and future manipulated input moves on the future plant behavior and the constraints can be forecasted online and used while deciding the input moves in an optimal manner. MPC has four important tuning parameters which controls the formulation of algorithm. They are weight matrix, the output weight matrix the prediction horizon and the control horizon. Only the first step of the control strategy is implemented, then the model state is sampled again and new calculations are repeated starting from the new current state, yielding new control & predicted path. MPC starts adjusting the control signal head of the reference changes. This model works in two modes of operation

Grid connected mode: In this mode, Microgrid is connected to the Main grid through a static switch. The references for the DS controller can be obtained from the Grid. The reactive power is compensated with the help of the Predictive control algorithm. The Diagram in the Grid connected mode is shown in figure 3.2.

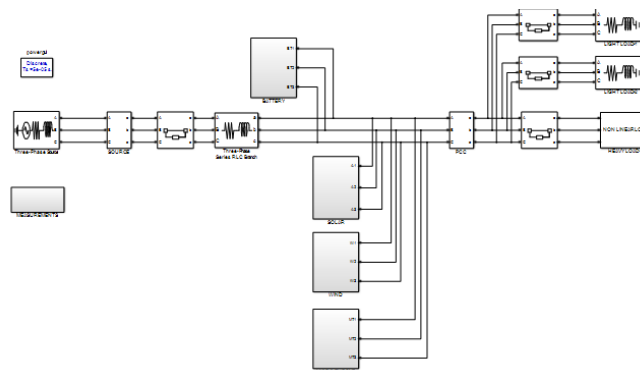


Fig.3.2:- Grid connected mode

Islanded Mode: Islanding of the Microgrid can take place by unplanned events like faults in the grid side or by planned actions like maintenance requirements. In this mode also three DGs are modeled using MPC controller and connected to the PCC. The main grid is disconnected from the Microgrid by using the static switch. In this case, the local generation profile of the microgrid can be modified in order to reduce the imbalance between local load and

generation. The Microgrid has to work autonomously feeding the loads. MPC generates references for the function of the Microgrid. After 0.2s the Grid is added to the Microgrid. The diagram of the isolated mode is shown in the figure 3.3.

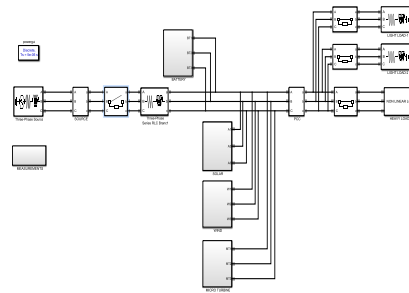


Fig.3.3:- Isolated mode

In the isolated mode of operation due to the presence of non-linear loads all the DG's have to compensate the loads, in the absence of the Grid. The present network has a dominant impact in the quality of power delivered to the loads in the case of Isolated mode while considering the non-linear load. For the improvement of reliability of the present system, a modification in the Microgrid is proposed. DSTATCOM was proposed to be added to the existing system. The need for the CPD in the distribution side of the Microgrid was for the smooth function whose performance was very sensitive to the quality of power delivered to the loads. The result of the nonlinear loads in the distribution side makes the waveform of suppliers to be in the distorted form.

A DSTATCOM is a custom power device which is utilized to eliminate the harmonics from the source currents and also balance them in addition to providing reactive power compensation to improve the power factor or regulate the load bus voltage. With the shunt connected controllers (DSTATCOM), it is possible to control the power flow in critical lines. DSTATCOM was also modeled with the Predictive controller to regulate the power flow at the point of common coupling. This system was checked with the two modes of operation and in addition to that fault analysis mode too. The effectiveness of the proposed system was verified using the MATLAB/SIMULINK program.

The Predictive controller is been incorporated in each distributed sources and the DSTATCOM which injects voltages and currents to the point of common coupling in both the modes. MPC controller is shown in figure 3.4.

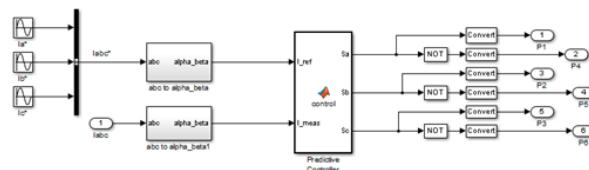


Fig.3.4:- Model Predictive Controller

The reference currents are produced by the MPC by the help of three reference signal constants converted to alpha-beta version and is given to the Predictive control block from the Simulink tool box. Actual currents are also sensed and given to the controller block. Controlled output signals will be fed to the PCC from each modeled Distributed resources. Modeled Solar with MPC is shown in the Figure 3.5.

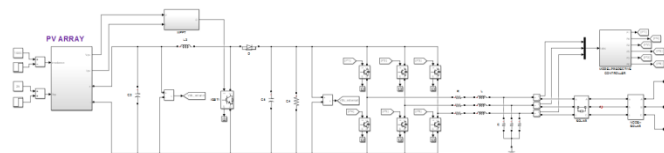


Fig.3.5:- Solar Model with MPC

It consists of a PV array which is fed to six bridge inverter and given the output to the PCC. The pulses to the thyristors are given from the MPC controller.

Results & Discussions:-

The Simulink diagram of the proposed Predictive control based DSTACOM is shown in the figure 4.1.

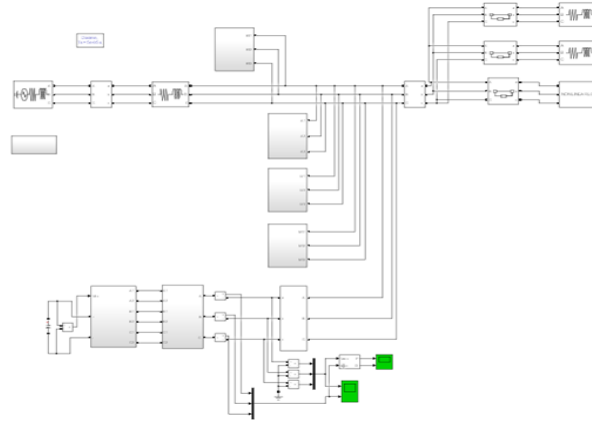


Fig.4.1:- Grid connected mode with DSTATCOM

The test model contains three Distributed energy resources which are modeled with the Predictive controller and the power ratings of the solar and wind models with a varying output of 5kW and reactive power of 10kW varying with respect to time. Three loads are connected to the PCC with two light loads of 25 kW each and a non linear load of 18 kW with 8.5KVAR and 12.3 KVAR respectively. The grid rating is 2500×10^6 VA and 60 Hz frequency.

The test micro grid was simulated in two different modes of operation.

Grid Connected mode:-

The simulation sequence was as follows. Load1 was decreased from 25 kW and 18 kW to 20kW and 10 kW at 1.2 s to investigate power sharing in terms of load variation during grid-connected operation. At 2.0 s, the static switch was opened so that the Microgrid was islanded from the grid. Output Active & Reactive power at the Grid and load side is shown in the simulation graphs from the figure 4.2& 4.3.

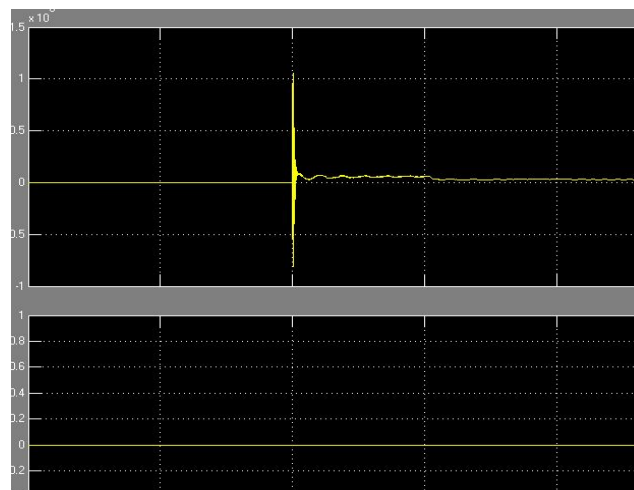


Fig 4.2:- Grid Side Active & Reactive power

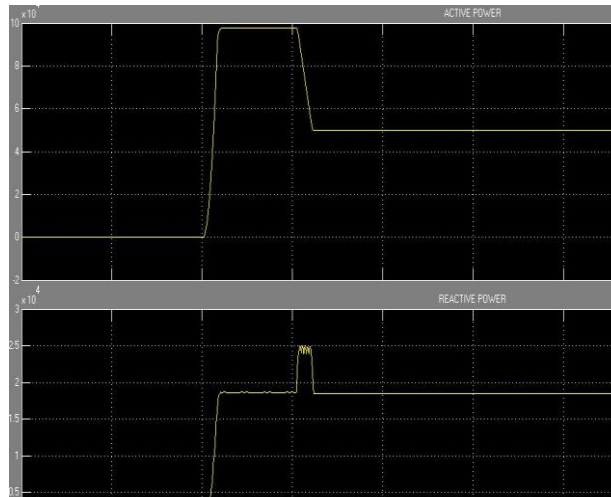


Fig .4.3:- Load side Active & Reactive power

From the above shown graphs, when all the loads are connected to the grid the active power becomes 95kW and reactive power becomes 190 kvar with a small spike.

Islanded Mode:

The static switch was opened and the grid will be disconnected from the AC bus. Load is being varied to appropriate values. The IGBT inverters which are modeled in each Distributed sources with the MPC will operate in Islanded mode. If there is any decrease in the DG output to load, the controller compensates the DS output. Figure 4.4 shows the Microgrid operation in Isolated mode of operation.

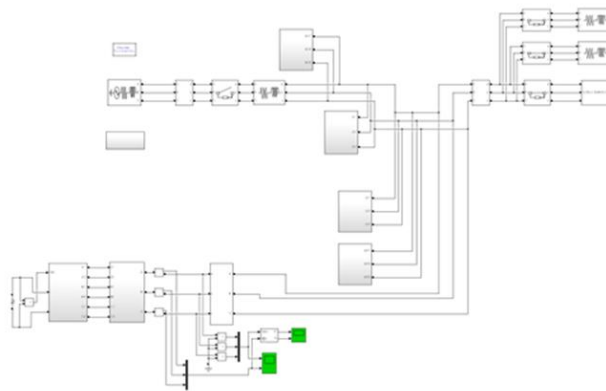


Fig.4.4:- Islanded Mode with DSTATCOM

In the islanded mode of operation, when all the **loads** are connected to Microgrid, load power is compensated by the MPC based DSTATCOM at the PCC. From the figure 4.5..a. It's clear that the DSTATCOM compensates the active and reactive power in the Islanded mode up to 0.2s of the Microgrid and loads. Its negative value is of 2.5×10^4 VAR of reactive and 500 W of active power. Figure 4.5 shows the load side Active and reactive power in the Isolated mode of operation.

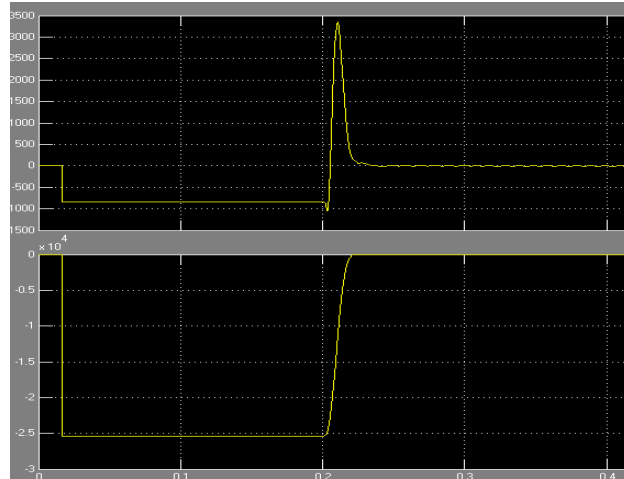


Fig.4.5.a:- DSTATCOM output in Islanded Mode

Voltage variations were seen in this mode of operation when non-linear load is connected. This is shown in the figure 4.5.b.

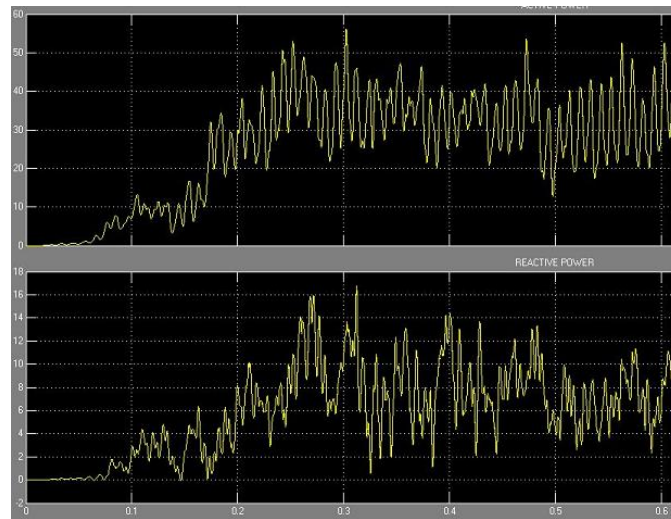


Fig.4.5.b:- load side active and reactive power

Due to the presence of second order harmonics with Non-linear load in the isolated mode of operation all the DG's are connected to a single modelled structure as shown in Figure 4.6 to improve the system stability when the Grid is disconnected.

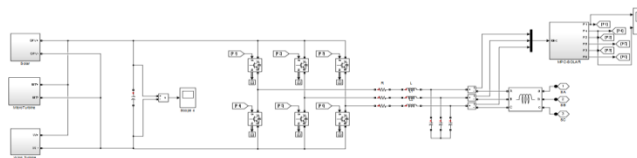


Fig.4.6:- Incorporated view of DG's

All the modelled DG's are connected to a single MPC controller and it has been connected to the point of common coupling (PCC) with the DSTATCOM. The output of the current view is shown in the figure 4.7. It is clear that Reactive power is being compensated in the islanded mode of operation completely. Negative reactive power is caused by capacitive loads that include lighting ballasts, variable speed drives for motors, computer equipment, and inverters.

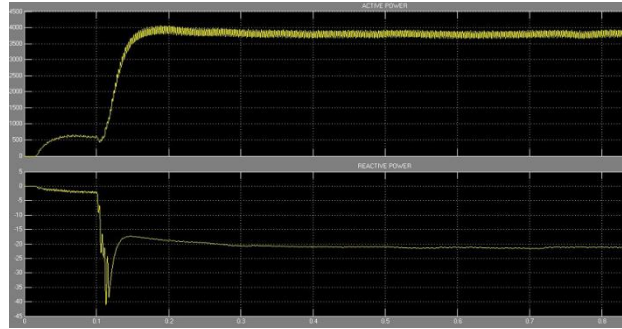


Fig.4.7:- Load active and Reactive power.

Conclusion:-

In this paper an MPC based DSTATCOM was implemented in the Microgrid. The Microgrid reliability and the compensation of the reactive power were examined with the simulation results. Predictive controller was successfully implemented in the Distributed resources and the power quality issues were studied, analyzed and compensated in the proposed Microgrid. The control algorithm was implemented in the conventional distribution system; it is also applicable in the case of Microgrids too. MPC based DSTATCOM was incorporated in the Microgrid and comparative results were done in both Grid connected and Islanded modes with the help of Simulation. In

Grid-connected mode, DGs compensates the non-linear and unbalanced loads and shares with utility grid in a pre specified ratio. During islanded mode harmonics were present on the system with non-linear load, on basis of the future analysis advantage characteristics and the variations of voltage and frequency, an incorporated view is being suggested and the satisfactory results were produced. Battery source was also added to the Microgrid to compensate the loads in the islanded mode. MPC offers a good improvement from the conventional PI and PR controllers in the presence of linear and non linear loads. A future development is to check the MPC controllers with the fault analysis mode in the proposed Microgrid.

APPENDIX

DC VOLTAGE	400V
SWITCH FREQUENCY	16Hz
GRID VOLTAGE	400kV, 2500MVA
VOLTAGE IN ISLAND	390kV, 60Hz
LIGHT LOAD1	25kW ,8.5kVAr
LIGHT LOAD2	25kW, 10kVAr
NONLINEAR LOAD	18kW , 12.3kVAr
X/R RATIO	10
V_{dc}	350 V (Vary)

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RESEARCH ARTICLE

AMELIORATING EFFECTS OF TOMATO PUREE AGAINST ARSENIC INDUCED ALTERATION IN OVARY OF SWISS ALBINO MICE.

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Key words:-

Arsenic (As), tomato puree, ovary and follicle.

Abstract

Arsenic compound are known to induce toxicity in mammalian system. However reproductive toxicity is comparatively less expedited. Arsenic has a long environmental persistence and never loses its toxic potential. Arsenic affects the reproductive systems of female Swiss mice. In the present study, toxic effects of arsenic are observed on histopathology of ovary studied after administration of low (1mg/kg body weight) dose of arsenic trioxide to the Swiss albino mice; along with this tomato puree on arsenic induced toxicity has also been examined. A significant weight loss of ovary was also observed. Moreover, arsenic was also responsible for ovarian follicular degeneration and vacuolization in follicular cells. The results of the present study indicated a marked effect of recovery of the follicle cells of the ovary after exposure to tomato puree with higher dose as it was dose dependent.

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Introduction:-

Arsenic in drinking water is one of the top most environmental threats worldwide as it is associated with numerous diseases (Tapio and Groshe, 2006). Environmentally relevant forms of arsenic are inorganic and organic existing in the trivalent or pentavalent state (ASTDR 2007b). Presently, due to enhanced human activities like mining, smelting, coal combustion, etc., the level of arsenic has crossed its permissible limit (Sharma and Singh, 2016) and Chronic intake of arsenic is strongly associated with an increased risk of skin, lung, liver and other cancers, type 2 diabetes, cardiovascular diseases, neurological, cognitive defects, reproductive and developmental problems (Kargas *et al.*, 2002; Prozialerk *et al.*, 2008; Walker *et al.* 2009). But recently arsenic intoxication in experimental animals has been associated with hepatic tumors (Waalkes *et al.*, 2003), inhibition of testicular steroidogenic function (Sarkar *et al.*, 1991) and spermatogenesis (Sukla and Pandey, 1984) as well as with severe metabolic disorders such as diabetes in humans (Longnecker and Daniels, 2001; Tseng *et al.*, 2002). Long term exposure of arsenic is associated with abortion, low birth weight and reduced lactation (Donald *et al.*, 1995) as well as with embryonic cells toxicity in vitro (Lee *et al.*, 1985). Though, there is scanty literature on the effect of arsenic on ovarian steroidogenic functions, particularly at the dose levels occurring in drinking water in wide areas of India and in other countries where this trace element is present in the range above the admissible limit of 0.01 ppm, (Rahman *et al.*, 2003). Chattopadhyay *et al.*, (1999) considered arsenic to be responsible for the inhibition of ovarian steroidogenesis and as an elevator of adrenocortical steroidogenesis (Ghosh *et al.*, 1999) even when the level of arsenic was within the range in drinking water of West Bengal.

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Chemotherapeutic drugs are effective against cancer cells because they are designed to interfere with rapidly dividing cells. Many chemotherapeutic agents induce the production of free radicals. These free radicals can be bound and detoxified by antioxidant compounds such as lycopene (Sahin, *et al.*, 2010). Lycopene accumulates in relatively few tissues and is found in ripe tomato fruit, watermelon and pink grapefruit giving them a characteristic red pigmentation. Tomatoes and processed tomato products such as juice, ketchup, paste, sauce and soup all are good sources of lycopene and may account for over 85% of dietary lycopene in the North American diet. Lycopene is considered as cardioprotective and anticarcinogenic (Chen *et al.*, 2007). The lycopene content of tomatoes varies with the variety and increases with fruit ripening. Several studies (Bohm *et al.*, 1995; Lu *et al.*, 1995; Dimascio *et al.*, 1989) have indicated that lycopene is an effective antioxidant and free radical scavenger. The present work is aimed to evaluate the protective role of tomato puree on arsenic induced toxicity in ovaries of albino mice.

Materials And Methods:-

Animal selection and care:-

Female Swiss albino mice having weight 20-30g were procured from CENTRAL RESEARCH INSTITUTE, KASALI and DISTRICT SOLAN (H.P.). The present research work has been carried out with the permission of Institutional Ethical Committee (107/99/CPCSEA/2013-02).

They were kept and acclimatized to laboratory conditions for 2 weeks under optimal condition of light and temperature. The animals were given standard mice feed and *ad libitum* access to R.O water. The animals were handled with humane care in accordance with National Institute of Health guidelines.

Chemicals: - The Arsenic trioxide was obtained from QUALIKEMS FINE CHEMICAL PVT. LTD., NEW DELHI. Its stock solution was prepared by dissolving 1.32g of As₂O₃ and 4g of NaOH in one litre of distilled water (1ml of stock solution contained 1mg of As) and was administered intraperitoneally to mice. Tomato puree marketed by KISSAN HINDUSTAN UNILEVER PVT. LTD. MUMBAI was used as a source of lycopene. It contained 36.8% tomato paste.

Experimental design: - Mice were divided into following groups:

Group I- Control animals were given equal volume of distilled water only.

Group II- Albino mice were administered arsenic trioxide at a dose of 1mg/kg body weight (b.wt.) intraperitoneally (i.p).

Group III- Mice were injected an acute dose of 1mg/kg b.wt. of arsenic trioxide i.p. followed by a daily dose of 20mg/kg b.wt. of lycopene for 15 days.

Group IV- Mice were injected an acute dose of 1mg/kg b.wt. of arsenic trioxide i.p. followed by a daily dose of 40mg/kg b.wt. of lycopene for 15 days.

Autopsies from each group were done on 15 days post treatment. Ovaries were removed, freed of adipose tissue, blotted dry so as to remove blood and were weighed separately.

Histopathological studies:-

The ovaries were fixed in alcoholic Bouin's fixative and embedded in paraffin wax (58-60°C) and the sections were cut at 5-7µ thick sections were stained with Heamatoxylin-eosin stains.

Statistical analysis: - Data were subjected to Student's *t*- test is used to compare the control group vs. treated (arsenic) group and antioxidant treated groups.

Results And Discussion:-

Heavy metals toxicity is a worldwide problem which adversely affects the growth, health, reproductive performance and life span of living organisms (Dhir, 2011). The present study showed a histopathological alterations in ovary of arsenic treated mice were in the form of ovarian follicular degeneration, vacuolization in granulosa cells, loss of uniformity in granulosa cell layer. Chattopadhyay *et al.*, (2003) suggested that As influences hypothalamic pituitary ovarian axis which leads to deficiency in secretion of growth hormone and hence reduction in body weight. Similarly Chattopadhyay *et al.*, (2003) observed ovarian follicular and uterine cell degeneration which was also accompanied by decrease in dopamine levels in the midbrain and diencephalon as well as increased arsenic levels in ovaries, uterus and plasma. Low dopamine levels could decrease gonadotrophin synthesis and secretion. Thus, it attributes to decrease in the number of healthy follicles and increase in the number of apoptotic follicles. The authors suggested that uterine cells degeneration may be due to low ovarian estradiol after arsenic treatment.

Histological changes in Ovary:-

The histopathological alterations observed in ovary of arsenic treated mice were in the form of ovarian follicular degeneration, vacuolization in granulosa cells, loss of uniformity in granulosa cell layer loose germinal layer, hyperaemia in medulla and vacuolization in germinal epithelium cells, (Fig.2,6,8) and degenerative changes in the oocyte and reduction in the secondary and graafian oocytes along with an increase in the number of atretic oocytes. These findings are supported by the results of Chadhopadhyay *et al.*, (2003); Islam *et al.*, (2011) and Dezfouli *et al.*, (2014).

Moreover, it is shown that As is responsible for vacuolization, atrophy and hyperaemia in the medulla along with the decrease in the number of follicles (Jhala *et al.*, 2004; Dezfouli *et al.*, 2014).

Estrogen plays a critical role in regulating the sex organs as well as sex gonadal development. This implies a direct impact on the growth of ovary cells, particular folliculogenesis. Furthermore, estrogen elevates the effects of FSH on granulosa cells (Hegele-Hartung *et al.*, 2003) and in this way performs an important role in follicular development. Arsenic reduces the level of estrogen and as an endocrine disruptor can cause ovarian follicular degeneration (Dezfouli *et al.*, 2014).

In the present study, the ovary of TP (20mg) treated mice showed normal histology as compared to arsenic treated mice (Fig 3.). Fig.9 depicted moderate signs of protection as primary follicles showed vacuolization in granulosa cells the number of primary follicles was observed to be more as compared to arsenic treated mice. Group IV As depicted mice co-administration with 40mg/kg b.wt. of TP showed a great protection to the ovary against arsenic toxicity. The graafian follicles as well as germinal epithelial layer retained their original shape, structure and thecal layers were also properly discernible (Fig. 4,10).

The existence of LH (Luteinizing hormone) receptor sites in granulosa cell membrane is induced by FSH (Follicle stimulating hormone) that prepares the graafian follicles for a preovulatory LH surge. Furthermore, induction of LH receptors by FSH is necessary for interactions between granulosa cells, which is important in cell differentiation. It is also believed that estrogen enhances the effects of FSH on gene expression in granulosa cells (Erickson *et al.*, 1979). As granulosa cells play important role in development and maturation of oocytes, the dysfunction of granulosa cells may lead to disorder of folliculogenesis and steroidogenesis (Lan *et al.*, 2013). It has been demonstrated that LH stimulates follicular maturation and induces follicular atresia. Sharma *et al.*, (2012) recently suggested that LH is also a stimulant for early stages of follicular growth. Heavy metals not only exerted inhibitory effect on follicular growth but also stimulated the atretic follicle formation (Tajima *et al.*, 2007; Mori *et al.*, 2009).



Fig.1

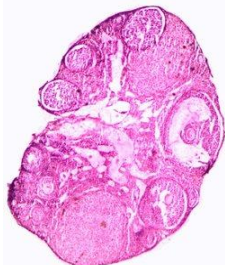


Fig.2



Fig.3

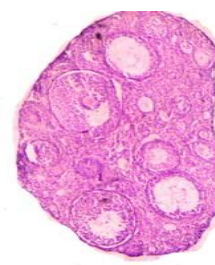


Fig.4

Hematoxylin and eosin-stained sections of mice ovary (40×): **Fig. 1** Normal control mice ovary: normal medulla matures graafian follicles and normal surface epithelium. **Fig.2** arsenic (1mg) — treated mice ovary: atretic follicles. **Fig.3** As 1mg+ TP 20mg —treated mice ovary: vacuolization in granulosa cells. **Fig.4** As 1mg+ TP40 mg— treated mice ovary: almost normal graafian follicle with normal architecture of medulla.

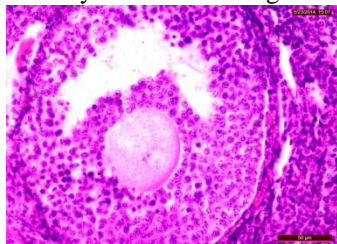


Fig.5

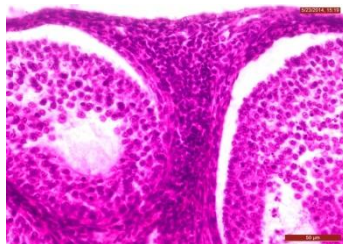


Fig.6

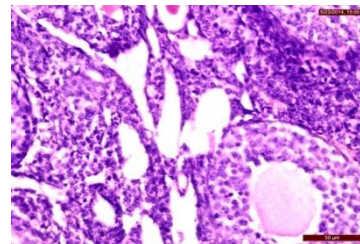
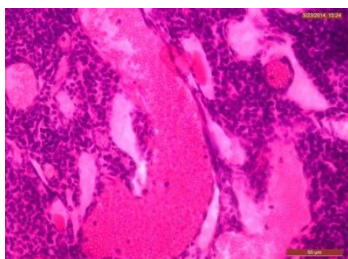
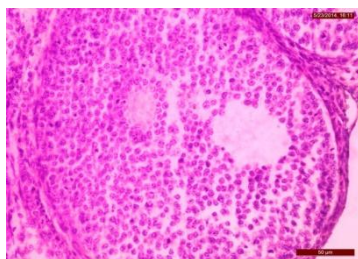
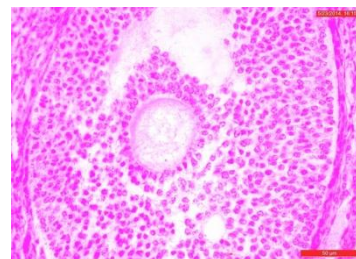


Fig.7

**Fig.8****Fig.9****Fig.10**

Hematoxylin and eosin-stained sections of mice ovary (400×) **Fig.5** Normal control mice ovary: normal cells of graafian follicle, compact layer of granulosa cells. **Fig.6** Arsenic (1mg/kg) —treated mice ovary: vacuolization loosening of granulosa cells, vacuolization in germinal epithelium cells. **Fig.7** Normal control mice ovary: normal architecture of medulla. **Fig.8** Arsenic (1mg/kg)— treated mice: hyperaemia in medulla. **Fig.9** As 1mg+TP 20mg— treated mice ovary: vacuolization in granulosa cells, the layer is not uniformly compact (400×). **Fig. 10** As 1mg+ TP 40mg— treated mice ovary: compact layer of granulosa cells, oocyte and almost normal architecture of theca externa.

Conclusion:-

There are many evidences supporting the antitoxic and anticancer action of tomato puree. The results of the present study demonstrates that arsenic induced histomorphological and oxidative stress is responsible for promoting deleterious effects of arsenic in the ovaries, but tomato supplementation (tomato puree) which is considered as one of the most potent antioxidants among dietary carotenoids showed very promising and encouraging ameliorative effects on arsenic induced oxidative stress in female reproductive organs of swiss albino mice.

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RESEARCH ARTICLE

CREATING CONSUMER PURCHASE DECISION : ANALYSIS OF BRAND IMAGE, SERVICE QUALITY AND MARKETING COMMUNICATION. (A Case Study on Mal Artha Gading/MAG Jakarta)

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Abstract

The research aimed to examine and analyze either partial or simultaneous influence of brand image, service quality and marketing communication on purchase decision of Mal Artha Gading consumers. Of 40.000 populations, 100 samples were obtained, based on Slovin's sampling technique. For data collection, the research relied on questionnaires and interviews. Descriptive and inferential statistics were applied for data analysis. The research findings have revealed that brand image, service quality and marketing communication influence consumer purchase decision simultaneously. Partially, brand image variable gave the greatest and the most significant influence on consumer purchase decision, while service quality did not influence consumer purchase decision very much; marketing communication had a significant impact on consumer purchase decision. The implication is that in order to have consumers make a purchase, the management had better improve its brand image, service quality and marketing communication.

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Introduction:-

Shopping malls in Indonesia have evolved so fast. Starting from Sarinah which is the first mall in Indonesia with about 5,000 m² width of retail area in 1962 nowadays Jakarta becomes a city with the greatest number of malls and shopping centres in the world. It has 173 shopping centres spreading in 5 areas of its, with 4,232,100 m² retail area used.

The condition is experienced by Mal Artha Gading, one of malls and shopping centres in Kelapa Gading which has wider total net leaseable area than other shopping centres in Jakarta have. It is 373,228 m² width in which the shopping centres built have different concept but same catchment area.

Mal Artha Gading is located in Kelapa Gading, precisely beside Cawang – Tanjung Priok toll road. Since it was opened to the public on 27 October 2004, the mall has greatly competed with other malls such as Mal of Indonesia (MOI) built in the first year of Mal Artha Gading operation, Bay Walk Pluit, Kota Kasablanka, Gandaria City and Mal St.Moris Puri which are more modern. The competition continues until the building Mal Kelapa Gading 3.5 and Gading Walk. This makes Mal Artha Gading keep competing with other malls and shopping centres.

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A success measurement of a shopping center is the increasing number of its visitors indicated by the achievement of total visitor numbers targeted by its management. It is experienced by Mal Artha Gading with 38,000 visitors per day, below its target which is 40,000 to 43,000 visitors per day. Table 1 shows data of unachieved daily visitor target of Mal Artha Gading

Table 1:- Average Number of Daily Visitor of Mal Artha Gading from January to May 2015

Visits	Daily	Weekday	Weekend	Total	Target	Dev
Customer	38,000	30,000	53,000	1,150,000	1,250,000	-8%
Cars	8,000	7,000	11,000	240,000	262,500	-9%
Motorcycle	2,500	2,700	3,000	78,000	87,500	-11%

Source : BP. MAG 2015

In 2012 – 2015 the number of visitors decreased; the management tried to find the cause, whether it is internal factors or external factors. In the last three years, the number of its visitors is presented in Table 1.3.

Table 1.3:- Average Number of Monthly Visitor from 2013 to 2014

Years	Average Number of Monthly Visitor	Target	Deviation (%)
2012	1,048,800	1,100,000	-5%
2013	1,037,875	1,150,000	-10%
2014	1,092,500	1,200,000	-9%

Source : BP. MAG 2015

Based on the initial survey conducted in Central Jakarta, South Jakarta, West Jakarta, and East Jakarta, Mal Artha Gading was found less popular than the same level Mal in the same area. Therefore, brand image and marketing communication strategy have important roles. In addition, service quality also takes part in making consumers visit the mall.

In accordance with the introduction above, three identified problems are: 1) unachieved visitor target; 2) unpopularity of Mal Artha Gading; 3) image of Mal Arta Gading identical with a second class mall resulting in low interest of visitors; 4) poor service quality.

In addition to the phenomena which have been explained, this research was supported by the findings stated in international journals involving: 1) Ong and Sugiharto (2012) , Evelina, Handoyo, and Listyorini (2012), and Nurlisa and Sofiyah (2010), suggesting that brand image, service quality and promotion positively and significantly influence purchase decision; 2) Kusumah Ridwan (2011) and Tristania Widiyanti (2008) revealing that service quality has significant impact on purchase decision; 3) Zikmund, William G. (2003), in Hapzi Ali (2012), finding that service quality positively and significantly influence brand image.

Based on the problems, this research aims to analyze the influence of:

- 1) Brand image on purchase decision;
- 2) Service quality on purchase decision;
- 3) Marketing Communication on purchase decision, and
- 4) Brand image, service quality, and marketing communication simultaneously on purchase decision

Literature Review:-

Brand Image:-

Ditcher (1985) defines image as an illustration that in detail explains of the entire entity impression of object owned by society. Kotler and Armstrong (2001:225) state that brand image refers to a set of consumer believe in a certain brand. Brand image can be defined as a perception about brand as reflected by the brand association held in consumer memory (Keller, 1998:93).

In this research the context of image measured is the image of a shopping centre. A study on image of a shopping centre reveals that communication and consumers' shopping experience in a shopping centre and consuming goods or service experience influence their purchase interest. Communication among consumers known as word-of-mouth

communication, promotion and communication in media influence customer perception (Zeithmal and Bitner, 1996).

In according to the theories, this research analyzed brand image variable using theory proposed by Chang and Tu (2005) that is considered to be relevant for studying a shopping centre with four available dimensions which are facility, service, activity, and convenience.

Service Quality:-

Quality is a dynamic condition that has impacts on products, service, human being, process, and environment that meets or exceeds the expectations (Tjiptono, 2001). Thus service quality can be defined as an effort to meet the need and desire of consumer and its delivery directness in balancing the consumer expectation (Tjiptono, 2001).

Parasuraman, et.al (1998) concludes that there are five dimensions of service quality, which are: 1. tangibles, 2. reliability, 3. assurance, and 4. Empathy. According to Kotler and Keller (2008:156), there are some dimensions of quality in the context service quality which can be used as reference; they include responsiveness, reliability, empathy dan assurance serta tangibility.

Based on the theories, this research analyzed service quality variable using theory by Parasuraman (2001) that is considered to be relevant for studying a shopping centre with five available dimensions which are tangibility, reliability, responsiveness, assurance, and empathy.

Marketing Communication:-

Mahmud Machfoedz (2010:16) states that marketing communication refers to all elements of marketing that have value and communicate the value to consumer and stakeholder of a company. Kotler and Kevin Lane Keller (2007:204) suggest that marketing communication is a mean used by a company to inform, persuade, and remind consumer either directly or indirectly about products and brands that they sell.

Furthermore, Prisgunanto (2006:8) state that marketing communication refer to all promotion elements of marketing-mix involving any form of communication between organization and its target audience for marketing performance. In accordance, it is concluded that marketing communication is an effort of a company to send its message about the existence of its products in market to the consumer and stakeholder

Based on the theories, this research analyzed marketing communication variable using theory by Prisgunanto (2006) that is considered to be relevant for studying a shopping centre with eight available dimensions which are direct marketing, sales promotion, marketing public relation, personal selling, advertising, publicity, event/sponsorship, and interactive marketing.

Purchase Decision:-

Phillip Kotler (2005:204) suggests five stages of purchase decision process, which are need recognition, information search, alternative evaluation, purchase decision, and post purchase behavior. This model emphasizes that process starts before the purchase and has further impacts after the purchase.

Kotler (2006:129) mentions five types of decision which customers make, which are product options, brands, distributor choices, time of purchase, and number of purchase. Consumers can take purchase decision on the number of purchase to make in a time. In addition, Kotler (2012:166) states that purchase decision is a process consisting of five stages passed by a consumer before making decision to purchase and after making a purchase.

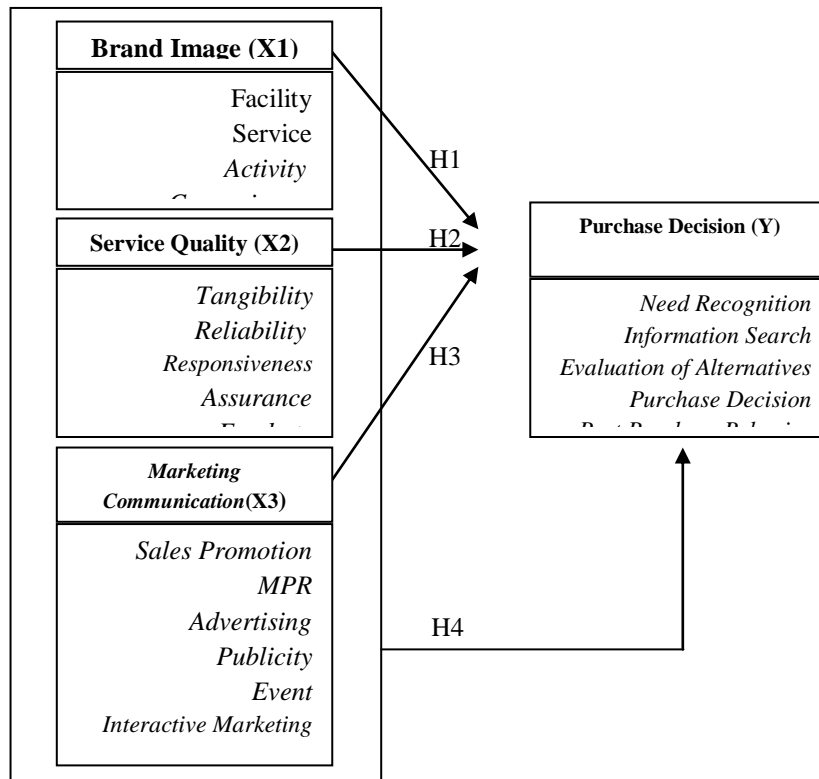
Based on the theories, this research analyzed purchase decision variable using a theory by Philip Kotler (2005) that is considered to be relevant for studying a shopping centre with five available dimensions, which are need recognition, information search, alternative evaluation, purchase decision, and post purchase behavior.

Conceptual Framework:-

Conceptual framework has a function to design the influence or relationship between variables, that brand image, service quality, and marketing communication positively and significantly influence consumer purchase decision either partially or simultaneously.

The conceptual framework of this research was formulated based on background of the study, some proposed theories, and some previous researches involving: 1) Ong and Sugiharto (2012), Evelina, Handoyo, and Listyorini (2012), and Nurlisa and Sofiyah (2010), suggesting that brand image, service quality and promotion positively and significantly influence purchase decision; 2) Kusumah Ridwan (2011) and Tristania Widiati (2008) revealing that service quality has significant impact on purchase decision; 3) Zikmund, William G. (2003), in Hapzi Ali (2012), finding that service quality positively and significantly influence brand image. The conceptual framework of the research is illustrated in Figure 1.

Figure 1:- Conceptual Framework



Based on the conceptual framework, the hypotheses of this research are:

1. Brand image positively influences the consumer purchase decision.
2. Service quality positively influences the consumer purchase decision.
3. Marketing communication positively influences the consumer purchase decision.
4. Brand image, service quality, and marketing communications simultaneously positively influence the consumer purchase decision.

Methods:-

This research is a quantitative research which analyzes numerical data and processes the data using statistical method (Azwar, 2009a: 5). It employed correlation approach. It examined the correlation of brand image, service quality, and marketing communication and number of Mal Artha gading visitors. Arikunto (2010: 4) explains that correlational research aims to find the relationship between two or more variables without either changing or manipulating the available data.

Survey method was applied in this research. Of 40.000 populations, 100 samples were obtained, based on Slovin's sampling technique (Husein Umar, 2003:120). The populations of this research were visitors who did shopping in Mal Artha Gading. They were assumed to be loyal customer indicated by their visit frequency, at least once a week, and by their age, age of 18-24 years old that tends to try new things (Wood, 2004, in Setyaningrum, 2007: 110)

Quantitative analysis using regression analysis with SPSS 2.33 was applied. Before analyzing the data, test of questionnaire validity and reliability and test of classical assumption were performed (Solimun, 2008). After Path analysis, the data were analyzed using R^2 Test. T Test and F Test with 5% error tolerance were performed to test the hypotheses.

Result & Discussion:-

Reliability test was done to find and measure if the questionnaires used in the research were relevant and if the reliability of the questionnaires was strong as *composite reliability* > 0.6 . the result is shown in table 4.1.

Table 4.1:- Statistical Reliability of Each Variabel

	Cronbach's Alpha	N of Item
<i>Brand Image</i>	.924	20
<i>Service Quality</i>	.902	20
<i>Marketing Communication</i>	.866	20
<i>Purchase Decision</i>	.831	20

The table shows that all dependent variables and independent variables have cronbach's alpha > 0.6 . It indicates that the variables are reliable and can be used in measurement for data collection.

The following is histogram to investigate that the dependent variable, purchase decision, is normally distributed. One of the requirements to continue to the next stage, regression analysis, is that the dependent variable is normally distributed. The result is shown in Figure 2.

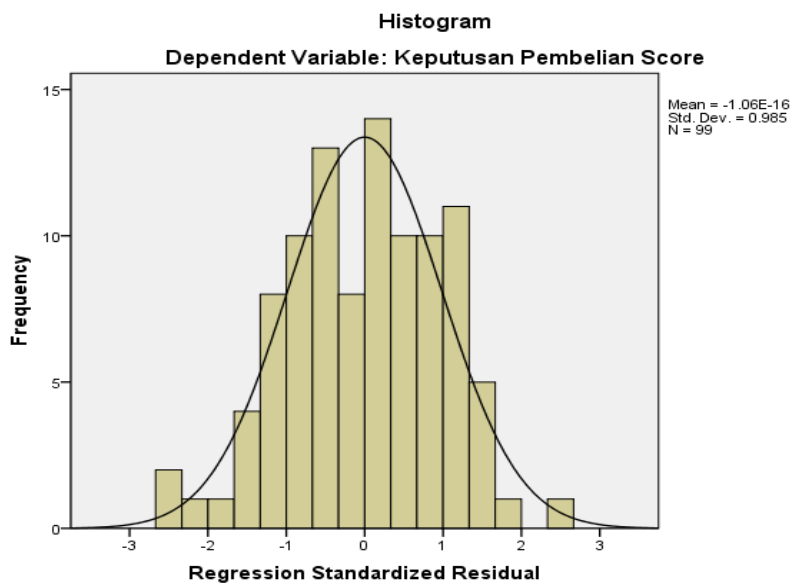


Figure 2:- Histogram

Figure 2 shows that the available data can be used for the next stage that is regression analysis.

Research Findings:-

The Alpha value of the variables of brand image, service quality, marketing communication and purchase decision are 0.924, 0.920, 0.866 and 0.831 respectively. This indicates that reliability of four variables is very strong, which is measured by 20 indicators.

Hypothesis Testing:-

a. Simple Regression Test of brand image on purchase decision

Brand image variable and purchase decision variable have a relationship (coefficients $7.656 > 1.9887$ and $0.000 < 0.05$) and they have a strong relationship (model summary 0.624) and it can be generalized to the population level (ANOVA table 0.000).

b. Simple regression test of service quality on purchase decision

Service quality and purchase decision have a relationship (coefficients $5.897 > 1.9887$ and $0.00 < 0.05$), and they have a strong relationship (model summary 0.514) and it can be generalized to the population level (ANOVA 0.000).

c. Simple regression test of marketing Communication on purchase decision

Service quality and purchase decision have a relationship (coefficients $5.995 > 1.9887$ and $0.00 < 0.05$), and they have a strong relationship (model summary 0.520) and it can be generalized to the population level (ANOVA 0.000).

F test:- Table 4.2. ANOVA

Model	Sum of Square	Df	Mean Square	F	Sig.
1 Regression	2060.479	3	686.826	25.951	.000 ^a
Residual	2514.249	95	26.466		
Total	4574.727	98			

a. Predictors: (Constant), Marcom Score, brand image Score, service quality Score

b. Dependent Variable: purchase decision Score

Table 1 shows that the variables of brand image, quality of service and marketing communication simultaneously affect purchase decision variable.

T test:- Table 4.3. T test

Model	Unstandardized Coefficients		Standardized Coefficients	T	Sig.	Collinearity Statistics	
	B	Std. Error	Beta			Tolerance	VIF
1 (Constant)	11.177	4.711		2.373	.020		
Brand image	.390	.093	.513	4.178	.000	.383	2.610
Service quality	.033	.102	-.041	.322	.748	.362	2.760
Marcom Score	.194	.060	.291	3.208	.002	.705	1.418

a. Dependent Variable: purchase decision Score

Table 2 shows that: 1) brand image has a positive and significant impact on purchase decision, 2) Service quality does not affect significantly on purchase decision, and 3) marketing communication has a positive and significant impact on purchase decision.

Multiple Regression Modeling:-

Based on multiple regression modeling, there is a relationship between brand image and marketing communication, and purchase decision and they have a strong relationship (model summary 0.671). This data can explain 45% of purchase decision variance and it can be generalized to the population level (table ANOVA 0.000).

Modeling of Regression Equations:-

- Brand image has a positive effect on purchase decision. If the variable of brand image increases, assuming marketing communication variable is constant, the purchase decision will also increase.
- Marketing communication has a positive effect on purchase decision. If the variable of marketing communication increases, assuming the brand image variables is constant, the purchase decision will also increase.
- The variable of brand image has the most significant effect on purchase decision.

Exploration of Correlation Per Dimension:-

- Brand image, quality of service, marketing communication and problem identification

The variables of brand image, service quality, marketing communication and problem identification do not have a relationship. It can be seen from the table of Significance, which is greater than 0.05.

- Brand image and information search

There is a relationship between brand image and information search (coefficients $2.987 > 1.9887$ and $0.04 < 0.05$), and its relationship is weak (model summary 0.290).

- Service quality and information search

It reveals that there is no relationship between the variable of service quality and information search dimension (coefficients $1.792 < 1.9887$ and $0.76 > 0.05$) and its relationship is weak (model summary 0.290).

- Marketing communication and information search

It reveals that there is a relationship between the variable of marketing communication and information search dimension (coefficients $4.423 > 1.9887$ and $0.00 < 0.05$), and it has a strong relationship (model summary 0.410).

e. Brand image and alternative evaluation

It shows that there is a relationship between brand image and alternative evaluation dimension (table of coefficients $7.683 > 1.9887$ and $0.00 < 0.05$) and it has a strong relationship. (model summary 0.613).

f. Service quality and alternative evaluation

It shows that there is a relationship between service quality and alternative evaluation dimension (coefficients $6.135 > 1.9887$ and $0.00 < 0.05$), and it has a strong relationship (model summary 0.527).

g. Marketing communication and alternative evaluation

It shows that there is a relationship between marketing communication and alternative evaluation (coefficients $5.115 > 1.9887$ and $0.00 < 0.05$), and it has a strong relationship (model summary 0.459).

h. Brand image and purchase execution

It shows that there is a relationship between brand image and purchase execution dimension (coefficients $5.285 > 1.9887$ and $0.00 < 0.05$), and it has a strong relationship (model summary 0.471).

i. Service quality and purchase execution

It shows that there is a relationship between service quality and purchase execution dimension (coefficients $4.277 > 1.9887$ and $0.00 < 0.05$), and it has a weak relationship (model summary 0.397).

j. Marketing communication and purchase execution

It shows that there is a relationship between service quality and execution purchase dimension (coefficients $3.830 > 1.9887$ and $0.00 < 0.05$), and it has a weak relationship (model summary 0.361).

k. Brand image and purchase satisfaction

It shows that there is a relationship between brand image and purchase satisfaction (coefficients $8.187 > 1.9887$ and $0.00 < 0.05$), and it has a strong relationship (model summary 0.637).

l. Quality Service and purchase satisfaction

It shows that there is a relationship between service quality and alternative evaluation dimension (coefficients $5.919 > 1.9887$ and $0.00 < 0.05$), and the strength of relationship is sufficient (model summary 0.513).

m. Marketing Communication and purchase decision

It shows that there is a relationship between the variable of marketing communication and purchase satisfaction dimension (coefficients $4.820 > 1.9887$ and $0.00 < 0.05$), and the strength of relationship is sufficient (model summary 0.438).

Discussions and Research Analysis:-

Visitor perception to the MAG brand image:

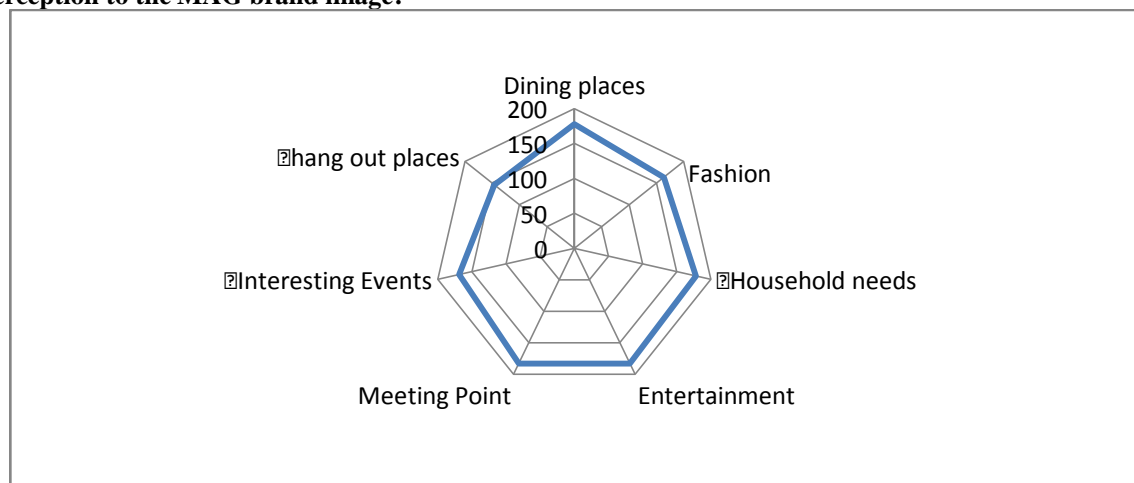


Figure 3:- Brand Association of MAG

Based on Figure 3, the perceptions of visitors to the MAG position as a shopping center in North Jakarta are as follows:

- For the top position, MAG is a mall with excellent entertainments (183). It is one of strong points where MAG has an "Amazon" children ride, which is considered as the biggest game zone located on the ground floor, first floor and second floor. Other supporting aspects are the provision of NAV Karaoke, Bowling center with 30 Lines, cinema XXI, and others.

- b. MAG is in the second place as a meeting point (183) since it is located in a very strategic place, in front of the main road of Yos Sudarso that has an easy direct access to toll road, particularly for private cars. For those who take public transport also can get off just right in front of mall. In addition, MAG provides various coffee shops such as Starbucks, Coffee Bean, Bangi Kopitiam, J'Co Coffee, Choco Corner, and many more.
- c. As the third place of visitor perceptions, MAG is a mall providing complete household stores (178). Some of them are ACE Home Center and Informa Furnishing, which are regarded as the biggest equipment and household stores. Furthermore, Home Furnishing is the most complete furnishing items in Jakarta occupying all over the third floor. The presence of Diamond, as the most popular supermarket, equips the shopping needs visited by housewives around Kelapa Gading and the surrounding community.
- d. Meanwhile, in the fourth place, MAG is a mall that has a fine dining place (178). This is supported by the availability of a variety of branded restaurants and coffee shops, like; Marugame Udon, Ci Wei, Ta Wan, Red Bean, Warung Tekko, Sate Khas Senayan, Imperial Lamien, XO Suki, Pizza Hut, KFC, A & W, Singapore Tau Fu, Batam Fish Soup, Fiesta Steak, etc. As for the coffee shops, among others are Starbucks, Bangi Kopitiam, Bread Talk, J'Co, Beard Papa, Dunkin Donuts, chattime and Choco Corner.
- e. For the brand association, MAG lacks interesting events (169) and less complete branded fashions (163). So this issue needs to get more attention if MAG has a willing to improve its brand image.
- f. As for the lowest brand association that needs serious attention to improve brand image is that MAG is necessary to make a trendy hang out place for young people (146).
- g.

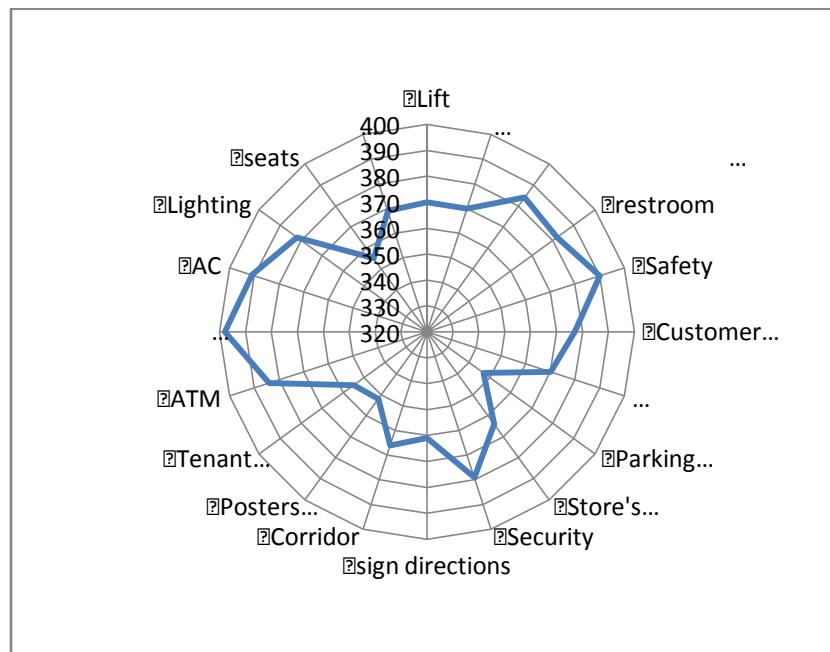


Figure 4:- Perceptual Mapping Image of MAG

Based on Figure 4, it can be revealed that visitors generally have a somewhat good image on MAG in spite of the fact that there are still some lacking points that need serious attention and improvement to enhance the brand image of MAG.

The analysis on perceptual mapping image:

- a. The highest visitor Image is on cleanliness (398), which is in line with the research finding that the janitors' quality services rank above average position.
- b. The second image is the comfort of mall and excellent security (390). Based on data from the operational department of MAG that crime rate decreases for 16% such as theft, and fraud. However, it still lacks the security officer services to visitors. That's why it needs more attention and improvement.
- c. Restrooms' condition (382), lighting (382), escalators' conditions (384), and availability of ATMs (384) rank third.
- d. The following points that need to be addressed and improvement are the decoration of the mall (369) that seems monotonous. The elevators (lift capsules) are always full because the location of other elevators (north and south elevators) is quite far from the atrium. Parking space is less convenient due to lack of parking capacity.

(370) especially during weekends and holidays. The last one is redemption counter (370) that is located in the distance.

- e. Next, the things that should be a priority for immediate improvements, such as the arrangement of the corridor (366) which seems rather chaotic, low store employees' services (364), and lack of sign directions (361), tenant directory (355) and posters publicizing the events (352).
- f. The last one is the most urgent thing that needs to be done, namely, the less availability of seats (355) inside the mall causing discomfort which affects the image and perception; and less responsive of parking attendant services.

The effect analysis of brand image, service quality and marketing communication on the stages of purchase decision at MAG.

- a. The purchase decision process begins when a prospective buyer is aware of the need, which is due to internal and external stimuli. At this stage, a brand image, service quality and marketing communication have not had any effect on a potential buyer. This is consistent with the research findings about the correlation of brand image, service quality and marketing communication and the problem identification dimension. It shows that there is no relationship between three variables and the dimension of problem identification.
- b. At this stage, a consumer, who is driven by needs might look for further information. Therefore, the most influential variable for a person to look for information is a marketing communication variable. If we look closely at the correlation matrix table among six dimensions, marketing public relation is the most influential variable to attract visitors in the stage of information search. While the variable of service quality has no effect on the visitor. This might be because the service quality of a mall does not have any effect on a person's need.
- c. At a later stage, a consumer will process all information about the selection of the brand to make the final decision where the consumer will give different weight to each attribute of the product in accordance with his/her interests. In this stage, brand image is the most influential variable in making alternative choices. Before making decisions, normally the consumer will pay attention thoroughly, efficiently and effectively in selecting the desired product. And the most decisive factors are brand image and today's trend. The service quality ranks second place and marketing communication ranks the last, this is because a person is not too affected by a marketing communication in this stage.
- d. The fourth stage as the evaluation stage is the execution of a purchase where consumers lists brands in the set selection and do purchase intentions. In the execution of the purchase phase, a consumer would select preferred brands. On the other word, brand image becomes the most affecting variable in the execution of purchase. Indeed there are other affecting factors such as attitudes and unforeseen factors. However, in this study, the variable of brand image has the highest impact.
- e. After the purchase of a product, the consumer will experience some degree of satisfaction or dissatisfaction. In this stage, brand image becomes the most influential variable on the purchase satisfaction as the goods have been tested and have proven quality. Brands can have a high value due to brand building activity that is not only based on the communication but it is also all sorts of other efforts to strengthen the brand. Of communication, the brand can promise something and can signal something (brand signaling) too.

Conclusion:-

- a. In general brand awareness of Mal Artha Gading is good, and its association has started to appear. This can be seen from the brand image of the mall as the most influential variable on purchase decision of customers. To build a strong brand image MAG provides various facilities, like fashion, entertainment, children playground, meeting point and sport centres; this aims to build an image of MAG as a mall for family. The mall also provides high class service so that an image of high class, sociable, comfortable, and safe mall is created; this will form perception of the society and its visitors toward the mall that it is the main choice to do their shopping dan to fulfill their daily need. Whereas there are some aspects including event, brand fashion add, trendy hangout places among today's young generation, which still need improvement.
- b. Service quality has the lowest influence on purchase decision in Mal Artha Gading. This is a result of the quality of its employees who do not meet the needs of the visitors
- c. Marketing communication applied by the management of Mal Artha Gading influences the improvement of mall awareness of people in Jakarta. This influences the purchase decision of the customers. The research result shows that the use social media, internet infrastructure, and mobile application in Mal Artha Gading is effective in informing activities done in the mall. However, the use of advertising, billboard, and promotion media has not been maximum. As a result, the impact of advertising the events is not significant.

- d. Brand image and marketing communication influence the purchase decision simultaneously. Meanwhile, the other variable, service quality, influences partially, and it is not significant.
- e. This research is considered to be valuable for the management of MAG since by referring to each influential variable the most important dimension and the less influential indicator can be found. As a result, the management can make a required policy for increasing number of visitors.
- f. The strength of this research is that it is able to reveal the most important dimension or indicator to be improved so that the number of visitors can increase. Referring to the finding of the research, some substantial and valuable inputs for further theoretical and practical development can be made.

Recommendation:-

- 1) To improve the number of visitors, the service quality through the availability of alternative places for eating and meeting point is required. Then to increase its trendy image, regarding its customer buying power, to provide Korean and Japanese restaurant becoming trend and affordable brand is highly recommended.
- 2) Service quality of front liner needs improvement. Providing a special officers who actively approach the customers needing help can be implemented; this results in a more sociable and friendlier image of the mall.
- 3) For future research, variables including product quality, customer satisfaction, people, and other processes can be examined and analyzed. Besides, to obtain more accurate results and to reveal more findings, applying other analysis technique such as path analysis is suggested. This research is expected to help retails company and similar companies improve their sells in this fast changing business era.

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RESEARCH ARTICLE

MULTI-CHANNEL MEDIA ANNOTATION USING SOCIAL ACCESSIBILITY COMPUTING.

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Abstract

towards technology. Low vision persons make use of vision and other senses to understand and learn. Low vision means severe visual impairment; they cannot read the content at a normal viewing distance, even with the help of power glasses or with lenses, not with necessarily limited to distance vision. Carrying out of their daily activities is often a major problem. Activities that are previously taken for granted, such as dressing, eating, writing, travelling and simple communications or interacting with other people are hampered. One of the mobile applications was proposed to facilitate them, to identify and tell the things or information through voice by a social networking people using photos, videos or by sending portable document format [pdf] image which is to be uploaded by the low vision people. This will help them to identify things which are before them without interacting with their neighboring people so that they can proceed in a hopeful manner. This paper was proposed with a mobile application developed for low vision people.

Today, we are in modern world;
it is changing at neck speed

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Introduction:-

Internet and mobile computing bring much convenience to most people. Some may have a wrong opinion that persons with disabilities, those visually impaired or low vision, are unable to use mobile devices and mobile applications. Mobile application developers are not aware of the disability groups in using mobile devices of their special needs. Persons with disabilities are using touch screen mobile devices nowadays. Mobile devices and applications, Information and Communications Technology are used by visual impairment people anytime and anywhere more effectively, live more independently and participate fully in the society.

Accessibility of Mobile Applications:-

A high responsibility was given for mobile communications for doing businesses and for delivering a wide range of applications. So making accessible applications in mobile not only benefits the people with disabilities but also fulfills legal understanding and responsibilities, building corporate image and widening customer base. For enterprises and organizations opportunities opens to develop accessible applications on mobile platform in order to develop the fast growing digital economy and harness the technology innovation. It is necessary for mobile

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application developers that there is need and importance of accessibility requirements of mobile applications with the different needs of different segments of the community. There are two types of accessibility in mobile apps

- Automated. E.g.: using algorithms.
- Social network. E.g.: involves humans.

A. *Mobile Applications*

Mobile applications are of 3 different types which are as follows

- A web app is launched by mobile browser and it was an internet based application.
- A native app can be downloaded it is a platform specific software and it can be used even though in offline.
- A hybrid app is a combination of both native app and web app.

Mobile applications are usually referred to native apps or hybrid apps which require downloading and installation on a mobile device

Literature Survey:-

Low vision is ubiquitous problem for people having difficulty in viewing objects even with correct lenses. Persons with low vision use mainstream computing devices, to access information when accessibility tools provide sufficient support remains understudy [37] findings delivers the needs of low vision people and improved design opportunities for low vision accessibility tools. It is a major problem in our day to day society. Even then technology is always one step before and new inventions are found before we address the accessibility problems for low vision people. Addressing these issues is very difficult for making new inventions are mobile devices and tablets where there is no proper set of guidelines [21]. Independence and mobility are other important problems with people having visual impairment, they often face difficulties in their navigation [8] accessibility collaborative makes improvement to make web more adaptive to improve process with invited users [10]. Social Accessibility project is a new driven improvement for collaborative metadata authoring technologies [38] interface and applications are written by developers [1]. Several middleware models are proposed and graph sketching tool, blind screen reader, node-link diagrams are shared with sighted people [3] with a real time. 3D's and accessibility dimensions of the built environment [5] specifically, they highlighted that human computer interfaces at the time generally failed to take into account the needs of disabled users. Appliance reader broadly describes the hybrid approaches, which combines human and machine intelligence for interactive access technology [7]. An exploratory study describes the development of access lens hardware and software, the interactive design of access lens in collaboration with blind computer users[12] the characteristics of CS in context makes possible pathways in the field[13]. In addition to compatibility having screen readers, refreshable braille displays and magnification software is designed to low vision users [17]. The mobile cloud computing is becoming a dominant way for various mobile applications [19]. SIMON use ICT services to promote mobility impaired people in the context of on-street public parking areas and multiple transport modes [22] access to information by disabled people has been hampered by short sightedness on the part of computer and communication systems designers [23]. Information and communication technologies (ICTs) become diffuse, with developers and designers to consider users with disabilities and aging populations [27]. Front-end users with mobile devices and back-end cloud servers includes mobile cloud computing. This module helps to access a big volume of storage with distributed portable devices [29] research reveals creativity to CS as a pathway [30]. As ASSETS attendees, it is clearly promoting accessibility computing. Most such courses are aimed at upper-level students [31] Helping users with visual impairments understand the actual issues is a crucial and challenging topic, and will lead to improved accessibility[32] a combined Cloud-Internet of Things (IOT) paradigm provides scalable on-demand data storage and resilient computation power at the cloud side as well as anytime, anywhere[33] The need to study representative users is widely accepted within the human-computer interaction (HCI) community[34] present a case study with the evaluation of four e-government mobile applications.

EXISTING SYSTEM:-

Visual impairment:-

Persons with visual impairment who are having low vision, color blindness or color deficiency, are unable to see the screens and controls in mobile applications. Due to their visual constraints, they cannot see the buttons and use a touch screen to access and navigate the mobile application functions. Most persons with visual impairment rely on a screen reader to operate computers as well as mobile devices. A screen reader is software for converting information displayed on the device screen into speech or Braille on a Braille display. Some mobile device has gesture features with screen readers so that persons with visual impairment can hear functions and operate the functions through touch.

Accessibility features:-

- Screen reader – Nowadays, mobile devices have built in screen reader to facilitate the needs of people.
- Adjustable font sizes – it is used to enlarge the font size in mobile applications through built in feature.
- Screen magnifier – to enlarge the display screen.
- Adjustable brightness/contrast controls – This is used to increase the background color of the screen according to the individual needs.
- Backlit display – it is used to change the contrast of the screen in outdoors.
- Voice recognition –it is used to control the mobile devices through voice commands.

Accessibility Inspection Tools:-

Some of accessibility issues can be detected using inspection tools and/or simulators. It is recommended to test the mobile application with these tools during development stages as early as possible.

- Color Contrast Check
- WCAG Contrast checker (Firefox plugin)
- Mobile Speak for Windows Mobile (Third-party)
- Talkback for Android (Third-party)
- Voiceover for IOS (Built-in)

PROPOSED METHOD:-

This application is developed especially for low vision people to enhance information through voice by social network people through photos, videos or by sending **portable document format** [pdf] image by them. When this application is created for mobile phones, it is very useful for low vision people to identify the object which can't be identified by them. Developing android application for this challenging environment results in enhancement of low vision people for their virtual vision. This App is named as "SOMDVIC" which works for the low vision people. It works with text message along with voice.

How the app Works:-

- Open the app and touch the screen to recognize your voice for photo or video routine
- Touch to take photo
- Once again touch to send sms.
- Admin will receive a message with a link
- Through that link photo gets uploaded for view
- Admin replies a text
- User receives a voice message about that photo.
- Video gets opened at last to save the thing in that mobile phone.

Features

- Voice message
- Speech Recognition
- Photo capture
- Video capture
- Sending and receiving message through server.

Advantages over an existing system:-

- This app will facilitates the need of low vision people by sending the image of a thing.
- This app provides voice message for low vision people to be clear in their proceedings.
- This app created in Android mobile, will help low vision people anywhere in the surroundings to know their needs.

EXPERIMENT AND RESULT ANALYSIS:-*Implementation Tool:-*

It is an android based mobile application so one should have android mobile phone to use this application.

Least Hardware Requirement Environment**TABLE 1:-**

Hardware	Least requirements
System	Windows 7[64 or 32 bit]
Hard Disk	500 GB
Monitor	15 inch color
RAM	2 GB

Software Requirements:-

This Application is supported by android mobiles having lower version of API 8: ANDROID 2.2 [Froyo] to API 18: ANDROID 4.3 [JELLY BEAN].

Software used:-

adt-bundle-windows-x 86-20130917

The bundle comprises of eclipse android development tool with SDK manager build: v22.2.1-833290

Development tool:-

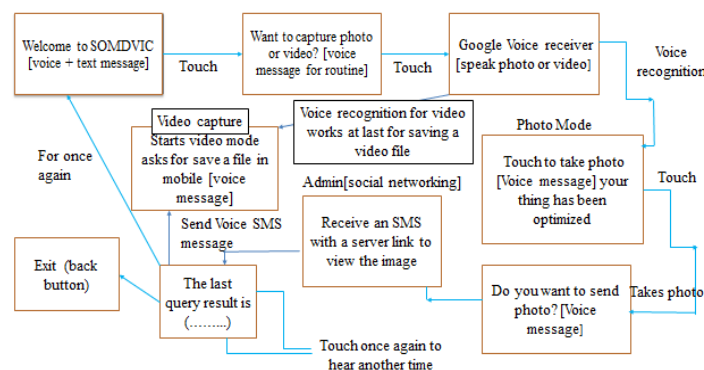
Eclipse android development tool with an android Emulator.

SOMDVIC Mobile application development:-

The SOMDVIC application is developed using eclipse android development kit. Code was made using java programming language. The user interface is created using XML language.

Server:-

Mobile application server consists of operating system and server hardware. These functions on side by side allow the server to remote access and provide services to apps, which contains security features, authentication and updates. In this, the mobile app server communicates with the client to enhance data and balancing load. It makes the admin to configure settings, make out updates and wipe the data's from current location. Because this app is a type of hybrid mobile application we can use any type of server side technology, like a classic web server technology (using JAVA, PHP or .NET) or some kind of cloud technology. Eg: Parse.com

**Fig. 1:-Overall Module****Server used for this App:-**

In this App, server is used as a kind of cloud technology for loading and storing of data. The Cloud Technology used in this App is Heptotechnologies.com

A. Modules

The modules involved in the design of this application are as follows

- Media Capture
 - Image Capture.
 - Video Capture.
- Admin
 - SMS activity [Sending and receiving SMS]
 - Server Link

Media Capture:-**Image Capture:**

This app is developed for low vision people to capture images hence photo mode is used in such a manner to take photo. In this photo module, the resolutions are set out in pixels to capture images in good looking manner. If the persons changes in different directions also it takes the image in pixel format. Touch makes to take photo.

Video Capture:

Video Capture is done in this app for saving things which are captured as an image. People having low vision are not able to see some views clearly so this app facilitates image but it is not saved in their app hence for future reference this app provides an option of video capture to capture videos. Video mode is carried out by voice recognition at the beginning of the application, at last it takes to this mode to save videos. Touch to start, end and save videos. If you don't want a video option, leave it as such don't speak when voice is recognized.

Admin Module:-**Sms activity:-**

After image capture is over, the image has to be send to the Admin mobile. The Admin is the social networking person who helps the low vision people to understand the image send by them through voice. After taking image, the person has to touch the screen to send sms to admin. Admin will receive the sms as an server link. Hence after viewing the image the admin will send the reply as an message. It will be received in this app through voice. Steps involved are

- Sends sms after the image was taken.
- Admin receives sms.
- Admin opens the image through server.
- Admin reply through sms.
- Low vision identify by voice message through this app.

Server link:-

As soon as the message is received by the admin, he/she views the message as a text with link when the admin touches the link it opens the cloud server link and displays the image. Cloud server is the one which stores data in its server. After viewing the image the admin came to know about the image. The admin will reply through sms to that application.

Procedure to use the app:-

This is the first viewing screen for SOMDVIC app, next touch the screen Figure 2.

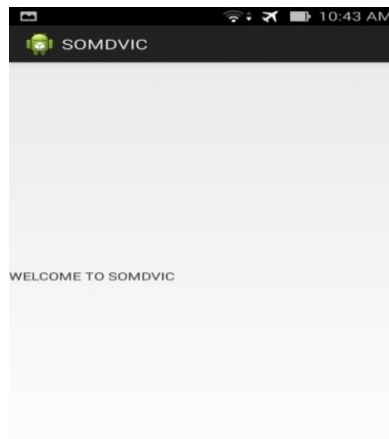


Fig. 2:- Application

It will show Google voice recognition to speak Figure 3. Next this Google voice recognition is used for finding photo or video mode.

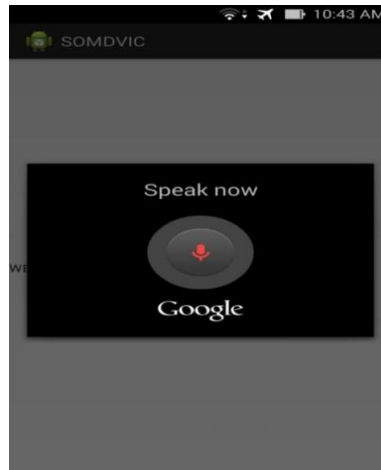


Fig. 3:- Google Voice Recognition

Sometimes it may fail also because of insufficient network facilities but it used for mode change in this application it is shown in Figure 4.

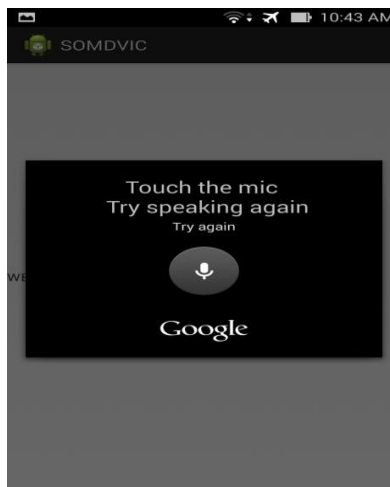


Fig. 4:- Google Voice fails to capture your voice

If you speak photo it takes you to the photo mode as shown in the figure 5.



Fig. 5:- Photo mode

If you speak video, it takes you to the video mode as shown in the figure 6.



Fig. 6:- Video mod.

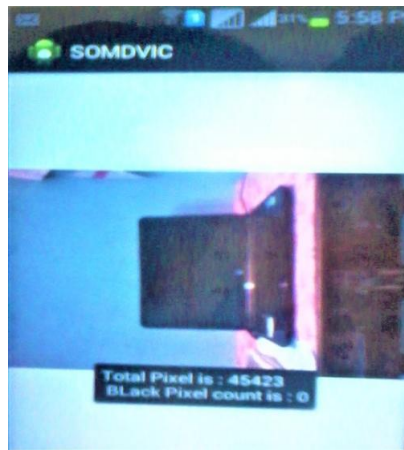


Fig. 6:- Laptop photo capture in this App

Now the app takes this laptop in the photo mode and again touches to send the SMS to server. The laptop is taken as a picture in photo mode and it was shown in the figure 6.

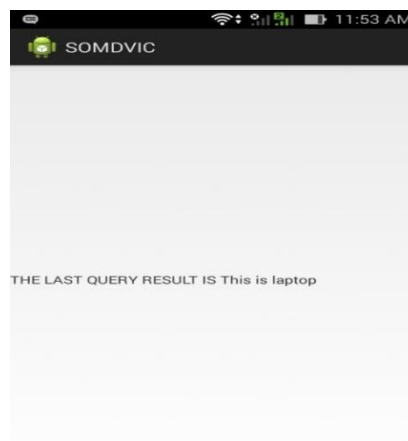


Fig. 7:- Server reply through App

Social networking people receive this message to give back the reply. If they type it is a lap it will be received by this SOMDVIC app and it is replied through voice in that app as shown in Figure 7.

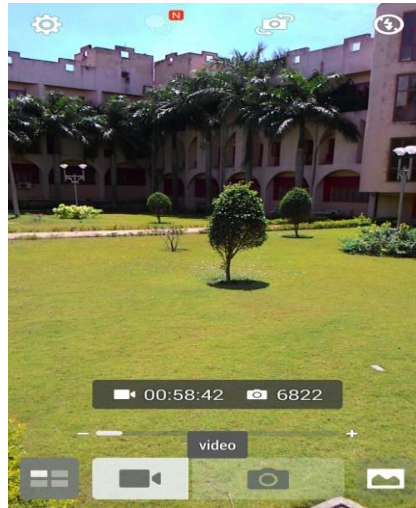


Fig. 8:- Saved video can be watched.

Video can be also be captured using this application and it was saved using options in this application and it can be viewed later or send it to server to get the information used in that video. The image used for video capture shown in Figure 8. We can watch the saved video.

Conclusion:-

It is difficult to adopt a technology for people with physical disabilities compared with normal people. The mobile app had been developed for low vision people across the world. Android applications are not very much common for low vision people, but they are using and accessing it in their daily activities to some extent. Many smartphones app has usability issues for low vision people. In this app, we evaluate the image of anything, through which low vision people came to know about the image for further enhancement process. This will help them to identify things which are before them without interacting with their neighboring people. This app was created as a guidance to increase the smartphone application for low vision people. The result of using this app, is better in terms of usability, persons have low vision find their routine daily works easily to accomplish it.

Future Work:-

This application was proposed, it provides a way for better application development to help low vision users to carry out their routine tasks efficiently and smoothly with the help of latest technologies. In Future, this app can be connected to what Sapp, twitter etc., to enlarge social networking environment for low vision people.

Acknowledgment:-

GAYATHRI.S received the degree in master of computer science from Pondicherry University in 2015. Currently, she is an M.Tech student at Pondicherry University. Her interests are in Advanced Mobile Technologies and AI.

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RESEARCH ARTICLE

TRIPLE LAYER SECURE ENCRYPTION: BY COMBINED RSA, IMAGE STEGANOGRAPHY & DIGITAL SIGNATURE.

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Cryptography, Steganography, RSA encryption, Digital Signature.

Abstract

With increase in use of Internet among public and availability of digital data sharing has taken industry professionals and researchers to give independent focus on information security. With growing internet users frequently required digital media store space to receive and send private information and this information requires protected against unauthorized attacks & access. In this paper I have presented a new technique for providing secure encryption to the user data. New proposed secure encryption will consist of a combination of RSA Encryption, Steganography along with digital signature.

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Introduction:-

Cryptography offers us the ability to transmit critical data between resources in a way that protect from third party reading it. Cryptography also provide authentication of someone and data. Following types of algorithms are used to specify cryptographic algorithms:

1. Secret key: Encryption and decryption done by single key.
2. Public Key: Two separate key are used for both process.
3. Hash Functions: A mathematical transformation to irreversibly encrypt information^{1,2}.

Steganography technique overlaps the existence of the secret data from an observer into a cover media. Three primary accessories are needed before performing steganography. First is information or data, Secondly Cover data medium and most important hiding technique. The cover medium can be a text file, an image, an audio file or video file but the most popular is the Image steganography. Primary objective of this technique is to avoid attention from the transmission of hidden information. If intruder detected any changes in the content or material, then the objective that has been processed to achieve the security level of the secret data is at risk, intruder will anyhow try to get the hidden data inside the message.

Background:-

RSA Algorithm is designed by Ron Rivest, Adi Shamir, and Leonard Adleman in 1978 is represented in figure 1. It uses two big prime numbers randomly generated to define the public and private keys. The two different keys are used for encryption and decryption purpose. Sender, when the message gets transmit to receiver, encrypts the message using Receiver public key and then receiver can decrypt it using his own private key^{3,4}. RSA operations can be divided into three broad steps; key generation, encryption and decryption.

Key Generation⁵:-

1. Select two distinct large random prime numbers p & q such that $p \neq q$.

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2. Calculate: $n = p \times q$.
3. Calculate: $\phi(n) = (p-1)(q-1)$.
4. Select an integer e such that $1 < e < \phi(n)$
5. Calculate d to satisfy the congruence relation $d \times e = 1 \pmod{\phi(n)}$; d is kept as private key exponent.
6. Public key is (n, e) and private key is (n, d) .

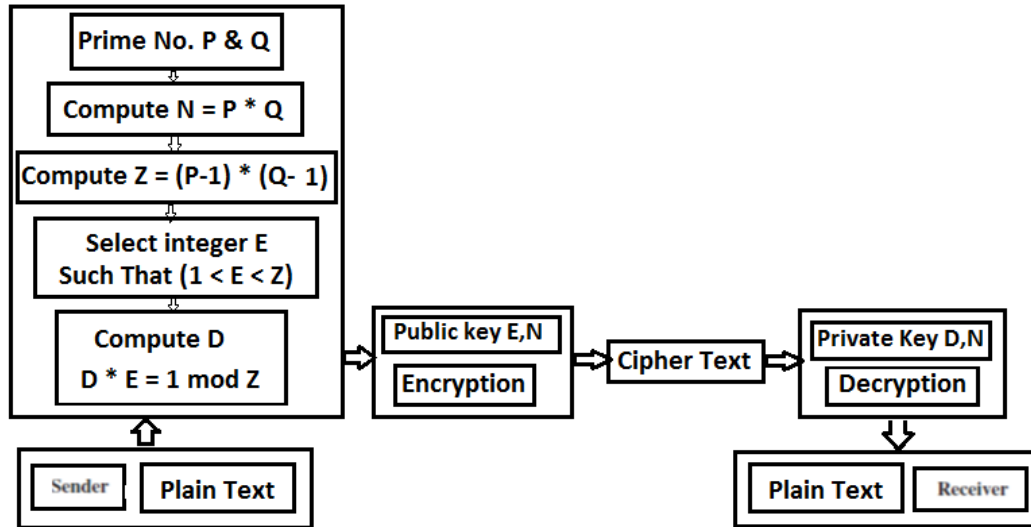


Figure 1:- Block Diagram for RSA

Encryption:Plain text: $P < n$ Cipher text: $C = P^e \pmod{n}$.**Decryption:**Cipher text: C Plaintext: $P = C^d \pmod{n}$.**Proposed Methodology:-**

Required robustness and security can be achieved; steganography & cryptography is combined along with additional digital signature that provide third layer of security. For cover media I will take Image for steganography and RSA algorithm will be used for encryption and is represented in figure 2. By combining, the data encryption will be done by RSA and then the cipher text will be used as input with Image media with the help of digital signature. The combination of these two methods along with digital signature will enhance the security of the data embedded.

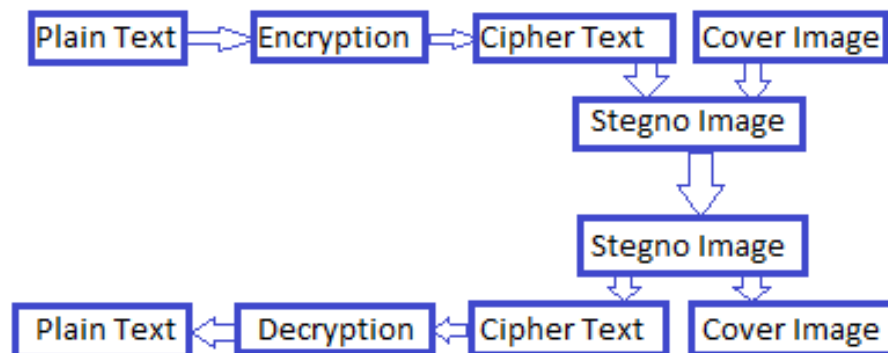


Figure 2:- Combination of Steganography and Cryptography

Working Architecture:-

A development of the complete proposed model is completed and the implementation of Encryption Process is also completed. As encryption is lifelong work to secure data. Day after day new and advanced techniques will be required to secure our data from hacker.

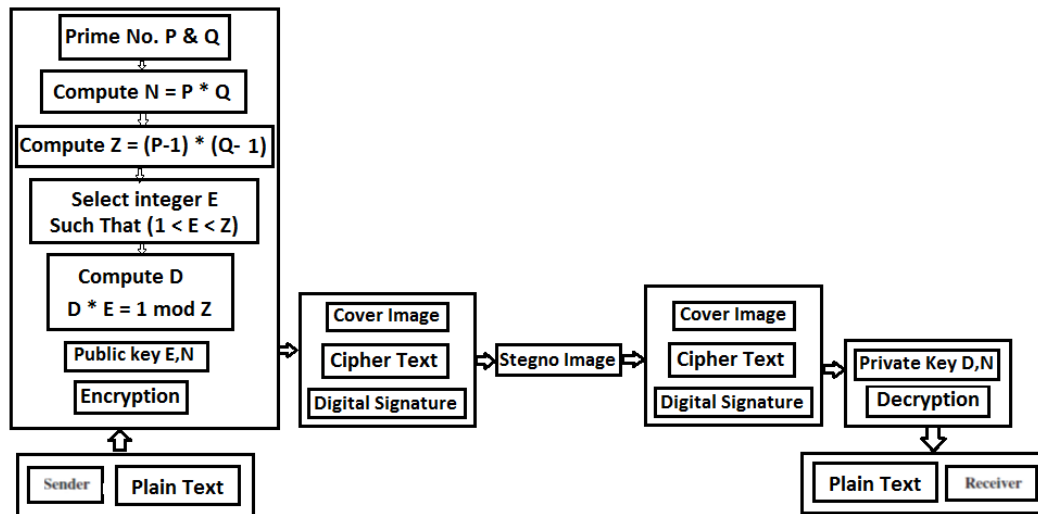


Figure 3:- Detailed Architecture for Proposed Technique

As details architecture is represented by in the figure 3. The Process is stated by converting plain text of uses into the cipher text by using RSA encryption process, Cipher text as plain text for Steganography, Cover Image required is input by user, Digital Signature that is of user choice. Here in implementation I have considered four – digit numeric value for digital signature and that is entered by user. Encryption Process is completed as per the detailed Architecture.

Results:-

According to the details architecture of the proposed new techniques shown in figure 3 I have developed new model. Figure 4 contains the complete encryption process. In Figure 4(a) contains the public & the private key randomly generated with the help of RSA encryption process and Figure 4(b) contains the data to be in secured. Figure 4(c) contains the cipher text that is obtains from RSA encryption process. Further Figure 4(d) contains the source of the cover media along with the last layer of security that is digital signature. And the final image Figure 4(e) contains the final message with the save new media that contains data to be hidden.

Decryption process is illustrated in figure 5. Figure 5(a) contains the source media that contains data along with the digital signature both value input by the user. Further figure 5(b) contains the cipher that is obtains from the step one of decryption process that separates cipher text from cover media with the help if digital signature.

Conclusion& Future Work:-

By this extra layers of security will be available to provide security to our document in today's words. If any how intruder is able to detect the text in the image. Then also that text is no more than cipher text and is also secured by digital signature. Thus triple Encryption is provided to the user data. Decryption process is not completed in the development of this model. My encryption process is working as per proposed model.

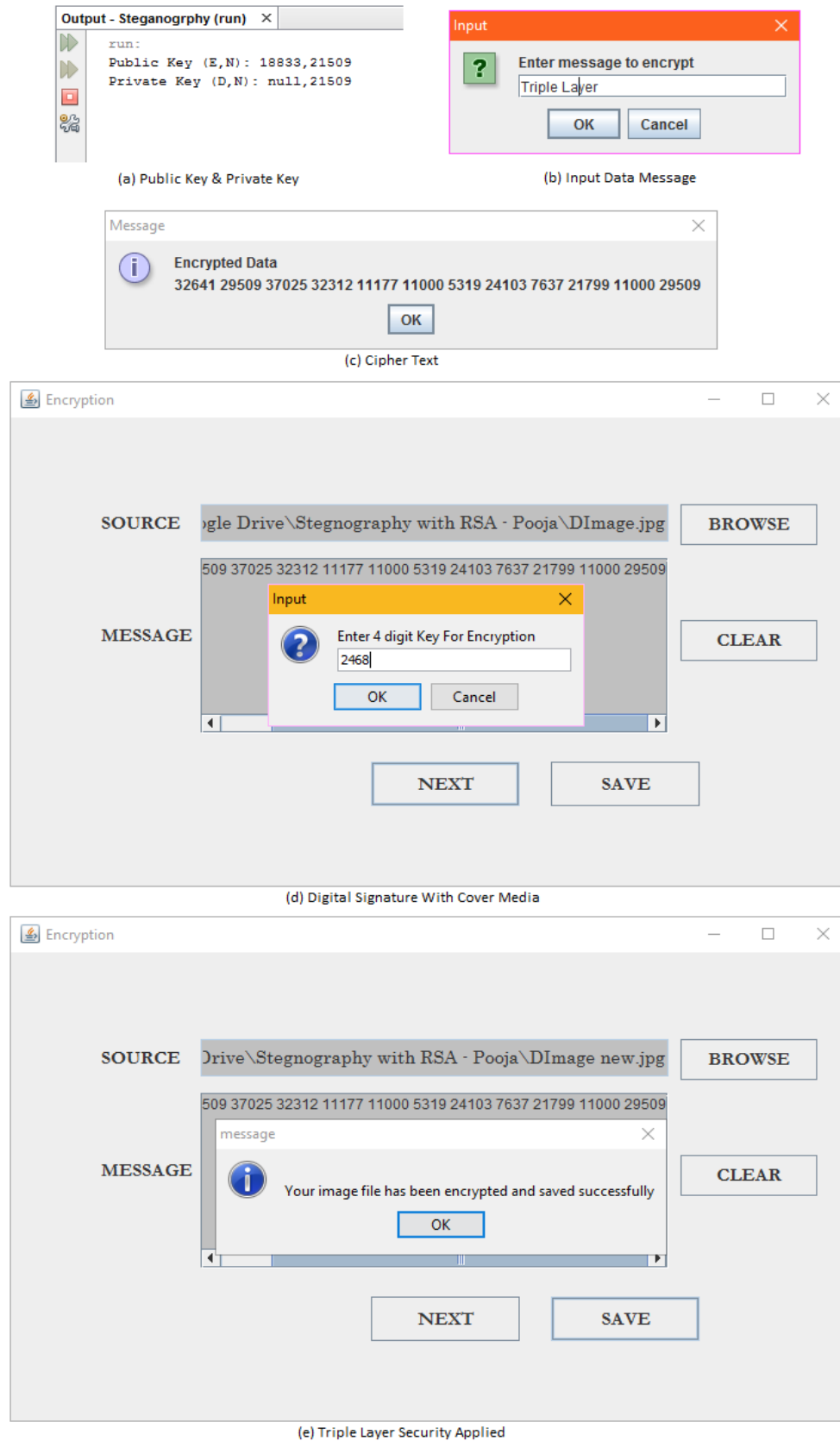
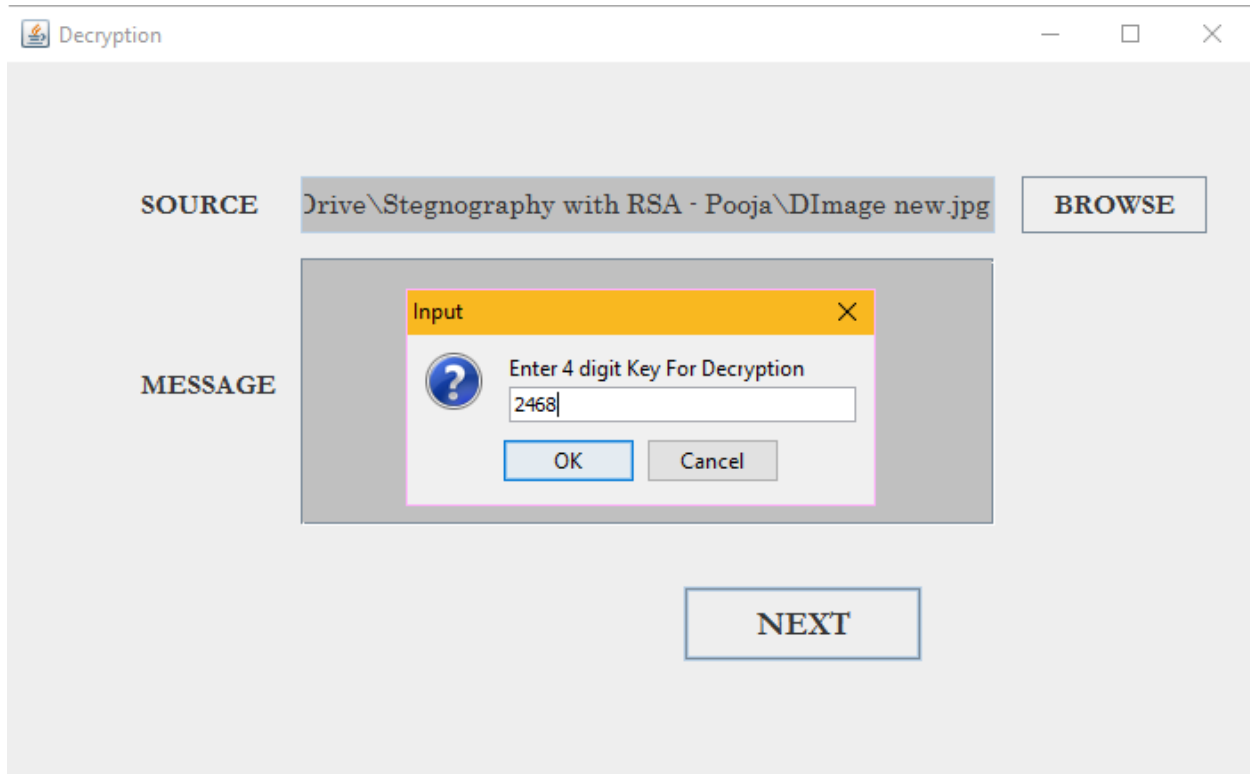
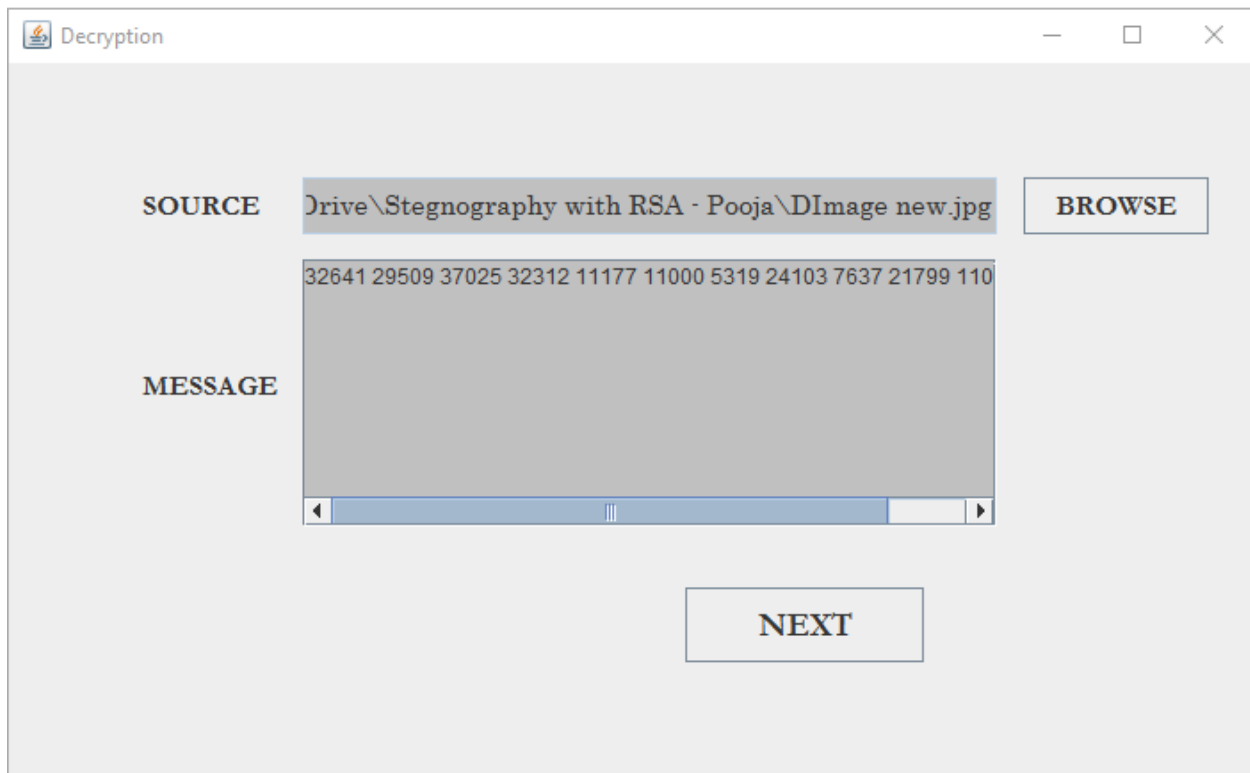


Figure 4:- Detailed description of encryption process of new proposed Technique



(a) Input Data Source with Digital Signature



(b) Cipher text

Figure 5:- Description of decryption process of new proposed Technique

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REVIEW ARTICLE

HAEMONCHUS CONTORTUS AND OVINE HOST: A RETROSPECTIVE REVIEW.

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parasite-environment interaction;
haemonchosis

Abstract

Gastrointestinal (GI) parasitic infections are a world-wide problem for both small- and large-scale farmers. Infection by GI parasites in ruminants, including sheep and goat can result in harsh economic losses in a variety of ways: reproductive inefficiency, decreased work capacity, involuntary culling, diminished food intake, poor animal growth rates and lower weight gains, treatment and management costs, and mortality in heavily parasitized animals. Among the GI parasites that cause losses to the farming industry, the barber's pole worm *Haemonchus contortus* is the predominant, blood-sucking, highly pathogenic, and economically important nematode that infects small ruminants. Here, we review the historical and recent literature on the ovine-parasite-environment interaction for *H. contortus* to bring avenues where advances in the understanding of these interactions is an indispensable to develop a cost effective control strategies as potential options for the haemonchosis control in sheep and the proper management of sheep in various production systems.

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History of Parasitology:-

The Egyptian Ebers papyrus of 1500 BC illustrates parasitic disease descriptions, which have been proven by the calcified helminth egg disclosure in mummies dating back to 1200 BC. Our earliest ancestors could be knowledgeable of the larger species, such as the roundworm *Ascaris* and tapeworm *Taenia*, which can be observed with the naked eye [1].

In the 17th century, Robert Hooke invented the microscope and our life understanding and disease increased. Further understanding of the invisible, microscopic world has been occurred after the establishment of modern microbiology in the 19th century by Louis Pasteur and Robert Koch [2]. Thanks to modern technology, there have been nearly 300 species of parasitic helminths known to be human parasite. Additionally, livestock, crops and pets are all victims of parasitic helminths, which cause extreme effects on the human population as well [3].

Helminth Classification:-

Parasitic helminths are contained in the following groups: flatworms (platyhelminthes), namely cestodes (tapeworms) and trematodes (flukes), and roundworms or nemathelminths (nematodes). The name helminth was mainly used to stand for worms of the phyla platyhelminthes and nemathelminths; however, it has acquired an extensive sense, being now commonly employed for all worm parasites mentioned above. Below is a simple flow chart to aid in parasitic helminth classification of [4; 5](Fig. 1).

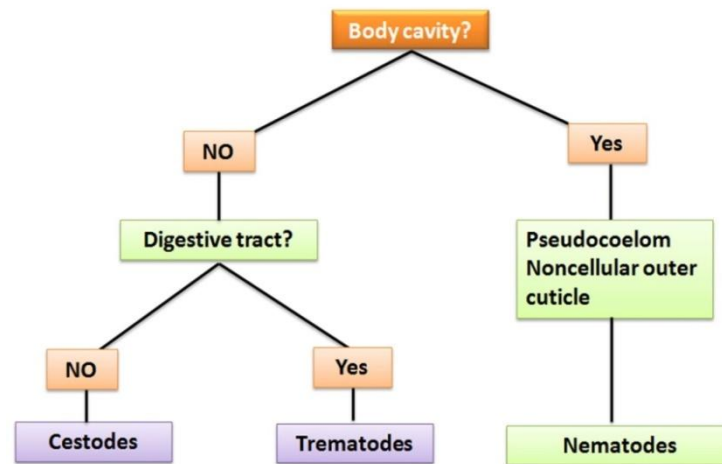


Figure 1:- A simple algorithm for the identification of parasitic helminths.

Nematodes are one of the most diverse groups of organisms on the planet (Fig. 2). Within the animal kingdom, nematodes are second only to the arthropods both in the species numbers and in the individual numbers present. Some are free-living, and many are parasitic, causing devastating diseases and socioeconomic problems world-wide [6].



Figure 2:- An approximate percentage of nematode genera in various habitats.

Gastrointestinal nematode (GIN):-

Livestock are an exceedingly valuable agricultural commodity in developing countries worldwide. They are raised under diverse husbandry systems ranging from large-scale intensive commercial programs to conventional small-holder and village production systems. When ruminants graze on natural pastures, which are common aspect of approximately all production systems, climate plays an essential role in the gastrointestinal nematode (GIN) transmission. GINs are dominant contributors to diminished yield and can reduce the meat, milk and wool production. All grazing ruminants are vulnerable to GIN infections at pasture and any future intensification of pasture-based systems will likely augment the risk of GIN diseases. The restricting environmental variable controlling the worm egg development to infective larvae is rainfall; because temperatures are always warm (i.e. tropical and subtropical regions) enough to facilitate this process. Therefore, there is a direct correlation between the harshness of GIN problems and rainfall in these regions during the wet periods of the year where livestock are raised in the developing countries. They have direct life-cycles with the following patterns: nematode eggs are passed out with faeces of affected animals. The nematode eggs develop into 3rd stage larvae on the pasture. These infective larvae migrate onto the surrounding herbage to facilitate larval pick-up and are ingestion by the grazing ruminants for the parasitic phase initiation of the life-cycle. The infective larval-stage numbers present in the host environment at any given period is related to nematode egg numbers passed by the grazing animal, and this widely determines the parasite numbers potentially established in a susceptible grazing host owing to the absence of pre-parasitic and their infective stage multiplication [7]. Moreover, some of these GINs have diverse development times and stages outside and inside the definitive host; knowledge of which is crucial for effective control measures. Nevertheless, factors such as age, breed, and nutritional status of the animal as well as characteristics of the environment also have a reasonable impact on the GINs and their capacity to infect and inflict deterioration to the grazing host [8; 9]. Grazing sheep are perpetually infected with a wide-range of nematode species, which have been the subject of several reviews [10; 11; 12; 13; 14]. The most important GINs found in sheep and goats are the tricho-strongylids *H. contortus*, *Trichostrongylus axei*, *T. colubriformis*, *T. vitrinus*, *T. capricola*, *Cooperia curticei*, *Nematodirus filicollis* and *N. spathiger*, the hookworm *Bunostomum trigonocephalum*, the strongylids *Oesophagostomum columbianum* and *Chabertia ovina*, the trichurids *Trichuris* spp. and the oxyurid *Skrjabinema ovis*. Moreover, it has been reported that the most important nematode parasite in small ruminants that are reared in warm climates is *H. contortus* [15; 16; 17], which is also important in countries with a temperate climate, including Sweden and Canada [18; 19]. One has to bear in mind that *H. contortus* and *Oe. columbianum* are of considerable clinical and economic importance on sheep and goat production. *H. contortus* and related species belong to a large order of GINs (Strongylida) of animals, including humans. Haemonchosis caused by *H. contortus* represents ~15% of all gastro-

intestinal diseases of small ruminants world-wide and results in extensive financial losses (<http://www.fao.org>). *H. contortus* is a blood-feeding worm that leads to anaemia and associated complications, causing death in extremely affected animals [20]. It causes diminished production and economic loss due to treatment costs and control measures [21; 22].

History of *H. contortus* nomenclature:-

H. contortus was first characterized in 1803 by Rudolphi [23]. Primarily, the parasite was termed *Strongylus contortus* [24] and It was however, not until the 1900s that *H. contortus* became the approved nomenclature [25; 26]. There have been different common names associated with *H. contortus*, including barber's pole worm, twisted stomach worm, and wire worm [27; 28; 29].

Taxonomy:-

Haemonchus contortus belongs to the family Trichostrongylidae as outlined below[30]:

Class: Secernentea

Subclass: Rhabditia

Order: Strongylida

Superfamily: Trichostrongyloidea

Family: Trichostrongylidae

Scientific name: *Haemonchus contortus*

Common name: Barber pole worm

Haemonchus contortus life cycle:-

H. contortus is an abomasal [31] and compartment 3 (C-3) blood feeding nematode of small ruminants (a four-chambered stomach/sheep) and pseudo-ruminants (a three-chambered stomach/ camelids) [32], respectively. The common parasite name is the barber-pole worm on account of the white reproductive tract is wrapped around the red blood-filled intestine give a twisted or barberpole appearance [33](Fig. 3).



Figure 3:- *H. contortus* showing barber-pole appearance with white ovaries twisted around red, blood-filled intestine (A), and male and female *H. contortus* adult recovered after necropsy (B).

As shown in Figure 4, the parasite has a small buccal cavity with a single tooth, called the lancet, which is made of cuticle and is used for feeding by slicing the ovine mucosa [31].

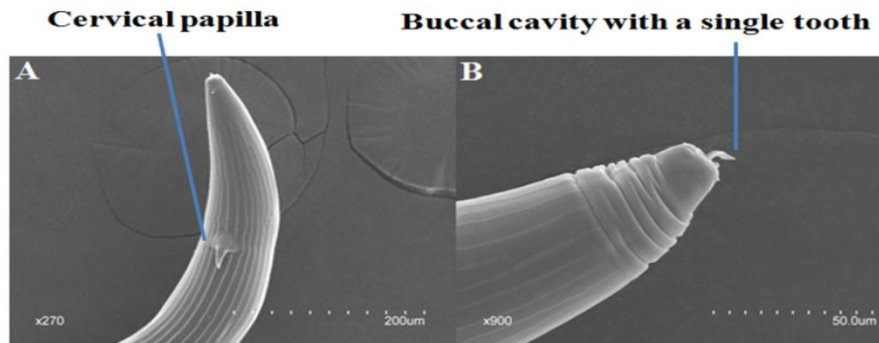


Figure 4:- Scanning electron micrographs (SEMs) of *H. contortus* -anterior end with cervical papilla and lancet tooth, which is used to initiate blood flow for feeding.

H. contortus has separate sexes and adults can be visualized with the naked eye. Adult females are 20 to 30 mm in length, tapered at both ends, and usually have a vulvar flap. Males are 10 to 20 mm in length and are tapered at the anterior end. The posterior end of the male has a copulatory bursa and spicules with a barb at the end used to hold open the female's genital opening during mating[31]. The adult males possess a peculiar three-lobed copulatory bursa involving two symmetrical lateral lobes and one asymmetrical dorsal lobe [29]. The *H. contortus* life cycle is direct, and the prepatent period is 16-21 days in small ruminants. The life cycle pattern can be categorized into four stages; these are: the parasitic stage (the interaction between the sheep and the parasite); the contamination stage (eggs are passed in the faeces during defecation); the free-living stage (larval-stages develop and survive); and the infection stage (infective larvae are consumed during grazing). Adult worms only live for a few months and reproduce sexually in the host. Females may lay thousands of eggs per day that pass out in the host faeces. *H. contortus* egg are regular, large, ellipsoidal, slightly flattened at the poles and morula not fully filled cavity of the egg with an average size range of 76-81 x 44-46 µm [34; 35]. Under ideal conditions (temperature:31°C-34°C and relative humidity: above 85%), larvae hatch from eggs in the faeces [36]. As mentioned above, the first-stage larvae (L_1) develop further and molt to become the second-stage larvae (L_2), which occur within the manure where the larvae feed on bacteria. After a second molt, larvae become the infective third-stage (L_3) about 5 days after passing out in the faeces. These larvae maintain the cuticle of the second stage as a protective sheath, which protects the L_3 from harsh environmental conditions and prevents it from feeding [37] (Figure 5).

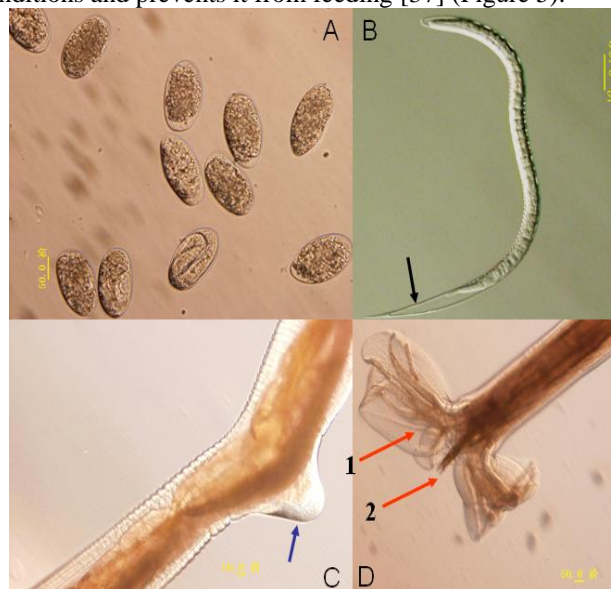


Figure 5:- *H. contortus* life-cycle stages [38].

(A) *H. contortus* egg. (B) *H. contortus* L3 larva. Slender larva, tail sheath medium length ($> 50\mu\text{m} < 100\mu\text{m}$), tapering to point and often kinked. (C) *H. contortus* adult female vulval flap. The blue arrow is pointing to the vulval flap. (D) *H. contortus* adult male bursa. The red arrows are pointing to the capulatory bursa (1) spicules used to hold open the female worms genital opening (2).

Infective L₃s rely on stored energy as they travel from the faecal mass to the vegetation to be swallowed by sheep [33]. In supportive warm and humid climates, transmission can occur year round. L₃s can survive in the pasture up to one year under these conditions. Nevertheless, survival is usually weeks to a few months in cold climates. Infective L₃s undergo an exsheathment process in the forestomach and the exsheathed L₃s penetrate the abomasal mucosa and molt to the fourth-stage larvae (L₄), which return to the abomasal lumen and develop to dioecious haematophagous adults within ~3 weeks following the ingestion of L₃s [39]. The L₄s are distributed widely in the anterior fundic area and are almost completely absent in the pyloric region; however, adult worms are distributed in the posterior fundic region of the abomasum closer to the pyloric area, which may be a selection by adult worms of a more supportive feeding site, or an avoidance of the host reaction in the area parasitized by earlier larval-stages [40]. Between 9 and 12 days after infection, the L₄s migrates back out into the lumen and molt one final time to the pre-adult stage (L₅s) under normal development conditions), which then develop into the reproductive mature adult to finalize the life cycle.

Hypobiosis:-

Haemonchus, *Trichostrongylus* and *Teladorsagia* have developed an extraordinary adaptive mechanism, referred to as hypobiosis or arrested larval development, to survive during harsh environmental conditions and ensure persistence (winter cold or summer heat)[41]. Hypobiosis [42] can be defined as “the temporary cessation of development of nematodes at a precise point in early parasitic development, where such an interruption contains a facultative element, occurring only in certain hosts, certain circumstances, or at certain times of the year and often affecting only a portion of the worms.” There are two types of arrested development that occur in GIN parasites. The first is designated immune-mediated arrest (non-specific). It is elicited by either host-related or parasite-related factors at any time of the year. The second is designated seasonally induced arrest and occurs at the same time each year. This type of hypobiosis is similar to diapause in insects and elicited by an external environmental stimulus [43]. The latter seems to be the prevailing form of hypobiosis in *H. contortus* [44]. Factors that may act separately or in combination with each other to provoke hypobiotic L₄s include: a) host-related factors, such as host resistance, acquired immunity, and age [45]; b) parasite-related factors, including population density and genetic predisposition[46]; and c) environmental factors, such as temperature, humidity, and photoperiod length. Environmental factors are most likely to be the triggers involved because of hypobiotic L₄s appear to be more seasonal than immunological [46; 47]. Haemonchine hypobiosis is similar to the arrested development phenomenon (diapause) in insects. Diapause is an inhibition of development induced by environmental factors and considered to be genetically controlled. This arrest in development is temporarily irreversible and may continue until either a specific stimulus presents itself or a predetermined period of time has elapsed [48]. The L₄s of *H. contortus* is able to undergo a state of arrested development [49]. They remain in the abomasal mucosa in a dormant (metabolically inactive) for 3 to 4 months, at which time they resume development. The end of hypobiosis coincides with changes in the weather, which causes the environment to once again become conducive to development and survival of free-living parasitic stages. Hypobiosis is beneficial to the *Haemonchus* infection as it is a means of delaying egg production until the external environment is likely to permit larval development in spring, or when moist conditions return; therefore, it is a forceful element in the condition known as spring rise in egg counts. The resumed development of the hypobiotic larvae often results in the events known as “spring rise” and “periparturient rise.” A sudden pronounced surge in nematode egg counts is the characteristic features of these events as a result of the arrested larvae massively reach the reproductive adult stage [50]. Spring rise occurs during the spring and although it is commonly associated with parturition, it is also seen in non-reproducing hosts [51]. On the other hand, periparturient rise harmonize with parturition during the spring lambing/kidding season but may also occur at other times of the year.

Spring lambing (i.e. ewes typically breed in fall and lamb in spring) occurs concurrently with the spring rise (periparturient rise/first rise). Therefore, the rise in the population of L₃s concurs with the augmented availability of susceptible neonates, thus ensuring transmission. During winter months, most L₃s on pasture will die and adult worms inside of the host will senesce, leaving the only surviving parasites as hypobiotic L₄s, which contribute to parasite burdens in the spring. Peri-parturient ewes will begin to produce eggs in faeces. About one month after introduction to pasture, the ewes, and more importantly lambs will begin to have high fecal egg output as a result of hypobiotic larvae emerging from the ewes and acting as a source of infection for the lambs [52]. Infected lambs can

in turn serve to establish a second rise in eggs and larvae in late summer and fall. The seasonality of *Haemonchus* parasite load (i.e. generally heavier following these peaks) provides intervention points for treatment of sheep flocks with anti-parasitic drugs during these crucial times. Environmental conditions, including season and climate, host immunity, and genetics have been implicated [53]. Furthermore, the photoperiod, temperature, humidity, and host immune system relaxation due to periparturition and/or lactation are the triggers for hypobiotic larval emergence. Immunity relaxation in late gestation or immunity suppression by reproductive endocrine activity are hypothesized to play a role in larval emergence from hypobiosis [54]. However, the larval development may also reinitiate spontaneously without the influence of a stimulus after a predetermined length of time [46; 47]. This aspect further exposes the resemblance between haemonchine hypobiosis and insect diapauses [55]. Winter hypobiosis of *H. contortus* has been reported in the northern U.S.A. as an obligatory survival mechanism [53]; however, adult worms would be likely to survive during the unfavorable dry season (November to March) as adults with no serious hypobiosis [56]. Hypobiotic parasites are more resistant to drugs intended for their destruction; nevertheless, macrolide anthelmintics are the only class anthelmintics that is effective against hypobiotic L₄s [41]. The anthelmintic resistance emergence in worm populations reduces the control level of hypobiotic L₄s during the extreme environmental condition. The short life cycle and the survivability of the larvae have implemented *H. contortus* to be a highly infective GIN parasite able to cause a reasonable amount of damage to an entire ovine population. Knowledge of the lifecycle and environmental conditions provide producers with a number of tools to manage GIN parasite infection in their lambs and ewes.

Pathogenesis:-

Haemonchosis is the disease associated with severe *H. contortus* infections of the ruminant abomasum world-wide [57]. Clinical signs of disease may become more frequent following these peaks owing to higher worm burdens. They include severe anemia, seen as pale mucous membranes, periorbital and submandibular edema resulting from hypoproteinemia caused by the *Haemonchus* bloodsucking activity, lethargy, emaciation, weakness, wool loss, and even death [58]. The adult worms feed on host blood and move from one feeding site to another, leaving behind wounds that continue to hemorrhage and resulting in anemia, which is the most common clinical sign. The volume of blood that is lost and consumed with a heavy worm burden may result in the host death [29]. The degree of anemia relies on the abomasal worm numbers. Clinically, haemonchosis can be categorized into three types; hyperacute, acute and chronic. The hyperacute form occurs in young and/or unhealthy lambs exposed over a short period of time to heavy infection (ingested a massive number of L₃s > 10,000), and is rare and results in lamb death [59]. The faecal color from these animals usually becomes dark due to digested blood and sudden death may occur owing to tremendous blood loss. Acute cases usually occur in young lambs that get heavily infected with or without diarrhea, but the host mounts an erythropoietic response resulting in the partial compensation for the blood loss. The parasite burden is mild, 1,000-10,000 individuals, and all ages of sheep are affected, regardless of present health status. Furthermore, anemia is accompanied by hypoproteinemia and edema, which may contribute to death. A common remark in these cases is sub-mandibular edema designated "bottle-jaw" [60]. The L₄s and L₅s larvae and adult worms are the robust blood-sucking GIN parasite; movement of the worm causes wounds and secretion of anti-coagulants resulting in a continuous haemorrhage from the abomasal wall [61]. A 2-month-old male lamb was identified with the history of anorexia, weight loss and dark color diarrheic faeces from a local farm in Jin Zhan village, Chaoyang, Beijing, China (Fig. 6). Clinical examination revealed severe emaciation, lateral recumbent position, and pale and anaemic mucous membranes. Parasitological examination showed numerous *H. contortus* eggs. The animal was died one week after treatment with with single dose of ivermectin (Shijiazhuang Fengqiang Animal Pharmaceutical Co., Ltd), and levamisole (Hebei New Century Pharmaceutical Co., Ltd). Post-mortem examination disclosed serious congestion, pinpoint petechial haemorrhages, and watery bloody contents with diverse minute hair like *H. contortus* worms in the abomasum. Gross lesions, such as petechial haemorrhages on account of the *Haemonchus* parasite attachment and feeding behavior, and severe congestion in the abomasal mucosa were corresponded with the earlier observations [62].



Figure 6:- Two-month old lamb died after treatment and post-mortem examination gross anatomy.

Bloated appearance face (bilateral periorbital oedema) and ovine hind-quarter is soiled with faeces due to diarrhea (A); Abomasal mucosa was severely congested; pin point petechial haemorrhages (B).

The possible pathogenic mechanism, which is responsible for cause of death in haemonchosis is hemorrhagic anaemia, hypoproteinemia and oedema due to vital blood sucking by both *L4s* and adults. Moreover, diarrhoea causes fluid loss and dehydration resulting in hypovolaemic shock [63]. Chronic haemonchosis is noticed in lambs infected with comparatively few worms (100-1,000 individuals), and distinguished by high morbidity and low mortality. Infected lambs are unthrifty, weak and emaciated with or without anemia relying upon the ovine erythropoietic status. Pregnant ewes and does have elevated faecal egg counts (FEC) around the parturition time known as the periparturient rise. Immune response against parasitic infection shortly before and after parturition appears to be compromised owing to reasons yet to be validated [64]. Haemonchosis early diagnosis is fundamental for the infected flock or herd treatment. Generally, clinical signs as well as history are essential for the diagnosis for the acute and chronic forms of this disease. Furthermore, faecal examination can also be conducted to confirm the diagnosis. However, necropsy and looking for adult worms in the deceased animal abomasum for the diagnosis of hyperacute haemonchosis [65].

Common diagnostic techniques-determining how "wormy" sheep are!

Many diagnostic techniques have been currently made available, particularly in the field of molecular biology, biochemistry and immunology. Nonetheless, they are still expensive and not applicable in conventional practice [66]. Parasitized animals can demonstrate several infection signs depending on the GIN parasites present. The general signs include rough hair coat, diarrhea, depression, weight loss, bottle jaw and anorexia. Laboratory diagnostic findings may involve anemia (low packed cell volume), elevated faecal egg count (FEC) and loss of plasma protein.

FAMACHA® eye color chart system

The FAMACHA card was named FAMACHA® after its originator Professor Francois 'Fafa' Malan's Chart. The FAMACHA chart, which depicts five illustrations of ocular membrane colors: 1: deep red (nonanemic), 2: red-pink (nonanemic), 3: pink (mild anemia), 4: white-pink (anemic), and 5: white (severely anemic) [67; 68]. The FAMACHA test is a sheeP- side test that allows approximation of the animal's PCV. It is performed by everting the lower lid of the eye and examining the color of the conjunctival mucosa (mucous membrane). Level of anemia can be roughly evaluated by observing the color of mucous membranes which are areas where there are a lot of capillaries (very small blood vessels) close to the surface so that tissue color reflects blood color. In general, pale or white membranes correlate with varying degrees of anemia, which correlate well with the burden of blood - sucking parasites (Fig. 7). These observations are part of a minimal database that will be an important aid in establishing the nature of disease. These measurements should be made repetitively and recorded, which over time will allow determination of improvement or exacerbation of the underlying condition. In settings in which anemia causing nematodes (mainly *H. contortus*) are predominant, blood PCV and FAMACHA score both are good indicators of the level of blood loss and associated problems. The FAMACHA © system is a selective program for managing haemonchosis in sheep and goats [69]. This system was developed in South Africa and allows owners and producers to rank an animal's level of anemia [70; 71]. The FAMACHA card depicts five colors from red (healthy) to white

(very anemic), which is then matched to the color of the inside of the lower eyelid of the affected animal. The score correlates well with PCV and can be used easily in the field [70]. The FAMACHA © system works by scoring animals on a scale of A to E and is performed by looking at the conjunctival membranes. A score of A or B is indicated as normal, while a score of D or E is anemic. The membranes of the eye are very pale at a score of E. It has been shown to be an effective way of determining, which animals need deworming for haemonchosis [69; 70; 72]. Employing this method diminishes the necessity to deworm all animals in a herd [71]. It is best to establish routine parasite evaluations, including FEC, PCV, FAMACHA, or weight gain at specified intervals (mostly 2 to 4 weeks, depending on expected severity of problems) from parturition throughout the grazing season. Because *Haemonchus* is the only parasite monitored with FAMACHA, attention should be paid to identifying and controlling other parasites when needed (e.g., by faecal egg count [FEC] or fecal culture). Recently, strong correlation among FAMACHA© eye scoring, PCV, and faecal culture positivity has been observed. Additionally, the FAMACHA© anaemia scoring guide can be employed to treat only severely anemic sheep reducing mass treatment and chance of drug resistance[73].



Figure 7:- FAMACHA© eye color chart being used to check level of anemia.

In this sheep the conjunctiva of the eye is being evaluated for color (A), on a scale of A to E (with 1 = normally pink and 5 = very pale) (B).

Body condition scores:-

Determination of body condition score (BCS) is an effective tool for managing both individual animals and herds. In an individual animal, low BCS may indicate disease or poor access to feed. In a flock or herd, a trend toward low BCSs may be indicative of inadequate feed quantity or quality or of management-related diseases, for example internal parasitism. Hands-on examination can be conducted to determine BCSs. BCSs were measured by palpating (physical feeling) the level of muscling and fat deposition over and around the vertebrae in the loin area behind the ribs and in front of the pelvis, and assigning a score of A-E, where a score of A is considered emaciated and E is considered grossly obese [74] (Fig. 8; Fig. 9; Table 1). It has been reported that changing in the body scores are good indicators of the intensity of GIN infection in the Nigerian WAD sheep [75].

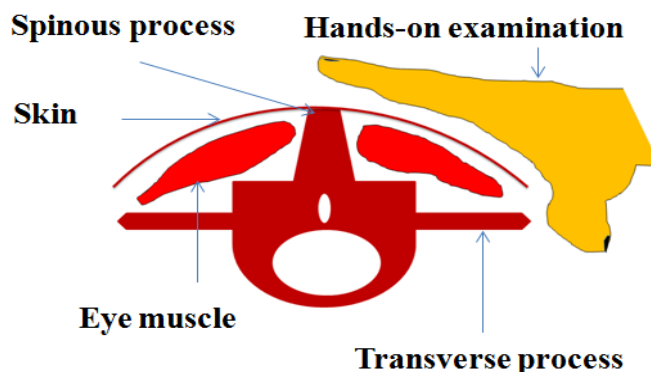
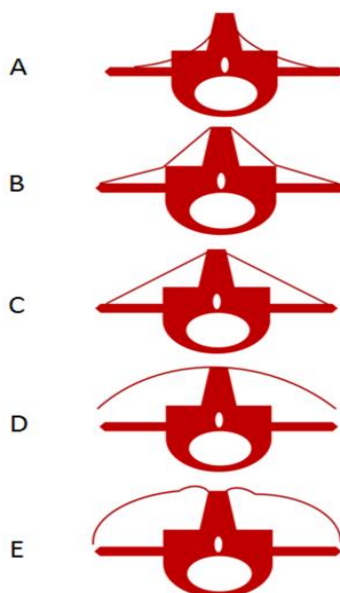


Figure 8:- Assessing body condition score by physical feeling.

Table 1:- Body condition scoring in sheep [76; 77; 78].

Assigned score	Condition	Physical Finding			
		Spinous processes	Transverse processes	Loin eye muscle	Fat cover over loin eye muscle
Condition A	Emaciated	Sharp and prominent	Sharp	Shallow	None
Condition B	Thin	Sharp and prominent	Smooth, slightly rounded	Medium depth	Little
Condition C	Ideal	Smooth and rounded	Smooth, well covered	Full	Medium
Condition D	Fat	Palpable as firm line with pressure	Not palpable	Full	Thick
Condition E	Overly fat	Not palpable	Not palpable	Very full	Very thick

**Figure 9:-** Body condition scores (BCSs) for sheep. These drawings demonstrate a cross-section through the lumbar region and characterize the fat covering.**Modified McMaster egg counting for nematode egg quantitation:-**

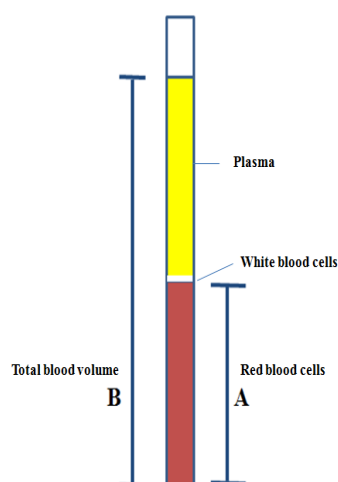
The coprological approach is the most extensively utilized method to detect helminth eggs and larvae, protozoan oocysts and cysts [79; 80; 81]. FEC plays an essential role in monitoring helminth burdens in herds and flocks, determining the pasture contamination degree and identifying anthelmintic resistance [81; 82; 83]. The McMaster method, which developed at the McMaster laboratory of the University of Sydney, is the most world-wide utilized FEC technique in veterinary and medical parasitology, and is supported by the World Association for the Advancement of Veterinary Parasitology for assessing the anthelmintic drug efficacy in ruminants [84], as well as for the anthelmintic resistance detection [82]. This method has improved diagnostic parasitology, both animal [85; 86; 87; 88; 89] and human [90; 91]. Several McMaster method modifications, including faecal weight and volume and flotation solution type, sample dilution, flotation times, an additional centrifugal application, centrifugal duration and speed, numbers of chambers of the McMaster slide counted and different coefficients for interpretation have introduced [88; 92; 93]. Three selected modifications of the McMaster counting technique, namely the McMaster method modified by Wetzel (W) and Zajčėk (Z), as well as the concentration McMaster technique according to Roepstorff and Nansen (R&N) have been chosen to compare their sensitivity and reliability by [94], however the method reported by Roepstorff and Nansen proved to be the more sensitive and reliable technique (Table 2).

Table 2:- Evaluation parameters of McMaster egg counting technique modifications [94].

	McMaster technique modifications		
	Wetzel (W)	Zajíček (Z)	Roepstorff and Nansen(R&N)
Amount of faeces (g)	2	1	4
Flotation solution type	NaCl	MgSO ₄ + Na ₂ S ₂ O ₃	NaCl + glucose
Solution specific gravity	1.2	1.28	1.3
Centrifugation (RPM)	None	2,000	1,200
Centrifugation time (min)	None	2/1	5
Flotation time in chamber (min)	2–3	5	3–5
McMaster counting chamber	3	2	2
Multiplication factor	67	33	20

Packed cell volume:-

The packed cell volume (hematocrit) is the ratio of the red blood cell volume (A) to the total blood volume (B) (Fig.10). PCV is an essential indicator of resistance of sheep to *Haemonchus* infection, and is negatively correlated with FEC in sheep (Shakya et al. 2011).

**Figure 10:-** Packed cell volume diagram**Immune response to GIN parasites:-****Ovine immunity:-**

It has been reported nearly 10-35% of neonatal lamb mortality within the first 6 months of life, a mortality rate nearly duplicated that of mature sheep [95] owing to their naïve immune system. Neonatal lambs are almost completely reliant upon passive transfer of maternal antibodies through colostrum in the first 3-4 months of life. Even though maternally-derived antibodies provide protection, they synchronously hamper the newborns from mounting their antibody response [96], due to this inhibition, newborns are not immunized against infectious diseases until they are at least 3-4 month old [97]. Neonatal lambs are especially vulnerable to infection when maternal antibodies are no longer present and have not yet begun to adequately mount their own immune response. The immune system consists of a wide range of diverse effector cells, tissues and organs with complex interactions that allow a rapid response to any foreign invaders [97]. The production of cytokines, and low weight signaling proteins involved in immune cell communication by activated CD4⁺ T helper cells in response to intracellular or extracellular pathogens resulting in the differentiation of naïve T helper lymphocytes into two subsets, T-helper type 1 (Th1) and type 2 (Th2) [98]. Th1-type cells are responsible for the cellular immunity activation, and a Th1-type response is generally noticed in response to viral infection, intracellular pathogens, and the delayed-type hypersensitivity activation. A Th1-type response usually responds to viruses and vaccinations, and is characterized by the cytokines IL-2, tumor necrosis factor (TNF), and interferon- γ (IFN γ) resulting in the mobilization of macrophages, T cells, and natural killer (NK) cells to promote proliferation and inflammation [99]. A Th2 response is distinguished by the elevated production of immunoglobulin (Ig) secretion by B cells, especially IgG and IgE and

the cytokine IL-4, as well as enlarged eosinophils and mast cell recruitment [100]. A Th2-type response is associated with allergic reactions [98], and is also activated upon parasitic infection [101]. There is a balance between the expression of both the Th1 and Th2 responses, even though challenges with diverse pathogens may modify the type and ratio of cytokines expressed, for example the Th2-type cytokine increase during parasitic infections.

Host response to *Haemonchus* infection:-

Inflammation during parasitic infection is necessary in larval and adult stage mucosal worm expulsion [99]. It has been reported that a clear Th2-type response in lambs repeatedly exposed to *H. contortus* [102], discriminated by Th2 cytokine surges, eosinophil, mast cell and globule leucocyte (intra-epithelial mast cell) recruitment, and parasite-specific IgA, IgG1 and IgE elevated production [103; 104]. Antibodies, lymphocytes, eosinophils, globule leucocytes and mast cells act upon larval stages of *H. contortus* embedded within the mucosa and adult worms residing the gastrointestinal tract lumen upon parasitic infection. Effector cells degranulate, releasing vasoactive molecules, which physically expel GINs from the abomasal mucosa [9; 99]. Eosinophils are closely associated with GIN infection, and activated by Th2 cytokines, such as IL-3 and IL-5 [105]. Sheep elicit an early immune response to infection with *H. contortus* larvae, represented by the CD4 T-cell and B-cell activation in the draining lymph nodes and eosinophil, CD4(+) and gamma delta-TCR, WC1(+) T-cell and B-cell recruitment. However, increases in mast cell numbers prevail the local response during infection with the adult parasite [105]. Mediators released by eosinophils include major basic protein, peroxidases, and neurotoxins, and have been demonstrated to have lethal effects against GIN parasites *in vitro* [106]. Mediators act as robust vasodilators, enhancing smooth muscle contraction and subsequent abomasal parasite expulsion [107]. It has been found that sheep with low egg counts had higher numbers of circulating eosinophils, which have been also been associated with larval migration inhibition [108]. Eosinophils and other microenvironmental factors, such as globule leucocytes and IL-4 can effectively kill GIN larvae *in vivo* [109]. Infection with *H. contortus* is associated with the production of mucosal mast cells and globule leukocytes within the GI tract of the sheep [110; 111]. Sheep that are more genetically resistant to parasites have more globule leukocytes than susceptible breeds [112]. Mucosal mast cells, recruited from bone marrow by Th2 cytokines, such as IL-4, histamine release, are eventually becoming globule leukocytes after degranulation [110]. Several studies have recorded the role of mast cell degranulation in worm expulsion [113; 114]; however, quantifying mast cell numbers upon necropsy is not a precise reflection of activation status and can therefore be ambiguous [115]. On the other hand, globule leukocyte quantification has been demonstrated to contribute a more accurate measure of mast cell activity [115].

Factors affecting host response to parasitic infection:-

Age is the most substantial factor in an animal's ability to effectively fight parasitic infection. The immune system develops with age; therefore, young lambs harbor the most serious *H. contortus* infections and account for the bulkiest commercial losses among producers as a result of diminished weight gain and death in extremely infected hosts [99]. The effects of haemonchosis are also more obvious in pregnant or lactating, undernourished, diseased or stressed animals due to a compromised or underdeveloped immune system [116; 117]. Genetic factors also play a role in an animal's resistance to parasitic infection. Several tropical and subtropical breeds of sheep are documented to have more natural resistance to GIN parasites. Resistant breeds include St. Croix [118], Florida Native [119; 120], and Gulf Coast Native [8]. Resistance in these breeds is defined as diminished establishment of larvae and their subsequent development into adult stages [121]. Nutritional status is a key component of the host immune response and the host's subsequent ability to mount an effective defense against GIN infection [122]. It is well-documented that animals on diets deficient in protein or key vitamins have a weakened capacity to combat parasitic pathogenesis, and concomitantly GINs significantly restrict the host's ability to efficiently exploit nutrients [123]. Lambs fed a diet high in protein harbor less severe GIN infections owing to heightened immune response. Furthermore, Immunoglobulin (Ig) A-mediated suppression of worm growth and fecundity is influenced by the quality of the diet [124]. Several studies have shown that supplementation with protein, energy [125], and trace elements [126] enhances host resistance and resilience to GIN infection. Resilience is another concept relating to the ability of the sheep to thrive in the presence of *Haemonchus* and reflects the host response to *Haemonchus* infection (Gray and Gill 1993).

Distribution and importance:-

H. contortus is distributed world-wide in tropical and subtropical regions with concentrations in the tropics and subtropics where there are high temperatures and a lot of rainfall. The parasite can also be found in more temperate areas, such as the United States. It dominates over other GINs, such as *Teladorsagia* and *Trichostrongylus* species in the southeast U.S because of the warm and humid environment. The environmental temperatures (31°C-34°C) are optimum for *H. contortus*; however, it can be existed in temperatures as low as 10°C and as high as 36°C [37; 127]. The hot and dry summer, and relatively cool winter conditions frequently constrain *H. contortus* development to short periods of the year in Mediterranean climates [128], diminishing the risk severity and duration. *H. contortus* is of relatively lesser importance in higher latitudes, owing to shorter and cooler summers, and more severe winters in studies, including Canada [129], Sweden [18], and South Dakota [130]. On the other hand, the existence of *H. contortus* in arid and semidesert areas is likely to witness its survival capacity (hypobiosis) and potential for rapid population expansion [131]. Nearly 60 different species of both domestic and wild ruminants have been identified as hosts for this GIN parasite [58]. Cross-host transmission is possible between wild and domestic hosts, for example white-tailed deer and domestic cattle and sheep [132]. It has a wide host range, including cattle, white-tailed deer, bison, antelope, giraffes, and camels; however, sheep and goats tend to be the favored hosts [133]. Domestic sheep appear to be the most exceptionally overwhelmed, which may be due to their grazing behaviors and strong flocking [58].

Ruminants (pregastric and postgastric microbial action):-

The gastrointestinal tract (GIT) of animals enables them to digest food, absorb nutrients and excrete waste products. The GIT nature is mainly determined by the food type. The stomach of carnivorous and omnivorous animals is relatively simple (i.e. hydrochloric acid and pepsin for primary digestion). However, the simple-stomach animals have a modification of the lower tract, the caecum, which is inhabited by microbes, whose function is to degrade cellulose. Some herbivores (hind-gut fermenters) have developed especially large caeca (e.g. the horse) or have other mechanisms for maximizing microbial intervention in the fiber digestion (e.g. coprophagy in rats and rabbits). The GIT of the ruminant animals has, in addition to the caecal (postgastric) fermentation, a large organ (i.e. rumen), inhabited by complex microbial populations, whose function is to degrade fibrous feed before it reaches the true stomach. The anatomy of the ruminant stomach is well-documented and the interested reader can find detailed descriptions in diverse specialized texts [134].

Only a brief and somewhat simplified description will be presented here. Ruminants are any of the diversified cud (fermented ingesta) chewing cloven-hoofed quadrupeds of the order *Artiodactyla*, obtain their food by browsing or grazing, such as cattle, sheep, goats, buffalo, deer, elk, giraffes and camels that usually have a stomach that are divided into three or four compartments. Ruminants are even-toed animal that regurgitates and masticates its food after swallowing (rumination). A diagram of feed conversion in simple stomached animals versus ruminants is presented in Figure 11. Unlike simple-stomach animals (monogastrics), such as poultry, ruminants have a digestive system designed to ferment foodstuffs and provide energy. True ruminants, including cattle, sheep, goats, deer, and antelope, characterized by their four-chambered stomach, namely the rumen, the reticulum, the omasum, and the abomasum as viewed from outside, in Figure 5, and "cud-chewing" behavior. The reticulo-rumen chambers have a wide variety of microorganisms that are essential for the digestion of plant cell walls and the production of volatile fatty acids (VFAs). On the other hand, the omasum compartment is characterized by the presence of many folds involved in the absorption of nutrients and water. The abomasum is the "true stomach" of a ruminant, and it is comparable to the non-ruminant stomach.

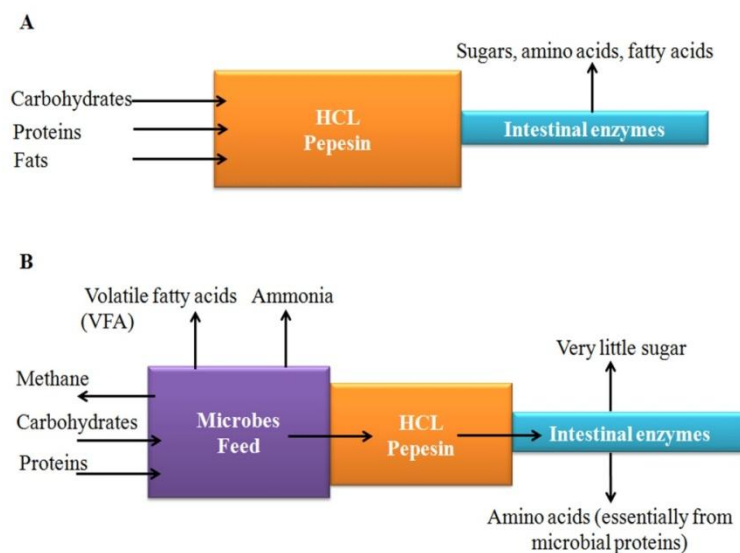


Figure 11:- Food digestion in multi- chambered stomach animals (microbial fermentation prior to digestion in ruminants) versus mono-gastric animals (non-ruminant animals)[135].

In the newborn lamb, the rumen is undeveloped and is small in comparison with the abomasum. The immature small ruminants do not have a functional rumen and reticulum, and undergo reticulorumenomasal growth. Rumen papillae (sites of nutrient absorption) lengthen and decrease in numbers as part of rumen development. The reticulum resembles a "honey comb" in appearance. Digesta mingle continually between both sections. Relatively little digestive activity occurs in the omasum (many piles) (Fig. 12; Fig. 13).

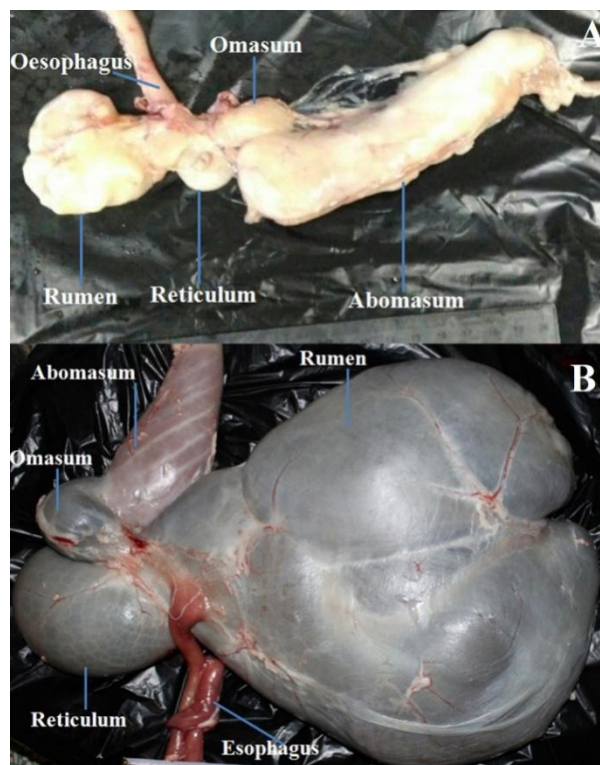


Figure 12:- Illustrating the external structure of the *rumen*, the *reticulum*, the *omasum*, and the *abomasum*, and the relative proportions of stomach compartments in sheep at different ages [7-day-old lambs (A) versus 140-day-old lambs (B)].

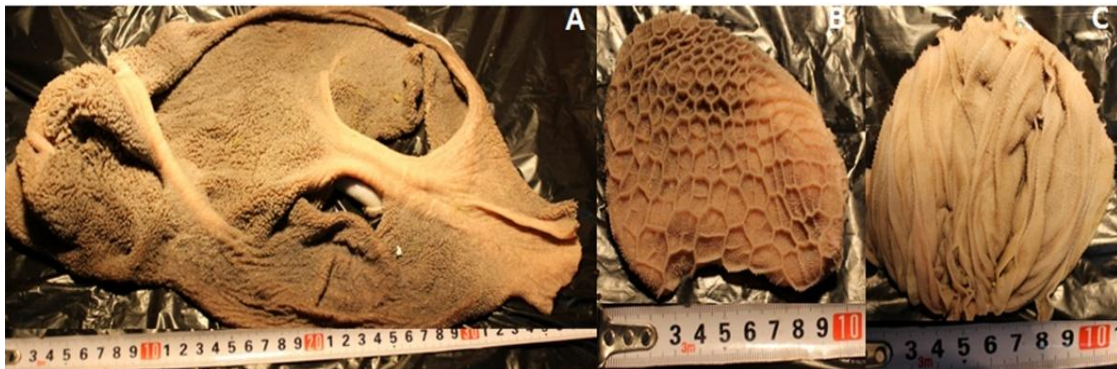


Figure 13:- Illustrating the internal structure of the *rumen* (A), the *reticulum* (B), and the *omasum* (C).

History of Sheep:-

The first livestock species to be domesticated were sheep and goats about 11,000 years ago in southwest Asia. They were primarily reared for meat; nevertheless, wool was discovered to be a beneficial product by the 4th and 5th millennium B.C. in southwest Asia and Europe [136]. Sheep then spread across Europe, Asia, and into Africa, developing into numerous breeds.

The mouflon (*Ovis orientalis orientalis* group) is a subspecies group of the wild sheep (*Ovis orientalis*). Populations of *O. orientalis* can be segregated into the mouflons (*orientalis* group) and the urials (*vignei* group). All modern domestic sheep breeds are most likely descended from the wild mouflon of Europe and Asia [137; 138; 139]. *Ovis aries* is an entirely domesticated animal that is broadly reliant on humans for its health and survival.

Characteristics of Sheep:-

Sheep belong to the order Artiodactyla with camelids, suborder Ruminantia, family Bovidae, and subfamily Caprinae [140]. Sheep have an array of sizes, colors, and breeds globally and are considered to be intermediate grazers (i.e having nutritional requirements midway between grazers and browsers).

Economic importance of sheep:-

Sheep are a crucial part of the global agricultural economy. China, Australia, India, and Iran have the bulkiest modern flocks, and serve both local and global requirements for wool and mutton. However, New Zealand has smaller flocks and retains an enormous international economic impact because of sheep product exportations. The conventional essential markets, including European Union (EU) and United States of America (USA) will be enlarged by augmenting demand from developing countries, such as China, Saudi Arabia, Jordan, United Arab Emirates, India, Turkey and Qatar [141]. According to Food and Agriculture Organization of the United Nations (FAO), the ten countries with the largest number of heads (expressed as a percentage) of sheep were: mainland China, Australia, India, Iran, Nigeria, the former Sudan, United Kingdom, Turkey, New Zealand, and Ethiopia. Interestingly, mainland China has changed dramatically from the top three producers in 1961 to the top first in 2014 (Fig. 14) (<http://www.fao.org/faostat>).

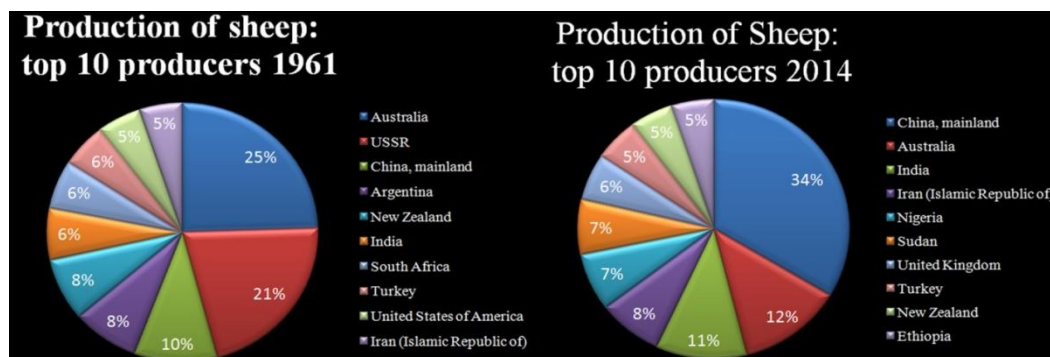


Figure 14:- Top 10 sheep producers world-wide.

Sheep versus goats:-

Goat and sheep industry have made monumental gains in productivity globally. Both species provide humans with meat, milk, wool and skins. Goats are well-known as a tough animal that can be fruitful in extreme environments that are not appropriate for other ruminants, such as sheep and cattle [142]. By comparison with sheep, goats have an exceptional ability for walking long distances, are recognized to select the most nutritive plants and can utilize bushes and shrubs [143] (Fig. 15). Several reports showed that sheep and goats share the same species of GIN parasites [144; 145]. Additionally, both sheep and goats can readily transmit GIN parasites to each other [146; 147]. Cross-infection of resistant GIN parasites between these two hosts may occur and must be a significant cause for concern in countries like China where goats and sheep may share the same farming system. Goats prefer browsing, while sheep will rely almost completely on grazing pastures. Browsing diminishes L_3 intake owing to higher numbers of infective larvae are only found very nearby the base of pastures [148]. Similarly, higher faecal egg counts were reported in the Angora goats (grazer) compared to Saanen goats (browser) due to the differences in feeding behavior [149].

Therefore, sheep are more likely to contract GIN parasite infections due to grazing behavior; however, goats do not mount the same level of immune response to the GIN parasites as sheep. Several studies have compared the level of GIN parasite infection in goats and sheep during common grazing when they placed in diverse conditions. The higher faecal strongylate egg counts of New Zealand feral goats are significantly different from those of adult Romney sheep grazing mixed grass and clover swards with no access for browsing [150]. Furthermore, other studies have reported that goats are generally more succumbed to parasitic infection than sheep under grazing systems [151; 152]. Results from another study involving Coopworth lambs and Saanen kids between 6- and 8- month-old grazing naturally infected pasture indicated that after approximately 6 months, Saanen kids had significantly higher faecal egg counts than those of the Coopworth lambs [153]. Previous research comparing sheep and goats has found that the average strongyle faecal egg counts of sheep reduced from the age of 8 months onwards whereas this only happened in goats from 12 to 18 months onwards under the traditional husbandry system in peninsular Malaysia [154]. Faecal egg counts in sheep declines more rapidly due to the earlier development of an effective immune response to GINs compared to goat [155]. A similar picture documented that goats are highly susceptible to GIN infections in general and to *H. contortus* in particular compared to sheep [156]. Poorly developed immunological responses are the primary cause of their relative inability to control GIN infections, and the associated pathophysiological consequences [149].

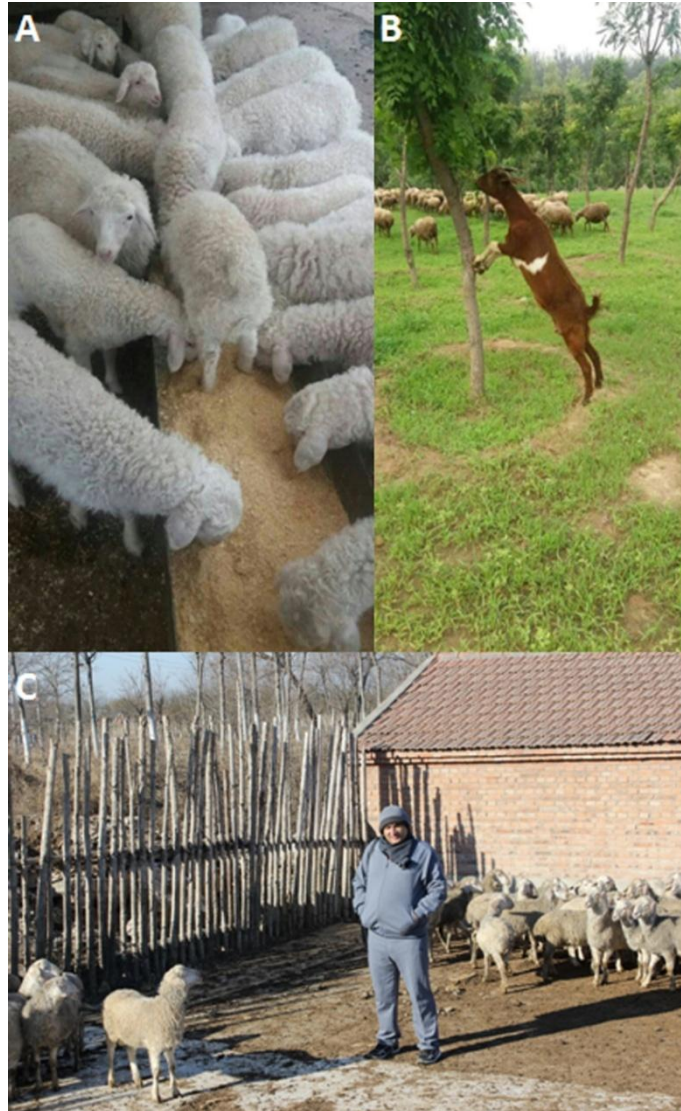


Figure 15:- Traditional method of goat and sheep housing and management.

The figure shows lambs fed concentrate diets (A), browsing goats and grazing sheep shared the same farming system on pasture in rangeland near their owner's premises at Jin Zhan village, Chaoyang, Beijing, China (B), and sheep and goats confined in their shelter (C).

Abomasal anatomy:-

The abomasum is separated into three obvious anatomical parts. These are: the cardiac region, which contains a small circular area around the omaso-abomasal junction, the fundic/corpic area, which is distinguished by luminal folds covering the major part of the abomasal surface, and the pyloric area, which is the most distal part, is marked by the existence of rugae (Fig. 16).

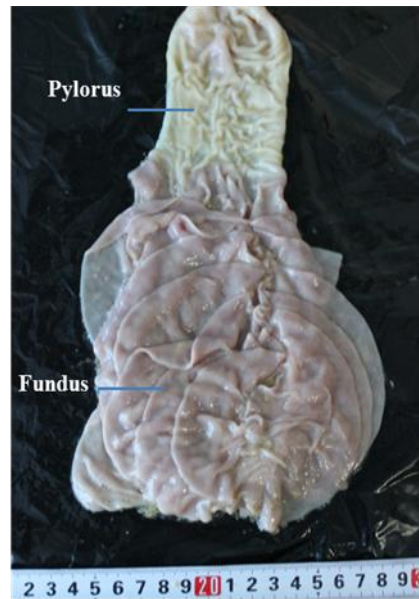


Figure 16:- Inner structure anatomy of the ovine abomasum.

Histologically, the abomasal wall, like the rest of the gastrointestinal tract, is organized into four concentric layers from the outside inward, which are: the *tunica serosa*, the *tunica muscularis*, the *tunica submucosa* and the *tunica mucosa*. The *lamina propria* has a connective matrix supporting the gastric epithelium and the feeder blood vessels. The gastric epithelium is composed of surface epithelial cells (cuboidal to columnar cells) and gastric glands. The cellular layer forms numerous invaginations, called gastric pits or foveolae serving as draining duct of adjacent glandular secretions to the lumen. The abomasum has two types of gastric glands according to their anatomical location, namely the pyloric and fundic glands (Fig. 17; Fig. 18).

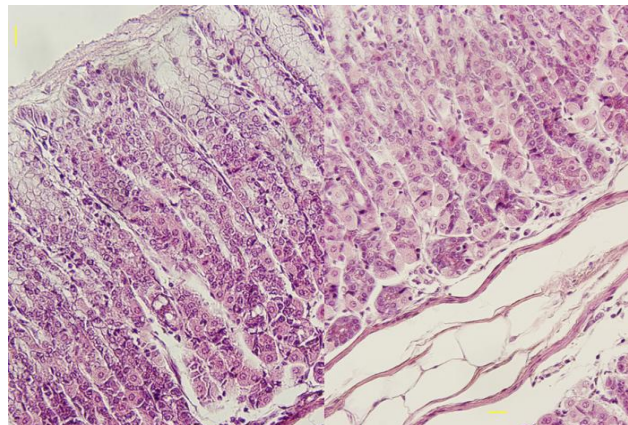


Figure 17:- Normal architectures were seen in the ovine abomasal tissue (fundic region). HE 40X. Bar = 10 um.

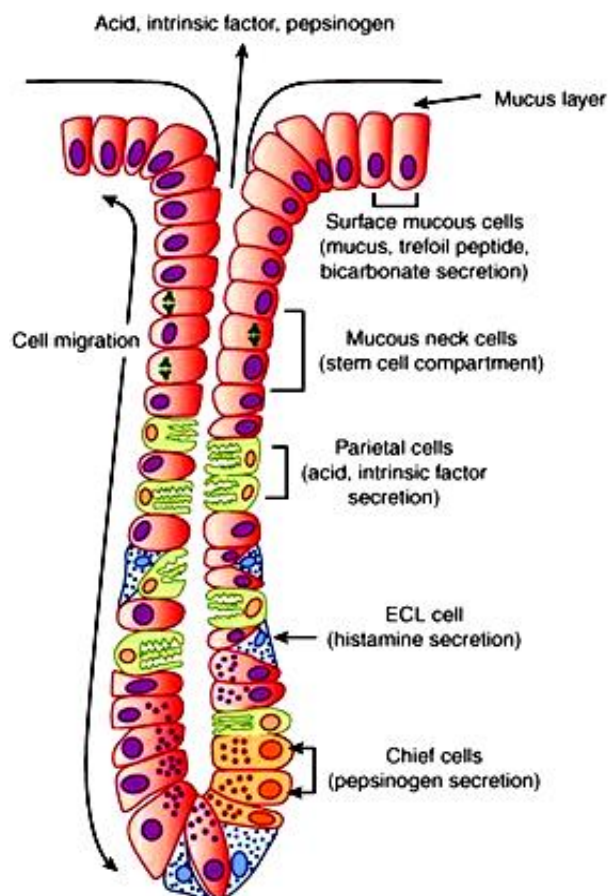


Figure 18:- Gastric gland structure from the fundic area (Adapted from [157].)

Abomasum is one of the most important sites for living bursate nematodes belonging to Trichostrongylidae family in small ruminants, because it is the site location for 3 pathogen species of GI nematodes e.g., *Haemonchus* spp., *Teladorsagia* spp., *Ostertagia* spp. and

Trichostrongylus spp. [158]. The histopathology of haemonchosis in sheep has been the subject of intense investigations during the last decades [61; 159; 160; 161]. Abomasal nematode larvae enter the gastric pits and glands, where they cause local lesions, the nodules, which have at their centre developing larvae in distended glands lined by many flat undifferentiated cells, but few secretory cells. These nodules are diverse on the abomasal folds [162; 163]. Adjoining parasitized glands demonstrate mucous cell hyperplasia with fewer parietal cells and chief cells. These changes become more broad after the L₄s or immature adults emerge [163; 164; 165; 166; 167; 168]. Luminal stages stay close to and within the superficial mucus [169]. The infected sheep abomasa are much heavier than control animals and the mucosal thickness is much greater [170; 171]. The pits are expanded and there are fewer parietal cells and chief cells, however, more mucous neck cells (MNCs) and undifferentiated cells [172]. Similar generalized histological changes are observed after adult *T. circumcincta* [168; 173] or *H. contortus* transfer [174] even though there are no damaged glands where larvae have grown. There may be an association between the pathology, histopathology and inflammatory responses to the nematode presence, especially through the parietal cell loss and inhibition. Infection provokes the release of cytokines, including IL-1 β , IFN- γ and TNF- α , which are inhibitors of parietal cells [175; 176; 177]. Parietal cell loss not only results in diminished acid secretion and increased abomasal pH, although fewer chief cells, owing to parietal cells produced growth factors necessary for mucous neck cells (MNCs) to develop into chief cells [178]. In line with these findings, this occurs in other pathologies where parietal cells are lost, for example transgenic mouse models [179; 180]. The increase in mucus-producing cells in general, and MNCs in particular, is consistent with the increased mucin secretion seen in *H. contortus*-infected sheep [181].

Pathophysiology:-

H. contortus- or *T. circumcincta*-infected sheep have increased gastric pH, elevated serum concentrations of pepsinogen and gastrin, and diminished acid secretion [166; 174; 182]. The abomasal pH of uninfected sheep is approximately 2.8, nevertheless the pH may be 4 and above in *T. circumcincta* infected sheep [166; 182]. Previously challenged sheep with *Ostertagia circumcincta* may be able to acidify their abomasal fluid and maintain the pH low, while naive animals lose the ability to maintain abomasal pH [183]. Hypergastrinaemia began at the same time as abomasal pH became increased, however stayed high and continued to augment when pH had reached a maximum and had returned towards normal. Serum gastrin was depressed when abomasal pH exceeds 5.5 in *O. circumcincta*-infected sheep [166; 184].

Hyperpepsinogenaemia is frequently employed in ruminants as an indicator of abomasal parasitism [166; 174; 182; 185; 186; 187]. Serum pepsinogen usually elevates at the time of parasite emergence from the glands or a little earlier and remains increased for up to 30-60 days after a single infection while the parasites are still present in the abomasum in *O. circumcincta*- and *H. contortus*-infected sheep [166; 168; 174; 188]. The magnitude of the increased plasma pepsinogen levels in ovine gastrointestinal trichostrongyles, including *Ostertagia leptospicularis* and *O. circumcincta* varies between animals, from approximately zero [166; 174; 182; 187; 189] to remarkable increase in immune animals, even with small worm burdens [190]. After transfer of adult *H. contortus* [174], *Ostertagia ostertagi* [165], or *T. circumcincta* [164; 166; 168; 188], abomasal pH, serum gastrin and pepsinogen concentrations increase within the first day. These effects on abomasal secretion are proposed to be initiated by abomasal parasite chemicals [164; 166; 167; 168], but later also to include the host response. *H. contortus* excretory/secretory products (ES), which are released by GINs to their environment, consist of diverse bioactive components, including chemotaxins, metabolic end-products, enzymes, immunomodulators and growth factors. ES has been reported to inhibit acid secretion *in vitro* [191], as well as restrain the enterochromaffin-like cells, which may indirectly cause diminished acid secretion by parietal cells [187; 192]. Ovine gastrointestinal nematode chemotaxins recruit granulocytes *in vitro* [193], which could be another way of damaging parietal cells *in vivo* [163; 167].

Conclusion and future directions:-

We have attempted to provide a comprehensive overview about the haemonchine parasite and the ovine host as well to reveal intervention points that could be exploited to discover novel therapies, vaccine strategies and prophylactic intervention points for parasites, and to understand the parasitic disease progression and the infection consequence.

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Competing interests:-

The authors declare that they have no competing interests.

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RESEARCH ARTICLE

GALLERY WALK TECHNIQUE IN TEACHING SELECTED TOPICS IN ENGLISH AND LEARNERS' PERFORMANCE.

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Abstract

The study aimed to determine the effectiveness of Gallery Walk Technique in teaching selected topics in English among Grade Four learners. The hypothesis stating that there is no significant difference between the performance of the experimental and control groups was tested at .05 level. Quasi-experimental research was adopted by the study employing match-pairing of participants. Pre-test was administered to the learners and scores utilized to identify the match-pairs. T-test for independent means was employed to determine if there is a significant difference between the performance of the two groups. For two quarters, the experimental group was taught using Gallery Walk Technique whereas the control group was taught using traditional method. Formative tests were administered during the implementation of the study. Post-test was given after implementing the two methods of teaching. Results revealed that there was a significant difference in their performance. The experimental group performed better than the control group.

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Introduction:-

English is a learning area in elementary level in the Philippine educational system. Pupils' performance is gauged based on the results of the National Achievement Test (NAT) administered yearly by the Department of Education. The test aims to provide empirical information on the achievement level of students at certain grade levels. The low Mean Performance Scores (MPS) in English of pupils from 2009-2013 (DepEd, 2013) specifically in reading and grammar indicated that the teaching of English is very much wanting of improvement.

The primary purpose of teaching at any level of education is to bring a fundamental change in the learner. To facilitate the process of knowledge transmission, teachers should apply appropriate teaching methods that best suit specific objectives and level exit outcomes. Identified teaching strategies all over the world are the visual aids, modelled spoken language, lesson outlines, skim and scan, and the like contingent on the set curriculum to improve pupils' performance. Because teaching and learning now come in different styles and forms, educators need to try on adopting new methods in teaching and learning which aim at improving the quality of education. Although some teachers shudder at the thought of having all of the students out of their desks moving around the classroom at the same time, but in reality, it can be a very effective technique for classroom management especially in the elementary level. Meyers, et al (1993) consider active learning as learning environments that allow students to talk

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and listen, read, write and reflect on course content. This is enhanced through informal small groups, simulations, problem-solving exercises and other activities.

Stewart (2009) cites in his theory of basic learning that learners had their foundation in English at the elementary level and enhanced in the high school level. Bloom, et al. (1956) in his Taxonomy of Educational Objectives said that students proceed from a lower level of learning such as remembering concepts, comprehension and application to a much higher level. Mastery of the lower level should be achieved first before they are able to analyze and evaluate concepts, processes and procedures.

The five skills of English as a discipline such as reading, listening, speaking, writing and viewing need to be amplified. Effective teaching for tomorrow demands teaching strategies in which teachers creatively organize a variety of content which must be taught to a variety of learners with different backgrounds, needs and interests. Compared to traditional methods, multi-sensory integrated strategies make pupils more enthusiastic, thereby more motivated to learn. Gagne's (1985) Conditions of Learning Theory asserts that there are several different types or levels of learning. He identified five major categories of learning: verbal information, intellectual skills, cognitive strategies, motor skills and attitudes. All categories of learning cited by Gagne are included in the Gallery Walk. Gallery Walk technique encourages students out of their chairs and actively involve themselves in the discussion. Tewksbury, et al. (2014) cite that Gallery Walk is cooperative learning strategy. Since it is a cooperative learning because students are formed in groups, they share thoughts in a more intimate and supportive setting. (<http://serc.carleton.edu/introgeo/gallerywalk/what.html>).

Materials and methods:-

The study adopted the quasi-experimental research where participants were match-paired based on the scores in the administered pre-test. One group was taught using gallery walk technique and the other group the traditional method of teaching. The study covered the selected topics scheduled in English subject for two quarters. Three phases were considered for the conduct of the study. The first phase was the development of learning materials, tests, and lesson plans which were submitted to the experts for validation. The second phase was the implementation involving the administration of pre-test to the learners to identify the match-pairs for the two groups. Mean scores of the two groups in the pre-test were subjected to t-test to determine if there was a significant difference in their prior knowledge before the implementation of the study. The analysis revealed that there was no significant difference, therefore there was no bias in the selection of the participants. The same sets of questions and exercises were given covering the same lessons. Quizzes as formative tests were also administered. The third phase of the study was the post-implementation where quarterly (summative) tests and post tests were given to the learners. Significant difference in the performance of learners in these tests was determined using t-test for independent means.

Results and Discussion:-

The table below illustrates the performance of the two groups in the formative tests for the two quarterly periods. For the first quarter a mean difference of 2.88 was obtained in favor of the control group. A computed t-value of 1.72 was not significant at .05 level with 48 degrees of freedom. This accepts the hypothesis that no significant difference exist in their performance during the first quarterly period. However, in the second quarterly period, the computed t-value of 2.83 suggests that there was a significant difference in their performance.

Table 1:-Formative Means of Control and Experimental Groups in the Two Quarterly Periods

Groups		t	df
Control	Experimental		
First Quarterly	76.44	73.56	1.72*
Second Quarterly	72.44	78.32	2.83*48

Note. *=p<.05

In Table 2, the control group garnered a mean score of 18.76 and a standard deviation of 5.38. The experimental group got a mean score of 22.72 with a standard deviation of 5.57. This conveys a mean difference of 3.96 in favor of the learners in the experimental group further suggesting that they performed better than their counterpart. This is affirmed by the computed t-value of 2.56 at .05 level.

Table 2:-Post-test Means of Control and Experimental Groups

Groups		t	df
Control	Experimental		
Post-test	18.7622.722.56*	48	
	(5.38) (5.57)		

Note. *=p<.05

Mean differences of 5.60 and 5.96 in the first and second quarterly periods respectively were both in favor of the experimental group's performance. As shown in Table 3, the computed t-values of 2.79 and 3.88 were significant at .05 level, therefore there was a significant difference between the performance of the two groups. It implies that learners in the experimental group performed better.

Table 3:-Quarterly (Summative) Test Means of the Control and Experimental Groups in the Two Quarterly Periods

Groups		Control	Experimental	t	df
First Quarterly		17.96	23.56	2.79*	48
(4.26)	(8.87)				
Second Quarterly		17.12	23.08	3.88*	48
(4.11)	(6.03)				
Total	35.08	46.84	3.79*	48	
(7.45)	(13.30)				

Note. *=p<.05

The findings revealed significant differences in the mean scores of learners in the two groups. These are evident in the formative tests, summative or quarterly tests and the post tests. Mean differences in scores were in favor of the experimental group taught with gallery walk technique. The analysis suggests that the technique was more effective than the conventional method of teaching. Since gallery walk involves the use of multi-sensory strategies it is an effective means of bringing out the best in the students, hence optimum learning is achieved.

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REVIEW ARTICLE

NEXT GENERATION SEQUENCING AND MICROBIAL COMMUNITY ASSOCIATED WITH EUKARYOTES INCLUDING PARASITIC HELMINTHS: A REVIEW ARTICLE.

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Abstract

Molecular techniques, such as 16S rRNA sequencing and whole microbial genome sequencing, have revolutionized the standard microbiological practice by disclosing the remarkable diversity, composition and identity of bacterial communities associated with eukaryotes. Here, we outline the development from the single-gene analysis in an ecosystem to the compounded genetic information of the entire ecosystem employing high-throughput sequencing technologies. We also provide the microbial communities associated with eukaryotic organisms, including some parasitic helminths. We hope that the information provided here will be useful to widen our understanding of the techniques in most reasonably equipped molecular biology laboratories, and bacteria associated with eukaryotes, involving parasitic helminths.

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The necessities of microbial world:-

Prokaryotes represent the largest proportion of life forms on earth, which comprises 10^6 to 10^8 different genotypes. The largest reservoir for carbon (350–550 Petagrams), nitrogen (85–130 Pg) and phosphorous (9–14 Pg) is the prokaryotic organisms that inhabit our planet [1]. These microorganisms are responsible for most of the chemical cycles on earth, which are essential for our existence. In addition, microorganisms serve human beings by maintaining our health, fermenting food and producing drugs [2]. Most of these microorganisms live in communities; many of those communities are complex with high magnitude of diversity with thousands of interacting members [3] where they will compete for basic needs like space, air, etc. For the better understanding of life, it is essential to understand the diversity of these microorganisms in the community. Most of the research on microorganisms is based on culturing organisms in the laboratory. Major difficulty encountered by researchers in the field of microbial study was, how to study those organisms which do not grow under standard culturing conditions [4]. The term metagenomics was first coined by Handelsman in 1998, for habitat based investigation of mixed microbial population at the DNA level. Metagenomics provides a culture-independent way to access unculturable microorganisms and is now possible to study the genome of all those organisms. Thus metagenomics revolutionized the field of microbiology which offers a window to understand previously unknown and uncultivable microorganisms. Life on earth was flourished due to a transition from the anaerobic to aerobic forms of photosynthetic bacteria. Due to this transition oxygen began to accumulate in the atmosphere until it was sufficient to support the life of aerobic organisms. Once oxygen concentration reached at a very high concentration, oxygen molecule began to collide and produced ozone. Later this ozone gas accumulated in the stratosphere and protected the life forms on earth from ultraviolet light [5]. Yet another group of microorganisms evolved are nitrogen fixers. These bacteria could break triple bonded nitrogen molecule and fix atmospheric nitrogen for the usage of terrestrial living beings. Human health is under constant check by human microbiome such as gut microflora, when the balance of gut microbial community is compromised, many diseases like colon cancer, inflammatory bowel disease, obesity and diabetes may occur. All these microbes coevolved with the human species, produces an intertwined web of dependency and communication. A large proportion of the drugs available today are synthesized from bacteria and fungi. The discovery of antibiotics has transformed human existence by providing an outstanding way for the treatment of infectious diseases. In addition microorganisms play an important role in providing industrial enzymes and polymers, cleaning up toxic waste products and can be employed in the process of fermentation etc. Tabulated below are the differences between cultivable and uncultivable microbes [6]. In the late 1970s, the microbial genome study began with the sequencing of bacteriophage genomes MS2 RNA [7] and QX174 [8]. *Haemophilus influenza* genome was sequenced in 1995 [9].

History and technology evolution:-

There are many examples in the scientific literature of bacterial groups co-existing in the animal and human body. Their composition and activities are thought to be closely involved in shaping our health and have been examined over several years. The classical approach to explore the animal and human microbial community is relied on culturing and isolating a single microorganism, and identifying its physiological, biochemical, ecology, life history and serum reaction index characteristics. Nevertheless, a large proportion of the gut microbiota is uncultured microbes, and it would be logistically laborious to grow the diverse species that can be cultured. With the development of molecular biology and the extensive application of modern technologies, many new genomic approaches have emerged during the past decade for the animal and human microbe investigations. These approaches involve terminal restriction fragment length polymorphism (T-RFLP), PCR based denaturing gradient gel electrophoresis (DGGE), automated ribosomal intergenic spacer analysis, fluorescence in situ hybridization (FISH), and microarray.

Sanger sequencing and next generation sequencing (or high-throughput sequencing):-

Sequencing technology has become the common process in researching the animal microbial community, and metagenomic sequencing has firmly transferred from conventional Sanger sequencing technology to be next-generation sequencing (NGS). The disadvantages of Sanger sequencing are the laborious cloning process, the cloning bias and the expensive cost of a single giga base of data generated. The most conspicuous source of this bias, which produces a low read coverage in these regions, is adenine-thymine (AT) richness. NGS, which eliminate the cloning bias, has entirely altered sequencing efficiency, allowing high-throughput analysis of complex microbiota via the capture of short DNA sequence (amplicons or random fragments) in which a myriad of samples may be multiplexed employing short DNA sequence “bar codes” [10]. This provides adequate sequencing depth in each sample to characterize the top 99.99% of the microbiota, permitting the unravelled micro-organism discovery with no prior knowledge of their sequences. In addition to analyzing the microbial genomics, NGS is also

appropriate for transcriptome sequencing, which determines mRNA quantitatively, enabling new insights into genome expression and how it may be altered in healthy and diseased individuals. The NGS technology holds promise for a contemporary understanding of infectious disease and for diseases not previously known to have a microbial element.

The different sequencing strategies:-

In recent years, various types of sequencing strategies have been applied successfully in a wide range of metagenomic projects that have enormously revolutionized our knowledge of the microbial communities living in and on animal and human bodies. A large and growing body of literature on the microbial community is relied primarily on three kinds of sequencing strategies: amplicon sequencing, shotgun sequencing and transcriptome sequencing, whose cardinal goals are to study microbial communities *in vivo*. The most common application of amplicon sequencing with NGS technology in the microbiome is 16S rRNA gene sequencing, which is usually employed to assess the bacterial general composition. Metagenome shotgun sequencing provides more intricate information of the microbiome, including gene content and its potential functions. Finally, the active members of the microbial community and its functionality can be investigated by transcriptome sequencing.

Amplicon sequencing:-

Complex microbial communities, such as the animal and human gastrointestinal tract (GIT) microbiome, are presently receiving increasing interest, owing basically to technological advances in culture independent approaches in recent years. Nevertheless, in surveys of tremendously diverse ecosystems, the size of clone libraries (typically 100–500 clones each) only allows for identification of the community members that are present in huge abundance [11; 12; 13; 14; 15; 16]. In addition to failing to detect rare members of the ecosystem, these relatively small data sets provide inaccurate estimates when used for computing species richness within an ecosystem. Regardless of the approach used to estimate species richness, the estimates acquired are highly dependent on sample size, and smaller data sets usually result in the miscalculation of species richness [17; 18]. However, it was not until the end of the 1970's that ribosomal ribonucleic acid (rRNA) sequences were observed to provide a key to prokaryotic phylogeny. The pioneering research and view of [19; 20] was approved and taken up by others [21; 22]. The rRNA molecule is universally distributed, functionally homologous across all prokaryotes and has regions that are conserved-thus permitting sequences to be aligned for comparison and thereby fulfilling the criteria of an exceptional chronometer [19]. In contrast to physiological and phenotypic traits, the genetic material is not impacted by culture conditions. The number of differences in nucleotide sequences in a specific gene that are counted, the greater the difference between genes, the more evolutionary separated are the two organisms. The evolutionary distances, taken as a fraction of sequence differences between pairs in a collection of sequences, is employed to assemble phylogenetic trees [23]. Because of the slow rate of evolutionary change of small-subunit rRNA gene sequences, along with ease of extraction and manipulation, they have become the molecule of choice, and form the "gold standard" in the creation of phylogenetic classification strategies. 16S rRNA gene sequence analysis has not only facilitated new insights into the phylogenetic interrelationships of microorganisms but has provided molecular systematists with an immensely powerful means for describing new diversity within any given microbiological environment. The molecular approach has had major repercussions not only in taxonomy but also, for example, microbial ecology. Studies by [22] have demonstrated that microorganisms could be identified directly in their habitats through a combination of rRNA gene cloning and sequencing. The 16S rRNA gene is a constituent of the 30S small subunit of prokaryotic ribosomes. It is approximately 1.5 kb (or 1500 nucleotides) in length (Fig. 19) and is employed for phylogenetic studies [24] as it is extremely conserved between different bacterial and archaeal species [25].

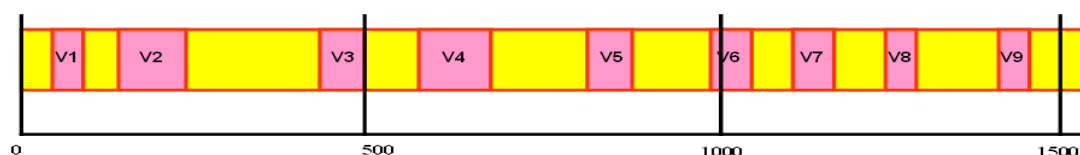
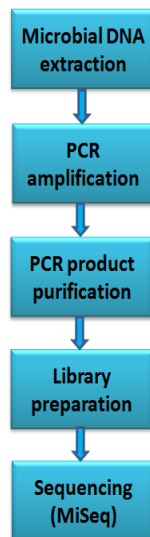


Figure 19:- Nearly 1.5 kb 16S rRNA gene of *E. coli* illustrating the nine variable regions.

The 16S rRNA gene consists of conserved and variable regions (Fig. 18). The variable regions, which are nine different variable 16S rRNA gene regions being flanked by conserved sites in most bacteria [26], allow discrimination between different microorganisms. 16S rRNA gene methods rely on the PCR (polymerase chain reaction) using 'universal' primers targeted at the conserved regions and designed to amplify as wide a range of

different microorganisms as possible [27; 28]. As a result, 16S rRNA gene sequencing has become ubiquitous in microbiology and microbial ecology as an expeditious and precise alternative to phenotypic bacterial identification methods [29]. Currently, the sequencing of 16S rRNA gene variable regions is the most common application of amplicon sequencing on NGS platforms (Fig. 20).

It provides a quantitative description of the bacterial community present in a complex biological mixture, allowing examination of entire communities and the identities of their constituent members. Phylogenetic mapping of rRNA variation was first used to create the three domain of life [30]. The gene conserved nature was subsequently exploited to develop more rapid methods for determining relationships between organisms directly from environmental DNA and RNA extracts [27; 31]. The coupling of 16S rRNA PCR with next-generation sequencing enables the study of a wide range of samples at low cost [32]. There are three benchtop high-throughput sequencing instruments, which are the 454 GS Junior (Roche), MiSeq (Illumina), and Ion Torrent PGM (Life Technologies). They are laser-printer sized and offer modest set-up and running costs. The MiSeq had the highest throughput per run (1.6 Gb/ run, 60 Mb/h) and lowest error rates. The 454 GS Junior generated the longest reads (up to 600 bases) and most contiguous assemblies but had the lowest throughput (70 Mb/run, 9 Mb/h). The Ion Torrent PGM, which runs in 100-bp mode, had the highest throughput (80–100 Mb/h). In contrast to the MiSeq, the Ion Torrent PGM and 454 GS Junior both produced homopolymer-associated indel errors (1.5 and 0.38 errors per 100 bases, respectively) [33]. 16S rRNA data analysis depends on related sequence clustering at a specific level of identity and counting the representative number of each cluster. Similar sequence clusters are known as operational taxonomic units (OTUs). A level of 95% sequence identity is frequently chosen as being representative of a genus and 97% for a species when using partial 16S rRNA gene sequences [34]. Identification accuracy is reliant on the selected reference database. Curated databases, for example the Ribosomal Database Project [35], GreenGenes [36] and SILVA [37], where quality assessment and alignment of MiSeq sequences are manually optimized, are substantial for optimal phylogenetic analysis. There are two common analysis pipelines employing for analyzing 16S rRNA gene sequence data. These are: QIIME [38] and Mothur [39]. Figure 1 and Table 1 present the summary statistic of the community structure, and the commonly employed ecological and NGS term explanation in the microbial community research field. The commonly employed ecological and NGS term explanation in the microbiota research field is illustrated in Figure 21 and Table 3.



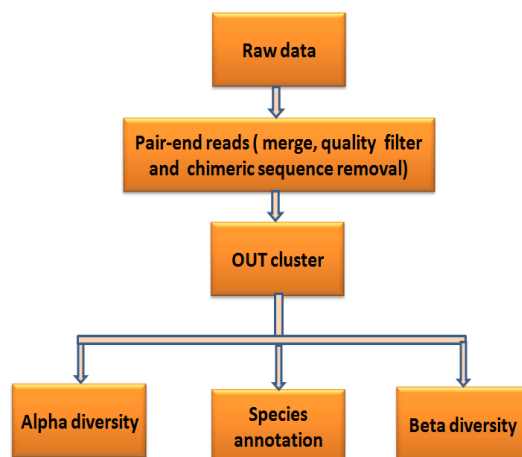


Figure 20:- Flow chart summarizing the 16S ribosomal RNA (rRNA) NGS

Table 3:- The commonly employed ecological and NGS term explanation in the microbiota research field.

Term	Explanation	Reference
Operational taxonomic units (OTUs)	Clusters of 16S/18S small subunit (SSU) rRNA gene similarity, are used as theory-agnostic approximations of microbial taxa	[40]
Microbiome	It literally means small biome, the ecosystem comprising all microorganisms in a particular environment together with their genes and environmental interactions.	[41]
Microbiota or microbial community	The assemblage of microorganisms themselves and can include bacteria, archaea, viruses, phage, fungi and other microbial eukarya.	[41]
Bacterial community	The composition of bacteria living within a particular part of the body.	[42]
Microflora	The community of microorganisms, including bacteria, fungi and algae, which live in a special habitat or in or on another living organism.	[43]
Flora	Referring specifically to plants, rather than microbes	[41]
Bar code	For processing a large number of samples with multiplex sequencing on a high-throughput instrument, individual "barcode" sequences are added to each sample so they can be distinguished and sorted during data analysis.	[41]
Evenness	A measure of the skew in abundance of community members. Is there one dominant organism or are all evenly represented?	[41]
Richness	The number of different types of organism present.	[41]
Diversity	A combination of richness and evenness can be considered to be a summary statistic for community structure.	[41]
Simpson index	A common diversity index indicating the probability that two individuals taken at random from a population are the same. Often presented as the inverse so that increasing diversity is mirrored by an increasing index value.	[41]
Shannon index	Alternatively, Shannon entropy-another common diversity index that quantifies the uncertainty of predicting the next individual taken from a sample.	[41]
Alpha diversity	Within sample diversity.	[44]
Beta diversity	Between sample diversity.	[44]

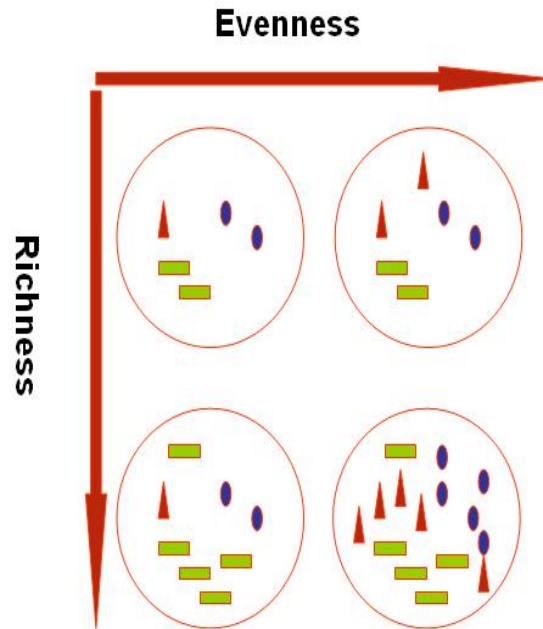


Figure 21:- A diagram describing species richness and evenness and how they characterize the community composition. Each shape represents an individual and the color and nature of the shape represents a different type of organism. Increased numbers of diverse types of organism is characterized as augmented species richness. The community is described as even, when no one organism is dominant.

Factors affecting gastrointestinal tract (GIT) microbiota:-

The ruminant gut microbiota is crucial in shaping several of its host's functional attributes. The bovine rumen microbiota is necessary for the proper physiological development of the rumen and for the animal's ability to digest and convert plant mass into food products, making it highly significant to animals. Factors affecting gastrointestinal tract (GIT) microbiota composition and activity in ruminants are presented in Figure 21. The bacterial profiles of the rumen microflora is known to be highly responsive to changes in diet, age, antibiotic use and health of the host animal and varies according to geographical location, season and feeding regimen (feed and feed additives) [45; 46; 47; 48; 49; 50]. Additionally, it has been reported that the host gut microbiota varies across species and individuals; however, it is relatively constant over time within an individual [51]. However, it has been reported that the detectable bacterial structure in the rumen is remarkably conserved among diverse locations and over time, while the quantity of individual bacterial species may alter diurnally in response to the feeding regimen [29]. The diversity and within-group similarity augmented with age, suggesting a more diverse but homogeneous and specific mature community, compared with the more heterogeneous and less diverse primary community. In addition, the establishment of this microbial population and the alterations occurring with the host's age was observed [52]. The composition of the intestinal microbiota can be modulated as a result of dietary exposure (ovine milk, formulas) as well as of intentional diet supplementations (prebiotics or probiotics). Rumen microbial community composition of cattle, bison, and buffalo (bovines), sheep and goats (caprids), deer (cervids), and alpacas, llamas, and guanacos (camelids), including diverse breeds of domestic cattle, sheep, and goats varies with diet and host [53]. It is well-established that the microbial community structure and composition is affected by diet [54]. The feeding operation is a more essential determinant of the bovine microbiome than is the geographic location of the feedlot [55]. Probiotics/prebiotics have the ability to modulate the balance and activities of the GI microbial ecosystem in ruminants [56]. Host genotype is among the factors that influence the microbial composition of the gut [57; 58]. Some authors studied the similarity degree in the predominant faecal microflora of identical twin pairs, fraternal twin pairs, and unrelated controls; the highest levels of similarity were observed in genetically identical twins [59]. Some authors studied the development of rumen microbial populations in young calves and lambs [60; 61]. The diversity indices of bacterial 16S rDNA and the change in soil bacterial community composition increased under intensive grazing [62]. A growing body of evidence shows that parasitic infections are associated with the alterations of porcine proximal colon microbiota at 21-days of infection with *Trichuris suis* [63], and the changes in the caprine abomasal microbial composition induced by *H. contortus* at 50-days infection [64]. Interactions among helminth parasites, bacterial microbiota, and host immunity have been reviewed in detail elsewhere [65; 66; 67] (Fig. 22).

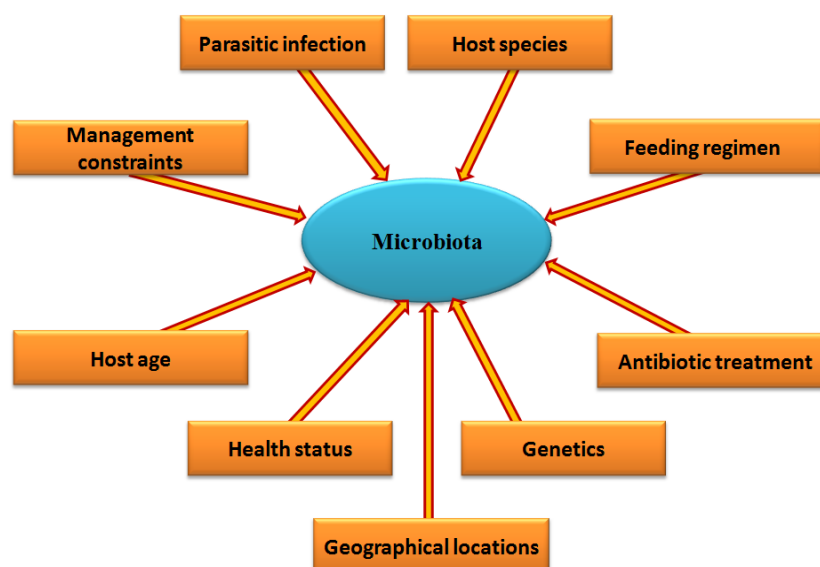


Figure 22:- Factors influencing the development and composition of microbiota in ruminants.

Microbial community associated with eukaryotes:-

A microbiota is an ecological community of symbiotic microorganisms found in and on all multicellular organisms. The synonymous term microbiome literally means small biome, the ecosystem including all microorganisms in a particular environment together with their genes and environmental interactions. The assemblage of microorganisms themselves is referred to as the microbiota or microbial community and can involve bacteria, archaea, viruses, phage, fungi and other microbial eukarya [68; 69]. Microflora can also be existed as a general term in the literature; nevertheless, flora refers particularly to plants, rather than microbes. The animal microbiota consists of microorganisms that exist upon, within or in close proximity to the animal body. Bacteria live in a wide variety of environments and have evolved close relationships with invertebrates, vertebrates, plants, and even with other bacteria [70; 71; 72]. The interaction between host organisms and prokaryotic microbes can range from pathogenic to mutually beneficial [71]. Symbiosis is an intimate interaction between two different biological species [73]. Such associations involve a variety of more specific relationships: parasitism (The symbiont benefits and the host is harmed), commensalism (the symbiont benefits and the host is unaffected), and mutualism (both the host and the symbiont benefit). It can be categorized according to their location in the host as either ectosymbionts (living on the surface) or endosymbionts (residing within host tissues). The latter can further be classified as primary, obligate *P*-endosymbionts (vital for host survival), or secondary, facultative *S*-symbionts (not necessary for host survival). Some *P*-endosymbionts reside in particular host structures or organs called bacteriomes; however, others are more extensively distributed in host tissues [74; 75]. The symbiosis continuation through host generations depends on symbiont transmission. Horizontally transmitted symbionts are recently taken up from the environment by each host generation, and vertically transmitted symbionts are frequently transferred through the female germ line. Mixed mode of transmission also exist [76]. For horizontally transmitted bacteria, symbiotic life is facultative: a free-living population serves as the inoculum for the symbiosis. Such free-living populations occur in soil [77], marine shallow waters and the deep sea [78]. In some cases the free-living population is restored by symbiont release from the host [79]. Vertical transmission frequently involves no aposymbiotic phase and transmission through the female germ line, even though there may be acquiring during mating. Moreover, there are unusual ways of maternal transmission, as in the stinkbug (*Megacopta punctatissima*), which harbor extracellular symbiotic gammaproteobacteria in the midgut cavity [80]. These bacteria are deposited with the eggs on plants in “symbiont capsules”, are eaten by the newly developing nymphs and colonize the insect midgut [81].

Symbiotic bacteria:-

A variety of microbes colonize invertebrate and vertebrate mucosal surfaces, including digestive, respiratory, and reproductive tracts [82].

Gut symbionts:-**Invertebrate-microbe associations:-**

The life cycle of five invertebrate-microbe interactions has been reviewed by [83]. In each of the symbioses demonstrated below, the animal acquires a specific symbiont (or symbionts), which colonizes the host in a specific location: a) the squid obtains its symbionts (e.g. *Vibrio*) from sea-water populations, which colonize the nascent light organ; b) the nematode brings its symbiont (e.g. *Xenorhabdus-Photorhabdus* and nematode worms) into the insect host (e.g. *Drosophila*), where both proliferate. The bacteria then recolonize the nematodes, which escape from the carcass; c) juvenile leeches obtain symbionts (*Aeromonas*, *Rikenella* and the leech) after hatching from their cocoon (perhaps from the cocoon itself). They then take up residence in the crop, where they digest their blood meal; d) the tsetse fly (*Sodalis glossinidius* and the tsetse fly) can either pass the symbionts maternally to the eggs or pick up new strains from the environment; e) specific symbionts on the food of the fruit fly colonize and persist in the enteric tract (i.e. *Enterococcus* and the fruit fly) (Fig. 23).

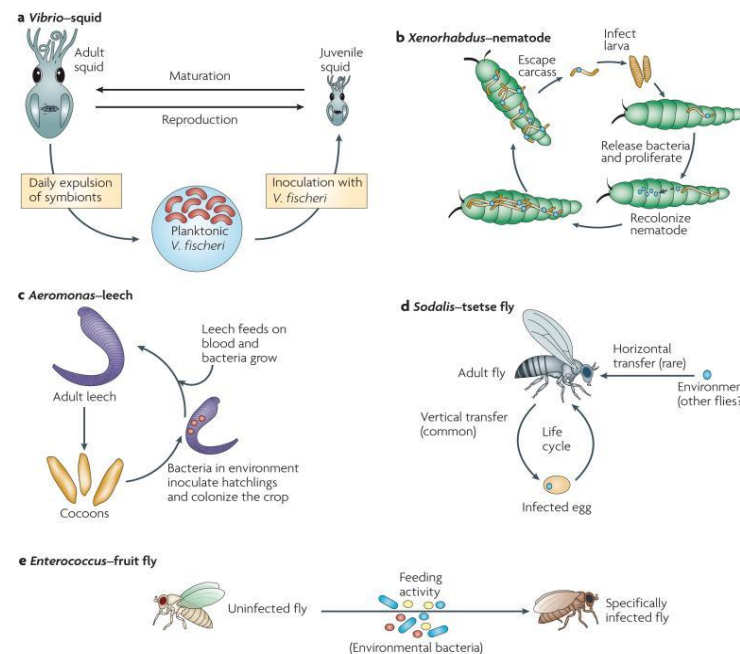


Figure 23:- Simplified life cycles of five symbioses (adapted from [83])

Nematode-microbe associations:-

Little is recognized about the symbiotic gut bacteria of nematodes and their role in host nutrition; however, there are ectosymbiotic microbes of marine nematodes known to supply nutrients to their hosts [84]. While abomasal nematodes are exposed to the abomasal and ruminal microbiota, the nematode gut symbionts may reflect the external microorganisms, which continually pass through their guts.

Rumen-microbe associations:-

The rumen microbes involve many species of bacteria, protozoa, fungi, and archaea, and are dominated by bacteria. These microbes are crucial for the fermentation, digestion, and conversion of indigestible foods and mucus into short-chain fatty acids and microbial protein [85; 86]. The rumen microbiota starts establishing in the lamb rumen soon after birth, before the rumen is functional. In the rumen of lambs, rigorous anaerobic bacteria prevail as early as 2 days of age. Cellulolytic bacteria emerge around the fourth day. Protozoa become established later (2 wk) and their populations have maintained by 4 mo [87]. In field-raised lambs, individual bacteria sequentially colonize the rumen: strict anaerobes prevail at day 2, from day 2 to 10, the strict aerobes and facultative anaerobes are 10 to 100-fold lower than the anaerobes and continue to decrease thereafter; methanogens and cellulolytic bacteria appear by day 3 and reach adult levels by the end of the first week [88]. The prevailing bacterial species in the immature rumen belong to *Bacteroides*, *Propionibacterium*, *Clostridium*, *Propionibacterium*, *clostridium*, *Peptostreptococcus* and *Bifidobacterium* [60], while in adult sheep and goats, the dominant species belong to the genus *Bacteroides* and the phylum *Firmicutes*, including *Clostridium* and *Prevotella* [89; 90]. The rumen bacterial population has been found to be predominantly composed of two phyla: *Firmicutes* (54%) and *Bacteroidetes* (40%) [91]. Similarly, it has

been reported that the rumen bacterial sequences are assigned to 19 phyla, with the dominance of *Firmicutes*, *Bacteroidetes*, and *Proteobacteria* [92].

Human gut microbes:-

The total population of human gut microbes (10^{14}) exceeds the body somatic and germ cells (10^{13}). The microbial density increases from mouth to the large intestine, with nearly 70% of gut microbes colonizing the colon [1]. The gut is antiseptic at birth, colonization starts immediately after the contact with the mother, and the bacterial number and diversity of the gut flora increases during postnatal development [93]. The microbial population is determined by the delivery mode (vaginal versus caesarean), diet, age, host genetics (lean versus obese), country of birth (developing world [Pakistan] versus developed world [Sweden]) [94; 95].

Bacterial enumeration and molecular analysis of human gut samples have shown that the gut microbiota is comprised of approximately 35,000 bacterial species belonging to at least 50 phyla [96]. The gut microbiota are dominated by members of phyla Firmicutes and Bacteroidetes, and there are relatively small proportions of Proteobacteria, Verucomicrobia, Actinobacteria, Fusobacteria, and Cyanobacteria [97]. The bacterial diversity in the gut lumen is different from the bacterial population attached to the mucus layer, and closely associated with the epithelium [98]. Aside from fermenting indigestible foods and mucus into valuable nutrients for the host, gut microbial community is crucial to the host in diverse ways, including the contribution to the immune system development and homeostasis, epithelial cell proliferation and differentiation, and protection against pathogens [99; 100].

Ecdysozoa microbial community:-

Ecdysozoa is a group of protostome animals, including Arthropoda, and Nematoda [101]. Even though arthropods and nematodes are morphologically different, they have several similar features in their endocrinology and physiology, including molting during development. Bacteria, archaea, and eukaryota, are the gut microbes of insects [102]. Related insect groups tend to share relevant species of symbionts, for example most aphids have primary endosymbionts of the genus *Buchnera* [103] and tsetse flies harbor *Wigglesworthia* in particular cells known as bacteriocytes or mycetocytes [104]. When the gut is simple tube or there is a high-throughput of digesta, the microbial diversity is low [105].

Insect gut microbes:-

The gut bacteria provide essential nutrients for the host, as well as particular enzymes [106], for example cellulase in the termite [107]. The microbial population in the insect hind guts changes with diet [108]. In this context, the bacterial community of wild ground beetles and field-caught *Anopheles stephensi* mosquito larvae are more diversified than those of laboratory-reared beetles and mosquito larvae, respectively [109; 110]. Bacterial community related to *Erwinia herbicola* and *Pantoea agglomerans*, which are ectoparasites of several plants, has been identified in pea aphid excreta cultured on nutritional agar [111]. Obligate and facultative anaerobes are the predominant hindgut microflora of cockroaches and termites [112; 113].

The denaturing gradient gel electrophoresis (DGGE) fingerprinting techniques and visualization with fluorescence *in situ* hybridization (FISH) have been employed to illustrate the microbial diversity associated with insects. Phyla, including Firmicutes and Bacteroidetes are the predominant. Similarly, the gut microbial population identified in ground beetles includes Bacilli, Fusobacteria, Gammaproteobacteria, Alphaproteobacteria, Clostridia, and Bacteroidetes, of which Bacilli and Gammaproteobacteria were dominant in wild and laboratory-raised beetles, respectively [114]. The gut bacterial community of the mosquitoes *A. aegypti* and *A. albopictus* included Actinobacter, Pseudomonas, Asaia, uncultured Gammaproteobacteria [110].

Microbial manipulators of reproduction:-

The best studied endosymbiont microbe that changes the host reproduction in arthropods is *Wolbachia pipientis*, which exploits the host insect to disseminate their progeny [115]. Gut bacteria of fruit fly *Drosophila melanogaster* impacts host mating preferences through pheromone synthesis [116]. Symbiotic *Spiroplasma* protects female *Drosophila hydei* from parasitic wasp attack [117]. Reproductive endosymbionts, including *Wolbachia*, *Rickettsia*, *Arsenophonus*, *Cardinium*, and *Flavobacterium*, which are maternally transmitted, are common among insects. They manipulate host reproduction to facilitate their transmission in diverse ways, such as parthenogenesis induction, feminization, and male killing [118; 119; 120; 121]. *Wolbachia* were early identified microscopically as Rickettsia in insect and arachnid tissues, involving eggs in 1920s [122]. From then on, strains of *Wolbachia pipientis* have been

known as important symbionts of filarial nematodes, while a variety of effects on insect reproduction are associated with the symbiotic *Wolbachia pipientis* strains of insects [123].

Microbial manipulators of host fitness:-

Symbiotic bacteria trigger the host immunity resulting in the regulation of the symbiotic population [124; 125]. Endosymbionts (*Regiella insectocola*) of aphids provide protection to their host against the fungal pathogen *Pandora neoaphidis* [126]. Similarly, the facultative symbionts *Serratia symbiotica* and *Hamiltonella defense* protect the host pea aphid from parasitic infection by *Aphidius ervi* and *Aphidius eadyi* [127; 128]. Symbiotic strategies include competition with the invaders for resources inside the host, synthesis of toxic chemical to assist host defenses, and augmentation of host heat tolerance [129; 130; 131; 132].

Bacteria associated with nematodes:-

Free-living terrestrial nematodes:-

Studies of the interactions of soil nematodes with bacteria have mainly focused on bacteria as a food source [133; 134], model for host-pathogen interactions, and possible nematode biocontrol strategy [135; 136]. Symbiotic bacteria, including Alphaproteobacteria, Gammaproteobacteria dominated in the soil nematode bacterial profiles with absence or very low presence of Actinobacteria [137; 138].

Free-living marine nematodes:-

A number of authors have studied the associations between sulphur-oxidizing, chemoautotrophic bacteria with marine nematodes. The nematode parasites provide oxygen and sulphide to the symbiont, which in turn provide food for the host [139; 140]. The cuticle of Stilbonematinae and Desmodoridae harbors sulphur-oxidizing ectosymbionts, which are a source of food for the nematode [84; 86]. The ectosymbiont microbes may be predominated by *Laxus oneistus* and *Robbea* sp. [141].

Entomopathogenic nematodes:-

Entomopathogenic nematodes (EPN) are lethal insect parasitoids, which have been employed effectively for the biological control of lepidopteran, dipteran, and coleopteran pests [142]. The symbiotic bacteria of *Photorhabdus* and *Xenorhabdus* of nematode families Steinernematidae and Heterorhabditidae, respectively are pathogenic in insect hosts and mutualistic in the nematode [143; 144]. These bacteria secrete toxins that are not only toxic to a range of insects, but also to other plant parasitic nematodes, indicating their roles for parasite biocontrol [145].

Mammalian nematodes:-

Wolbachia in filarial:-

Symbiotic *Wolbachia* have been observed in the majority of filarial nematode species, involving the human parasites *Brugia malayi*, *Onchocerca volvulus*, *Wuchereria bancrofti* and *Mansonella ozzardi* [146][146]¹⁴⁶[145][145][145][356], the dog heart worm *Dirofilaria immitis* [147], and the bovine parasite *Onchocerca ochengi* [148]. They are obligate mutualistic endosymbionts, which are essential for worm embryogenesis, development and adult survival, and essential nutrients to the nematodes [149; 150; 151]. Antibiotic therapy has confirmed beneficial in treating human and animal filarial infections and doxycycline treatment is now extensively recommended in endemic areas [150; 152; 153].

Ascaris suum:-

The facultative anaerobic from the intestine of *Ascaris suum*, including *Escherichia*, *Enterobacter*, *Klebsiella*, *Actinobacter*, *Citrobacter*, *Pseudomonas*, *Aeromonas*, and *Shigella* contribute to the possible serotonin synthesis [154; 155; 156].

Heligmosomoides polygyrus:-

The effects of L₃ larvae and adult worms of *Heligmosomoides polygyrus* on the ileum and caecal bacterial population were investigated, and the adult worm bacterial profiles were similar to those in the infected ileum with the bulk of bacterial species belonging to the phylum Firmicutes. However, the L₃ larval-stage profiles were different from the aforementioned two profiles and composed mainly of phylum Proteobacteria [157].

Trichuris muris:-

The literature has emphasized the necessity of host gut bacteria for the development of GIN parasites. By reducing the bacterial number in the mouse intestine using antibiotic treatment, the number of hatched *T. muris* eggs was

substantially diminished [158]. Several species are known to be affected by the germ-free status of the host include *Nippostrongylus brasiliensis* [159], *H. polygyrus* [160], *Trichinella spirallis* [161].

***Haemonchus contortus*:-**

Few studies designed to identify bacteria naturally associated with GIN parasites of sheep. L₃ obtained by *in vitro* culture of faeces collected from animals infected with *O. ostertagi*, *Cooperia onchophora*, and *H. contortus* was exploited to identify the bacterial profiles associated with these parasites. The bacterial species identified *Sphingobacterium multivorum* (opportunistic pathogen) and *Streptococcus macacae* (commensal bacterium). The fundamental focus of previous studies on the bacterial interactions with *H. contortus* has not been on symbiotic bacteria, but rather on potential pathogens, such as the soil bacterium *Bacillus thuringiensis*, which could be exploited to control the GIN parasites. The toxic *B. thuringiensis* are commonly employed in the insect biocontrol [162; 163; 164]. More recent attention has focused on the establishment of the molecular identity of abomasal bacteria associated with genetic resistance and susceptibility to *H. contortus* infection in sheep. The observed bacterial phyla with marked differences between resistant or susceptible sheep were Firmicutes (61.4% and 37.2%, respectively), Proteobacteria (10.2% and 37.2%, respectively), Bacteroidetes (12.8% and 5.8%, respectively), and unclassified bacteria (12.8% and 17%, respectively) [165]. It has been demonstrated that PCR-DGGE short sequences and clone libraries from three stages of *Haemonchus contortus* life cycle (eggs, L₃s, and adults) contained sequences belonging to *Weissella*, *Lactococcus*, *Leuconostoc* and *Streptococcus* [166]. More recent attention has focused on the haemonchine microbiome [167]. The dominant bacterial genera belonged to *Escherichia-Shigella*, *Pseudomonas* and *Ochrobactrum*, which were shared in all the stages of the parasite life-cycle using V3-V4 and V5-V7 amplicons. Moreover, the parasite microbiome could reflect the external micro-organisms (i.e. micro- and macro-habitats).

Conclusion and future directions:-

We have attempted to provide an overview about the high-throughput sequencing and bacteria associated with eukaryotes including parasitic helminths. There is abundant room for further progress in comparing microbiome of different parasitic helminths, which has, and will continue to offer an important parallel goal for the removal of a wide-variety of devastating animal and human diseases.

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Competing interests:-

The authors declare that they have no competing interests.

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RESEARCH ARTICLE

SYNTHESIS AND CHARACTERIZATION OF 16, 17-MEMBERED TETRA AZA MACROCYCLIC COMPLEXES.

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Abstract

Oxo tetra aza macrocyclic transition metal complexes $[M(C_{15}H_{25}N_5O)Cl_2]$ or $[M(C_{16}H_{27}N_5O)Cl_2]$; M= Co(II), Ni(II), Cu(II) and Zn(II) have been synthesized by the template condensation reaction of O-amino benzoic acid, triethylene tetramine, and 1,3-dibromo propane or dibromo methane in methanol medium. All the complexes have been characterized on the basis of elemental analysis, IR, NMR, EPR, UV-vis, conductivity and magnetic moment data. All the complexes are non ionic in nature and exhibit an octahedral geometry.

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Introduction:-

The pioneering work on metal template synthesis by Curtis⁽¹⁾ and Busch⁽²⁾ exposed the field of macrocyclic chemistry as the major area of research interest. Polyamide macrocycles are of particular interest in view of their two possible donor sites, nitrogen and oxygen atoms. Further more, many of polyamide macrocyclic complexes function like porphyrins in catalyzing organic oxidation reactions⁽³⁻⁸⁾. Amide macrocycles bear the dual structural features of macrocyclic tetra amines and oligopeptides and can stabilize higher oxidation states of some of the metal ions^(9,10). Some of these compounds have been used as metal ion carriers^(11,12). Transition metal(II) complexes of amide macrocycles have interesting properties and can be considered as models for metalloproteins and oxygen carriers^(13,14). The present work describes the synthesis and characterization of a new series of oxotetraamine macrocyclic complexes obtained by the template reaction of o-aminobenzoic acid, triethylene tetramine and dibromo propane.

Material and methods

o-Amino benzoic acid, 1,3-diamino propane, diamino ethane and triethylene- tetra amine (all E. Merck were used as purchased. The metal salts were AR grade reagents obtained from BDH.

Preparation of dichloro (3-oxo-1,2-benzo-4,7,10,13-16 penta aza cyclo-hexadecane) metal(II) $[ML^1 Cl_2]$ [M = Co^{II}, Ni^{II}, Cu^{II} and Zn^{II}]

To a methanolic solution of metal salt (0.01 mol) were added simultaneously triethylenetetra amine (0.01 mol) and o-aminobenzoic acid (0.01 mol) through the each neck of a two-necked round bottom flask. The resultant mixture was stirred for about 7 hrs and then dibromo ethane was added. The stirring was continued for another 3 hrs which resulted in the formation of the title compounds. The title compounds were isolated by filtration, washed several times with methanol and then with Et₂O and dried in vacuo. The purity of the final products was confirmed by TLC as follows: The complexes were dissolved in DMF using EtOAc (80%) MeOH (15%) and MeCO₂H as eluent. A single spot was seen in each case on developing in an iodine chamber.

Preparation of dichloro (3-oxo-1,2-benzo-4,7,10,13,17 pentaazacyclohepta) decane metal(II), $[ML^2 Cl_2]$ [M = Co^{II}, Ni^{II}, Cu^{II} and Zn^{II}]

The procedure used for these compounds was similar to the above method except that 1,3-dibromo propane was added instead of dibromo ethane in the final step.^(15,16)

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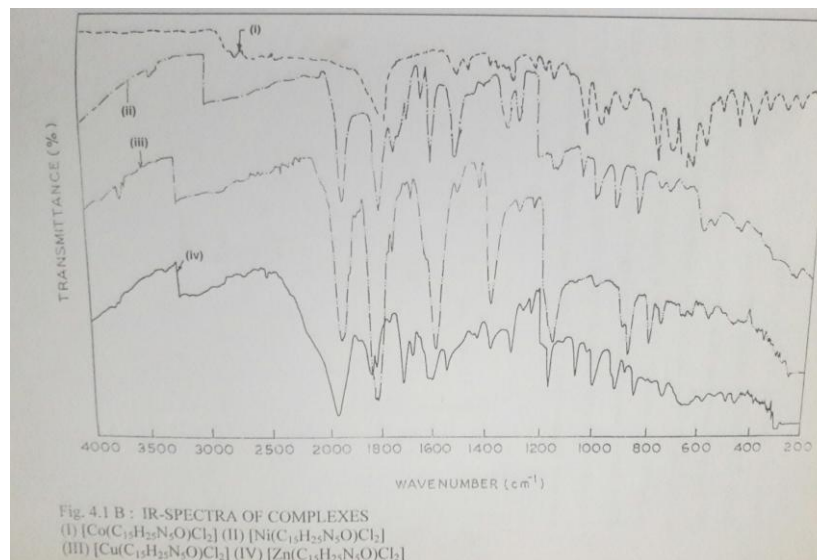
Address:- S.S.M.V. (Pg) College, Shikarpur Bulandshahr, India.

Results and Discussion:-

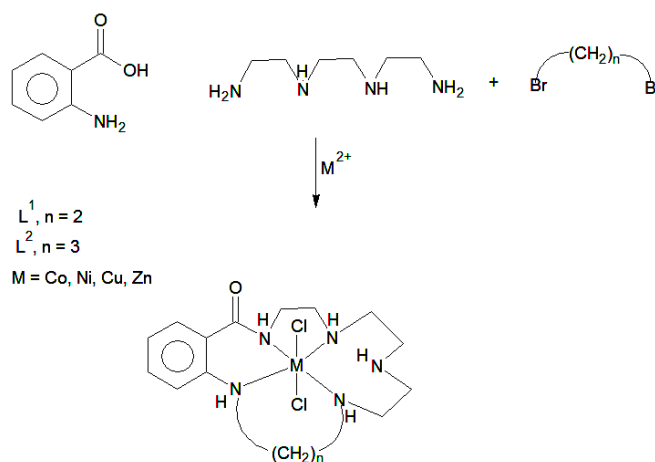
The template condensation reaction of triethylene tetraamine, o-aminobenzoic acid and dibromo alkane resulted in the formation of macrocyclic complexes as shown in Scheme I. All these compounds were obtained as polycrystalline solids. They are freely soluble in dimethyl sulphoxide, dimethylformamide and acetonitrile. The results of elemental analysis are consistent with their proposed 1:1 (metal to ligand) stoichiometry. All the complexes exhibited low molar conductivity values in DMSO ($13\text{--}20\text{ ohm}^{-1}\text{ cm}^2\text{ mol}^{-1}$) which support⁽¹⁷⁾ the non-ionic nature of these compounds.

IR-Studies:-

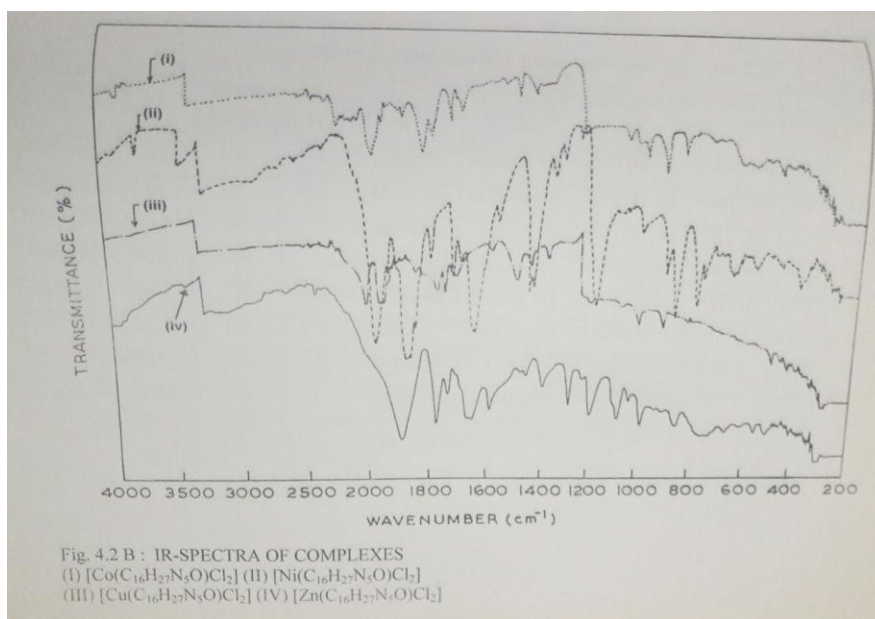
The IR spectra of all the complexes exhibited a single sharp band in the region $3250\text{--}3270\text{ cm}^{-1}$ which may be assigned to coordinated NH stretching vibration. This information together with the appearance of four bands in the ranges $1680\text{--}1710$, $1530\text{--}1570$, $1250\text{--}1260$ and $650\text{--}670$.



cm^{-1} assignable⁽¹⁸⁾ to amide I ($\nu\text{C=O}$); amide II ($\nu\text{C-N} + \delta\text{NH}$), amide III (δNH) and amide IV ($\phi\text{C=O}$) bands respectively, suggests the formation of the proposed macrocyclic framework. The absorption bands appearing in the region $2880\text{--}2930$ and $1410\text{--}1460\text{ cm}^{-1}$ in all the ligands may reasonably be assigned to the CH stretching and CH bending vibrational modes, respectively. The appearance of a low medium intensity band in the region $410\text{--}450\text{ cm}^{-1}$ assignable to $\nu(\text{M-N})$ further confirms the involvement of nitrogen in coordination. However, no complex gave bands assignable to primary amines or carboxylic acid groups.



SCHEME - 1

**Table 1:-** Characterization data of the compounds.

S. No.	Compound	Melting point (°C)	$\mu_{\text{eff.}}$ (B.M.)	Found (Calcd.), %				
				M	Cl	C	H	N
1.	$[\text{Co}(\text{C}_{15}\text{H}_{25}\text{N}_5\text{O})\text{Cl}_2]$	140	4.03	13.82 (13.99)	16.70 (16.86)	42.61 (42.76)	5.80 (5.93)	16.50 (16.64)
2.	$[\text{Ni}(\text{C}_{15}\text{H}_{25}\text{N}_5\text{O})\text{Cl}_2]$	145	3.10	13.80 (13.95)	16.70 (16.87)	42.70 (42.78)	5.80 (5.94)	16.50 (16.63)
3.	$[\text{Cu}(\text{C}_{15}\text{H}_{25}\text{N}_5\text{O})\text{Cl}_2]$	163	1.72	14.80 (14.93)	16.53 (16.68)	42.20 (42.29)	5.73 (5.87)	16.32 (16.44)
4.	$[\text{Zn}(\text{C}_{15}\text{H}_{25}\text{N}_5\text{O})\text{Cl}_2]$	170	—	15.19 (15.29)	16.50 (16.61)	42.0 (42.11)	5.70 (5.84)	16.27 (16.37)
5.	$[\text{Co}(\text{C}_{16}\text{H}_{27}\text{N}_5\text{O})\text{Cl}_2]$	145	4.09	13.40 (13.55)	16.12 (16.32)	44.0 (44.14)	6.10 (6.20)	15.90 (16.09)
6.	$[\text{Ni}(\text{C}_{16}\text{H}_{27}\text{N}_5\text{O})\text{Cl}_2]$	154	3.10	13.39 (13.50)	16.20 (16.33)	44.0 (44.14)	6.10 (6.21)	15.92 (16.10)
7.	$[\text{Cu}(\text{C}_{16}\text{H}_{27}\text{N}_5\text{O})\text{Cl}_2]$	162	1.73	14.30 (14.45)	16.0 (16.15)	43.60 (43.68)	6.0 (6.14)	15.78 (15.92)
8.	$[\text{Zn}(\text{C}_{16}\text{H}_{27}\text{N}_5\text{O})\text{Cl}_2]$	173	—	14.70 (14.81)	15.89 (16.08)	43.40 (43.50)	6.0 (6.11)	15.69 (15.85)

NMR-Studies:-

The ^1H NMR spectra of zinc macrocyclic complexes showed a multiplet, in the region 8.41–8.56 ppm which can be assigned⁽¹⁹⁾ to amide (NH–CO) protons. This clearly shows that the condensation of o-aminobenzoic acid and triethylene tetraamine, has indeed taken place. A multiplet appearing in the region 3.35–3.61 ppm may be ascribed to the methylene protons (CO–N–CH₂). The complexes showed a multiplet in the region 6.98–7.26 ppm corresponding to the phenyl ring protons. Another multiplet observed for all the complexes in the region 2.58–2.60 ppm may be assigned to the methylene proton between the nitrogen atoms (–CH₂–N). The spectra of all the complexes showed multiplet in the region 6.21–6.40 ppm which corresponds⁽²⁰⁾ to the secondary amino protons (C–NH). No compound exhibited signal corresponding to the carboxylic or secondary amino protons, which supports the proposed structure shown in Scheme 1.

EPR-Studies:-

The EPR spectra of polycrystalline copper(II) macrocyclic complexes at room temperature show a single band with g_{\parallel} and g_{\perp} values 2.24, 2.30 and 2.13, 2.09 respectively. None of the complexes exhibited hyperfine splitting which may be due to the fact that the paramagnetic centres are not diluted. The existence of $g_{\parallel} > g_{\perp}$ values 2.24, 2.30 and 2.13, 2.09 respectively. None of the complex exhibited hyperfine splitting which may be due to the fact that the paramagnetic centres are not diluted. The existence of $g_{\parallel} > g_{\perp}$ suggests⁽²¹⁾ that dx^2-y^2 is the ground state. The g values are related to the axial symmetry parameters, G by the following expression, $G = (g_{\parallel} - 2)/(g_{\perp} - 2)$. The G values measure the extent of exchange interaction between copper centres in the polycrystalline solid. If $G > 4$, exchange interaction is negligible, and if $G < 4$, considerable exchange interaction exists in the solid complexes⁽²²⁾. In the present case, G values were found to be 1.85 and 3.33.

Electronic Spectral Studies:-

The electronic spectra of cobalt complexes exhibited two ligand field bands in the ranges 13800–14000 and 22200–22300 cm^{-1} which are assignable^(23,24) to ${}^4\text{T}_{1g}(\text{F}) \rightarrow {}^4\text{A}_{2g}(\text{F})$ and ${}^4\text{T}_{1g}(\text{F}) \rightarrow {}^4\text{T}_{1g}(\text{P})$ transitions respectively in an octahedral geometry⁽²⁵⁾. The appearance of two bands in the electronic spectra of nickel complexes in the ranges 11550–11600 and 17500–17650 cm^{-1} may be assigned⁽²⁵⁾ to ${}^3\text{A}_{2g}(\text{F}) \rightarrow {}^3\text{T}_{1g}(\text{F})$ and ${}^3\text{A}_{2g}(\text{F}) \rightarrow {}^3\text{T}_{1g}(\text{P})$ transitions, respectively suggesting an octahedral geometry for Ni^{2+} complexes. The electronic spectra of copper complexes exhibited two well-discernible bands in the ranges 16400–16700 and 18800–19100 cm^{-1} assignable^(26–28) to ${}^2\text{B}_{1g} \rightarrow {}^2\text{B}_{2g}$ and ${}^2\text{B}_{1g} \rightarrow {}^2\text{E}_{2g}$ transitions, respectively which are characteristic of an octahedral geometry. The high intensity bands observed around 37000 cm^{-1} for all the complexes in the ultraviolet region of the spectra may be assigned as metal to ligand charge transfer transitions. The magnetic moment data (Table 1) for all the complexes are consistent with the high spin octahedral geometry around these metal ions which further supports the electronic spectral results.^(29–31)

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RESEARCH ARTICLE

INTER-DISTRICT DISPARITIES IN INDUSTRIAL GROWTH OF ASSAM.

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Abstract

The level of industrial development across different districts of the state has been worked out with the help of composite indices. The composite indicators are obtained with the help of two different methods. The district-wise data in respect of forty three indicators are used for twenty three districts of the State. The data on most of the indicators are for the year 2013-2014. Ranks of the districts in the level of industrial development are obtained from both the methods, which are later compared. It is observed that there is no significant difference between the ranks obtained from the two methods used. The study has brought out the huge disparities that exist among the districts which have accentuated over a period of time. The district Kamrup is ranked first and the district Hailakandi is ranked last. For bringing out uniform regional development, potential targets have been estimated for low developed districts. These districts require improvements of various dimensions in some of the indicators for enhancing the level of overall socio-economic development.

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Introduction:-

The problem of inter-district disparity in the levels of development is not of recent origin and is almost universal. Its extent may differ in different economics but its existence can hardly be challenged. Economic development is a concern of all societies as well as the nations. During the course of economic development, the determinants of developmental process shape up in the form of agrarian transformation towards non-agricultural sectors. Also, at some stage of economic development, increasing urbanization, improved quality of infrastructure, increased standard of living of human beings and the improved quality of overall governance take place. However, in this process of development, some regions of the economy grow faster whereas others tend to lag behind.

Assam had a great legacy of modern industrial development. Along with tea, oil refineries and coal mines were developed post-independence. A number of major industries based on the rich mineral and forest resources were set up under the central public sector. Fertilizer industry, Namrup; Cement factory, Bokajan and Paper mill at Jagiroad etc. were also set up. Assam produces three unique varieties of silk; the golden muga, the white pat and warm eri. Sericulture is one of the biggest contributors of state income. It is practiced in more than 9373 villages and provides employment to 2.4 lakh families. Eri contributes Rs 31.5 crores where as muga contributes Rs 40 crores and pat silk contributes Rs 120 crores out of total Rs 190 crores generated annually through the silk industry in Assam. The Assam Handloom Industry is known for its tradition of making handloom and handicraft products. At present, 11570 villages of the state with 1319754 weavers are covered by the directorate of Handloom and Textiles for its

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handloom activities. The Small Scale Industrial (SSI) Sector is a vital constituent of the total industrial sector. Electricity, good roads and transport system are the key indicators of development in a region. A better transport system increase income, employment opportunities, tourism, and industrial development in many ways. But there are differences in all the districts of the State as far as industrial growth is concerned. All the districts of the State are not equally developed. So, we have made an attempt to throw light on the developmental disparities in industrial growth for twenty three districts of the State. Development is a process which improves the quality of life. Its impact cannot be fully captured by a single indicator. Hence there is need for building up of a composite index of development, combined in an optimum manner. A deep analysis using the district level data on socio-economic indicators was made by Narain et. al. for the States Orissa [1992-1993], Andhra Pradesh [1994], Kerala [1994] Uttar Pradesh, [1995] Maharashtra [1996], Karnataka [1997], Tamil Nadu [2000], Madhya Pradesh [2002]. Similar study had been made by Borah and Nath for Assam [2004, 2010]. In all, the study for evaluating the level of socio-economic development was conducted in two hundred twenty eight districts belonging to the states of Andhra Pradesh, Karnataka, Kerala, Madhya Pradesh, Maharashtra, Orissa, Tamil Nadu and Uttar Pradesh and it was found that 73 districts were low developed which require special attention for undertaking future developmental programmes.

Methods of Analysis:-

Development is a multi-dimensional continuous process. For this study, the districts have been taken as the unit of analysis. Twenty three districts of the state of Assam are included in the study. Two methods have been separately used to rank the districts of the state, viz. Narain et. al. method and Michela et. al. method.

Narain Et. Al. Method:-

Let a set of n points represent districts $1, 2, \dots, n$ for a group of indicators $1, 2, \dots, k$, which can be represented by a matrix (X_{ij}) ; $i = 1, 2, \dots, n$ and $j = 1, 2, \dots, k$. As the developmental indicators included in the analysis are in different units of measurement and since our objective is to arrive at a single composite index relating to the dimension in question. There is a need for standardized as shown below [Narain et. al., 1991]:

$$Z_{ij} = \frac{X_{ij} - \bar{X}_j}{S_j},$$

$$S_j^2 = \frac{\sum_{i=1}^n (X_{ij} - \bar{X}_j)^2}{n}$$

and

$$\bar{X}_j = \sum_{i=1}^n \frac{X_{ij}}{n} \quad (i=1,2,\dots,n), \quad (j=1,2,\dots,k)$$

Let $[Z_{ij}]$ denote the matrix of standardized indicators. The best district for each indicator (with maximum/minimum standardized value depending upon the direction of the indicator) is identified and from this the deviations of the value for each district has been taken for all indicators in the following manner:

$$C_i = \left(\sum_{j=1}^k (Z_{ij} - Z_{0j})^2 \right)^{1/2},$$

where Z_{0j} the standardized value of the j th indicator of the best district and C_i denotes the pattern of development of i^{th} district.

The pattern of development is useful in identifying the districts which serve as 'models' and it also helps in fixing the potential target of each indicator for a given district. In this study, the composite index of development is obtained through the following formula:

$$D_i = \frac{C_i}{C},$$

$$C = \bar{C} + 2S$$

$$\text{where } \bar{C} = \sum_{i=1}^n \frac{C_i}{n}$$

$$S = \left(\sum_{i=1}^n \frac{(C_i - \bar{C})^2}{n} \right)^{1/2}$$

and

“ D_i ” gives the composite index of development with which ranking of the districts is done.

Michela Et. Al Method:-

Theoretical frame work and methodology is followed from [14]. The aggregated values give the composite index of development to rank the districts. A theoretical framework should be developed to provide the basis for the selection and combination of single indicators into a meaningful composite index. The indicators should be selected on the basis of their analytical soundness, measurability, country coverage, relevance to the phenomenon being measured and relationship to each other. The use of proxy variables should be considered when data are scarce.

A multivariate analysis should be done to investigate the overall structure of the indicators, assess the suitability of the data set and explain the methodological choices. The next step is normalization in which the indicators should be normalized to render them comparable.

$$Z_{ij} = \frac{X_{ij} - \bar{X}_j}{S_j}$$

A correlation study is done to find the redundancy in the indicators, and the correlation co-efficient is given by:

$$r_{z_i, z_j} = \frac{\sum (z_i - \bar{z}_i)(z_j - \bar{z}_j)}{\sqrt{\sum (z_i - \bar{z}_i)^2} \sqrt{\sum (z_j - \bar{z}_j)^2}}$$

We discard the indicators having high correlation co-efficient with other indicators and as such the number of indicators reduces.

Finally weighting and aggregation is done in which the indicators should be aggregated and weighted according to the underlying theoretical framework.

$$A_i = \sum_j z_{ij}$$

The aggregated values give the composite index of development to rank the districts.

Relative Share of Area and Population under Different Level of Development:-

A simple ranking of district on the basis of composite indices is sufficient but a suitable classification of districts formed on the basis of mean and standard deviation of the composite indices will provide a more meaningful characterization of various stages of development. For relative comparison it appears appropriate to assume the districts having composite index less than or equal to (Mean - SD) as highly developed districts. And the districts having composite index greater than or equal to (Mean + SD) be low developed districts. Similarly districts with composite index lying between (Mean and Mean - SD) are classified as middle level developed and district with composite index lying between (Mean and Mean + SD) are classified as developing districts.

Fixation of Potential Targets:-

Using the standardized variates $[Z_{ij}]$, the economic distance between different districts may be obtained as follows:

$$D_{ip} = \left(\sum_{j=1}^k (Z_{ij} - Z_{pj})^2 \right)^{1/2},$$

(i=1,2,...,n and p=1,2,...,n).

Here $D_{ii} = 0$ and $D_{ip} = D_{pi}$.

The distance matrix will take the form:

$$\begin{bmatrix} 0 & d_{12} & d_{13}\dots & d_{1n} \\ d_{21} & 0 & d_{23}\dots & d_{2n} \\ & & \cdot & \\ d_{n1} & d_{n2} & d_{n3}\dots & 0 \end{bmatrix}.$$

The minimum distance for each row, (d_i , $i=1,2,\dots,n$) will be obtained from the distance matrix for computation of upper and lower limits (C.D.) as indicated below:

$$C. D. = \bar{d} \pm 2\sigma_d,$$

where

$$\bar{d} = \sum_{i=1}^n \frac{d_i}{n},$$

and

$$\sigma_d = \left(\sum_{i=1}^n \frac{(d_i - \bar{d})^2}{n} \right)^{1/2}.$$

The distance matrix can also be used for fixing targets for different districts on each indicator, which would be in the direction of reducing the disparities. The districts should be identified which are homogeneous with a close proximity to each other with the district under consideration, in terms of considered indicators. For setting out the targets, the model districts are to be identified on the basis of composite index and individual distance with districts. The best values among the model districts will be taken as potential target for a particular district for a given indicator. This procedure will be repeated for a given district for all indicators considered. This would give the extent of improvement required in different indicators for balanced development in the district. It also provides avenues to bring about uniform regional development in the state. Such information helps the planners and administrators to readjust the resources to reduce inequalities in level of development among different districts of the state.

The study utilizes data on most of the industry sector indicators for the year 2013-2014. A total of forty three development indicators have been included in the study.

Developmental Indicators:-

Each district faces situational factors of development unique to it as well as common administrative and financial problems. The composite indices of development for different districts have been obtained by using the data on the following indicators. The different indicators are classified into the following categories [16].

[X] Handloom

x_1 : No. Of HTC

x_2 : No. Of trainees in HTC

x_3 : No. Of WESU

x_4 : Production of WESU

x_5 : No. of HDC

x_6 : Production of HDC

x_7 : No. of weavers engaged in HDC, part time

x_8 : No. of weavers engaged in HDC, full time

[Y] Sericulture

- y₁ : No. of Sericulture village
- y₂ : No. of family engaged in eri
- y₃ : No. of family engaged in muga
- y₄ : No. of family engaged in mulberry
- y₅ : Total area under silkworm food plants (eri)
- y₆ : Total area under silkworm food plants (muga)
- y₇ : Total area under silkworm food plants (mulberry)
- y₈ : Yield of Eri Cocoons
- y₉ : Yield of Muga Cocoons
- y₁₀ : Yield of Mulberry Cocoons
- y₁₁ : Production of silkyarn (eri raw silk)
- y₁₂ : Production of silkyarn (muga raw silk)
- y₁₃ : Production of silkyarn (mulberry raw silk)

[Z] SSI

- z₁ : Achievement of khadi and village industry (unit functioning)
- z₂ : Achievement of khadi and village industry (production)
- z₃ : Achievement of khadi and village industry (employment)
- z₄ : Number of registered bill fisheries.
- z₅ : Production of fish seed
- z₆ : Total number of MSME units
- z₇ : Total workers of MSME units
- z₈ : Number of registered factories
- z₉ : Number of registered workers
- z₁₀ : Production of rubber in area.
- z₁₁ : Production of rubber
- z₁₂ : Tapping area.
- z₁₃ : People in household industry (main worker)
- z₁₄ : People in household industry (marginal worker)

[V] Infrastructure

- v₁ : State High way in km
- v₂ : Major district road in km
- v₃ : Rural road in km
- v₄ : Urban road in km
- v₅ : Total road length
- v₆ : Road length in km per lakh population
- v₇ : Road length in km per (00) sq km of geographical area
- v₈ : Number of villages electrified.

Comparison of Ranks:-

We have used Spearman rank correlation co-efficient to test if there is any significant difference in the ranks obtained by the two methods. The rank correlation co-efficient is given by Ronald et. al.(1985)

$$r_s = 1 - \frac{6 \sum_{i=1}^n d_i^2}{n(n^2 - 1)}$$

where d_i is the difference between the ranks assigned by the two methods and n is the number of pairs of data [15].

Results and Discussions:-

The Level of Development:-

The districts have been ranked on the basis of development indices. Table-1 presents the composite indices of development along with the ranks of different districts. It may be seen from the above table that out of 23 districts of the State, the district Kamrup is ranked first whereas the district Hailakandi is ranked last in the overall industrial growth.

Table 1:- Ranks of all the districts of Assam obtained from the two methods.

DISTRICTS		INDUSTRY									
		HANDLOOM		INFRASTRUCTURE		SERICULTURE		SSI		OVERALL	
		N	M	N	M	N	M	N	M	N	M
1.	KOKRAJHAR	23	19	12	14	6	1	22	20	4	18
2.	DHUBRI	20	16	16	17	23	23	7	7	20	21
3.	GOALPARA	9	9	15	13	16	10	15	3	10	12
4.	BARPETA	3	1	9	10	17	18	9	12	11	4
5.	MARIGAON	7	14	21	19	15	16	14	14	14	8
6.	NAGAON	15	3	2	2	8	12	2	2	2	5
7.	SONITPUR	16	4	4	6	9	13	3	8	6	11
8.	LAKHIMPUR	12	11	17	18	1	2	18	21	8	10
9.	DHEMAJI	14	22	14	12	2	4	23	22	16	16
10.	TINISUKIA	21	17	11	11	11	15	6	11	15	19
11.	DIBRUGARH	6	10	10	9	4	8	5	10	9	3
12.	SIVSAGAR	5	8	6	4	5	3	11	15	5	6
13.	JORHAT	18	15	8	8	3	5	8	9	7	14
14.	GOLAGHAT	10	12	5	7	13	9	12	19	13	13
15.	K.ANGLONG	13	6	1	1	14	7	19	4	3	15
16.	D.HASAO	22	18	7	3	20	17	13	23	21	23
17.	CACHAR	11	13	18	20	12	14	4	6	14	9
18.	KARIMGANJ	17	21	19	21	18	22	10	5	18	20
19.	HAILAKANDI	19	23	23	23	19	21	21	17	23	22
20.	BONGAINGAON	8	20	22	22	21	19	20	16	22	17
21.	KAMRUP	1	2	3	5	7	6	1	1	1	1
22.	NALBARI	2	5	20	16	22	20	17	18	17	2
23.	DARRANG	4	7	13	15	10	11	16	13	12	7

* N denotes Narain et. al. method and M denotes Michela et. al. method.

It is seen that, for most of the districts, ranks calculated by the two methods are almost same whereas for a few other districts, ranks calculated by the two methods are different.

We have tested the hypothesis that the correlation between the ranks obtained by Narain *et. al.* method and the Michela *et. al.* method is zero against the alternative hypothesis that it is greater than zero. At both 0.01 and 0.05 level of significance, it is observed that the two methods are correlated and there is no significance difference between the ranks obtained from the two methods.

Area and Population in Different Stages of Development:-

It would be quite interesting and useful to find out the relative share of area and population affected under different levels of development in the State. The area and population covered by the districts falling under different levels of development are presented in Table 2.

Table 2:-Area and Population under Different Levels of Development

Level of Development	No. of Districts	Population (%)	Area (%)
High (≤ 0.679)	3	17.7	23.8
Medium (0.679-0.786)	7	32.8	27.4
Developing (0.786-0.892)	10	41.7	37.7
Low (≥ 0.892)	3	7.8	11.1

It is evident from the table that about 23.8% area consisting of about 17.7% population of the State fall in the districts which are high developed. About 11.1% area and 7.8% population fall in the districts which are low developed in the industrial sector. The low developed districts which have been found in this study are Hailakandi, Bongaingaon and Dima-Hasao. List of model districts for these low developed districts is presented in Table-3.

Table 3:-Model Districts for Low-developed Districts

S. No.	Low Developed Districts	Model Districts
1.	Hailakandi	Morigaon, Kamrup, Golaghat, Nalbari
2.	Bongaingaon	Morigaon, Kamrup, Nagaon, Cachar
3.	Dima-Hasao	Kamrup, Karbi-Anglong, Cachar, Nalbari, Nagaon

The districts Kamrup, Nagaon and Morigaon are found to be the model districts for most of the low developed districts.

Potential Targets of Indicators for Low Developed Districts:-

It would be useful to examine the extent of improvements required in different indicators of the low developed districts for enhancing the level of development. The best values of the indicators of better developed districts will be taken as potential targets for the low developed districts. The extent of improvement needed in various indicators of the low developed districts is given below:

Table 4:-Estimate of Potential Target and Actual achievement (given under the bracket).

Indicator s	Hailakandi	Bongaingaon	Dima-Hasao	Indicators	Hailakandi	Bongaingaon	Dima-Hasao
x ₁	238.4(16.9)	1709.5(12.55)	1709.5(14.52)	z ₂	3007.1(3369.8)	3007.76(2943.95)	3007.7(163.3)
x ₂	2549(1000)	2549(1815)	2549(607)	z ₃	19392(9358)	24262(5800)	24262(280)
x ₃	672(90)	672(29)	672(21)	z ₄	12(0.2)	10(0.4)	10(0.9)
x ₄	230(99)	326(16)	326(199)	z ₅	1(0.45)	0.9(0.75)	.69(.12)
x ₅	5(1)	8(3)	8(.4)	z ₆	620.02(326.25)	620.02(321.62)	620.02(43.25)
x ₆	100(15)	122(27)	122(40)	z ₇	185(25.95)	90.14(33.15)	90.14(90.23)*
x ₇	7(2)	7(2)	7(5)	z ₈	234.2(71.75)	213.22(109.88)	213.2(878.57)*
x ₈	11(5)	17(5)	17(11)	z ₉	95.59(35.6)	92.86(46.66)	95.59(38.38)
y ₁	1807(98)	1807(59)	1807(46)	z ₁₀	2916(351)	2916(712)	2916(1237)
y ₂	10829(2950)	10829(2996)	10829(5045)	z ₁₁	160(17)	297(41)	297(380)*
y ₃	1850(98)	1850(59)	1850(46)	z ₁₂	89.4(49.14)	675.25(462.3)	675.25(38.24)
y ₄	84(35)	134(47)	134(1)	z ₁₃	489.6(180)	584.9(198.6)	584.9(358.8)
y ₅	1343(39)	1343(64)	1343(26)	z ₁₄	868(243)	868(259)	868(178)
y ₆	42770(2957)	42770(5331)	42770(617)	v ₁	350(205)	375(265)	375(285)
y ₇	1304(306)	1327(844)	1304(496)	v ₂	183(47)	183(36)	183(2)
y ₈	695(357)	675(903)*	1327(198)	v ₃	3235(473)	3235(805)	3235(1876)
y ₉	72430(10570)	724.3(14929)	72430(390)	v ₄	1681(263)	1681(222)	1681(16)
y ₁₀	551(245)	1185(156)	1185(145)	v ₅	33(6)	88(36)	88(60)
y ₁₁	106803(12000)	93020(38543)	106803(13945)	v ₆	600(10)	500(20)	500(45)
y ₁₂	39654(4049)	39654(4729)	39654(742)	v ₇	9(4.05)	8.1(6.75)	8.1(1.04)
y ₁₃	29452(3264)	29452(4756)	29452(750)	v ₈	240.5(33.74)	117.18(43.36)	117(117.68)*
z ₁	2150.8(1300.8)	575775(900)	575775(1400)				

*Indicates actual achievement that already better than the potential target.

It is found in the industrial growth sector, that the districts of Kamrup, Nalbari, Dibrugarh and Sibsagar are high developed districts. The districts of Hailakandi, Dhubri and Dima-Hasao are low developed districts. The remaining are middle developed and developing districts.

Conclusion:-

The broad conclusions emerging from the study are as follow:

Two ranking methods, viz. Narain *et. al.* and Michela *et. al.* are used to rank the districts of Assam on the basis of industrial development of the state. It is observed that both the methods gave almost the same ranking. A ranking test is carried out and it is observed that there is no significant difference between the two methods.

With respect to overall development in the industrial progress, the districts of Kamrup, Karbi-Anglong and Nagaon are found to be better developed as compared to the remaining districts of the State. Similarly the districts of Hailakandi,

Bongaingaon and Dima-Hasao are low developed districts. The level of development in the rest of the districts is of average order but most of these districts are having the tendency to make improvements in the pattern of development.

As compared to the previous study, this study reveals significant changes in the status of the districts Nagaon, Kokrajhar and Karbi-Anglong by improving the ranks and Dibrugarh, Golaghat and Tinisukia by deteriorating their ranks, which may be understood in the light of two different parameters; i) introduction of SSI as one new parameter, ii) improving the status of existing parameters.

Nagaon and Karbi-Anglong gain their rank as SSI index were 2 and 4 which boosted up the overall rank from 6 and 11 to 2 and 3 respectively. Because of consideration of SSI in the new study which was not in the earlier study. Even both the districts show downfall in the indices of handloom in the later study. For the district of Kokrajhar, the better socio-political scenario boosted up the sericulture productivity indices from 20 to 1 and hence overall tabled at rank 4.

For the district of Dibrugarh, deteriorating sericulture sector and average SSI pulled it back and placed it at the rank 9 in lue of 2 in the previous study. Same is the scenario for the district of Golaghat. For Tinsukia district, deteriorating hand loom sector added with poor sericulture and SSI indices pulls back its rank from 10 to 15.

Despite of all changes in our procedure and timely developmental activities between two studies, Kamrup district holds its original rank.

In order to reduce the disparities, district level studies or setting the objective in the district level may not be a wise idea. So, looking for the potential areas for development in taluka or block level may be of great importance and emphasis on over all developmental indexes will be of good use to reduce the developmental disparities

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RESEARCH ARTICLE

EFFECT OF CRYOGENIC TREATMENT ON TOOL STEELS.

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Abstract

The objective of this review is to understand the effect of cryogenic treatment on tool life and its performance. As cutting tool life is an important factor in reducing the cost of production. In this process of cryogenic treatment, tools are generally subjected to the process such as heat treatment for achieving the hardness which will improve the performance of the tool to some extent. Cryogenic treatment also known as cold treatment, in which the phase transformation of the material takes place, is mainly responsible for changes in the wear resistance, tensile strength, toughness etc. which increases the tool life. From the review it was understood that has great effect on cutting tool properties. Especially soaking period varied from 6 hours to 36 hours are giving different effect on the phase transformation of the material, consider as one of the important aspect for tool life improvement. Various soaking range namely shallow cryogenic treatment (SCT), Cryogenic treatment (CT) and deep cryogenic treatment (DCT) produce different properties in tool material. This treatment or process can be very useful in case of machinability of work-piece, quality of machining in terms of surface finish and economics of the tooling.

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Introduction:-

In metal working industries, the tool materials used for cutting tools are all belong to a separate class of steels called as Tool Steels. Tool steels are called so because of a few very important machinability properties they possess such as 'abrasion resistance' 'resistance to deformation' 'high hardness' 'hot hardness' and so on. Because of these, tool steels are used for applications involving shaping other materials. High quality of the tool steels are achieved through controlled chemical compositions which give them the desired properties for shaping materials. The type of tool steel depends on the percentage of carbon (usually 0.1% to 1.6%) and the alloying elements used. Molybdenum, vanadium, chromium are very commonly used and their benefits are very well documented and widely used in the metal industry. Tool steels are widely used for applications such as die casting, plastic molding, making cutting tools, various sheet metal tooling's such as blanking, shearing, forming and so on. Tool steels offer better choice than common construction and engineering steel materials because of their corrosion resistance, durability, stability at high temperatures.

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In steel making industries there has been special attention set on producing tool steels particularly the high alloyed steels intended for the purpose of plastic molding, forming, blanking, die casting, forging extrusion, recycling and wood- working. Steels are also produced by following the Powder Metallurgy. Tool steels can be procured in soft annealed condition because of which they are easily made into cutting tools. The soft annealed condition tool steel are also well suited for hardening. The microstructure provides pockets to embed the carbide particles.

Generally Iron Carbides Fe_3C are the particles which give high strength to carbon steels. Whereas in alloy steels Tungsten (W), Vanadium (V), Molybdenum (Mo), Chromium (Cr) carbides depending on the composition of the steel. The carbides are characterized by high hardness. The wear resistance improves with the increase in percentage of various carbides and their distribution in the steel crystal structure. Non carbide forming elements when alloyed, get absorbed into the metal matrix. Cobalt and Nickel are one such alloying elements. Cobalt improves the red hardness in high speed steels, nickel improves the hardening properties as well as the toughness in hardened condition. Other inclusions in steel like manganese are kept low to avoid cracking due to thermal load during water quenching

Metal cutting is the backbone of manufacturing industry. Each and every product ever made in history can trace its origin to the basic metal cutting process. The cutting tool therefore is the important element for any metal cutting operation to realize its true potential. Since the last few decades, in a bid to make the processes economic, a lot of effort has been put into the research of metal cutting. The results have thus given birth to newer tool materials with marked improvement in performance thereby improving productivity. Lighter yet stronger tool materials have been the catch words among the cutting tool manufacturers. Some of the other properties which are highly desired are

- Resistance to diffusion.
- Resistance to brittle fracture.
- Ease and economy of fabrication of the tool.
- High resistance to thermal and mechanical shocks.
- Ability to retain strength at high speeds and withstand the heat generated during cutting operation.

The enormous developments taking place in the cutting tool technology is driven by the knowledge about the capabilities of the tool materials to work under different conditions of stress, high temperatures at the interface between tool and work piece. The tool wear occurs by different mechanisms which include thermal cracking, chipping at the cutting edges of the tool and abrasive wear. The tool materials should therefore have good mechanical, physical, chemical properties at such elevated temperatures¹⁻⁴

Tool steel grades:-

Various tool steel grades have been developed over the years and are well established in AISI and SAE tool steel grades. The different alloy tool steels in each of the grades are given a number to be identified with. In total, there are 6 different grades of tool steel material. The choice of the tool steel grade is influenced by the cost, surface hardness required, working temperature, toughness, shock resistance, durability, and strength. If the working conditions are expected to be severe, the respective alloying element is chosen depending on the properties they enhance on the steels.

Tool steels prepared by water quenching are known as water hardening group, these steels can have a maximum hardness of 66HRC. O - series and A - Series are also based on quenching medium used (Oil for O - series and Air for A - series). The Oil hardening series have up to 7 types numbered from O1 to O7 and D - series (High Carbon High Chromium steels) in which the chromium content is up to 13%. Next is the shock resistance alloyed tool steels which are prepared by alloying elements such as chromium, silicon, tungsten, molybdenum, manganese and approximately 0.5 % C is maintained where higher toughness is expected. Most widely used is High speed steels which are used in cutting tools such as turning tool, drill bits, saw blades. These are used predominantly in cutting operations of mild steels and other softer materials. When the materials are to be cut, formed at high temperatures, a special type of tool steels called Hot Working tool steels is used. The hot hardness and wear resistance are provided because of the presence of carbide content of the alloying element. This is the most populated group of tool steels with close to 60 types. The types numbered from H1 to H19 are high chromium content (up to 5%), H20 to H39 are tungsten alloy (9% to 18%), H40 to H59 being molybdenum based. These have low carbon content.

Introduction to Cryogenic Process:-

Cryogenic is a term used to refer to things at very low temperatures. The idea of very low temperatures is not a modern one. It has been used as back in the time as the 16th century

when it was used to enhance some mechanical properties. Cryogenic treatment induced positive effects on the key parameters such as tool performance, tool wear. Mohan et al have achieved a 110% gain in tool life after studying the effects of cryogenic treatments. Its applications are very wide in range as it has been used for ferrous, non - ferrous metal alloys, ceramics, plastics, carbides. In conventional heat treatment methods, the changes take place on the surface or only up to certain depth. Whereas in Cryogenic treatments the changes take place at the basic crystal structure of the material,¹ As a bonus there is no negative influence on the corrosion resistance of the material. The effect of DCT on various other grades of steel like the stainless steel type by conducting Qualitative salt spray corrosion test confirming the same.² Thus Cryogenic treatment has been a very effective process to obtain many desirable properties in metals.

The actual process of Cryogenic treatment is quite simple. The material samples will be cooled to very low temperatures by means of a cooling system regulated by a control unit. Gases like nitrogen, helium are used to cool to extremely low temperatures like -80°C to -196°C . The abundant availability of nitrogen in the atmosphere and its suitability for carrying out cryogenic treatment has made it the quintessential component of Cryogenic Treatment. It has a melting point of -210°C and boiling point of -196°C . The samples will be kept at cryogenic temperatures for some time. This period is called 'holding time'. It is during this holding time that the change to the microstructure takes place. After this the material samples are brought gradually to room temperatures. Figure 1 shows the Schematic presentation of cryogenic treatment applied in a controlled manner³.

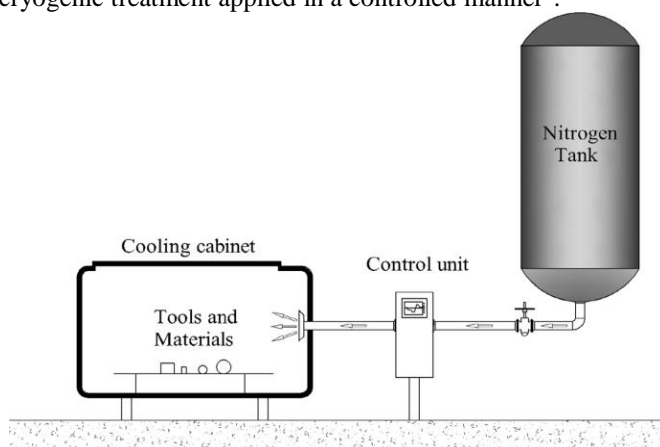


Figure 1. Schematic presentation of cryogenic treatment applied in a controlled manner⁴

The results of cryogenic treatment are obvious and their improvements over untreated samples are astounding. Marked improvements in abrasion resistance, toughness, hardness, electrical conductivity can be seen in cryogenic treated samples. These can be attributed to generally the two underlying mechanism (i) conversion of austenitic structure into martensitic. (ii) Finer distribution of the carbides in the metal matrix. The presence of nanosized carbide particles through the study of microstructures. As a result Cryogenic treatment is widely used to increase the performance of chip breaker tools. When undertaken on bigger scale these methods also offer economy over other methods such as coating. Cryogenic treatments are also applied on cutting tools, race car engines, tool steels, brake discs, composites and on some plastics.⁴ Figure 1 shows the Schematic presentation of cryogenic treatment applied in a controlled manner⁵

Basically the Cryogenic treatments can be broadly classified into two types. (i) Shallow Cryogenic Treatment – SCT & (ii) Deep Cryogenic Treatment – DCT. The treatment in which the cryogenic treatments are kept at -80 to -149°C is called SCT and DCT involves even lower temperatures at -140°C to -196°C . Figure 2 shows the Variations of temperatures during Cryogenic Treatment⁵

Rate of cooling:-

The material should be brought to the cryogenic temperature and then it undergoes soaking at same temperature for period of 2 hrs to 48 hrs depending on requirement, figure 2 shows the variations of temperatures during cryogenic

treatment. In rate of cooling there are two main approaches, in the first approach the samples slowly brought to cryogenic temperature in between (-80°C to -196°C). in the second approach, the samples are slowly brought to cryogenic temperature. According to the study the second approach is recommended as some damages like micro cracks are developed on the sample due to thermal shock.

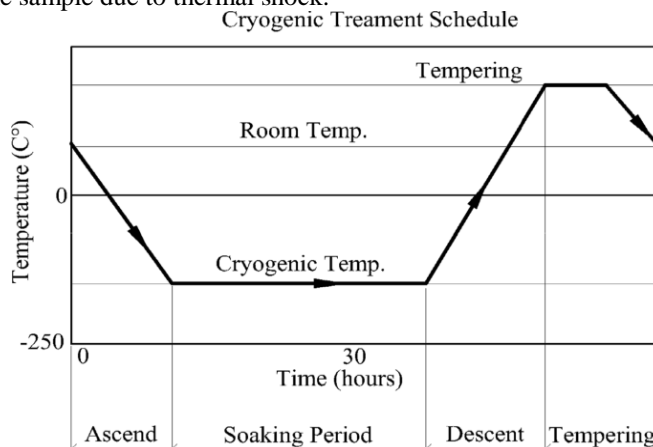


Figure 2. Variations of temperatures during Cryogenic Treatment ⁴

The used carbide cutting tools at two different rates of cooling/heating ($0.5^{\circ}\text{C}/\text{min}$ and $1^{\circ}\text{C}/\text{min}$) and completed the cryotreatment in 8h and 4h and reported that the wear resistance was better in the sample ($0.5^{\circ}\text{C}/\text{min}$, 8h) compared to ($1^{\circ}\text{C}/\text{min}$, 4h). sudden decrease in temperature (thermal shock) reduces the cost of cryotreatment but increases the risk of occurrence of micro cracks in the microstructure of the specimen due to which the specimen may not give better performance. So in cryotreatment the slow cooling/heating process is recommended. ⁶

Soaking Period:-

After the literature review of cryotreatment (DCT) on the tool steels the samples were subjected to soaking period in between 1 and 40 h were the conversion of material from austenite to martensite takes place and the formation of new carbides takes place for carbide distribution. The soaking of Ti-6Al-4V alloy up to 72h (DCT) at -196°C . Therefore the soaking period should be optimized to increase the wear resistance efficiency and reduce the cost of cryogenic treatment. ⁷

Soaking Temperature:-

Cryogenic treatment also known as sub-zero heat treatment shows the variation according the type of tool steel material. Cryogenic treatment temperatures applied to cutting tools range between -80°C and -196°C . Taking as -80°C - Cryogenic treatment, -120°C - Shallow Cryogenic treatment and -196°C - Deep Cryogenic treatment. The study shows that to increase the wear resistance to the samples the soaking temperature plays an important factor. From the literature studies it is found that -196°C (DCT) gives the best result in the improvement of mechanical properties of the specimen.

Tempering Process:-

Tempering is usually done after the cryogenic treatment. The process is mainly performed to remove the internal stresses of the specimen that occur due to excessive cooling in cryogenic treatment. Generally, the process is applied to cutting tools by holding the specimen at $150-200^{\circ}\text{C}$ for 1.5–2 h. In some previous studies has under gone double tempering after cryogenic treatment and a fine distribution of Carbide particles have been obtained by double Tempering after CT ⁸ as represented in Figure 3. In some studied the results show that Tempering indeed maximizes the gain in tool life after DCT (86% without Tempering, 126% with Tempering). ⁹ through investigations have found that tempering before actually decreases wear resistance as against increasing it. On a hind note it has been observed that multiple cycles of Tempering after DCT hasn't improved on the wear resistance, but has a negative impact. Also there has been decrease in wear resistance after multiple cycles of Tempering, compared to CT and HCT samples. ¹⁰

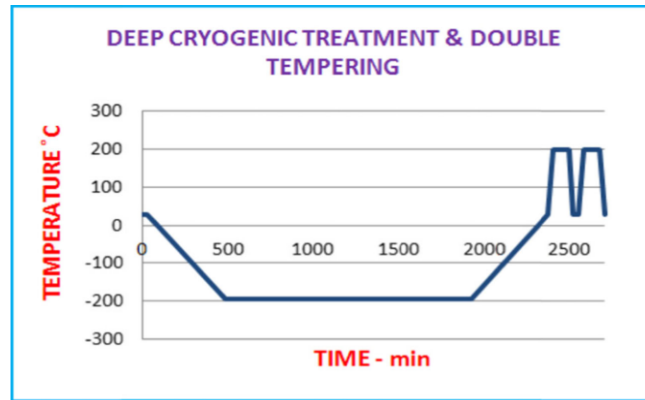


Figure 3. Variations of temperatures during the Tempering process, after Cryogenic Treatment ⁶

Results and Discussion:-

Tool life:-

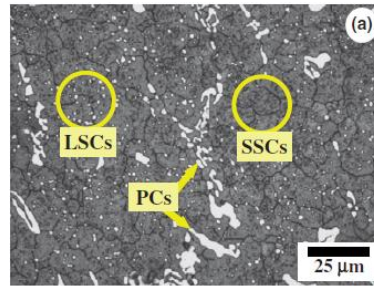
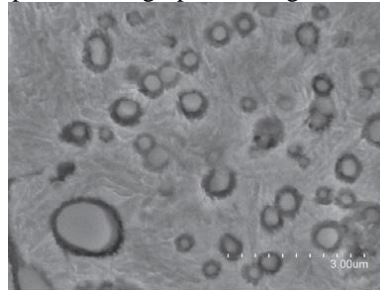
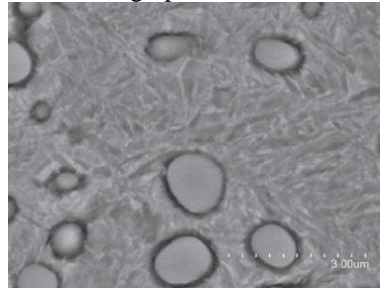
The actual length of the cutting time that the tool can be used is known as tool life. Generally the tool failure happens due to sudden breakage of tool tip or due to the gradual wear of the tool is known as flank wear or crater wear, the tool wear is mainly caused due to increasing cutting forces and cutting temperature, which leads to the poor surface finish and accuracy of the components. The tool life was found increasing generally by the research conducted there was the improvement of about 86% for M2 tool steel and 48% for D3 tool steel compared to heat treated tool steel.¹²

There are mainly three stages of flank wear rapid growth region, steady state region (Temperature insensitive region) and Catastrophe failure (Temperature sensitive region). Flank wear formation mainly depends on cutting condition i.e. Parameters like speed, feed and depth of cut as well as the properties of the work and tool material.

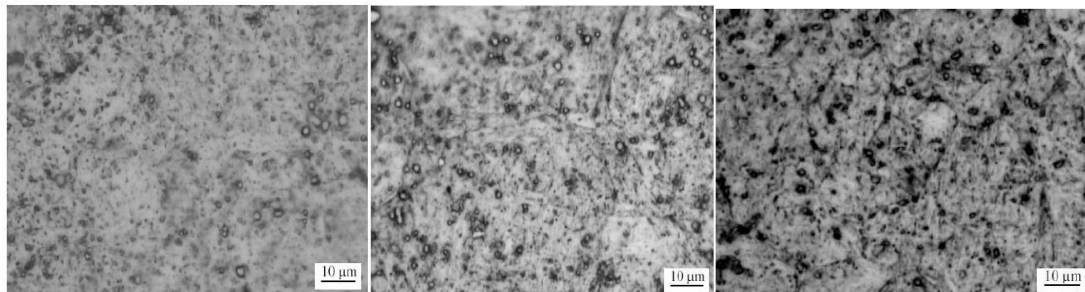
Microstructure:-

A detailed study of the mechanism which improves the mechanical properties in Deep cryotreated materials has been provided where presence of different alloyed carbide has been absorbed in the sample even before, have also reported about various carbides being present.^{13,14,15} The carbides have been classified into primary carbides and secondary carbides. Primary carbides are large, spherical in shape and their sizes are in excess of 5µm whereas secondary carbides are in irregular shapes. Secondary carbides are further classified into small (i) Secondary carbides (SSC) of size from 0.1 µm to 1 µm and (ii) Large secondary carbides (LSC) of size from 1µm to 5 µm. All studies point to benefits of DCT in multiple ways. A great reduction in the amount of retained austenite is seen – in most cases retained austenite content is reduced to as low as 4%. The microstructural changes increase the wear resistance of the material.

The martensitic phases in the steels are further conditioned during DCT leading to precipitations of high amount of ultrafine secondary carbides. Secondary carbides are generally hard and thus contribute to improved wear resistance. The micro structural changes that occurred during DCT as shown in figure 4, 5 and can be compared to the figure 6 which is of cryoheat-treated (CHT) for D2 tool steel. From these illustrations it can be seen that DCT enables precipitation of fine and ultrafine carbides in the material. The reduction in the amount of Retained Austenite is confirmed from XRD profile obtained for such DCT material specimen. A very scant amount of retained austenite has been detected by TEM studies from the DCT specimen. The secondary Carbides are made up of M₂₃C₆ where as eutectic M₇O constitutes primary carbides along with small amount of C₇Cr₃ carbides.¹⁶

Figure 4. Optical Micrograph showing various carbides ¹³Figure 5. SEM Micrograph of DCT on AISI D2 Steel ¹³Figure 6. SEM Micrograph of CHT on AISI D2 Steel ¹³

The reciprocator friction and wear monitor test on SCT and DCT samples of En 52 and 21-4N valve steels. The results have shown that the wear resistance has increased in DCT sample compared to SCT samples. The microstructure of the En-52 and 21-4N (CHT, SCT and DCT) variants is shown in figure 7 and 8. A very large white patches can be seen in CHT sample in which martensite and retained austenite both exist together where as in cryotreated specimens (DCT) the austenite is fully transformed into martensite form, in SCT the austenite is retained in small fraction. Since the martensitic phase is the last phase that enhances the hardness uniformly throughout the material which increases the wear resistance of the material. The clusters of primary and secondary carbides are also seen in SCT and DCT samples. ¹⁷

Figure 7. The microstructure of En 52 CHT, SCT and DCT ¹⁴

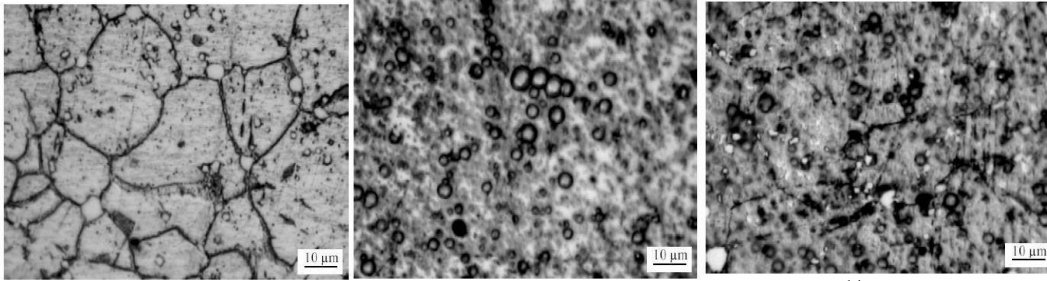


Figure 8. The microstructure of 21-4N CHT, SCT and DCT ¹⁴

The optical microstructure studies ¹⁸ on CHT and DCT samples for D2 tool steel also confirmed the same as in other studies. The DCT sample again exhibit higher carbide precipitation and a finer distribution of secondary carbide than none at all in CHT samples which shows irregular shaped primary carbides. Comparative microstructural images are given figure 9.

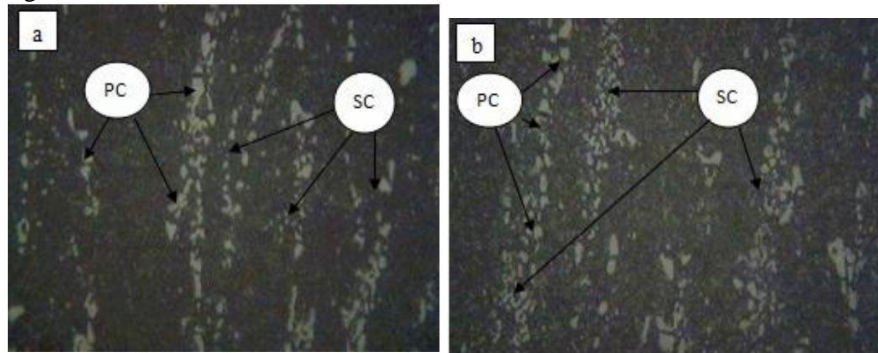


Figure 9. The microstructure of D2 CHT, DCT ¹⁵

Various sequences of hardening and tempering process along with DCT were carried out on D3 tool steel by SimranPreet ¹⁹ the results of experimentation were studied through microstructural studies. The various sequences of heat treatment process are tabulated below. The figure 10(a) shows the microstructure of the as the received state of D3 large accumulation of carbides of all irregular shapes can be seen carbides get dissolved in the medium after hardening process and the materials takes different changes based on the subsequent process carried. It can be seen through comparison between the figure 10(b) and 10(c) for B-I and B-II sequence that the size of carbides have been reduced. When cryotreatment was followed by tempering and double tempering the carbides can be seen to getting more cores through a phenomenon called “Ostwald ripening” (smaller particles getting deposited onto larger carbides).

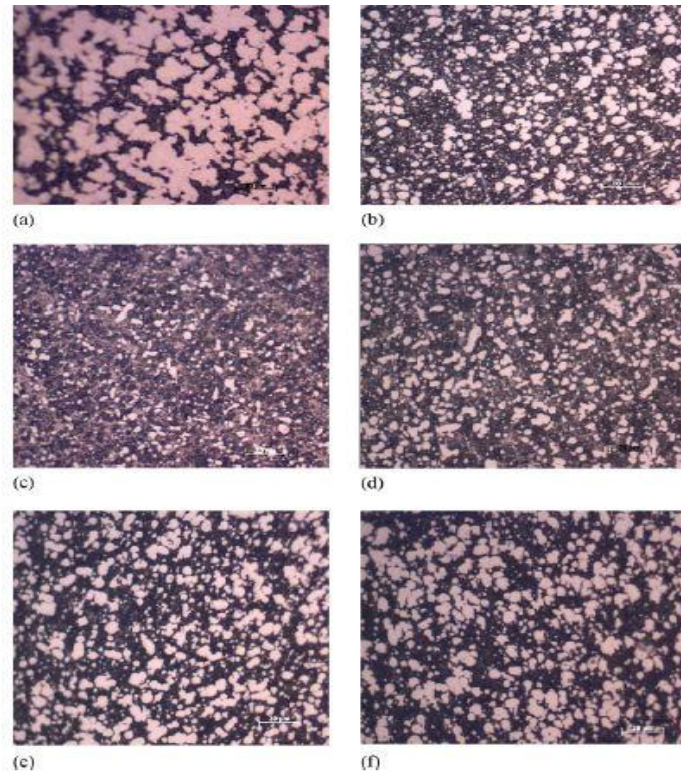


Figure 10. Microstructure of D-3 steel for treatments B-I, B-II, B-III, B-IV and B-V: (a) raw (500×); (b) HT (500×); (c) HC (500×); (d) HCT (500×); (e) HCTT (500×); (f) HCTTT (500×).¹⁶

The changes in D2 and D3 tool steel after cryogenic treatment, Globular shaped carbides of 9 micron and nodular shaped carbides of 11X4 micron can be seen in the microstructure of CHT of D2 in figure 11(a) and untempered martensite and retained austenite was seen of about 10% and 50% respectively there has been an decrease in carbides size both in globular and nodular shape as compared to just austenizing, quenching and tempering process. The distribution of carbides is more uniform figure 11(b). the carbide sizes in both forms was found to increase in AQCT process after tempering however multiple tempering process after cryogenic treatment increases the retained austenite there by neglecting the overall benefits of cryogenic treatment therefore multiple tempering treatment is not advisable after DCT.²⁰

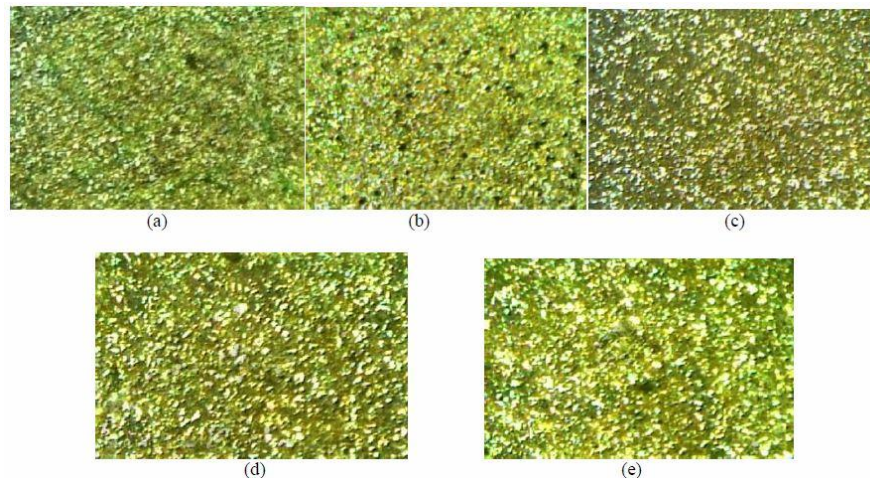


Figure 11. Microstructure of various combinations of treatments for the specimen of D2 steel a) CHT b) AQC c) AQCT d) AQCTT e) AQTCT¹⁷

Figure 12(a) which shows the microstructure of D3 tool steel of CHT, there has been up to 20% of the globular shaped carbide content of size 3x6 microns. The well distributed structure was observed with around 7% of untempered martensite the carbide content is up to 4% in AQC sequence figure 12(b) which shows the positive effect

of tempering after cryogenic temperature. The medium contains up to 95% of unstable austenite figure 12(c) the carbide distribution is more uniform. Nodular carbides of 2X4 microns are also seen amounting to 15%, on further tempering multiple times the retained austenite content goes on reducing and the untempered martensite is increasing in content from 5% to 7% in further sequences in AQCTT and AQTCT sequences.²⁰

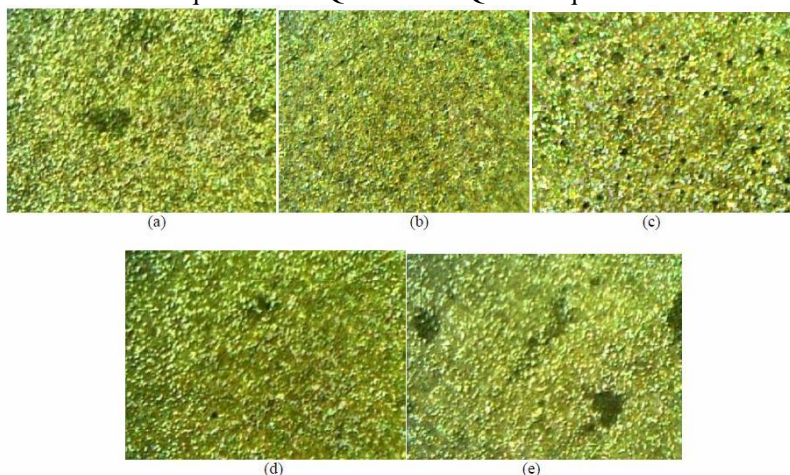


Figure 12. Microstructure of various combinations of treatments for the specimen of D3 steel a) CHT b) AQC c) AQCT d) AQCTT e) AQTCT¹⁷

Huang²¹ studied the effects of cryogenic treatment and subsequent tempering had on the precipitation of carbide in M2 tool steels using the TEM micrographs of untreated and cryotreated M-2 tool steel as shown in figure 13 and has concluded that the distribution of carbide as well as their sizes remained same in treated and untreated cryogenic samples, but the density of the carbide content was different during the treatment as the retained austenite is converted into martensite, internal stresses are developed in the samples this is caused due to the chain reaction the stress are mainly caused due to the changes in the composition and microstructure which causes dislocation and twinning in the crystal structure. High internal stresses are developed when the samples are cooled below certain temperatures, when the holding time is long enough the carbon and other alloying elements diffuses to the defected sites thus creating a cluster of carbon and alloying elements. As the martensite becomes supersaturated due to decreasing temperatures during cryogenic treatment the resulting thermal instability and lattice distortion drive the alloying atoms and carbon atoms clustering near defects. It is around these clusters where carbides are formed when the tempering process is followed after cryogenic treatments.

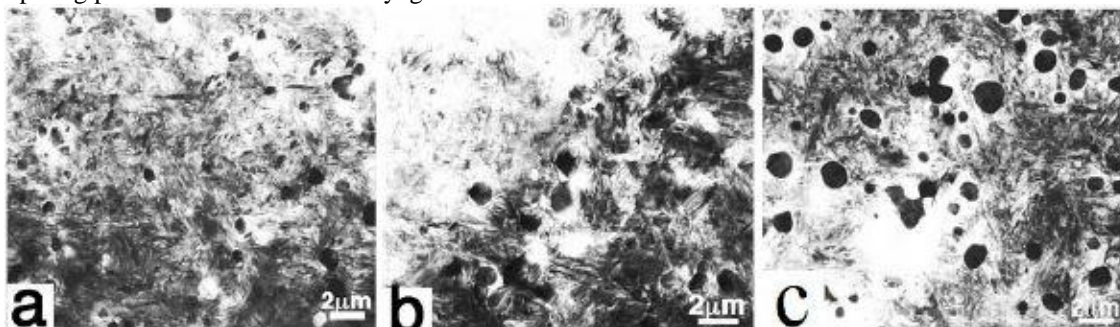


Figure 13. TEM micrographs of M-2 steel (a) and (b) Representative regions from the same untreated sample (c) Cryotreated sample¹⁸

In the series of experiment^{22,23,24} significant precipitation of secondary carbides was observed. As in most studies of cryogenic treatments on tool steels mechanical properties have improved figure 14 shows the SEM micrographs of various cryotreatment, In cryogenic treatment holding time has a significant effect on wear resistance properties. It is found that the wear resistance increases as the holding time increases up to 36 hours and decreases beyond that. Thus a function between the density, distribution of carbides and holding time can be established which agrees in the case of M2 tool steel strongly.

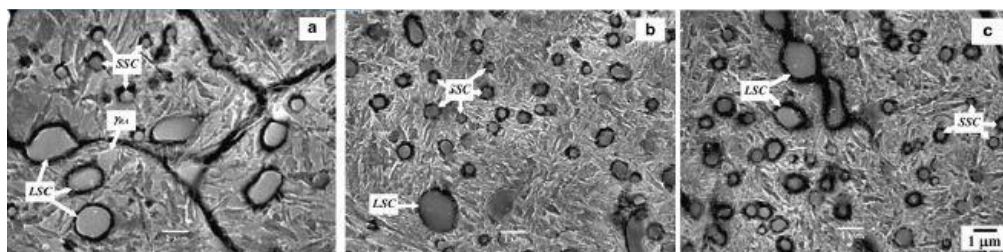


Figure 14. High-magnification SEM micrographs: (a) conventionally quenched and tempered; (b) cryogenically treated for 36 h; (c) Cryogenically treated for 84-h²¹

The authors also concludes that the wear resistance on the material will have a positive effect only till the holding time is 36 hours only, and beyond that the material will show the reverse effect i.e. decrease in the properties.

Wear behavior:-

The wear of tool during a wide range of industrial applications is a serious issue in the production prospective. Cryogenic treatment is a best solution to reduce the tool wear which is also effectively proved by the authors. There was an increase in wear resistance by 18.54% and 14.04% for a load of 50N and 25N respectively.²⁵ In some cases an improvement of wear resistance in D3 tool steel by 80%. The co-efficient of friction was also found to decrease which point to the quality surface finish was obtained in cryotreatment.²⁶

Conclusion:-

- The cryogenic treatment has been used effectively to enhance the mechanical properties like wear resistance, hardness of various grades of steel.
- The core benefits of cryogenic treatment are increase in wear resistance and hardness.
- Soaking period and temperature also plays an important role in varying the properties of the tool.
- These benefits are effectively applied to various tool steels to maximize the tool life.
- The underlying mechanism had been conversion of austenite to martensite structure. The tool life also gets enhanced through increased wear resistance caused by the deeper distribution of carbide particles in the material.
- Maximum benefits from can be achieved in cryogenic treatment by controlling the parameters such as (holding temperature, holding time, identification of heat treatment to be applied before or after, etc.) which should be applied under optimum conditions.

Declaration of conflicting interests:-

- There are no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

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RESEARCH ARTICLE

A COMPERATIVE STUDY ON THE PERFORMANCE OF THE STUDENTS IN MATHEMATICS AND SOCIAL SCIENCE.

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Manuscript Info

Abstract

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Introduction:-

Being a Mathematics Teacher in a Senior Secondary School of our locality, I personally observe the fact that students of Secondary Sections pay less attention as well as they are less interested in the Subject General Mathematics in comparison to other Subjects. They have a fear in mind about Mathematics and so always hesitate to apply their intellect in solving sums. As the subject itself can't produce any interest among the students unlike to literature, history, Social Science etc from which the students get something interesting going through the lessons and hence they pay less attention to that subject.

To make Mathematics interesting, I myself have tried to help them learn with the help of different models which I have made myself of angles, triangles, quadrilaterals, geo-board, Models of Venn diagrams with simple things like ball pens, riffles, straw pipes, Card-boards bangles and plywood etc. The result I got is very interesting. The students get interested in Practical Classes but still they are lagged behind in theoretical knowledge. Taking this matter as a subject of my project, I have collected the marks obtained in Annual Examination of last two years of the students of Class – IX and Class – X in mathematics and Social Science and a thorough Statistical Study has been done.

Methodology:

A statistical survey has been performed among the students of Class – IX and X in a High School in our locality and their marks in General mathematics and Social Science in Annual Examination for 2015 and 2016 respectively and the marks obtained by them have been tabulated in grouped data and the result has been thoroughly studied to find the fact if they are lagged behind in Mathematics in Comparison to Social Science. The marks obtained by the students in General Mathematics and Social Science is tabulated in grouped data and the results are shown graphically.

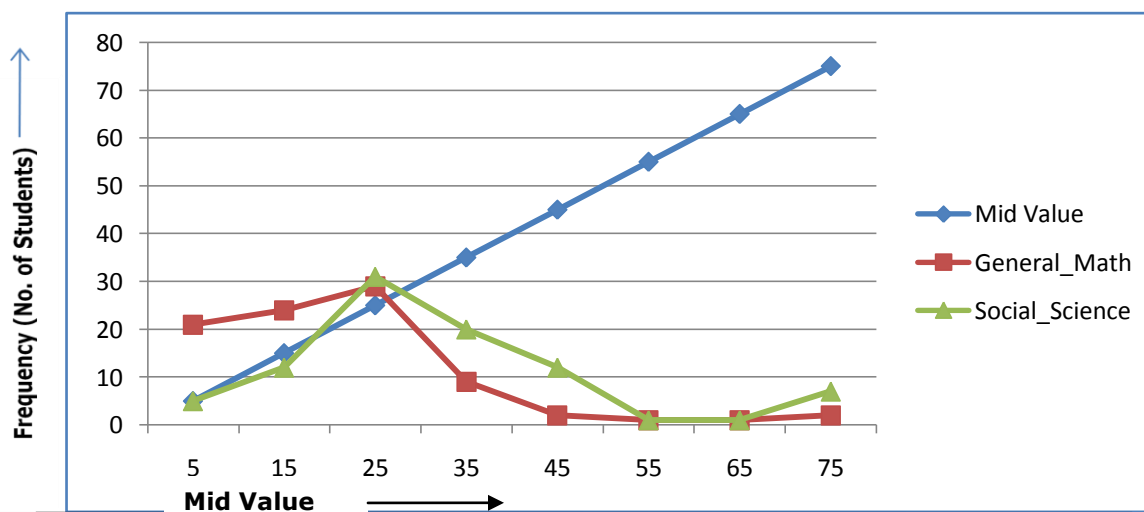
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Results Of Class Ix In Mathematics And Social Science, 2015.

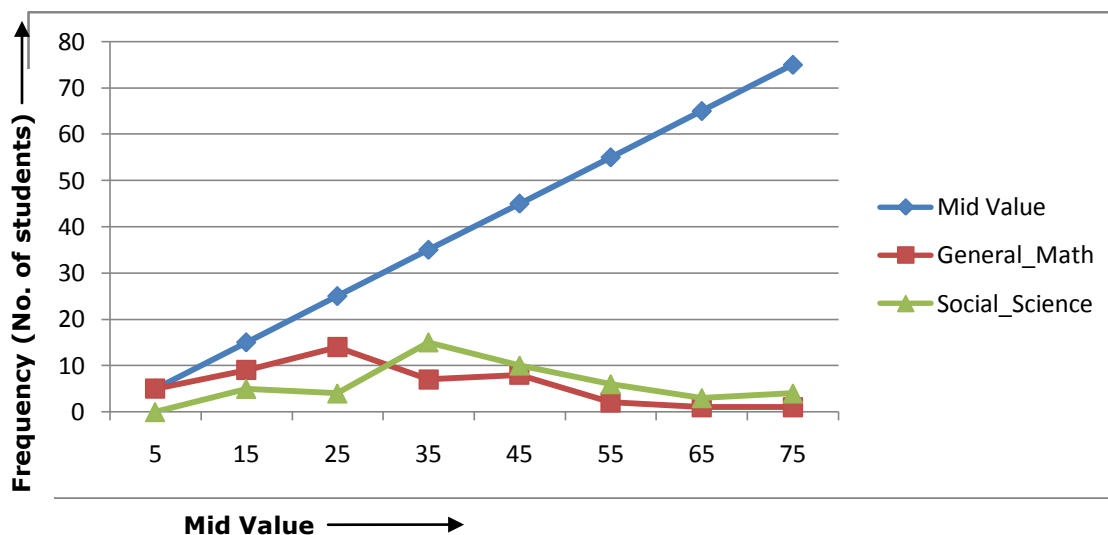
Only marks in theory (Full marks = 80) is considered here. 20 marks in practical is not considered in tabulation.

Class Interval (Marks obtained)	Frequency (Number of students)	
	General Mathematics	Social Science
0-10	21	05
10-20	24	12
20-30	29	31
30-40	09	20
40-50	02	12
50-60	01	01
60-70	01	01
70-80	02	07
Total:	89	89

**Results of Class X in Mathematics and Social Science, 2015.**

Only marks in theory (Full marks = 80) is considered here. 20 marks in practical is not considered in tabulation.

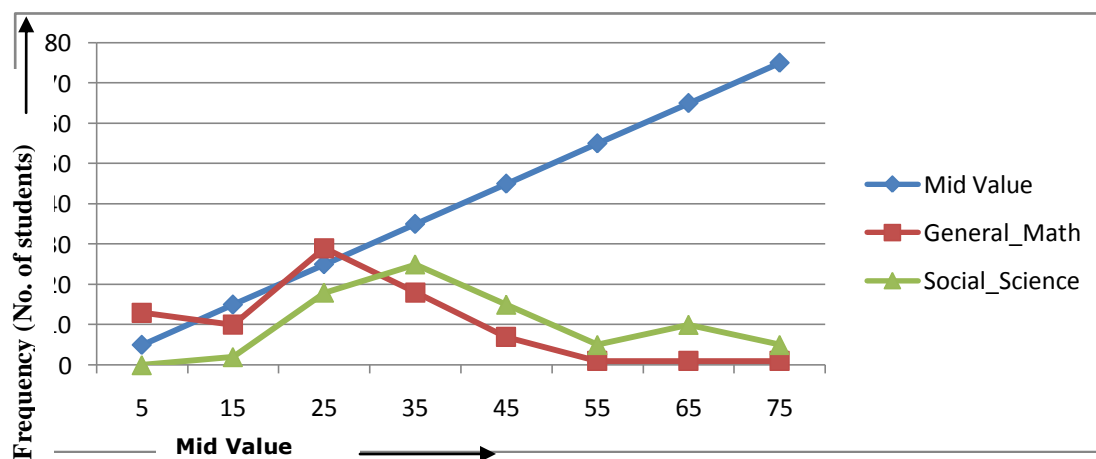
Class Interval (Marks obtained)	Frequency (Number of students)	
	General Mathematics	Social Science
0-10	5	00
10-20	9	05
20-30	14	04
30-40	07	15
40-50	08	10
50-60	02	06
60-70	01	03
70-80	01	04
Total:	47	47



Results of Class IX in Mathematics and Social Science, 2016:-

Only marks in theory (Full marks = 80) is considered here. 20 marks in practical is not considered in tabulation.

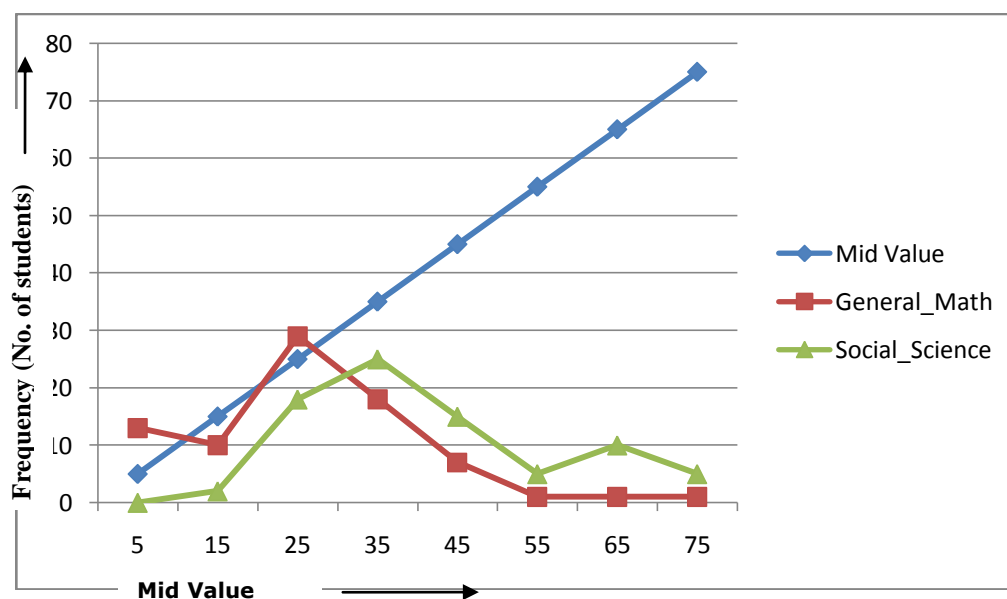
Class Interval (Marks obtained)	Frequency (Number of students)	
	General Mathematics	Social Science
0-10	13	00
10-20	10	02
20-30	29	18
30-40	18	25
40-50	07	15
50-60	01	05
60-70	01	10
70-80	01	05
Total:	80	80



Results of Class X in Mathematics and Social Science, 2016:-

Only marks in theory (Full marks = 80) is considered here. 20 marks in practical is not considered in tabulation.

Class Interval (Marks obtained)	Frequency (Number of students)	
	General Mathematics	Social Science
0-10	13	00
10-20	10	01
20-30	30	18
30-40	19	25
40-50	07	14
50-60	01	05
60-70	01	10
70-80	01	05
Total:	82	78

**Observations:-**

The above statistical data shows that less Number of Students obtain 60% or above marks in General Mathematics in comparison to Social Science. The number of Students getting 60% or above in Mathematics in the Annual Examination in 2015 is 03 in Class IX comparison to Social Science where the number is 8. In Class X also the number of Students getting 60% and above marks in Mathematics is only 02 (Two) and in Social Science the number of such students is 07 (seven). In 2016 also the number of students getting 60% and above marks in Mathematics in Class IX is only 02 (Two) where as in Social Science the number is 15 (fifteen). The result of the observations is as follows –

Percentage Of Marks	In 2015				In 2016			
	No Of Students Of Class IX		No Of Students Of Class X		No Of Students Of Class IX		No Of Students Of Class X	
Subjects	G. Math	Social Science	G. Math	Social Science	G. Math	Social Science	G. Math	Social Science
Below 30%	48	17	26	06	47	12	19	02
50% And Above	15	41	12	23	10	35	11	30
60% And Above	05	20	05	15	06	24	07	27
80% And Above	02	08	02	07	02	15	02	05

Causes for weakness in Mathematics:-

According to my observation, some of the causes for weakness in Mathematics in comparison to other Subjects are –

1. Theory part of Mathematics is itself not interesting and some of the topics can also be not represented by different models or diagrams.
2. In lower primary as well as upper primary classes the basic Concepts of different mathematical terms have not been explained in easy language and with sufficient diagrams in text books.
3. There is Lack of sufficient elaborate explanations of different mathematical terms and definitions in Text books.
4. Insufficient Equipments or instruments for Practical Classes in Mathematics in the Senior Secondary Schools

Remedial Suggestions:-

From my own experience, I had innovated myself some methods to grow interest in Mathematics –

1. In lower and upper primary classes the basic concepts can be explained with the help of different games among the students.
2. Geometric concepts can be explained with the help of different models and Geo-boards.
3. Difficult problems of Set theory can be made easy to understand with the help of models of Venn diagrams of different set operations
4. To make the subject interesting audio – video classes, different workshops and seminars should be frequently arranged in the schools.

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RESEARCH ARTICLE

A PROSPECTIVE STUDY OF FACTORS RELATED TO PLATELET YIELD AMONG DONORS UNDERGOING PLATELETPHERESIS.

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Abstract

Background: The transfusion of blood products is life saving especially at emergency conditions. The platelet transfusions helps in prevention of bleeding related complications and thus prevents morbidity and mortality in thrombocytopenic patients. Platelets are transfused by two methods i) by fractionation of whole blood and ii) by platelet apheresis. The quality of single donor platelets (SDP) in terms of yield influences platelet recovery in the recipient.

Material and Methods: A sample of 360 donors were included in the study over a period of one and half year. Various donor-related factors were meticulously recorded prior to performing plateletpheresis. The aim was to identify donor factors that influence platelet yield. The plateletpheresis procedures were performed using Trima accel machine. A relationship between pre-donation donor variables and yield of platelets was studied using the Pearson correlation.

Results: The mean platelet yield was $2.29 \pm 0.43 \times 10^5/\mu\text{L}$ per donor. A positive correlation was observed between platelet yield and pre-donation platelet count, body mass index (BMI; Kg/m²) of the donor, while a negative correlation was observed between age and the platelet yield.

Conclusion: Donor pre-donation platelet count, BMI and donor age influence platelet yield. Young healthy donors with a high platelet count and better BMI can give a better platelet yield in the SDP.

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Introduction:-

Platelets are essential for the formation of primary haemostatic plug and maintenance of haemostasis. Approximately 2.2 million platelet doses are transfused annually in the United States (1). A high proportion of these platelet units are transfused prophylactically to reduce the risk for spontaneous bleeding in patients who are thrombocytopenic after chemotherapy or hematopoietic progenitor cell transplantation (HPCT) (2,3). Either a pool of 4 to 6 units of random donor platelet or 1 unit of single donor platelet (SDP) is transfused (4). One unit of SDP should contain a minimum of 3×10^{11} platelets as per American Association of Blood Bank (AABB) guidelines (11) while European guidelines advocate that one SDP should contain 2×10^{11} platelets (5,6). Unlike other blood components, platelets must be stored at room temperature, limiting the shelf life of platelet units to only 5 days because of the risk for bacterial growth during storage. Therefore, maintaining hospital platelet inventories is logistically difficult and highly resource-intensive (7,8). Platelet transfusions can be done either as i) a pool of 4 to 6 units of random donor platelet, or ii) 1 unit of single donor platelet (SDP) is transfused (4).

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Plateletpheresis (apheresis= to remove) is a procedure designed to collect large number of platelets from single donor thereby giving more consistent product (9). As per guidelines of American association of blood banks plateletpheresis (Single Donor Platelets) unit must have platelet count of 3×10^{11} which in turn raises platelet count by 30,000-60,000 per micro litre and is equivalent to 4-6 units of random platelet concentrates (10). Compared to whole blood donation, apheresis has advantages for the donor, such as there is lesser loss of red blood cells. However, apheresis can also lead to specific adverse events such as citrate toxicity or, for single needle devices, extracorporeal circulation reactions (11-13). Platelet transfusion success depends on rational use of platelet components and on the quality of the component. Platelet recovery in a patient is influenced by the transfused dose of platelets which in turn is dependent on the platelet yield.(14) The possibility of obtaining higher platelet yields has important clinical implications: it reduces frequency of platelet transfusions and number of donor exposures with important consequent clinical and economic advantages.(15,16)

Material and Methods:-

This is a prospective case study conducted from August 2015 to December 2016 at tertiary care centre in North India. A total of 360 healthy donors comprising of 359 men and 1 woman with mean age of 41.8 years were selected from plateletpheresis donor registration. With the exception of the donation interval, the requirements for apheresis and whole blood donation are the same. Apheresis donors must meet the requirements for whole blood donation and additionally satisfy criteria that are particular to the selected apheresis. Donors that were less than 18 years and more than 60 years, or were on any medical treatment 7 days prior to the procedure were excluded in this study. Blood samples were collected from donors before and after apheresis procedure. Informed consent was obtained from all donors. Donor screening is required to ensure a safe transfusion for the recipient. Prospective donors must complete several steps before actual platelet donations, including physical examination, a donor history questionnaire, and testing for transmissible diseases. The American Association of Blood Banks (AABB) recommends that prospective donors receive physical examinations including an assessment of weight: hemoglobin, hematocrit, ABO and Rh typing; and inspection for marks from intravenous drug use. Tests are performed for the presence of syphilis, human immunodeficiency virus (anti-HIV-1/2 and HIV-1 RNA), hepatitis C virus (anti-HCV and HCV RNA), hepatitis B virus (HBsAg and anti-HBc). Further, the platelet counts of plateletpheresis donors must be $>150 \times 10^3$ per μl [32]. Individuals can donate 3 days after ingesting aspirin-related medications.

Apheresis was performed using Trima (Gambro BCT, Lakewood, CO). Acid-citrate-dextrose formula was used as anticoagulant during procedures according to the manufacture's recommendations.

All procedures were completed successfully with peripheral venous access . Cubital vein was used for access. All procedures done were single needle. The procedure was continuous and automated. The blood pump speed was set at of 60-80 ml/min . Blood pressure (BP) and pulse were monitored at frequent intervals during the sessions and donors were closely observed for development of any complications and overall status.

Results:-

The results obtained are tabulated below :

Table 1:- The distribution of hemoglobin in the donor of study sample (n=360).

Hb (g/dl)	No. of donors	%age
12-13	27	7.5
13-14	51	14.1
14-15	111	30.8
15-16	105	29.1
16-17	57	15.8
>17	9	2.5

The maximum number of patients had hemoglobin in the range of 14-15 g/dl and 15-16 g/dl, depicting the donors overall had been derived from healthy population. Only where donor constraint was an issue , selection was done in the Hb range 12-13 g/dl and > 17 g/dl.

Table 2:- The distribution of Hematocrit in the donor of study sample (n=360)

Hct (%)	No. of donors	%age
<40	48	13.3
40-45	198	55
45-50	99	27.5
>50	15	4.1

The haematocrit of the donor population were between 40-45% in maximum number of donors.

Table 3:- The distribution of pre-platelet count in the donor of study sample (n=360)

PRE-PLATELET COUNT(/cumm)	No. of Donors	%age
<150	30	8.3
150-200	141	39.1
200-250	108	30
250-300	57	15.8
300-350	24	6.6

The platelet count in our donor population fall in lower range of normal platelet count i.e., 150-200/cumm and 200-250/cumm .

Table 4:- The distribution of platelet yield in the donor of study sample (n=360).

PLATLET YIELD ($\times 10^{11}$)	No. of donors	%age
<2	36	10
2-3	120	33.3
3-4	204	56.6

The platelet yield was between $2-3 \times 10^{11}$ in maximum number of donors.

Table 5:- The distribution of platelet yield in the donor of study sample (n=360)

Adverse effects	Number	%
Citrate reaction	12	3.3
Hematoma	6	1.6
Vasovagal reaction	2	.55
Hypovolemia	1	.27
Mech. Issue	3	.83

The side effects were less observed in the procedure done. The most common reaction had been citrate reaction followed by hematoma formation at vascular access site.

Table 6:- Correlation between various donor factors with Platelets yield.

Platelet Yield	Age	BMI	Hb	Hct	Pre-platelet count
	- 0.329 (<0.01)	+ 0.268 (<0.01)	0.063 (<0.01)	0.021 (<0.541)	0.284 (<0.01)

Discussion:-

Various studies have shown that transfusion of high platelet doses could reduce number of platelet concentrates required by thrombocytopenic patients even in patients with adverse clinical factors in which refractoriness to transfusion is common(17,18). Nevertheless, there are very few studies related to donor clinical and laboratory factors that may influence number of platelet yield (4,19). Identification of these factors would allow for better selection of donors resulting in higher platelet yield and consequently a lower number of donor exposures to the patients. Instruments which collect SDP are programmed to calculate the yield from the donor's haematocrit, platelet count, height and weight.

In our study, the mean pre-donation platelet count was $2.29 \pm 0.43 \times 10^5/\mu\text{L}$ and the mean platelet yield was $2.86 \pm 0.52 \times 10^{11}$. A direct linear correlation was obtained between the pre-donation platelet count and the platelet yield ($r = 0.284$, $p < 0.01$). Ravi et al (20), found the mean pre-donation platelet count was $2.69 \pm 0.65 \times 10^5/\mu\text{L}$ and the mean platelet yield was $3.16 \pm 0.62 \times 10^{11}$. A good direct linear correlation was obtained between the pre-donation platelet count and the platelet yield ($r = 0.284$, $p < 0.01$). Chaudary R et al, reported that out of 94 plateletpheresis procedures, the mean platelet yield was $3.65 \pm 0.80 \times 10^{11}$ when the pre-donation platelet count was $< 3 \times 10^5/\mu\text{L}$, while the mean yield was $2.5 \pm 0.59 \times 10^{11}$ when the pre-donation platelet count was $> 2 \times 10^5/\mu\text{L}$. They also observed a direct relationship between platelet count and platelet yield ($r = 0.50$, $p < 0.001$) (21). In a study, 2708 plateletpheresis procedures having a mean pre - donation platelet count of $2.37 \pm 49 \times 10^5/\mu\text{L}$ resulted in a platelet product with mean yield of $4.24 \pm 1.1 \times 10^{11}$. A direct linear correlation was observed with all the procedures (4). In a study (22) donor pre-donation platelet count was found to positively correlate with the platelet yield ($r = 0.51$, $p < 0.001$). A direct positive correlation was also observed in another study (17) ($r = 0.512$) (23). Our observations were similar.

In our study, 204 donors gave a platelet yield of more than 3×10^{11} per unit and thus 56.6% of our platelet yield met AABB guidelines. As per European guidelines 324 of our donors i.e., 93% had a platelet yield of $> 2 \times 10^{11}$ (20). Chaudhary et al., found only 41.5% of the SDPs met AABB guidelines (19).

Our study showed negative correlation between the donor age and platelet yield ($r = -0.329$, $p < 0.01$). Das SS et al., has showed a significant negative correlation between the donor age and platelet yield ($r = -0.229$, $p < 0.01$) but no such observation was reported in another study (21). The decreasing platelet yield with increasing age found in our study may be explained the physiological decrease that occurs to platelet count with increasing age (22).

We found a positive correlation between platelet yield and either haemoglobin ($r = 0.063$) or haematocrit ($r = 0.021$). While as positive correlation between platelet yield and donor haemoglobin and haematocrit have been found in some studies (25,17). No correlation was observed between the haemoglobin concentration and platelet yield in another study. They reported that donors with a haemoglobin levels of 16 g/dL or more gave a comparatively lower platelet yield. This could be related to the higher plasma volume processed in donors with low haemoglobin concentration thereby giving a higher platelet yield (19).

Furthermore, a positive correlation between BMI and platelet yield ($r = 0.268$, $p < 0.01$).

Similar observations were reported in another study (18) where BMI correlated consistently with a good platelet yield. But, in another study (19) where the quality of SDP in relation to low weight (40.8- 49.9 Kg) of the donors was studied, donor weight was not found to have any effect on platelet yield. Since there was only one female donor, we are not able to calculate the correlation between the platelet yield and gender. In a study (21) it was found that gender also influences platelet yield and women had higher yields. This was possibly because there is a higher prevalence of iron deficiency among women with consequent increase in platelet count; hormonal influence could also play a role.

Conflict of Interest : None .

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RESEARCH ARTICLE

PHYTOCHEMICAL SCREENING, ACUTE TOXICITY AND CARDIOVASCULAR EFFECTS OF WATER EXTRACT OF *HALLEA LEDERMANNII* (K. KRAUSE) VERDC (RUBIACEAE), MEDICINAL PLANT FROM GABON.

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Phytochemical screening, acute toxicity, *Hallea ledermannii*, secondary metabolites, cardiovascular effects.

Abstract

Hallea ledermannii is a medicinal plant commonly used in traditional Gabonese medicine to fight against high blood pressure. The objective of this study was to determine the chemical profile and evaluate the acute toxicity and effect of the aqueous extract of *Hallea ledermannii* on cardiovascular parameters in normotensive rats. Phytochemical screening was carried out by using the usual methods of Bouquet. Acute oral toxicity was evaluated in mice at doses of 50, 300, 2000 and 5000 mg / kg in accordance with OECD Guideline 423. Tests on blood pressure and heart rate were realized by the invasive method in normotensive rats anesthetized at urethane 15%. The results show that *Hallea ledermannii* contains many secondary metabolites. Its polar extracts have of the low contents in total phenol (3.55675 ± 0.083 , 3.27425 ± 0.101 , 2.78675 ± 0.110 mg GAE/g of drugs) and total flavonoids (1.2272 ± 0.197 ; 0.7232 ± 0.187 ; 0.8072 ± 0.129 mg QE/g of drugs). Aqueous extract of *Hallea ledermannii* is not toxic up to the 5000 mg/kg dose. This extract (10, 20 and 30 mg/kg) causes a low in systolic blood pressure and heart rate. These results suggest that the aqueous extract of *Hallea ledermannii* can be used orally up to the 5000 mg / kg dose without risk of toxicity and has chronotropic negative and hypotensive effects that could explain its use in traditional Gabonese medicine against high blood pressure.

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Introduction:-

"Soft" medicine know considerable success in many parts of Africa. Surveys reveal that 80 to 85% of developing countries use medicinal plants for public health care (Organisation Mondiale de la Santé, 2002). The recourse of medicinal plants know up to this day a craze that could be explained by several reasons: The return to supposed natural products without adverse effects unlike conventional medicines; difficulties in accessing pharmaceuticals and their increasing costs; the importance given to medicinal plants by the pharmaceutical industry. Despite the place of antihypertensive drug products on the market, high blood pressure constitute up to this day public health problem whole world and in particular in developing countries. Today, high blood pressure has become a major

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cardiovascular risk factor representing a public health problem for African populations. In Gabon, the prevalence is about 25 to 30% (Kearney et al., 2004). The Gabonese flora it is full still several species of plants little or not studied, but endowed with true pharmacological properties, it is the case of *Hallea ledermannii* still called *Mitragyna ciliata*. This medicinal plant belongs to the family Rubiaceae. It is very present in marshy areas. It is rich in alkaloids (Burkill, 1997). *Hallea ledermannii* is a large tree can reach 20 to 35 meters tall with a diameter rarely reaching 0.8 to 1 meter, cylindrical, and without thickening at the base. Gabonese population uses this plant in the treatment of female infertility, for douching as a local antiseptic and bark decoction in association with other plant species is administered as an enema in painful menstruation (Raponda-Walker et Sillans, 1961; Gassita et al., 1982; Wagner, 1986).

Material and Methods:-

Plant material:-

The trunk bark of *Hallea ledermannii* were harvested in Franceville, in the Province of Haut-Ogooué (South-Eastern of Gabon) in March 2014. Identification of the species was carried out at the Institute of Research in Tropical Ecology and deposited at the National Herbarium of National Center of Scientific and Technical Research (CENAREST) of Libreville (Gabon). The bark was dried at Laboratory of Natural Substances and Organometallic Synthesis (LASNSOM) at Department of Chemistry of the Faculty of Sciences of USTM (Franceville) protected from light and at ambient temperature (27-29 °C) during 5 weeks. The dry bark was crushed and preserved in bottles out of glass safe from the light and moisture for later analyses.

Animal material:-

The present study was carried out on male swiss albino mice aged 8 to 10 weeks and of weight including between 25 and 30 g and wistar albino rats from 12 to 14 weeks and of weight including between 250 and 350 g.

Alimentation and habitat of animals:-

The animals were supplied by the pet shop of the Faculty of Health Sciences of the Marien Ngouabi University where they were raised under natural aeration and lighting conditions. They were fed by a standard alimentation in the form of croquettes and were drinking *ad libitum* with drinking water.

Preparation of plant extract:-

The total extracts were prepared from the dry powder of the plant material. 100 g of powder were set to macerate in 1 L of solvent, respectively ethanol, water ethanol (500/500, v/v) and water, with magnetic stirring for 48 hours. The macerate was then filtered and then the solvent was evaporated to dryness under reduced pressure at 75 °C at help BÜCHI R-210 rotary evaporator. The extracts were concentrated and then kept in glass bottle for analysis.

Preparation of the aqueous extract:-

The aqueous extract was prepared by decoction from the dry powder of the plant material. 100 g of plant material powder were immersed in 1000 mL of distilled water. The mixture was heated with a hot plate with magnetic stirring for 30 minutes. After cooling, the extract was filtered and then the solvent was evaporated to dryness under reduced pressure at 75 °C at help BÜCHI R-210 rotary evaporator.

Phytochemical screening:-

The chemical characterization of the secondary metabolites contained in the plant extract was carried out according to the conventional methods described by Bouquet (1972). Thus, we carried out the tests of alkaloids (reactive of Dragendorff and Mayer), anthocyanins (HCl 20 %), anthraquinones (KOH 10 %), flavonoids (reaction to the cyanidin), triterpenoids (reaction of Liebermann-Bouchard), saponosids (foam index), gallic tannins and catechic tannins (ferric chloride, Stian), reducing compounds (reagent of Fehling) and carotenoids.

Phenolic content:-

The total phenolic contents of the different extracts were determined by the method of Folin-Ciocalteu (Singleton et Rossi, 1965). A quantity of 200 µL of the extract is mixed with 1 mL reagent of Folin-Ciocalteu coldly prepared (10 times diluted) and 0.8 mL sodium carbonate solution (7.5 %). The unit is incubated at ambient temperature during 30 min and the reading is carried out against a white using a spectrophotometer with 765 nm. All analyses were done in triplicate and results (average of triplicate analysis) are expressed in milligrams equivalent of gallic acid per gram of dry vegetable matter.

Flavonoid content:-

The content in flavonoids of the extracts was given by using the colorimetric method with aluminum chloride (Kim et al., 2003). A quantity of 100 μ L of the extract was mixed with distilled water 0.4 mL and thereafter with 0.03 mL sodium nitrite solution (5 %). After 5 min, 0.02 mL aluminum chloride solution (10 %) was added. One adds with the mixture 0.2 mL sodium carbonate solution (1 M) and 0.25 mL distilled water after 5 min with rest. The unit is agitated using a vortex and the absorbance was measured to 510 nm was recorded after 30 min of incubation. A standard calibration plot was generated using known concentration of quercetin. The results are expressed in milligrams equivalent of quercetin per gram of dry vegetable matter.

Acute toxicity:-

After three days of acclimatization in the laboratory, the mice were fasted for 4 hours before administration of the products without removing them from the water. Acute oral toxicity was assessed in accordance to the OECD Guideline n° 423 (2001). Five (5) batches of three (3) mice each were constituted and treated orally in the following manner: Control batches were treated at distilled water (0.5 mL/100 g) and the other batches treated at aqueous extract of *Hallea ledermannii* at the respective doses of 50, 300, 2000 and 5000 mg/kg. After administration of the products, each mouse was placed in an individual cage for ½, 1, 2, 3 and 4 h observations on spontaneous motricity, aggressiveness, responses to external stimuli, Stools, ptosis, piloerection, convulsions. Batch mortality was assessed within 48 hours of product administration. The body weight of each animal as well as food consumption were recorded every other day for 14 days (OECD, 2001).

Evaluation of cardiovascular effects:-**Anesthesia of the animal and catheterization of the femoral vein and the carotid artery:-**

The general anesthesia of the rats was carried out by intra-peritoneal injection of urethane 15 % at 1.5g/kg at the rate of 1 mL/100 g of rat body weight (Dimo et al., 2003). The anesthetized animal was fixed in dorsal decubitus, by means of pins planted on these legs on a cork board. The femoral vein and then the carotid artery were exposed and then catheterized, respectively, using fine polyethylene catheters attached to a syringe and introcan 24 G catheters as described by Etou Ossibi and al (2010). The 10 % heparinized NaCl (0.9 %) solution was injected at a rate of 0.1 mL/100 g by the femoral vein to prevent blood coagulation. When the clamp is removed, the blood engulfs into the catheter Introcan 24 G and transducer of type Biopac Student Lab MP 36 transmits the variations of the blood pressure at recorder of the same type that converts the waves into traces (records Blood pressure and heart rate) that can be viewed on the computer screen and whose values are read directly (Nguelefact, 2008).

Administration of the aqueous extract of Hallea ledermannii:-

After a period (about 30 min) of blood pressure stabilization, a physiologic solution of NaCl (0.9 %) and the aqueous *Hallea ledermannii* extract (10, 20 and 30 mg/kg) were administered to the normotensive rats (n = 5) at a rate of 0.1 mL/100 g. After administration of the products, their effects on systolic blood pressure and heart rate were observed for 30 minutes.

Statistical Analysis:-

The results were analyzed at help of the Excel and expressed as mean \pm ESM. Analysis of variance (ANOVA) was done for batch comparison. The difference between the batches was significant at least at the $p < 0.05$ threshold.

Results:-**Phytochemical screening:-**

The phytochemical screening was carried out from the total extracts to identify the main chemical groups present in the plant. Due to the results obtained, shown in table 1, it appears that the total extracts of *Hallea ledermannii* contain flavonoids, terpenoids, saponosides, cathechic and gallic tannins, anthraquinones, anthocyanins, alkaloids and reducing compounds. These plant extracts tested do not contain carotenoids.

Totals phenolic and flavonoid Contents:-

The results of the quantitative analysis of the total phenols and flavonoids are reported in table 2. The total phenol contents of the extracts are expressed in milligrams equivalent of gallic acid per gram of dry plant material (standard equation of the calibration curve: $Y = 0.0004X + 0.0053$, $R^2 = 0.9929$) and those of total flavonoids are expressed in milligrams quercetin equivalent per gram of dry plant material (standard equation of the calibration curve: $Y = 0.0005X + 0.0044$, $R^2 = 0.9963$). The polar extracts of *Hallea ledermannii* present low totals phenol and flavonoid

contents. Water-ethanol extract has the highest total phenol content (3.55675 ± 0.083 mg GAE/g of drug) and the total ethanolic extract in total flavonoids (1.2272 ± 0.197 mg Q E/g of drug).

Acute toxicity:-

The aqueous extract of *Hallea ledermannii* (50, 300, 2000 and 5000 mg/kg) does not modify spontaneous motility, reactions to external stimuli, stool condition, nor does it provoke aggressiveness, ptosis, piloerection, convulsions of mice. No mortality of mice was noted at the doses studied. The LD₅₀ is greater than 5000 mg/kg. Regarding the weight evolution, the results obtained show that the mice which received the aqueous extract of *Hallea ledermannii* gain weight compared to the control mice (distilled water). The weight gain of the mice is greater with the dose of 300 mg/kg (figure 1). The results of figure 2 show that the food intake in the mice treated with the aqueous extract of *Hallea ledermannii* at the different doses studied is greater compared to that of the control mice having received distilled water at 0.5 mL/100 g.

Effects of the aqueous extract of *Hallea ledermannii* on cardiovascular parameters:-

Intravenous administration of the aqueous extract of *Hallea ledermannii* (10, 20 and 30 mg/kg) causes a significant immediate decrease in systolic blood pressure respectively of 12.83 ± 1.33 (p <0.05); 21.86 ± 1.86 (p <0.01) and 27.37 ± 1.71 % (p <0.01) followed by rising and then permanently relapsing below the initial values from the 10th minute (figure 3). At these doses, significant decreases in heart rate are respectively 4.97 ± 1.63 (P > 0.05); 7.28 ± 1.17 (p <0.05) and 12.78 ± 1.93 % (p <0.01). In rats treated with the NaCl (0.9 %) physiological solution, immediate decreases in systolic blood pressure (SBP) and cardiac frequency (CF) are not observed (Figure 4).

Table 1:-Phytochemical screening of total extracts of *Hallea ledermannii*

Chemical groups	<i>Hallea ledermannii</i>
Alkaloids	++
Anthocyanins	+++
Anthraquinones	+++
Flavonoids	++
Terpenoids	+++
Saponosids	+++
Cathechics tannins	+
Gallics tannins	++
Reducing compounds	+++
Carotenoids	-

+++ = Very abundant; ++ = Abundant; + = not abundant; - = Not detected

Table 2:- Total phenolic content (TPC) and Total flavonoid content (TFC).

Extracts	Yields (%)	TPC (mg GAE/g of drug)	TFC (mg QE/g of drug)
EE de <i>Hallea l.</i>	6.545	$2.78675 \pm 0,110$	$1.2272 \pm 0,197$
EHE de <i>Hallea l.</i>	13.245	$3.55675 \pm 0,083$	$0.7232 \pm 0,187$
EA de <i>Hallea l.</i>	7.172	$3.27425 \pm 0,101$	$0.8072 \pm 0,129$

Hallea l. = *Hallea ledermannii*; EE = Ethanol extract; EHE = Water-ethanol extract; EA = Aqueous extract; All measurements were reproduced three times.

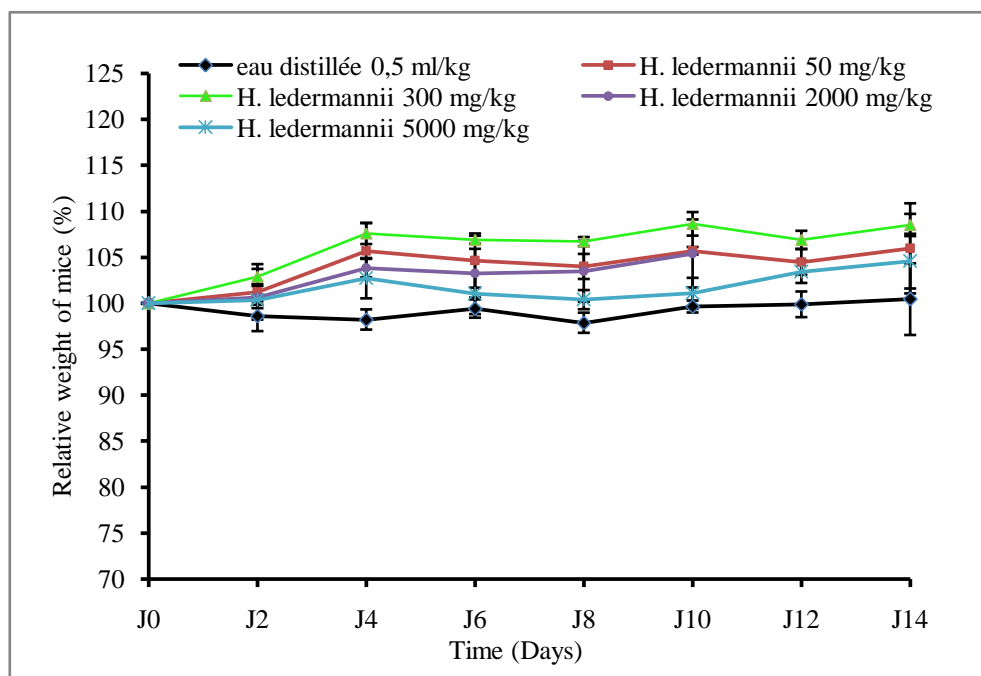


Figure 1:- Effects of the aqueous extract of *Hallea ledermannii* on the weight evolution of mice. Each point represents the mean \pm ESM, with $n = 3$. * $p < 0.05$, ** $p < 0.01$ and *** $p < 0.001$.

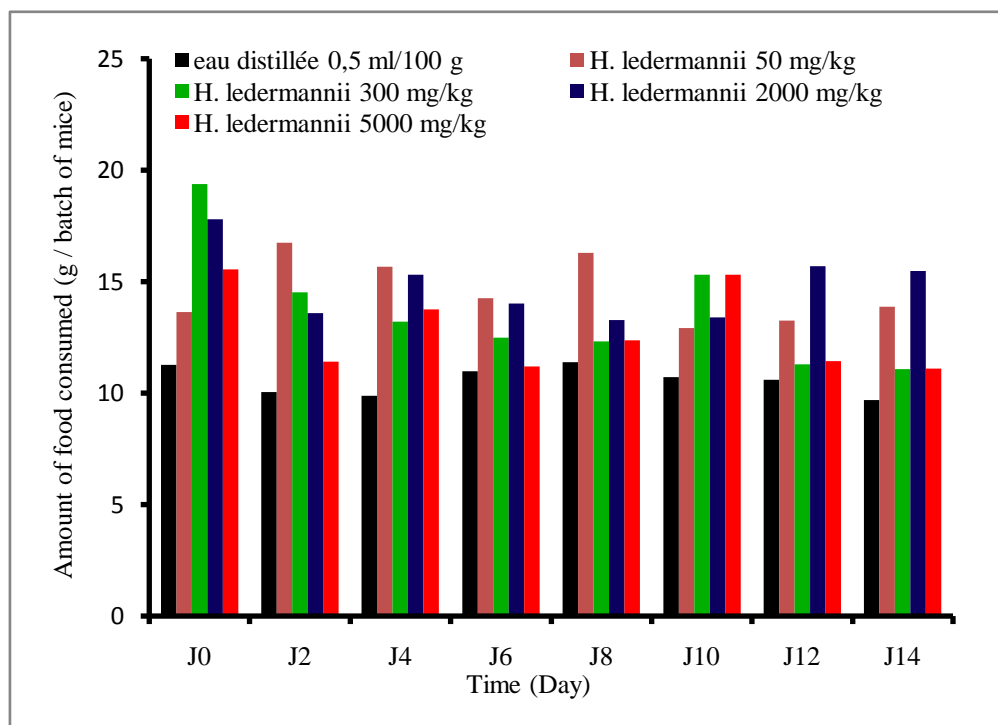


Figure 2:- Effects of aqueous extract of *Hallea ledermannii* on the feed intake of mice. Each value is a mean \pm ESM, with $n = 3$.

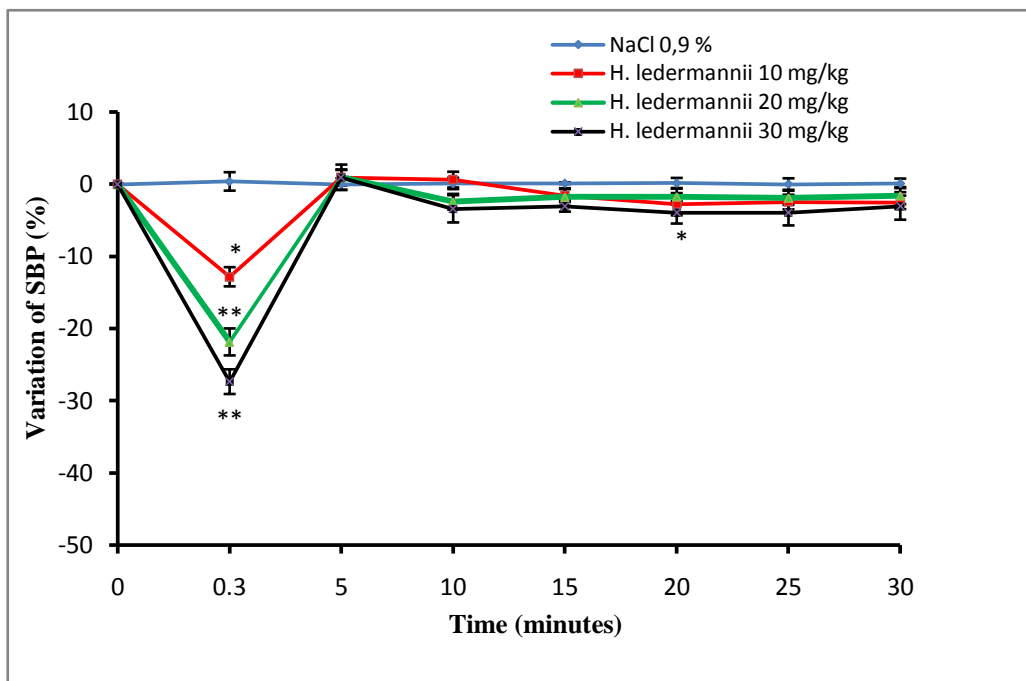


Figure 3:- Effects of the aqueous extract of *Hallea ledermannii* on systolic blood pressure in NTR. Each point represents the mean \pm ESM, with $n = 5$. * $p < 0.05$ and ** $p < 0.001$ significant difference from the initial value.

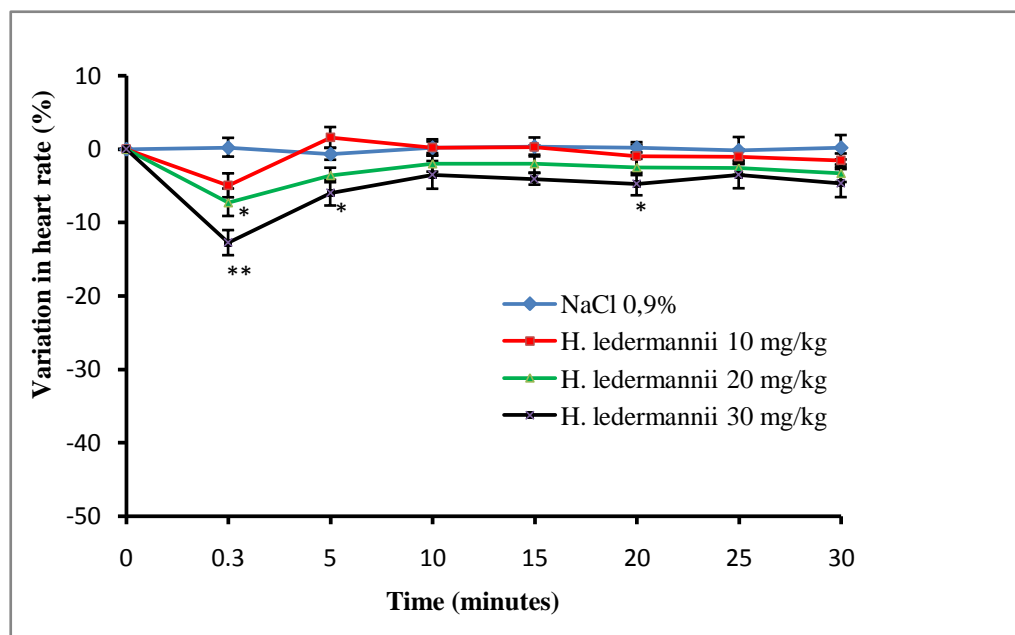


Figure 4:- Effects of *Hallea ledermannii* aqueous extract on heart rate in NTR. Each point represents the mean \pm ESM, with $n = 5$. * $p < 0.05$ and ** $p < 0.001$ significant difference from the initial value.

Discussion:-**Phytochemical Screening:-**

Eight chemical families have been identified: alkaloids, anthocyanins, anthraquinones, flavonoids, terpenoids, saponosides, tannins and reducing compounds. The test carried out on the carotenoids was negative. *Hallea ledermannii* is therefore a medicinal plant rich in secondary metabolites. These results are in agreement with those obtained by Mibindzou (2004) and Souleymane (2004) on the bark of *Hallea ledermannii*, harvested respectively in Libreville (in the South of Gabon) and in the Region of Abidjan (Côte d'Ivoire). On the other hand, BIDIE et al (2011) mentioned the total absence of gallic tannins in the methanol extract of the *Hallea ledermannii* bark harvested in the central western forest region in the Issia department of Côte d'Ivoire. This difference in chemical composition could be explained by the environment, geographical origin and harvest period, drying time, climate, parasites and extraction method (Svoboda and Hampson, 1999).

Totals phenolic and flavonoid contents:-

The results of the quantitative study show that the polar extracts of *Hallea ledermannii* have of the low contents of total phenols and total flavonoids. Water-ethanol and ethanol extracts have the highest contents respectively of total phenols and flavonoids. These results are different from those obtained by Adeleke et al (2012), on *Hallea ledermannii* leaves harvested in Nigeria, with a high total phenol content (88 mg GAE/ g of drug). This difference is probably due to various conditions especially plant maturity, environment, organ used, place of harvest, drying time, extraction method, climatic and environmental parameters (Svoboda And Hampson, 1999).

Acute toxicity of aqueous extract of Hallea ledermannii:-

The study of acute oral toxicity shows that the aqueous extract of *Hallea ledermannii* does not modify the behavior or the general state of the mice. Also, this extract does not cause the death of the mice up to the dose of 5000 mg/kg. The aqueous extract of *Hallea ledermannii* is well tolerated and according to the System of Classification harmonized globally (GHS), the LD50 of this extract would be greater than 5000 mg/kg (OECD, 1998). Moreover, the results of the present study show that this extract causes an increase in the body weight of the mice and their feed intake compared to the controls (distilled water). The increase in food intake could be due to the stimulation of appetite by this extract. The weight gain of the mice which received the aqueous extract of *Hallea ledermannii* could be explained by their high food consumption. Indeed, it is reported that weight gain is proportional to food consumption (Guyton, 1989).

Effects of the aqueous extract of Hallea ledermannii on cardiovascular parameters:-

The aqueous extract of *Hallea ledermannii* causes immediate decreases in systolic blood pressure (SBP) and heart rate (HR) in normotensive rats. This extract therefore causes hypotension and a negative chronotropic effect. The negative chronotropic effect could in part explain the observed hypotensive effect of this extract. Indeed, it has been shown that the decrease in heart rate following the administration of plant extracts would be the cause of the drop in blood pressure (Etou Ossibi and al., 2010, 2014 and 2016). The rapid rise in systolic blood pressure could be explained by a reflex phenomenon consecutive the increase in the discharge of catecholamines (Guyton, 1989). Moreover, the presence of flavonoids, tannins and alkaloids in this plant could also explain the hypotensive activity of this extract (Souleymane, 2004; Bruneton, 1999). Previous studies have shown the vasculo-protective and vasodilating effects of polyphenols (flavonoids and tannins); effects involved in decrease blood pressure (Martin and Andriantsitohaina, 2002; Falleh and al., 2008; Xu and al., 2007).

Conclusion:-

Hallea ledermannii is a medicinal plant rich in secondary metabolites and well tolerated in mice with an LD50 greater than 5000 mg/kg. It is therefore relatively non-toxic and therefore can constitute a good drug for pharmacological studies. The abundance of phenolic compounds this plant, reputed as vasodilators, could justify its use in traditional Gabonese medicine in the treatment of high blood pressure. These preliminary results could be very useful in the search for new active medications capable of effectively fighting cardiovascular diseases. Subsequent analyzes will enable to highlight the molecules responsible for this hypotensive activity and study their action mechanisms.

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RESEARCH ARTICLE

HYDROGEOCHEMICAL STUDIES AND GROUNDWATER QUALITY EVALUATION IN HANUR WATERSHED, KOLLEGAL TALUK, CHAMARAJNAGAR DISTRICT, KARNATAKA STATE, SOUTH INDIA.

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Abstract

Groundwater is becoming a scant resource in part of Kollegal taluk, Chamarajnagar District, Karnataka State. This area receives an average annual rainfall of 696 mm. Groundwater has been the major sources of irrigation and drinking purpose for several years almost 80 percent of area is underlain by hard rocks. The study area is drought prone, most of the people in this area depend on groundwater for drinking water and agriculture. Therefore, constant monitoring and assessment of water resource is necessary. The study area is bounded by Yelandur and T. Narasipura Taluk to the west, Mandya and Bangalore to the North, and East and South it is bounded by Dharamapuri District of TamilNadu this area is located in the south eastern corner of Karnataka. The area is located between $77^{\circ} 51'$ to $77^{\circ} 30'$ East longitude $11^{\circ} 45'$ to $12^{\circ} 15'$ North latitude with an areal extent of 1026sq km covering 138 villages coming under the Survey of India Toposheet (SOI) numbers are 57 H/4, 57H/7, 57H/8,, 58E/1 and 58E/5. The study area comes under the semiarid region. In recent years, this region has been facing water scarcity as well as water quality problems. The major sources of employment are sericulture, agriculture and horticulture, engaging almost 70% of the workforce. Water samples have been collected from 68 locations of the study area, during the monsoon year 2016, The collected samples were analysed for chemical parameters. In the study area Ca-Mg-HCO₃ type of water predominates during the monsoon season of year 2016. On the basis of hydro-chemical facies classification the water is found to be suitable for irrigation. The other evaluated parameters are SAR, RSC, salinity sodium hazard and USSL Classification.

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Introduction:-

Water is the most important commodity for all living things, in rural as well as urban areas. Because of increase, in agricultural and domestic activities, the demand is increasing. Quantity of water without quality is of no utility and hence chemical quality of water has gained equal importance along with quantity. The quality of any water may be altered due to over usage of fertilizers and other biogenic activity. The chemistry of host rock through which the groundwater flows below the earth surface and the total time of residence of the water with the host material control the chemistry of groundwater. The present study aims to obtain comprehensive information on the quality of

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groundwater of Hanur Watershed which lies on South Eastern parts of Kollegal Taluk, Chamarajnagar District, Karnataka state, India. The study area comes under Southern Dry Zone-VI Groundwater samples were collected and analyzed for determining its quality in the laboratory.

Study Area:-

Hanur Watershed of Kollegal taluk, Chamarajnagar District is located in the south eastern corner part of Karnataka state between $77^{\circ} 51'$ to $77^{\circ} 30'$ East longitude $11^{\circ} 45'$ to $12^{\circ} 15'$ North latitude (**Figure 1**). It covers an area of 1026 sq.km. Groundwater is influenced by underlaying lithology, structures, soil types thickness and structure of rock formations. Weathered and fractured charnokites, hornblende/ampibolite schist form the main aquifer in Hanur watershed. Groundwater in the study area occurs under water table conditions in the weathered and fractured charnokites, hornblende/ampibolite schist (**Figure 2**); there is no perennial river in the study area. Most of the people depend on groundwater resource for their domestic and agricultural requirements, the major ion chemistry of groundwater of Hanur watershed has not been studied earlier. During the monsoon period in the year 2016, 68 Samples were collected from the different locations of the study area. The collected water samples were transferred into precleaned polythene container for analysis of chemical characters. Chemical analyses were carried out for the major ion concentrations of the water samples collected from different locations using the standard procedures recommended by APHA-1994. The analytical data has been used for the classification of water for utilitarian purposes and for ascertaining various factors on which the chemical characteristics of water depend.

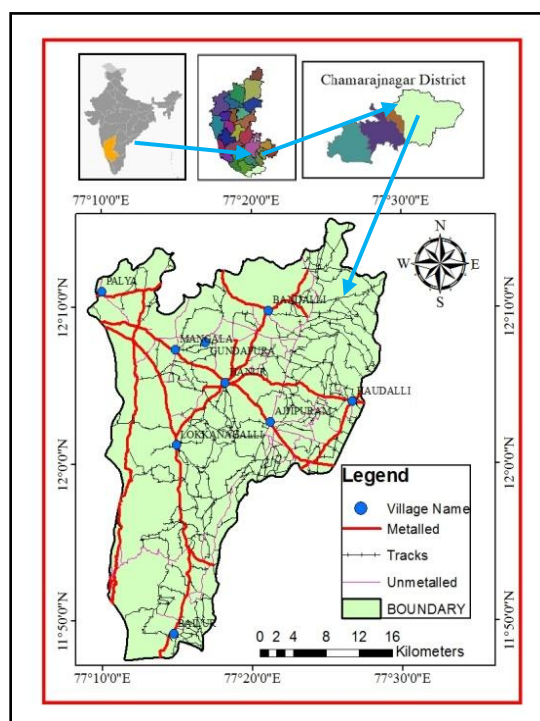


Figure 1:- Location map of the study area.

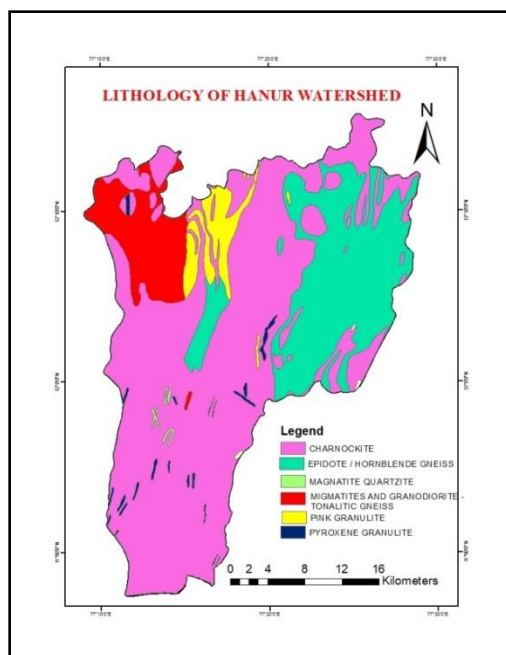


Figure 2:- Lithology of Study area.

Methodology:-

As stated earlier, 68 locations Groundwater samples were collected from Hanur watershed during monsoon period 2016 (**Figure 3**). The collected water samples were stored in pre-treated polythene bottles and transferred laboratory for analysis of major chemical elements - Ca^{+2} , Mg^{+2} , Cl^{-1} , HCO_3^{-1} , Na^{+1} , K^{+1} , and SO_4^{-2} . Using the standard procedures recommended by APHA-1994.

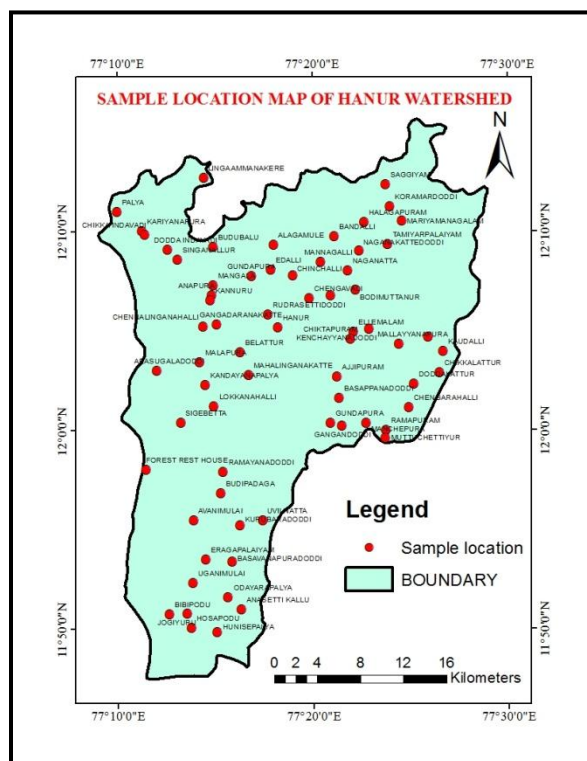


Figure 3:- Sample Location of study area

Results and Discussion:-

The Groundwater quality reveals that the pH varies from 6.28 to 9.4, EC ranges from 86 to 2670 $\mu\text{mho/cm}$, total dissolved solids(TDS) values range from 385 to 1926 mg/l, calcium varies from 20 to 179 mg/l, Mg varies from 14 to 109 mg/l, Na+k varies from 21 to 546 mg/l, Cl values range from 14 to 376 mg/l, Co3 values range from 0 to 93 mg/l, HCO3 values range from 152 to 646 mg/l, SO4 values range from 0.5 to 552 mg/l, NO3 values range from 0 to 62 mg/l in water of the study area. Chemical data of water samples are given in Table 1. Year -2016

Table 1:- Different chemical parameters of Hanur watershed -2016 (PPM)

Sl no	TDS	EC	Ca	Mg	Na+K	HCO3	CO3	Cl	NO3	SO4	pH
1	795	1220	165.0	73.0	98.0	530.0	10.0	230.0	62.0	89.0	7.09
2	786	1200	162.0	75.0	92.0	526.0	10.0	234.0	1.0	1.6	7.06
3	480	733	33.6	47.0	66.5	310.0	10.0	88.0	0.2	0.6	8.1
4	478	730	20.0	38.0	57.0	300.0	10.0	40.0	0.5	13.8	7.65
5	485	736	22.0	42.0	62.0	320.0	10.0	52.0	0.4	14.0	7.68
6	500	740	22.0	42.0	63.0	294.0	10.0	63.0	0.8	22.8	7.72
7	490	746	21.0	37.0	58.0	264.0	10.0	55.0	10.0	15.6	7.75
8	680	860	69.0	42.0	86.0	450.0	35.0	58.0	7.0	20.0	7.6
9	700	1020	160.0	75.0	62.0	486.0	10.0	198.0	18.8	75.4	6.85
10	810	1185	157.0	71.0	62.0	505.7	10.0	210.0	14.5	81.8	6.28
11	495	650	56.0	33.0	90.0	436.4	24.0	42.0	2.6	12.0	8.6
12	838	2320	16.0	68.0	465.0	646.0	42.0	322.0	12.0	225.0	7.25
13	760	1100	32.0	38.0	266.0	520.0	36.0	204.0	22.0	28.0	7.02
14	554	2050	50.0	36.0	186.0	486.0	10.0	143.0	14.2	16.8	7.36
15	618	642	62.0	28.0	126.0	455.0	50.0	34.0	18.0	26.0	7.8
16	834	1516	50.0	69.0	160.0	484.0	40.0	138.0	25.0	94.0	7.78
17	615	86	60.0	28.0	126.0	340.0	38.0	66.0	16.0	54.0	8.32
18	250	380	34.0	15.0	40.0	152.0	43.0	17.0	10.0	10.0	8.7
19	400	826	44.0	24.0	71.0	280.0	45.0	22.0	13.0	22.6	8.6
20	447	596	42.0	25.0	84.0	316.0	50.0	30.0	10.0	0.5	8.56
21	640	978	80.0	35.0	74.0	295.0	44.0	96.0	16.0	42.0	7.82
22	1136	1016	90.0	97.0	312.0	712.0	10.0	86.0	14.0	552.0	7.76
23	580	628	60.0	25.0	84.0	378.0	48.0	14.0	15.0	16.0	7.82
24	725	610	48.0	30.0	112.0	318.0	35.0	54.0	16.0	48.0	7.8
25	552	884	28.0	35.6	138.0	398.6	54.4	38.2	3.4	24.0	7.4
26	518	860	44.0	47.0	136.0	390.0	72.0	50.0	16.0	78.0	8.2
27	442	820	38.0	62.0	99.0	420.0	40.0	62.0	8.2	62.0	7.6
28	600	830	45.0	43.0	260.0	573.0	72.0	76.0	12.0	120.0	8.3
29	1082	1020	71.0	33.0	101.0	442.0	0.0	54.0	15.2	82.0	8.4
30	858	984	61.0	29.0	99.0	360.0	28.0	37.0	13.2	86.0	8
31	718	480	44.0	42.0	111.0	432.0	36.0	49.0	10.0	38.0	8.2
32	624	782	48.0	30.0	144.0	434.0	58.0	46.0	10.0	32.0	7.72
33	496	548	58.0	34.0	90.0	435.0	26.0	44.0	3.8	14.0	8.48
34	510	992	62.0	82.0	101.0	588.0	46.0	70.0	14.2	45.0	7.8
35	420	834	48.0	58.0	72.0	410.0	34.0	58.0	6.4	36.0	7.62
36	518	712	56.0	34.0	80.0	395.0	46.0	16.0	18.0	16.0	8.08
37	560	860	46.0	48.0	100.0	355.0	43.0	40.0	14.0	80.0	8.7
38	775	1200	61.0	48.0	145.0	196.0	24.0	216.0	14.0	100.0	7.97
39	324	566	58.0	16.0	45.0	214.0	26.0	22.0	14.0	20.0	7.6
40	808	1040	140.0	16.0	124.0	438.0	50.0	110.0	16.0	52.0	7.92
41	1926	2080	46.0	68.0	546.0	905.0	93.0	205.0	14.0	342.0	7.8
42	520	840	69.0	28.0	78.0	328.0	48.0	59.0	13.0	20.0	8.43
43	528	995	30.0	48.6	72.0	328.4	33.8	47.8	10.0	30.0	9.4
44	560	1000	126.0	37.0	50.0	421.0	29.0	78.0	15.0	50.0	7.66
45	642	998	54.0	43.0	98.0	310.0	50.0	96.0	16.0	45.0	8.8
46	600	782	62.0	36.0	112.0	408.0	54.0	28.0	34.0	30.0	8.5

47	618	1018	48.0	79.0	50.0	437.0	43.6	60.0	12.0	24.6	8
48	676	812	75.0	40.0	105.0	478.0	40.0	50.0	18.0	10.0	7.62
49	576	912	86.0	43.0	115.0	578.0	44.0	52.0	19.0	10.0	8.15
50	680	1250	135.0	42.0	48.0	433.0	52.0	85.0	21.0	50.0	7.15
51	635	1215	129.0	38.0	51.5	415.0	25.0	78.0	35.0	47.0	7.69
52	598	896	43.0	28.0	110.8	192.0	0.0	188.0	1.4	56.0	7
53	605	1100	128.0	38.0	51.0	425.0	30.0	80.0	41.0	52.0	7.56
54	825	1050	140.0	14.0	126.1	437.0	48.0	110.0	40.0	50.0	7.79
55	721	910	48.0	31.0	122.0	205.0	0.0	201.0	1.8	61.0	7.5
56	545	710	30.0	41.0	97.0	450.0	0.0	30.0	30.0	21.0	8.5
57	1010	1700	158.0	30.0	163.6	470.0	78.0	172.0	56.0	67.0	8.5
58	545	715	53.0	47.0	96.0	400.0	48.0	38.0	31.0	22.0	8.5
59	889	1453	88.0	57.0	73.0	428.0	0.0	116.0	25.6	78.0	6.7
60	1838	2670	179.0	109.0	116.0	544.0	0.0	376.0	4.7	164.0	7.1
61	410	510	35.0	21.0	76.0	260.0	35.0	19.0	14.0	17.0	7.93
62	385	635	44.0	27.0	76.0	268.0	6.0	30.0	58.0	55.0	8.43
63	710	535	36.0	33.0	73.0	275.0	14.0	33.0	59.0	44.0	7.4
64	690	875	71.0	40.0	106.0	480.0	40.0	50.0	9.0	12.0	7.61
65	720	1150	45.0	41.0	30.0	335.0	0.0	51.0	0.2	2.1	7.4
66	650	1200	35.0	31.0	21.0	210.0	0.0	52.0	5.0	14.0	7
67	733	1100	70.0	52.0	33.0	410.0	0.0	50.0	55.0	12.0	7
68	644	981	98.0	28.0	53.0	333.0	62.0	71.0	0.0	3.0	7.1

Total Dissolved Solids:-

As groundwater moves and stays for a longer time along its flow path, increased in total dissolved concentrations and major ions normally occur (Norris et al. 1992). Higher TDS shows longer residence period of water (Davis and De Viest, 1966). TDS content is usually the main factor, which limits or determines the use of groundwater for any purpose (Nordstrom, 1987). The analysis of samples for the seasons has shown that there is a general tendency of increase of TDS, based on TDS, for various uses like general household, drinking, irrigation and understanding and it could be seen from the (table.1) that a majority of the portion is occupied by 500 to 1000 ppm TDS followed by <500 ppm concentration and finally >1000 ppm. During the stay or movement of the groundwater in the subsurface the TDS concentration slowly gets enriched and shown in figure 4.

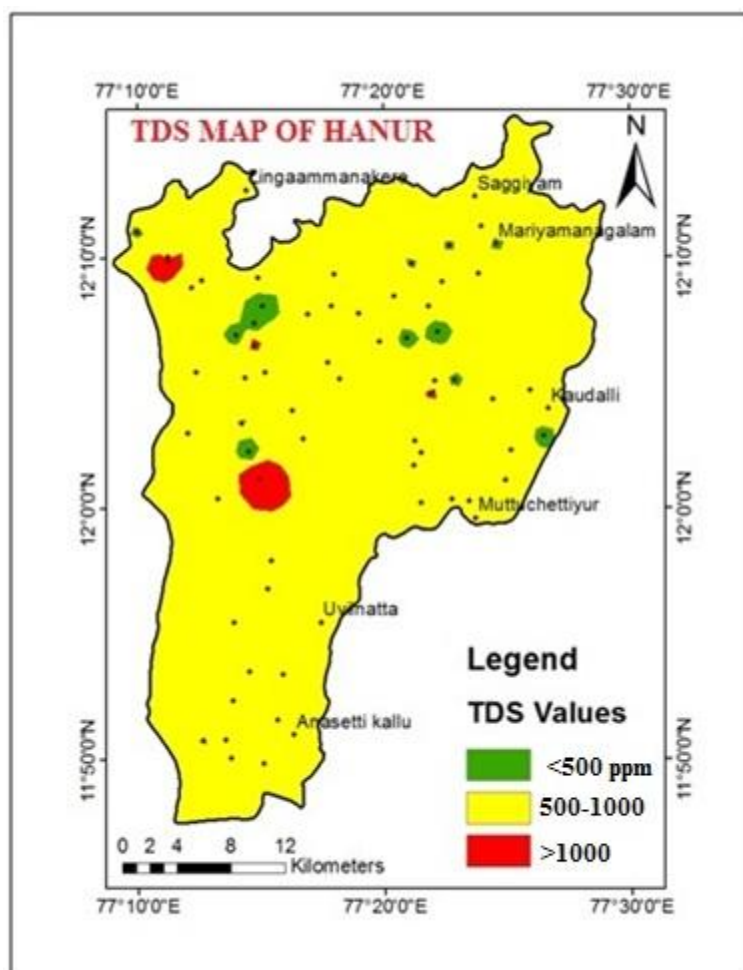


Figure 4:- Total dissolved solids of study area.

Hydrochemistry of Groundwater:-

Piper (1944, 1953) introduced a trilinear diagram which is used to evaluate the geochemical evolution of groundwater and relationship between rock type and water composition. Analysis of piper's trilinear plot shows that most of ground water samples are of Ca-Mg - HCO_3 type (Figure 5).

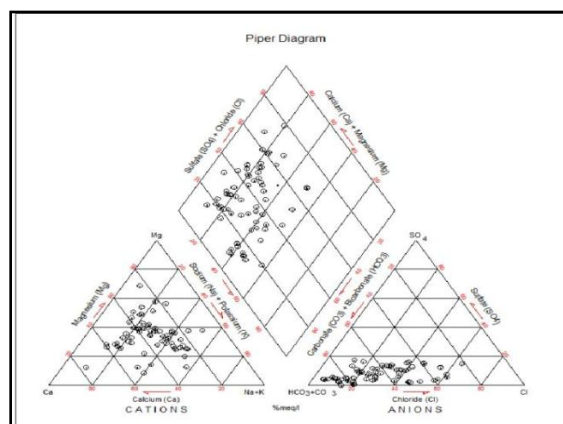


Figure 5:- Plotted in piper-Trilinear diagram.

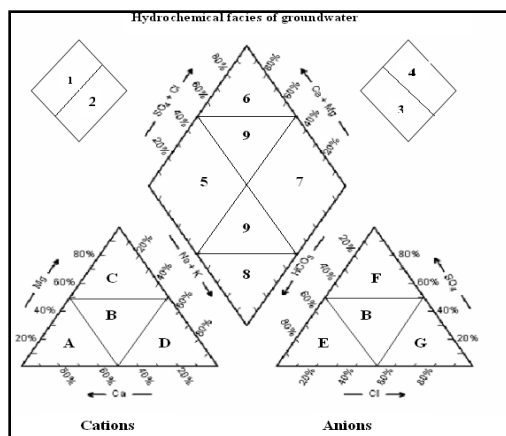


Figure 6:- Classification diagram for anion and cation facies in the form of major –ion percentages. Water types are designed according to domain in which they occur on the diagram segments.

Legend:-

A- Calcium type, B- No Dominant type,
C- Magnesium type, D- Sodium and potassium type
E- Bicarbonate type, F- Sulphate type,
G- Chloride type

Hydrochemical facies are distinct zones that projects the dominating cation and anion concentration in water. To define the composition class, Back and co-workers suggested various sub divisions within the tri-linear diagram (**Figure 5**). The interpretation of distinct facies from the 0 to 10% and 90 to 100% domains, on the diamond shaped cation to anion graph, is more helpful than using equal 25% increments. It clearly explains the variations or domination of cation and anion concentrations. Groundwater Hardness is important parameters for water use and consumption. Hardness is caused primarily due to the presence of cations such as calcium and magnesium and anions such as carbonate, bicarbonate, chloride and sulfate in water. Wilcox classified groundwater for irrigation purposes based on % sodium and electrical conductivity. Eaton recommended the concentration of Residual Sodium Carbonate to determine the suitability of water for irrigation purposes. The US-Salinity Laboratory of the Department of Agriculture adopted certain techniques based on which the suitability of water for agriculture is explained. The sodium in irrigation waters is usually denoted as per cent sodium and can be determined using the following formula.

$$\% \text{Na} = (\text{Na}^+) \times 100 / (\text{Ca}^{2+} + \text{Mg}^{2+} + \text{Na}^{+1} + \text{K}^{+1})$$

That is the quantities of Ca^{2+} , Mg^{2+} , Na^{+} and K^{+} are expressed in milliequivalents per liter (epm).

Table 2:- Classification of water based on % sodium

%Na	Water Class	No. Samples	%	Sample Nos
Up to 20	Excellent	10	14.70	9,10,44,47,50,51,53, 65,66,67
20 - 40	Good	24	35.30	1,2,3,4,5,6,7,8,18,21,27,34,35,36,39,40, 42,43,48,49,58,59, 60,68
40 - 60	Permissible	30	44.12	11,14,15,16,17,19, 20,22,23,24,25,26,29,30,31,32,33,37,38,45,46,52,54,55,56,57,61,62,63,64.
60 - 80	Doubtful	4	5.88	12,13,28,41
Total		68	100	

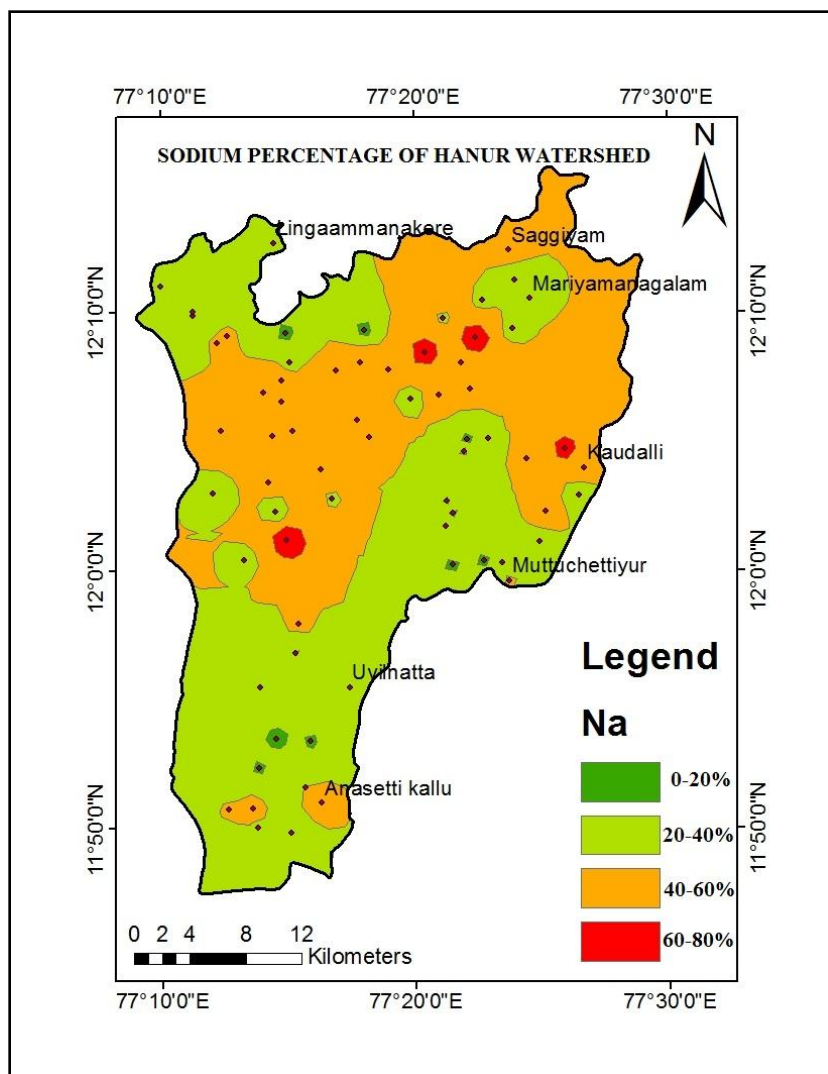


Figure.7 Sodium Percentage of the study area

Table 3:- Groundwater quality based on RSC (Residual Sodium Carbonate).

RSC (epm)	Water Class	No Samples	%	Sample Nos
< 1.25	Good	26	38.24	3,4,5,6,7,16,18,21,22,27,29,35,37,39,40,42,43,45,47,54,62,63,65,66,67, 68.
1.25 - 2.5	Doubtful	22	32.35	8, 11, 17,19,24,30, 33,34,36,44,46,48,50,51,52,53,55,56,58,59,61,64.
>2.5	Unsuitable	20	29.41	1, 2,9,10,12,13,14, 15,20,23,25,26,28,31,32,38,41,49,57,60.
Total		68	100	

The classification of groundwater samples with respect to per cent sodium is shown in (Table 2). It is observed that about 10 samples are excellent, 24 samples are good, 30 samples are permissible in this limit and 4 samples are doubtful. In waters having high concentration of bicarbonate, there is tendency for calcium and magnesium to precipitate as the water in the soil becomes more concentrated. As a result, the relative proportion of sodium in the water is increased in the form of sodium carbonate. RSC is calculated using the following equation.

$$\text{RSC} = (\text{HCO}_3^- + \text{CO}_3^{2-}) - (\text{Ca}^{2+} + \text{Mg}^{2+})$$

That is all ionic concentrations are expressed in epm.

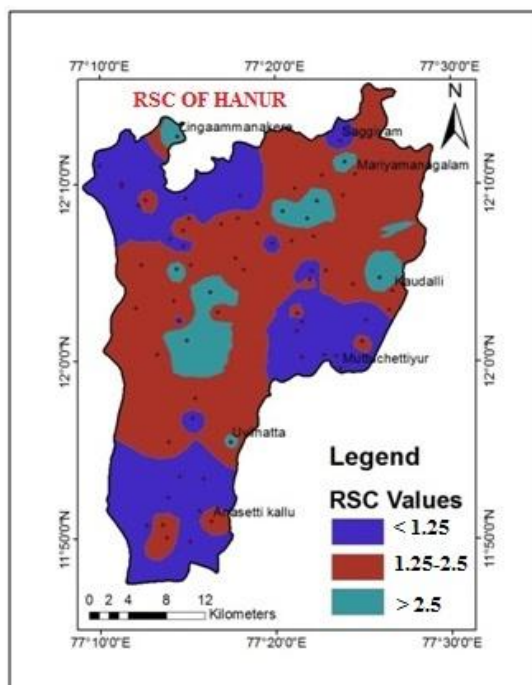


Figure 8:- Residual Sodium carbonate of the study area.

According to the US Department of Agriculture, water having more than 2.5 epm of RSC is not suitable for irrigation purposes. Groundwater of the study area is classified on the basis of RSC and the results are presented in **(Table.3)**. Based on RSC values, out of 68 samples, 8 samples have shown values less than 1.25 and are safe for irrigation.

The most important characteristics of irrigation water in determining its quality are: (i) Total concentration of soluble salts; (ii) Relative proportion of sodium to other principal cations; (iii) Concentration of boron or other element that may be toxic, and (iv) Under some condition, bicarbonate concentration as related to the concentration of calcium plus magnesium. These have been termed as the salinity hazard, sodium hazard, boron hazard and bicarbonate hazard. In the past, the sodium hazard has been expressed as per cent sodium of total cations. A better measure of the sodium hazard for irrigation is the SAR which is used to express reactions with the soil. SAR is computed as where all ionic concentrations are expressed in epm.

$$SAR = \frac{Na^+}{\left\{ \frac{Ca^{2+} + Mg^{2+}}{2} \right\}^{1/2}}$$

Table 4:- Classification of Sodium Adsorption Ratio.

SAR	Water class	No. of Samples	%	Sample Nos
<1	Excellent	4	5.88	50,65,66,67
>1	Good	64	94.12	1,2,3,4,5,6,7,8,9,10,11,12,13,14,15,16,17,18,19,20, 21,22,23,24,25,26,27,28,29,30,31,32,33,34,35,36,37,38,39,40,41,42,43,44,45,46,47,48,49,51,52,53,54,55,56,57,58,59,60,61,62,63,64,68.
Total		68	100	

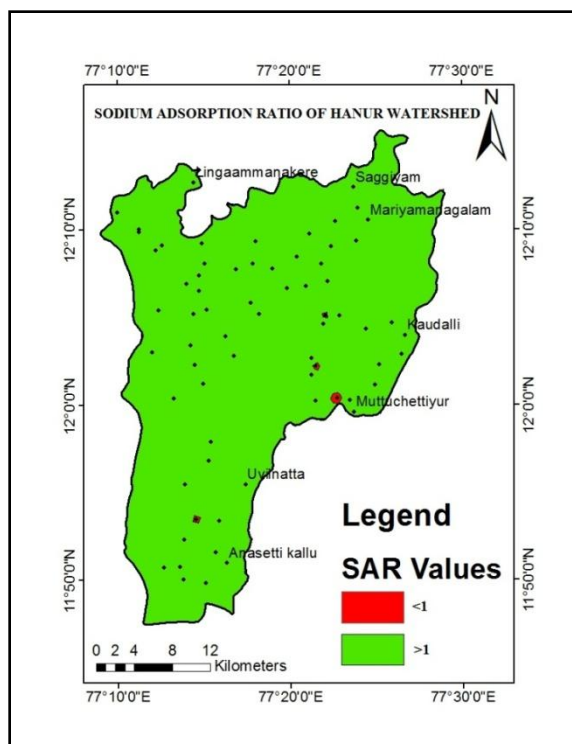


Figure 9:- Sodium Adsorption of the study area.

The classification of groundwater samples from the study area with respect to SAR is represented in (Table 4). The SAR value of all the samples are found to be less than 6, and are classified as excellent for irrigation. When the SAR and specific conductance of water are known, the classification of water for irrigation can be determined by graphically plotting these values on the US salinity (USSL) diagram (**Figures 10&11**). The groundwater of Hanur watershed is in general Ca-Mg-HCO₃ type during year 2016. About 88.24 % of the samples are grouped within C2S1 and C3S1 classes.

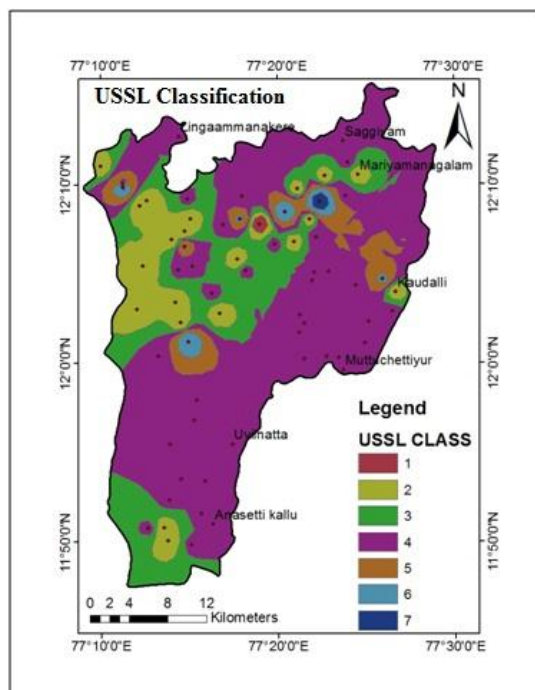
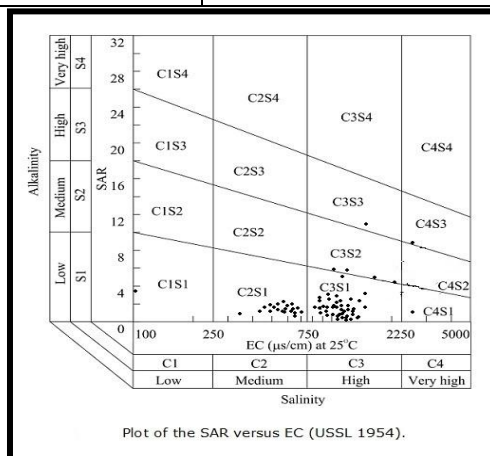


Figure 10:- Salinity Sodium hazard of study area.

Table 5:- Stastical data of Salinity Sodium hazard (USSL Classification)

Rank	Quality Parameter	No. of Samples	Percentage
1	C ₁ S ₁	1	1.47
2	C ₂ S ₁	21	30.88
3	C ₃ S ₁	39	57.36
4	C ₃ S ₂	4	5.88
5	C ₃ S ₃	1	1.47
6	C ₄ S ₁	1	1.47
7	C ₄ S ₃	1	1.47
Total		68	100

**Figure 11:-** USSL classification of water samples.

For the purpose of diagnosis and classification, the total concentration of soluble salts (salinity hazard) in irrigation water can be expressed in terms of specific conductance. Classification of groundwater based on salinity hazard is presented in Table 5. It is found from the EC value; only 2 samples are found to be unsuitable for irrigation purposes. **Table 5** shows the Salinity hazard classes. In the study area, the groundwater is generally Ca- Mg-HCO₃ type, which is mainly due to the geology of the area which comprises igneous rocks of crystalline nature, in which the major units are Charnokites and hornblende schist. Groundwater in the study area occurs under water table conditions in the weathered and fractured Charnokites and hornblende schist.

Table 6:- Comparison of the quality parameters of groundwater of the study area with WHO and ISI standard for drinking purpose.

S.N o.	Water Quality Parameters	WHO (1984)		ISI (1991)		No. of locations which exceed max. permissible limit (WHO)	Concentration in Study Area	Undesirable Effect Produced Beyond Maximum Allowable Limit
		Max Desirable	Max. Permissible	Max. Desirable	Max. Permissible			
1.	pH	7.0 to 8.5	6.5 to 9.2	6.5 to 8.5	No relaxation	1	6.28 to 9.4	Taste, effects mucus membrane and water supply system
2.	TDS mg/l	500	1500	500	1000	2	250 to 1926	Gastrointestinal irritation
3.	Ca mg/l	75	200	75	200	0	16 to 179	Encrustation in water supply, scale formation
4.	Mg ml/l	30	150	30	100	0	14 to 109	Encrustation in water supply and adverse effect on domestic use.
5.	Na mg/l	-	200	-	200	2	15 to 248	--
6	Cl mg/l	200	600	250	1000	0	14 to 322	Salty Taste
7.	SO ₄ mg/l	200	400	150	400	1	0.5 to 552.	Laxative effect.

Conclusions:-

- Ca-Mg-HCO₃ type of water predominates in the study area during the Year 2016,
- Though the suitability of water for irrigation is determined based on SAR, %Na, RSC and Salinity hazard, it is only an empirical conclusion. In addition to water quality, other factors like soil type, crop type, crop
- Pattern, frequency and recharge (rainfall), climate, etc. have an important role to play in determining the Suitability of water.
- Water that is not suitable based on the above classification may be suitable in well-drained Soils.
- The suitability of water for irrigation is evaluated based on SAR, %Na, RSC and salinity hazards. Most of
- the samples in Study area fall in the suitable range for irrigation purpose either from SAR, % Na or RSC
- Values.
- About 98% of the samples are grouped within C₂S₁ and C₃S₁ classes (Figures 10 & 11).
- Most of the samples in study area fall in the suitable range for irrigation purpose from USSL diagram.

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RESEARCH ARTICLE

HUMANIZING THE IMPACT OF DISRUPTIVE CLASSROOM ENVIRONMENT ON NIGERIAN STUDENTS DISCIPLINE.

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Disruptive behavior, Insecurity, Students' discipline, School environment and Classroom management.

Abstract

Disruptive classroom environment has been a worrisome problem challenging school systems in Nigeria. It is filled with high incidence of disruptive behaviors ranging from noise making, eating, sleeping, inattentiveness, playing, etc. The paper looks into the concept of disruptive classroom behavior, forms, causes and ways of ameliorating disruptive behavior. The study concluded that when teachers use better instructional programs, there will be producing academic progress. Finally the paper recommended among others that teachers should endeavor to manage their classroom activities effectively and build positive interpersonal relationship in students, use viable instructional approach which directs them to concentrate in other to avoid disruptive classroom behaviour, assess students properly and counsel them from displaying social misfits' attitudes.

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Introduction:-

Disruptive classroom behavior is prevalent this 21st century. The school environment does not exist in isolation of the prevailing situations of insecurity inherent in the Nigerian context. Most disruptive behaviors that start from early school age sometimes lead to higher school age. Effective teaching and learning instruction will go down the drain without proper discipline of students. Huge investment in education according to Willy (2002) is not worthwhile without discipline. Training of a child goes hand in hand with discipline. Hence, formal education without discipline is worthless.

Disruptive behavior in the school systems has been identified as a challenge hindering a positive school environment. (Oliver, 2011). Disruptive behavior in the classroom affects the classroom environment as well as the educational experience for students enrolled in the course negatively. Disruptive behavior is referred as any behaviors that hamper instructors. Conducive classroom environment is the cornerstone of getting a good education and instilling discipline in students. Nakpodia (2010) stated that good behavior makes one to be relevant to the society. Classroom with incessant disruptive behaviors waste time and yield poor teaching and learning outcome. Continuing, a nation's building and behavior maintain peaceful environment for learning.

Conceptual Framework:-

Disruptive behavior obstructs teacher and instruction. Disruptive classroom behavior is the behavior which does not allow the teachers and students for sound teaching and learning process. Most often poses a hazardous low achievement trait for students. It is obvious that classroom with disruptive behavior have less academic engaged time. A teacher who is grossly inadequate in classroom management skills is probably not going to accomplish

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much (Marzano, 2003). This empowered Borden (2013) to posit that disruptive behavior is associated with a wide range of cognitive, educational, constitutional, and family problems. Disruptive behavior in the classroom is one of the most serious challenge hampering effective teaching and learning environment especially in secondary schools due to lack of disciplinary measure for aggression and unpleasant situations.

Insecurity denotes varied phenomena; it entails distinct meanings ranging from: anxiety, fear, un-sureness, vulnerability, unsafe, danger, hazard and uncertainty. AkporRobaro, M. (2013) sees insecurity from two perspectives identified two types of insecurity. Beland (2005) connotes insecurity with absence of protection from crime and its consequential harm. From the aforementioned points one can describe insecurity as a psychological feeling of distress or threat to a person's life; the state of being vulnerable to danger. Insecurity this is the direct opposite of the term security.

Discipline is a very necessary condition for any venture to be successful. It is the training of an individual behavior in accordance with the rules of conduct. It is a systematic method of obtaining obedience in a person. It is the correction procedures for misbehavior. The free dictionary online describes discipline as the "training expected" (www.thefreedictionary.com/discipline). It is targeted to influence an individual actions and behavior positively. Students' discipline is the systematic application of school rules, punishments, and behavior. Students' discipline has been identified as one of the essential factors that foster practices and the main purpose of individual development. Creek School District, 2015). Students' discipline presupposes a positive learning environment. (Bodens, 2013).

Classroom environment encapsulates the physical class -"personality" of teachers. Classroom is known to yield effective teaching and learning experience which is cherished by the teacher and enjoyable by the students. Some of the ways of developing a healthy classroom environment include; building caring relationships amidst staff and students, rewarding academic excellence, provision of classroom rules and regulations, encouraging participatory / cooperative learning and counseling mal-adjusted students.

Causes of Disruptive Behavior in the Classroom:-

There are three main reasons which foster disruptive classroom behavior and they are; classroom related issues, personal issues and Social issues. Most often students seem to be unsecured, bored, uninteresting, confused, inattentive, annoyed, lost, asleep, resentful and not concentrating when they perceive that the teacher is not communicating effectively in carrying them along in the teaching and learning context. Personal problems of students ranging from anxiety, depression, eating disorders, attention deficit disorder, learning difficulties, emotional (www.ul.ie). Antisocial behaviors more or less influence their dispositions for teaching and learning outcome. More still, social issues which emanate from Factors outside the classroom such as; family breakdown, changes in the society, parents' role, schools' culture, teachers' education, schools' curricula, and examination points system pose a challenge to effective teaching and learning context.

Forms of Classroom Disruptive Behavior:-

There are different types of classroom disruptive behavior in secondary schools display by students and each one has negative impact on classroom management. Some of them include; entering classroom with screaming sound, eating in class, sleeping during teaching, and chatting with one another during when lesson is going on. Others are coming to classroom habitually late, unnecessarily arguing with teachers, bringing noisy electric devices in the classroom, shouting loudly to create thrill in classroom, sluggish wandering on veranda to distract students' attention and making complains against their fellow etc.

Effective Classroom Management Techniques:-

Management skills are crucial and fundamental. Effective classroom management establishes preventive techniques towards students' indiscipline rather than engaging in procedures. Effective Classroom management entails the establishment and maintenance of teaching simultaneously smoothly. Oliver (2011) pointed out that rules and routines are powerful preventive and powerful component to classroom organization and management plans and re-taught if inappropriate behavior occurs. Planning is a condition sine qua non for effective classroom management. (www.teqjournal.org/TEQ%20Website/Back%20Issue..)

Construction of Classroom rules is also an essential, powerful and preventive technique to effective classroom organization and management. They help to coordinate students, reinforced behaviors and consequences of inappropriate behavior. The rules should be minimal, precise and positively stated and so on (www.gtlcenter.org)

Classroom Arrangement is a condition sine qua non for effective classroom management and organization technique. It promotes order and decency for the smooth flow of teaching and learning experience. Dunbar (2004) in conformity suggested decongestion of high traffic areas and that good relationship with students should be maintained.

Constructive Discipline Techniques in Overcoming Disruptive Behavior in the Classroom:-

Motivation and communications are keys in overcoming disruptive behavior in classrooms. Others include teachers' techniques, protection of students' dignity, active listening and avoidance of confrontation. Corporal punishment should be totally avoided.

Conclusion:-

It is a hallmark of an effective classroom teacher to react positively to disruptive classroom behaviour or undesirable behaviors. Teachers should establish positive to classroom management and instruction, concentrate more on well planned and implemented instruction for it goes a long way in preventing classroom problems. They should ensure self-discipline so that students can be assisted to be disciplined in order for them to understand and appreciate the teaching and learning content. Finally, teachers will have to be effectively concerned with students' academic and social disposition and manage disruptive classroom behavior with caution.

Recommendations:-

The paper recommends that:

1. Teachers should endeavor to manage their classroom activities effectively and build positive inter-personal relationship in students.
2. They should use viable instructional approach which directs them to concentrate more on well planned and implemented instruction which prevents classroom problems.
3. Teachers should carry out proper assessment of students and elementary emotional disturbance.
4. The administrators should endeavor to renovate and improve dilapidated school physical facilities.
5. Government should provide adequate classroom spaces for proper accommodation of students to avoid crowdedness that gives room to classroom disruptiveness.
6. Government should also provide enough furniture for comfortable seating arrangements to avoid disruptive classroom behaviors that may arise from struggling for seats.
7. Teachers should embark on self-discipline in order to be a role model to the students under his/her control.

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RESEARCH ARTICLE

A COMPARATIVE CLINICAL AND RADIOLOGICAL STUDY USING BIOGRAFT-HABG ACTIVE AND BIOGRAFT –CPC.

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Bioactive composite granule, Calcium phosphate cement, periodontal endosseous defects.

Abstract

Background and objectives: This clinical study was carried out on 20 systemically healthy subjects to evaluate and compare, clinically and radiographically, the efficacy of Bioactive composite granule (Biograft-HABG Active) and Calcium phosphate cement (Biograft-CPC) in the treatment of periodontal endosseous defects clinically and radiographically and compare them.

Methodology: 20 intrabony defects present in 20 patients, who fulfilled the inclusion and exclusion criteria were selected and divided into two groups. Group I received bioactive composite granule, Group II received calcium phosphate cement. Clinical and radiological parameters such as Plaque Index (PI), Gingival Index (GI), Probing Pocket Depth (PPD), Clinical Attachment Level (CAL), Gingival recession (GR) and Depth of the bone defect (BL) were assessed at baseline 3, 6 and 9 months post operatively. The results were averaged for each clinical and radiological parameter at baseline, 3 months, 6 months and 9 months.

Results: Statistically significant difference in mean values of the plaque index, gingival index, pocket depth reduction, clinical attachment level, gain amount of defect resolution were observed in both the groups at subsequent time periods.

Conclusion: Clinical and radiographic assessments reveal that both bone graft materials were efficacious in the treatment of periodontal endosseous defects and have nearly comparable effects. However, long-term, multicenter randomized, controlled clinical trials will be required to discern the definite clinical and radiographic effects of these graft materials and to arrive at an explicit conclusion.

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Introduction:-

Periodontitis, evoked by the bacterial biofilm (dental plaque) that forms around teeth, progressively destroys the periodontal tissue supporting the teeth, including the periodontal ligament, cementum, alveolar bone and gingiva (Darout et al, 2014). It is characterized by the presence of gingival inflammation, periodontal pocket formation, and loss of connective tissue attachment and alveolar bone around the affected teeth (Hanna et al, 2004). Several

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therapeutic modalities have been considered to arrest the disease progression and to regenerate the lost tissue. Periodontal surgical procedures have focused on the elimination of hard and soft tissue defects (i.e., probing depths and osseous defects) by regenerating new attachment (Froum et al, 1998). Reconstructive modalities that appear to have merit and have demonstrated significant gain of clinical attachment include: surgical debridement with adjunctive root surface or wound conditioning, implantation of bone, bone derivatives and substitutes, and placement of barrier membranes for guided tissue regeneration (Becker et al 1999).

One of the best approaches to achieve periodontal regeneration is the use of bone replacement graft techniques, which are based on the concept of filling the infrabony defects with a number of grafting materials, including autografts, xenografts and alloplastic materials (Kasaj et al 2008). The objectives of periodontal bone grafts are: probing depth reduction, clinical attachment gain, bone fill of the osseous defect and regeneration of new bone, cementum and periodontal ligament.

Alloplasts are synthetic bone substitutes that are readily available and also eliminate the need for a patient donor site. BIOGRAFT -CPC is an osteostimulative and osteoconductive bioactive device used for grafting osseous defects. BIOGRAFT - HABG ACTIVE (Bioactive composite granule) are bioactive glasses composed of SiO_2 , CaO , Na_2O , P_2O_5 and bond to bone through the development of a surface layer of carbonated hydroxyapatite. It is thought that the bioactive properties guide and promote osteogenesis, allowing rapid and quick formation of new bone.

A paucity of studies comparing the effects of Bioactive composite granule (BIOGRAFT-HABG ACTIVE) with Calcium phosphate cement (BIOGRAFT - CPC) was noted, thus, this study was designed to evaluate and compare, clinically and radiographically, the efficacy of bioactive composite granule versus calcium phosphate cement in the treatment of periodontal endosseous defects.

Aims And Objectives:-

1. To evaluate clinically, the efficacy of Bioactive composite granule (BIOGRAFT -HABG Active) and Calcium phosphate cement (BIOGRAFT -CPC) in the treatment of periodontal endosseous defects.
2. To evaluate radiographically, the efficacy of Bioactive composite granule (BIOGRAFT -HABG Active) and Calcium phosphate cement (BIOGRAFT -CPC) in the treatment of periodontal endosseous defects.
3. To compare clinically and radiographically, the efficacy of Bioactive composite granule (BIOGRAFT-HABG ACTIVE) and Calcium phosphate cement (BIOGRAFT -CPC) in the treatment of periodontal endosseous defects.

Materials and Methods:-

Subjects and study groups:-

A randomized, longitudinal interventional study involving a total of 20 systemically healthy subjects, contributing to a total of 20 surgical sites was designed and conducted on a study population selected from the subjects visiting the out-patient section of the Department of Periodontics, D A Pandu Memorial R V Dental College, Bangalore. The ethical clearance for the study was obtained from the ethical committee and review board of the institution.

Patients aged between 25-55 years, who were systemically healthy and had no contraindications for periodontal therapy met the inclusion criteria. A patient was not considered eligible if gingival index score was >2.1 .

Two and combined 3-wall intrabony periodontal defects with a probing pocket depth (PPD) ≥ 5 mm, radiographic defect depth ≥ 3 mm were included in the study.

The 20 surgical sites were identified and were randomly divided into 2 groups; Group I and Group II.

The groups were:-

Group I (n=10): Those to be treated with Bioactive composite granule (BIOGRAFT-HABG ACTIVE).

Group II (n=10): Those to be treated with Calcium phosphate cement (BIOGRAFT -CPC).

Clinical and Radiographic Assessments:-

Oral hygiene status was assessed using Plaque Index (Sillness and Loe (1964)) and Gingival index (Loe and Sillness (1963)). Probing pocket depth (PPD), clinical attachment level (CAL), and marginal recession (GR) were measured to the nearest millimeter with a calibrated periodontal probe using an occlusal stent as a reference point for probe

placement. Occlusal stents for positioning measuring probes were fabricated with cold-cured acrylic resin on a cast model obtained from an alginate impression. Measurements were recorded from:

- ☐ Stent to cemento-enamel junction
- ☐ Stent to gingival margin
- ☐ Stent to deepest probing depth at test sites

Surgical Procedure:-

After local anesthesia, an intrasulcular incision aiming to preserve the papillae was performed. Mucoperiosteal buccal and lingual access flaps were then reflected. Granulation tissue adherent to the alveolar bone was removed to provide full access and visibility to the root surfaces. Any subgingival calculus was removed gently by using hand instruments. Defects in group I received Bioactive composite granule (BIOGRAFT-HABG ACTIVE) and defects in group II patients received Calcium phosphate cement (BIOGRAFT -CPC). Finally, the flaps were replaced and sutured appropriately with a 3-0 silk material using interdental suture technique. After a healing period of 10 days, the sutures were removed.

Postoperative Care:-

All patients received systemic antibiotic therapy for a period of 5 days postoperatively (amoxicillin 500 mg three times per day for 5 days). In addition, all patients were advised to avoid tooth brushing and hard chewing in the surgical areas and to rinse twice daily with a 0.2% solution of chlorhexidinedigluconate for 2 weeks. Recall appointments were scheduled every second week during the first 2 months after the surgical procedure, and all patients were recalled once a month for the remaining observation period.

Post-Surgical Evaluation and Review:-

Gingival Index (GI) and Plaque Index (PI) were re-evaluated at 3 months, 6 months and 9 months. Probing Pocket Depth (PPD), Clinical Attachment Level (CAL), Gingival Recession (GR) were also re-evaluated at 3 months, 6 months and 9 months using the previously used acrylic stents to provide a reproducible insertion axis. Depth of the defect was re-assessed at 3 months, 6 months and 9 months using digital radiography.

Results:-

Statistical test used included Kruskal Wallis test and Student unpaired t-test.

Clinical evaluation of post-surgical healing revealed a good soft tissue response to the combinations with no adverse complications. Both groups presented similar baseline characteristics in terms of PPD, GR, CAL, plaque index, gingival index.

All patients maintained a good level of oral hygiene and gingival status throughout the recall periods. Intergroup differences were found to be insignificant ($P > 0.05$) in terms of plaque index and gingival index (Table I and Table II).

At 9 months, all the groups presented a significant improvement in terms of PPD reduction and CAL gain (Table III and Table IV). The intergroup differences were found to be significant (Table III and Table IV). Gingival recession levels had also improved, however, the difference was not statistically significant (Table V).

Evaluation of the hard tissue findings indicated that all treatment modalities resulted in bone gain at 9 months in both groups (Table VI).

Discussion:-

The results of the present study show that treatment of intrabony defects with Bioactive composite granule (BIOGRAFT-HABG ACTIVE) / Calcium phosphate cement (BIOGRAFT -CPC) leads to significant PPD reduction, attachment, and radiographic bone gain compared to baseline values. Statistically significant differences in all of the investigated parameters were found between the treatments. Being alloplastic in nature, these graft materials do not increase the patient morbidity and do not require a second surgical site as in the case of autografts.

Approximately 60% of the bone graft substitutes currently available involve ceramics, either alone or in combination with another material. These include calcium sulphate, bioactive glass, and calcium phosphate. The use of ceramics, especially calcium phosphates, is driven in part because of the fact that the primary inorganic

component of bone is calcium hydroxyapatite, a subset of the calcium phosphate group. In addition, calcium phosphates are osteoconductive, osteointegrative (the newly formed mineralized tissue forms intimate bonds with the implant material), and, in some cases, osteoinductive. They often require high temperatures for scaffold formation and have brittle properties; therefore, they are frequently combined with other materials to form a composite. They may be porous or non-porous. Calcium phosphate compounds, currently most widely used in periodontal surgery are hydroxyapatite (HA) and tricalcium phosphate (TCP) (Pandit et al, 2010).

Bioactive glasses are composed of bone silicon dioxide (46 mole %), sodium oxide (24.4 mole %), calcium oxide (26 mole %), and phosphorous pentoxide (6 mole %) and bond to bone through the development of a surface layer of carbonated hydroxyapatite. It is thought that the bioactive properties guide and promote osteogenesis, allowing rapid and quick formation of new bone. Fetner et al in 1994 suggested that these materials have properties of superior manageability, hemostatic effects, and osteoconductive properties (Fetner 1994).

In a study by Lindhe et al, it was shown that, surgical procedures would induce loss of attachment if done in pockets shallower than 4.2mm (Lindhe et al, 1982). Hence, probing depth greater than 5mm was considered for the study. Laurell et al, in his study, has shown that, to benefit from regenerative procedures, depth of defect should be at least 3-4 mm (Laurell et al, 1998).

Probing Pocket Depth (PPD), Clinical Attachment Level (CAL) and Gingival Recession (GR) were assessed using a UNC 15 probe positioned along the grooves on a customized acrylic stent which was fabricated for each patient for providing a reproducible insertion axis for the probe. Similar technique has been adopted in other studies (Sharma, 2011; Subbaiah 2011, Sollazzo et al 2010).

Preoperative and postoperative comparability of probing measurements that do not use this standardized method may be open to question (Carranza, 2006). The depth of the angular bone loss was assessed to the closest 0.5 mm on the intraoral periapical radiograph taken using the paralleling cone technique with the radiographic grid in position which allowed for standardization. Other studies have also used this technique for assessment of bone defect and bone fill (Subbaiah, 2011; Cardaropoli, 2002).

The results of the study showed a statistically significant decrease in the plaque index and gingival index from baseline to 3 months, baseline to 6 months and at the end of 9 months in Group I as well as in Group II which is in accordance with the study conducted by Stein et al who used biphasic calcium composite grafting material in the treatment of human periodontal intrabony defects. However, no statistically significant difference was recorded between the two groups suggesting that all patients were very well motivated and there was a good maintenance of oral hygiene throughout the study in both the groups.

There was a statistically significant reduction in mean values of pocket depth and clinical attachment level at three months and six months in the two groups. These findings are in agreement with the results of Froum and Gupta (Froum et al 1998).

The mean amount of defect resolution from the baseline to three months and six months in all the groups was statistically significant. The mean percentage of defect resolution at three months and six months in the two groups was statistically significant. These results were in accordance with the study conducted by Meffert and Pepelassi.

Group I

Figure 1



Figure 2

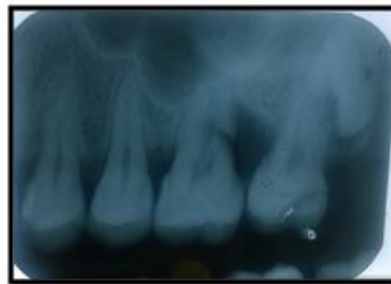


Figure 3



Figure 4



Figure 5



Figure 6



Figure 1& 2:- Pre-Operative Clinical And Radiographic Examination

Figure 3 & 4:- Intraoperative View Of Defect, Graft Placement (Biograft Habg)

Figure 5:- Post- Suturing

Figure 6:- Periodontal Dressing Placed

Figure 7



Figure 8



Figure 7 & 8:- Clinical and radiographic evaluation at 9 months.

Group II

Figure 1



Figure 2



Figure 3



Figure 4

Figure 5



Table I:- Comparison Of Mean PI Scores Between Two Study Groups At Different Time Intervals

Time	Group	N	Mean	SD	Mean Diff	t	P-value
Baseline	BCG Group	10	1.56	0.14	0.06	0.896	0.38
	CPC Group	10	1.50	0.16			
3 Months	BCG Group	10	1.32	0.12	0.00	0.001	1.00
	CPC Group	10	1.32	0.14			
6 Months	BCG Group	10	1.19	0.09	-0.01	-0.287	0.78
	CPC Group	10	1.20	0.07			
9 Months	BCG Group	10	1.03	0.02	0.00	-0.29	0.77
	CPC Group	10	1.03	0.02			

denotes significant difference (P value <0.05)

Table II:- Comparison Of Mean GI Scores Between Two Study Groups At Different Time Intervals.

Time	Group	N	Mean	SD	Mean Diff	t	P-value
Baseline	BCG Group	10	1.77	0.36	0.00	0.001	1.00
	CPC Group	10	1.77	0.43			
3 Months	BCG Group	10	1.30	0.11	-0.02	-0.314	0.76
	CPC Group	10	1.32	0.11			
6 Months	BCG Group	10	1.16	0.07	-0.02	-0.570	0.58
	CPC Group	10	1.18	0.08			
9 Months	BCG Group	10	0.83	0.21	0.00	-0.010	0.99
	CPC Group	10	0.83	0.21			

*denotes significant difference (P value <0.05)

Table III:- Comparison Of Mean PPD Scores Between Two Study Groups At Different Time Intervals

Time	Group	N	Mean	SD	Mean Diff	t	P-value
Baseline	BCG Group	10	9.22	1.00	0.36	0.891	0.38
	CPC Group	10	8.86	0.79			
3 Months	BCG Group	10	5.95	0.59	0.08	0.323	0.75
	CPC Group	10	5.87	0.51			
6 Months	BCG Group	10	4.97	0.45	0.05	0.270	0.79
	CPC Group	10	4.92	0.37			
9 Months	BCG Group	10	4.26	0.30	0.12	1.134	0.27
	CPC Group	10	4.14	0.14			

*denotes significant difference (P value <0.05)

Table IV:- Comparison Of Mean RAL Scores Between Study Groups At Different Time Intervals Using Student Unpaired 't' Test

Time	Group	N	Mean	SD	Mean Diff	t	P-value
Baseline	BCG Group	10	10.12	0.54	0.04	0.155	0.88
	CPC Group	10	10.08	0.61			
3 Months	BCG Group	10	7.07	0.42	0.14	0.607	0.55
	CPC Group	10	6.93	0.60			
6 Months	BCG Group	10	5.39	0.41	0.11	0.567	0.58
	CPC Group	10	5.28	0.46			
9 Months	BCG Group	10	4.37	0.28	0.05	0.470	0.64
	CPC Group	10	4.32	0.19			

*denotes significant difference (P value <0.05)

Table V:- Comparison Of Mean GR Scores Between Two Study Groups At Different Time Intervals.

Time	Group	N	Mean	SD	Mean Diff	t	P-value
Baseline	BCG Group	10	1.67	0.13	0.03	0.485	0.63
	CPC Group	10	1.64	0.14			
3 Months	BCG Group	10	2.05	0.16	-0.06	-0.845	0.41
	CPC Group	10	2.11	0.16			
6 Months	BCG Group	10	1.81	0.13	-0.01	-0.172	0.87
	CPC Group	10	1.82	0.13			
9 Months	BCG Group	10	1.70	0.14	-0.10	-1.732	0.10
	CPC Group	10	1.80	0.12			

*denotes significant difference (P value <0.05).

Table VI:- Comparison Of Mean Depth Of The Defect As Measured In IOPAR Within Subjects At Different Time Intervals In Two Study Groups.

Study Groups	Time	N	Mean	SD	Greenhouse-Geisser		Diff	P-Value
					F	P-Value		
BCG Group	BL	10	11.35	0.93	556.642	<0.001*	I Vs II, III, IV	<0.001*
	3 Months	10	8.07	0.73			II Vs III	<0.001*
	6 Months	10	6.07	0.41			II Vs IV	<0.001*
	9 Months	10	4.19	0.39			III Vs IV	<0.001*
CPC Group	BL	10	10.94	1.15	375.542	<0.001*	I Vs II, III, IV	<0.001*
	3 Months	10	7.15	1.16			II Vs III	<0.001*
	6 Months	10	5.40	0.67			II Vs IV	<0.001*
	9 Months	10	4.14	0.54			III Vs IV	<0.001*

*denotes significant difference (P value <0.05).

Conclusion:-

Clinical and radiographic assessment reveal that treatment with bioactive composite granule and calcium phosphate cement both are efficacious in the treatment of periodontal endosseous defects. These two appear to have nearly comparable effects, treatment with both bioactive composite granule and calcium phosphate cement improve the healing outcomes regarding probing depth reduction, CAL gain, and bone fill. However, long-term, randomized, controlled clinical trial and histomorphometric studies employing a greater number of patients will be needed to arrive at a definitive conclusion.

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RESEARCH ARTICLE

AN ANALYTICAL STUDY ON THE RESULTS OF POSTERIOR FOSSA DECOMPRESSION AND LAX DUROPLASTY IN CHIARI 1 MALFORMATIONS.

Dr. R Saravana santosh kumar, Dr. M M Sankar and Dr. S Rajkumar.

Manuscript Info

Abstract

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Introduction:-

Chiari 1 malformation has been approximately diagnosed in 1% to 4% of patients undergoing brain or cervical spine magnetic resonance imaging (MRI) studies,^{1,2}. Chiari malformation type 1 (CM-1) is a common and often debilitating neurosurgical condition. In recent years, there has been a growing emphasis on outcomes research, including efforts to compare the results of various surgical approaches.^{3,4} To help facilitate more objective comparisons, several groups have produced and/or validated different metrics to evaluate treatment outcomes.⁵⁻⁸ However, despite this growing interest in improved research methodology, evidence-based guidelines for surgical intervention are undefined. The decision to treat CM-1 surgically may be based on a variety of considerations, such as anticipated syrinx improvement and stabilization of spinal cord damage, as well as the risk of postoperative complications. The objective of this study was to use clinical and neuroimaging features to predict long-term patient-reported Quality of life outcomes in patients undergoing posterior fossa decompressions.

Methods:-

Patient population:-

This study is an analysis on the 28 patients evaluated and managed in our department of Neurosurgery in Govt Stanley medical college hospital, Chennai for Chiari 1 malformations between 2011-2016. The study was proceeded following the approval from the ethical committee from our institution and consent from the patients. All patients had a detailed neurological examination followed by MRI scans of the brain with whole spine screening to evaluate the clinical presentations. Chiari 1 malformation patients had been graded based on the chiari severity index and underwent posterior fossa decompression with lax duroplasty.

Data collection:-

Preoperative signs and symptoms were recorded on admission. These findings were subsequently grouped into 3 categories 1) classic Chiari headaches (including occipital, Valsalva-induced, posttussive, and exertional headaches), 2) atypical presentation (frontotemporal headaches; and poorly localized headaches) 3) myelopathic predominant. We defined myelopathic symptoms as numbness, weakness, hyperreflexia, or unsteady gait.

Radiological analysis with MRI craniovertebral junction was used mainly to analyse the extent of descent of cerebellar tonsils and the size of the syrinx. In addition to these, we recorded the following neuroimaging parameters potentially related to CM-1 pathology: (1) tonsillar ectopia, as the perpendicular distance from the tip of the cerebellar tonsil to McRae's line^{9,10}; (2) clivus-canal angle, as the angle between

Wackenheim's clivus baseline and the posterior C2 vertebral body line¹⁸; (3) dens angle, as the angle formed by a line through the C2 synchondrosis and a line through the odontoid tip measured from the posterior aspect of the C2 vertebral body¹¹; (4) pB-C2, as the perpendicular distance between the ventral dura and a line joining the basion to the inferoposterior C2 body^{17,19,20}; (5) obex position, as the distance between the obex and foramen magnum (basion-opisthion line)¹²; and (6) basilar invagination, as present when the tip of the dens was 5 mm above the Chamberlain's line.¹¹ We defined a syrinx as a contiguous spinal cord cavitation measuring at least 3 mm in maximum diameter on T2-weighted MRI.

Outcome assessment:-

We evaluated patient-defined outcome as improved, stabilised and worsened on the basis of postoperative follow up during the reviews. Any improvement from the presenting complaints was considered as IMPROVED. Stabilised was defined as non-progression of complaints following the procedure. Any worsening of symptoms was to be taken as WORSENERD.

Results and Discussion:-

Out of the total of 28 patients undergoing posterior fossa decompression for Chiari 1 malformation, the mean age of presentation was 24.5 years. 13 patients were male and 15 were female. Headache was the presenting complaint in all the 28 patients with classical chiari type of headache in 24 and atypical presentation like generalised or fronto temporal headache in 4 patients. Features suggestive of myelopathy were seen in 14 patients. Scoliosis was present in 4 patients and Charcot's joint in 4 patients (as shown in table 1). Radiological findings found were syringomyelia in 14 patients, interhemispheric cyst in 2 patients, corpus callosal agenesis in 4 patients, block vertebra involving C2C3 in 1 patient and hydrocephalus in 3 patients (as shown in table 2).

Based on these presentations, clinical grading was done as grade 1 – typical presentation, grade 2 – atypical presentation, grade 3 – predominance of myelopathic features. Table 3 shows the distribution of patients based on the clinical grading. Radiological grading was done on the basis of syrinx. If the diameter of the syrinx was greater than 6mm, it was grade B and if it was less than 6mm or absent, it was considered as grade A as shown in table 4.

Patients were graded in to 3 based on chiari severity index. Grade 1 as clinical grade 1, Grade 2 as clinical grade 2/3 + radiological grade A, Grade 3 as clinical grade 2/3 + radiological grade B. There were 14 patients in grade 1 of chiari severity index, 4 patients in grade 2 of chiari severity index and 10 patients in grade 3 of chiari severity index. All the patients underwent posterior fossa decompression with removal of posterior arch of C1 with lax duroplasty. The 3 patients with hydrocephalus underwent ventriculoperitoneal shunt procedure followed by PFD. Postoperatively, csf leak was observed in 1 patient, aseptic meningitis in 1 patient, pseudomeningocele in 3 patients and hydrocephalus in 1 patient. Postoperative period was uneventful in 22 patients.

Postoperative outcome categorised as Improved (18 patients 64.2%), Stabilised (9 patients 32.1%) and worsened (1 patient 0.35%) as shown in table 5.

Conclusion:-

Posterior fossa decompression with lax duroplasty despite its complexity of the procedure is safe and effective way of reducing the symptomatology of chiari malformations with a lesser complication rate.

Table 1:-showing the distribution of symptoms.

Symptoms	No Of Patients	Percentage
Headache	32	100 %
Occipital	26	
Frontotemporal headache	6	
Dissociated/ suspended sensory loss	14	44 %
Spasticity of lower limbs	14	44 %
Small muscle wasting of hands	12	38 %
Bladder / bowel disturbances	—	—
Trigeminal neuralgia	1	
Scoliosis	4	12.5 %
Charcot joint	4	12.5 %

Table 2:-showing the MRI findings

Mri Findings	No Of Patients
Syringomyelia	17
Interhemispheric cyst	2
Corpus callosal agenesis	4
Block c2- c3 body	1
Hydrocephalus	3

Table 3:- showing the distribution of patients on clinical grading.

Clinical grade	Number of patients
1 (typical presentation)	12
2 (atypical presentation)	2
3 (myelopathy)	14

Table 4:-showing radiological grading based on syrinx

Radiological grade	Number of patients
A (No syrinx or syrinx <6mm)	18
B (Syrinx > 6mm)	10

Table 5:-showing postoperative outcome following PFD

C.S.I	Number of patients	Postoperative outcome		
		Improved	Stabilised	worsened
1	14	12	2	-
2	4	-	3	1
3	10	6	4	-
Total	28	18 (64.2%)	9 (34.1%)	1

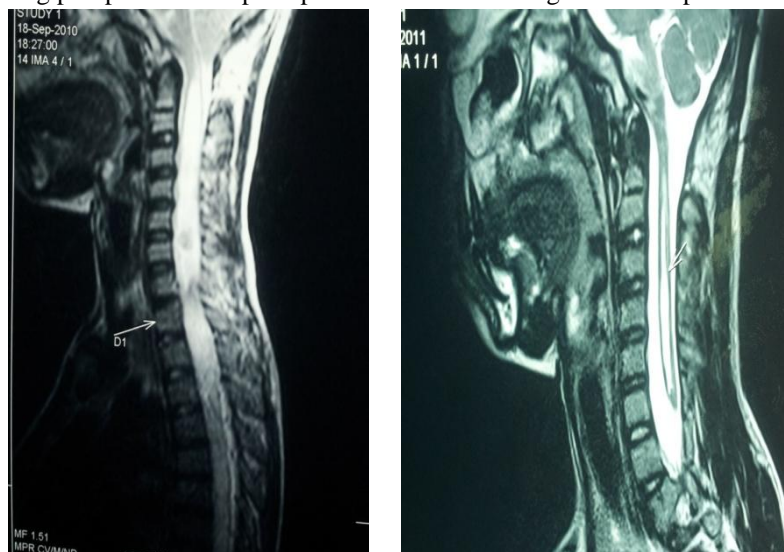
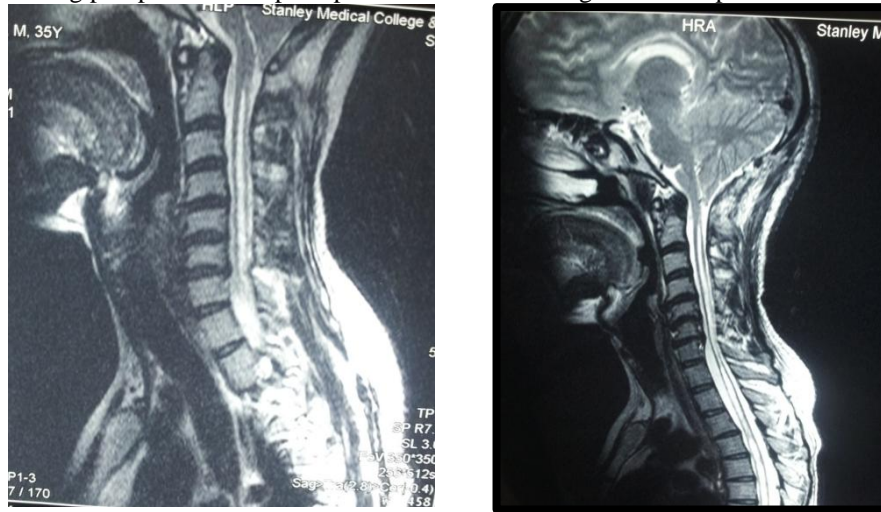
Figure 1:-showing preoperative and postoperative scans following successful posterior fossa decompression

Figure 2:-showing preoperative and postoperative scans following successful posterior fossa decompression



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RESEARCH ARTICLE

NOISE POLLUTION STUDY ATMHASWADYATRAMAHOTSAV, 2016 MAHARASHTRA, INDIA.

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MhaswadYatraMahotsav, crackers, noise pollution, monitoring Sites.

Abstract

The unwanted or disturbing sound is called as noise. This reported study shows noise quality of firing the crackers at “MhaswadYatraMahotsav- 2016”, which held every year at Mhaswad, Dist. Satara in Maharashtra. The noise level due to bursting of crackers is measured by using standard method. For present study ten different sites were selected as noise monitoring sites on “Palakhi Marg” and noise of crackers was noted at ten different timings with the interval of half an hour in each recording. The result concluded that, in the residential zone and silence zone at night condition, the noise level exceeded the prescribed standard level of 65 dB as per the Noise Pollution (Regulation and Control) Rules, 2000.

In the present study, the average noise level at all the recorded timings was found to be above the prescribed limits of Central Pollution Control Board (CPCB).

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Introduction:-

Noise pollution is a major concern throughout the world. Noise pollution in different areas is coming from wide variety of sources. Noise is an unpleasant and unwanted sound which is indeed annoying and harmful too. Its effects are more directly on the receiver i. e. Man. It is said that “you may forgive noise but your body will never” (Agrawal and Yadav, 2013). Noise pollution damagingly affects the human being leading to loss of hearing as well as mental stress (Jhanwar, 2016). Although noise pollution is not fatal to human being but its effects cannot be overlooked. Noise causes stress reactions, increased blood pressure, sleep disorders, some hormonal changes and general quality of life (Bhat, 2003). The effect of noise has been studied in humans by different workers (Belojevic *et al*, 2008; Babisch, 2000 and 2005; Lundberg, 1999). At present noise pollution is considered to be one of the key problems that has numerous hazardous effects on environment which deals with cost of society (Martin *et al*, 2006).

Indian festivals are traditionally celebrated by song and dance in large groups. During festival loudspeakers, bursting of crackers produce intermittent and high frequency impulse noise (Concha-Barrientos *et al*, 2004). Diwali is an important festival when lot of crackers are used in almost every part of the country (Singh and Joshi, 2010). Mhaswad is famine and low rainy area which is traditionally known for Lord Siddhnanthyatra. This yatra festival is celebrated for one month starting from ‘laxmipoojan’ of Diwali and ends in kartik month of Marathi year. This yatra is a celebration of marriage ceremony of God Siddhnanath and Goddess Jogeshwari. Traditionally there is a faith that when God and Goddess pass in their “palakhi”, citizens are trying to stop that “palakhi” in front of their houses. To stop the “palakhi” peoples are using to burst the fire crackers and thus there is a competition in the peoples for bursting crackers. Traditionally it is called as “Diwali Maidan”.

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In the present investigation, an attempt has been made to monitor the noise created by bursting of crackers in “Diwali Maidan”.

Working Area:-

The present work has been carried out at the “Palakhi Marg” at Mhaswad City, Tal. Man, Dist. Satara. This “Palakhi Marg” begins from Shri Siddhanth Temple and passes from main market area and ends back to Shri Siddhanth temple.

Material and Method:-

The present study was conducted at ten different timings during the “Palakhi Marg” in “Diwali Maidan”. The ambient noise level was monitored with the help of sound level meter during 8 pm to 12.30pm. Precaution was taken to avoid echo or resonance of sound by selecting suitable distance from the crackers. Readings were taken after every half an hour interval. Ambient sound levels were compared with that of the standards prescribed in the Environment Act, 1981 and standards of CPCB (Tripathi, 1999). As well as the noise limits given by, The Noise Pollution (Regulation and Control) Rules, 2000 were used to compare the noise levels in areas under study.

Table 1:- Noise standards for ambient noise level

Sr. No.	Area Code	Category of Area/ Zone	Sound Level Limit at Day Time (dB)	Sound Level Limit at Night Time (dB)
1)	A	Industrial Area	75	70
2)	B	Commercial Area	65	55
3)	C	Residential Area	55	45
4)	D	Silence Zone	50	40

Source: Lad *et al* (2012)

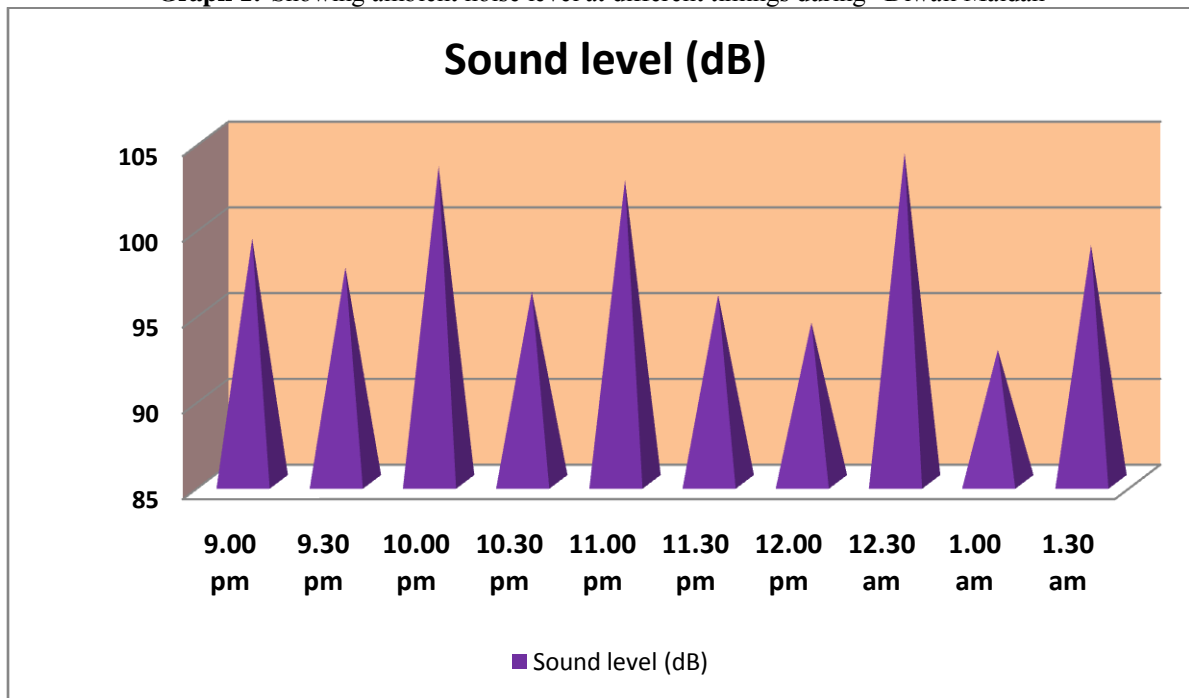
Result:-

The results of ambient noise pollution monitored in the YatraMahotsav at Mhaswad have been summarized in table 2.

The ambient noise level noted at all the monitored sites was very high as per given by the CPCB. The “Palakhi Marg” is having silence zone, which is also showing ambient noise level more. The highest ambient noise level recorded was 104.2 at 12.30 am which is very high than the recommended noise level at night.

Table 2:-Noise level in different zones of “Palakhi Marg” at Mhaswad city in “Diwali Maidan”.

Zone	Names of the sites	Time	Sound level (dB)
Residential	Mhaswad Main Peth	9.00 pm	99.2
		9.30 pm	97.5
		10.00 pm	103.4
Silence	Hospital and Bank	10.30 pm	96.1
		11.00 pm	102.6
		11.30 pm	95.9
Residential	BajarPeth	12.00 pm	94.3
		12.30 am	104.2
		1.00 am	92.7
		1.30 am	98.8

Graph 1:-Showing ambient noise level at different timings during “Diwali Maidan”**Discussion:-**

In the present study, the average noise level at all recorded timing was 98.47 dB which is found to be above the prescribed limits of CPCB (Central Pollution Control Board, New Delhi) during the “Yatra Mahotsav” day at 2016. This average recording of ambient noise level at residential and silence zone during night conditions is very high. In the beginning of “Palakhi” noise level is above the average which is slightly declined in the silence zone but again inclined at residential zone at last phase.

According to Mishra *et al* (2017), the ambient noise level increased due to different human activities during Bhopal VigyanMela 2016. The study report of Patel and Bhawe (2014), shows increased noise level due to bursting of crackers during diwali festival. The noise level during festive and non-festive days in Haridwar city were measured and compared by Sharma and Joshi (2010) which is found to be very high during Diwali season. At different parts of Meerut city on the night of Diwali festive, noise pollution was studied by Singh and Joshi (2010), which shows extreme ambient noise level during Diwali. A study report of Pathak *et al* (2008) showed traffic noise became main reason of headache, high BP and other stresses among the peoples in the working places at Varanasi city. In Bareilly metropolitan city, the noise level was slightly higher than the prescribed limit of the Central Pollution Control Board (Gangware *et al.*, 2006).

Conclusion:-

The present study is an attempt to assess the level of ambient noise pollution during the particular day of “Diwali maidan”. Peoples in the residential area were suffered from this high levelled noise pollution. Noise pollution affects and damages the human being leading to loss of hearing as well as mental stress. Efforts are made through this article to make the citizens aware about the preliminaries of noise pollution and its hazardous effects.

Recommendations:-

- Proper supervision should be followed to make sure that cracker seller in and around Mhaswad, following the noise standards set for crackers.
- There should be ban on noisy crackers during “Yatra Mahotsav” in and around Mhaswad.
- A separate area to be made available to the peoples for firing of crackers by the municipal corporation.
- The time limit should be given for bursting of the crackers which should be strictly followed by the citizens.

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RESEARCH ARTICLE

EVALUATION OF GRADIENT GEOMETRY FOR MULTIMODAL MEDICAL IMAGE FUSION

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Medical image fusion, geometry, SSIM, PSNR.

Abstract

Fusion of medical images such as MRI and PET by gradient selection is attempted in this paper. The efficiency of the fusion process depends on the selection of the appropriate image geometry. This work analyses different gradient geometries for medical image fusion. From the experimental analysis, it is found that the images fused by the Southwell geometry produced better results when compared to Fried geometry and Hudgin geometry.

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Introduction:-

Image fusion integrates information from multiple individual images into a single image so that the obtained image contains more details of the source images. In static image fusion, it is necessary that the input images are aligned and there exists coincidence in terms of depth or field of view of the imaged scenes. Image registration is done to make the images suitable for static image fusion [1]. Image fusion algorithm can be classified into pixel-based and regional-based algorithm [2]. In pixel-based methods, each pixel of the input images are combined using various rules such as averaging, finding maximum to form corresponding pixels in the fused image [3]. In region-based techniques, wavelet transforms are used to represent the input images in a multi-resolution framework, and then each region of the image is applied a different fusion rules that are used to desegregate the information into a more comprehensive image [4].

Medical image fusion [5] aids in the clinical diagnosis and treatment by providing useful information especially for lesion location, making treatment and pathological study. MRI image gives information about anatomical structure of soft tissues such as brain, organs and blood vessels [6,7], while PET image suffers from low resolution but provides physiological information [8]. Hence fusion of these two images offers the advantage of better diagnosis of diseases [9] particularly of the cancer tumours.

Socolinsky and Wolff proposed an image fusion approach in which the information from a multi-spectral image dataset is integrated to produce a one band display of the image. They generalize image gradients are those which closely related to image contrast [10], by defining it for multi-spectral images in terms of differential geometry. The desirable gradient field is reconstructed using the contrast information, to produce the fused image. Later, Wang *et al* fused the images in gradient domain using weights dependent on local variations in intensity of the input images [11]. At each pixel position, they construct an importance-weighted contrast matrix.

The algorithm proposed by Sujoy Paul *et al* combines gradient fusion as well as pixel-based fusion [12]. It works for the fusion of colour or grayscale image for multi-focal, multi resolution [13] and multi-exposure [14] images. In the

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case of colour images, the luminance and chrominance of the images are fused in a different manner. By doing so the computational time for running the algorithm is greatly reduced. Also, due to the fact that the luminance (Y) channel represents the image brightness and contrast information and it is in this channel where variations and details are most visible, since the human visual system is more sensitive to luminance (Y). The chrominance (Cb,Cr) [15] channels contain only colour information, to which the human visual system is less sensitive. Comparison between Hudgin, Fried, Southwell geometries proved the advantage of each for noise reduction. Roopashree *et al* reported that the fried geometry produced worst performance, as its consistency is poor [16].

In this paper, the fusion of the medical image using the gradient selection is proposed. The quality of the image is analyzed by using different geometry. The performance assessment test such as Structural Similarity Index (SSIM), and Peak Signal to Noise Ratio (PSNR) are used for testing the accuracy of the fusion process.

Materials and Methods:-

The static image fusion requires input multi-modal images of same size and in the same depth of view. Hence input image must be registered before processing [17]. Here, pre-registered MRI and PET images are taken for further processing.

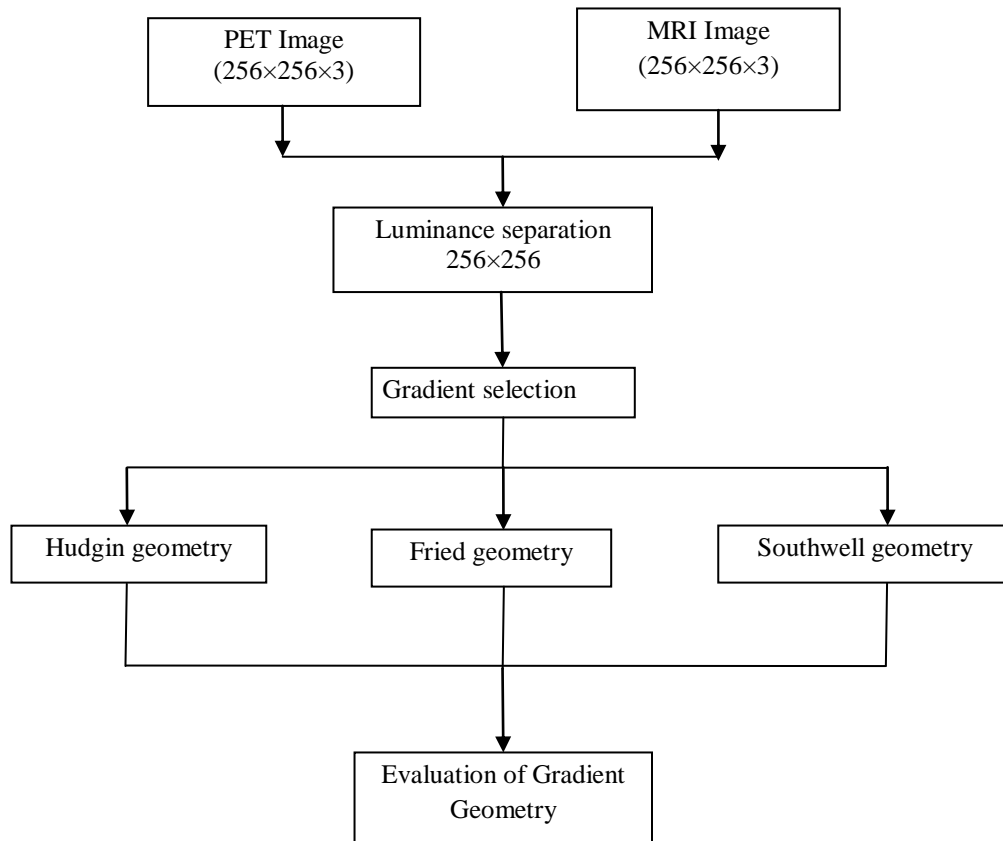


Fig. 1:- Steps involved in the evaluation of gradient geometry for medical image fusion

The input images are taken from www.med.harvard.edu. The Fig. 1 explains the gradient selection process. Initially, luminance channel is separated from each of the images. From that the gradients are selected using various geometries such as Hudgin geometry, Fried geometry, Southwell geometry. After fusion of images using any reconstruction [18] algorithm, the parameters PSNR and SSIM are calculated for each fused image. Finally, the performance of the fused image is compared and analyzed.

Fusion technique proposed by Sujoy paul *et al* for colour image the luminance part is separated from the chrominance part (RGB to YCbCr) as different fusion technique is followed for each. Gradient fusion is carried out

for luminance channel. This involves taking the gradients with the maximal magnitude at each pixel position will lead to an image which has much more detail than any other image in the stack. Luminance channel for N input images are stored in a stack as $I'=\{I_1, I_2, \dots, I_N\}$ where $N > 2$. The gradient value for each image is found for each pixel position using Hudgin geometry. Those values are taken mean with the neighbouring pixels and are made equivalent for the gradient for the respective pixels which is the Southwell geometry explained later in this paper. The gradient of the luminance channel of an image may be $\Phi_n^x(x, y)$ and $\Phi_n^y(x, y)$ where Φ_n^x and Φ_n^y are the gradient components along the x and y directions respectively.

The magnitude of the gradient may be

$$H_n(x, y) = \sqrt{((\Phi_n^x(x, y))^2 + (\Phi_n^y(x, y))^2)} \quad (1)$$

The magnitude of gradient at all the pixel positions of each image is compared with that of the other images and the maximum value of that is stored as

$$P(x, y) = \max_{1 \leq n \leq N} H_n(x, y) \quad (2)$$

The fused luminance gradient is

$$\Phi^x(x, y) = \Phi_{p(x, y)}^x(x, y) \quad (3)$$

$$\Phi^y(x, y) = \Phi_{p(x, y)}^y(x, y) \quad (4)$$

where $\Phi^x(x, y)$ and $\Phi^y(x, y)$ are the values of gradient components in the x and y gradient components of the image with index $P(x, y)$ at pixel location (x, y) and the fused luminance gradient is $\Phi = [\Phi_x \Phi_y]^T$. The relationship between the fused gradient (Φ) and the luminance channel (I) is $\nabla I = \Phi$, where $\nabla = [d/dx, d/dy]^T$.

As the fused gradient is a combination of multiple luminance channel, there are possibilities for high peaks in the adjacent pixels. Hence it needs to be optimized using poisson solver which satisfies the zero curl condition to remove artifacts. The reconstruction of the luminance channel from the gradient data is done with a wavelet based algorithm proposed by Haar-Hampton et al followed by image enhancement using gamma correction. The fusion procedure for grayscale images is the same as followed for the luminance channel.

For chrominance part, the individual chrominance channels (C_b, C_r) are taken and the fusion is carried out by taking the weighted sum, with the weights depending on the channel intensities of input chrominance channels. Their values will range from 16 to 240. For grayscale images this process does not apply as they have values of both C_b and C_r equal to 128. The weights must be selected for pixel positions such that how far the chrominance value is from 128. Computation is less intensive than gradient fusion.

Concept of Geometries:-

Local gradients are measured at discrete locations of the wavefront. Depending on the position at which phase differences are estimated with reference to the location of slope measurements, there exist three main classifications in wavefront sampling namely Fried, Hudgin and Southwell (or Shack Hartmann) geometries. The illustration of relationship between the slope and phase measurements for the above geometries are shown in fig(1).

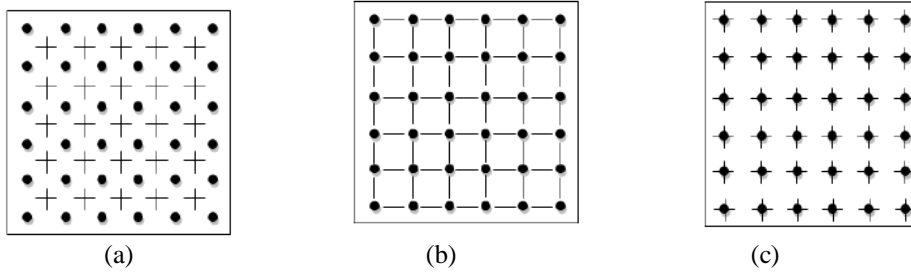


Fig.2:- Sampling Geometry (a) Fried (b) Hudgin (c) Southwell

The horizontal and vertical lines represent positions of slope measurements in x and y directions respectively and dots represent positions of phase estimation. The difference between the geometries are shown in Fig.2. In Fried and Southwell configurations both x and y slope measurements are made at the same point. In the Southwell (Shack Hartmann) configuration, slope measurement points coincide with the phase evaluation points whereas in Fried configuration, phase evaluation grid is displaced by half the sensor pitch with respect to slope measurement grid. In Hudgin geometry, x and y slope measurement points are displaced from one another and phase is evaluated at the edges of the slopes [19]. The slope to phase relation for different configurations are as follows

For Fried geometry,

$$\Phi_n^x(x, y) = \frac{\left[\frac{\Phi_{(x+1,y)} + \Phi_{(x+1,y+1)}}{2} - \frac{\Phi_{(x,y)} + \Phi_{(x,y+1)}}{2} \right]}{h} \quad (5)$$

$$\Phi_n^y(x, y) = \frac{\left[\frac{\Phi_{(x,y+1)} + \Phi_{(x+1,y+1)}}{2} - \frac{\Phi_{(x,y)} + \Phi_{(x+1,y)}}{2} \right]}{h} \quad (6)$$

Where $x = 1, 2, \dots, N-1$ and $y = 1, 2, \dots, N-1$.

For Hudgin geometry,

$$\Phi_n^x(x, y) = \frac{\Phi_{(x+1,y)} + \Phi_{(x,y)}}{h} \quad (7)$$

$$\Phi_n^y(x, y) = \frac{\Phi_{(x,y+1)} + \Phi_{(x,y)}}{h} \quad (8)$$

Where $x = 1, 2, \dots, N-1$ and $y = 1, 2, \dots, N$.

For Southwell geometry,

$$\frac{\Phi_n^x(x+1, y) + \Phi_n^x(x, y)}{2} = \frac{\Phi_{(x+1,y)} + \Phi_{(x,y)}}{h} \quad (9)$$

$$\frac{\Phi_n^y(x, y+1) + \Phi_n^y(x, y)}{2} = \frac{\Phi_{(x,y+1)} + \Phi_{(x,y)}}{h} \quad (10)$$

Where $x = 1, 2, \dots, N$ and $y = 1, 2, \dots, N-1$.

Any of the above geometries can be applied to find the gradient values from the input images each producing different quality of fused image when the gradient data is used for reconstruction.

Image Quality Assessment Tests:-

(i) PSNR:

The Peak Signal to Noise Ratio (PSNR) can be computed by calculating MSE (Mean Square Error).

It can be calculated by squaring the intensities of original input images and the final output image[20]. Then the average is taken. It is given by

$$MSE = \left(\frac{1}{NM}\right) \sum_{m=0}^{M-1} \sum_{n=0}^{N-1} e(m,n)^2 \quad (11)$$

Where $e(m,n)$ is the error difference between original and distorted images. PSNR is obtained as the pixel difference between the reconstructed and the original image. It is given by

$$PSNR = 10 \log \frac{S^2}{MSE} \quad (12)$$

where $S=255$ for images having 8-bit.

(ii)SSIM:

The Structural SIMilarity Index (SSIM) is a method for measuring similarity between two images. The SSIM index can be viewed as a quality measure in which the fused image is compared with the image that is considered as perfect quality [21]. Its formula is given as

$$SSIM(x,y) = \frac{(2\mu_x\mu_y + C_1)(2\sigma_{xy} + C_2)}{(\mu_x^2 + \mu_y^2 + C_1)(\sigma_x^2 + \sigma_y^2 + C_2)} \quad (13)$$

Results and Discussion:-

The fusion of medical images (MRI and PET) based on gradient approach is presented in this paper. The gradient approach is done only to Luminance channel. The input gradients are taken using three different images such as Hudgin geometry, Fried geometry, Southwell geometry. Among these three methods Southwell geometry provides the best PSNR and SSIM values. The comparison among the geometries for five different cases are given below.

In Table 1, the normal brain images from MRI and PET are taken and fused using three different geometries. From the fused image of Fried geometry it is shown that the local features of the input images are not obtained clearly. The fused images of Hudgin geometry and Southwell geometry seem to be similar, but the parameters PSNR and SSIM is high for Southwell geometry compared to Hudgin geometry.

In Table 2, the PSNR value of fused image of Fried geometry is very much lower compared to Southwell geometry. While considering Fried geometry, there are more undesirable artifacts and so it is better to use Southwell geometry. SSIM of Southwell geometry is also greater to some extent when compared with other geometries. It can be seen that the tumour is more clearly visible in Southwell geometry image.

In Table 3, the twists and turns of the brain tissues are clearly visible in the fused image of Southwell geometry. But they are not visible in Fried geometry. Southwell geometry is having PSNR and SSIM value very much better than Fried geometry. SSIM value of Southwell geometry is nearly 100 times better than Fried geometry.

In Table 4, Carcinoma is visible more obvious in the fused image of Southwell geometry. Noise ratio and similarity index are higher when compared to other two geometries which may result in better diagnosis of the disease.

In Table 5, Similarity index is very poor in Fried geometry. The features of the original images are not clearly visible. It will be difficult for doctors to diagnose.

Table 1:- Fusion of MRI/PET images of Normal Brain

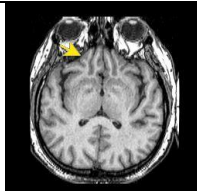
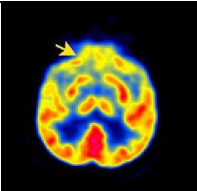
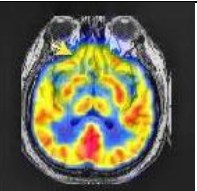
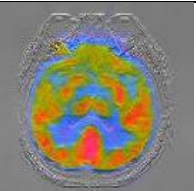
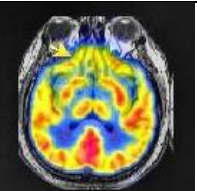
CASE	MRI	PET	HUDGIN GEOMETRY	FRIED GEOMETRY	SOUTHWELL GEOMETRY
NORMA L BRAIN					
PSNR			18.3559	12.2044	18.9041
SSIM			0.4686	0.0069	0.4872

Table 2:- Fusion of MRI/PET images of Brain Tumour

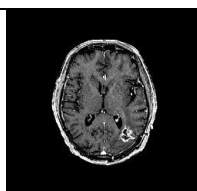
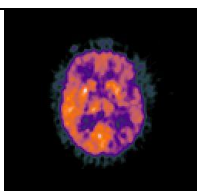



CASE	MRI	PET	HUDGIN GEOMETRY	FRIED GEOMETRY	SOUTHWELL GEOMETRY
BRAIN TUMOU R					
PSNR			18.024	14.0082	22.5549
SSIM			0.2320	-0.0324	0.2775

Table 3:- Fusion of MRI/PET images of Motor Neuron disease

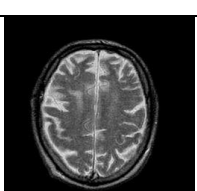
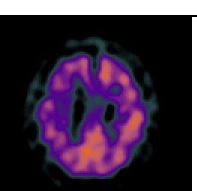
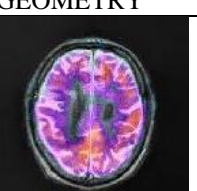
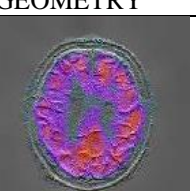
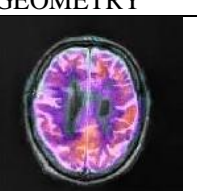
CASE	MRI	PET	HUDGIN GEOMETRY	FRIED GEOMETRY	SOUTHWELL GEOMETRY
MOTOR NEURON DISEASE					
PSNR			21.6072	14.3681	21.9384
SSIM			0.4041	-0.0421	0.4213

Table 4:- Fusion of MRI /PET images of Metastatic Bronchogenic Carcinoma

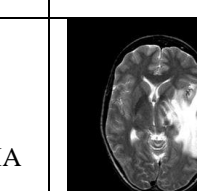
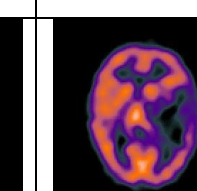
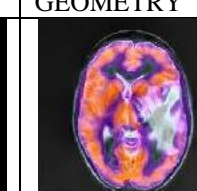
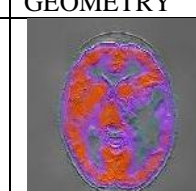
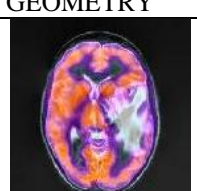
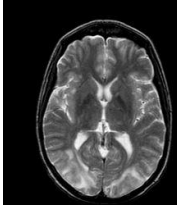
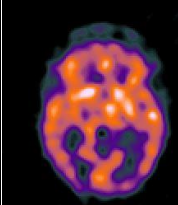
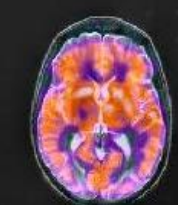


CASE	MRI	PET	HUDGIN GEOMETRY	FRIED GEOMETRY	SOUTHWELL GEOMETRY
META STATIC BRONCHO GENIC CARCINOMA					
PSNR			19.4366	13.1369	19.9160
SSIM			0.3505	0.0176	0.3629

Table 5:- Fusion of MRI/PET images of Hypertensive Encephalopathy

CASE	MRI	PET	HUDGIN GEOMETRY	FRIED GEOMETRY	SOUTHWELL GEOMETRY
HYPERTENSIVE ENCEPHALOPATHY					
PSNR			19.7199	13.2178	20.2520
SSIM			0.3759	-0.0324	0.2775

Conclusion:-

This work has demonstrated the fusion of MRI and PET images using Gradient Selection method. The luminance channels of MRI and PET are taken and the gradients are selected using three geometries namely Hudgin geometry, Fried geometry, and Southwell geometry. The performances of the three geometries are analyzed using the parameters PSNR and SSIM. Based on the comparative analysis, it is found that the Southwell geometry provides better results for fusion of MRI and PET images.

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RESEARCH ARTICLE

A STUDY ON RURAL MARKETING PERSPECTIVE OF FAST MOVING CONSUMER GOODS (FMCG). (WITH SPECIAL REFERENCE TO COIMBATORE DISTRICT)

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Abstract

India is a synonym with a wide range of diversity. Even in the state and union territories, which have been carved out based on culture and lingual communality, witnessed a marked variation in the economical socio-cultural background communality distinct. The rural consumers are not always “distant cousins” of the urban fold. They have their distinct personality and independent thinking based on the strong set of beliefs and values. Thanks to television and developments in transportation, today, a customer in a rural area is quite literate about the countless products that are on offer in the market place. The sector is excited about burgeoning rural population whose incomes are rising and which is willing to spend on goods designed to improve lifestyle. Also with near saturation and cut throat competition in urban India, many producers of FMCG are driven to chalk out bold new strategies for targeting rural consumers in a big way.

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Introduction: -

The Indian Fast Moving Consumer Goods (FMCG) industry began to get shaped during the fifty years. The FMCG sector is a cornerstone of the Indian economy. This sector touches every aspect of human life. Indian FMCG market has been divided for a long time between the organized sector and unorganized sector. Unlike the US market for FMCG presents a tremendous opportunity for makers of branded products who can convince consumers to buy branded products. FMCG refers to consumers' non-durable goods required for daily or frequent use

With the liberalization and growth of the Indian economy since, the early 1990s, the Indian customer witnessed an increased exposure to new domestic and foreign products through different media. Apart, from this, social changes such an increase in the Indian consumers' personal consumption.

Generally, a consumer buys FMCG goods at least once in a month. This sector is considered as the lucrative sector with a total market size of around Rs. 52,000 crores. In any country, the good performance of FMCG segment reflects the growing incoming level among the consumers. At the beginning of the privatization process in our country, many MNC's had given priority to FMCG sector.

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Objectives: -

The objectives of the study are as follows

- ❖ To understand the socio-economic profile of the respondents
- ❖ To study the perception of respondents on FMCG products.

Scope of The Study: -

Rural consumers have customs and behavior and marketers may find it difficult to serve them as they are mostly equipped with urban-based marketing skills. This study has confined its scope in understanding the level of awareness about FMCG products, examine the purchase pattern of selected FMCG products, the purchase decision taken by the rural consumers. In addition, it also analyses the factors influencing the same. Further this study also takes into consideration regarding the perception of respondents on FMCG products and their level of satisfaction towards them.

Limitations of The Study: -

The study is based on the opinion of the sample respondents from rural area of Coimbatore district. All possible efforts have taken to see all the respondents reveal the true facts and figures and what they feel about selected FMCG products.

- ❖ The present study deals with some selected FMCG products via Washing powder, Toilet soaps and Health drinks. It has not covered the entire FMCG products.
- ❖ Only individual consumers have been chosen as sample respondents.
- ❖ The study has covered only the rural parts such as Chettipalayam, Aalaandurai, Iruttupallam and Madhampatti which is in the interior parts of Coimbatore district.
- ❖ The study is based on the feedback of the respondents. It always calls for credibility issues.
- ❖ The result may not be generalized to other villages.

Review of Literature: -

- ❖ Purba Basu (2004), explains the need of understanding the rural consumers among the marketers and fine tune of their strategy to reap benefits in the coming years except a few life styles based products which depends mainly on urban India. The greatest hindrance to reap the opportunities in rural India is understanding the consumer behavior.
- ❖ Nandha and Chinnaiyan (2003) found that product quality was the first factor influencing the brand preference of soft drinks in the rural areas of Tamil Nadu followed by the availability and retail price. Rural price, advertisement, word of mouth and retail shop keepers are the other factors which influence the brand of soft drinks in the study area.
- ❖ Dhillon et al., (1999) identified that the number of all durable goods possessed by urban home makers was more than the rural ones. The desire to acquire the latest model of durable goods and to enhance the works efficiency for reducing the drudgeries of daily household chores were found the major factors for replacement of durable goods for both the rural and urban home makers.
- ❖ Keshav et al., (2002) found the rural consumers believed in joint buying decisions making in consultation with the elders and the ladies of the house. However, the youngsters purchase items of their personal use as per their own independent buying decisions. Advertisement with rural culture and regional / local language attracted the audience. All the respondents felt strongly about their customs and traditions. The respondents were aware of their availability of the products. They preferred quality to price.
- ❖ Ghosh (2007), pointed out in his study on, "FMCG companies in rural Indian Markets", that there are key challenges for FMCG companies who are penetrating the rural markets, which include infrastructure issues like poor distribution system, improper logistics of the fragmented rural market, given the heterogeneity of the Indian population. The companies who understand these challenges and tune their strategies accordingly will surely be the winners in the years to come, taking advantages of this economic boom in the rural sector in India.
- ❖ Samuel (2005), in his study on rural consumer behavior towards selected non-durable goods in Coimbatore district, revealed that the rural people change their tastes and preferences in procuring non- durable goods, the purchase decision pattern and consumption of certain non- durable goods in the daily market.
- ❖ Ambika (2007), revealed in her study that advertising itself cannot sell the product. It cannot sell the product of poor quality, product which is too costly or items which do not come up to the expectations of the consuming public. The product that we use in day to day life like butter, potato chips, tooth paste, household care products, packaged food and beverages etc.,

Methodology: -

- ❖ The study has used values based on primary data which has been collected from the individual respondents using interview schedule.
- ❖ Convenient sampling method was adopted to collect the responses from 200 respondents who are the individuals purchasing household care products, personal care products and food & beverages at various POP outlets including grocery stores, weekly market, nearby stores and medical shops located in the rural areas of Coimbatore.
- ❖ The data collected have been analyzed with SPSS package based on the objectives of the study.
- ❖ The statistical tools applied for the studies are
 - Simple percentage analysis
 - Garrett ranking

Analysis and Interpretation: -**Table 1: -Socio - economic profile of the respondents**

Particulars	Variables	No.	%
Age	Below 20 years	41	20.5
	20-25	34	17
	25-30	34	17
	30-35	22	11
	Above 35	69	34.5
Educational Qualification	Illiterate	21	10.5
	School Level	111	55.5
	College Level	47	23.5
	Professional	21	10.5
Occupation	Business	44	22
	Agriculture	29	14.5
	Employee	46	23
	Professional	18	9
	Others	63	31.5
Marital Status	Single	66	33
	Married	134	67
Family Type	Joint	56	28
	Nuclear	144	72
Family Size	2 Members	4	2
	3 Members	40	20
	4 Members	56	28
	5 Members	54	27
	Above 5 Members	46	23
Family Income	Below Rs. 5000	58	29
	Rs. 5000 – 10000	55	27.5
	Rs. 10000 – 15000	57	28.5
	Rs. 15000 – 20000	15	7.5
	Above 20000	15	7.5

It is observed from the above table that among the total respondents,

- ❖ Majority of the respondents belongs to the age group of 35 years and above.
- ❖ Majority of the respondents have studied up to school level.
- ❖ Majority of the respondents have other occupations.
- ❖ Majority of the respondents are married.
- ❖ Majority of the respondents belong to nuclear family
- ❖ Most of the respondents have 4 members in their family.
- ❖ Majority of the respondents earn up to Rs.5000.

Perception of respondents on FMCG products: -**Table 1: -** Perception of Respondent on Washing powder

Quality	Score		Rank
	Total	Mean	
Removes stain	11175	55.88	5
Washes well in any water	12087.5	60.44	2
It washes more clothes	12162.5	60.81	1
Consumes less water	11512.5	57.56	4
Easy to handle	6000	30	7
Non-allergic	10225	51.13	6
Keeps the clothes fresh	12062.5	60.31	3
Good fragrance	4175	20.88	8

Interpretation: -

It is seen clear from the above table that among the perception on FMCG- washing powder, the perception 'Washes more clothes' has secured the higher mean score and stood at the top, followed by 'Washes well in any water' received the secured second place, 'Keeps clothes fresh' stood at third by securing the highest mean score, 'Consumes less water' stood at fourth, 'Removes stains' stood at fifth, stood 'Non allergic' at sixth, 'Easy to handle' stood at seventh and finally, 'Good fragrance' has secured last score and stood at last.

Majority of the respondents prefer washing powder because of it washes more clothes.

Table 2: - Perception of Respondents on Toilet Soap

Quality	Score		Rank
	Total	Mean	
Good cleaning	10887.5	54.44	3
Good for skin	10337.5	51.69	4
Good fragrance	6387.5	31.94	7
Easy availability	13137.5	65.69	1
More lather	100337.5	50.19	5
More refreshing	9287.5	46.44	6
Low price	7087.5	35.44	8
Brand image	12812.5	64.06	2

Interpretation: -

It is seen from the above table that the perception of the respondents on Toilet soaps, the perception on 'Easy availability' has secured higher mean score and stood at the top, followed by 'Brand Image' has secured next higher at second place, 'Good cleaning' has secured next higher score and stood at third, 'Good for skin' stood at fourth, 'More lather' stood at fifth, 'More refreshing' stood at sixth, 'Good fragrance' stood at seventh and finally, 'Low price' has secured last score and stood at last. Majority of the respondents prefer toilet soaps keeping in mind of its availability.

Table 3: -Perception of Respondents on Health Drinks

Perception	Score		Rank
	Total	Mean	
Flavor	8237.5	41.19	3
Taste	7612.5	38.09	6
Color	7550	37.75	7
Refreshment	7175	35.88	8
Smell	8587.5	42.94	1
Packing	7837.5	39.19	5
Brand image	8075	40.38	4
Durability	8375	41.88	2

Interpretation: -

It is understood from the above table that the perception of respondents on Health drinks, perception under FMCG category, 'Smell' has secured higher mean score and stood at top, followed by 'Durability' has secured next higher and stood at second, 'Flavor' has secured next higher score and stood at third, 'Brand image' stood at fourth, 'Packing' stood fifth, 'Taste' stood sixth, 'Color' stood seventh and finally, 'Refreshment' has secured last score and stood at last.

Majority of the respondents prefer health drink because of its smell.

Suggestions: -**❖ Price Reduction**

The study revealed that people with formal education give top priority to price of products. Being a low involvement product of regular purchase, the prices of personal care products may be reduced by avoiding unnecessary expenditures, instead of frequent price discounts and extra quantity offers which disturbs the brand loyalty.

❖ Need for more products research

The women consumers who increasingly dominate the decision making and consumption of FMCG products having increasingly started looking for specialized products to offer customized solutions to their needs. This requires increased thrust on product research and innovation.

❖ Promotion and distribution

The FMCG players need to concentrate on increased consumer awareness through advertisement campaigns, new brand advertisement, free samples and expansion of franchisee network.

Conclusion: -

It is concluded from the study that FMCG sector is growing and shall continue to grow very fast. The future for the FMCG sector looks extremely encouraging. This sector having undergone a structural change is all set to emerge stronger in future.

One of the greatest achievements of the FMCG industry has been the 'sachets', which helped companies to introduce the product in small package size, at lower price points, to reach new users and rural consumers to expand the market share for value-added products in India.

The FMCG market remains highly fragmented with almost of the market representing unbranded, unpackaged home-made products, who can prompt consumers to use their branded products. If the suggestions given in the study is carried out by the FMCG, then the sector will not only grow by 50% in the forthcoming year, but also grow to newer heights soon.

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RESEARCH ARTICLE

CRYOGELS POREUX A BASE DES FIBRES DE LIN TRAITEES PAR UN MELANGE LIQUIDE IONIQUE EMIMAC / DMSO.

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Abstract

Les cryogels lignocellulosiques ultra poreux sont reconnus comme des matériaux fonctionnels, en raison de ses différences qualitatives dans les propriétés, la densité et la porosité, et diverses compositions chimiques. Les fibres de lin sont très prometteuses car elles présentent une résistance mécanique élevée, une stabilité chimique, une biodégradabilité, et une non-toxicité sont constituées principalement par de cellulose (72%). Les cryogels lignocellulosiques ultraléger et très poreux sont préparés par dissolution des fibres de lin dans un mélange de solvant à base de 1-éthyle-3 méthylimidazolium (EMIMAc) et de Diméthylsulfoxyde (DMSO). Le biomatériau à base de lin brute présente des propriétés texturales, morphologiques et cristallines remarquables. La dissolution des fibres de lin dans l'EMIMAC / DMSO est partielle, par contre, sa dissolution dans les mêmes paramètres avec de l'EMIMAc pur a été parfaite, et aucun trace des fibres observée par microscopie optique et image morphologique.

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Introduction:-

Les politiques actuelles de synthèse des matériaux se basent sur trois grands principes: le respect de l'environnement et des hommes, la réduction des déchets et des matières polluantes et le remplacement partiel ou total des dérivés pétrochimiques par des ressources renouvelables lignocellulosiques. Ainsi, la valorisation de la biomasse lignocellulosiques est favorisée par deux facteurs. D'une part, l'industrie des matières plastiques doit faire face à une diminution des dérivées pétrochimiques, d'autre part à une prise de conscience grandissante des populations de l'impact des matériaux plastiques sur notre environnement (dégagement de CO₂). Alors des stratégies connues pour convertir la lignocellulose en matériaux poreux impliquant différentes étapes d'élaborations suivies par des traitements chimiques sont recommandé. Parmi ces stratégies nous avons pensé à la solution suivante : La synthèse des matériaux poreux sans recours à des traitements chimiques.

C'est pour cela que nous avons pensé à une matière lignocellulosiques brute qui sera traité par le procédé que nous allons décrire. L'utilisation des matières primaires lignocellulosique pour la fabrication de produits respectueux de l'environnement a généré beaucoup d'études, avec différentes étapes nécessaires pour l'obtention d'un produit tel que

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Tous les axes de recherche sont d'une grande importance dans le domaine de la lignocellulose. Cependant, une attention particulière a été donnée aux aspects d'accessibilité de la Fibre de lin. Il est à noter que cette fibres est mondialement connue, elle est utilisé actuellement pour la fabrication des plusieurs bio composites (les chaises, les cadres de lunette...).

D'autant plus, beaucoup des procédés chimiques qui sont utilisés pour séparer la cellulose et l'hémicellulose de la biomasse lignocellulosique, car la lignine molécule très solide, sont à la fois lourds et coûteux. Nous tenons à préciser qu'aucun travail n'a été effectué dans ce domaine en utilisant la dissolution des fibres de lin brute dans l'EMIMAc/DMSO.

Dans cet article nous résumons les différentes étapes de synthèse et caractérisation que nous avons suivie pour atteindre notre but.

Etudes experimentales :-

Materiels et produits chimiques:-

Nous avons utilises:-

- Des fibres de lin de longueur 0.5 mm de la part de Dehondt-France,
- Un liquide ionique : 1-Éthyle-3-méthylimidazolium acétate 97% (EMIMAc, Sigma-Aldrich) ,
- Un co-solvant : Diméthyle sulfoxide (DMSO),
- Des coagulant non solvant : de l'eau dé-ionisée et de l'acétone absolue
- Tous les réactifs ont été utilisés tels que reçus sans aucune purification.

Procedes De Préparations Des Solutions Lin/EMIMAc-DMSO (80-20):-

Etapes de prétraitement et dissolution des fibres de lin:-

Nous avons séché les fibres de lin avant dissolution à une température de 50°C sous vide pendant 2 heures, à fin d'éliminer tous trace d'eau adsorbé. Puis ces fibres ont été dissoutes dans un mélange EMIMAc/DMSO (80/20) en poids à 70°C pendant environ 124 heures sous agitation magnétique de 500 rpm. Les rapports des fibres de lin: solvant mixte étaient 1:99, 2:98 et 3:97 (% masse / masse). Les solutions ont été respectivement marquées comme FL1, FL2 et FL3.

Enfin, lorsque des solutions colorées, homogènes et visqueuses ont été obtenues, une centrifugation des solutions obtenues a eu lieu pour bien séparer la phase gel de la phase non dissoute. En fin ces solutions ont été stockées à température ambiante et protégées contre l'absorption d'humidité.

Etapes de Régénération:-

L'acétone et l'eau dé-ionisée ont été utilisées comme non-solvants pour les bains de régénérations. Les solutions obtenues ont été versées dans des tubes en polypropylène, puis de l'acétone pure y a été ajouté soigneusement. Ce mélange a été mis au repos pendant 6 jours pour favoriser la diffusion solvant co-solvant. Ces solutions pré-régénérées obtenues ont été recouvertes de l'acétone périodiquement changée (4 fois/jour) pendant 10 jours. Une étape de changement de non solvant a eu lieu après la régénération des solutions dans l'acétone, dans des tubes en polypropylène remplie d'eau bi-distillées pendant 48h à fin de remplacer tous traces d'acétone par celui de l'eau. Durant cette mise en forme la teneur en solvant dans les solutions FLX(x : 1, 2 ou 3 % en Fibres de lin) varie de très important lors de la dissolution, elle passe à très faible lors de la fin de régénération.

Etapes de Séchage: Lyophilisation:-

Tous les échantillons ont été séchés à 40 mTorr et -80 ° C pendant 48h dans un lyophilisateur (Cryotec Comos 80). Après la lyophilisation, on obtient des cryogels de dimensions définies. Les cryogels LIN/EMIMAc-DMSO ont été marqués comme CF01, CF02 et CF03, respectivement.

Caractérisations:-

Des observations microscopiques ont été effectuées après avoir placé les fibres de lin en contact avec le solvant mixte entre deux lames de verre, par un microscope optique métallographique inversé (Olympus PMG3) de hautes résolutions. Les photos d'agrandissement 500X ont été prises par un logiciel Archimed.

Aussi les mesures rhéologiques en écoulements des solutions de Fibre de lin/EMIMAc-DMSO ont été effectuées sur un rhéomètre Bohlin Gemini équipé d'une géométrie cone-plan (4°-40mm) et d'une platine Peltier pour le contrôle

de la température. La dépendance de la viscosité en fonction du taux de cisaillement pour les solutions de cellulose (Lin)/EMIMAc a été enregistrée à températures fixes de 0 à 100°C, par incrément de 5°C.

Pour savoir si ces travaux ont eus des résultats nous avons procédé par une spectroscopie infrarouge à transformée de Fourier (ATR-FTIR, Spectrum Two Perkin Elmer), pour étudier la composition initiale des fibres et les cryogels. Le traitement spectroscopique a été enregistré dans la gamme de longueur d'ondes de 4000-500 cm^{-1} . Les échantillons ont été balayés 10 fois avec une résolution spectrale de 4 cm^{-1} .

D'autre par, la morphologie des échantillons métallisés a été examinée au microscope électronique à balayage (SEM Zeiss Supra 40 FEG) à une tension d'accélération de 3 kV.

Tandis qu'aux surfaces spécifiques des Cryogels (SBET) ont été calculées selon la théorie de Brunauer-Emmett-Teller (BET), en utilisant des isothermes d'adsorption-désorption d'azote (ASAP 2020, Micromeritics) à -196 °C.

Les densités apparentes des échantillons ont été mesurées en utilisant le pycnomètre à poudre Geopyc 1360 de Micromeritics.

La synthèse ainsi que les caractérisations des échantillons ont été étudiées au centre de mise en forme des matériaux, l'école supérieur des mines de Paris, Sophia Antipolis-Nice, France.

Résultats Et Discussion :-

Gonflement et dissolution des fibres de lin : vue microscopique:-

Dès le début et par différents grossissement nous avons remarqué que le système lin/solvant a provoqué un gonflement des fibres de lin. Plus précisément avec l'analyse microscopique, il s'est avéré que ce gonflement est en réalité un ballonnement sous forme de chapeler de petit ballon (Fig.1).

En effet les fibres de lin gonflent de façon homogène, le solvant mixte réagit comme agent gonflant. Le mécanisme de gonflement et de dissolution est identique à celui de la cellulose dans les liquides ioniques [4]. Cela indique que le processus de dissolution des fibres de lin brute n'est pas contrôlé uniquement par la nature chimique du solvant mais également par la qualité lignocellulosiques de la matière (72% de cellulose dans le lin).



Fig. 1. Gonflement des fibres de lin dans EMIMAC/DMSO

Les paramètres clé influençant les phénomènes de gonflement et de dissolution des fibres de lin sont liés à leur composition lignocellulosiques, la structure morphologique et le degré de polymérisation élevés ($DP = 9000$) [5]. Après 10h de traitement sous agitation magnétique à température égale à 70°C, les ballons ont commencé à éclater, pour favoriser la dissolution et la formation d'une matière visqueuse.

Ce prétraitement permet une solubilisation quasi-totale de la lignine (coloration brunâtre des solutions) et des hémicelluloses avec le temps. Le degré de polymérisation (DP_{lin}) et le taux de cristallinité de la cellulose du lin sont aussi diminués. Les fibres de lin, composées majoritairement de la cellulose (72%) [6], elles prennent majoritairement les mêmes caractéristiques de gonflement que celui de la cellulose. Il a été rapporté que se phénomène a été déjà étudiée pour le coton et les fibres de bois [7], en liquide ionique avec quelques grammes de DMSO suivent la même séquence de dissolution suivant quatre phases :

- Phase 1: Ballonnement,
- Phase 2: Éclatement du ballon,

- Phase 3: dissolution des sections non ébouriffées (Rupture),
- Phase 4: dissolution des restes de membrane des ballons.

La structure supramoléculaire des polysaccharides de lin est détruite d'une façon irréversible par l'effet des paramètres de dissolution (solvant, température, temps de réaction et agitation).

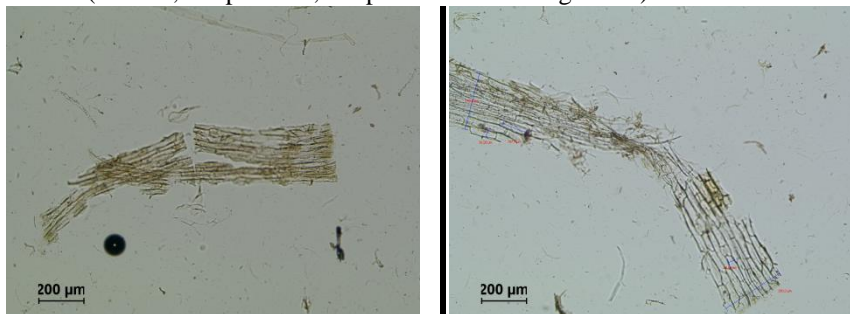


Fig. 2. Rupture des fibres au cours d'agitation

L'intérêt des liquides ioniques dans le cadre de notre étude repose sur leurs remarquables capacités à dissoudre des composés lignocellulosiques ou polymériques brutes, polaires ou non polaires, organiques ou inorganiques. Il a ainsi été montré que certains liquides ioniques pouvaient dissoudre des polysaccharides telles que la chitine [9] et la cellulose [10].

La rupture (Fig.2) n'apparaît jamais de façon brutale mais elle se déroule progressivement après que la fibre ait subi des étapes de gonflements successifs. Les fibres se déchirent peu à peu, suite à des efforts d'agitation et de température (Fig.2). La rupture des fibres de lin ne se déroule pas facilement. Elle se manifeste sous l'effet d'un phénomène de fatigue, après une certaine accumulation du cisaillement et des contraintes dans les liaisons internes des composées lignocellulosiques du lin.

Néanmoins, des solutions parfaites des polymères dissous totalement sont rarement observables même macroscopiquement. Ce pendant lorsque nous n'avons pas utilisé le co-solvant « DMSO » nous avons obtenues une dissolution parfaite. Ce qui montre que ce co-solvant entrave la perfection de dissolution.

Courbes d'écoulements:-

Généralement le comportement rhéologique des solutions dépend largement de la nature de la matière qui les constitue (la concentration et de la température). La Figure 4 présente les courbes d'écoulement des solutions de lin qui se comportent comme des gels très visqueux après l'étape de dissolution.

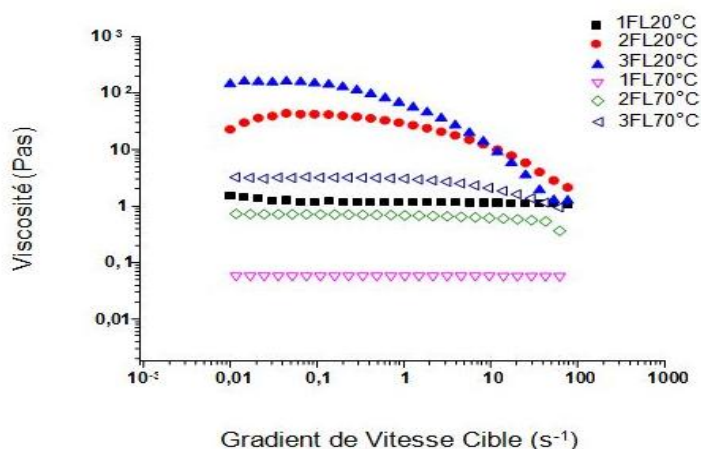


Fig. 3. Courbes d'écoulement des fibres de lin DANS EMIMAC/DMSO.

Il est à noter aussi qu'à concentration et à température faible (1% massique, 20°C), nous obtenons des courbes de tendances linéaires (courbe noire), ainsi que pour des valeurs de températures élevées quel que soit le degré de

concentration. Par contre pour un degré de concentration élevé et une température faibles nous avons obtenues courbes présentent un comportement Newtoniens.

L'augmentation de pourcentage de lin conduit à une augmentation progressive de viscosité. De plus l'influence du DP des fibres de lin sur la viscosité élongationnelle est aussi très importante pour des faibles déformations. Cela corroborait, par plusieurs travaux antérieur, qui ont montré que la viscosité est très importante [11], si le DP est plus élevé (degré de polymérisation de lin égale a 9000) [5].

Caractérisations texturales des cryogels:-

La lyophilisation produit des cryogels hautement poreux et à faible densité mais à un effet destructeur sur la structure du réseau sur tout pour les concentrations massique faibles (pour une concentration massique de 0,5g en fibres de lin l'échantillon sa tient pas malgré la dissolution totale du lin).

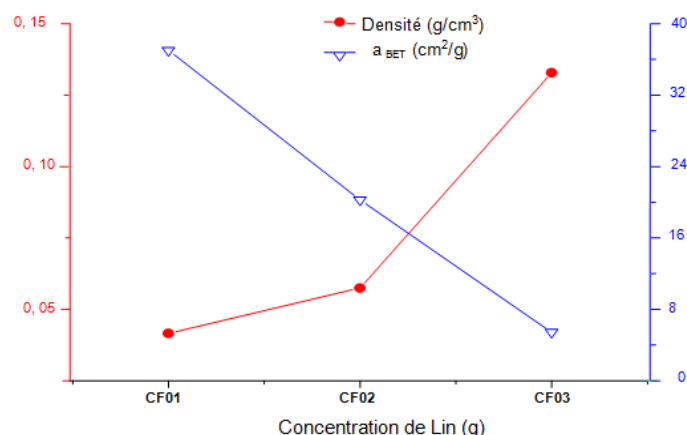
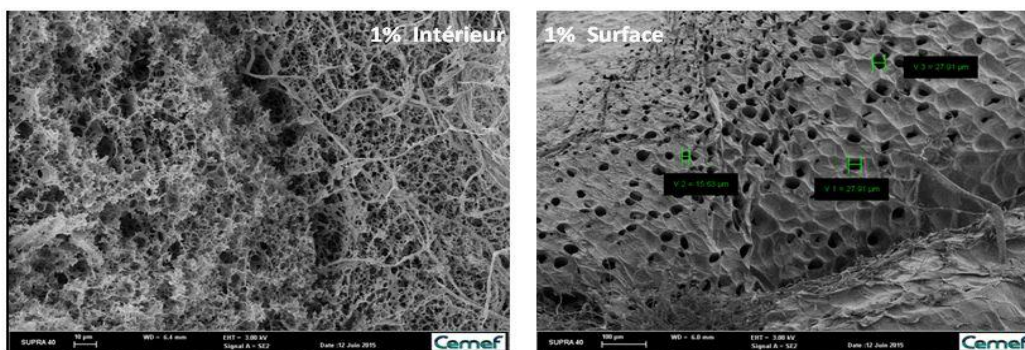


Fig. 4. Courbes de densité et de surface spécifique des cryogels en fonction de la concentration massique de fibre de lin.

L'augmentation de la concentration en fibres de lin à toujours pour conséquence la densification du réseau (Fig.5), et une diminution de porosité et de la surface spécifique des cryogels de lin. La porosité des cryogels synthétisées a été aussi déduite par les images morphologiques.

Morphologies des cryogels de lin:-

La morphologie des échantillons lyophilisés a été examinée par MEB (Fig.6). Les échantillons cryogels ont été étudiés en morphologie à l'intérieur et à la face supérieure pour les trois concentrations déjà mentionné.



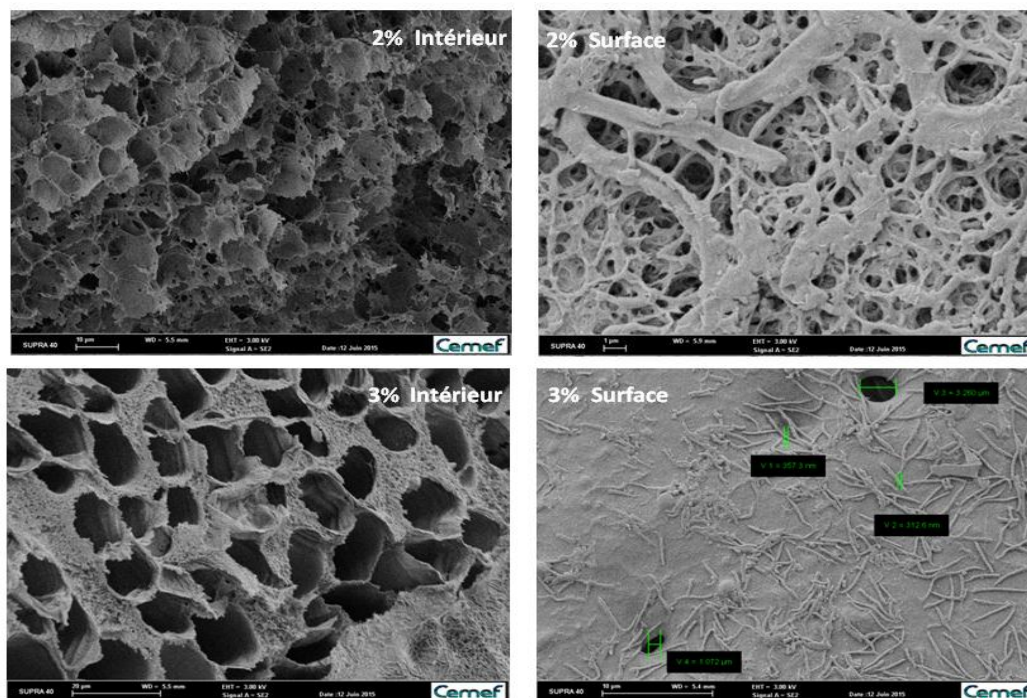


Fig. 6:- Les images morphologiques de coupes intérieures et la surface supérieur des cryogels CF01, CF02 et CF03.

Les faibles grossissements permettent de discerner un réseau poreux à l'intérieur et un réseau solides avec apparition des traces des fibres de lin non dissoutes à la surface supérieur des cryogels (Fig.6). A ce niveau de grossissement, on peut distinguer la haute porosité des cryogels élaborées ainsi que les zones plus foncées (noir), qui doivent correspondre à du vide, donc vraisemblablement à un réseau poreux des matériaux.

La formation du réseau poreux apparaît pour les différentes concentrations des fibres de lin. On peut distinguer que pour une concentration égale à 1% en masse de lin (1 : 99) le réseau n'a aucune structure ordonnée (zone feuilleté et zone fibrillaires), par contre pour celui de 2% en masse un réseau poreux se présente sous forme des feuilles, attachés les uns aux autres. Le réseau poreux feuilleté se densifie avec l'augmentation de la concentration pour former un réseau poreux de structure ordonné en nid d'abeilles de parois poreux.

On peut déduire que les parois de cryogel CF1 sont poreuses et fibreuses. De ce fait la porosité des cryogels apparaît à différentes échelles de micro à nano-pores. La densification des cryogels avec l'augmentation en teneur de lin a été déjà mentionnée (Fig.5), et les images morphologiques confirment cette densification avec la diminution progressive de porosité (nombre des pores sur la surfaces de cryogels 3%) par rapport à celui de 1% en lin. L'apparition des traces de fibres de lin pour les cryogels synthétisées à différentes concentrations exprime que le type de solvant EMIMAC/DMSO n'est pas un bon solvant pour le lin. Cette dissolution partielle est influée par l'ajout de co-solvant diméthyle-sulfoxyde.

Études spectrales des cryogels ATR-FTIR:-

Les spectres infrarouges obtenus pour les fibres de lin et le cryogel de lin sont présentés sur la figure 7. Le but de cette partie est de savoir la différence entre la matière primaire brute et le cryogel synthétisée à partir des fibres de lin par spectroscopie infrarouge.

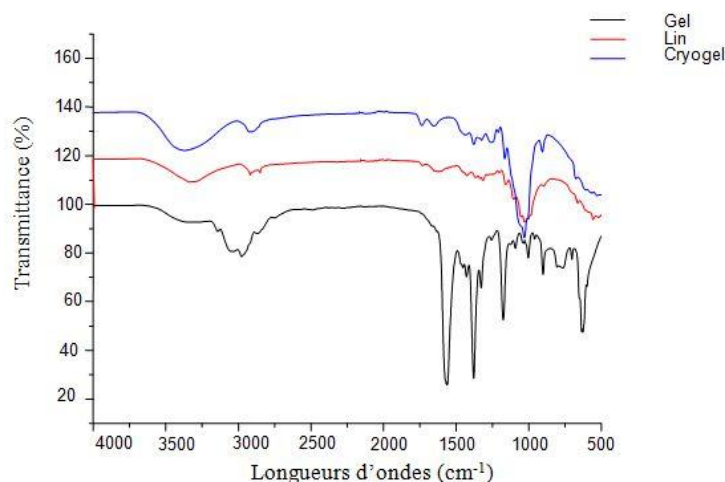


Fig.7. Les Spectres infrarouge de lin brute, solution lin/solvant et cryogel élaborées.

La comparaison des deux états de synthèses (fibres de lin brute, gel de Lin-Solvant) avant le séchage par lyophilisation confrontées afin d'évaluer l'effet de solvant sur le comportement structurale des fibres de lin.

L'apparition d'un enchevêtrement dans l'intervalle $1750-500\text{ cm}^{-1}$ pour les gels de lin en comparaison avec le spectre des fibres de lin brute. Cette enchevêtrement présente les pics caractéristiques de liquide ioniques, et due à la présence des traces de lignine, hémicelluloses et des extractibles dans la solution lin-solvant, qui disparu avec le changement successive de bain de régénération (coloration foncé du premier bain exprime la présence de forte quantité de lignine soluble dans les gels, cette coloration devient claire avec le changement successifs des bains de régénération jusqu'à ce que le bain sera incolore : couleur de coagulant). Il est toute fois possible de distinguer le pic de la ligne de base aux alentours de 1730 cm^{-1} [12].

Cette intensification des pics de cryogels dans les deux intervalles $1000-2000\text{ cm}^{-1}$ et $2750-3500\text{ cm}^{-1}$ par rapport à celui de spectre du lin brute peut être interpréter par l'importance de la méthode de séchage sur la structure spectrale finale de cryogel. On peut déduire que le spectre infrarouge de cryogel possède des intensités des pics plus importants par rapport aux fibres de lin brut. Cette intensification peut être retenue par la formation des cristaux de glace le long de l'étape de sublimation qui influe sur la structure interne du matériau et sur l'ordre latérale des matériaux (formation d'un réseau ordonné de nid d'abeille (Fig.6).

Conclusion:-

Les premiers échantillons élaborés dans le cadre de ce travail ont été considérés comme des matériaux de haute porosité à base de fibre de lin brute. Néanmoins la dissolution est partielle, le pourcentage de dissolution est de l'ordre de 85% de grandeur de celle des fibres de lin dissoute classiquement dans l'EMIMAc pur.

Les observations effectuées à l'aide de la microscopie optique montrent des importantes étapes de changement de la morphologie des fibres lors de l'étape de dissolution (des fibres gonflé par ballonnement, éclatement des ballons, dissolution des membranes externes des ballons...). Le gonflement du lin est observé au premier 6h de dissolution, mais après les 6 heures le gonflement disparaît avec le temps (éclatement des ballons), qui correspondent également à la dissolution progressive des fibres de lin.

La microscopie électronique à balayage a permis de compléter les informations obtenues par microscopie optique, sur la dissolution partielle des fibres de lin, par l'apparition des traces des fibres de lin non dissout à l'intérieur et la surface des cryogels. En effet, ce que l'on a pu observer c'est un trace des fibres de lin non dissolu avec diminution de leurs longueurs et largeur initiale cela est à cause du fort degré de polymérisation de lin ($DP_{lin} = 9000$) et l'effet de l'ajout de DMSO.

Il semble que la structure cristalline soit plus affectée à une plus grande concentration en lin.

Les résultats préliminaires de notre étude sur les moyens d'augmenter l'accessibilité de convertir les fibres de lin en matériaux hautement poreux. Des analyses plus détaillées doivent être effectuées. Par exemple, il serait intéressant de dissoudre la fibre de lin brute avec d'autres types de solvant plus efficaces pour minimiser le temps de traitement et le pourcentage de dissolution.

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RESEARCH ARTICLE

ETHNOGRAPHIC STUDY OF THE MAO NAGA TRIBE OF MANIPUR, INDIA

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Abstract

The Mao Naga is a major Naga tribe in Manipur, India. They bear an important place in the history of migration and dispersion of the Nagas. Makhel being the first settlement area of the Nagas and the *Chitebu-Kajü*, the place of dispersal of the Nagas are located in the Mao territory, the Mao Naga can be considered as the pioneer settler among the Nagas. Till today, the Maos continue to play an important role in the politics of the Naga for their strategic location and as a permanent member of the Tenyimi Peoples' Organization (TPO).

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Who are the Mao Naga?

The Mao Naga is a major Naga tribe in Manipur, India. They are a recognized 'Scheduled Tribe' under the Constitution Scheduled Tribes order of 1950. They are known as *Ememei* in Mao language. They occupy the northernmost part of the state spread across Senapati district, bounded by similar Nagas like Angami and Chakhesang in the north, Maram and Zeme in the west and south, and Tangkhul and Poumai in the east. The main area of their habitation falls under Mao-Maram Sub-Division. According to 2011 census, the total population of Mao Naga under Mao-Maram Sub-Division is 1, 16,374. There are 58 Mao villages, out of which 38 villages are revenue village and the remaining 20 are federal unit.

The Maos are also found in large numbers in the town areas of Senapati and Dimapur and in capital cities of Imphal and Kohima. They have formed unions to unite and coordinate among themselves in the town areas where they have settled permanently as Mao Senapati Union, Mao Imphal Union, Mao Kohima Union, Mao Hoho Dimapur and Mao Union Jalukie in the respective town. However, for administrative purpose, the Maos are recognised only in Manipur state as a 'Scheduled Tribe'.

The Maos speak 'Mao' or '*Emela*' language which belongs to Angami-Pochuri group of Tibeto-Burman language family (Burling, 2003). Mao language is an officially recognised language which is taught as a subject up to class ten standards in the state. In 2016, the Mao language is recognised to be taught up to class 12 levels by the Council of Higher Secondary Education, Manipur. On August 12, 2016, Mao language is selected as a language subject for 'Language Certificate Programme in North Eastern Language' at Namaste, Centre for Studies on North Eastern States, Nitte University, Derlakatte, Mangalore.

The traditional habitation area of the Mao Naga is also known as 'Mao'. Mao area is a strategically important location connecting the two north eastern states of Manipur and Nagaland. National highway 2 pass through the heart of the Mao inhabited area. This highway is the only effective land connection of the landlocked Manipur state with the rest of the country.

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Origin and Migration:-

The origin and migration of the Mao Naga in particular and the Nagas, in general, is shrouded in mystery since they practiced oral tradition. Thus, it is difficult to separate the myth and history as per the oral tradition of the Mao Naga. The origin of the Mao Naga is known through the popular myth '*Dzüliamosüro*'. According to the myth, the godly and the mysterious woman *Dzüliamosüro*, was already there at Makhel (also known as '*Makhräi Rabu*', considered as the ancestral home of the Nagas), while she was resting under the 'Sacred Banyan Tree' (also known as '*Charasü Marabo Kaji*', still standing tall at the heart of Makhel village), a cluster of cloud enveloped her and she conceived and gave birth to *Ora* (spirit/gods), *Okhe* (tiger) and *Omei* (man), (Daniel, 2008; Mao, 2009; Nepuni, 2010; and Salew, 2014). The three brothers lived in harmony however when their mother became old and sick, dispute broke out between the three brothers each claiming to inherit her land. To settle the dispute, the mother devised a contest among the brothers. The contest was a race. For this purpose, she made a simulacrum with grass-like ball at a distance and said that the one who touched the simulacrum first would inherit the mothers' land. Man being the youngest and the most noble was favoured by his mother; told him to make a bow and arrow to shoot at the simulacrum as she knew that he could not compete with the powers of the spirit/gods and tiger in such a race. The man following his mother's instruction succeeded in touching the simulacrum target first by using bow and arrow and finally inherited his mother's land. The three brothers had erected a monolith each at the time of their departure at *Chazhelophi*, about 3 kilometres south of Makhel village. The monolith of spirit/gods and man is still standing erect however the tiger's monolith had fallen down.

Nepuni Salew, the present *Movou* (village Chief) of Makhel village writes that *Omei* (Man), the youngest sons of the mysterious woman *Dzüliamosüro* was known as *Alechameiwo* and he had three sons, namely, *Asüpfu Alapha/Asüpfu Alapha*, *Tuttowo/Chütuwo* and *Khephio* (2014). There are variations in terminology since they practice oral tradition and the myths were passed down from one generation to another through oral narrations and due to variation in dialects from one village to another. Whatever might be the differences, it was said that the three brothers lived together at Makhel for a very long time. It was said that there were seven hundred houses on the eastern side and seven hundred houses on the western side (Onia, 1995). There came a time when they had to move out as population flourished. At the time of their departure, they held feast and had erected a monolith. The monolith is still standing tall and located on the eastern side of the village written as '*Pinoumei Kosü Tamara Tu*', meaning '*dispersal stone of the three brothers*'.

Most Naga writers and scholars accepted that Nagas had migrated from a distant place and first made a settlement at Makhel. Heshu writes that majority of the Naga tribes considered *Kheso* as their original place from where they had migrated following a river name *Chuhrürü* as their main route and settled down at *Makhräi Rabu* (Makhel) where they can view clearly both the topography of east and west of the place (2008). While Lhousa claimed that Naga ancestors hailed from *Heziera* where there was no difference between day and night and a daughter given out in marriage grows unrecognisably changed in a year due to strong wind. He further writes that the Naga forefathers migrated from the eastern part of Asia/Indo-China through Burma and settled at Makhel while some Nagas came straight from Chindwin river direction and settled in northeastern part of Nagaland (2015). Sanyu is of the view that the Nagas were among those tribes who migrated from China and took the route of Patkai section and settled down on the way in the Naga Hills (1996).

While there is lack of clarity about the migratory routes taken to reach Makhel, it is accepted by most of the Naga tribes that the Nagas first settled at Makhel and then dispersed from it (Hodson, 1989 and Banee, 1995). Even recent writers like Lhousa (2015), Lotha (2010), Nepuni (2010); Mao (2009) and Heshu (2008) write that most of the Naga tribes first settled at Makhel and then dispersed from it. Nagas did not migrate at the same time but the migration was a continuous process in ripples of one tribe pushing the other while, in turn, being pushed by another (Lotha, 2010).

Chütebu-Kajü, the 'The Great Naga Wild Pear Tree' is a living proof that before the Nagas dispersed from Makhel; they had planted this tree to remember that they first settled at Makhel and then dispersed from it. This tree is still standing tall at Charanho village about one kilometre to the north of Makhel. It was said that in the past when a branch of this tree fell, those people who dispersed from Makhel observed a *genna* (also known as '*mani/mane*' in Mao language; it is a day of prayer and religious observance day according to the Mao traditional religion and people abstain from going to field) in their own respective village even if the information reached to them a month or so later. This tree marked a symbolic representation of the dispersals of the Naga brothers and more importantly symbolises the oneness of the Nagas (see also Heshu, 2008).

The Naga tribes who trace their dispersal from Makhel are known as 'Tenyimi' or 'Tenyimia'. Angami, Chakhesang, Mao, Maram, Poumai, and Zeliangrong are the tribes who originally traced their dispersal from Makhel. They are united under the Tenyimi Peoples' Organization (TPO), the apex body of the Tenyimi. Its unit is spread over three states namely – Nagaland, Manipur, and Assam. At present, the members' tribes of the TPO are Angami, Chakhesang, Impui, Mao, Maram, Pochury, Poumai, Rengma, Thangal, and Zeliangrong. It is formed to protect the welfare of the Tenyimi brothers and sisters and to foster unity and brotherhood among them as well as the other Naga tribes. They share a deeply rooted cultural heritage and inherent values. Ura Academy, an academic institution is set up to study Tenyidie to preserve and promote the Tenyimi language, literature, cultural values, traditional heritage and customary practices. It is located at Kohima. Tenyidie is the first Naga language to be introduced at the University level. It was introduced in the post-graduate level in 1997 under Nagaland University. In 2005, Tenyidie was further introduced for Ph.D. level. This institution had celebrated 75 years of its establishment in 2014 (1939-2014). On the 11th April 2014, some members of the TPO and Ura Academy came to Makhel and *Chütebu-Kajü* (Charanho village) seeking blessing from their ancestral place with the banner '*We are here to celebrate our common roots*' before the celebration of its Platinum Jubilee.

Ancestors of Mao Naga:-

According to the myths, *Dzüliamosüro*, *Alechameiwo* and *Khephio* were the great ancestress and ancestors of the Mao Naga. However, according to the folktales and folksongs of the people, the forefather of the Maos was known as 'Shüpfumei'. Nepuni writes that Shüpfuwo was the eldest son of Khephio and his descendants were known by the name Shüpfumei in the past (2010). The word Shüpfumei means 'the people of Shüpfuwo'. The Mao people practice oral traditions and the only source of their history is folktales and folksongs. In their folktales and folksongs, the Mao people are referred by the name Shüpfumei. Even in the early writings about the community written by the British administrators has referred them as 'Sopvomah/Sopvoma' or 'Mau/Mao' by Dun (1886), Grierson (1903) and Hodson (1989) and 'Sopvoma' or 'Memi' or 'Mao' by Hutton (1921). 'Sopvomah/Sopvoma' is a corrupt form of Shüpfumei. Heshu writes that 'the difference in the spelling of the earlier writers could be because the writers are unable to capture the exact pronunciation' (2008: 2).

Shüpfuwo had three sons namely, Kapewo, Toliwo and Choro (Saleo cited in Nepuni, 2010). Choro was the youngest and so inherited his father's land and settled down at Makhel while his two brothers went out in search of a better place for settlement in the neighbouring areas. Kapewo first settled down at Saranamei village, (a Poumai village) and his descendants are known as Kapemata/Paomata. Toliwo first settled down at Koide (a Poumai village) and his descendants are known as Tolüpemata/Lepaona (Nepuni, 2010). The descendants of Kapewo and Toliwo are together known as Poumai Naga and the descendants of Choro are known as Mao Naga. In the past, the present Mao and Poumai Naga were together known as Shüpfumei.

Khrasi, Ahrowo, Kade and Memüwo/Memüo were the four sons of Choro. The descendants of Khrasi are known as *Ekhramei* and settled down on the western side; the descendants of Memüwo/Memüo are known as *Imepadeina* and settled down on the northern and eastern side and the descendants of Ahrowo and Kade on the southern side of the Mao territory. They settled down in different villages however in close proximity to one another. At present, the descendants of Choro have settled down in 58 villages under Senapati district of Manipur.

Village Settlement:-

As population increases, within a social structure adaptations must be made in order to prevent the community from fissioning (Caneiro, 1967) so for better life the Mao people looked out for new settlement site. In the past, while selecting a new site for settlement, the Maos were very particular about the location of the site. A new site for settlement was chosen on hilltops since it had a strategic position during the warfare time since they practiced head hunting tradition. Secondly, the Mao people looked out for signs and dreams seeking divine consent before they settle down at a new place. They believe that human being lives along with benevolent and malevolent gods and spirits and so if anybody wanted to settle at a new place, he or she should first claim the place and became the owner of the place before gods/spirit claim it. Even when a child is born, the parents have to first claim the child; otherwise, if gods first claim the child, the child might not survive. Claiming the child means taking the ownership by the parents (Nepuni, 2010). After a site is chosen, they kept simulacrum and go home and see their dreams. If dreams were not good and the simulacrum too falls then they took this signs as the place is not good for settlement and they would leave the place and move on searching for new place.

When a place was finally chosen to settle, on the first day, they carried a shield, a spear, a rooster and materials needed to make fire which is known as *milasü milareï* in Mao language. It may be mentioned here that in olden days the Maos did not use match stick but they practiced indigenous technique known as '*milasü*' to light fire. Even today during cultural festivals, it is frequently witnessed that the traditional art of making fire '*milasü*' is replicated in the form of game or competition. When they reach the particular place which is decided to make settlement; first they claimed the place by saying 'I want to make this as my place and my land' and then they sacrificed a rooster to supreme god and start lightning fire through *milasü* and let the fire and the smoke reached the sky. If the smoke goes straight to the sky, it is taken as a good omen that they will have a prosperous life in the village (Saleo, 2008).

In order to depict the traditional way of making new settlement, the story of how two brothers first settled down at the present Charanho village (also known as Shajouba is one of the biggest Mao villages at present) is explained here. One day two brothers came across the present area of the village and they wanted to settle there, and so the brothers, each kept a bundle of paddy straw on a branch of a tree for the night and went home. The next day they came back to that particular place to see the sign on the paddy straw; it was found that the elder brother's paddy straw was dying out and the younger one's look fresh and lively. From this sign, it was taken that this place can be a good place for settlement however, from the sign of the paddy straw it is taken that the younger brother would be a better leader among the two. Thus, they made their settlement and as per the omen, the younger brother became the *Movou*, village Chief of the village (Personal Interview with Daiho Kholi, the present *Movou* of Charanho village).

In present times, national highway becomes a preferred choice for a new settlement. With the development of national highway 2, the Mao people began to move towards the national highway. National highway 2 passes through the heart of the Mao territory. Some of the Mao villages which are established along the national highway are Kayinu, Song Song, Kalinamei, Phikomei, Tadubi, Khridzüphi, Makhan Tabio, Makhan Centre, Makhan Lovadzüngho, Makhan Khuman, Khongnem Thana, Puni Pfosemei, Mao Karong, Taphou Pudunamei, Mayangkhang Khunou and Daili village.

Social Organization:-

The Mao Naga follows typical patriarchal and patrilineal social set up. They practice monogamy and type of family is nuclear family. Father is the head of a family. His decision is final and binding in all respect. The rule of descent is traced through the father's line or male line. The succession and inheritance of property is through sons or male offspring. The eldest son inherits his father ancestral paddy field known as *opfu lophre* and the youngest son inherit his parent's house. The Mao community gives more importance to the eldest and the youngest sons in a family since it is them who inherit their father's ancestral property. If there is no son in a family, the ancestral/parental house and the father's ancestral paddy field goes to the nearest male relative from the father side and the daughters can inherit only the remaining property of the family. The legacy of a family is carried down by male offspring in a family and so the ancestral properties are passed down only through the male line.

The next level of social organization is 'clan organization'. It is known as '*pfuta*' in Mao language. The clan is an endogamous group in a village. Inter marriage between the clan members is a taboo. Every clan have its own gate known as '*koro*', built nearby the main clan habitation area in the village. Most clan have community land. The members of every clan select their own clan representatives to administer them in the village since time immemorial. The clan representatives are known as *Enulei Kotsümei* or Village Council or Village Authority (at present). The Mao Naga is known to have numerous clans and sub-clans. As they make new settlement site and form a village, they tend to use a different name like their lineage ancestors' name or sub clan as their clan and in this way, they develop new clans with the establishment of a new village. At present, the Maos has more than 60's clans. Some commonly used clan name are Arücho, Arüna, Chachei, Chalai, Chakre, Ekhe, Kadena, Kapena, Kayina, Krichena, Lirina, Piku, Poji, Pfoze, Rajina, Maheo, Saprüna, etc.

Religion:-

The Maos have their own traditional religion known as *Opfufe Chiina* – religion of the forefather. They believe in the existence of a supreme god called *Oramei*. They also believe in the existence of benevolent gods/spirits and malevolent gods/spirits. The benevolent spirits are considered as good and helpful while the malevolent spirits are evil one which causes sickness and suffering to mankind. Thus, their religion can be called as nature-centric and animistic. They performed rituals, observed *gennas* and offered sacrifices to propitiate their gods and made appeasement. Taboos also constitute an important religious element in the Mao traditional religion. It is not only a mere prohibition with a superstitious belief but it guided and governed their socio-religious life. It has two connotations, '*moli chünoe*' – taboos of commission and '*li chünoe*' – taboos of omission (Loli, 2011). *Movou*, the

village Chief been the village priest announced the *gennas*, performed rituals and offered sacrifices for the well being of the people. Sometimes the head of the family or the woman of the family too offer sacrifices and performed rituals for the individual family.

With the coming of Christianity, the traditional religion of the Maos lost its importance and significance. At present, the Maos are 99 percent Christian. Baptist, Roman Catholic, Christian Revival, Seventh Day Adventist and Good Samaritan are the main denomination practiced by the Mao community.

Economy:-

Traditionally, the Mao Naga is an agrarian community. Agriculture is the main stay of the people. The Maos are hard working people. They work from dawn till dusk. They make terrace fields at the slopes or foots of the mountains and along the river bank. The hard work of the people can be judged from the exquisite craftsmanship of converting the hilly terrains into delicately beautiful and continuous terraces covering ranges for the purpose of rice cultivation. Every family of the Mao community has their own terrace fields and cultivate for their sustenance and sustainability (Heshu, 2008).

The Maos also practice shifting/jhum cultivation and permanent garden cultivation. They cultivate different kinds of crops. Rice, maize, potatoes, cabbages, chillies, tomatoes, tree tomatoes, cho-cho (squash), beans, pumpkins, cucumbers, brinjals, oriental onions, spring onions, onions, yam, etc are the main cultivated crop. The Regional Potato Farm, first sponsored scheme of North Eastern Council is in Mao area, located at *Pfukhro* (Song Song). This farm is wholly owned by the state government and targeted to supply potato for the whole north east regions. The Maos are the only community producing potato for commercial purpose in the state. They also supply to the neighbouring state of Nagaland. The Mao people also cultivate fruits like plum, peach, pear, guava, banana, passion fruit, kiwi, grapes, etc for consumption as well as for selling.

The Maos also rear varieties of animals like fowls, cat, dog, pig, cow, buffaloes, duck, rabbit, etc. Cows and buffaloes are used for ploughing the fields. Pisciculture is also practiced since time immemorial. The Mao people used to rear fish in the middle of the wet terrace field known as '*khochübvü*'. Fish is an important food item of the Maos. The penalty for person stealing fish from this *khochübvü* is very severe. In the past, before the introduction of monitory system, the penalty was ten *oji* (a traditional kind of basket to measure and carry paddy).

Floriculture is developing in a big way in Mao area. It was initiated by the womenfolk. They rear flowers for both beautification and for commercial purposes. The Maos has celebrated 'Flower Festival' in the year 2015 and 2016 which is first of its kind in the region. Floriculture is giving a big boost to the economy of the Maos.

In present days, the Maos begin to adopt new occupations. National highway 2 passes through the heart of the Mao inhabited area. The strategic location of the Maos on the national highway 2 has brought a lot of avenues to the Mao community especially in the field of economy. The Maos have taken up entrepreneurship, small time business men and women. And also the agricultural products could be easily transported to state capital Imphal and Kohima and other commercial hubs such as Senapati or Dimapur and sold off at good prices. At three places on the national highway 2, the Maos have developed into tourist place, open up food and tea stall and sells locally produced agricultural products, mostly of organic vegetables and flowers. The three tourist places are Mao Gate (Song Song), Vanee (Tadubi) and Oprüingho (Makhan).

The Maos are pursuing in public sector undertaking like banking, insurance, BPO, call centres, etc apart from government jobs both state and central. Another very prominent occupation adopted by the educated Maos is in the field of teaching. We can see the Maos teaching in private educational institutions in the whole of north eastern states. The economy of the Maos is slowly growing with the passage of times.

Conclusion:-

The Mao Naga bears an important place in the history of migration and dispersion of the Nagas. Makhel being the first settlement area of the Nagas and the *Chütebu-Kajü*, the place of dispersal of the Nagas are located in the Mao territory, the Mao Naga can be considered as the pioneer settler among the Nagas. Till today, the Maos are playing an important role in the politics of the Nagas for their strategic location and as a permanent member of the Tenyimi Peoples' Organization (TPO). The TPO has being playing an active role in Naga's politics fostering peace and unity among them and also other neighbouring tribes in the region. The strategic location of the Mao Naga between

Manipur and Nagaland, the Maos have been playing active role in the Naga's politics for instance - in the year 2010, when the Manipur government banned Mr Thuingaleng Muivah, General Secretary of NSCN-IM from visiting Somdel, his village in Ukhrul district, Manipur after more than four decades of being away and he got stranded at Viswema, the Maos organised peace rally to show their solidarity. Mr Muivah came with the consent of the government of India however Manipur government denied the permission on the grounds that it might cause 'communal disharmony endangering the peaceful co-existence of the different ethnic communities' (Bhatia, 2010: 38) and so the government of Manipur deployed armed forces at Mao Gate, the interstate boundary on the national highway 2 to prevent Muivah's entry to the state. The deployed Manipur armed forces assaulted a civilian at Mao Gate on 5th May 2010. The already tensed situation was aggravated by this incident. The next day i.e. 6th May while the Mao people was going a silent peace rally against this assault and demanding that the Manipur armed forces to vacate the place, the peace rally group were tear-gassed and then fired upon by the Manipur armed forces. This incident had killed two young college going students and injured many, including men, women and children. Every year on 6th May, the Maos observe the death anniversary as 'Martyr's Day' and declared special holiday and all economic activities are restricted on this day in the Mao area. The Mao Council, the apex body of the Mao Naga fought alongside with the United Naga Council (UNC), the apex civil body of the Manipur Nagas during the introduction of Naga People's Front (NPF), a regional political party which originated from Nagaland state in the year 2011.

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RESEARCH ARTICLE

A COMPARATIVE STUDY ON INTERNET USE BY RURAL AND URBAN COLLEGE STUDENTS.

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Abstract

This world is termed science and technology. Using digital devices could be a vast part of their everyday expertise. In each facet of life, we have a tendency to use technology. Technology creates our life quicker, better and easier. With the assistance of technology, we are able to simply create the teaching-learning method softer and on the market. There's no facet of human life that isn't bitten by technology. Numerous technologies have been employed in education, the internet is one in every of them. The Internet is that the backbone of this society. The Internet plays a very important role in developing the communication or interaction method. Present days is that the world of web or internet. The Internet has created our lives batter. Through the internet, we are able to simply communicate with our knowledgeable or guide everywhere in the planet. It's true that the employment of internet in rural and urban areas is increasing day by day. This study was conducted to compare the use of the internet by rural and urban college students. The sample consists of 150 female and 150 male students from four colleges in Purulia district, West Bengal, India selected randomly. The study revealed that urban college students use the internet quite rural college students in terms of daily or $\frac{3}{4}$ times of every week as calculated criteria. Once analysis it's additionally found that male college students use internet quite the feminine college students in terms of daily or $\frac{3}{4}$ times of every week. The present study also indicates that Science college students use internet over the arts college students in terms of daily or $\frac{3}{4}$ times of every week. However, the humanities college students use the internet over science counterpart in terms of $\frac{3}{4}$ times of a month or at least one time of a month. Gift study additionally indicates that Hindu college students use the internet quite Muslim college students in terms of daily or $\frac{3}{4}$ times of every week and a lot of students use internet for the purpose of social media and news.

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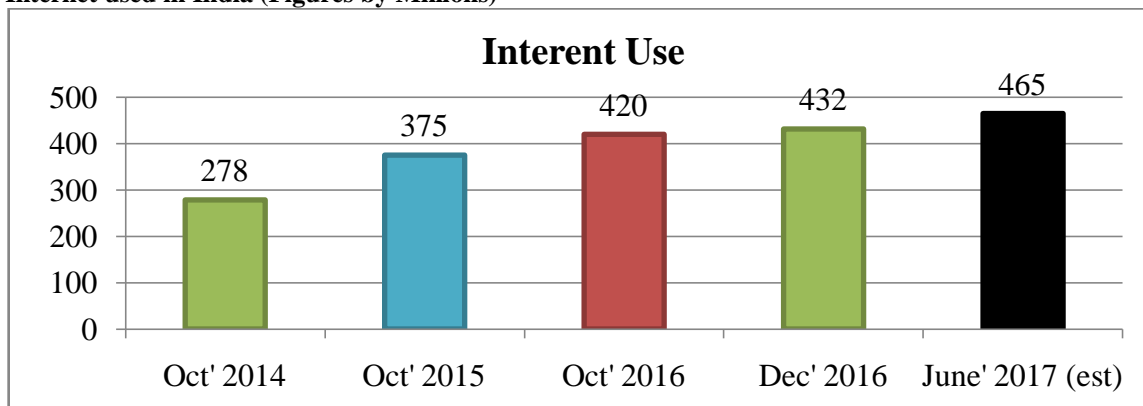
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Introduction:-

Internet is one of the powerful parts of technology and also essential for whole human society. Internet provides 24/7 access information at one click. Today without internet we can't think about our daily life. Internet is the core part of education system. Today more people are on the internet than anywhere else. With over 460 million internet users, India is the second largest online market, ranked only behind China. By 2021, there will be about 635.8 million internet users in India. Despite the large base of internet users in India, only 34.8 percent of the Indian population accessed the internet in 2016. This is a significant increase in comparison to the previous years, considering the internet penetration rate in India stood at about 10 percent in 2011. Furthermore, men dominated internet usage in India with 71 percent to women's 29 percent (www.statista.com/topics/2157/internet-usage-in-india). At present, the study noted, rural Indians use the Internet mainly for social network websites (around 70% of the Internet users living in rural areas) such as Facebook, and for emailing. A report titled 'Internet in India 2016' by the Internet and Mobile Association of India (IAMAI) jointly published by the IMRB, mentions that the number of internet users in India is expected to reach between 450-465 million by June 2017. It adds that the country had 432 million mobile internet users in December 2016, of which 269 million or 62.3% were from urban India and 163 million, or 37.7% were from rural India (Pai, 2017, <http://www.medianama.com/author/vivek/>).

Internet used in India (Figures by Millions)



Source: IMRB 1-cube.

By gender, 40% of daily internet users from urban areas consisted of females, where only 25% of daily internet users from rural areas were females.

Today the use of internet is the common matter in India. It is assumed that the college students in India feel more dependent on the Internet for their class assignments and for the latest information about their subject areas than conventional resources of information. A good number of the youth particularly college students use the internet for social interaction and communication as well as for their education. But, just as they use the internet to take help of their education, they use social sites to enrich their social lives (Jones, 2002). College also feels a bit handicapped in updating their knowledge base quickly without using the Internet for their research and classroom teaching activities (Sridevi & Indrani, 2015). Internet came to India in early 1990 for a restricted group of users only. But, now it is open to everyone (Singh, 2014). Jackson, et al. (2011) remarked that the Internet would increase the educational level due to its reach and availability to everyone, everywhere and anytime. The internet has created the opportunities for people of all ages of students to contribute and access information.

Definition of Internet:-

Cambridge International Dictionary of English defines Internet as "large system of many connected computers around the world which people use to communicate with each other". (Network of networks) The internet knowledge is the knowledge of the basic theoretical aspects of the internet and its practical application. According to Douglas E. Comer (2003) internet is "the collection of networks and routers that use the TCP/IP protocol suite and function as a single, large network. The internet reaches government, commercial and educational organization around the world.

Statement of the Problem:-

The problem for the present study is specifically stated as below:

A Comparative Study on Internet Use by Rural and Urban College Students.

Literature Review:-

Bimber, B. (2000) after measuring the differences in men's and women's use of the Internet in U.S. found the existence of gender gap in their Internet use. **Ono, H. & Zavodny, M. (2003)** also found women to be less frequent and less intense users of the Internet. Concern about gender inequality has now shifted from access to intensity. **Mishra, O.P., Yadava, N. & Bisht, K. (2005)** conducted a research study to learn the Internet utilization patterns of undergraduate students at the G B Pant University of Agriculture and Technology. The findings of the study revealed that a majority of the students (85.7%) used the Internet in which male students use Internet in greater numbers than females. **Kumar, R. & Kaur, A. (2005)** disclosed that majority of the respondents (69.4%) used the internet for educational purpose, (51.9%) for research purpose, (47.4%) for communication purpose and 34.7% for entertainment purpose. **Mishra, et al. (2005)** indicated that a majority of the students (85.7%) use internet in which that male students are more than female students. **Loan, A.F. (2011)** revealed that majority of the students are frequent users of internet using internet from daily to week basis in which use by urban students is more than their rural counterparts. **Thanuskodi, S. (2013)** found that internet was used by boys and girls to some extent. Both boys and girls had equal access to Internet but there was a difference in usage pattern. **Sharma, et.al. (2014)** conducted a survey on 391 and found that male's students were more addicted to the internet use than female.

Delimitations of the Study:-**(A) Geographical Area**

The investigation was delimited to only Purulia district of West Bengal India.

(B) Level of Education

1. The study was restricted to the college students (UG level) in Purulia district.
2. Among the College students, only the science and Social Science students were considered as the subjects of the present study.

Objectives of the Study:-

The study was conducted to find out the following objectives.

1. To point out the frequency of the internet use by college students
2. To compare the use of internet by rural and urban college students.
3. To compare between Male and Female college students in respect of their using internet.
4. To compare between Arts and Science college students in respect of their using internet.
5. To compare between Hindu and Muslim college students in respect of their using internet.
6. To know the purpose of internet use.

Methodology of the Study:-

The survey method of research was used to conduct the study and a structured questionnaire, specially drafted for the purpose is used for collecting the data. Data were analyzed by different quantitative techniques and presented in the appropriate formats.

Population of the Study:-

All the college students (UG level) in Purulia district of West Bengal (India) are the population of this study.

Sample and Sampling:-

Four Colleges of Purulia district, West Bengal were selected randomly. The researchers selected 300 students randomly. Here stratified random sampling technique was adopted. The sample profile is given in Table- 1.

Table 1:- Sample Profile.

Male	Female	Rural	Urban	Science	Arts	Hindu	Muslim
150	150	130	170	100	200	260	40
Total 300		Total 300		Total 300		Total 300	

Data Analysis and Discussion:-

In this chapter, the researchers analyzed and interpreted the data collected. The data are collected through questionnaire method. After checking the questionnaire for completion and editing the entire, the researchers analyzed the data.

Objective-1: To point out the frequency of use internet by college students.

Table 2:- Show the frequency of use internet by college students

Use Internet	College Students
Daily	100/300 (33.33%)
$\frac{3}{4}$ Times of every week	120/300 (40%)
$\frac{3}{4}$ Times of a month	60/ 300 (20%)
One time of a month	20/ 300 (6.67%)

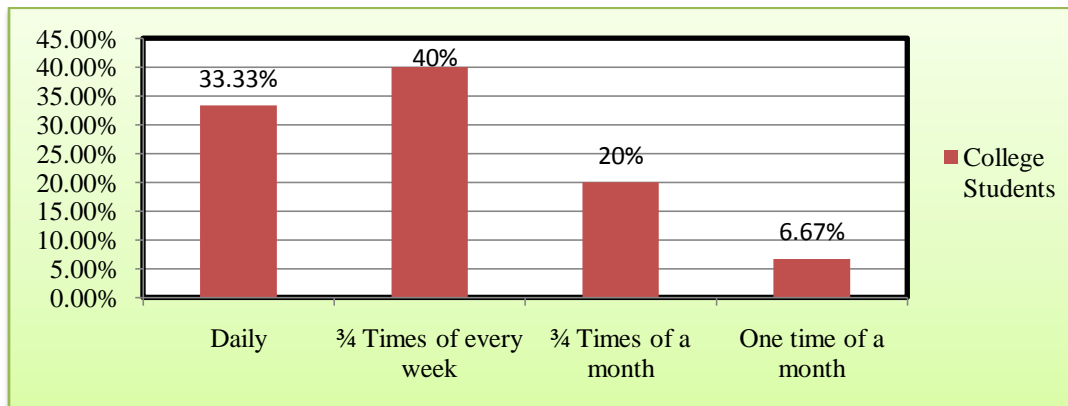


Figure 1:- Percentage of using internet by college students.

Table 2 shows that 33.33 percentage college students using internet from daily to $\frac{3}{4}$ times of every week and 26.67 percentage college students using internet from $\frac{3}{4}$ times of a month to at least one time of a month.

Objective 2:- To Compare the use of internet by rural and urban college students.

Table 3:- Show use of internet by rural and urban college students

Use Internet	Rural	Urban
Daily	55/130 (42.31%)	80/170 (47.06%)
$\frac{3}{4}$ Times of every week	45/130 (35.62%)	65/170 (38.24%)
$\frac{3}{4}$ Times of a month	20/130 (15.38%)	15/170 (8.82%)
One time of a month	10/130 (7.69%)	10/170 (5.88%)

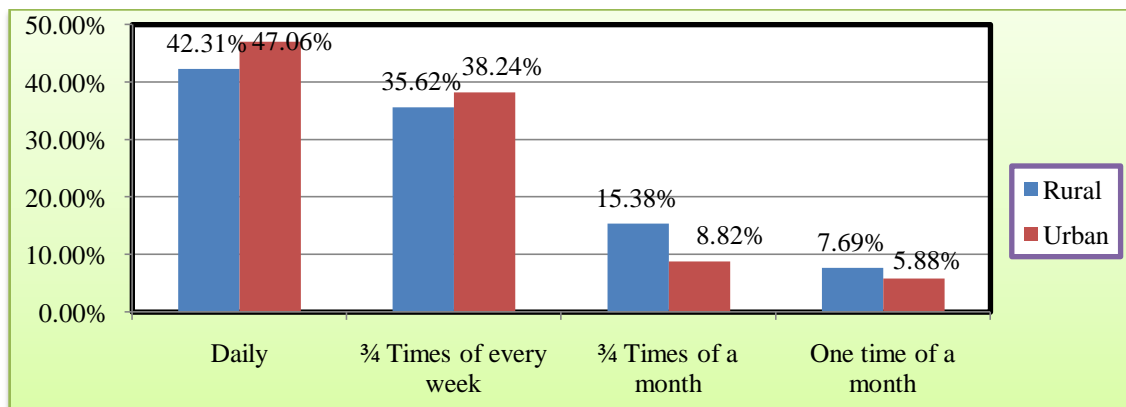


Figure 2:- Percentage of using internet by rural & urban college students.

Table- 3 shows area wise using internet. It's noted that 77.93 percentages rural college students using internet starting from daily to $\frac{3}{4}$ times of every week. Whereas 85.3 percentages urban college students using internet starting from daily to $\frac{3}{4}$ times of every week. On the opposite hand it's noted that 23.07 percentages rural college students using internet starting from $\frac{3}{4}$ times of a month to at least one time of a month compared to 14.7 percentages of urban college students.

Objective-3: To compare between male and female students in respect of their using internet.

Table 4:- Show internet using by male and female college students

Use Internet	Male	Female
Daily	90/150 (60%)	40/150 (26.67%)
$\frac{3}{4}$ Times of every week	30/150 (20%)	70/150 (46.67%)
$\frac{3}{4}$ Times of a month	20/150 (13.33%)	30/150 (20%)
One time of a month	10/150 (6.67%)	10/150 (6.67%)

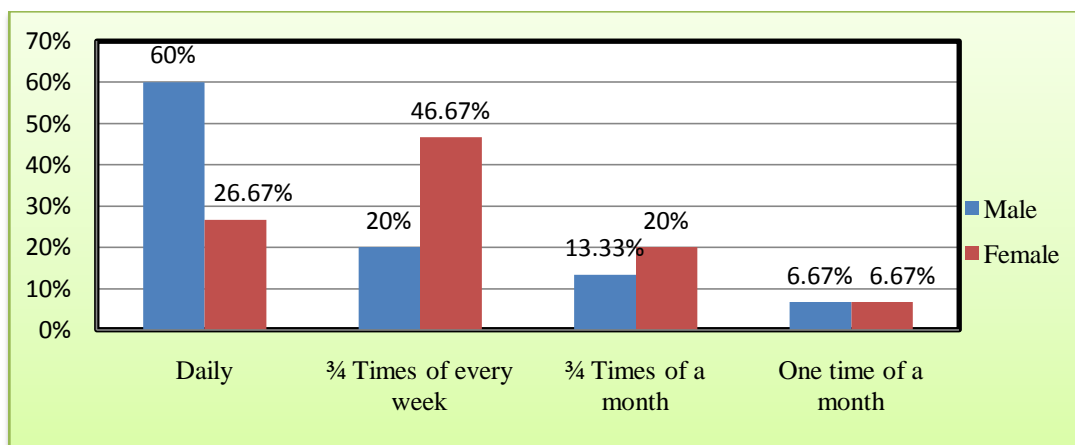


Figure 3:- Percentage of using internet by male & female college students

Table- 4 shows gender wise using internet. It's noted that 80 percentages male college students using internet ranging from daily to $\frac{3}{4}$ times of every week. Whereas 73.34 percentages female college students using internet ranging from daily to $\frac{3}{4}$ times of every week. On the other side it's noted that 20 percentages male college students using internet ranging from $\frac{3}{4}$ times of a month to at least one time of a month compared to 26.67 percentages of female college students.

Objective 4:- To compare between arts, science students in respect of their using internet.

Table 5:- Show using internet by Arts and Science college students

Use Internet	Arts	Science
Daily	90/200 (45%)	70/100 (70%)
$\frac{3}{4}$ Times of every week	70/200 (35%)	20/100 (20%)
$\frac{3}{4}$ Times of a month	35/200 (17.05)	10/100 (10%)
One time of a month	10/200 (5%)	00/100 (0%)

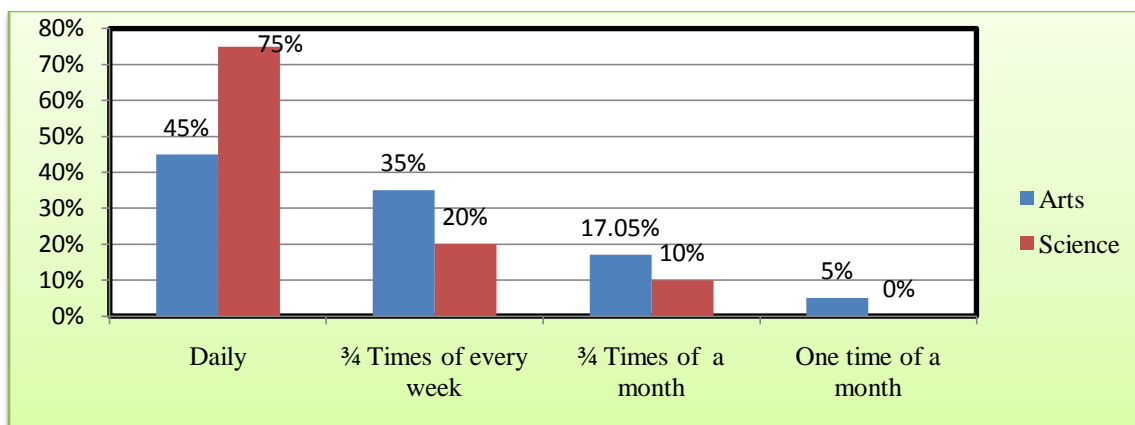


Figure 4:- Percentage of using internet by arts & science college students.

Table- 5 shows stream (Arts & Science) wise using internet. It's noted that 80 percentages arts college students using internet ranging from daily to $\frac{3}{4}$ times of every week. Whereas 90% percentages science college students using internet ranging from daily to $\frac{3}{4}$ times of a week. On the other side it's noted that 22.05 percentages arts college students using internet ranging from $\frac{3}{4}$ times of a month to at least one time of a month compared to 10 percentages of science college students.

Objective 5:- To Compare between Hindu and Muslim college students in respect of their using internet.

Table 6:- Show using internet by Hindu and Muslim college students

Use Internet	Hindu	Muslim
Daily	140/260 (53.85%)	10/40 (25%)
$\frac{3}{4}$ Times of every week	60/260 (23.08%)	20/40 (50%)
$\frac{3}{4}$ Times of a month	40/260 (15.38%)	5/40 (12.5%)
One time of a month	20/260 (7.69%)	5/40 (12.5%)

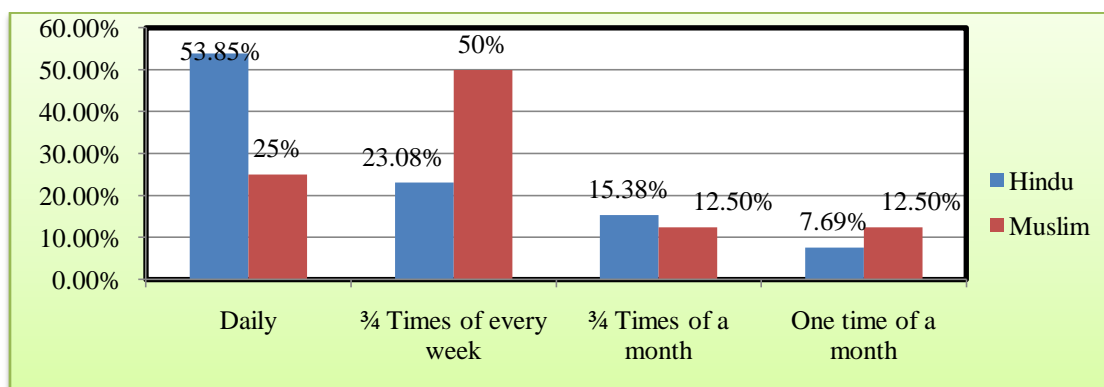


Figure 5:- Percentages of using internet by Hindu & Muslim college students.

Table- 6 shows religious (Hindu & Muslim) wise using internet. It's noted that 76.93 percentages Hindu college students using internet ranging from daily to $\frac{3}{4}$ times of every week. Whereas 75 percentages Muslim college students using internet ranging from daily to $\frac{3}{4}$ times of every week. On the other hand it's noted that 23.07 percentages Hindu college students using internet ranging from $\frac{3}{4}$ times of a month to at least one time of a month compared to 25 percentages of Muslim college students.

Objective 6:- To know the purpose of internet use.

Table 7:- purpose of internet using by college students

Purpose of Internet Use	Total
Educational Information	62/300 (20.67%)
Social Media and News	150/300 (50%)
Audios and Videos	38/300 (12.67%)
Gaming	30/300 (10%)
Other	20/300 (6.67%)

From the table-7 it's clear that the college students used internet primarily social media and news (50%) followed by educational information (20.67%), audios and videos (12.67%), gaming (10%) and a small number of students used internet for other purpose like online shopping or searching (6.67%).

Conclusion:-

Present study indicates that hundred percent college students use internet starting from daily to at least one time of a month.

It is conjointly found that urban college students use internet over rural college students in terms of daily or $\frac{3}{4}$ times of every week as calculated criteria. However, on the opposite hand, it's noted that rural college students are exploitation internet over the urban college students in terms of $\frac{3}{4}$ times of a month or just one occasion of a month. After analysis, it's conjointly found that male college students use internet over the feminine college students in terms of daily or $\frac{3}{4}$ times of every week. On the opposite facet, feminine college students use internet over male college students in terms of $\frac{3}{4}$ times of a month or just one occasion of a month.

The present study indicates that Science college students use internet over the arts college students in terms of daily or $\frac{3}{4}$ times of every week. On the other hand humanities college students use internet over science counterpart in terms of $\frac{3}{4}$ times of a month or just one occasion of a month as a calculated criteria.

Present study conjointly indicates that Hindu college students use internet over Muslim college students in terms of daily or $\frac{3}{4}$ times of every week. However on the opposite facet, it's conjointly found that Muslim college students use internet over Hindu college students in terms of $\frac{3}{4}$ times of a month or just one occasion of a month as calculation criteria.

Most of students use internet for social media, news and 20.67% students using internet for educational information.

On the idea of this analysis work government ought to organized better internet facility in rural and concrete areas and intended the college or school students to most exploitation internet for the aim of education. The internet has emerged because the single most powerful medium for providing access to unlimited data. The internet is associate indivisible a part of today's academic system. The internet facility has enabled the lecturers and also the students to reinforce their educational excellence by providing them the foremost up-to-date data and access to the worldwide data.

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RESEARCH ARTICLE

KEYWORD EXTRACTION: A COMPARATIVE STUDY USING GRAPH BASED MODEL AND RAKE.

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Abstract

In this paper we introduce Rapid Automatic Keyword extraction an unsupervised, domain independent and language independent method for extracting keywords from individual documents and compare this model with a graph based ranking algorithm (TextRank). In general TextRank consists of two unsupervised methods for both keyword and sentence extraction. Also we conduct a simple study regarding TextRank with the previously published methods.

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Introduction:-

Keyword extraction (KE) is termed as the process that automatically identifies a set of elements that best matches with the subject of document. To represent most relevant information contained in the document: key phrases, key segments, key terms or just keywords different approaches were used. Keyword extraction connects different areas of text mining, information retrieval and natural language processing. The process of keyword extraction can be classified into two categories; quantitative and qualitative.

Quantitative techniques are based on statistical relations in addition to formal linguistic processing. Methods like frequency, TF*IDF, co-occurrence are used as word statistics. The basic idea behind this approach is that important terms are most often referenced within the text. But sometimes the most frequent terms are not enough to represent a meaningful keyword. Certainly further modifications are needed for this approach.

Qualitative methods are based on semantic relations and analysis. Semantic analysis relies on semantic description of lexical terms. Such kind of extraction provides highly structured conceptual relations to the content of the text. This method is more reliable than quantitative approach because sometimes it is not compulsory to appear the most important keyword as a frequent item.

This paper presents a new extraction model RAKE operates on individual documents as well as multiple types of documents. The method starts with the selection of candidate keywords and a graph of word co-occurrences is constructed. After that a score is calculated for each candidate keyword. Rather than that we include a discussion on an unsupervised keyword extraction.

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Extraction Methods:-

Keyword assignment tasks can be divided into two subclasses: (1) Keyword assignment and (2) keyword extraction, both will finally results a set of keywords. In keyword assignment, keywords are chosen from a predefined class of terms, keyword extraction searches a document with keywords that are explicitly contained in the document.

According to Zahang, automatic keyword extraction methods can be classified into four.

1. Simple statistical approaches
2. Linguistics approaches
3. Machine learning approaches
4. Other approaches

Simple Statistics Approaches:-

Which is the simplest method of keyword extraction, which do not require training data. In addition such methods are language and domain independent. The disadvantage is that in information regarding health and medical, the most important keyword may appear only once.

Linguistic Approaches:-

In such types of methods which pay attention to linguistic features such as parts of speech, syntactic structure and semantic qualities tend to add value, Functioning sometimes are filters for bad words.

Machine Learning Approaches:-

This type of keyword extraction can be seen as supervised learning. In machine learning approach the keywords are extracted from training documents. This approach includes Naïve Bayes, Support vector machines.

Other Approaches:-

Other approaches combines the above mentioned methods with some heuristic knowledge such as position length, etc...

TextRank:-

TextRank is a graph based ranking model that can be used in text processing for the fruitfulness of natural language applications. It consist of 2 unsupervised methods for keyword and sentence extraction.

1. Graph based – Kleinberg's HITS Algorithm
2. Ranking Algorithm Google's PageRank

Let $G = (V, E)$ be a directed graph with a set of V and set of edges E , where E is a subset of $V \times V$. $In(V_i)$ be the set of vertices that point to it and $Out(V_i)$ be the set of vertices that V_i points to. The score of a vertex is,

$$S(V_i) = (1 - d) + d * \sum_{j \in In(V_i)} \frac{1}{|Out(V_j)|} S(V_j)$$

d is a damping factor and its value within the range 0 and 1. During the time of Web surfing, TextRank implements the "random surfer model". When a user clicks on a link with a probability 'd' and the probability of jumping to a new page is 1-d. The value of d usually is 0.85. Starting from any arbitrary node, the computation iterates until convergence below a given threshold is achieved.

Graph Representation of text:-

Depending on the context, text units of various sizes / characteristics can be added as vertices. The important steps followed in the ranking algorithm can be as follows.

1. Identify text units, and add them as vertices in the graph.
2. Identify relationships between these text units and by using these relationships draw the edges
3. Iterate until convergence.
4. Score vertices based on their final score.

Text Rank Keyword Extraction is fully unsupervised. First the text is tokenized and decorated with parts of speech tags. We consider only single words as candidates. Now an edge is added those lexical units that co-occur within a window of n words. After the graph constructed, the initial score of all vertices are set to an initial value of 1, and the above ranking algorithm is applied for several iterations until it converges usually 20-30 iterations and a

threshold of 0.0001. Once the final score is obtained for each vertex in the, vertices are sorted in reverse order of their score, and the top T vertices in the ranking are retained for post processing. T can be set to any fixed values usually ranging from 5 to 20 keywords.

Rake:-

The input parameter of RAKE are a stop list, set of phrase delimiters, set of word delimiters. By using phrase and word delimiters the doc. Is partitioned into candidate keywords. To identify co-occurrences of words within this candidate keywords, no arbitrarily sized sliding window is needed.

E.g.: Compatibility of systems of linear constraints over the set of natural numbers.

Manually assigned keywords:

Linear constraints, set of natural numbers

Candidate keywords parsed:

Compatibility – systems - linear constraints – set - natural number

After identifying every candidate keyword a graph of word co-occurrences is build and a score is evaluated for each candidate keyword. For this several metrics were used. (1) Word frequency, $\text{freq}(w)$, (2) word degree ($\text{deg}(w)$) (3) ratio of degree to frequency ($\text{deg}(w) / \text{freq}(w)$)

After candidate keywords are scored, the top T scoring candidates are selected. We assume T as one-third the number of words in the graph as in Mihalcea and Tarau.

RAKE does not require a training set. TextRank applies syntactic fillers to a document text to identify content words and accumulates a graph of word co-occurrences in a window size of 2. A rank for each word in the graph is calculated through a series of iterations until convergence below a threshold is achieved.

TextRank damping factor 0.85.

Threshold 0.0001

Not have access to the syntactic filters

Advantages:-

- Total time needed for TextRank to extract keywords compared to RAKE is over to times the time of RAKE.
- RAKE If the number of content words increases, the performance also increases because it can score keywords in a single pass whereas TextRank requires repeated iterations to achieve convergence on word ranks.

Related Works:-

According to Rada Mihalcea Dept of CS University of North Texas[1] ;their paper presents unsupervised method for automatic sentence extraction using graph based ranking algorithm in the context of text summarization task. They discussed a set of graph based Ranking algorithms.

HITS (Hyperlinked Induced Topical Search) Kileinberg 1999

Iterative algorithm for ranking web pages according to their degree of authority

Positional power function:-

Introduced by (Herings et al 2001) that determines the score of a vertex as a fun that combines both the number of its successors and the score of its successors.

1. Page Rank used for Web link analysis
2. Weighted graphs

Content of web surfing and citation analysis. TextRank model the graphs are build from natural language texts and may include multiple / partial links between the units that are extracted from text. Consequently, we introduce new formulae for graph based ranking that take into account edge weights when computing the scores associated with the vertex.

TextRank sentence extraction algorithm is evaluated in the context of a single document summarization task using 567 news articles provided during the document understanding evaluations 2002.

Martic Dostal and Karel Jezek [2] conducted a study on automatic keyword extraction based on NLP and statistical methods. Key phrase candidates are extracted by a combination of graph methods (TextRank) and statistical methods (TF * IDF). During the text preprocessing phase, the content is partitioned into meaningful tokens and non-significant characters are removed. The remaining tokens are considered as candidate keywords. Then calculate the TF * IDF score only for these candidate keywords.

Feifan Liu, Deana Pennel, Fei Liu and Yang Liu [3] approaches for automatic keyword extraction using meeting transcripts. The paper suggest that the important part of keyword extraction is to assign a score to a word depending on its importance. Also the article compare different methods for weight calculation the TF IDF framework and the graph model.

TF – IDF FRAMEWORK:-

Basic TF IDF weighting:-

The TF (term frequency) for a word w , in a doc is the no of times the word occurs. The IDF value is $IDF I z \log (N / N_i)$ where N_i denotes the no of documents containing word w_i , and N is the total number of documents.

Parts of speech filtering:-

In addition to using a stop word list to remove words from consideration we also leverage POS information to filter unlikely keywords. Only verbs, nouns and adjectives are likely to be keywords.

Integrating word clustering:-

By using SRILM toolkit words with the same semantic meaning can be identified.

Guangyi Li, Houfeng Wang [4] suggest an improved automatic keyword extraction based on textrank using domain knowledge. They focused keyword extraction for Chinese scientific articles, they used a framework for selecting candidate keywords by Document Frequency Accessor Variety (DF AV) and a TextRank algorithm to improve the performance of keyword extraction, they considered keywords for a specific domain.

Automatic keyword extraction from documents using conditional random fields. (Journal of Computational Information Systems 2008) [5] is another work published on a journal. In this paper keyword extraction based on a CRF model. Conditional Random Fields (CRF) is a state of the art sequence labeling method also their implementation results guarantee that the CRF model is more better than support vector machine, multiple linear regression model etc.

Conclusion:-

In this paper we have proposed and compared two keyword extraction methods: TextRank and RAKE. RAKE gathers high performance when compared with existing keyword extraction techniques. RAKE automatically extracts keywords in a single pass. Also RAKE is high simple and efficient. But TextRank can be used for large number of natural language applications. TextRank consists of two unsupervised method for keyword and sentence extraction. Advantages of TextRank is that it doesn't demand linguistic and domain knowledge.

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RESEARCH ARTICLE

INTRAUTERINE INSEMINATION OF SEMINAL PLASMA AT THE TIME OF OVUM PICKUP: RELATIONSHIP WITH SUCCESSFUL OUTCOME.

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Abstract

There is a debate about the role of seminal plasma for improved implantation rate and pregnancy rate during in vitro fertilization.

Objective:- The aim of this study is to investigate whether the intrauterine application of SP at the time of the ovum pick-up has any influence on the pregnancy rates in IVF or not.

Methods:- This prospective double-blind placebo-controlled randomized study

Was conducted to compare two groups. The first group represents the cases whom seminal plasma insemination was done and the other group no plasma insemination but placebo. Clinical pregnancy rate and implantation rate were the outcomes.

Results:- 400 patients were randomized after fulfilling the inclusion criteria. 200 patients were included in the study group and 200 patients were included in the control group. Our results found no statistically significant difference in the two groups regarding implantation rate or pregnancy rate (57.3% versus 49.7%, p value=0.39) and (39.2% versus 31.9%, p value=0.69).

Conclusion:- Intrauterine insemination of seminal fluid at the time of ovum pickup is unlikely to increase the pregnancy rate....

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Introduction:-

Although the gradual increase in the success of reproduction, over the years, many couples also had been left frustrated following repeated failed attempts. Only 20 to 25 % of the transferred embryos lead to a pregnancy. Endometrial function is a limiting factor of the pregnancy[1]. This percentage cannot be further increased by the improvement of the embryo transfer technique and culture conditions or by an optimal selection of embryos, the endometrium is receptive to implantation in the mid-luteal phase during the so-called "implantation window". Its duration is determined by the sex hormones which regulate the expression of several cytokines., endometrial function and receptivity have been accepted to be the major limiting factors of IVF pregnancy rate[2]. Human endometrium is a complex, multicellular tissue that is regulated by steroid hormones (estrogens, progesterone, androgens and glucocorticoids) and has different characteristics in the various phases of the menstrual cycle. These changes include restructuring of the cellular architecture, expression of specific cell-surface molecules as well as a secretion of biologically active factors such as cytokines, chemokines and growth factors. The process of the regeneration of the endometrium is currently viewed as a process of cell proliferation and a consequent differentiation of endometrial multipotent stem cells[3]. The presence and the characterization of multipotent stromal stem cells, epithelial progenitor cells and endothelial progenitor cells in the human endometrium and decidua have been reported by a number of research groups. These cells reside in both basal and functional layers of the endometrium and can be identified and isolated even from menstrual blood. It was recently demonstrated that endometrial stem/progenitor cells can induce proliferation and this report substantiates the hypothesis on the role of the stem cells in endometrial regeneration[4]. The indisputable biological purpose of the endometrium is to secure the successful development of pregnancy. Embryo implantation is only possible for a short period of time when the hostile uterine lining transforms to a hospitable surface to accept the embryo. The newly acquired capacity of the endometrium to welcome the embryo is termed "endometrial receptivity" and it is viewed as a dynamic process of genotypic and phenotypic changes of the endometrial cells. The result is that they are capable of participating in two-way cross-talk with the embryo which may or may not lead to successful opposition, attachment, penetration and implantation and possibly development and growth of a viable conceptus[5,3]. The molecular mechanisms behind this complex and sophisticated process have been studied using animal models and knock-out (KO) mouse studies have positively identified genes for receptivity (leukemia inhibitory factor-LIF, Homeobox protein X3), responses to the embryo (Cyclooxygenase 2-COX 2) and decidualization (Interleukin 11 Receptor-IL-11R). Additional information has derived from in vitro studies with human endometrial cells and explants cultures, human trophoblasts, and placental explant cultures. Members of the cell adhesion molecules family (integrins, etc.) are expressed on the surface of the epithelial cells during the WOI in humans. Extensive studies are being currently carried out on the timed restricted expression of a number of molecules such as mucin (MUC-1), trophinin, L-selectin, Wingless (Wnt) family members, etc. in reference to the possibility of using them as biomarkers for endometrial receptivity[2]. In addition to exploring the value of endometrial secretion analysis, N. Macklon has employed a human co-culture model, consisting of decidualized endometrial stromal cells and single hatched blastocysts to identify the soluble factors involved in implantation and to correlate these to embryo development. The cytokines and chemokines produced and secreted by the endometrial cells have been discussed in an extensive review. It is pointed out that numerous cytokines such as IL-11, LIF, IL-15, IL-1 and members of the superfamily of the transforming growth factor (TGF) are important factors in establishing the optimal interactions between the embryo and the endometrium[6]. COX-2 (cyclooxygenase-2), EGF (epidermal growth factor) and LIF (leukemia inhibiting factor) Schematic diagram of the implantation process. Exposure to seminal plasma (SP), the fluid component of ejaculate produced by seminal vesicles, may play a beneficial role in implantation[2].

SP, which contains paternal alloantigens and high concentrations of cytokines, growth factors and prostaglandins, induce a state of active immune tolerance, essential for the embryo to be implanted. Animals that become pregnant through artificial insemination or embryo transfer without being exposed to SP have substantially lower rates of implantation than those exposed to SP[7]. Excision of the seminal vesicle glands from males diminishes the tolerance-inducing effect of mating, while the vasectomy to remove sperms from the seminal fluid does not substantially impact the response[8]. These findings have led to the conclusion that the use of SP or its ingredients may stimulate endometrium in IVF cycles in order to improve implantation rates. Several studies have investigated the role of SP in implantation with controversial results, some reporting benefits, whereas others showing no effect[5,3,8]. The purpose of this study is to investigate whether intrauterine application of SP at the time of the ovum pick-up has any influence on the pregnancy rates in IVF or not.

Subjects and Methods:-

This a randomized,-controlled, a double-blind clinical study investigating superiority of Seminal Plasma administration at the time of oocyte pick-up.

Setting;

MIRU,miniauniversity,MIVF.

All couples undergoing IVF or ICSI treatment between January 2010 and December 2013 were considered for enrolment in the trial.Of these, 400 women were eligible for enrolment and consented to participate in the study. Participation was limited to one treatment cycle per couple.Patients were included and randomized at the first consultation in our center, before the start of the treatment. Concealed allocation by computerized randomization. Eligible patients were categorized into two groups. Group A who inseminated by seminal plasma and Group B (placebo group).

The exclusion criteria included the presence of hepatitis B, C, leukocytopenia or other signs of infection and men with <300 µl of SP.

The study was approved by the local ethical committee and each patient's approval was given by written consent. Stimulation was performed using the long agonist protocol, pituitary down-regulation was done using GnRH agonist 0.1 IU Leuprolide acetate S.C. (Leucine, Abbviepharmaceutical) starting in the luteal phase of the previous cycle. After down-regulation of the pituitary gland, patients received in addition either 150–300 IU of recombinant FSH (GonalFw, Merck Serono) or highly purified HMG (Fostimon, IBSA pharmaceutical, Switzerland) per day.Once an adequate ovarian response had been confirmed, 10 000 IU of urinary hCG (Choriomon, IBA pharmaceutical, Switzerland) was used to trigger ovulation. Ovum pick up was done 34–36 h after hCGadministration under sedation. Fertilization was achieved by ICSI. Day 5 grade 1 embryos were transferred back into the uterus. Luteal support was done using vaginal progesterone (Prontogest 400 bid, IBSA pharmaceutical, Switzerland).

Preparation and application of SP:-

Semen samples were obtained before follicle aspiration by masturbation from the patient's partner and were collected in sterile flasks. SP was extracted by centrifugation of ejaculates at 600 g for 15 min. The second round of centrifugation was performed at 10 000 g for 15 min to extract all spermatozoa.

SP was analyzed several times to exclude remaining contamination with spermatozoa. None of the samples contained any spermatozoa after centrifugation.

Then 400 µl of supernatant were diluted with 1600 µl of sterile sodium chloride and mixed. From this solution, 1500 µl was stored in 2 ml syringes at –20°C (serum). For the placebo, 1500 µl of sterile sodium chloride was also stored in 2 ml syringes at –20°C.

Blinding of the samples was achieved by using a placebo (sodium chloride), which could not be differentiated from SP due to the same optical appearance.

An insemination catheter was introduced just at the end of the cervix and 1500 µl SP or placebo was injected.

Statistical Analysis:-

Data were analyzed using Statistical Package for the Social Sciences (SPSS), Version 19. Quantitative data were presented as a range, mean and standard deviation, qualitative data presented as a frequency distribution. Chi-square test, independent sample t-test, ANOVA, and correlation test were used. The probability of less than 0.05 was used as a cut-off point for all significant tests.

Results:-

Table I shows the demographic criteria of the two groups.400 participants were similar in their characteristics so there is no statistical difference between the study and the placebo groups.

	Group (A)	Group (B)
Mean Age (Years)	31 (23.2: 40.1)	32 .1 (21.2: 40.9)
Cause of infertility		
• Male Factor	52	63
• Tubal Factor	30	17
• Endometriosis	67	43
• PCO	18	57
• Unexplained	33	20
Type of infertility		
• Primary	113	141
• Secondary	87	69
No. of ICSI failure	2.3 (1-3)	1.7 (0-2)

Table 2 shows outcomes and embryological data of both groups. There is no statistical difference in the pregnancy rate in the two groups. Also, the number of embryos transferred were similar in both groups so there is no statistical difference between the study and the placebo groups.

	Group (A)	Group (B)	P-Value
Median No. of Transferred embryos	2.71 (1:3)	2.89 (1:4)	-
Clinical Pregnancy Rate	39.2%	31.9%	0.69 NS
Implantation rate	57.4%	49.7%	0.39 NS

Discussion:-

This study disputes the hypothesis that intrauterine instillation of SP at the time of oocyte retrieval increases pregnancy rate. We found that there is no difference in the pregnancy rate between the two groups (39.2% in group A versus 31.9% in group B p=0.69) which is statistically insignificant. Also, we found that there is no difference in the implantation rate between the two groups (57.4% in group A versus 49.7% in group B p=0.39) which is statistically insignificant. In agreement with us, Qasim who found that application of SP in the vagina during intrauterine insemination does not improve the pregnancy rate [9]. Also, the same results found by Fishel who declare that there are no differences in the pregnancy rates between the two groups (32 % versus 33 % and 21 % versus 17 %) [10]. Similarly, VonWolff applied thawed SP at the time of OPU during IVF-ICSI cycles and found a non-significant increase in the pregnancy rate (37.3 % versus 25.7 %) [11]. In contrast, Bellingue who deposited semen in the vagina of patients undergoing IVF at the time of the oocyte fertilization and found an implantation rate of 53 %, compared with 23 % in the control group [12]. Also, Coulam and Stern performed a placebo-controlled clinical trial, depositing into the vagina capsules containing SP or placebo. They described an implantation rate of 80 % in the SP group, compared with 67 % in the placebo group [13]. Therefore, the previous studies are diverse and difficult to compare.

These different results of the previous studies could be explained by the followings

Firstly, different sites of insemination whether cervical, vaginal or intrauterine. In the study published in 2009, pure SP was installed into the cervix, which is very close to the physiological situation during sexual intercourse. In this study, SP was injected into the uterine cavity, which is nonphysiological. Gutsche et al. found that seminal plasma induces endometrial inflammatory responses only *in vitro* in human [3], whereas cervical inflammatory responses

following exposure to ejaculate have been demonstrated in vivo [8]. Also, in vitro experiments demonstrated that this effect is due to SP and not due to semen [14]. Second, trauma induced by the catheter while introduced through the cervix into the uterus may increase the pregnancy rate without any pathophysiological effect. This matches several studies that have demonstrated an increased pregnancy rate after the endometrial injury before IVF treatment (Nastri et al., 2012). This effect was not seen after endometrial injury at the time of follicle puncture.

However, as the introduction of a transfer catheter possibly also causes some cervical trauma, it might be speculated that this would have a positive effect in our new study, although in reality such an effect was not observed. Finally, there is the possibility that intracavitary treatment with SP does not have any clinically relevant immunomodulatory effects on the endometrium at all or that the dilution of SP reduced the effect to such an extent that it was not pronounced enough to have a substantial effect on endometrial function. Indeed, the present study suggests that SP installation into the uterine cavity is unlikely to increase the chance of clinical pregnancy, at least not by a rate $\geq 14\%$ as initially hypothesized.

In conclusion besides the quality of the embryo, endometrial receptivity plays an important role in the establishment of pregnancy. Endometrial functional should be considered of great relevance in a physiological context, e.g. spontaneous conception in a natural cycle and implantation in assisted conception. Finally, there is the possibility that insemination of SP does not have any clinically relevant immunomodulatory effects on the endometrium at all, or have a substantial effect on endometrial function. Intrauterine instillation of diluted SP at the time of ovum pick-up is unlikely to increase the pregnancy rate.

In conclusion, the results of this study cast doubt on the validity of the concept that SP increases endometrial receptivity and thus implantation in humans.

Clinically relevant differences cannot reliably be excluded based on this single study. Finally, the answer to the question about the role of seminal plasma insemination at the time of OPU is still unclear, therefore further studies are still needed, with a larger number of patients; this will further justify the exact place of IU insemination of SP in modifying the endometrial function.

Conflict of interest:-

We declare no conflict of interest.

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RESEARCH ARTICLE

PREGNANCY OUTCOME IN SHORT CERVIX: PROGESTERONE VS CERVICAL ENCLERCLAGE.

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Key words:-

Cervical Enclerclage, Pre-term Birth,
Progesterone, Short cervix.

Abstract

Introduction: Preterm birth (PTB), defined as birth before 37 weeks of gestation, is the leading cause of perinatal morbidity and mortality. A sonographic short cervix has emerged as a powerful predictor of preterm birth. The present study was conducted to compare the outcome of pregnancy with short cervix with natural micronized progesterone and cervical cerclage.

Materials & Methods: A Prospective Comparative study was conducted in the Department of Obstetrics and Gynaecology of a Tertiary Care Hospital and Medical college. A total of 50 cases of short cervix were included in the study. Out of 50 cases, 25 cases each were divided in two groups by simple random sampling: **Group A:** Given natural micronized progesterone; and **Group B:** Underwent cerclage procedure. Detailed history, ANC check-up, USG and appropriate investigations were carried out for each subject. The cases were followed thereafter in antenatal clinic till delivery and pregnancy outcome was noted.

Results: The mean maternal age was 22.44 years in progesterone group and 22.64 years in cerclage group ($p=0.514$). The outcome of pregnancy with natural micronised progesterone reflected 36% births between 28-32 weeks of gestation, 28% after completion of 37 weeks and only 16% before 28 weeks of gestation whereas outcome with cerclage 36% between 28-32 weeks, 44% after completion of 37 weeks and 8% before 28 weeks ($p=0.56$). The difference in secondary outcome measures (PPROM, LSCS rate) and neonatal outcome (mean birth weight, APGAR, duration of NICU admission, respiratory distress, development of IVH, NEC and neonatal sepsis) in cerclage and progesterone group was also statistically non-significant ($p>0.05$).

Conclusion: Natural micronized progesterone is as effective as cervical cerclage in prevention of premature labour in women with short cervix. Use of natural micronized progesterone is more preferable in clinical practice because it is non-invasive technique, easy to administer and the patients do not suffer from surgical and anaesthesia procedure related adverse effects. It is also not associated with any hospital stay and is very economical.

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Introduction:-

Preterm birth, defined by the World Health Organization (WHO) as birth prior to 37 completed weeks (259 days) after the first day of the last menstrual period preceding the pregnancy,¹ is a major global public health problem.² Approximately 15 million neonates are born premature worldwide yearly, implying a rate of 1 out of 10 neonates and nearly 1 million children die each year due to complications of preterm birth.³ In developed countries the preterm birth rate is about 8% of all the pregnancies.³ Among the 10 countries with greatest number of preterm births India has the highest number. Preterm births accounted for 75.3% of all perinatal mortality with mortality risk of 82.6 per 1000 births.^{4,5}

A sonographic short cervix has emerged as a powerful predictor of preterm birth. It is unlikely that this condition is due to a single cause, and a multiple causation model of a sonographic short cervix has been proposed (e.g. a short cervix is syndromic in nature). Such model would have biological, diagnostic, prognostic and therapeutic implications. Indeed, patients may have a short cervix after DES exposure in utero, a cervical conization, a LEEP procedure, intrauterine infection/inflammation, a decline in progesterone action, and the challenging condition clinically referred to as idiopathic cervical insufficiency⁶.

Progesterone is considered a key hormone for pregnancy maintenance, and a decline of progesterone action is implicated in the onset of parturition. If such a decline occurs in the midtrimester, cervical shortening may occur, and this would predispose to preterm delivery.⁷⁻¹⁴ Various randomized clinical trials and data meta-analysis showed that vaginal progesterone decreases the rate of preterm delivery and neonatal morbidity/mortality in women with a sonographic short cervix¹⁵⁻¹⁷.

Cervical encerclage has also been widely used as a surgical method to prevent recurrent mid-trimester pregnancy loss in women at risk. Elective cerclage placement may benefit some women with proven cervical insufficiency. Although highly contentious, more recent data suggest that cervical encerclage may reduce the risk of preterm delivery in that subgroup of asymptomatic singleton pregnancies with both shortening on TVS and a history of spontaneous preterm birth¹⁸.

The Present study aimed at comparing the effects of micronized natural progesterone and cervical encerclage in pregnancy with short cervix.

Material & Methods:-

Type of Study & Study Area:-

A Prospective Comparative study was conducted in the Department of Obstetrics and Gynaecology of a Tertiary Care Hospital and Medical college. The study was conducted over a period of two years from August 2014 to August 2016. A written informed consent was obtained for this study from all subjects after obtaining approval of the ethical committee.

Inclusion Criteria:-

1. Gestational age between 18-28 weeks.
2. Singleton pregnancy.
3. Clinically short cervix 1.5-3 cm or sonographically short cervix or any sign suggestive of incompetent cervix.
4. Previous preterm delivery
5. Previous history of os tightening

Exclusion Criteria:-

1. Congenital anomaly
2. Multi fetal pregnancy
3. Extremely short cervix (<1.5cm)
4. Leaking per vaginally
5. Any major medical disorder (severe heart disease, severe pre-eclampsia, uncontrolled diabetes mellitus and patients in renal failure).
6. Placenta previa, intrauterine infection or inflammation

7. Non consented patients

Sample Size:-

A total of 50 cases of short cervix were included in the study. Out of 50 cases, 25 cases each were divided in two groups by simple random sampling:

Group A: Given natural micronized progesterone (200mg bd /300 mg SR per vaginally capsules of micronized progesterone); and

Group B: Underwent cerclage procedure.

Methodology:-

Study patients at the time of recruitment were subjected to:

1. Detailed clinical history
2. Clinical examination
3. Per abdomen examination
4. Per speculum examination
5. Per vaginal examination
6. Routine ANC profile
7. Ultrasonography

The cases were followed thereafter in antenatal clinic till delivery and outcome was noted. The primary outcome measures were preterm birth <32 weeks of gestation and composite perinatal morbidity and mortality (defined as the occurrence of any of the following events: respiratory distress syndrome, grade III/IV intraventricular haemorrhage, necrotizing enterocolitis, neonatal sepsis, bronchopulmonary dysplasia, or perinatal death).

Secondary outcome measures included preterm birth at <37, 34-37, and <28 weeks of gestation, respiratory distress syndrome, necrotizing enterocolitis, grade III/IV intraventricular hemorrhage, neonatal sepsis, bronchopulmonary dysplasia, perinatal mortality, a composite neonatal morbidity outcome (defined as the occurrence of any of the above mentioned neonatal morbidities), birth weight <1500 g and <2500 g, and admission to the neonatal intensive care unit (NICU).

Data Analysis:-

Data was analyzed using SPSS 21.0 (SPSS Inc., Chicago, IL, USA) using appropriate statistical tests.

Results:-

The mean maternal age was 22.44 years in progesterone group and 22.64 years in cerclage group (p=0.514). Maximum number of women, 28% of progesterone group and 44% of cerclage group, were noted between 18-20 weeks gestation while 20% of progesterone and 44% of cerclage were between 20-22 weeks (p=). Maximum number of women (44%) of progesterone group had cervical length of 1.5-2 cm whereas 44% of cerclage group had cervical length of 2.1-2.5cm (Table 1). The outcome of pregnancy with natural micronised progesterone reflected 36% births between 28-32 weeks of gestation, 28% after completion of 37 weeks and only 16% before 28 weeks of gestation whereas outcome with cerclage 36% between 28-32 weeks, 44% after completion of 37 weeks and 8% before 28 weeks (p=0.56; Table 2). The difference in secondary outcome measures (PPROM, LSCS rate) in both the group were also statistically non-significant (p=1.0; Table 3). Our study also compared neonatal outcome (mean birth weight, APGAR, duration of NICU admission, respiratory distress, development of IVH, NEC and neonatal sepsis) in cerclage and progesterone group. No statistically significant difference was observed between the groups regarding neonatal parameters (p>0.05; Table 4).

Discussion:-

The first randomized clinical trial to examine the effects of vaginal progesterone on the prevention of preterm birth in women with a short cervix was reported by da Fonseca et al.¹⁹ The primary outcome of the trial was the frequency of spontaneous preterm delivery at <34 weeks of gestation. Patients allocated to receive vaginal progesterone had a lower rate of preterm delivery (<34 weeks) than those in the placebo group [19.2% (24/125) vs. 34.4% (43/125)]. In another trial, termed as "PREGNANT" trial²⁰, It was estimated that 14 women with a cervical length between 10–20mm would need to be treated with vaginal progesterone to prevent one case of preterm birth before 33 weeks of

gestation. In addition, there was a significant decrease in the rate of preterm delivery <35 and <28 weeks of gestation. Since then, various trials and meta-analysis have shown the efficacy of vaginal progesterone to prevent preterm birth in cases with short cervix²¹⁻²⁹.

Cervical cerclage was introduced in 1955 by V. N. Shirodkar, Professor of Midwifery and Gynecology at the Grand Medical College in Bombay, India.³⁰ The procedure was developed in response to his observation that “some women abort repeatedly between the fourth and seventh months, and no amount of rest and treatment with hormones seemed to help them in retaining the product of conception.” Despite the 50 years that have elapsed since the introduction of cerclage as a procedure, there is conflicting evidence about its efficacy for standard indications (i.e. prophylactic) or for some patients with a sonographic short cervix. Several randomized clinical trials have been conducted to date which have yielded mixed results³¹⁻³⁸. A meta-analysis of randomized clinical trials of patients with a prior history of preterm birth and a short cervical length (<25mm) suggests that cervical cerclage is effective in reducing the rate of preterm birth and perinatal morbidity/mortality.¹⁵⁸ A different meta-analysis has suggested that women with a prior spontaneous preterm birth and singleton gestation may be monitored safely with transvaginal sonographic cervical length measurements³⁹.

In present study, we observed that both vaginal progesterone and cerclage in patients with a short cervix was associated with a significant reduction in the risk of preterm birth. The key finding is that both are equally effective for the prevention of preterm birth and adverse perinatal outcomes.

Very few studies has directly compared cervical cerclage and vaginal progesterone for the prevention of preterm birth in women with a sonographic short cervix. Most previous randomized trials allocated to receive vaginal progesterone versus placebo /no treatment, or cerclage versus no cerclage for the prevention of preterm birth.

Keeler et al.⁴⁰ compared between patients with short cervix on transvaginal ultrasound between 16 and 24 weeks' gestation treated with McDonald cerclage and those treated with weekly intramuscular injections of 17 alphahydroxyprogesterone caproate. Spontaneous Pre-term Birth (PTB) prior to 35 weeks' gestation occurred in 16/42 (38.1%) of the cerclage group and in 16/37 (43.2%) of the 17OHP-C group (relative risk, 1.14 95% CI, 0.67, 97 1.93). A post hoc analysis of patients with a prior PTB showed no difference in spontaneous PTB <35 weeks between groups. Conde-Agudelo and co-investigators performed an indirect comparison of vaginal progesterone versus cerclage, using placebo/no cerclage as the common comparator. Four studies evaluating vaginal progesterone versus placebo (158 patients) and five evaluating cerclage versus no cerclage (504 patients) were included. Both interventions were associated with a statistically significant reduction in the risk of preterm birth <32 weeks of gestation and composite perinatal morbidity and mortality compared with placebo/no cerclage. Adjusted indirect meta-analyses did not show statistically significant differences between vaginal progesterone and cerclage in reducing preterm birth or adverse perinatal outcomes⁴¹.

The strength of this study is that it is first study in this region to compare directly between vaginal progesterone and cervical cerclage. All previous studies compare either vaginal progesterone with placebo or cervical cerclage with placebo or conducted an indirect comparison. Our study compares two methods directly to be more practical and to avoid the hidden bias such as selection bias which affect indirect comparing.

Table 1:- Distribution of subjects based on baseline characteristics

Characteristic (n=50)	Progesterone group (n=25)	Cerclage group (n=25)	p- value
Maternal age (in years)	22.44± 1.91	22.64± 1.99	0.719
Parity	0.48±0.65	0.36± 0.63	0.514
Gestational age at start of treatment (weeks)	23.2±3.26	22.08±3.13	0.222
Cervical length (mm)	21.92±3.48	22.08±2.98	0.862

Table 2:- Distribution of subjects based on Primary Pregnancy Outcome

Pregnancy Outcome	Group		p- value
	Progesterone group (n=25)	Cerclage group (n=25)	
< 28 weeks	4 (16%)	2 (8%)	0.56
28-32 weeks	9 (36%)	9 (36%)	
33-37 weeks	5 (20%)	3 (12%)	
> 37 weeks	7 (28%)	11 (44%)	
Total	25 (100%)	25 (100%)	

Table 3:- Distribution of subjects based on Secondary Pregnancy Outcome

Pregnancy Outcome	Group		p- value
	Progesterone group (n=25)	Cerclage group (n=25)	
PPROM	1 (4%)	2 (8%)	1.0
LSCS	2 (8%)	1 (4%)	1.0

Table 4:- Distribution of subjects based on Neonatal Outcome

Outcome	Group		p- value
	Progesterone group (n=25)	Cerclage group (n=25)	
Birth weight (g)	2.1±0.18	2.1± 0.19	0.19
APGAR Score	9±0.91	9.4± 0.70	0.09
Days in NICU per admission	15.83±1.01	11±0.92	0.14
NICU admission	6 (24%)	3 (12%)	0.46
Intraventricular hemorrhage	1 (4%)	1 (4%)	1.00
Respiratory Distress	3 (12%)	2	0.67
Necrotizing Enterocolitis	1 (4%)	1 (4%)	1.00
Neonatal sepsis	1 (4%)	1 (4%)	1.00
Bronchopulmonary dysplasia	0	0	NA
Perinatal death	0	0	NA

Conclusion:-

The observations made in present study suggests that natural micronized progesterone is as effective as cervical cerclage in prevention of premature labour in a women with singleton pregnancy with short cervix. Use of natural micronized progesterone is more preferable in clinical practice because it is non-invasive technique, easy to administer and the patients do not suffer from surgical and anaesthesia procedure related adverse effects such as pain, headache, vomiting and other complications. It is also not associated with any hospital stay and is very economical. Using vaginal progesterone saves time for patients as well as doctors.

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Conflict Of Interest:-

None declared

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RESEARCH ARTICLE

RECOVERY OF URANIUM FROM SULFATE LEACH LIQUOR USING ETHANOLIC EXTRACTANT OF AERIAL PART OF *HELIANTHUS ANNUUS* PLANT.

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Abstract

The possibility of using *Helianthus annuus* extractant (sun flower) as a solvent extractant for uranium from its sulfate liquor as means for their recovery has been studied. Several experiments were conducted to determine the relevant factors affecting both the extraction and stripping of the uranium from a synthetic solution. Under the optimum conditions, the achieved uranium capacity has attained about 22 mg/g for the ethanolic extractant. The optimum stripping conditions for uranium content from the loaded ethanolic extractant which is rich of some active compound to form complex with uranium included stripping agent type 1M NaCl /1M HCl, aqueous to organic phase ratio (A/O) 1:3, stripping time 15 min and at room temp. From the prepared sulfate leach liquor of Abu Zeneima ore material (South Eastern Sainai, Egypt) uranium has then been recovered using the working ethanolic extractant under the previously studied optimum conditions, with an efficiency of 98% The obtained strip liquor was then subjected to precipitation using NH₄OH solution at pH of 7.5 and analysis of the obtained precipitate ammonium uranium oxide hydrate [UO₃NH₃H₂O] was found assay 76% uranium which has been confirmed using XRD and EDAX analysis.

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Introduction:-

Twenty one century, high lights are focused now upon the environmental concerns. One of the important concerns is the useful employment and recycling of the plant and animal remains, on their burning, cotton stalk, rice husk, sun flower stalk and plant cellulosic remains....etc. are essential pollutants to the atmosphere. Thus, the possibility of using the sunflower as extractants for the recovery of lanthanide could be considered as one of improvement of the using of waste management.

Perennial sunflower species are not as popular for gardens due to their tendency to spread rapidly and become invasive. (Subashini and Rakshitha 2012) evaluated the methanolic extract seeds of *Helianthus annuus* L. for the phytochemical present in the plant and results shown that, the carbohydrates, flavanoids, tannins, alkaloids, sapanins, phytosterols, steroids and fixed oils were present in the extract. (Macias et al. 2008).

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In nature, uranium can be found in the Earth's crust at an average concentration of about 2.5 mg/kg but anthropogenic activities, such as utilization of depleted uranium in munitions and nuclear accidents, introduce uranium to the environment on a larger scale. Uranium resulting from mining, reprocessing and disposal activities related to the nuclear industry is a problematic environmental pollutant. The monitoring of the movement of uranium from soil to plants especially to edible plant parts is very important due to possible contamination of the food chain. Uranium presents an exposure hazard due to its chemical toxicity as well as the radioactivity resulting from its decay and its decay products. In plants, uranium is stored mainly in roots, which was demonstrated on hydroponically cultivated sunflower (Dushenkov et al., 1997). Dushenkov et al., (1997) reported that sunflower plants were found to have a high affinity for uranium extraction and were selected for treatment of contaminated water.

Blanco et al. (2006), tested the linearity assumption of the validation of soil-to-plant transfer factors of uranium and Ra226 using *Helianthus annuus* L. grown in a hydroponic medium in addition, transfer of the studied species in both the aerial parts of plants and in the overall seedlings. The results show that the linearity assumption can be considered valid in the hydroponic growth of sunflowers for the radionuclides studied. The ability of sunflowers to translocate uranium and Ra226 was also investigated. In this sense, the removal percentages obtained for uranium and Ra226 were 24% and 42%, respectively. Practically all the uranium is accumulated in the roots. However, 86% of the Ra226 activity concentration in roots was translocated to the aerial part. Shahandeh and Hossner (2002), investigated uranium accumulation from uranium contaminated soil among sunflower plant species. They found also that, uranium accumulated mainly in the roots of plant species.

Generally uranium is influenced by its speciation and low pH conditions (Lauria et al., 2004). Uranium is present as a positively charged uranyl ion UO_2^{2+} in low pH samples and is very mobile U accumulation and distribution in plants has been reported by several authors (Straczek et al., 2010; Véra-Tome et al., 2008, 2009, Ebbs et al., 1998). Sunflower heavily accumulates U in its roots where it is stored (Straczek et al., 2010; Véra-Tome et al., 2008). Conversely, wheat shows low U transfer factors in roots and shoots (Shahandeh and Hossner, 2002). This differential response with respect to plant species is explained by their cation exchange capacity (Dufey et al., 2001; Straczek et al., 2010).

The present work aims to study using the sun flower hexane extractant from its waste by mixing synthetic solution of uranium for its recovery. The obtained optimum conditions were applied upon real sulfate leach liquor of gibbsite ore materials as a case study.

Materials and Methods:-

Materials:-

Plant materials:-

The aerial part of *Helianthus annuus* were collected on April (2013) and identified by Botany department, Faculty of Science, Zagazig University.

Extraction of the ethanolic extractant (HAE2):-

The aerial part are dried in the shadow in air draft were comminuted to powder (2 kg) and exhaustively extracted under reflux over a boiling water bath with 5 liters of ethanol(80%) for 3 hours. The extract was filtered; the process was repeated 3 times. The solvent was removed under reduced pressure at about 60 °C. The process yielded finally 150 g of a sticky dark brown material.

Synthetic solutions of REEs have been prepared from their proper salts. REEs solutions were thus prepared by dissolving analytical grade metal salts in distilled water to obtain accurate concentration 200 ppm (mg/L) solution. The pH of this solution was measured and adjusted at 0.1 using sulfuric acid and all the adsorption experiments were run at room temperature.

Experimental procedures:-

Preparation of standard U (VI) and interferences solutions:-

A synthetic uranium solution assaying 200 ppm has been prepared by dissolving the required weight of the uranyl acetate salt in distilled water that slightly acidified with sulfuric acid solution. The solution pH was adjusted at 4 using sulfuric acid solution. On the other hand, for studying the possible metals interference that might be associated

with uranium in its solutions, proper weights of some of their compounds have been dissolved in the prepared synthetic uranium.

Uranium extraction procedure from sulfate solution by HAE2:-

Uranium extraction from either the synthetic solution or that of Abu-Zeneima leach liquor solutions by the ethanolic extractant has been undertaken after its dilution/kerosene to 10 vol % were carried out by shaking the two phases in separating funnels. The aqueous phase was analyzed for its uranium content and that in the organic phase was calculated by the difference. Several series of experiments were carried out to study the effects of the diluent type, HAE2 concentration, pH, the shaking time, the extraction temp., the interfering metal ions and the O/A phase ratio. On the other hand, the practical saturation capacity of the HAE2 for uranium has also been determined by using the multiple contact technique.

Uranium stripping procedure from the loaded HAE2:-

A number of mineral acids, alkalis and NaCl solutions have been used for studying the uranium stripping efficiency from uranium loaded upon the HAE2. For choosing the eluent, both the shaking time and temperature together with O/A phase ratio have been studied. The resultant strip solution is treated in subsequent circuit to produce a metal concentrate while the stripped solvent is returned for cycle to extraction circuit.

Analytical procedures:-

Ethanolic extractant characterization:-

As mentioned above, HAE2 was analyzed for its acid content in the quality control laboratory of Hashem Brothers for Essential & Aromatic Products using the Gas Chromatography Mass Spectrometry (GC-MS) HP 6890 Series A (Agilent). On the other hand, an infra-red analysis has been achieved for the ethanolic extractant using (FTIR) model Thermo Scientific Nicolet IS10, Germany.

Control Analysis for Uranium:-

Control analysis of uranium in the different aqueous stream solutions as well as in the product has been undertaken by the oxidimetric titration after its reduction using a standard solution of ammonium metavanadate (Mathew, 2009).

Results and Discussion:-

Characteristics of the ethanolic extractant:-

I.R spectrum of the ethanolic extractant:-

From the achieved IR spectral analysis of the the ethanolic extractant shown in Table (1) and plotted in Fig.(1) before complexation with REEs it is clearly evident that a number of characteristics peaks have been obtained.

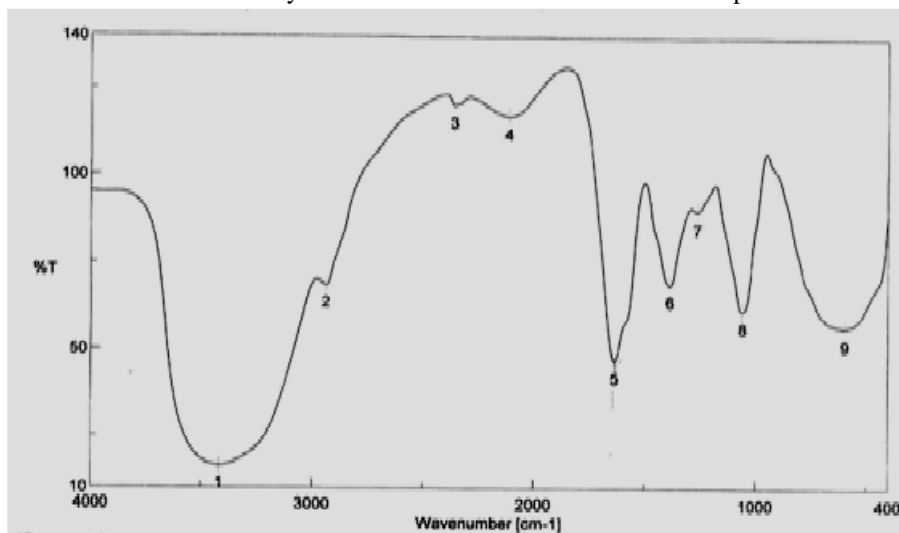


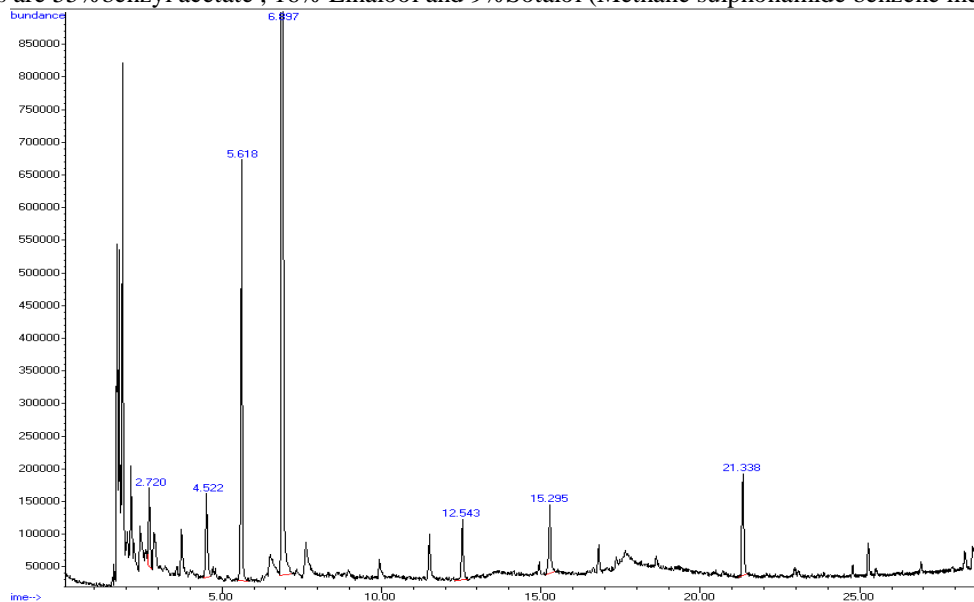
Fig. 1:- IR characteristic spectrum of the ethanolic extractant.

Table 1:- IR spectral analysis of the ethanolic extractant

Peak No	Spectra Value (cm ⁻¹)	Functional Group
1	3400	OH alcohol
2	2890	C-H
3	2350	X=C=Y (C,O,N,S)
4	2100	C=C
5	1750	C=O
6	1390	C-C
7	1250	C-N
8	1050	C-O

Chromatographic Analysis:-

The almost separated organic compounds were analyzed on Gas Chromatography Mass Spectrometry (GC-MSD). Retention time indices for all compounds were determined while, identification of the components was based on comparison of their mass spectra with those of internal (computer) library W9N11.L (Minimum Quality: 50) and NIST11.L (Minimum Quality: 50) libraries and some reference compounds. As shown in Fig.(2) the major components are 55% benzyl acetate, 16% Linalool and 9% Sotalol (Methane sulphonamide benzene methanol)

**Fig. 2:-** Gas chromatography-mass Spectrometry (GC-MS) of the ethanolic extractant.

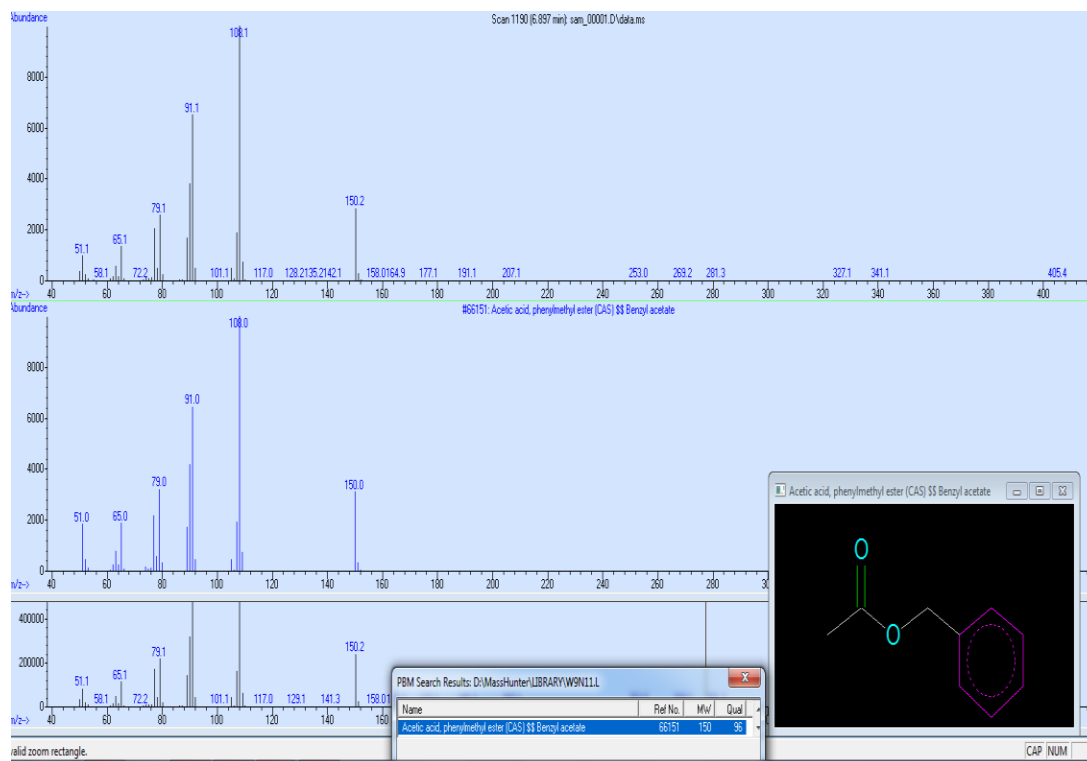


Fig 3:- Chemical structure of benzyl acetate.

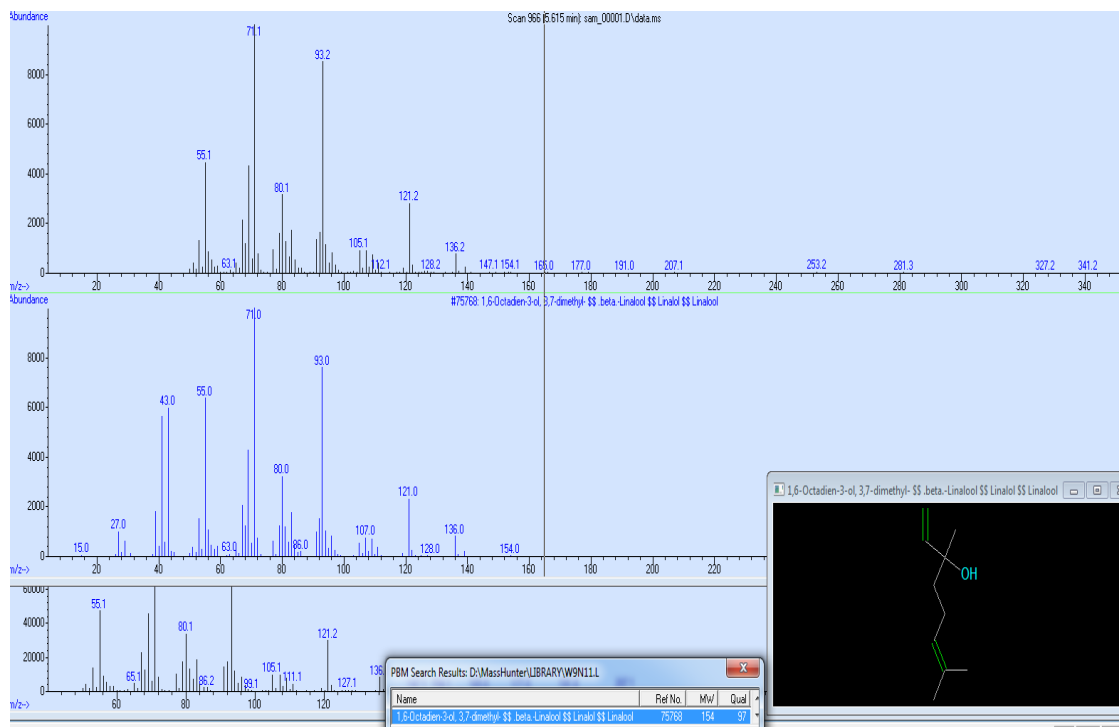


Fig 4:- Chemical structure of Linalool.

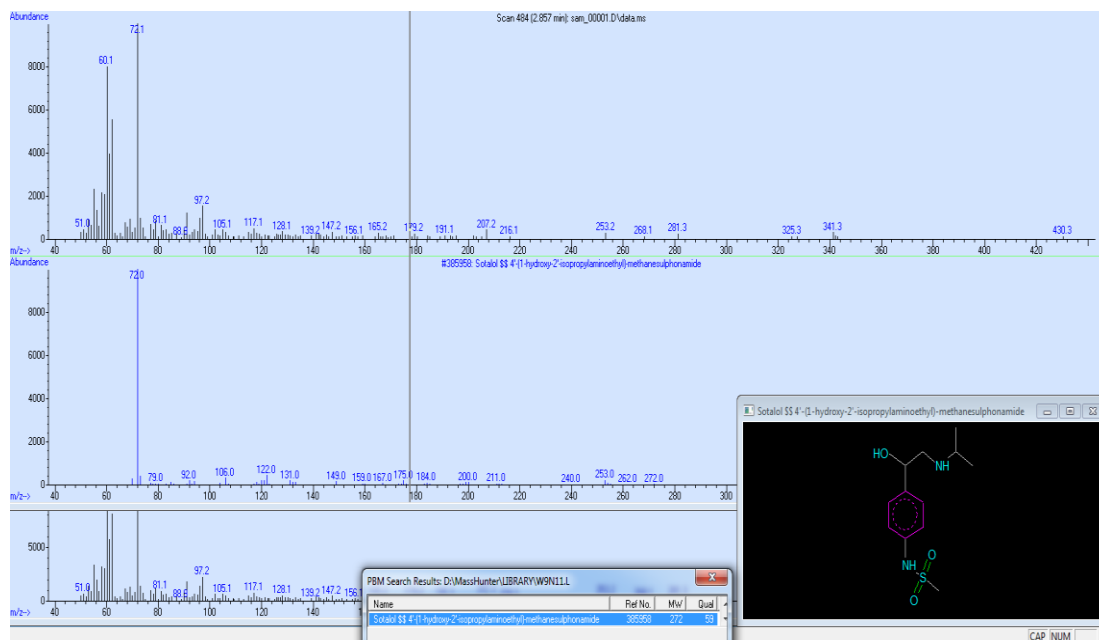


Fig 5:- Chemical structure of Sotalol.

Optimization of the uranium extraction:-

Effect of diluent type:-

To study the effect of the diluent type on uranium extraction from the prepared synthetic solution assaying 200 ppm of uranium three different organic diluents have been used namely; benzene, kerosene and carbon tetra chloride. The extraction process was performed under fixed conditions of a pH 4, a shaking time of 10 min. in an A/O ratio of 1:1 at room temperature and using 10 vol. % HAE. The obtained results indicate that kerosene is the best diluent in which 95% for uranium extraction has been achieved while the toluene and benzene diluents only 50% and 40% for uranium extraction were achieved respectively. Thus carbon tetra chloride was chosen as the best diluent in all extraction experiment.

Effect of pH:-

The effect of pH upon uranium extraction was investigated. For this purpose, a series of experiments were performed at different pH values of synthetic solution ranging from 1 to 5 at fixed condition of O/A 1:1, using 10% vol. HAE2 for 10 min. as contact time. From the resulted in **Fig. (6)**, it is clear that, the maximum uranium extraction efficiency 98% was achieved at pH 4. Rising the solution pH over 4, leads to decreasing uranium extraction efficiency.

Effect of solvent concentration:-

The effect of the HAE2 concentration on uranium extraction efficiency by contacting equal volumes of synthetic solution and solvents with concentration ranging from 2 to 20% at fixed condition. From the obtained data on Fig (7), the results indicated that, by increasing the concentration of the HAE2 from 2 to 8, the uranium extraction efficiency increased from 45 to 90 % while at 10% a 98% of uranium extraction has been achieved.

Effect of the O/A phase ration:-

To study the effect of the A/O ratio upon uranium by using HAE2, the same conditions were applied. The obtained result revealed an extraction efficiency of uranium attaining 90 and 96% respectively at the A/O ratios of 2/1 and 3/1, in a manner to indicate that 1ml of the HAE2 can adsorb about 3.6 and 5.7 mg U respectively.

Effect of contact time:-

To study the effect of contact time upon uranium extraction efficiency by the HAE2 a set of experiments have been performed at different contact times 1,2,3,5 and 10min. at fixed conditions of O/A 1/1, pH 4 and using 10vol of the

HAE2. The obtained results illustrated at **Fig.(8)** have indicated that uranium extraction is rapidly achieved where at 5 min., up to 95% have been obtained and 99% at 10 min. thus the latter considered as the optimum shaking time.

Effect of temperature:-

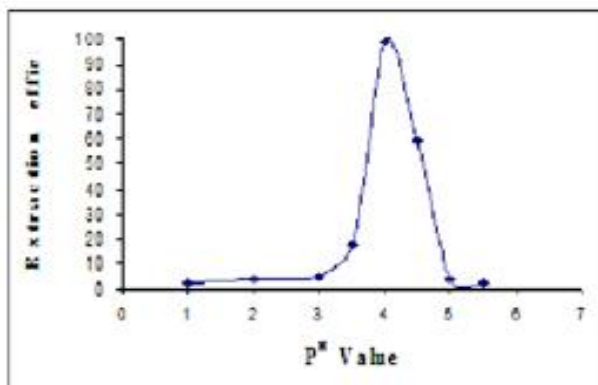
Using the previously optimum conditions of pH 4, O/A ratio of 1/1, a shaking time of 10 min. and 10 vol. % HAE2 for the studied system and diluted in carbon tetra chloride, a series of experiments was made to study the effect of temperature in the range from 25 to 70 °C. As shown in **Fig. (9)**, it was found that by progressively increasing the temperature an adverse effect upon the uranium extraction efficiency from 98 down to 40% has been obtained. This is most probably due to the decomposition of the components of the organic solvent under high temp.

Effect of interfering metal ions upon uranium extraction:-

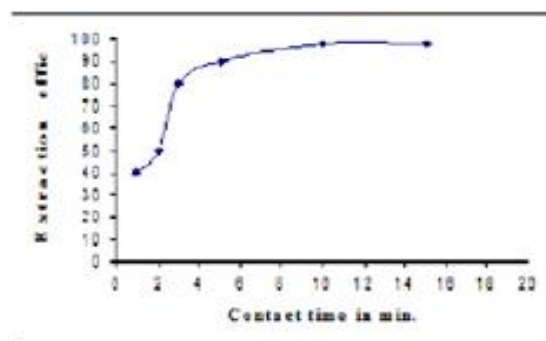
In order to investigate the possible and extent the interference of some metals ions- that might be associated with uranium in their ores, solution of Cu, Ni, Fe, Zr, V, Ba, and La, Ce, and Y as REEs have been prepared (1000 ppm) concentration for each. To study the effect of latter, 10 ml of the mentioned 9 metal solutions together with 10 ml of a uranium solution assaying also 1000 ppm have been mixed in a manner that each of which would assay 100 ppm. The concerned experiments was then performed under different pH values varying from 1 to 4 at the previously determined optimum conditions. All the obtained results are summarized in **Table (2)** for the studied metal values at the different pH values.

Table 2:- Effect of pH upon the extraction percent of uranium with some interfering metal ions.

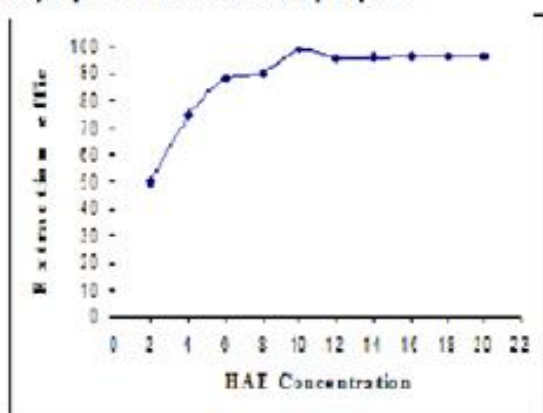
pH	Extraction %									
	U	Y	Ce	La	V	Fe	Zr	Ba	Ni	Cu
1	2.5	2	5	3	17	12	15	14	18	44
2	4	1	2	2	15	9	12	9	14	32
3	5	3	6	2	10	7	8	7	8	21
3.5	18	3	3	3	8	4	5	4	6	11
4	96	2	1	3	1	20	1	2	3	3
4.5	60	1	1	2	1	2	1	2	3	3



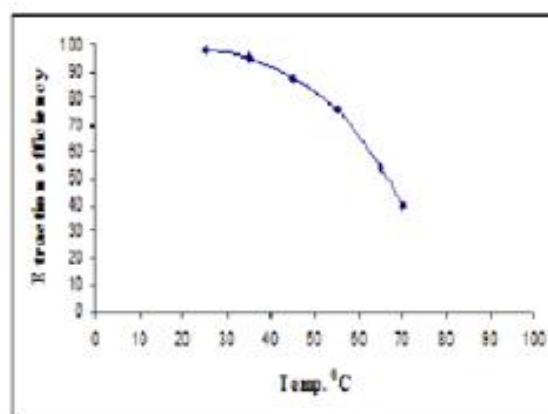
Fig(6): Effect of pH upon uranium extraction efficiency % by HAE



Fig(8): Effect of contact time upon uranium extraction efficiency % by HAE.



Fig(7): Effect of HAE Concentration upon uranium extraction efficiency, %.



Fig(9): Effect of temperature upon extraction efficiency % by HAE.

From the above studied of uranium extraction factors, it can be concluded that the optimum extraction conditions for about 98% of uranium content from sulfate solution via HAE2 would be summarized as:

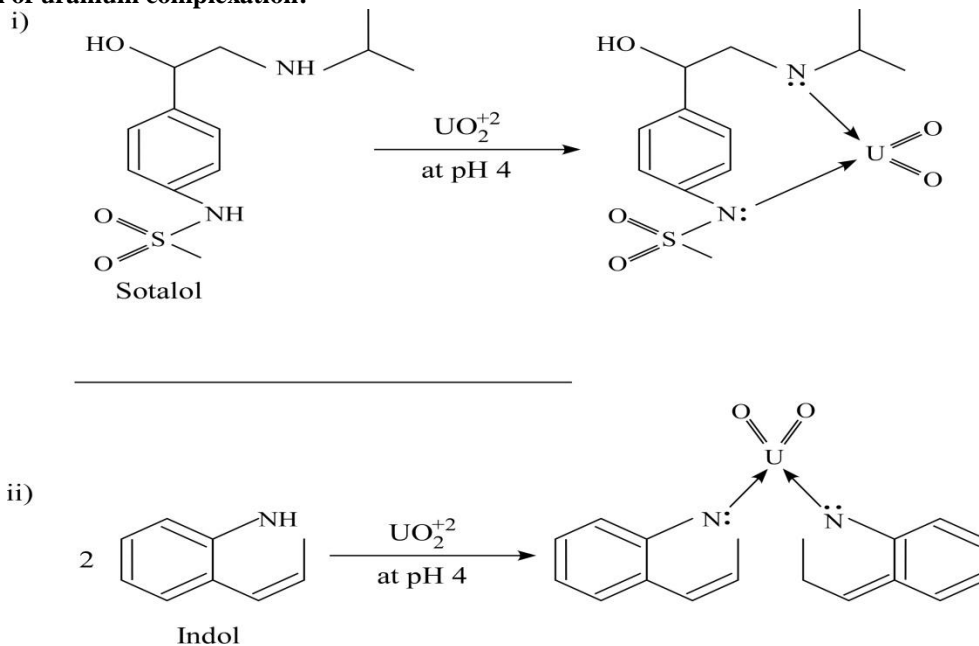
A/O	: 1:1
Contact time	: 10 min.
Ethanollic extractant Concentration	: 10 %
Temperature	: room temp.
pH	: 4

Uranium saturation capacity:-

To determine the saturation capacity of the HAE2 an known volume of the extractant has repeatedly been contacted with the prepared synthetic uranium solution under the previously determined optimum conditions using 1g of the HAE2 (10 vol.% in carbon tetra chloride). After each contact, uranium was analyzed in the obtained raffinate till almost saturation of the extractant. From the results shown in Table (3), it is clearly evident that the saturation capacity of the HAE2 attains about 22mg /1g of extractant for uranium. The obtained data support indeed the possible mechanism of uranium species extraction by the HAE2.

Table 3:- Uranium saturation capacity by the HAE2

Contact number	Uranium distribution	
	Assay in raffinate , ppm	Loaded amount mg/1g HAE2
1	Nil	6
2	Nil	6
3	200	4
4	300	3
5	450	1.5
6	500	1
7	450	0.5
8	600	Nil
Total		22

Mechanism of uranium complexation:-**Physical parameters:-****Thermodynamic characteristics of uranium extraction:-**

Variations of uranium extraction data with temperature for uranium extraction from the aqueous phase by used HAE2 in kerosene was used to calculate the thermodynamic constants including the standard enthalpy (ΔH), and the standard entropy (ΔS) based on Van't Hoff plot using the following formula:

$$\ln K_d = \frac{\Delta S^\circ}{R} - \frac{\Delta H^\circ}{RT}$$

Where K is the equilibrium constant $R = 8.3145 \text{ J mol}^{-1}\text{K}^{-1}$ and T = absolute temperature in Kelvin. ΔH and ΔS were determined from the slope and intercept of $\ln K_d$ versus $1/T$ graph. **Fig. (10)** Plots $\ln K_d$ versus $1/T$, K^{-1} (in case of HAE) which give a straight line whose slope equals $(-\Delta H / R)$ for the extraction of uranium. The ΔH and ΔS values for uranium, in case of HAE2, were -91.26 kJ/mol and -272 J/mol k respectively as calculated from the slope and intercept using the Van't Hoff equation. These values of ΔH and ΔS have then been used to obtain the corresponding free energy ($\Delta G = -10.2 \text{ kJ/mol}$) at 298°K for HAE using the following equation:

$$\Delta G^\circ = \Delta H^\circ - T\Delta S^\circ$$

The negative value of ΔH indicates that the extraction of uranium in this system is an exothermic process and that the reaction becomes more favorable at room temperature. The negative value of ΔG indicates that the reaction is

spontaneous. On the other hand, the observed decrease in the negative values of ΔG with elevated temperature implies that the reaction becomes more favorable at room temperatures.

Kinetic characteristics of uranium extraction by HAE:-

In terms of kinetic modeling, the pseudo-first-order and pseudo-second-order equations were used for the mathematical interpretation of the uranium extraction rate from the aqueous phase by HAE2. The pseudo-first-order equation is represented as follows:

$$\log(q_e - q_t) = \log q_e - \frac{k_1 t}{2.303}$$

Where q_e and q_t are the amounts of metal ions adsorbed at equilibrium and at time t respectively, and k_1 is the equilibrium rate constant of the pseudo first-order equation (1/min). On the other hand, the pseudo-second-order kinetic model is represented as follows:

$$\frac{t}{q_t} = \frac{1}{k_2 q_e^2} + \frac{t}{q_e}$$

Where k_2 is the equilibrium rate constant of pseudo second-order equation. The slope and intercept of the plot t/q_t versus t were thus used to calculate the pseudo second-order rate constants k_2 and q_e . The pseudo-first order kinetic model was found to best fit the experimental results of uranium extraction by HAE in kerosene with correlation coefficients very close to unity other than pseudo second-order kinetic model (Figs. 11 and 12) and the result agree with that reported by (Mckay et al., 1988).

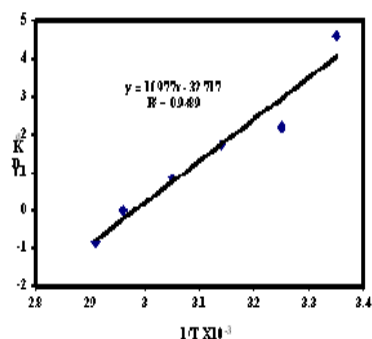


Fig. (10): Plot of $\ln K_d$ against reciprocal temperature for uranium from the 200 ppm by HAE2

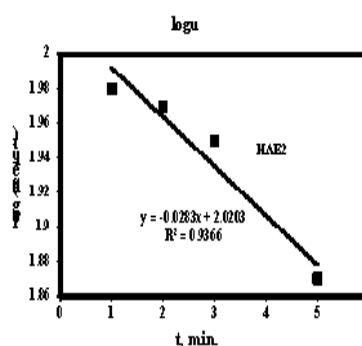


Fig. (11): Plot of $\log (q_e - q_t)$ versus time (t) for kinetic extraction of uranium from the 200 ppm aqueous solution by HAE2 at 25°C

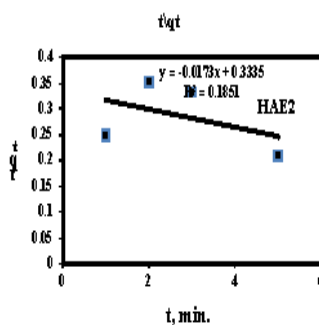


Fig. (12): Plot of t/q_t versus time (t) for kinetic extraction of uranium from the 200 ppm aqueous solution by used HAE2 at 25°C

Table 4:- Pseudo First-order constants of uranium extraction by used ethanolic extractant at 25°C.

Pseudo First-order constants	Result of HAE2
K_1	0.065
q_e	104.71
R^2	0.94

Optimization of the uranium stripping factors loaded upon the ethanolic extractant:-

Uranium stripping was carried out at room temperature by shaking equal volumes of the loaded solvent with a suitable stripping solution for a proper time. Factors influencing the stripping efficiency include stripping agent type and its concentration, aqueous to organic phase ratio (A/O), stripping time and temperature.

Effect of the stripping agent type:-

Different stripping agents were applied to strip uranium from the loaded 10% the HAE/kerosene. These included distilled water and 1 M of mineral acids HCl, H₂SO₄ and HNO₃ as well as NaCl/mineral acids (HCl, H₂SO₄ or HNO₃) mixtures. The stripping experiments were carried out in an A/O ratio of 1/1 for 10 min. contact time at room temp. The obtained results presented in **Table (5)** reveal that NaCl acidified H₂SO₄ at the mentioned molarity is actually the most efficient stripping agent for uranium from the loaded with 95% efficiency.

Table 5:- Effect of stripping agent type on uranium stripping efficiency loaded upon HAE2

Stripping agent type/ conc.,	U stripping efficiency, %
Water	20
HNO ₃ (1mole)	65
HCl (1mole)	67
H ₂ SO ₄ (1mole)	86
HNO ₃ /NaCl (mole / mole)	75
NaCl/HCl (mole / mole)	90
NaCl/ H ₂ SO ₄ (mole / mole)	95

It is interesting to mention here, that the uranium will stripped with NaCl/H₂SO₄ solution (mole/mole).

Effect of molarity NaCl/H₂SO₄ upon uranium stripping efficiency from the loaded solvent:-

The effect of NaCl and H₂SO₄ molarity ratio on uranium stripping from the loaded 10% HAE2 was studied while keeping the other stripping factors of contact time for 10 min., the settling time for 5 min. in an A/O phase ratio of 1/1. From the obtained results given in **Table (6)**, it is obvious that NaCl/H₂SO₄ ratio equal 1 has resulted 95% uranium stripping efficiency.

Table 6:- Effect of NaCl and H₂SO₄ molarity on uranium stripping from the loaded HAE2

Molarity of NaCl/H ₂ SO ₄ ratio	U stripping efficiency %
H ₂ SO ₄ / NaCl (mole / mole)	95
H ₂ SO ₄ / NaCl (0.5 mole/1 mole)	85
H ₂ SO ₄ / NaCl (0.25 mole/ 1 mole)	70

Effect of O / A phase ratio:-

In a manner to increase the uranium concentration in the stripping aqueous phase the effect of the A/O phase ratio upon the uranium stripping efficiency from the loaded HAE2 was studied up to the ratio of 3/1 using 1M NaCl /1M H₂SO₄. In these experiments, the other stripping factors were fixed at their studied optimum values. From the obtained results, it was shown that upon increasing the O/A ratio from 1/1 to 2/1, about 98% of uranium stripping was achieved while at the ratio of 3/1 give the same result as the A/O ratio of 2/1 thus the A/O of 3/1 is the optimum phase ratio.

Effect of contact time:-

The effect of a lower contact time upon the uranium stripping efficiency by mixed of 1M NaCl/1M H₂SO₄ from the loaded 10% HAE was studied for 5, 10 15 and 20 min. using the above mentioned optimum molarity of NaCl / H₂SO₄ at an O/A ratio of 3/1 at room temperature and using a settling time of 5 min. The corresponding uranium

stripping efficiencies indicate that a contact time of 15 min. as stripping time is the sufficient to achieve about 98% of the total uranium as shown in **Table(7)**.

Table 7:- Effect of stripping time upon uranium stripping efficiency for the HAE2

Stripping time	Uranium stripping efficiency%
5	66
10	75
15	98
20	98

From the above studied of uranium stripping factors, it can be concluded that the optimum stripping conditions for about 98% of uranium content from the loaded HAE would be summarized as following:

Stripping agent type : 1M NaCl /1M H₂SO₄
 A/O : 1:3
 Contact time : 15 min.
 Temperature : room temp

A Case study:, Abu-Zeneima ore material (South Eastern Sainai, Egypt) as a case study for uranium extraction using HAE2.

According to, the optimum leaching conditions of uranium from its mineralization at Abu-Zeneima ore material (South Eastern Sainai, Egypt) include a H₂SO₄ acid concentration 800 g/L, an agitation time of 4h, a S/L ratio of 1/2 at 100°C and using an ore grain size of – 60 mesh. (El Hazeek , 2008).

Characterization of Abu-Zeneima gibbsite ore:-

As previously mentioned, the working technological sample was provided from Abu Zeneima gibbsite ore lenses found in the shale beds. Results of the complete chemical analysis of the working sample are shown in Table (8).

Table 8:- Chemical analysis of Abu Zeneima ore material

Major oxide	%	Traces	ppm	REEs individual	
				Element	ppm
SiO ₂	21.07	U	560	La	209
TiO ₂	0.97	ΣREEs	3000	Ce	834
Al ₂ O ₃	30.43	Zn	3650	Nd	278
Fe ₂ O ₃	8.24	Cr	760	Sm	214
MgO	3.09	V	134	Eu	111
MnO	4.86	Cu	167	Gd	460
Na ₂ O	2.14	Ni	89	Tb	72
P ₂ O ₅	0.33	Mo	19	Dy	19
CaO	7.25	Cd	45	Ho	0.75
K ₂ O	1.10			Yb	0.98
L.O.I.*	20.17			Tm	0.67
Total	99.55			Y	500

From the prepared sulfate leach liquor of Abu-Zeneima mineralization uranium have then been recovered using HAE under the previously studied optimum conditions. Accordingly, a uranium extraction efficiency of 98% has been obtained. Subsequently, the uranium-loaded the HAE was subjected to uranium stripping using 1M NaCl /1M H₂SO₄ under the studied optimum stripping factors. These included an A/O ratio of 3/1 at room temperature for 15 min. contact time.

Uranium Precipitation:-

The stripping solution containing uranium would then be subjected to pH adjustment by using NH₄OH, which would be added to the solution, with continuous stirring to precipitate uranium at pH 7 as ammonium uranium oxide hydrate [UO₃NH₃H₂O] . After filtration, the produced filtrate would be re-acidified in order to be recycled in a new elution process. **Fig. (13)** Shows XRD analysis of the obtained [UO₃NH₃H₂O] product.

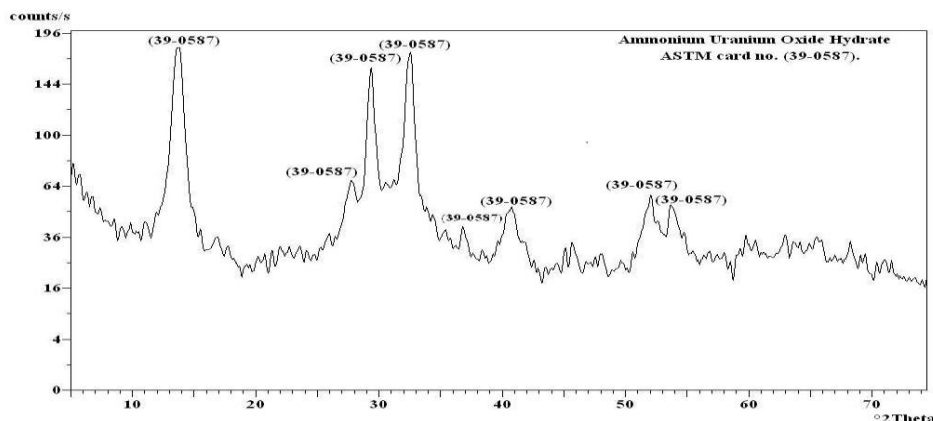


Fig. 13:- XRD analysis of the obtained $[\text{UO}_3\text{NH}_3\text{H}_2\text{O}]$ product.

Conclusion:-

The potentiality of the sunflower as extractant (HAE) for uranium from its sulfate solutions has actually been proven and the studied relevant factors have actually been optimized. These involved a 10 vol. HAE in kerosene, an O/A ratio of 1:1 for a shaking time 10 min. at the pH of 4. Under these conditions, the achieved uranium capacity has attained 33mg/g for uranium. The loaded uranium was afterward completely eluted using $\text{NaCl} / \text{H}_2\text{SO}_4$ solution. Finally, the working HAE was successfully applied for uranium recovery from an actual sulfate leach liquor mineralization in from the actual sulfate leach liquor of Abu Zeneima ore material (South Eastern Sinaï, Egypt).

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RESEARCH ARTICLE

“A STUDY ON HOW WE’RE INTERACTING WITH BRANDS ON DIFFERENT SOCIAL MEDIA”

Fahad Galib Ani, John Peacock and Shahanaz Akter.

Manuscript Info

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Abstract

A new study which investigates how users are interacting with brands across various social media. From this study we have found out that on average, people maintain accounts on three channels of social media. One hundred percent of all respondents stated that they actively follow brands on social media.

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Introduction:-

All things considered, most of the members are from three online networking groups. Respondents take after brands on various types of web-based social networking to fulfill distinctive requirements. Despite the fact that there are some key reasons they give for taking after brands by means of particular channels there are additionally many reasons which cover and are given paying little respect to the interpersonal organization picked.

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Other(s) Likert Scale	20

Executive Summary:-

On average, people are members of three social media communities. Respondents follow brands on different forms of social media to satisfy different needs. Although there are some key reasons they give for following brands via specific channels there are also many reasons which overlap and are given regardless of the social network chosen.

The most popular reasons for following brands on social media are displayed in Figure 1. The first three reasons are given more or less consistently for all social media networks analyzed. Similarly, the bottom three reasons are given less frequently among all social media platforms.

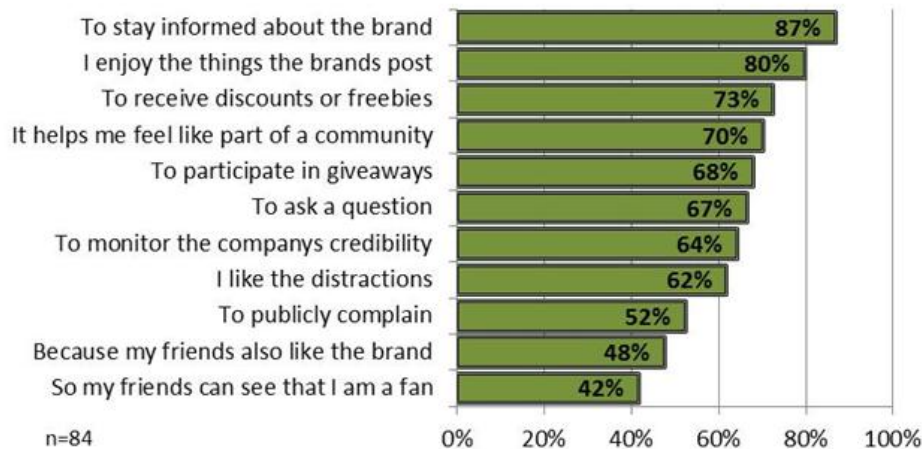


Figure 1:- Reasons for Following Brands on All Social Media

Despite these overlapping reasons for using social media, there also exist specific reasons people give for using certain platforms.

Facebook remains the most popular social network for following brands, perhaps due in part to its almost ubiquitous status as it is used by 90% of respondents. It is preferred more than twice as often as rival social networks for following brands. People use it to ask companies questions and check companies' credibility. The most popular brand category reported was news the social network is used relatively equally between the two genders.

Instagram was the second most popular social network analyzed for following brands on social media. This platform is used primarily by females and is particularly popular for following fashion brands. Restaurants and bars are also a popular brand category that is followed on Instagram. More so than on other networks, Instagram users express that the social media platform helps them to feel like part of a community.

Twitter was reported to be the least preferred social network for following brands. Although 34% of all respondents were members of this platform, on average users still preferred any other social media platform for following brands. Most notably, the brand categories bands and other(s) were ranked highly by users of twitter. The reasons given by twitter users for following brands fall in line largely with those of the other social media networks.

Hypotheses:-

The investigated hypotheses are reviewed in this section.

Hypothesis 1:-

The group's first hypothesis was as follows:

Respondents are more likely to use instagram to follow fashion and beauty product brands.

To check the validity of this statement, a statistical analysis of respondents' answers about following fashion and beauty brands on social media were reviewed and can be seen in Figure 2. As is displayed, Instagram users most consistently selected fashion brands as one of the brand categories they follow.

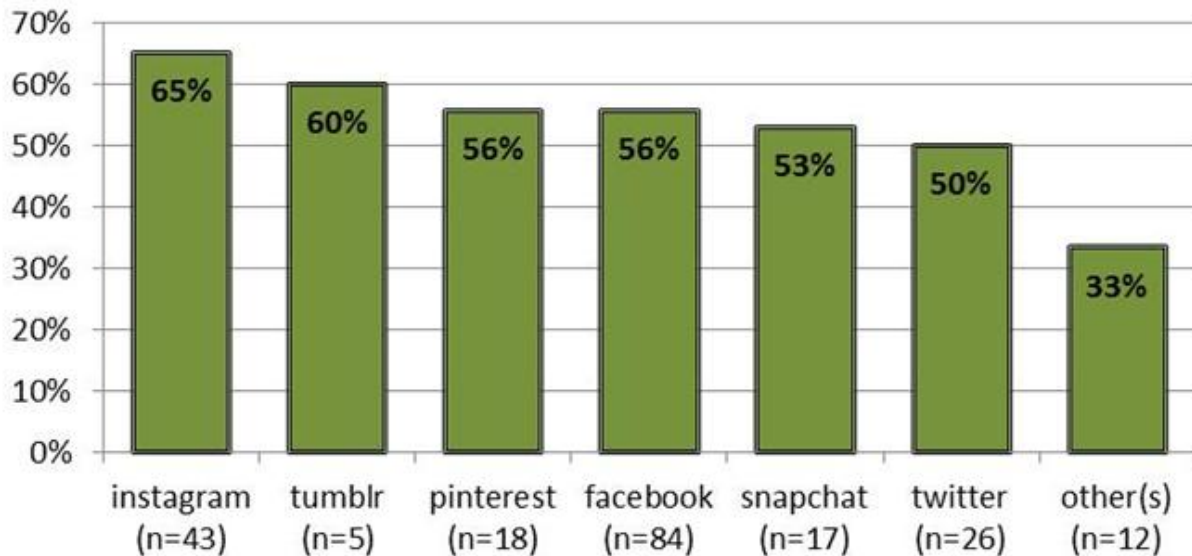


Figure 2:- Percentage of followers of fashion brands on all social media

For the second part of this hypothesis, the same review was carried out but with the data regarding the brand category “Beauty and Hygiene Products”. The responses split by social media channel can be seen in Figure 3. While Instagram does not show the highest percentage of users who selected this option, it is important to note that the only two higher ranking social networks had fewer respondents and therefore weight each response more strongly.

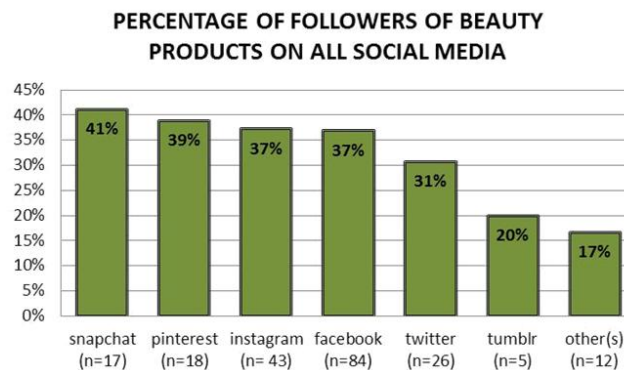


Figure 3:- Percentage of followers of beauty and hygiene products on all social media

Nonetheless, this survey only managed to partially support this hypothesis.

Hypothesis 2

The second hypothesis was as follows:

Facebook is used most for following offline brands (bars, restaurants etc.).

To answer this question, the brand categories “restaurants & bars” and “clubs & venues” were combined to analyze how users of the different social networks following “offline” brands. The results can be seen in Figure 4. As can be seen, Facebook scored well below most other social networks despite its advanced event coordination features.

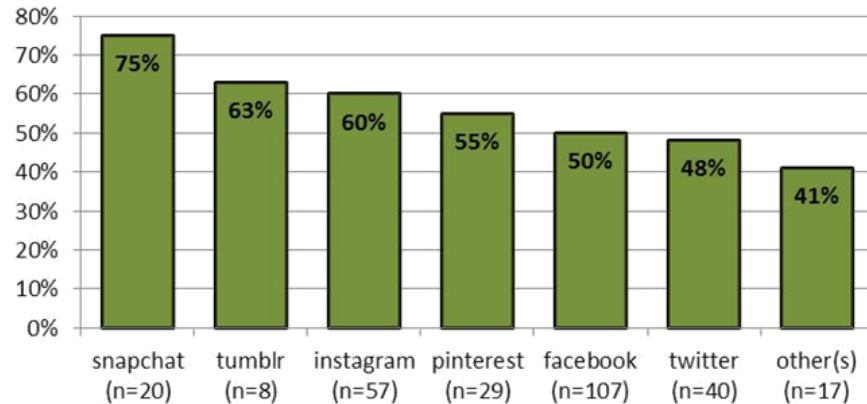


Figure 4:- Percentage of followers of offline brands on all social media

It was therefore concluded that this hypothesis was not supported by the collected data.

Hypothesis 3

The third hypothesis was as follows:

People are most likely to use facebook and twitter to complain, ask questions and check companies credibility.

In order to review this statement, the responses of all groups were compared for the listed reasons people provided for following brands. The results are displayed in Figure 5.

As can be seen, respondents use facebook to complain at a higher rate than reported for many other social networks, however asking questions and checking company credibility is reported on par with the rates reported for other social networks. For this reason, this hypothesis can be considered unsupported.

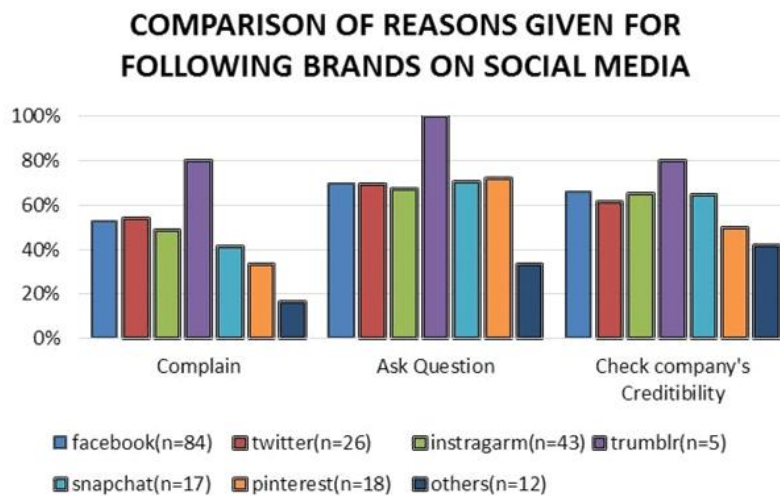


Figure 5:- Comparison of reasons given for following brands on social media

Hypothesis 4:-

The fourth hypothesis was as follows:

Females are more likely to use Instagram.

In order to test this hypothesis, the number of respondents who reported actively using Instagram was evaluated and split based on gender. The resulting percentages were as follows: 64% female users and only 36% male users. The entire sample of respondents, regardless of social media use, was 54% female and 26% male. Correcting for this slightly female skewed sample, it can nonetheless be concluded that Instagram is more popular among females than males.

Hypothesis 5:-

People are more likely to use facebook and twitter to stay up-to-date on brands.

Hypothesis 6:-

The sixth hypothesis is stated as follows:

People who follow trends closely are more likely to actively use newer forms of social media.

In order to test this hypothesis, a Pearson's correlation between the independent variable based on the Likert scale number derived from question 7 part 1 and the dependent variable derived from the number of social media networks respondents professed to using. The resulting r value is -0.065, i.e. a

Slightly negative correlation with almost no statistical significance. Further analysis on defined cohorts within the Likert scale further confirmed this as those who consider themselves to more closely follow trends had, on average, one less social media platform used as compared to the entire sample. It is therefore concluded that this hypothesis is not supported by the data collected.

Hypothesis 7:-

The seventh hypothesis is:

Respondents will prefer following social media brands on facebook.

In order to test this hypothesis, the answers to a set of analytical hierarchy process pairwise questions asking which social network was preferred among all social networks the respondents indicated they use were analyzed. The results can be viewed below (the median was used due to the high standard deviation):

Respondents' Median Preference of**Social Networks for Following Brands**

100%					
90%					
80%					
70%					
60%					
50%					
40%					No
30%	64%				Yes
20%					
10%		24%		12%	20%
0%	5%	7%	6%		

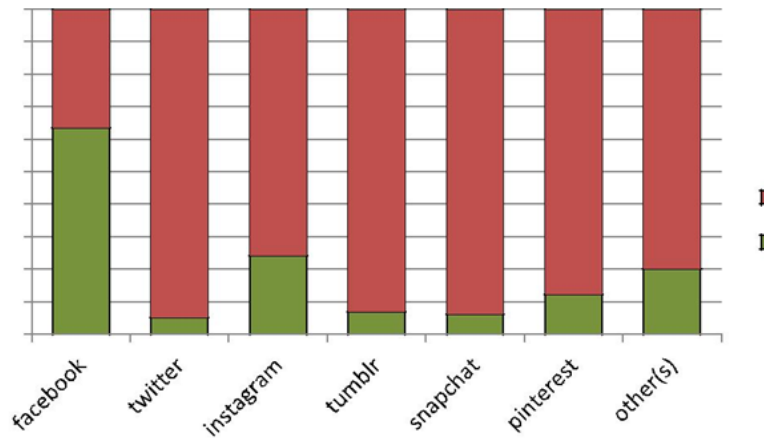


Figure 6:- Respondents' Median Preference of Social networks for Following Brands.

As can be seen, facebook is by far the most preferred social media network for following brands. This hypothesis is therefore confirmed.

Critical Self Reflection:-

Despite initial efforts to carefully plan and execute this marketing research project, some areas in which improvement can be achieved have been identified along the way. In general, the nature of these problems can be categorized as technical and conceptual.

Technical Difficulties:-

Unfamiliarity with some of the survey software used led to difficulties that affected the quality of responses received. Some questions (particularly multi-choice selections and decision tree scales) were improperly configured and as a result were not mandatory. The result is a portion of respondents simply skipped some questions. While the remaining results were nonetheless usable, this reduced the number of responses which could be analyzed for some questions and, specifically for the social media user groups investigated, which resulted in data for some groups which did not reach theoretical saturation.

Conceptual Difficulties:-

In terms of the questions asked of respondents, some of the underlying methodology could have been improved. Most importantly, the questions posed to users of specific social media platforms were not specific to that platform (i.e. each respondent answers the question “What categories of brands do you follow on social media” with only one answer for all social media platforms used or via the analytic hierarchy process). While creating this set of questions for each social network was considered, the group decided against doing so as it would:

1. exponentially increase the length of the survey, thus increasing the number of drop-outs,
2. increasing difficulty in understanding the question among respondents, resulting in a higher dropout rate, particularly because of a high reliance on respondents whose mother tongue is not English and
3. Assume the ability of respondents to easily differentiate their motivations for following brands differentiated by the social networks used.

Field Report:-

An English-language survey was conducted from 28.12.2017 to 03.01.2017. The methodology included dichotomous questions and question sets, 11-point Likert scales and an analytic hierarchy

process¹ to simultaneously gauge respondents' social media preferences while monitoring their response consistency. The full set of questions asked in the survey is available within the appendix.

Key response metrics are available in Table 1 and Table 2.

Table 1:- Survey Completion Statistics**SURVEY COMPLETION STATISTICS**

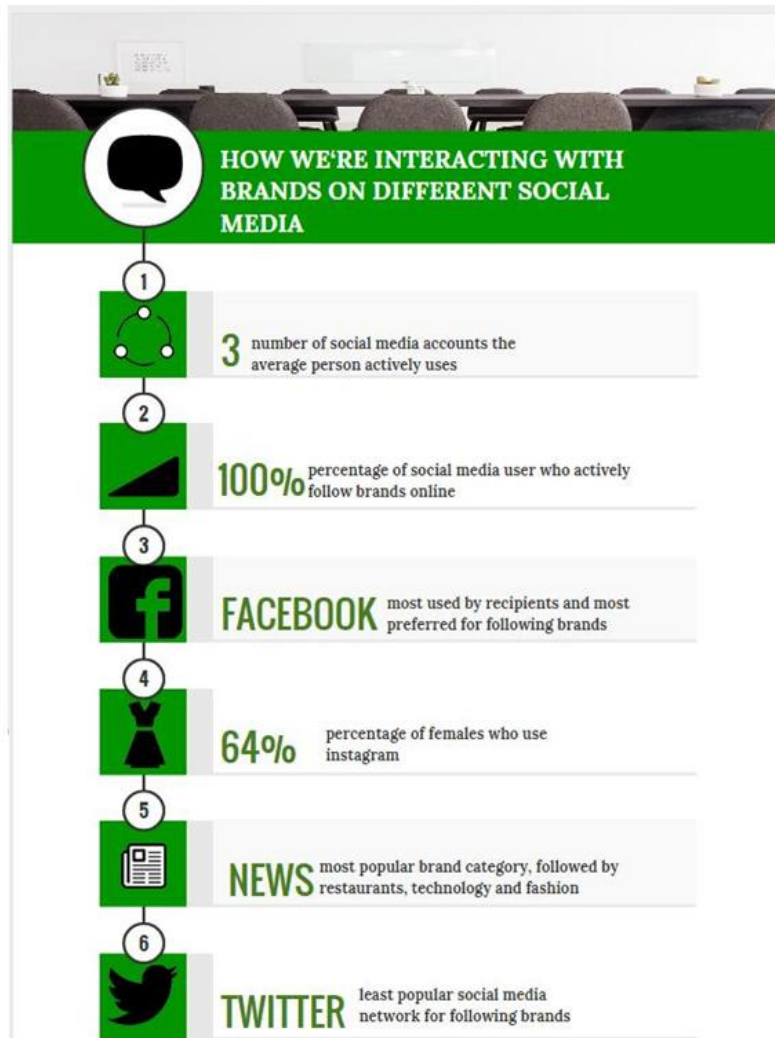
	Absolute	Percentage
Started	227	100%
Completed	120	53%
Excluded	2	1%
Usable Responses	118	52%

Table 2:- Survey Response Time (in minutes)**RESPONSE TIME (in minutes)**

Mean	02:57
Median	02:39
Minimum	00:41
Maximum	09:26

¹ Saaty, Thomas L. "Decision Making With the Analytic Hierarchy Process". International Journal of Services Sciences 1.1 (2008): 83. Web.

Appendix:-
Infographic:-



Conclusion:-

A new study which investigates how users are interacting with brands across various social media. From this study we have found out that on average, people maintain accounts on three channels of social media. One hundred percent of all respondents stated that they actively follow brands on social media.

Unsurprisingly, facebook remains the most popular social media network as ranked by percentage of members who responded to the survey and by user preference for following brands. Second in both aforementioned categories is Instagram. Twitter, while used by 34 percent of respondents, was the least preferred social network for following brands.

The king of fashion is predictably Instagram, where a whopping 65% of respondents – which coincidentally were mostly female – say they follow fashion brands. Twitter was most popular for news, though facebook users see themselves as, on average, more interested in the affairs around the world.

Despite these stark contrasts, a lot of similarities exist between the social media networks. Generally all users, regardless of the networks they use, stated they follow brands primarily to stay up-to-date about them, because they enjoy the things the brands post and to take part in sweepstakes and contests. Also across the board people said they were least likely to follow brands to show off to their friends.

These results can be used as a benchmark for leading brands and social media to improve their future strategies as these results clearly users or customers brand preferences by using different kinds of social media. Moreover, these results could be helpful for businesses to reach their potential customers easily.

SPSS Analysis

Membership by Gender

Report

Gender		facebook	twitter	instagram	tumblr	snapchat	pinterest	other(s)
Female	Mean	.922	.297	.563	.109	.219	.375	.172
	N	64	64	64	64	64	64	64
	Std. Deviation	.2705	.4605	.5000	.3146	.4167	.4880	.3803
	Median	1.000	.000	1.000	.000	.000	.000	.000
Male	Mean	.889	.389	.389	.019	.111	.093	.111
	N	54	54	54	54	54	54	54
	Std. Deviation	.3172	.4921	.4921	.1361	.3172	.2926	.3172
	Median	1.000	.000	.000	.000	.000	.000	.000
Total	Mean	.907	.339	.483	.068	.169	.246	.144
	N	118	118	118	118	118	118	118
	Std. Deviation	.2920	.4754	.5018	.2525	.3768	.4324	.3527
	Median	1.000	.000	.000	.000	.000	.000	.000

Membership by device type

Report

Mobile Device Background Check		facebook	twitter	instagram	tumblr	snapchat	pinterest	other(s)
Not a mobile device	Mean	.961	.431	.471	.039	.118	.157	.157
	N	51	51	51	51	51	51	51

	Std. Deviation	.1960	.5002	.5041	.1960	.3254	.3673	.3673
	Median	1.000	.000	.000	.000	.000	.000	.000
Mobile device without gyroscope	Mean	1.000	.000	.000	.000	.000	.000	.000
	N	2	2	2	2	2	2	2
	Std. Deviation	.0000	.0000	.0000	.0000	.0000	.0000	.0000
	Median	1.000	.000	.000	.000	.000	.000	.000
Mobile device with gyroscope	Mean	.862	.277	.508	.092	.215	.323	.138
	N	65	65	65	65	65	65	65
	Std. Deviation	.3481	.4510	.5038	.2917	.4143	.4713	.3481
	Median	1.000	.000	1.000	.000	.000	.000	.000
Total	Mean	.907	.339	.483	.068	.169	.246	.144
	N	118	118	118	118	118	118	118
	Std. Deviation	.2920	.4754	.5018	.2525	.3768	.4324	.3527
	Median	1.000	.000	.000	.000	.000	.000	.000

Membership by Employment Status

Report

Employment status		facebook	twitter	instagram	tumblr	snapchat	pinterest	other(s)
In training (student, apprentice or similar)	Mean	.875	.268	.446	.036	.161	.161	.196
	N	56	56	56	56	56	56	56
	Std. Deviation	.3337	.4469	.5016	.1873	.3706	.3706	.4009
	Median	1.000	.000	.000	.000	.000	.000	.000
Employed	Mean	.949	.359	.462	.077	.179	.308	.051
	N	39	39	39	39	39	39	39

	Std. Deviation	.2235	.4860	.5050	.2700	.3888	.4676	.2235
	Median	1.000	.000	.000	.000	.000	.000	.000
Self-employed	Mean	.833	.750	.750	.167	.167	.417	.083
	N	12	12	12	12	12	12	12
	Std. Deviation	.3892	.4523	.4523	.3892	.3892	.5149	.2887
	Median	1.000	1.000	1.000	.000	.000	.000	.000
Unemployed	Mean	1.000	.222	.333	.111	.222	.111	.333
	N	9	9	9	9	9	9	9
	Std. Deviation	.0000	.4410	.5000	.3333	.4410	.3333	.5000
	Median	1.000	.000	.000	.000	.000	.000	.000
Retired	Mean	1.000	.000	1.000	.000	.000	1.000	.000
	N	2	2	2	2	2	2	2
	Std. Deviation	.0000	.0000	.0000	.0000	.0000	.0000	.0000
	Median	1.000	.000	1.000	.000	.000	1.000	.000

Total	Mean	.907	.339	.483	.068	.169	.246	.144
	N	118	118	118	118	118	118	118
	Std. Deviation	.2920	.4754	.5018	.2525	.3768	.4324	.3527
	Median	1.000	.000	.000	.000	.000	.000	.000

Membership by Education Attainment Report

Education		facebook	twitter	instagram	tumblr	snapchat	pinterest	other(s)
Less than high school (or local equivalent)	Mean	1.000	.000	.000	.000	.000	.000	.000
	N	1	1	1	1	1	1	1

	Std. Deviation
	Median	1.000	.000	.000	.000	.000	.000	.000
High school (or local equivalent)	Mean	1.000	.600	.600	.400	.400	.800	.200
	N	5	5	5	5	5	5	5
	Std. Deviation	.0000	.5477	.5477	.5477	.5477	.4472	.4472
	Median	1.000	1.000	1.000	.000	.000	1.000	.000
Some college, no degree (or local equivalent)	Mean	.929	.357	.643	.071	.286	.357	.214
	N	14	14	14	14	14	14	14
	Std. Deviation	.2673	.4972	.4972	.2673	.4688	.4972	.4258
	Median	1.000	.000	1.000	.000	.000	.000	.000
Bachelor degree (or local equivalent)	Mean	.860	.349	.419	.116	.163	.302	.140
	N	43	43	43	43	43	43	43
	Std. Deviation	.3506	.4822	.4992	.3244	.3735	.4647	.3506
	Median	1.000	.000	.000	.000	.000	.000	.000
Master degree (or local equivalent)	Mean	.935	.261	.457	.000	.109	.087	.152
	N	46	46	46	46	46	46	46
	Std. Deviation	.2496	.4440	.5036	.0000	.3147	.2849	.3632
	Median	1.000	.000	.000	.000	.000	.000	.000
More Advanced degree (or local equivalent)	Mean	1.000	1.000	.800	.000	.400	.200	.000
	N	5	5	5	5	5	5	5
	Std. Deviation	.0000	.0000	.4472	.0000	.5477	.4472	.0000
	Median	1.000	1.000	1.000	.000	.000	.000	.000
I dont want to answer	Mean	.750	.000	.500	.000	.000	.500	.000
	N	4	4	4	4	4	4	4
	Std. Deviation	.5000	.0000	.5774	.0000	.0000	.5774	.0000
	Median	1.000	.000	.500	.000	.000	.500	.000

Total	Mean	.907	.339	.483	.068	.169	.246	.144
	N	118	118	118	118	118	118	118
	Std. Deviation	.2920	.4754	.5018	.2525	.3768	.4324	.3527
	Median	1.000	.000	.000	.000	.000	.000	.000

Facebook Likert Scales**Report**

facebook		How closely do you follow new trends?	How interested are you in current affairs in the world?	Are you interested in politics?	How well do brands help you express your personality?
	Mean	6.625	6.625	4.750	6.500
	N	8	8	8	8
	Std. Deviation	1.5980	1.5980	3.2404	1.5119
	Median	6.500	6.500	3.500	6.500
Yes	Mean	5.867	6.827	5.720	5.453
	N	75	75	75	75
	Std. Deviation	2.3035	2.4790	2.9342	2.5431
	Median	6.000	7.000	6.000	6.000
Total	Mean	5.940	6.807	5.627	5.554
	N	83	83	83	83
	Std. Deviation	2.2488	2.4015	2.9579	2.4755
	Median	6.000	7.000	6.000	6.000

Twitter Likert Scale**Report**

twitter		How closely do you follow new trends?	How interested are you in current affairs in the world?	Are you interested in politics?	How well do brands help you express your personality?
No	Mean	5.860	6.825	5.596	5.667
	N	57	57	57	57
	Std. Deviation	2.0996	2.2609	3.0110	2.3171
	Median	6.000	7.000	6.000	6.000
Yes	Mean	6.115	6.769	5.692	5.308
	N	26	26	26	26
	Std. Deviation	2.5819	2.7321	2.8951	2.8252
	Median	6.000	7.000	6.000	5.500
Total	Mean	5.940	6.807	5.627	5.554
	N	83	83	83	83
	Std. Deviation	2.2488	2.4015	2.9579	2.4755
	Median	6.000	7.000	6.000	6.000

Instagram Likert Scale Report

instagram		How closely do you follow new trends?	How interested are you in current affairs in the world?	Are you interested in politics?	How well do brands help you express your personality?
No	Mean	6.100	6.875	6.025	5.575
	N	40	40	40	40
	Std. Deviation	2.2165	2.3225	2.7408	2.5908
	Median	6.500	7.000	6.500	6.000
Yes	Mean	5.791	6.744	5.256	5.535
	N	43	43	43	43
	Std. Deviation	2.2945	2.4985	3.1327	2.3939
	Median	6.000	7.000	5.000	6.000
Total	Mean	5.940	6.807	5.627	5.554
	N	83	83	83	83
	Std. Deviation	2.2488	2.4015	2.9579	2.4755
	Median	6.000	7.000	6.000	6.000

Tumblr Likert Scale Report

tumblr		How closely do you follow new trends?	How interested are you in current affairs in the world?	Are you interested in politics?	How well do brands help you express your personality?

No	Mean	6.026	6.885	5.654	5.603
	N	78	78	78	78
	Std. Deviation	2.2734	2.2447	2.9180	2.4671
	Median	6.000	7.000	6.000	6.000
Yes	Mean	4.600	5.600	5.200	4.800
	N	5	5	5	5
	Std. Deviation	1.3416	4.3932	3.8987	2.7749
	Median	4.000	7.000	4.000	4.000
Total	Mean	5.940	6.807	5.627	5.554
	N	83	83	83	83
	Std. Deviation	2.2488	2.4015	2.9579	2.4755
	Median	6.000	7.000	6.000	6.000
			Report		
snatchat		How closely do you follow new trends?	How interested are you in current affairs in the world?	Are you interested in politics?	How well do brands help you express your personality?
No	Mean	5.909	6.742	5.424	5.439
	N	66	66	66	66
	Std. Deviation	2.1960	2.2757	2.9092	2.4059
	Median	6.000	7.000	6.000	6.000
Yes	Mean	6.059	7.059	6.412	6.000
	N	17	17	17	17

	Std. Deviation	2.5117	2.9041	3.1036	2.7613
	Median	6.000	8.000	7.000	6.000
Total	Mean	5.940	6.807	5.627	5.554
	N	83	83	83	83
	Std. Deviation	2.2488	2.4015	2.9579	2.4755
	Median	6.000	7.000	6.000	6.000

Pinerest Likert Scale Report

pinterest		How closely do you follow new trends?	How interested are you in current affairs in the world?	Are you interested in politics?	How well do brands help you express your personality?
No	Mean	5.938	6.538	5.385	5.631
	N	65	65	65	65
	Std. Deviation	2.3444	2.3788	2.9245	2.4467
	Median	6.000	7.000	6.000	6.000
Yes	Mean	5.944	7.778	6.500	5.278
	N	18	18	18	18
	Std. Deviation	1.9242	2.2895	2.9951	2.6303
	Median	6.000	8.000	6.500	5.000
Total	Mean	5.940	6.807	5.627	5.554
	N	83	83	83	83
	Std. Deviation	2.2488	2.4015	2.9579	2.4755

Fans of Brands – Submitted by John Peacock, Shahanaz Akter, Fahad Galib Ani

	Median	6.000	7.000	6.000	6.000
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Other(s) Likert Scale Report

other(s)		How closely do you follow new trends?	How interested are you in current affairs in the world?	Are you interested in politics?	How well do brands help you express your personality?
No	Mean	6.028	6.761	5.718	5.662
	N	71	71	71	71
	Std. Deviation	2.3236	2.4522	2.9722	2.4897
	Median	6.000	7.000	6.000	6.000
Yes	Mean	5.417	7.083	5.083	4.917
	N	12	12	12	12
	Std. Deviation	1.7299	2.1515	2.9375	2.3916
	Median	6.000	8.000	6.000	5.000
Total	Mean	5.940	6.807	5.627	5.554
	N	83	83	83	83
	Std. Deviation	2.2488	2.4015	2.9579	2.4755
	Median	6.000	7.000	6.000	6.000



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RESEARCH ARTICLE

EPISIOTOMY AND THE IMPACT ON THE PELVIC FLOOR: BIOMECHANICAL OVERVIEW.

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Key words:-

Episiotomy; Pelvic Floor Dysfunctions;
biocomputational birth simulator

Abstract

Aim: The aim of this review is to provide a comprehensive overview of the available literature on the use of episiotomy and its impact on the pelvic floor and to compare the current knowledge on the subject with recently developed bioengineer programs which allows us to further understand the labor process.

Method: We performed a literature research in the PubMed database for studies referring to the use of episiotomy published between 1990 and 2016, without restrictions for language and study type. We then assessed that knowledge to the findings of a new developing bioengineer program which simulates the childbirth, measuring the level of pressure on the pelvic floor and the outcomes.

Results: Biomechanical models of childbirth allow the simulation of several incisions on the pelvic floor muscles in order to quantify the effect and consequences of episiotomy. According to the analyzed studies, a mediolateral episiotomy reduces the stress on the muscles and the force required to successfully delivery. These models estimate also that the incision limited muscle injury. There was a strong agreement in the findings of the bioengineering program and the current knowledge about medicine. According to the results of the bioengineering study, a mediolateral episiotomy has a protective effect, reducing the stress on the muscles, and the force required during delivery. The intervention also has benefits on muscle injury, reducing the damage to a small zone.

Conclusion: Although there is a strong evidence that the cut applied mediolateral would have less impact on the pelvic floor and a protective effect during delivery, more studies should be made assessing the damage on the perineum and the consequences that may occur (blood loss, infections and others).

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Introduction:-

The pelvic floor is a muscular structure with an essential role in women's well-being and health. Its integrity is essential in maintaining urological, gynecological, gastroenterological and pulmonary functions. (Neels et al. 2016) There are numerous risk factors that can contribute to Pelvic Floor Dysfunction (PFD) which may lead to consequences such as Urinary Incontinence (UI), Fecal Incontinence (FI), Pelvic Organ Prolapse (POP) and sexual problems, like dyspareunia. These problems often deteriorate a women's quality of life and result in the need of complex surgical procedures, with high ratings of reoperation. Some of the main contributing factors to the occurrence of PFD are obesity, congenital factors, aging, pregnancy, childbirth, menopause and Chronic Obstructive Pulmonary Disease (Fritel 2010; Neels et al. 2016), which increase the pressure applied on the women's pelvic floor leading to muscular dysfunction or tears.

Approximately 54% of women have normal vaginal delivery, 21% instrumental delivery and 25% undergo caesarean section. (De Souza et al. 2015) The first two are responsible for the appliance of a higher degree of pressure on the pelvic floor, leading to a higher prevalence of PFD. It was therefore important to find a way of relieving this pressure.

Episiotomy is a surgical incision performed by the obstetrician during the second stage of labor (that ensues after a fully dilated cervix and ends with childbirth) on the musculature of the pelvic floor. It is the most common procedure in Obstetrics (Carroli and Mignini 2009; Graham et al. 2005), even though its routine practice is not supported by studies (Myers-Helfgott MG 1999) (de Tairac et al. 2006). Its main aim is to create a wider opening of the vagina preventing the hyperdistention and laceration of the perineum, vagina and, most specially, the muscles and aponeuroses of the pelvic floor, amongst which the levator ani muscle, minimizing post-partum complications. The procedure is done without the previous consent of the patient, being now considered a "obstetric violence" (Melo et al. 2014) (an act in which routine medical or pharmacological procedures are conducted without allowing the women to make the decision). There is controversy regarding when to preform episiotomy as a prophylactic measure amongst professionals. Even without this procedure around 23% of women with vaginal delivery need posterior suture of the perineum. (Leal 2014).

Currently the World Health Organization (WHO) recommends the reduction of episiotomy rate to 10%, encouraging physicians to use their judgement when to perform it. (Melo et al. 2014) The problem remains in establishing a protocol of when and which type to use.

The lack of consensus regarding this practice is an ongoing discussion, therefore the importance of biomechanical studies (Oliveira et al. 2016) to assess the impact on the women's pelvic floor and its capability in preventing PFD.

Methods and Materials:

Using the database of PubMed we acquired studies published from 1990 to 2016 about episiotomy and its diverse effects not only during but also after labor, comparing it to the data obtained by the computational model (simulator).

Results:**General knowledge:**

The pelvic floor of women is a complex muscular structure that must be kept intact as to prevent future severe damage, one of the most common being PDF which may in time lead to many other consequences that leave the woman unable to have the same life as before.

Childbirth is an occurrence when the pelvic floor undergoes a large amount of stress, sometimes unable to cope with it, leading to an increase likelihood of developing PDF. The highest point of this pressure is felt on the vaginal birth, leading to a much higher incidence of diseases such as urinary incontinence, organ prolapse, chronic pelvic pain and dyspareunia in vaginal childbirths than in cesarean. (Bertozi et al. 2011) (Victoria L. Handa 2012).

Perineal injury is the most common obstetrical consequence after vaginal childbirth (18,4% in primiparous women and 24,6% in multiparous). (Bertozi et al. 2011) (Mellier and Delille 1990) Therefore physicians started applying episiotomy as a mode of decreasing the pressure on the pelvic floor in order to attempt to decrease the negative effects vaginal delivery might have had on the pelvic floor. (Fonti et al. 2009; Memon and Handa 2013)

Its appliance leads to a significant reduction of first and second degree tears of the vagina (Lavesson et al. 2014) and anterior perineal lacerations (de Tayrac et al. 2006). It is also associated with a better outcome in assisted deliveries, since the appliance of forceps or perineal tear without episiotomy were associated with higher PFD prevalence 5 to 10 years after birth, unlike with the usage of episiotomy. The likelihood of developing PFD does not increase with the number of episiotomies performed, unlike those with assisted delivery by forceps. The risk of prolapse is much higher in women who suffer natural tears, due to the extensive stretching of the vaginal hiatus which leads to the avulsion of the levator ani. (Victoria L. Handa 2012) It is also performed to hasten the birth, preventing severe damage in a distressed baby, and to minimize or avoid complications from abnormal presentations.

Its use as a routine prophylactic measure was proved not to reduce perineal trauma or pelvic floor relaxation (Klein et al. 1992), thus leading to a more restricted approach to its use. Although being a constant source of discussion and contrary opinions, there is still to be established a consensus regarding its use.

Pelvic floor changes during childbirth:-

The women's pelvic floor is a complex structure that maintains the organs of the pelvic region in place allowing a normal functioning of different systems. This region undergoes an increased amount of pressure during pregnancy, and most notably, during stage II of childbirth. Therefore, it may result in tears and lead to a malfunctioning pelvic floor.

This pavement contains the perineum, the endopelvic fascia, the arcus tendinous fasciae pelvis, the coccygeus muscle, the levator ani muscles and a supporting layer consisting of the anterior vaginal wall and the connective tissue that attaches to the pelvic bones through the pubovaginal portion of the levator ani muscle, and the uterosacral and cardinal ligaments comprising the tendinous arch of the pelvic fascia.

The levator ani muscle along with its associated fascia make the pelvic diaphragm, responsible for maintaining closure of the urogenital hiatus. The anus hiatus on the other hand is composed by the pubic bones and levator ani muscles, the perineal body and external anal sphincter. (Ashton-Miller and DeLancey 2007).

It is easy to understand that the malfunctioning of one of these structures may lead to various consequences on the pelvic floors functions leading to PFDs.

During the II stage of labor the contraction of the abdominal wall and the respiratory diaphragm timed with the peak of uterine contraction greatly increase the pressure on the pelvic floor to successfully drive the fetal head through the hiatus. The pubococcygeus muscle, the shortest and most medial levator ani muscle, suffers the largest tissue strain. Tissue stretch ratios are proportional to fetal head size: increased fetal head diameter by 9% increases medial pubococcygeus stretch by the same amount.

It is also important to note that the pudendal nerve is also susceptible to injury during pregnancy and childbirth, suffering stretching and compression. This is a known risk factor for the development of PFDs, since it is responsible for nervous supply of most of the anatomic structures that maintain pelvic support and continence. (Ashton-Miller and DeLancey 2007; Herbert 2009).

Episiotomy - The procedure:

With the mother on the lithotomy position episiotomy should be performed on the second stage of birth at the time of maximum uterine contraction by inserting two fingers in the vaginal opening (protecting the fetus). If the woman has not received epidural beforehand, a local anesthesia should be applied before using the episiotomy scissor, or less commonly the Mayo scissors, to make an incision of 3-5 cm in length, either midline or mediolaterally. (Bettencourt B. 2003).

If the fetus does not descend, pressure should be applied in the incision site between contractions as to reduce the bleeding and the risk of postpartum hemorrhage.

After the delivery, all layers of the wound should be stitched, being the healing period of 4 to 6 weeks (depending on the length of the incision, type of incision, the suture material used and the proper wound care). The area should be assessed for any signs of trauma.

There is a crescent importance given to the procedure since the way the technique and material might have a preponderant impact on the outcome. (Bertozzi et al. 2011)

Types of Episiotomy:

There are two incisions commonly performed during the procedure: either a midline incision (from the vaginal opening towards the anus) or medio-lateral (diagonal cut from the vaginal opening towards the ischial tuberosity). The former is more used in the USA and preferred due mainly to the suture being easier to perform, a quicker healing process, lesser degree of pain, better sexual function, less blood loss and better cosmetic outcomes. The latter is preferred in South America and Europe because, although the cut does not occur in a natural anatomic plane it is associated with less high degree perineal tears (III and IV) (Thacker 2000).

It is important to access which method is better, or weather a different approach should be used in different settings.

Clinical variables of labor:

Situations that either decrease the compliance of the musculature of the pelvic floor, the vaginal opening or increase the pressure on the pavement might be indications for episiotomy. The variables commonly associated with the more frequent use of episiotomy during the vaginal delivery are primiparity, gestational age >41 weeks, augmented or induced labor, epidural analgesia use, oxytocin use, lithotomy position during fetal expulsion and instrumentation. On the other hand, maternal age <35 years or a neonatal weight < 2500g are associated with lower episiotomy rates. (Ballesteros-Meseguer et al. 2016)

Consequences and Complications:

Even though the execution of episiotomy is a relatively simple procedure it still can have some consequences or complications of either the procedure or the posterior wound or scar tissue. Although the health of the child does not suffer alteration from this procedure, there are a lot of unwanted events for the woman that may arise.

Episiotomy is associated with higher postpartum perineal pain levels in the immediate postpartum period, more stress urinary incontinence at 6 weeks and dyspareunia, but studies show recovery after 6 weeks and a complete normal function in 12 months after the procedure. (Macleod et al. 2013) A recent study (Bertozzi et al. 2011) also showed that episiotomy served as a protective factor of the pelvic disorders in terms of quality of life seeing they had better psycho-physical health status in the follow-up. It has also been speculated that it may result in an increase rate of clear cell carcinoma on the scar site (Han, Zheng, and Wang 2016) and bacterial vaginoses (Letouzey et al. 2015).

If a midline episiotomy is performed there is a higher risk of third-degree lacerations, with rare complications such as incontinence for stool and flatus and fistula-formation. If a mediolateral episiotomy is performed it is more frequently linked with impaired wound-healing. (Langer and Minetti 2006; Dimitrov, Tsenov, and Ganeva 2000; Hirsch 1997).

Other very rare consequences that may result from either the procedure or from the wound described in literature are infection, hematoma, cellulite, dehiscence, abscess formation, pudendal nerve lesion, necrotizing fasciitis and death. (Bettencourt B. 2003)

The Future:

Recent attempts have been made to decrease the need of episiotomy in vaginal birth.

Some studies have shown that exercise for the pelvic floor musculature during birth preparation might facilitate the delivery process and might result in reduced numbers of complications of PFD. (Haddow, Watts, and Robertson 2005) (Klein et al. 1997) Others have tried the appliance of a protecting device on the perineum at time of birth providing a structure to stabilize and prevent the extensive distension of the perineum, which might reduce the likelihood of developing a tear. (Lavesson et al. 2014) A device called EpiNo has also been tested to reduce the rate of episiotomy. (Kavvadias and Hoesli 2016) It consists of a device inserted in the vaginal cavity some time before birth, that will gradually distend the tissues, giving time for the musculature to adjust.

Although some research has been made it is still not clear weather their main goal is achieved and weather it leads to a better outcome than with episiotomy.

Biomechanical Overview (Oliveira et al. 2016):

With the advance in modern technology it is possible to simulate very accurately the human body and to experiment the outcome of different situations. There is a crescent tendency of using this model simulators to assess the outcomes regarding a wider range of variables.

Being that selective episiotomy is still a common procedure from either fetal indication (nonreassuring fetal heart, preterm delivery, breech delivery, macrosomia, shoulder dystocia) or if severe perineal trauma is imminent (instrumental delivery, thick inelastic perineum, previous lacerations, short perineal body), it is important to study the delivery techniques and the different outcomes.

A biomechanical numerical model widely used was applied to investigate the quantitative effects of episiotomy on the pelvic floor. For this model, it is used the pelvic floor muscles (levator ani muscles and coccygeus muscle) with a constant thickness of 2mm and assuming the muscles are fully relaxed due to anesthesia. In this study, we were able to see the resulting effect on the musculature of the pelvic floor of the mediolateral incision, varying the angle (30°, 45° and 60°), degree, length (1cm, 2cm and 3 cm, accounting for the absence of the perineum on the model) and time of incision (after the full descent of the fetal head). All these variables were compared to a delivery without episiotomy.

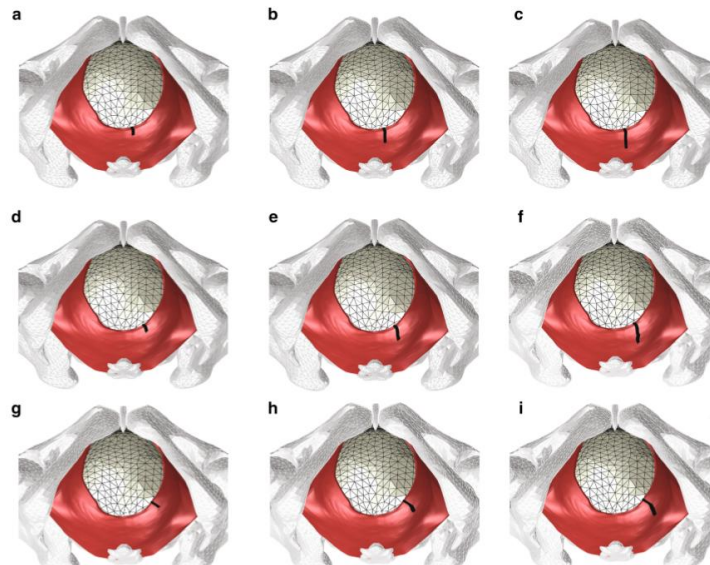


Fig1 – Inferior view of the female pelvic outlet at the moment that mediolateral episiotomy, with different extensions and angles, should be performed, i.e., after the full descent of the fetal head. The elements selected for the different situations are identified in *black color*. The figures on top are related to episiotomies carried out at 30°: **a** 10mm, **b** 20mm and **c** 30mm. The figures in the middle correspond to the episiotomies performed at 45°: **d** 10 mm, **e** 20 mm and **f** 30 mm incision. The last figures are from 60° episiotomies: **g** 10mm, **h** 20mm and **i** 30 mm incision. (Reprinted with permission from Ref 10).

It was observed that the distribution of the maximum principal stresses varies when the episiotomy is performed, changing also with the extension of the incision. In the non-episiotomy model the maximum value of stress was seen on the insertion points of the rectal area of the levator ani in the symphysis pubis. When episiotomy is simulated there is a shift of the peak stress observed on the posterior area of the levator ani.

In Fig2 it is evident that the episiotomy angle affects the amount of force experienced by the muscles, and the difference is even more evident the greater the extent of incision (longer incisions require less force of delivery).

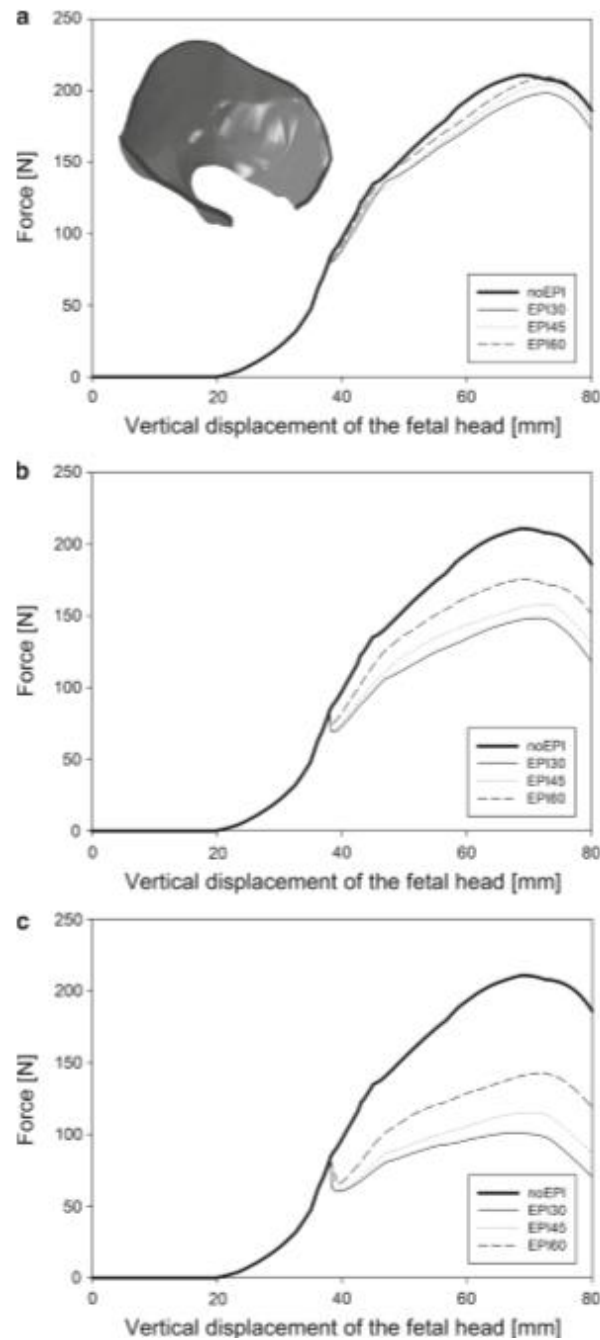


Fig2 – Antero-posterior reaction forces in the pelvic floor muscles during the passage of the fetus, measured in the nodes highlighted in the pelvic floor model, which represents the zone where boundary conditions were applied. noEPI, no episiotomy performed; EPI30, EPI45 and EPI60, episiotomy carried out at 30°, 45° and 60°, respectively. **a** 10 mm incision, **b** 20 mm incision and **c** 30 mm incision (*Reprinted with permission from Ref 10*)

Fig 3 shows us that the extension of the cut affects the amount of muscle damage, no matter which angle of incision is used. The longer the incision, the lower the damage experienced during delivery on the pelvic floor. When comparing the incision angle it is observed that the episiotomy performed at 30° angle is the most protective, since it is the one with less induced muscle damage. In all of the simulated cases, the levator ani is the most affected. If episiotomy is performed the damage is confined to the upper extremity of the incision, without episiotomy the whole muscular portion suffers.

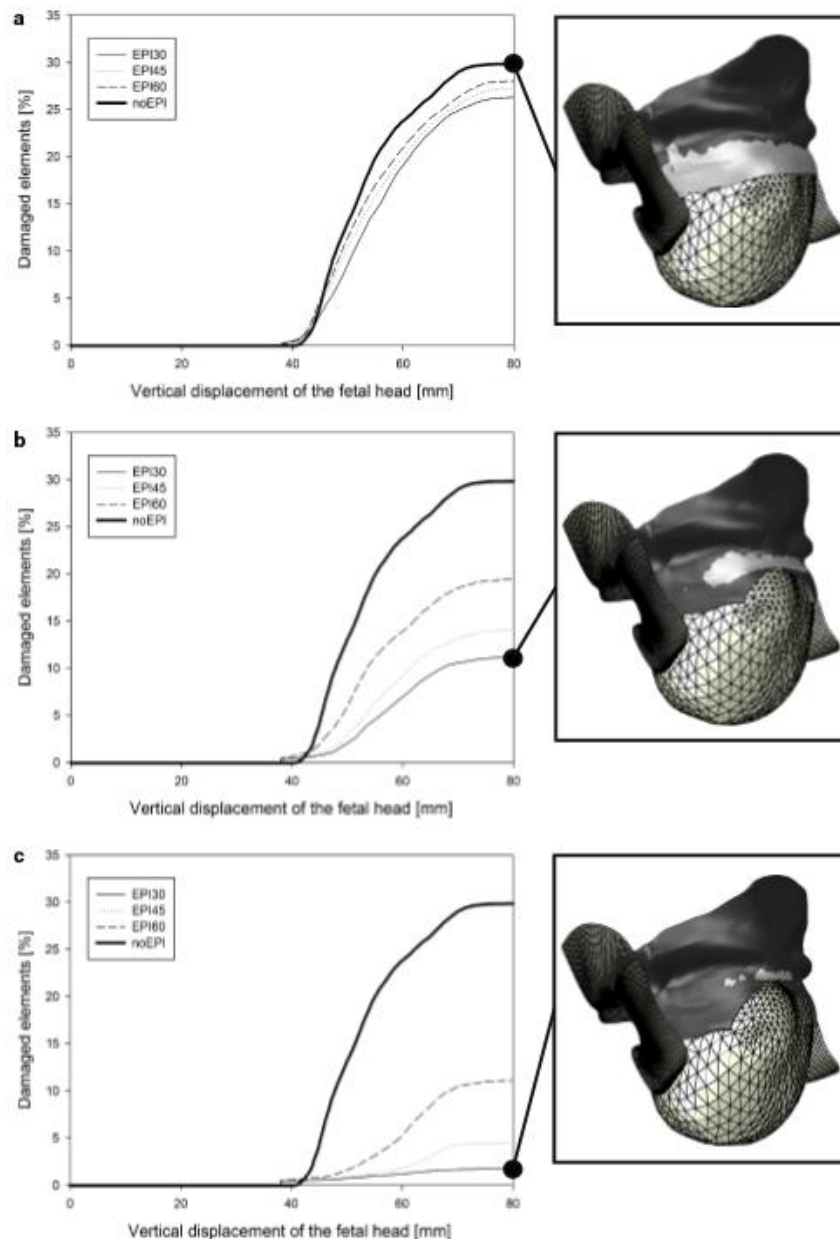


Fig3 – Evolution of the percentage of elements with fiber damage during normal vaginal delivery, and identification of the damaged region in the pelvic floor muscles, represented in light gray, at a vertical displacement of the fetal head of 80mm. noEPI, no episiotomy performed; EPI30, EPI45, EPI60, episiotomies carried out at 30°, 45° and 60°, respectively. **a** 10mm incisions and highlight of the damaged region in the pelvic floor muscles without episiotomy, **b** 20 mm incisions and highlight of the damaged region in the pelvic floor muscles with 30° episiotomy, **c** 30mm incisions and highlight of the damaged region in the pelvic floor muscles with 30° episiotomy. To simplify the images, only fetal head is showing (*Reprinted with permission from Ref 10*)

Episiotomy was associated with a decrease in the opposite forces against the fetal descent, suggesting that mediolateral episiotomy may facilitate birth, protecting the pelvic floor. This study also supports current knowledge that the pubovisceral muscles suffer greater damage and pressure than any other component of the pelvic floor. Some of the limitations for this study are the need to evaluate the long-term implications of intrapartum care (since this procedure is related to a slower recovery), the evaluation of the muscles of the pelvic floor with distinct characteristics (such as thickness and the need to include the perineum), the fact that the fetal head is rigid hindering fetal head modeling.

Discussion:

Episiotomy is still a controversial subject amongst physicians and the main aim of this study is to clarify some aspects of this procedure, contemplating the different biomechanical effects it has on the pelvic floor. The importance of such simulations is undeniable, and its clinical use has become more regular amongst investigators. It was clearly proved the role of episiotomy in reducing the stress forces on the pelvic floor. This leads to less strain for the muscles, which is a known risk factor for the development of PFDs. (Bozkurt, Yumru, and Sahin 2014; Chitra and Panicker 2011) As stated in previous studies, it should be seen as a protective measure when certain characteristics, either maternal or fetal, lead to a disproportional pressure of the fetal head on the pelvic floor. (Ballesteros-Meseguer et al. 2016; Fritel et al. 2008) Still, more studies and clinical trials should be made to establish clear guidelines when to perform this procedure.

Regarding the type of episiotomy (midline or mediolateral), due to the lack of representation of the perineum in this model, it was not possible to simulate the midline cut. Still, three different cut angles were performed (30°, 45° and 60°) and the model suggests that a more midline-like cut would be to greater advantage, since it would result in smaller forces against the musculature of the pelvic floor. This is not supported by current studies, that refer the mediolateral cut as the preferred approach, being that it leads to less complications and less risk of secondary tears or lacerations. (Carroli and Mignini 2009; Myers-Helfgott MG 1999; Necesalova et al. 2016; Thacker 2000) It is also known that the healing process occurs more naturally along the midline (due mainly to anatomical reasons). It would be therefore interesting to use a model with the inclusion of the perineum, and to study the post-cut effects on the pelvic floor and the healing process on both methods.

There aren't many known studies that compare the length of the cut used in episiotomy. In a recent study, *M Stedenfeldt et al (Kalis et al. 2012)* proved that scarred episiotomies with depth > 16 mm, length > 17 mm, incision point > 9 mm lateral of midpoint and angle range 30–60° are significantly associated with less risk of injury. The risk for injury decreased by 70% for each 5.5-mm increase in episiotomy depth, 56% for each 4.5-mm increase in the distance from the midline to the incision point of the episiotomy, and 75% for each 5.5-mm increase in episiotomy length. The biomechanical simulator showed a clear lessening of the force suffered by the pelvic floor structures with the increase of the length. But with longer cuts may arise more complications. A trial comparing the outcomes of the healing process with different episiotomies length would be important.

The effect of the timing of application of this procedure was not studied in this model, but according to recent studies (Rusavy, Karbanova, and Kalis 2016) there might be some advantage in applying the cut only when the head reaches its full descent in terms of additional vaginal trauma and blood loss.

Some limitations and prospective for future studies are the lack of consideration of tissue stresses, the simplified fetal head geometry and the lack of consideration of the difference that exists between the different components of the pelvic floor.

Conclusion:

Although it is clear the use of episiotomy is a contributing factor to the decrease of pressure on the pelvic floor during vaginal childbirth, leading to less PFD, it is still unclear the specific guidelines that should be used to apply this procedure, neither which technique should be used.

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RESEARCH ARTICLE

ACTIVITY OF ASPARAGINASE, PURIFIED AND CHARACTERIZED FROM FROZEN SEEDS OF PISUM SATIVUM.

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Abstract

Asparaginase was extracted from Frozen Plant seeds of *Pisum sativum* collected from a field crop after 3 months of freezing. Enzyme specific activity was (74.1 U/mg). Asparaginase was extracted from frozen seeds and purified by ion exchange chromatography using DEAE-Cellulose and gel filtration chromatography using sephadex G-200. We found that the molecular weight of asparaginase is 44000 Dalton, specific activity of Purified asparaginase was 226.5 U/mg. Purified asparaginase was characterized, the optimum pH for enzyme activity and stability was pH 8.5 and 9.5 respectively, while the optimum temperature for enzyme activity and stability was 40 °C and 37 °C respectively.

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Introduction:-

L-asparaginase (E.C.3.5.1.1) is the enzyme that catalyzes the hydrolysis of the amide group of L-asparagine releasing L-aspartate and ammonia. The enzyme plays important roles in the metabolism of all living organisms and the pharmacology as well (1). Asparagine predominates in the transport of nitrogen in many legumes studied so far and constitutes an important source of reduced nitrogen for developing seeds (2).

Pea (*Pisum sativum*) is a member of Leguminecea family that is widely grown in the world as a source of protein for human food (3). Green pea can be eaten as a cooked vegetable, or can be marketed fresh, canned, or frozen while ripe dried pea is used whole, split, or made into flour (4). Pea is reported to have potential antioxidant, anti-inflammatory and antimicrobial effect (5; 6).

The plant asparaginase from frozen *Pisum sativum* has been less studied. In plants, L-asparagine is the major nitrogen storage and transport compound (2). According to importance of L-asparaginase in different therapeutical treatment. Hence the necessity to find out a novel sources of this enzyme from plants this study was aimed to extract, purify and characterize the enzyme from frozen plant seeds of *Pisum sativum* in order to determine the activity of asparaginase from frozen sources of plant which could be considered as a unique case study

Materials and Methods:-

Sample collection:-

Plant seeds of *Pisum sativum* were collected during first season of 2016 from field of plant crops and freezed for (3) months at 0 °C. (7) 1

Extraction of asparaginase from frozen plant seeds:-

Extraction of asparaginase from frozen plant seeds was achieved according to Chang and Farnden (8) by homogenization, 10 grams of frozen plant seeds with three volumes of 0.05 M potassium phosphate buffer, pH 8.0 containing 1.5 M sodium chloride, 1mM PMSF, 1mM EDTA, and 10% (w/v) glycerol, then centrifuged at 10000 rpm for 20 minutes. Supernatant was regarded as crude enzyme.

Enzyme assay:-

Asparaginase was assayed according to Nesslerization method based on the conversion of L-asparagine to Ammonia and L-asparatate, as described by Ren *et al.* 2010 (9).

Protein concentration in plant extracts and enzyme concentrates was determined according to Bradford method (10).

Purification of asparaginase:-

Asparaginase from frozen seeds of *Pisum sativum* was purified by ion exchange chromatography using DEAE-Cellulose (2×23 cm) equilibrated with 50 mM potassium phosphate buffer (pH 8.0), then bound proteins were eluted with a flow rate of 3 ml/fraction by gradient concentrations of NaCl (0.1-0.5M) in 0.05 M potassium phosphate buffer solution (pH 8.0). Fractions represent asparaginase activity were collected, concentrated and applied onto gel filtration chromatography column using Sephadex G-200 (1.6×43 cm), and eluted with 0.1 M potassium phosphate buffer (pH8.0) with flow rate of 20 ml/hour. Fractions represent asparaginase activity were pooled, concentrated and kept at 4°C for further analysis.

Characterization of purified asparaginase:-

Characterization of purified asparaginase was achieved by determining the optimum pH and temperature for enzyme activity and stability, and the activation energy according to Segel (11), while the molecular weight of asparaginase was determined by gel filtration chromatography technique using Sephadex G-200 as described by Andrews (12).

Result and Discussion:-

Asparaginase activities were detected in plant seeds of *Pisum sativum* that was frozen for three months to determine the activity of enzyme. Al-Hussaini, (2012), showed that the quality of beans and peas increased after three months freezing, in spite of the content of vitamin C was decreased during freezing periods of 3 months (7).

Results indicated in Table (1) showed that asparaginase activity (571.0 U/ml), specific activity of (74.1 U/mg) and total activity (5710 U) were noticed in frozen plant seeds extract. According to these results frozen plant seeds were used for enzyme production, purification, characterization of asparaginase to determine the activity of the Frozen plant sources. Khalaf, et al., (2012), showed that asparaginase activity that extracted from fresh seeds of *Pisum sativum* var. Jof were (605.0 U) and specific activity (75.6 U/mg), (13).

Table 1:- Asparaginase activity and specific activity in the extracts of frozen plant seeds of *Pisum sativum*.

Plant part	Volume (ml)	Activity (U/ml)	Protein (mg/ml)	Specific Activity (U/mg)	Total activity (U)
Seeds	10	571.0	7.7	74.1	5710

Asparaginase extracted from frozen seeds of *Pisum sativum* was purified by ion-exchange chromatography using DEAE-Cellulose. Results indicated in figure (1) showed that one protein peak appeared in the washing step, while there are three protein peaks were appeared after elution peak as mentioned with gradient concentrations of sodium chloride, asparaginase activity was detected in the third peak in figure (1). The maximum asparaginase activity and specific activity in the asparaginase concentrate were 483.0 U/ml and 155.8 U/mg respectively, with 2.06 % fold of purification in the table (2).

Gel filtration chromatography technique was the next step for purification of asparaginase after the ion-exchange chromatography step. Results mentioned in figure (2) showed that only one protein peak represents asparaginase activity was appeared after elution with 0.1 M potassium phosphate buffer solution.

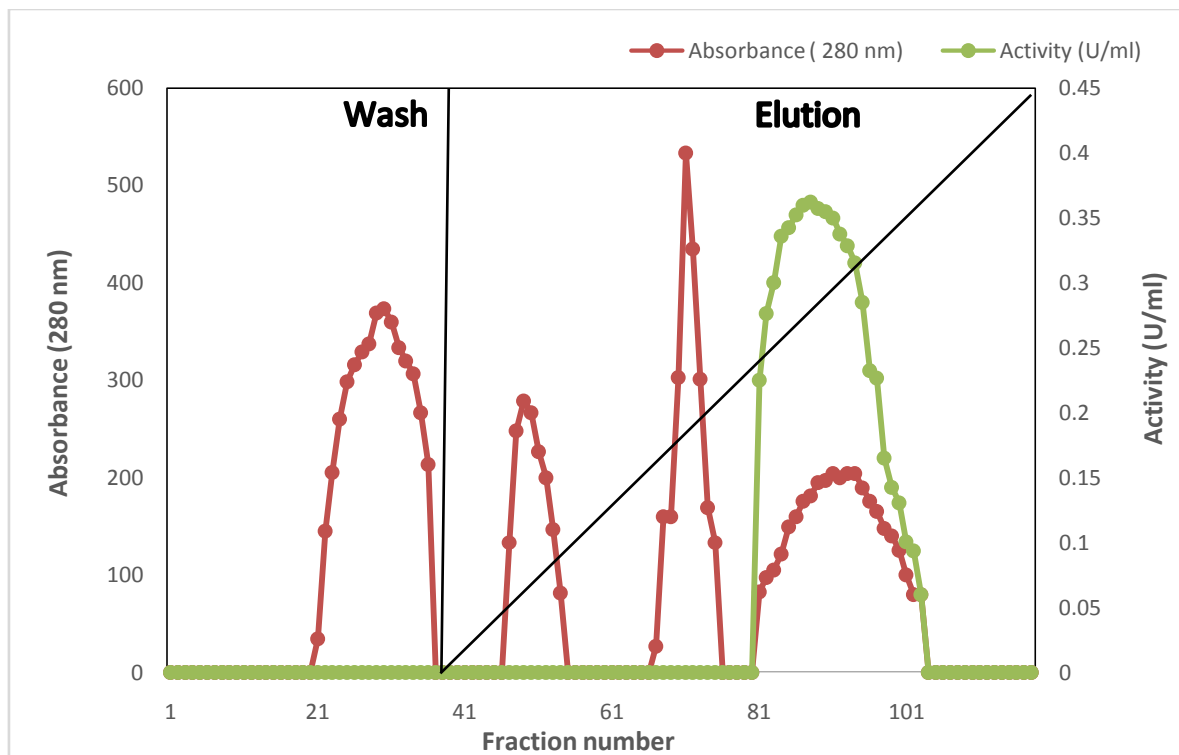


Figure 1:- Ion exchange chromatography for purification of asparaginase from frozen seeds of *Pisum sativum* using DEAE-Cellulose column (2×23 cm) with a flow rate of 20 ml/hour.

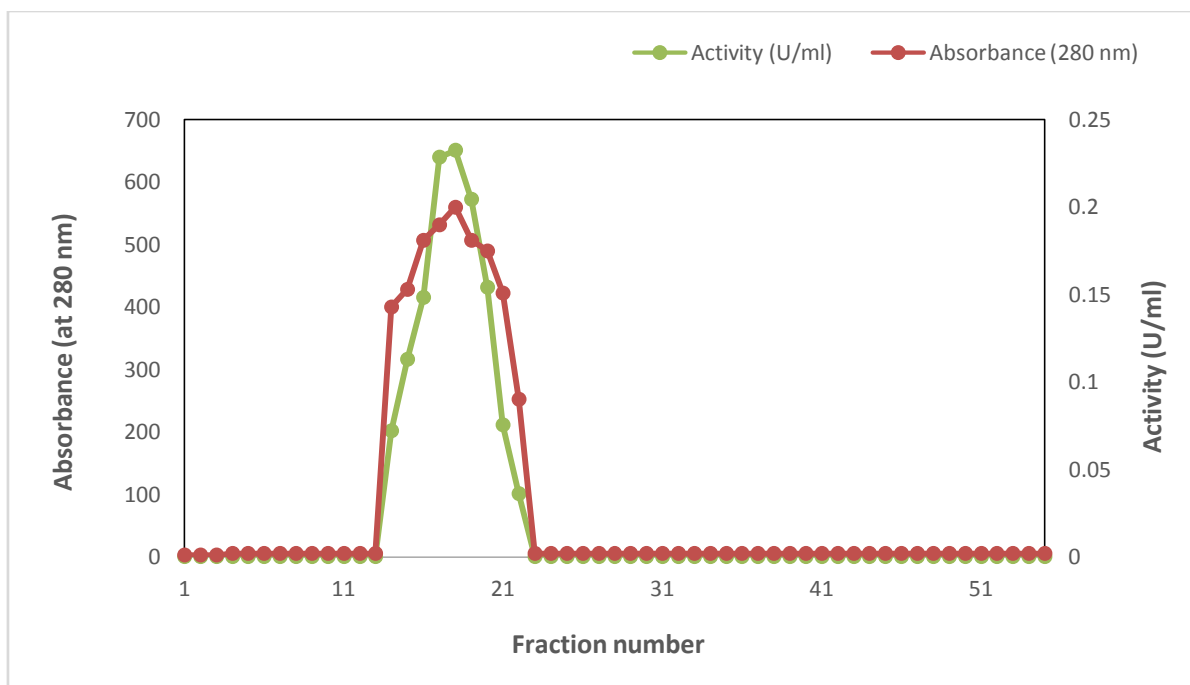


Figure 2:- Gel filtration chromatography for purification of asparaginase from frozen seeds of *Pisum sativum* using Sephadex G-200 (1.6×43 cm) equilibrated with potassium phosphate (pH 8.0), fraction volume was 5ml at flow rate of 20 ml/hour.

Results indicated in table (2) showed that there is an increase in both activity and specific activity of the purified enzyme with a purification fold of 0.013 and an increase in the yield of asparaginase to 3.05 %.

In other study, it was found that purification of asparaginase from fresh seeds of *Pisum sativum* using Sephadex G-200 as a second step (after DEAE-Cellulose Ion Exchange chromatography) gives the highest specific activity of 226.5 U/mg and 0.013 fold of purification with 3.05 % yield, (14).

Table 2:- Purification steps of asparaginase extracted from frozen seeds of *Pisum sativum*.

Step	Volume (ml)	Activity (U/ml)	Protein conc. (mg/ml)	Specific activity (U/mg)	Total activity (U)	Purification fold	Yield (%)
Crude enzyme	100	571.0	7.7	74.1	250098	1	1
Ion exchange	15	483.0	3.0	155.8	15939	2.06	0.06
Gel filtration	5	650.9	2.9	226.5	3284.5	0.013	3.05

Molecular weight of the purified asparaginase was determined by gel filtration using Sephadex G-200 in the presence of six standard proteins. Results indicated in figure (3) showed that asparaginase has a molecular weight of 44000 Dalton. Molecular weight of asparaginase differs according to the type of the produced genera, species, type of storage, and parts of plant. There was no evidence for dissociation into subunits on SDS-PAGE, and this suggests that asparaginase is a monomeric protein of molecular weight 69000 Da (15, 16).

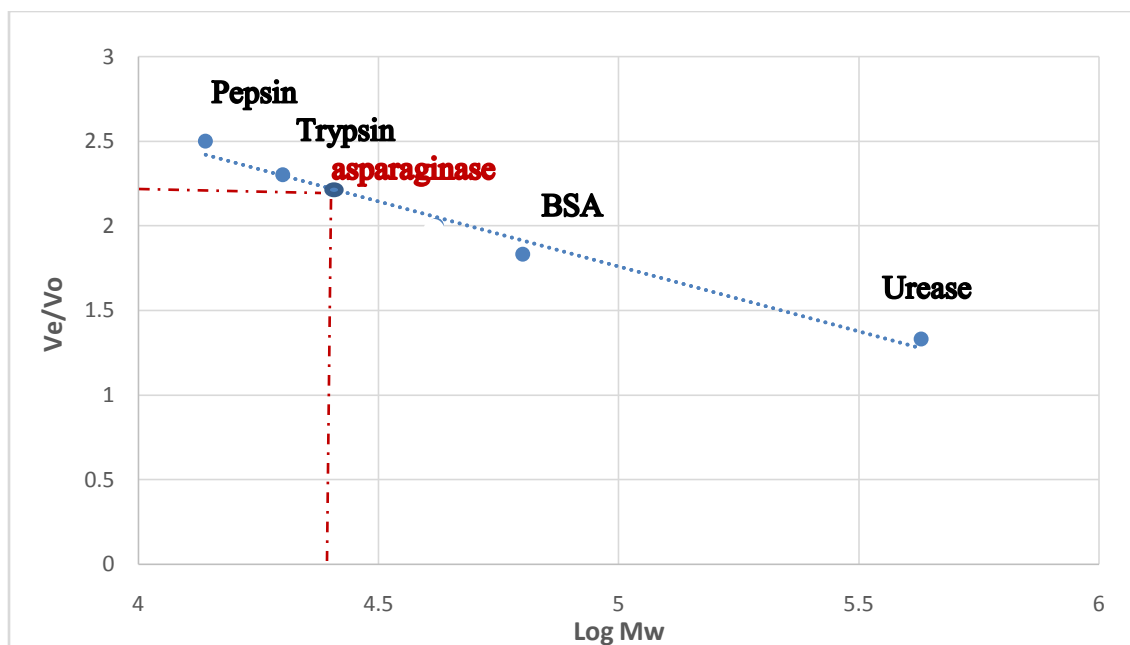


Figure 3:- Selectivity curve for determining the molecular weight of purified asparaginase extracted from *Pisum sativum* seeds by gel filtration chromatography using Sephadex G-200 (1.6×43 cm).

Optimum pH for activity of purified asparaginase from frozen seeds of *Pisum sativum* was determined by incubation with asparagine at different pH values ranging between pH4 and pH10.5. Results indicated in figure (4) showed that asparaginase was a maximum activity of 155.4 U/ml at pH 10.0. Effect of pH on the rate of hydrolysis of asparagine is due to the effect of asparaginase on the velocity of enzyme-substrate complex formation and breakdown in addition to its effect on the ionic state of the active site of enzyme since the change in the hydrogen ion concentration may affect the ionization state of amino acid side chains in the enzyme active site (17, 18).

On the other hand, results indicated in figure (5) showed that asparaginase was more stable at pH values ranged between 8.5 and 9.5, at this range enzyme gain both maximum activity and remaining activity, while the activity and remaining activity was lower when the enzyme was incubated at pH values less or more than optimum.

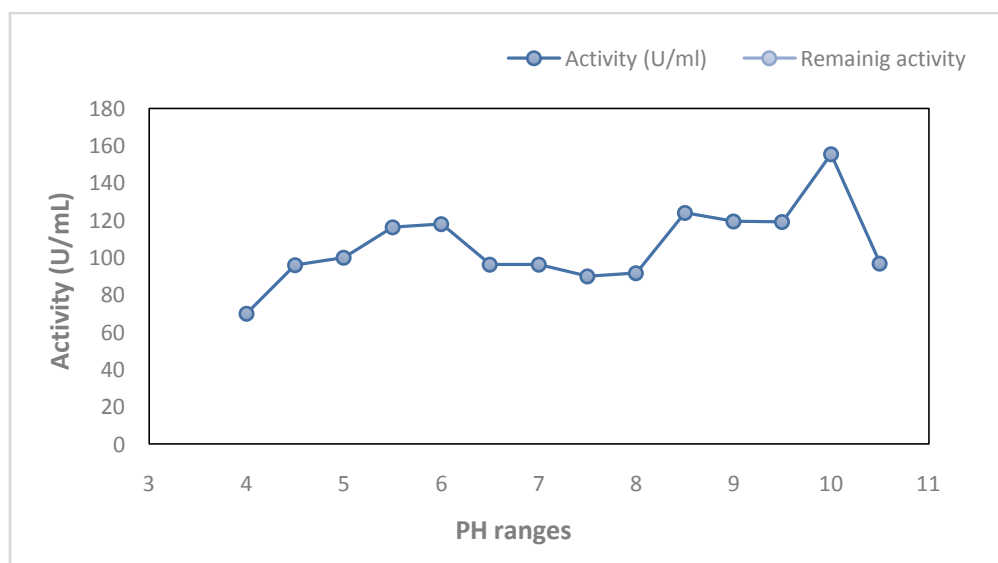


Figure 4:- Effect of pH on activity of purified asparaginase extracted from frozen seeds of *Pisum sativum*.

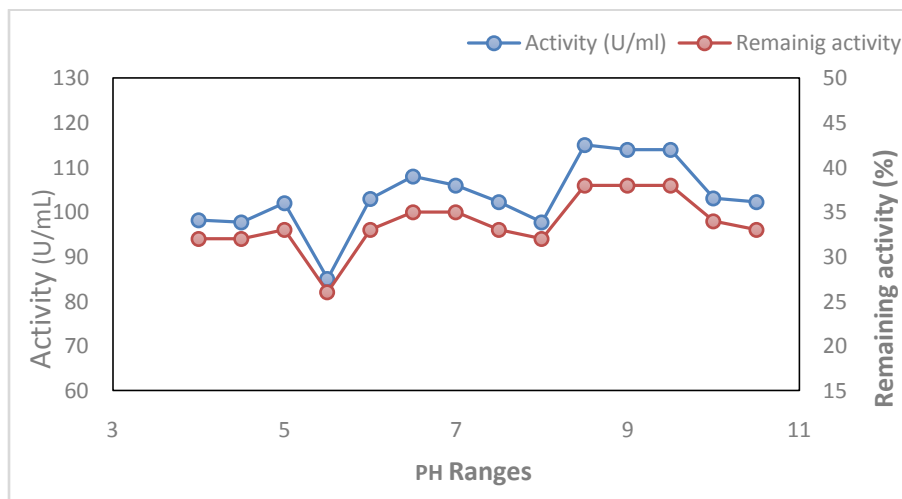


Figure 5:- Effect of pH on stability of purified asparaginase extracted from Frozen seeds of *Pisum sativum*

Saleh *et al.*, (2015) found that asparaginase from *Phaseolus vulgaris* seeds had optimum pH at 8.0. It showed that the tetrameric form of asparaginase from *Phaseolus vulgaris* seeds remain stable at pH values ranged between pH 7.5–9.0, it had high stability at alkaline pH when incubated for up to 24 h (19).

Effect of temperature on the purified asparaginase was also studied. Results indicated in figure (6) showed that the optimum temperature for enzyme activity was 524.6 U/ml at 40°C, while the activity was decreased at less or higher than this temperature. Most enzyme reactions were found to be accelerated with the increase in temperature for a limited range.

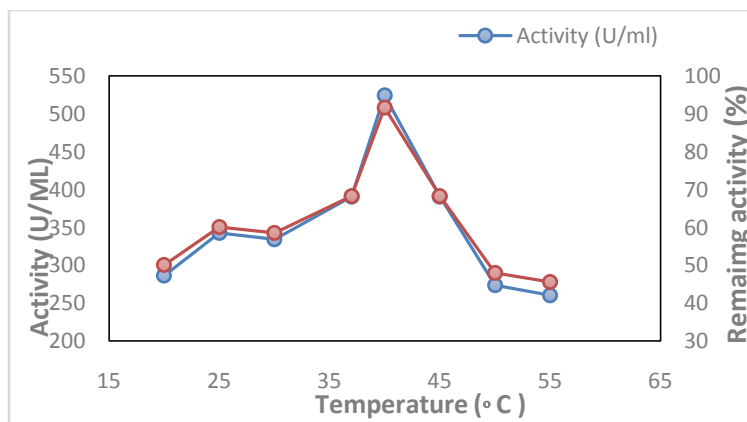


Figure 6:- Effect of temperature on the activity of purified asparaginase extracted from frozen seeds of *Pisum sativum*.

It was also found that the asparaginase stability was 624 U/ml at 37°C as shown in figure (7), at this temperature enzyme gain maximum activity and remaining activity, while they were decreased when the enzyme was incubated at temperatures less or more than the optimum temperature. The increase in temperature may lead to denaturation of enzyme by destruction the three dimensional structure of protein which leads to inactivation of the enzyme at higher temperatures.

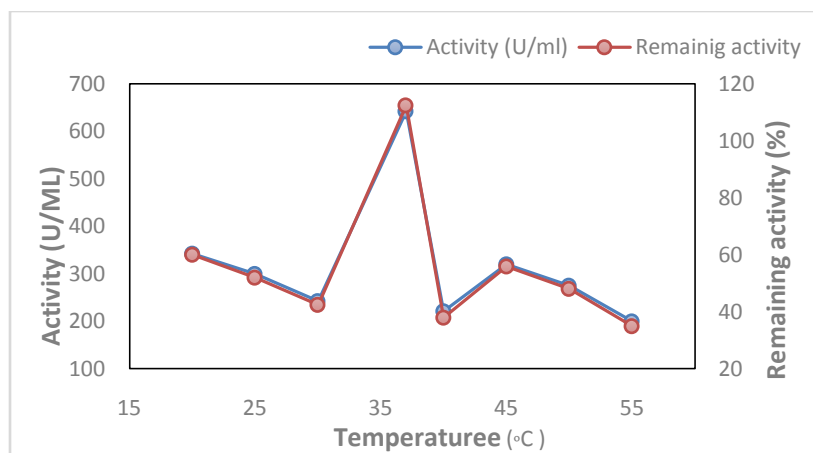


Figure 7:- Effect of temperature on stability of asparaginase purified from seeds of *Pisum sativum*.

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RESEARCH ARTICLE

DEMONETIZATION DRIVE IN INDIA AND ITS SOCIO-CULTURAL IMPLICATIONS.

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Black Money, Bribery, Corruption,
Demonetization, Fake Currency,
Surgical Strike and Terrorism

Abstract

One decision and every walk of life is deeply affected. Now life will never remain the same; the fake life, fake culture and fake currency is gone. It is affect the all shades of life. The black money plays an instrumental role in the lives of almost every section of the society in India. The black money occupies an indispensable position and it is extremely relevant at and every stage of life ranging from given gifts to all occasion or festivals, donation at school, transfer, postings in the services, bribery in recruitment, promotion, wedding of children, purchase of properties, jewelry, railway tickets, sanctioning of loans, tenders, filling of nominations, contesting elections etc. For a long term gain the short-term disruption and inconvenience and inevitable. It will pose a great challenge to our patience, resilience and character as well. This is history in making and we have to change our gears. In fact, changing of gears does not mean enhancing the speed. The gears can be change only brought by coordinated efforts at every level of management and participation.

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Introduction:-

Maximum all Indians were watching T.V. on 8th November 2016 when in the evening news spread that Prime Minister Mr. Narendra Modi was about to address the at 8p.m., no one had any idea what it was all about. After all, the first such address to the nation in his more than two years of rule, made everyone sit up and take notice. Either during the day Prime Minister had a meeting with the three services chiefs and Mr. Ajit Doval (Present National Security Advisor to P.M) in tow, so media was quick to speculate that, in view of the unabated heavy firing on the LOC and Kashmir by Pakistan terrorist attack. With media, everyone was expecting some major announcement in regard to national security and take some strong steps against Pakistani terrorism after a successful surgical strike. But Prime Minister was much more focused on the menace of black money and had come out with something that would have a deep and long-lasting impact on the country's economy. Like all, me was shocked too when P.M. announced 500 and 1000 would not remain legal tender from today's mid night after four hours they were and would be reduced to just a worthless piece of paper. He said,. " I said to my husband –**“ये सब आज पाकिस्तान के खिलाफ कुछ करने के लिए मिले थे या घर के अंदर ही सर्जिकल स्ट्राइक करने के लिए, हो गया बम बिस्फोट”**. And after that every thing changed for we all Indians. Common people run towards petrol pumps and daily needs shops. But everyone was welcoming this news. Next day morning something different for every one and all media mechanism print, electronic or social media started their TRP and humors game. "America is counting votes as India is counting notes." The announcement not only astonished the people of India but it also stole the thunder of the most waited USA presidential election. The news got attention in the almost all daily newspaper of International arena.

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New York Times- “Chaos as Millions in India Crowd Banks to Exchange Currency”

China Daily- “Indians run out of patience as banks & ATMs run out of cash, post-demonetization”

BBC- “Modi’s money medicine: kill or cure?”

The mass media is really become much powerful and in many ways they are influencing the government.

After one month, this country is in an uproar. While this is not the first time that India has attempted to demonetize in a professed bid to tackle the issue of the black money in the country, unlike the last time under Prime Minister Morarji Desai in 1978, this time Reserve Bank of India’s Governor Urjit Patel is in full support of the decision and has called it a bold move that “addresses the growing menace of fake Indian currency notes.”

It was momentous decision that sent shock waves down the whole system-as it was intended to do. The late evening announcement, and the narrow window offered before the order was to take effect, meant just about everybody was caught off-guard. Many wondered whether his move to bolster economy would pay off. But, in one fell swoop, P.M. Modi had sought to burnish his image as a decisive leader- a man given to big ideas and bold strokes – something that made him hero of the middle class with a massive electoral mandate two and half years ago. Significantly, the initiative comes only a few months before crucial assembly polls in Uttar Pradesh & Punjab, where huge amounts of illicit funds are believed to be in circulation. Sources close to the government let on that the demonetization policy-pitched as a ‘surgical strike against black money, corruption, and fake currency noted that fund terror.

Background:-

In the post –Bretton Woods world, government’s control over the supply of currency notes, has become a tool in the hands of government to control inflationary pressure by increasing or decreasing the supply of currency notes. The issues of currency notes itself was very important human invention, which monetized the world and facilitated the trade. But every good thing brings some evils with it.

For the last few years it was being observed the new currency notes issued by the RBI and infused into the banking system of the country were neither coming back to banks nor circulating in the economy. This indicated that people were hoarding currency notes as black money i.e. unaccounted and untaxed money. Due to the increase of black money in the economy government was suffering losses on following fronts:

1. Use of black money in the elections there by harming the very fundamental process of democracy undermining in the long term. Even student’s elections in universities and colleges are becoming prestigious and crossing limit of money.
2. Use of black money for funding of terror activities. After the announcement of demonetization media didn’t hype any terror activities in Kashmir. Humors spread in social media that all pelletters are busy with exchange currency.
3. Use of black money as bribes, resulting into influence of policy decisions as well leading to generation of further unaccounted money.
4. Losing track of the movement of money from one hand to another, there by facilitating unlawful and criminal activities.
5. Enabling the unscrupulous *Hawala* operators to take the wealth out of the country.
6. While black money fled out of the country or was stored in stashes, government has to resort to borrowing from World Bank and other international institutions and pay interest on it.
7. Fake currency trade has been haunting Indian securities establishment as well as economic intelligence agencies for a long time. The agencies believe that the fake Indian currency printed in Pakistan and bought through couriers via Bangkok, Kathmandu and Dhaka airports to fund terror activities on Indian soil.
8. The Political parties are spending huge unaccounted money in elections. This becomes mother of corruption. It also affects the quality governance and breed crony capitalism.

In the light of above factors, this step was a logical step waiting to be taken. Corruption, black money and terrorism are the factors that hinder the development of the economy and peace of the country. Since BJP government committed to development, these steps were considered necessary. A major economic reform was also needed, to increase the confidence of the global business community that India was open for business.

In this single move, the Government has attempted to tackle all the **three issues affecting the economy i.e. a parallel economy, counterfeit currency in circulation and terror financing**. There is no doubt that Prime Minister has pulled out a major coop and substantially enhanced his reputation as a strong leader. The P.M. hadn’t

been exactly idle on globalized corruption, and was continually exploring his menu of options. The P.M. also setup a Special Investigation Team (SIT) to look into the issue in his first cabinet meeting. His intentions were never in doubt but things take time. There were other elements that were key in his scheme of things. P.M. Modi talked on several occasions about promoting plastic money to pave the way for cashless transactions. The Jan-Dhan Project, The Voluntary Income Disclosure Scheme and GST – these were the part of grand plan. But perhaps ambitious voluntary disclosure scheme failed to unearth as much unaccounted money as the government had hoped for. “This might have prompted the push for demonetization. On September 2nd during TV interview, he said, “Don’t blame me if I take tough decisions after 30th September. For an economy where over 80% of transactions are in cash, this is traumatic shock. This is the first radical reform without a looming crisis. The last big reform of 1991 was in response to a foreign exchange crunch. In contrast, the economy now was growing at 7.1% and inflation was under control at 4.2%. There is no precedent in the world for an action on this scale and in these conditions. As a result there’s confusion about possible outcome of this decision.

A few things, however, are certain. It is difficult to oppose a move that attacks illegal money. Prime Minister Modi has flown with scheme on the wings of morality. Undoubtedly, the purge of black money and corruption will make a more honest nation out of us. This step has been taken as much with a view to curbing financing of terrorism through the proceeds of Fake Indian Currency Notes and use of such funds for subversive activities such as espionage, smuggling of arms, drugs and other contrabands into India, as with a view to eliminating black money, which cast a long shadow of parallel economy. One of the most important reason was to lower the cash circulation in the country, which is directly related to growing corruption in our country, as cash transactions are not likely.

After Demonetization:-

In order to implementation the decision of the government, RBI issued certain operational guidelines keeping in view the need to minimize inconvenience of the common people. Old high-denomination bank notes of aggregate value up to Rs. 4000/- could be exchanged at any bank branch or issue office of RBI provided a requisition slip as per the format specified by RBI was presented with a proof of identity. Similar services were also made available in post office. In starting the exchange process was supposed to close on 30th December 2016, it but closed earlier and people can deposit old currency in their account. Petrol pumps, gas, CNG, govt. hospitals, railways and airline booking counters, state govt. recognized dairies and ration stores and crematoriums were to accept old 500/- & 1000/- currency till 15th December 2016. However guidelines have been changed frequently depending on the changing circumstances.

As India lurches into the 2nd months of its tryst with demonetization, it has quickly evident that this is an unprecedented onslaught on the poor. The fallout of the decision of the NDA government to demonetize currency of higher denominations has been felt across all sections of people. There are concerns that it will lead to an overall economic slowdown given the acute shortage of currency for industrial and agricultural operations.

Immediate Problems:-

1. In rural economy “cash is king” and thus reduction in cash flow has hit the villagers badly. Poor penetration of Internet and low level of digital literacy are among the factors that make people depend on cash for transactions. Its impact would be on multiple spheres: agriculture, production, rural incomes, rural demand and rural credit.
2. Business has fallen from Rs. 8-10 crore earlier to just Rs. 4 crore at present. The ‘demon’ in demonetization is in the beginning. As almost 45 days passed and scenes such as this have become all too common, the country struggling to come to terms with Prime Minister Modi’s black money hunt, the biggest such since independence. Various agencies have estimated black money at roughly 20% of India’s GDP.
3. There has been apparent inconvenience to the common man initially. 8 million people every day in queue in front of banks, ATMs & post offices across the country patiently waiting to collect a fraction of what was legitimately theirs. December came and nothing changed. Considering the scale on which an entire population remained literally on its feet, which also resulted in nearly 100 deaths as a reported by media from across the country. As per Opposition parties, more than 50 people have died which can be considered as after effects of demonetization operation.
4. Economist said sectors with large exposures to cash like real estate, transport, and food could witness “downward price pressures”.
5. About 80% of the Indian working population is engaged with micro, small, enterprises and informal sectors. This sector could be adversely affected. The unintended disruption can be huge in the cash-driven economy- particularly the informal rural sector where millions of consumers and small traders rely on cash for the daily

transactions. Moreover, nearly half the population still does not have a bank account. Less than 300 million use the Internet, and therefore the overwhelming majority cannot make electronic payments.

6. It is also believed that demonetization is not a permanent solution but it is just like one shot and it will come back into the system again with full force. The demonetization does not solve the problem of the process, which created it. The system that created the black money will remain in place unless that becomes our next attention point. The biggest question is that if 13-15 crore money deposited in banks than where is black money? Might be the answer is here- after using following left out loopholes resulting into irreparable loss: –
 - Government has declared the deposits up to Rs. 2.5 lacs in normal bank accounts & up to Rs. 50000/- in Jandhan accounts without any scrutiny.
 - There are crores of accounts of various individuals in different banks. These accounts can easily be used for conversion of Black Money into White by allowing free flow of deposits below Rs. 2.5 lacs without scrutiny.
 - Our country has a population of 125 crores & if ID cards of 1% people is used than much more than the whole black money in cash can be converted by this mode only. It is pertinent to mention here that up to first 10 days there was no bar on use of one ID card in 'n' number of Banks / branches.
 - Black money can easily be deposited or used showing it Agriculture & other exempted incomes.
 - There is an increase in purchase of Gold in Tons & other costly items for cash both before & after the decision.
 - Another method of converting black money is deposits shown by Religious places like Temple, Gurudwara etc.
 - Forget class struggle. Trust became the social capital, as the poor stood in bank queues for the rich to deposit wads of cash – all below the unquestioned magic amount of Rs 2.5 lakh-into their Jan Dhan Account to be returned, with cut or commission, in due course.
 - India rush to railway counters to book expensive long-journey in old notes. Following cancellation, the refund came into new notes.
 - Since accounts of political parties do not come under RTI or Income Tax, they don't have to disclose the reasons of such deposits. The whole money in cash can be deposited in these types of accounts.
 - Businessmen have paid employees salaries for the next several months- all in old notes and below 2.5 lakh each, the deposit amount banks accept with out question. As media reported that in Gujarat some employees had to leave their debit cards with the as surety.
 - Tax-dodging doctors running flourishing hospitals didn't just pay advance salary to employees; they also placed massive orders for medical supplies and equipment-all in old notes.
 - Other means that are being used for converting black money into white is:
 1. Conversion of old currency into new one by commission agents at 20-40% conversion charges.
 2. Gold being purchased in the four hours window between the announcement and buying gold till mid night as media reported jewellery shops.
 3. Property being purchased & sold against old currency.
 4. Even after 8th Nov Govt allowed to use old money for various payments including Govt Payments / Taxes / Payment of Bank loans & some others. How Govt ensured that use of old currency notes at these places is not use of Black money.

It is pertinent to mention that if the Government does not stop generation of Black Money than this demonetization exercise will not be fruitful & may result into another Big Scam. Even no effect on big fishes & they may escape unhurt. Moreover as per recent estimate only 6% of the black money is kept in the form of cash and therefore much more can be converted & the said objective of the Government may not be achieved. A large quantity of new currency uncovered and caught by Income tax and Enforcement Directorate raids across the country after demonetization have confirmed the fact that demonetization is not the last and whole sole alternative of eradicating black money. Earlier also demonetization was implemented in nine countries & failed to achieve the desired objectives of curbing the black money in majority of cases.

Though this move is not less than “ Misguided Missile fired in Haste” just to have political gain specifically in forthcoming state election but I think we should support in the smooth implementation of the move considering bigger interest of “Great India” as decision has already been taken by Government elected by us only & roll back now will be another disaster.

Fake Currency & Terrorism:-

Terrorism is a frightening threat, so many have lost their lives because of it. Enemies from across the border are running their operating using fake currency notes. It is a part of a larger stratagem, a low-intensity war against the Indian economy by Pakistan's deep state. This strategy gathered pace over the past decade, and counterfeit notes

began flooding the Indian market. The NIA (National Investigation Agency) revealed a nationwide network injecting fake currency notes into the economy. It got the notes forensically analyzed and discovered they were manufactured in regular currency-making machines owned by a sovereign government. It didn't take the NIA long to establish that these machines were operating across the border, in Pakistan. The Indian Statistical Institute, Kolkata, in a report in June this year estimated, that Rs 400 Crore worth FICN (Fake Indian Currency Notes) was in circulation. Between 2012-2015, the government seized counterfeit currency. However it is the single biggest move in recent years to strike at fake currency. It disrupts the production of FICN in Pakistan, and makes redundant existing stocks of fake currency with a vast network of terror funders-the Hawala traders and money launderers. The phase out of these notes is double whammy for Pakistan. It limits the impact of counterfeit notes on the economy and curtails terror funding through FICN circulation in India. While removing black money, in the process it [demonetization] will definitely address the problem areas of terror financing and fake currency. There is illegal money behind terrorist funding. The channel through which the adversary sends in militants and arms is also used to send fake currency." demonetization would impact the terror financing, particularly in Jammu and Kashmir. Terrorist outfits like Hizbul Mujahideen collect donations in Pakistan and then route the money into the State through Hawala operators. "The terror funding module in place right now, will be affected as the terrorist operatives always store money in the form of big currency notes. That money has become a piece of paper now,"

No doubt there are fake currency notes in circulation. But fake currency, which is in circulation, is which good faith common man in good faith is receiving. So they will suffer not those who are actually culprits for printing & circulation. Other type of fake currency is, which is not in circulation will be dead money for them but this cost them just printing cost nothing else. Moreover Fake currency dealers will not print how Govt will ensure that New Currency Notes specifically Rs 2000 notes. We should understand this is not Rocket Technology. Even new currency notes of Rs. 2000 (though not fake) were found with the terrorists killed recently during encounter. Future stoppage not ensured. And daily we watch news in TV on new fake 2000 currency.

Media's Role:-

One wonders, why the media, especially the TV news-channels, do not focus more on the impact of demonetization on key sectors of the economy. The problem is that sections of the media appear to be obsessively focused on how the political parties will perform in elections which, in India, seem to be taking place at regular intervals in one state or the other. Some national TV news-channels keep coming up with opinion polls and not just in the states where the next round of assembly elections are due. These channels also give us once-in-six-months opinion polls which seem to tell us how many Lok Sabha seats the ruling and opposition parties and alliances could win if general elections were held at that point of time.

Which could be why many TV news-channel discussions focus not so much on the economics of demonetization as it affects the people but the political impact on the ruling and opposition parties. Instead of economists and representatives of non-political associations of farmers and of trade and industry (including those belonging to the medium and small-scale sector and which could be more affected by a liquidity crunch), the TV news-channels have spokespersons from the BJP, the Congress, the Left, the SP, the BSP, the JD (U) and the AAP. The next round of assembly polls could indicate how the people feel about demonetization in those states where elections are due. However, the real media focus could be on whether the people and especially the vulnerable and economically weaker sections throughout the country are able to get through the present before they reach the safety of a future when things will hopefully get better.

Five years ago, during the peak of Anna Hazare & Baba Ramdev led agitations against corruption, public anger across India was palpable. Literally millions of people in all cities and towns of India would throng local protest destinations with many of them joining in the evening after finishing their day jobs. 'Celebrated' news anchors like Barkha Dutt of NDTV who had then recently covered civil unrest in far off countries like Egypt were unable to even visit these protest sites because public anger was also directed towards them. Majority of the media narrative of that time was more stooped in disbelief laced with skepticism about the impact of these agitations on "real India".

After, 5 years, once again there are crowds thronging to banks and ATMs in millions every day mostly in an orderly fashion. Most Indians have been heavily inconvenienced due to government's demonetization, yet there are no mass protests in the cities and towns of India. Today, the media narrative though is the exact opposite to what it was some 5 years ago. Media, news anchors and columnists are working overtime to build a narrative of a manufactured anger

of the masses but with little or no impact on the ground. The contrast from 2011 to 2016 in the interplay between 3 primary players – media, public and government – couldn't have been starker. While public and govt. were at loggerheads in 2011, media was by and large with the govt. whereas in 2016, public and govt. seem to be in harmony while media is seen to be against the government (media here mostly refers to the dominant mainstream English language version). These are interesting inflection points, which suggest demographic pattern shifts in tandem with a media created dystopia.

The global attitude survey by PEW research shows a clear pattern of Indian public anger of 2013 turning into contentment of 2016, which has been consistent with our own polls and data over the last 3 years. Prime Minister Modi has consistently brought about an attitudinal shift in public's perception of the government. Globally, one of the biggest cause of public anger today has been because of the building distrust in institutions – we have seen just this month how voters in the US, the lone surviving superpower, have shown their massive distrust in institutions by electing a complete outsider to challenge the establishment.

“Yeh Modi kissi ka sagga nahi hai” averred a card carrying Youth Congress worker over a cup of tea near Meerut when he was explaining to us about how possibly the trader community which was supposed to be the core BJP-RSS support base would react adversely to the central government's demonetization move. That same Youth Congressi then added as an afterthought, *“Yeh (PM Modi) shayad desh ke liye kuch bhi kar sakta hai”*. His broader analysis was basically that Modi could have hurt his own party by doing the “right thing” due to the resultant anger among BJP's core voters.

In this reaction of even ideologically opposed sections of the populace towards the Prime Minister, we find that quantum leap that Modi has taken over other politicians. In an environment of cynicism where ordinary people whose default perception mode is to simply disbelieve a political leader meet the best of intentions by a politician with doubts and conspiracy theories, Modi is slowly bringing back that much needed trust. At a fundamental level, the demonetization of 500 and 1000 rupee notes is about bringing back faith in the institutions to ordinary Indians who simply believed everybody is corrupt.

“Very quickly all the pain of standing in queues would be forgotten, but these new 500 and 2000 rupee notes would keep reminding this generation every day about Modi's fight against black money” was how a retired government teacher standing in a long queue of an ATM in Mohali summed up the situation to me. Essentially, the reason why vast majority of ordinary Indians are willing to bear this short-term pain of sudden cash crunch despite no tangible gains is because they have taken that leap of faith with Modi who is trying to restore trust in institutions once again.

In fact, that same PEW research data (survey conducted before demonetization) reveals a subplot of the larger Modi story. Among various positive aspects of Modi's prime minister ship, there was one possible sore note – only 49% people believed that the PM had shown an ability to “get things done” while 33% felt he had failed in that aspect. A perception was gaining ground that despite his best intentions; the PM was being bogged down by the system around him. We believe that Modi understood this much better than anybody else and in a matter of 1 month he has probably completely changed that perception by first conducting a surgical strike against Pakistan and then boldly going ahead with demonetization.

The real focused concern of the media could be on how demonetization affects the people and not just political parties which rely on ‘anonymous’ cash donations and which may have been hit by the November 8 scrapping of 500 and 1,000-rupee notes. The print, electronic and social media has been praising Prime Minister's masterstroke by which he has reportedly destroyed the base of corruption in India. Before the advent of TV as a medium of information, the information requirements of most household was met by the daily newspaper, commentary and analysis through weekly or monthly magazines. Decades back, Marshall McLuhan, at infancy of the television medium had understood its intrusive character. Indian television, both English and vernacular, which started of mostly to entertain has now become increasingly didactic, stridently so. It perceives its role as one of ‘engineering change’. While many anchors pursue their own hobbyhorses, the compulsion to ‘convert’ the audience to there own perspective in increasingly evident. While TRPs the operative mantra, it is subtly intertwined with the anchor's desire to project loudly a particular point of view. It is interesting to see how each such public affairs chat show has its own character, depending on the anchor, participants, issues and relative political temperature in which is discussed. Some debates are dominated by emotions, some by facts, others by logic or reason- however, partnership is the permanent flavor. It is equally interesting to note how the nature of coverage by the media, subtly changes

with perceptions of changing political fortunes of parties and personalities. In case of demonetization media show different stories for example on Zee News 'nobody suffer' by this demonetization, AajTak showed people are suffering in front of ATMs and bank and people died in row reported by NDTV.

In social media lots of jokes and messages about the impending failure of government policy on demonetization are being shared. One point which is expressly missing in all these messages is the need for introspection by "we the society". No policy is perfect and no implementation can succeed without support from the very stakeholders for whose good it is meant. "We the people of India" are determined to make this initiative to failure. Here 'We' simply means, we Indian as a society. Every morning the newspapers report of cash being found (hoarded), in old and new denominations. The amounts are so large that I cannot relate to the numbers. Bankers, Bureaucrats, Politicians, Police, Builders and Cooperative Banks the list is unending. It seems that who so ever has got an opportunity to cheat the nation, has cheated, using his/her full potential. Even the poor common man, who allows his bank accounts to be used or the jeweler who back dates invoices. It appears that 'we' are determined to make the initiatives fail...and then gloat and "blame the government".

The failure will not be the defeat of the government, nor will it be a defeat of initiative. It will defeat of our India by 'us', its citizens. It's very sad. There are always two sides of coin. Don't try to look at things theoretically or from one side. Science also says "Balanced Diet is good for health." There are some channels, which are showing only good face of decision; some are showing mixed or other sides. But it's a practical approach to see real ground level reality.

Conclusion: -

It is no coincidence that 'Notebandi', the demonetization of high-value notes by the P.M. Narendra Modi Government, sounds a lot like 'Nasbandi', the infamous sterilization drive during the Emergency under Indira Gandhi. With long queues outside ATMs that seemed perpetually dry, the country has almost been cashless since P.M. Modi announced the demonetization on November 8, 2016. Ironically that timed perfectly with his campaign for a cashless India or as he twisted it, a less-cash India. Black money will soon be generated in new currency. This may be why the government narrative has changed from unearthing black money to making India a cashless economy. If it is to be considered a modern nation. Again, a laudable goal! At present, minuscule 2% transactions are digital, compared to advanced economy like the US, where it is 45%. It is not clear how much of the cash RBI intends to replace. But if the cash crunch continues, this will be a prolonged painful process, causing much suffering to the poor. The challenge for India to make more digital payments is enormous. The reflections are showing. India is a country of *Jugaad*, as seen with money launderers. As IT Minister Ravi Shankar Prasad is fond of saying: 'Indians first observe technology, then they adapt it, and after that they adopt it. Hopefully this is true. As cashless economy became the main focus, the talk of black money and counterfeit currency took a second place in the government's scheme of things.

Following are some effects after demonetization media didn't show and hid from people-

1. 100% Fake currency out of circulation in one stroke and Hawala sources from outside and Inside dried up for Funding of Terrorists, Maoists, Naxalites, Jihadis. Cash to create chaos and terror lying with Terrorists, Maoists, Naxalites, Jihadis gone waste.
2. Kashmir back to normal in one stroke. AAZADI forgotten. Children back to school. 95% turnout in J&K for exams and no schools have been burnt or no stone pelletiers to be found.
3. Jan Dhan accounts are full with money (more then 1 lakh transaction will cease to be BPL account)
4. India has around 13 lakh crore denomination of old 500 and 1000
5. 6000 crore cash deposited by only one Gujarati businessman. He has paid around 5k stipulated tax and penalty also on it.
6. All business men are depositing cash lying with them as current your income with ADVANCE TAX
7. All Jewellers are being issued Forms to declare their GOLD STOCK from 8th November onwards on day-to-day basis and all Jewellers whose stock doesn't not match with old record will be screwed big time.
8. Via Jewellers all those who have bought Gold will also not escape Income Tax net.
9. 55 lakh money disputes settled in one day in Lok Adalat.
10. Defaulters of Banks, Property Tax, Electricity bills, Telecom bills are clearing their long pending dues.
11. Defaulters of all kind of Govt taxes are clearing their dues
12. Govt moving towards Target of Cashless economy

13. Lot of small vendors has started using Apps like Paytm, Wallet facilities. Also installing Credit /Debit machines.
14. Banks are asking and guiding people to become cashless.
15. Businessmen with less black money declaring their money as Current your income and depositing with Advance Tax.
16. All with loads of Black money in cash either will surrender money with tax and Penalty or destroy it.
17. Next cleaning going to start with declaration of Benami Properties
18. Only 2 options –Either claim property and pay tax or give it up
19. All those people who are exchanging money of rich for 30% commission will lose their BPL status if transaction in their account is more then 1 lakh. All others will have to pay tax and penalty on money deposited above 2.5 lakh.
20. Fiscal deficit of India set to reduce. Even it will lower the prices of essential commodities, help in reducing the interest rate, contain inflation and not only fiscal deficit but also help in lowering CAD (current account deficit) and narrow down the trade deficit.
21. Banks have started mobile ATM for Hospitals.
22. 19 Banks have come together to start Unified online transaction.

The demon in demonetization is in the beginning. The need for the government to keep the move a secret so that tax evaders wouldn't be alert before the demonetization took place affected preparedness. This is no doubt that the coming months will be painful for the common man, small business housewives, etc. as there will be some shortage of legal tender /currency that will have an impact on them. Online trade can also be affected. There is some trouble for the economy, but not too long lasting said Ajit Ranade (Chief economist with Aditya Birla Group). Another rising issue will be that POS machine, debit cards which has to be resolved first, otherwise the measures will become an obstacle rather than the solution.

But after having discussed so much, will this major eliminate all the black money in the economy? This answer simply would be confirmative NO as it has been seen that black money is stored in various forms other than cash and as per 2012 report prepared by National Institute of Financial Management on unaccounted income found that cash was the least preferred option for storing unaccounted wealth. P.M. Modi has endeavored to strike at the root of black money menace and silence his critics to an extent. But his biggest challenge will be to ensure that the ship of state recovers quickly from these sudden shifts in policy direction. Demonetization, though welcomed by a largely enthusiastic public response in support of strike on black money, later invoked criticism on account of inconvenience caused by it to various section of the society. Yet, such criticism is also muted and overall, the sentiment has been of support to the government's initiatives with suggestions for improvement in implementation. It exhibits people's trust in the action of the government and it needs to be pointed that such trust is hard to be earned and easier to lost. The possibility of later would be a tragedy not only for the political dispensation at helm but also for the future of political and economic reform in India.

If these measures are implemented efficiently then we can expect higher collection of taxes, higher investments in the market, price corrections, improvements of India in some of the international rankings prevent corruption practices etc. and these are some of the reasons why people although are going through difficulties are lauding this measure of the government.

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RESEARCH ARTICLE

SCREENING OF ALKALINE PHOSPHATASE ACTIVITY OF PHOSPHATE SOLUBILIZING FUNGI FROM RHIZOSPHERE SOIL.

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Abstract

Soil is a mixture of organic nutrients, which provides nutrients for plant growth. Plants require N, P & K for their growth and vegetative propagation. Among vital nutrients, Phosphorus is less available to plants and microorganisms aid in phosphate solubilization. The present study is an ecofriendly method to induce phosphate solubilization in Rhizosphere soil. Totally 25 isolates were isolated from rhizosphere using Pikovskaya medium and screened for phosphate solubilization using Tricalcium phosphate supplemented minimal medium. Of these, *Aspergillus* sp and *Penicillium* sp were found to produce remarkable solubilization index. Hence these two isolates were used for further assays. Enzyme assay was carried out to determine the enzyme activity using cell free extracts and the activity was analyzed by spectrophotometry. In addition, HPLC also performed to determine the end products of phosphate solubilization. The results of HPLC studies revealed that tricalcium phosphate was solubilized into organic acids like citric acid, gluconic acid and malic acid. The results of present study are an evidence for the production of organic acids by *Aspergillus* sp and phosphate solubilization which induce crop improvement as well as sustainable agriculture.

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Introduction:-

Phosphate is an important nutrient for plant growth and development. All plants require phosphate for metabolic activities like nucleic acid synthesis, respiration, energy production, energy storage and transfer, cell division and cell growth. Phosphorus fertilizers when applied during the early stages of plant growth promotes early root formation, and is important for development of primordia for reproductive parts of plants. Seed formation requires phosphorus and its content is higher in seeds than in any other part of the plant. It improves survival of plants in winter climates. Phosphorus is available in rocks primarily as minerals like oxypatite, hydroxypatite and apatite. These are highly insoluble and constitute about 40% of total phosphorus in Indian soils. Indigenous microorganisms convert insoluble phosphates into soluble phosphates and aid in the bioavailability of phosphorus. Phosphate solubilizing microorganisms assimilate phosphorus and release them to plants. Phosphate solubilization is a complex phenomenon which highly depends on many factors such as nutritional, physiological and growth

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conditions¹. Bacteria such as *Pseudomonas*, *Alcaligenes*, *Rhizobium*, *Serratia*, *Erwinia* are capable of phosphate solubilization².

Mechanism of mineral phosphate solubilization by PSB strains is associated with the release of low molecular weight organic acids³, which through their hydroxyl and carboxyl groups chelate the cations bound to phosphate, thereby converting it into soluble forms. There is experimental evidence to support the role of organic acids in mineral phosphate solubilization⁴.

Phosphate solubilizers possess enzymes that convert insoluble phosphate into soluble phosphate. Phosphate solubilizers like soil fungi are capable to produce extracellular enzyme, i.e. group of phosphatase enzyme which are able to mineralize organic P into inorganic P so that high P is available for plant. There are several soil phosphatases and the most commonly determined are: phosphomonoesterases, phosphodiesterases and phytases. Phosphomonoesterases act on phosphate monoesters and according to their optimum pH are divided in acid and alkaline phosphomonoesterases⁵. Both are adaptive enzymes: acid phosphomonoesterase predominates in acid soils while alkaline phosphomonoesterase predominates in neutral and basic soils⁵.

An ecofriendly approach towards converting insoluble phosphates by potential phosphate solubilizing fungi from the soil is inevitable. The present study was designed to determine alkaline phosphatase activity of indigenous fungi from the natural rhizosphere soil.

Materials and methods:-

Isolation of phosphate solubilizing microorganisms:-

The serially diluted soil samples were placed on standard agar medium (pH 6.8-7.0) containing tricalcium phosphate (TCP) as sole phosphorus source for selectively screening the bacteria which have the ability to release inorganic phosphate from tricalcium phosphate. After 3 days of incubation at 25°C, phosphate solubilizing fungi developed clear zones around colonies. Colonies with clear zones were further purified by replating. The isolation of phosphate solubilizing fungi by serially diluting the soil sample and plating method using Pikosvikyos agar medium supplemented with Tricalcium Phosphate.

Identification of phosphate solubilizing Microorganisms fungal identification:-

The morphological identification of fungal isolates were done by the methods described in experiments in Microbiology, Plant Pathology and Biotechnology.

Phosphate Solubilization activity:-

(Ngugen *et al.*, 1992)

The bacterial and fungal isolates were screened for inorganic phosphate solubilization. A loopfull of fresh bacterial and fungal cultures were streaked on to Pikosvikyos agar medium inorganic phosphate and plates were incubated at 28 ± 2 °C for 3 days. After 3 days, the colonies showing the clear halo zone around them indicated solubilization of mineral phosphate. Phosphate solubilization activities were screened by measuring the clearing zone surrounding the developed bacterial colony via calculation of phosphate solubilization index (Nautiyal, 1999).

Phosphate solubilization Index = $A/B \times 100$.

A= total diameter (colony + halo zone).

B =diameter of colony.

Identification and Characterization of Phosphate Solubilizing fungi.

PDA was used to accelerate the growth rate and the production of enough conidia as reported by Diba et al. The characteristics of fresh cultures were compared with mycological identification keys and taxonomic description to identify the isolated fungi to the genus level. Identification was based on colony characteristics and microscopic features, among the colonial characteristics such as surface appearance, texture, and colour of the colonies both from upper and lower side. In addition, conidia, conidiophores, arrangement of spores, and vegetative structures were determined with microscopy.

The identified fungi were maintained on Potato Dextrose Agar (PDA) slant at (4°C) for further investigation. Slide culture was prepared in order to identify spores and mycelia of pure fungal isolates and identified by lacto phenol cotton blue staining using microscope and identified after growing them on slide according to Stevens et al

Results and discussion:-

Fungal isolates were identified by observing colony characteristics on RBCA plates. Growth pattern of the isolates were identified as *Aspergillus* sp and *Penicillium* sp (Fig 1,2). Further it was confirmed by microscopic analysis of colony using lacto phenol cotton blue staining method.

Solubilization index for the 24 isolates was in the range from 0.5cm-0.10cm. solubilization index for *Pseudomonas* sp was observed as 0.9cm^{10,11} and present study also revealed that fungal isolates was 0.6cm for *Aspergillus* sp and 0.5cm for *Penicillium* sp.

In liquid media, fungal isolates produced acid in high amount which is evident from decolourization of broth. Phosphate solubilization by these isolates was observed for 28 days. Quantitative estimation revealed that solubilization initiated after 3 days and was highest on the 7th day. Decrease in pH was observed in all the isolates. Phosphate solubilization was observed in the range of 100µg ml⁻¹ to 250µg ml⁻¹. Maximum phosphate was solubilized by *Aspergillus* sp (190µg ml⁻¹), and also it produced significant amount of citric acid, gluconic acid and moderate amount of oxalic acid. Organic acids were also produced including succinic, glycolic and malic acid in small amounts. *Penicillium* sp solubilized phosphate (150µg ml⁻¹) and also produced low levels of glycolic, citric, succinic, gluconic and oxalic acids.

Phosphatase activity:-

Phosphatase activity of fungi (*Aspergillus* sp and *Penicillium* sp.) were higher than bacteria¹⁰. Phosphatase activity of fungi (*Aspergillus* sp and *Penicillium* sp.) were higher than bacteria (*Pseudomonas mallei*, *Bacillus subtilis*) as reported earlier¹¹. *Aspergillus* sp solubilised phosphate into organic acid and accumulated organic acids having retention times of 5.5 (approximating the retention time of citric acid), and 7.1 min. *Aspergillus* sp isolate also accumulated a small amount of organic acids having retention time of 7.8 min (approximating the retention time of malic acid), and 8.3 min. *Penicillium* sp accumulated malic acid. The results of the present study revealed that *Aspergillus* sp proved to be an efficient strain for phosphate solubilization and *Penicillium* sp needs small genetic manipulation. In addition, no significant amounts of organic acid production was observed from a phosphate solubilizer fungus, *Penicillium* sp¹². Tricalcium phosphate induced extracellular phosphatase production (119U/ml) compared to other phosphate sources^{14,15}, similar result was observed when tricalcium phosphate was added (10g/l) in the medium. Phosphatase activity by fungal isolates was maximum in the pH 4.0-6.5, which was similar to the results obtained during optimization studies¹³ for *Trichoderma* sp

The physico chemical characteristics of soil such as pH was neutral, the nitrogen content of the rhizosphere soil was found to be low, while the amount of Potassium in soil was quite high and phosphorus content was also low^{14,16,17}. The results of the present study on soil properties were in agreement to the results obtained by¹⁵ which reported that the fertility of soil was moderate.

Conclusion:-

Two phosphate solubilizing fungi were isolated and identified as *Aspergillus* sp and *Penicillium* sp. Phosphorus solubilization activity of PSM is associated with the release of organic acid and a drop in the pH of the medium. In the present study, there is a decrease in the pH values of the Pikosvikiyos media because of the release of organic acids in the cultural medium. Furthermore, the results of HPLC studies revealed that fungi solubilizes Phosphate into organic acid viz., Citric acid, Gluconic acid, Glycolic acid, Malic acid, Succinic acid and oxalic acid, which is evident from their retention time. Consequently it was proved that the isolated *Aspergillus* sp and *Penicillium* sp have the ability to solubilize phosphate which was confirmed by its phosphatase activity. Further molecular characterization of the isolates will provide their genetic relationship with other phosphate solubilizers.



Fig 1:- Isolation of fungi on RBCA plates



Fig 2:- Blackcoloured colonies

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RESEARCH ARTICLE

TO ENHANCE THE PERFORMANCE OF SOLAR STILL WITH REFLECTORS

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Abstract

The solar still is an errorless well spring of freshwater for farming and drinking in far away, widely different areas or islands. There are great numbers of sort of solar stills the least difficult and most put examples on view of is the basin solar still. Observations put examples on view of that the common still has limited amount produced. Researches have taken determinations to make different designs of solar stills to get well the amount produced and got knowledge by reasoning that solar stills incorporated with reflectors are the most efficient and effective Designs The reflectors, either outside or inside, are a good and cheap modification to increase the solar irradiation given direction to the basin liner or the water along with the distillate efficiency of the still. In this paper, a wide measures taking for different solar stills with reflectors has been showed.

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Introduction:-

People existing in isolated areas or islands, where freshwater source by transport is costly, face the problem of water scarcity every day. Solar still offerings specific advantages to be used in these ranges due to its easier production, minimum skills of operation and preservation requirements, and responsiveness to the environment. The clean free and ecofriendly to the environment are two major benefits which strengthen the use of solar stills. The main drawback of solar stills is the low production of freshwater in comparison with the other distillation systems. The production volume for a simple type solar still is only between 2–5 /m²/day. This makes the solar stills uneconomic compared to the other conventional distillation systems [1]. Many researchers have reviewed, comprehensively, the latest work on solar stills such as classification [2], design [3], improvement techniques [4], passive [5], active [6], inclined [7], stepped [8], wick type [9], and condensers with solar stills [10]. There is no specific manageable survey on solar stills with reflectors. Along these lines, this work is to make a documental study on the solar stills combined with external reflectors and internal reflectors.

Conventional Solar Still with Reflectors:-

Internal reflectors are suitable tools to concentrate and transmit solar radiation. They are suggested when sunlight is weak or the local temperature is comparatively low. External reflectors are ideal to be used to change the direction of solar rays to improve the flexibility of the absorber plate configuration such as vertical solar absorber plate which helps to recover the vapor latent heat of condensation. External and internal reflectors are suggested when sunlight is weak or the local temperature is discreetly low shown in Figure no 1.

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Categorization of reflector with solar stills:-

To get high distillation production, scientists introduced many efforts to make different design of solar stills. They concluded the solar still integrate with reflectors is effective and efficient. The reflectors with solar stills can be distributed into three types (1) internal reflectors (2) external reflectors both top and bottom, and combination of internal and external reflectors. To utilize the external and internal reflectors can be an economical method to expand the solar irradiation incident on the basin liner to make high productivity as possible. A simple valuation of different solar stills with reflectors is showed in Table no 1. The evaluation based on location, daily productivity, yield improvement and efficiency.

Internal reflectors (IR):-

The reflectors inside the solar still affect significantly the output of distilled water, which is credited to the centralization of the reflected solar radiation incident on the water. Besides, reflectors decrease the waste heat energy from solar still. Tamimi [11] studied experimentally the performance of a single slope solar still

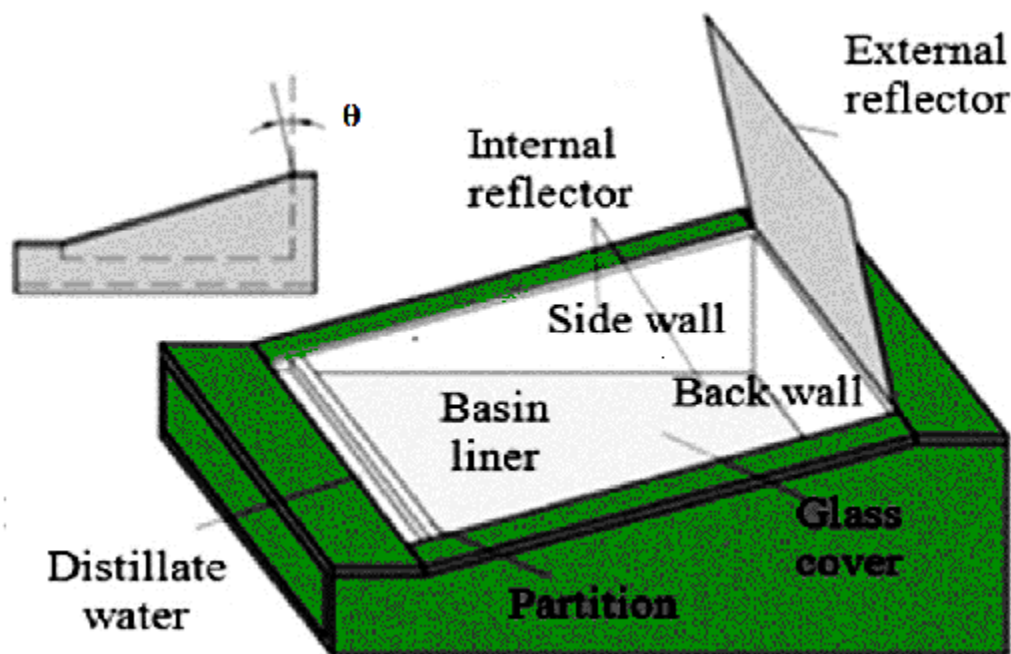


Figure 1:- A Solar Still basin with reflector

Single basin solar still with mirrors installed on the side walls of a still. They concluded that by using mirrors the basin efficiency increases during the whole day. El Swify and Metias [12] used scientific modeling and conducted experiments to find out the effect of internal reflectors on the back and side walls of a single slope solar still with its back wall acting as an additional condenser showed in Figure No.2. They mentioned that the distillation increase of 80% & 20% can be obtained from installing reflectors in the summer and winter season, respectively. New designs of solar still consist of a metallic cylindrical parabolic reflector studied by Minasian et al. [13]. The reflector was design to concentrate incident solar radiation on the black outside surface of a tray located on the focal line of the reflector. The tray was lined with blackened wick, which represents the evaporative surface of proposed still which was shown in Figure no. 3. They showed that the productivity of the latest still was 23–33% greater than that of a conventional basin type solar still. Abdallah et al. [14] provides the modified basin solar still. The design modification was fix the interior reflecting mirrors on the internal walls of the still to minimize the amount of energy lost shows in Figure no 4. The experimental result was shows that the use of internal mirrors improved the system thermal performance up to 32%. The solar still design was modified from flat basin to stepwise basin and the efficiency increases by a normal value of 185%. AlHayek and Badran [15] compared the performances of a double slope basin solar still and a single slope basin solar still. The inside surfaces of complete walls were made of mirrors. They performed their experiments during July and determined that using mirrors on the inside walls of the single slope basin solar still improved the production of distilled water by 18% higher than the double basin solar stills. The effect of an internal reflector on the production of a single slope solar still (during the winter and summer)

was examined experimentally and theoretically by Karimi et al. [16]. They presented a scientific model considering the effect of all walls (east, west, north and south) of the solar still on the amount of received solar radiation to brine. The model was validated with the experimental data. The model can calculate the yield of the still with and without Internal Reflector on various walls. The result shows the simultaneous use of Internal Reflector on front and side walls enhances the solar still efficiency by 16%. Though, installation of an Internal Resistance on the back wall can increase the annual efficiency by 20%. The installation of Internal Resistance on all walls in comparison to a still without Internal Reflector can increase the distillate production at winter, summer, and the entire year by 60%, 20%, and 31%, respectively. Modification of the stepped solar still over installing Internal Reflector on the vertical side which was introduced by Omara et al. [17]. The productivity of the stepped solar still with Internal Reflector was higher than that without the Internal Reflector. The results also indicated that the productivity of the modified stepped solar still with and without Internal Reflector is higher than that of the conventional solar still by almost 70% and 50%, respectively. Also, the efficiency of the stepped solar still with reflectors and without reflectors was 54% and 51% respectively, whereas, the efficiency of the conventional solar still was only 32%. The performance factors of the Corrugated Solar Still (CSS) and Conventional Solar Still (CnSS) were studied experimentally by Omara et al. [18]. The author's view concerns with using both double layer wick material and reflectors composed inside the CSS, Figure no 5. In addition, the influence of saline water depth (1.5, 2.5, and 3.5 cm) on CSS performance was also studied. During investigations, the product of CSS with wick and reflectors is about 146% higher than the CSS at a brine depth of 1.5 cm. In addition, the daily efficiencies of CSS and CnSS were approximately 60% and 35%, respectively. In other experimental study by Omara et al. [19], a hybrid solar distillation system covering of corrugated and wick absorbers of solar stills was combined with an external condenser and internal reflectors to study their performance. They showed that the productivity of corrugated wick still with reflectors and external condenser was improved by about 182% over CnSS at a brine depth of 1.5 cm.

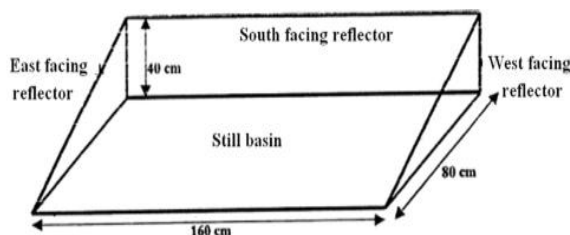


Figure 2:- Double Exposure solar still With Internal Reflectors.

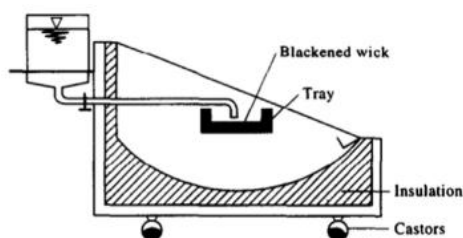


Figure 3:- Basin solar still with cylindrical parabolic reflector.

External reflector (ER):-

The external reflector used in the solar still is made up of greatly reflective materials such as mirror finished metal plate. The diffuse and direct beams transmitted over the glass cover are improved by using the ER. Scientist (Hiroshi Tanaka) is the most concerning to study the effect of reflector on the distillate of stills.

External top reflectors:-

Tanaka et al. [20–22] showed numerical investigation on a tilted wick solar still with a top mirror (vertical [20], forwards [21] and backwards [22]) extending from the upper edge of the still. They displayed geometrical models to compute the solar irradiation reflected from a top mirror and then absorbed on the evaporating wick. They concluded that the top mirror can increase the absorbed solar radiation by evaporating wick, and added, the amount of solar irradiation reflected from the top mirror and absorbed on the wick can be enhanced by inclining the top mirror to be backwards in summer and forwards in winter. A theoretical analysis was used by Tanaka and Nakatake [20] to study the effect of a vertical flat plate External Reflector on the productivity of a TWSS and shows the average increase of

10% in a year. Another theoretical analysis of a top External Reflector with TWSS on a winter was investigated by Tanaka and Nakatake [21]. Their results indicated that the productivity of a still with an inclined reflector would be around 12% or 25% over that with a vertical reflector when the reflector length is a half of or the same as the solar still length. In adding, a theoretical analysis was made on TWSS with external top reflector with the aim of determining optimum inclination for both reflector and solar still for different seasons by Hiroshi Tanaka [22], Figure No 5. Regarding to the results obtained, 30°N latitude was represented as the best inclination angle either for the still or the reflector monthly. In addition, he concluded that for any season the daily productivity of the solar still can be improved by adjusting the inclination of both the reflector and solar still, thus producing around 20% over the traditional TWSS throughout the year. Tanaka and Nakatake [23] displayed another theoretical investigation of one step azimuth tracking TWSS with a vertical flat plate reflector. The TWSS is assumed to be rotated manually just once a day at southing of the sun. They performed numerical analyses of heat and mass transfer in the still to determine the daily productivity of the still on four typical days: the spring and autumn equinoxes and summer and winter solstice days at 29°N latitude. For four days, results indicated that the increase in the productivity of TWSS would be around 40%, and can be accomplished by the simple modification of utilizing a reflector. El-Bahi and In an [24] examined a conventional still integrated with an outside condenser in Figure No 6.

Table 1:- Simple comparison of different solar still with reflectors

No	Categorization	Authors	Locations (Latitude)	Reflector material	- Daily yield (kg/m ² /d) - Improvement - Efficiency	Reflectors effect (%)	Observations in experiment
1. Internal reflectors							
1	Conventional still	El-Swify et al. [12]	Cairo, Egypt (25° N)	Mirrors	3.05–7.2 82.6% in winter 22% in summer –	82.6% 22%	The cooling effect of the back glass cover, back condenser, is improved due to the higher temperature difference as compared to the ordinary one.
2		Minasian et al. [13].	Baghdad, Iraq, (33.33°N)	Stainless steel	1.23–6 25–35% –	25–35%	The productivity of the conventional basin type solar still has been increased by using a stainless steel cylindrical parabolic reflector.
3		Abdallah et al. [14]	Amman, Jordan, (32°N)	Mirrors	1.64 30% ~	30%	Installing internal side mirrors gave better performance because these mirrors make use of the energy reflected on all sides of the solar still.
4		AlHayek and Badran [15]	Amman, Jordan (32°N)	Mirrors	– 20% 11%	20%	Performance characteristics of the stills showed that the temperature at the water surface is increased with decreasing water depth, and by the addition of dye.
5		Karimi et al. [16]	Iran. (30°N)	Mirrors	– 34% –	34%	The installation of IRs on all walls in comparison to a still without IR can increase the distillate production at winter, summer and the entire year by 65%, 22% and 34%, respectively.
6	Stepped solar still	Omara et al. [17]	Kafrelsheikh, Egypt, (31.07°N)	Mirrors	6.35 75% 56%	18%	Productivity of stepped still with and without internal reflectors was higher than the conventional still by 75% and 57% respectively.
7	Corrugated wick solar still	Omara et al. [18] and [19]	Kafrelsheikh, Egypt, (31.07°N)	Mirrors	4.1 145.5% 58%	55%	The yield of Corrugated wick solar still with reflectors when providing a vacuum was enhanced to about 180% higher than the conventional still.

2. External reflectors**External top reflectors**

8	Tilted wick solar still	Tanaka and Nakatake [20]	Kurume, Japan (33°N)	Mirrors	– 9% –	9%	The average daily amount of distillate for four days, (spring and autumn equinox and summer and winter solstice days), peaks when the angle of the still is 20° for the still with the reflector, and peaks at 30° for the still without the reflector.	
9		Tanaka and Nakatake [21]	Kurume, Japan (30°N)	Mirrors	4.2 15% –	15%		The inclined reflector can increase the distillate productivity of the still at any still's inclination, and the reflector's inclination should be set at about 15° from vertical.
10		Tanaka [22]	Kurume, Japan (30°N)	Mirrors	– 21% During the year –	21%		The distillate can be increased by inclining the reflector backwards in winter and forwards in summer, and the inclination angle of the reflector would be less than 25° throughout the year.
11	Conventional still coupled with external stainless steel reflector and outside condenser	E1-Bahi and Inan [24]	Ankara, Turkey, (39.6°N)	Stainless steel	–	–		The efficiency of the solar still was improved up to more than 70%, and the distilled fresh water was up to 71/m ² .d when a solar still coupled with an external stainless steel reflector and outside condenser
12	Conventional still	Shanmugan et al. [26]	Tamil Nadu, India (11.30°N)	Mirrors	4.2 – 45	–	The efficiency of the still was as high as 35%, and increased to 45% with the mirror booster. The twin reflector booster has to be reoriented only once in a day at mid-noon. The main advantage of the “V” type solar still is due to center collection and all the condensation are easily directed to the outlet.	
13	Conventional still With twin reflector	Srivastava et al. [27]	Rewa India (24.32°N)	Mirrors	79	79%		
14	“V” type solar still	Selva Kumar et al. [28]	India (22°N)	Mirrors	2.7 7.3 12	7.3%		
External bottom reflectors								
15	Double slope single basin solar still	Sebaii [29]	Tanta, Egypt (31° N)	Mirrors	– 19% and 30% summer and winter –	19% and 30% summer and winter	The daily productivity of the double slope single basin solar still with mirrors is higher than that without mirrors, due to the increased basin	

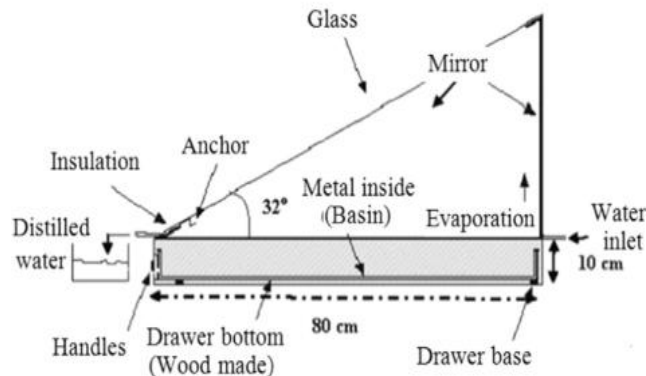


Figure 4: basin solar still with Internal Reflector

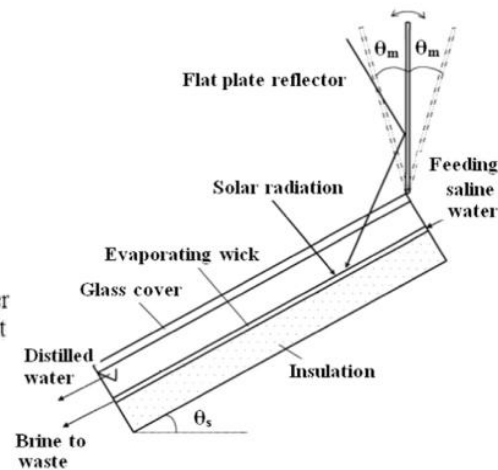


Figure 5: Top reflector with TWSS

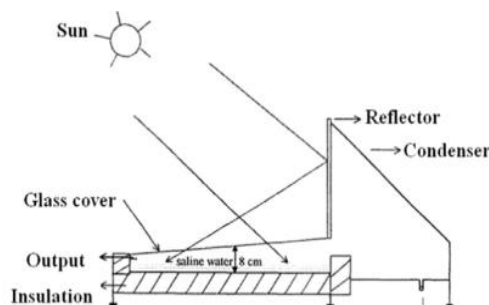


Figure 6:- Conventional Still integrated with top reflector.

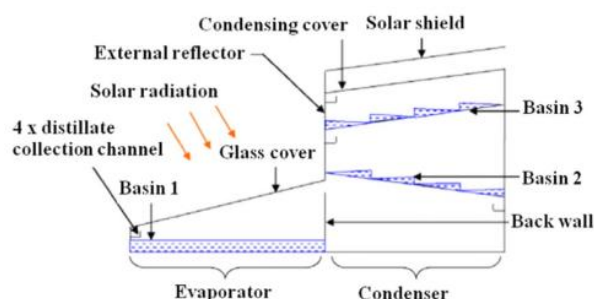


Figure 7:- Basin solar still with reflector and external condenser.

A reflector made of stainless steel was added to the glass cover to reflect the solar radiation into the still basin through the glass cover and to give a shadow for the condenser. The study led to the conclusion that the efficiency of the solar still was enhanced by about 72% and the daily productivity was up to 7 l/m^2 a day. Madhlopa and Johnstone [25] proposed a model to calculate the distribution of solar irradiation inside a conventional still with reflector and external condenser, Figure no 7. The system had one basin in the evaporation chamber and two other basins in the condenser chamber. It had a glass cover over the evaporator basin and an opaque condensing cover over basin 3. Water vapor from the first basin condenses under the glass cover while the remainder flows into the condenser by purging, diffusion, and condenses under the liner of basin 2. They found that distilled freshwater was about 2.1 kg/m^2 a day. The performance of an acrylic mirror boosted solar distillation unit was studied by Shanmugan et al. [26]. The performance of solar still in terms of distilled water collection has been analyzed and a booster mirror (acrylic) was attached just above the glass cover of solar still. The mirror boosting reflects the excess solar radiation to water and it is possible to adjust the booster mirror for perfect reflection depending upon the sun moving angle. The maximum amount of water collected per day was about 4.1 L. The efficiency of the still was as high as 33%, and increased to 43% with the mirror booster. Srivastava et al. [27] performed experiments of a modified solar still with multiple low thermal inertia porous absorbers, Figure no 8. The absorbers were made up of jute cloth and they float on the basin water with the help of thermocol insulation. The surfaces of the absorbers were always wet and hence there were no dry spots. The results showed that the productivity was 66% more than the conventional still and 33% more on cloudy days. The basin water below the floating absorber remained warm during offshine hours, and hence, the distillate was produced even at nights. A twin reflector booster was placed perpendicular to each other on the modified stills. The productivity increased by 77% over the still without booster. The thermal performance of a “V” type solar still with a charcoal absorber was analyzed by Selva Kumar et al. [28]. The main advantage of this type of still is that the distilled water collection was directed towards the central water collection channel as shown in Figure no 9. The production of the outlet distillate water had increased. The overall efficiency was found to be 25% and 12% for the still without and with the boosting mirror, respectively. He also conducted similar experiments which were carried out for the charcoal absorber in the still without and with the boosting mirror which yielded 30% and 14% respectively.

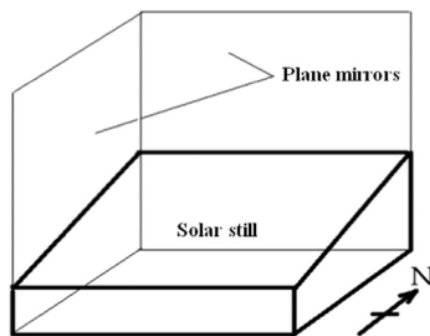


Figure 8:- Twin reflector booster applied to solar still.

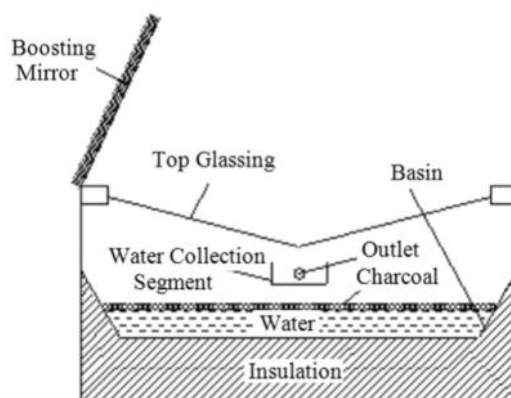


Figure 9:- V type solar still with External reflector.

Scope for further research:-

There are still some problems and challenges on the design of solar stills not being clearly analyzed for performance improvement, including solar stills with reflectors. This paper reviews the studies and developments of solar stills with reflectors. The results from the preceding work done clearly show that the enhancement of solar stills with reflectors performance differs greatly for different techniques due to different experimental conditions. The review represents specific inferences drawn from the analyses of solar stills with reflectors by various authors to pave way researchers to grasp the previous designs and to fabricate new designs with optimum design parameters for higher distillate output.

The following factors may be taken into consideration for further research of solar stills with reflectors:

For active solar still:

- 1.1 Combine solar still with solar water heater in order to increase the basin temperature.
- 1.2 For active solar stills, more research may be carried out with reflectors.
- 1.3 Further studies should be conducted to improve the solar stills productivity, especially in the fields of coupling with heat storage and various waste heat sources.

For tracking system:

- 1.4 Solar still with azimuth sun tracking and just an adjustment of the optimum elevation of the day.
- 1.5 Combine the solar still with sun tracking and the solar water heater.
- 1.6 The sun tracking system is more effective than fixed still and it is capable of improving the distillate output of the still with reflectors.

Conclusions:-

Reflectors are used to enhance the daily amount of distillate of the solar still. The accompanying conclusions can be inferred on the basis of above discussions

- Installing reflectors is more practical in places where solar radiation is weak and the ambient temperature is relatively low.

- Installation angle of the external reflector should be changed with the seasons to enhance the productivity through all the year.
- The external reflector is inclined backwards or forwards according to the month.
- The daily productivity can be increased by adjusting the inclination of both the still and reflector mirror in any season.
- The benefits of both the inclined external top and internal reflectors would be considerably less in summer than in winter.
- For the still with larger angle of the glass cover, the effect of the external top reflector would be smaller.
- The vertical flat plate external reflector would be less effective for the tilted wick solar still than for the conventional still.

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RESEARCH ARTICLE

ADHESION OF *STREPTOCOCCUS MUTANS* BIOFILM ON THE SURFACE OF INDIRECT RESIN COMPOSITES RESTORATIONS.

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Key words:-

Indirect Resin Composites,
Streptococcus mutans, bacterial
adhesion.

Abstract

Objective: To evaluate and compare *streptococci mutans* adhesion on the surface of indirect resin composites compared to direct.

Materials and Methods: A total of forty standardized disk shaped samples of the four tested material were constructed in special mold and cured according manufacturer's instructions and finished and polished with sofex finishing system, then subjected to bacterial biofilm colonization and the adhered bacteria were calculated and, the collected data were subjected to statistical analysis.

Results: One way ANOVA showed that there was a significant difference between the tested resin composites, and the post hoc Tukey's test showed that the lowest bacterial adhesion recorded for SR Nexco and Ceram X one with statistically significant difference compared to Z 250 and Herculite classic (p 0.05)

Conclusion: Indirect resin composites have lower susceptibility to bacterial adhesion than do direct one in the same context the smaller the filler particle size the lowest the bacterial adhesion.

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Introduction:-

Tooth colored restorative materials are today preferred due to the patient's demand for esthetic restorations and minimal invasive treatment modes. Although resin composite can be applied directly to restore posterior teeth, it can be also indicated for indirect restorations. In particular for medium to large sized cavities on the condition that sufficient tooth structure remains for adhesive cementation.¹ This restoration strategy is highly required due to the need of marginal adaptation, proximal contacts, anatomic form, color match, polymerization shrinkage and wear resistance controlling.^{1,2}

Indirect composites now offer an aesthetic alternative to ceramics for posterior teeth as inlays and onlays. The mechanical properties of lab composites in comparison to ceramics are inferior, however the aim is not that indirect composites replace ceramics rather that they supplement and complement them dependent upon each individual patient situation.

A brand of indirect resin composites, SR Nexco (Ivoclar Vivadent, Schaan, Liechtenstein) was introduced in 2012. SR Nexco Paste is a purely light-curing laboratory resin composite with high content of inorganic microopal fillers.

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This affords optimum benefits in terms of abrasion, discoloration, processing and surface gloss. The balanced ratio between the matrix and filler components results in outstanding physical properties achieved with polymerization units.³ The combination of microfillers plus prepolymer enables a very high filling ratio and excellent physical properties. The use of the prepolymer allows the advantages of large filler particles to combine with those of microfillers. This technology allows for a superior strength of resin composite that if only inorganic microfiller were used.³

The initial adhesion and early colonizing of bacteria to the surface is an important step in the process of plaque formation on solid substrate surfaces such as teeth and restorative materials.^{4, 5} Resin composites have surface characteristics different from those of teeth. Unlike tooth surfaces, the surface properties of resin composite materials related to bacterial adhesion and biofilm formation are affected by a myriad of factors — such as mechanical surface properties, material components such as filler particles and resin matrix, and curing conditions. In particular, matrix monomers might influence the growth of some cariogenic bacterial species^{6,7}

Many studies have reported that composite resins tend to accumulate more bacteria or plaque than do other restorative materials *in vitro* and *in vivo*. Differences in the amount of bacterial adhesion can be explained by the diverse surface characteristics of each type of composite resin.⁸⁻¹⁰

The success of the esthetic restorations on a long-term basis depends on the quality and amount of adhered biofilm are. The initial adherence and subsequent colonization of bacteria on the surface of composite resins is the key of the pathogenesis of the secondary caries promoted particularly by *Streptococcus mutans* (*S. mutans*) and *Streptococcus sobrinus* (*S. sobrinus*). *S. mutans* has been identified as the major etiological agent of human dental caries and composes a significant proportion of the oral streptococci in carious lesions.¹¹ *S. mutans* is harbored in mature plaque. Organic acids are trapped within the glucan barrier produced by these bacteria, resulting in a prolonged low pH around the enamel surface.¹²

Based on the facts described, the aim of this study was to evaluate *S. mutans* biofilm adherence on the surface of, nanohybrid, microfill direct resin composites, and nanofilled, microfill indirect resin composites.

Materials and methods:-

Materials:-

In this study these types of dental resin composite; SR NEXCO (microfill indirect composite Ivoclar Vivadent, Schaan, liechtenstein), Filtek Z 250 (Z 250) (microhybrid direct/indirect composite 3M Dental product, St Paul, MN, USA), Ceram X One Dentsply Deterey GmbH, De-Trey-Str. Germany and Herculite classic Kerr Italia S.r.l. via passanti, Scafati (SA) (microfill direct composite).

Methods:-

Specimens' preparation:-

A total of forty standard disc shape specimens were constructed from the four brands of the resin composite materials indirect microfill resin composite SR NEXCO, indirect microhybrid resin composite Z250 and direct microfill resin composite Herculite classic and nanohybrid resin composite Ceram X One (n=10).

A split Teflon mold was composed of an outer copper ring and split Teflon ring with an internal hole of 10mm diameter and 2mm thickness was used to construct the stranded disc shapes specimens, the mold can be split in two halves and reconstructed by the copper ring.

The selected shade was A3 shade to standardize the degree of polymerization. The filled split Teflon mold was assembled over a glass slide with a thickness of 2mm, covered with a 0.05mm transparent polyethylene film to standardize surface smoothness. The mold space was filled with each test material in a bulk pack technique, avoiding gross excess and entrapping of air, then covered with clear celluloid strip and another glass slide and pressed to remove excess material. The applied materials of the three tested groups was light cured for twenty seconds from each side using light emitting diode (LED) light-curing unit with an intensity of (900 MW/cm²). Then each specimens was light curried from each side for additional twenty seconds after removed of the glass slap and the polyethylene film. After that the indirect resin composite SR NEXCO there was subjected polymerization by special oven Lumamat-100* for ten minutes (Targis Power TP3 Upgrade, Ivoclar Vivadent AG Schaan,

Liechtenstein) using program P2 for 11 minutes in order to complete polymerization according to manufacturer's instructions

The surfaces were smoothed by rubber polishers and silicone polishing wheels, leather buff wheels and Universal Polishing Paste (Ivoclar Vivadent AG Schaan, Liechtenstein).³

Finally, all the specimens in the four groups were finished with flexible discs (Sof-Lex XT Pop On, 3M ESPE) following the recommended sequence of finishing and polishing discs (coarse, medium, fine and superfine) so as to have standardized a smooth surface.

Cultivation of the Microorganism and Preparation of Cell Suspensions:-

A standard suspension of *S. mutans* (ATCC 25175) containing 10^6 cells/mL was prepared. For this purpose, *S. mutans* was seeded onto blood (Oxoid, UK) and incubated for 24 hours. All incubation was carried out at 37°C in a CO₂ chamber. After incubation, the growth was suspended in sterile physiological solution (0.9% sodium chloride (NaCl)), and the number of cells in suspension was counted in a spectrophotometer (B582, Micronal, São Paulo, Brazil). The parameters of optical density and wavelength used were, respectively, 0.620 and 398 nm. These parameters were previously established by means of a standard curve with CFU vs. absorbance.

Biofilm Adhesion:-

Adherence testing was performed in an aseptic environment in a laminar airflow chamber. The broth used for adherence was trypticase soya broth, dissolved in 1000 mL of distilled water. The broth was sterilized by autoclaving at 121°C for 15 minutes. In each well of Sterile 24-well polystyrene tissue culture plates was placed one specimen, 1.5 mL of broth, and 0.1 mL of standardized *S. mutans* suspension. The plates were sealed and incubated at 37°C for 24 hours in a CO₂ chamber. Samples were then removed and washed twice with sterile physiological solution (0.9% NaCl) in order to remove loosely bound material. Following this, the samples were placed in tubes with 3 mL of sterile physiological solution (0.9% NaCl) and sonicated (Sonoplus HD 2200, 50 W, Bandelin Electronic, Berlin, Germany) for 30 seconds to disperse the biofilms. The suspension obtained was diluted 10, 100, and 1000 times, and aliquots of 0.1 mL were seeded in duplicate onto BHI agar and incubated for 48 hours at 37°C in a CO₂ chamber. After the incubation, the plates with 30 to 300 typical colonies of *S. mutans* were counted in a colony counter (Phoenix CP-600, São Paulo, Brazil), and mean values of CFUs were obtained (CFU/mL). Mean values of CFU/mL were converted into logarithmic (log₁₀) values and analyzed by ANOVA and the Tukey test. p value, 0.05 was considered to indicate a statistically significant difference.

Scanning Electron Microscopy (SEM) Evaluation:-

All the specimens were subjected for SEM evaluation. One specimen of each subgroup was prepared for the SEM (JSM-5310LV JEOL, Tokyo, Japan) evaluation. The specimens were coated with silver in a vacuum evaporator. The "retained biofilms" on the specimens after sonication were observed for each group. The samples were rinsed with PBS buffer and fixed in 4% paraformaldehyde with 1% glutaraldehyde in PBS for one hour. The samples were rinsed with PBS three times for two minutes each, and finally rinsed with deionized water three times for two minutes each. They were then dehydrated through an ethanol series (50, 70, 80, 95, and 100%) for 15 minutes each, desiccated, sputter-coated, and observed by a SEM. Photographs of representative areas of the polished surface were taken at 3000× magnifications. The results were analyzed by calculating the mean and standard deviations for each group. The data of each material were subjected to analysis of variance (ANOVA) followed by Tukey's high significant difference (HSD) test at a p-value of 0.05

Results:-

Mean and standard deviation values of the CFU/mL (log₁₀) of *S. mutans* within the biofilms formed are displayed in Table 1. The one way ANOVA test revealed that there was significant difference in terms of the level of adhesion between the tested the four groups (P=0.01); the highest value of bacterial adhesion were recorded for Herculite classic followed by Z 250, Ceram X One and, finally SR NEXCO. There was no statistically significant difference between SR Nexco and Ceram X one and, there was no statistically significant difference between Z 250 and Herculite classic. However, there was statistically significant difference between SR Nexco and Ceram X one in one side and, Z 250 and Herculite classic in the other side (p 0.05). Table 1

Type of Material	Mean Adhesion Level (colonies/mm ²)	P value
SR NEXCO	4.6±0.09 ^B	0.001
Ceram X One	4.7±0.08 ^B	
Z 250	4.97±0.06 ^A	
Herculite classic	4.99±0.06 ^A	

Table 1:- Mean bacterial adhesion in different groups

Scanning Electron Microscopy (SEM) Evaluation:-

SEM showed that *S. mutans* was observed to be accumulated on the surface of Herculite classic and Z 250 much more than SR NEXCO and Ceram X One. Figure 1&2 and 3.

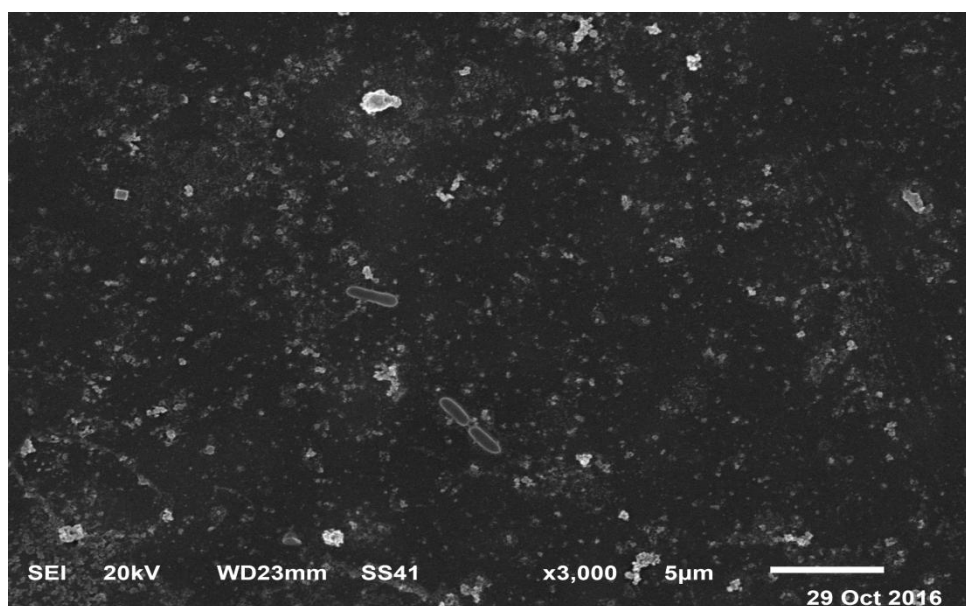


Figure 1:- showing SEM picture of *S mutans* on the surface of SR Nexco.

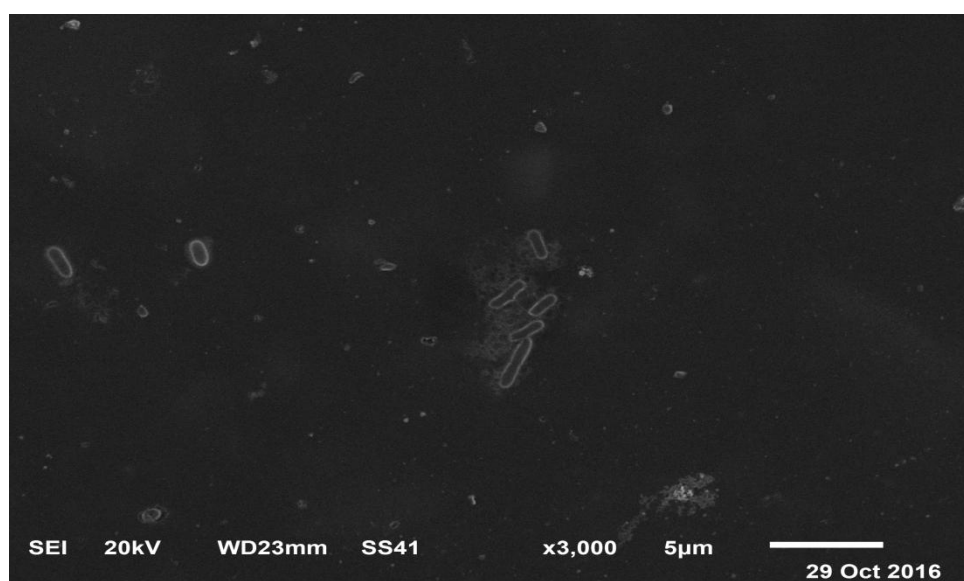


Figure 2:- showing SEM picture of *S mutans* on the surface of Ceram X One.

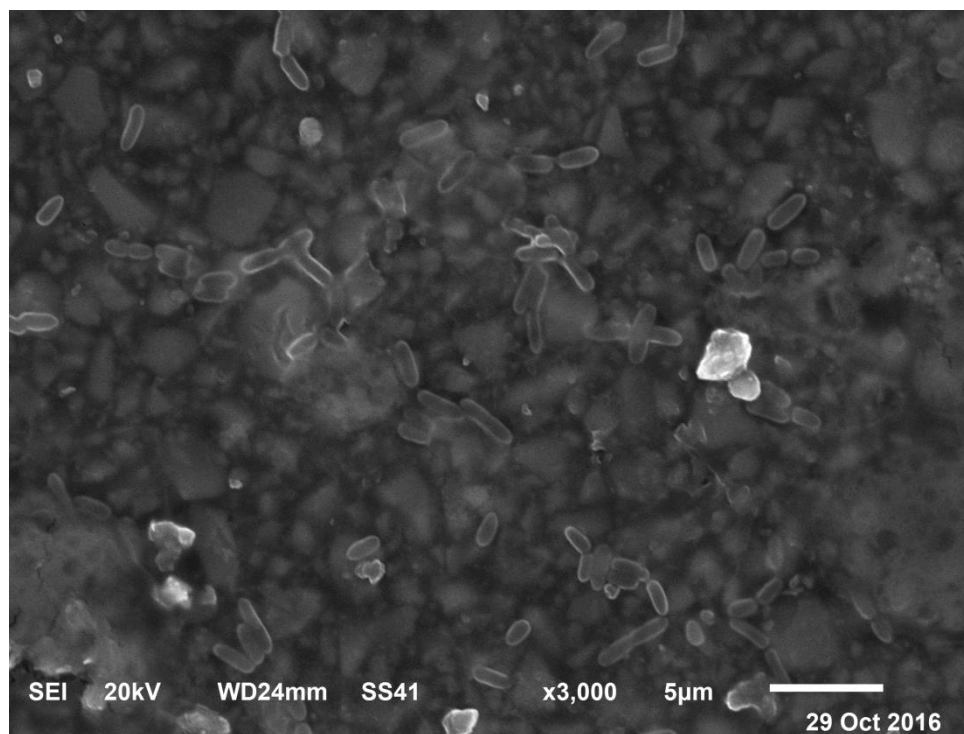


Figure 3:- showing SEM picture of *S. mutans* on the surface of Herculite classic.

Discussion:-

The quality and amount of adhered biofilm are important to the success of the esthetic restorations on a long-term basis. The initial adherence and subsequent colonization of bacteria on the surface of composite resins is the key of the pathogenesis of the secondary caries promoted particularly by *S. mutans* and *S. sobrinus*. *S. mutans* has been identified as the major etiological agent of human dental caries and composes a significant proportion of the oral streptococci in carious lesions.¹¹ *S. mutans* is harbored in mature plaque. Organic acids are trapped within the glucan barrier produced by these bacteria, resulting in a prolonged low pH around the enamel surface.¹²

Restorative materials may initiate plaque and secondary caries formation in the oral cavity. Therefore, it is important to observe for initial bacterial adhesion on restorative material surface, which is the first step in plaque formation. Initial bacterial adherence to solid surfaces is facilitated by several factors, namely electrostatic¹³⁻¹⁵ and hydrodynamic interactions¹⁶, thermodynamic binding parameters¹⁷, specific binding mechanisms including adhesion receptor interactions by which bacteria bind selectively to the surface, and cementation by polysaccharide matrices or glucans.^{5,17-19}

Although polyester matrix strips promote the smoothest surface most clinical situations require bulk removal of excess composite. Additionally, finishing and polishing techniques are necessary to remove the monomer rich surface layer of the composite material.²⁰ This eventually eliminates the organic matrix, exposing and dislodging the filler particles, thus increasing the surface roughness of polymer-based materials.¹⁶ Among finishing and polishing techniques, it has been reported that the Sof-LexTM aluminum oxide, discs technique provides a smoother surface on composites compared to carbide bur finishing followed by the Astrobrush TM technique.²¹

For this investigation, the samples were not coated with saliva because previous studies^{22,23} have found that saliva coating did not significantly alter the adhesion patterns of *S. mutans* and *S. sobrinus*. This phenomenon is consistent with other investigations showing that saliva coating does not significantly alter the adhesion of Streptococci to the underlying materials.^{24,25} However, other authors suggest emulating the conditions of the oral cavity, such as temperature, constant movement and saliva coating, which could represent an environment closer to reality.^{26,27}

In the present study it was found that SR Nexco and Ceram X one have the lowest bacterial adhesion with no significant difference between them and significant difference with the other two resin composite which may be in SR Nexco due to the enhanced polymerization of all resin composite by the furnace enhanced polymerization with

no or fewer double bond with subsequent enhancement of the surface characteristics minimizing bacterial adhesion which is in agreement with Derchi G who found that Indirect dental restorative composite resins less prone to biofilm adhesion than direct composite resins.²⁸, while in Ceram X one the lower bacterial adhesion may be due to the nano-ceramic filler used in its construction giving it a higher filler loading and smoother surface which yields a smoother surface with subsequent lower adhesion of bacteria, and this in agreement with the study conducted by

Ionescu A who concluded that the proportions of resin matrix and filler particles on the surface of resin-based composites strongly influence *S. mutans* biofilm formation in vitro, suggesting that minimization of resin matrix exposure might be useful to reduce biofilm formation on the surface of resin-based composites.²⁹

Both Z250 and Herculite classic have nearly the same tendency for bacterial adhesion also Z250 used as indirect resin composite it was cured by the direct LED without any furnace which may yield degree of conversion similar to that of Herculite, also they have similar filler size so under the same finishing and polishing procedure they gave nearly equal surface characters which explains the similarity in bacterial adhesion and this in accordance with Celik C et al who studied the effect of finishing and polishing procedures on surface roughness of tooth-colored materials and found that the smoothest surface was generated with Sof-Lex Pop-on disks for all the materials tested.³⁰

The surface micromorphology of resin composites after finishing and polishing has been shown to be influenced by the size, hardness, and amount of filler particles. Harder filler particles are left protruding from the surface during polishing, as the softer resin matrix is preferentially removed in hybrid composites. Filler particles should be situated as close together as possible in order to protect the resin matrix from abrasives.³¹ A nanocluster filler particle consists of loosely bound agglomerates of nano-sized filler particles. Eventually, the surface has smaller defects and better polish retention.³² Filtek Z250 showed the least surface smoothness compared with Ceram X and **SR NEXCO** composite restorative materials, probably because it contains large glass filler particles which can be plucked away, leaving voids or rougher surface after being polished.^{31,33} For these responses the *S. mutans* less attached to **SR NEXCO** and Ceram X than Filtek Z250 and **Herculite classic**.

Under the limitation of this study it was concluded that SR Nexco is a good choice for clinical situation that requires indirect esthetic restoration in regard to bacterial adhesion. Ceram X one is a good choice for clinical situation that requires direct esthetic restoration in regard to bacterial adhesion.

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RESEARCH ARTICLE

DOES INSPIRATORY MUSCLE TRAINER HAVE AN EFFECT ON DIAPHRAGMATIC EXCURSION IN HEMODIALYTIC PATIENTS?

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Abstract

Background: Hemodialysis patients suffer from severe muscular weakness and one of the most important muscle is the diaphragm – main muscle of respiration – that was experienced by dyspnea and decreased functional capacity due to a unique form of protein and energy malnutrition. **Purpose:** to determine the response of diaphragmatic excursion to inspiratory muscle trainer prior hemodialysis session. **Methods:** Thirty patients of both sexes (10 men and 20 women) were included in this study with mean age (\pm SD) was 35.47 (\pm 6.51). They were chosen from El Sahel Teaching Hospital, Nephrology department, dialysis unit, and undergoing dialysis from at least 2-3 years. Ultrasonography (U.S) was used to assess diaphragmatic excursion (D.E) during deep breathing maneuver before and at the end of the training program. All patients received the inspiratory muscle training for 12 weeks, 3 times/week. **Results:** The results of this study showed statistical significant difference between pre and post-training in diaphragmatic excursion ($p < 0.001$) with a percentage of improvement 65.74%. The percentage of improvement in men was 65.29% corresponds that in women was 66.08% compared to the pre-data status. **Conclusion:** Inspiratory muscle training by inspiratory muscle trainer device for 12 weeks in hemodialysis patients had significantly improved the diaphragmatic excursion (DE) that ensures the improvement in their mechanics of breathing and sense of dyspnea.

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Introduction:-

Chronic kidney disease is an evolving problem in modern countries. In 2010, 520,000 patients had routine dialysis. Each year, new patients (4.1%) are added to dialysis patients. Although hemodialysis has increased life expectancy of these patients, they suffer from many complications including restricted physical activity, muscle weakness, anemia, osteoporosis and metabolic disturbances. Muscle weakness is manifested as generalized weakness and particularly respiratory muscle weakness manifested by reduced vital capacity.¹

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Several decades of research efforts devoted to studying the patterns of chronic kidney disease-end stage renal disease (CKD-ESRD) progression as well as the impact of acute kidney injury (AKI) on this continuum of CKD-ESRD evolution but the full understanding of the process (es) of CKD-ESRD progression remains elusive². Significant muscle weakness and associated atrophy is seen in both dialysis patients and in patients with chronic kidney disease (CKD) stages 3–4. This is associated with increased morbidity and mortality^{3,4}.

Maintenance dialysis patients encounter multiple catabolic processes and experience a unique form of protein and energy malnutrition, which is characterized by muscle wasting and decreased visceral protein stores. The pathophysiology of muscle wasting in chronic kidney disease clearly is complex, multifactorial, and not fully elucidated. What is clear is that abnormalities in muscle function, exercise performance, and physical activity begin in earlier stages of chronic kidney disease and progressively worsen as ESRD ensues⁵.

The diaphragm is the major respiratory muscle, contributing up to 70% to resting lung ventilation. During dialysis the diaphragm as a skeletal muscle is affected by protein breakdown leading to its weakness and limitation of its movement⁶. These muscles show decreased in muscle strength and endurance properties resulting from uremic myopathy. Some authors who have studied the involvement of uremia in the diaphragm have concluded that loss of strength occurs through severe uremia. The ventilatory deficit due to this impairment in respiratory muscles, combined with other lung tissue impairments, compromises the functioning of this system, thereby contributing towards decreased lung capacity⁷.

The relationships between the lungs and the kidneys are clinically important ones in both health and disease. Chronic renal failure may affect respiratory function. Pulmonary dysfunction may be the direct consequence of circulating uremic toxins or may result indirectly from volume overload, anemia, immune suppression, extra osseous calcification, malnutrition, electrolyte disorders, and/or acid–base imbalances⁸.

As the respiratory muscles are morphologically and functionally skeletal muscles and therefore respond to training, just as any muscle of the locomotor system⁹. Inspiratory muscle training (IMT) is a technique used to increase strength or endurance of the diaphragm and accessory muscles of inspiration which improve inspiratory muscle functions, lung volumes, work capacity and power output¹⁰.

Methods:-

Participants:-

Thirty patients of both sexes (10 men & 20 women) undergoing hemodialysis from at least 2-3 years selected from El Sahel teaching hospital nephrology department, Dialysis unit with mean age (\pm SD) was 35.47 (\pm 6.51). They performed the dialysis 3 times / week. The purpose, nature and potential risk of the study were explained to all patients. All patients signed a consent form prior to participation in the study and confidentiality was assured. The inclusion criteria in the studied group were patients with creatinine level ranged from 4.8-7.8mg/dl and didn't participate in any physical activity prior the study by at least 6 months. Patients had the following criteria were excluded from the study: recent lung surgery (within 12 months), un-drained pneumothorax or large bullae on chest radiograph, marked osteoporosis (t-score $>$ -1.5) with a spontaneous rib fracture and developing cancer or smoking patients.

Protocol:-

All enrolled patients were evaluated by M-mode Ultrasonography to assess maximum diaphragmatic excursion pre and post-training by the same (U/S) machine and examined by the same U/S skilled radiologist. Diaphragmatic excursion (DE) were measured using a real time sonographic system with a 3.8 MHz convex probe (Aloka prosound 4000, Aloka, Japan ultrasound machine) at a depth of 22-24 Cm. The distance difference between the same leading edge of the diaphragm at the end expiration and end inspiration is calculated by M-mode tracing. During inspiration the diaphragm moves caudally towards the U/S probe and is represented by the upper peak on the M-mode, and during expiration the reverse occurs with the lower peak on M-mode represents the expiration. The difference between the peaks on M-mode is calculated by the machine and it represents the diaphragmatic movement (excursion) in the respiratory cycle that was ranged between 1.7- 2.5 cm.

Training Procedures:-

All patients in this study received the following instructions: Patients were instructed to wear loose, comfortable clothing especially around neck; they were rested in a sitting position. The training started by setting the maximum

load in which the patient could successfully execute ten breaths at maximum resistance depending on the patients' rate of perceived exertion, the starting load equal to 30% of the patient's maximum inspiratory effort. This individualized load increase progressively as the inspiratory muscle became stronger. The recommended pressure load determined by the 30% maximum 10-repetition method by using IMTdevice¹¹. The mouthpiece was attached to the device and nose clip on the patient's nose; so that all of the breathing was done through the mouth making sure the lips were sealed around the mouthpiece and the tongue not occlude it. The patients were asked to take full breath in (maximal and deep inspiration) then longer and slow expiration through mouthpiece. Continue this breathing pattern for 10-20 breath. Repeat this pattern for 4-6, 4-6 times, or about 10-15 minutes with rest in between 30 second. The session was repeated three times/week once daily for 12 weeks. The time of the session was at the morning prior to the dialysis session.

Results:-

Data obtained prior and following management including diaphragmatic excursion was statistically analyzed by SpSS. 20 computer statistical package.

Demographic data:-

Thirty patients (10 men & 20 Women) age ranged from 26 to 45 years old with mean age (\pm SD) was 35.47 (\pm 6.51), their height ranged from 1.56 to 1.8m with mean height 1.68(\pm 0.06), their weight ranged from 65 to 85 kg with mean weight 72.4(\pm 5.02) and their BMI ranged from 21.95 to 27.9 kg/ m² with mean BMI 25.63(\pm 1.46).

Studied parameters assessment:-

Table (1):- Statistical Analysis of Diaphragmatic Excursion measured pre- and post-training for the patients.

	Range	Mean \pm SD	Mean difference	Percentage of improvement	t-value	P value
Pre-training	1.70-2.50	2.16 \pm 0.25	-1.42	\uparrow 65.74%	-61.856	0.001**
Post-training	3.00-4.00	3.58 \pm 0.34				

**p< 0.01= highly significant.

Statistical analysis showed that increased significant improvement in diaphragmatic excursion that was shown in the mean difference (-1.42) with a percentage of improvement of 65.74% when compared to the pre- measured data that was shown in table (1) & figures (1&2).

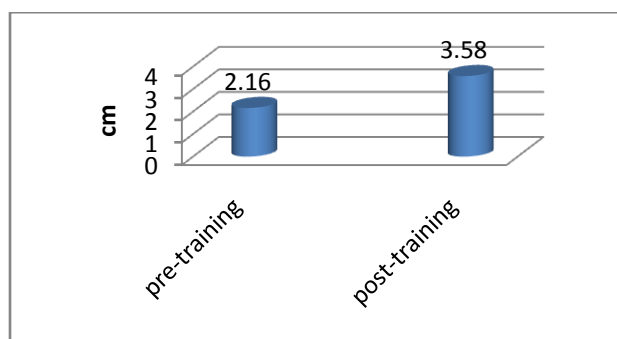


Fig.(1) :- Diaphragmatic Excursion of the Studied Group

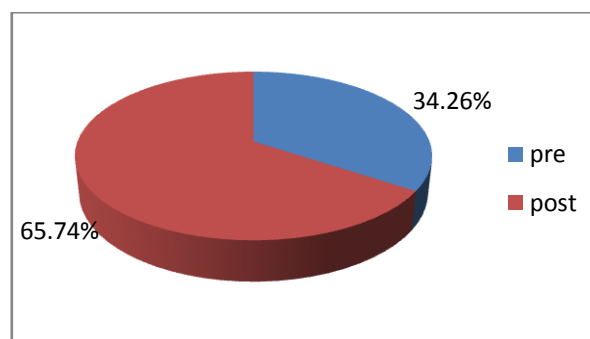


Fig.(2) :- Percent of improvement throughout the study

Table (2) :- Statistical Analysis of diaphragmatic excursion for men & women throughout the study:-

	Men (n=10)		Women (n=20)		MD	t-Value	P-value
	Pre	Post	Pre	Post			
Range	1.7-2.1	3-3.35	2-2.5	3.5-4			
mean \pm SD	1.93\pm0.16	3.19\pm0.13	2.27\pm0.22	3.77\pm0.21			
MD	-1.26		1.50		-0.24	11.343	0.001**
% of improvement	\uparrow 65.29		\uparrow 66.08				
t-Value	-61.688		-62.761				
P-value	0.001**		0.001**				

MD: mean difference .

**p< 0.01= highly significant.

As shown in table (2) & figures (3 &4) there was a significant difference between the pre and post-training results in men by 65.29% and in women by 66.08% that gives the resultant difference that women achieved a greater improvement than men did .

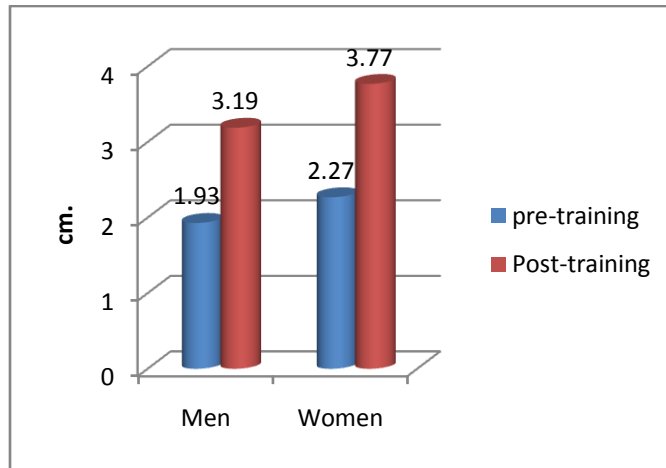


Fig.(3) : Diaphragmatic Excursion mean values pre- and post-training for men and women subgroups.

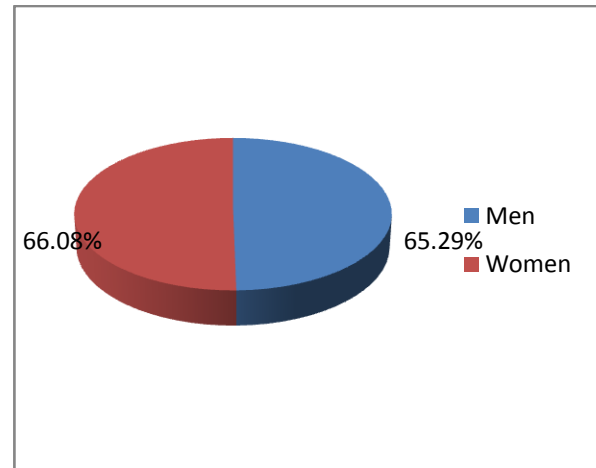


Fig.(4): The percentage of improvement represented in men and women.

Discussion:-

The analysis of the results of the current study showed that diaphragmatic excursion in hemodialysis patients that received inspiratory muscle training prior the session was significantly improved with percentage of 65.74% .This improvement in the diaphragmatic excursion may be due to improvement of diaphragm mechanics, power and inspiratory muscle strength. Studies involved the problem of strengthening of the diaphragm in hemodialysis patients is of great shortage.

The results of this study coincided with the results achieved by **Saad, 2013**¹² who **concluded** that inspiratory muscle training by IMT device for one month in post liver transplantation patients significantly improved the diaphragmatic excursion of these patients in addition to improvement of breathing mechanics for the study group.

The results of this study also agreed with the results achieved by **Saafan, 2013**¹³ who concluded that the inspiratory muscle training had significantly improved the diaphragmatic strength after upper abdominal surgeries.

A study results achieved by **Hamad , 2012**¹⁴ came in accordance with this study who concluded that inspiratory muscle training by IMT device for one month in postoperative thoracotomy patients significantly improved the diaphragmatic excursion of these patients in addition to improvement of breathing mechanics .

It was found that six weeks of post operative inspiratory muscle training using threshold IMT device significantly improved the ventilatory function and shortened the duration of postoperative hospitalization in patients gone through thoracic surgery¹⁵. This may be attributed to an improvement in the diaphragm strength, as evidenced by improvement in respiratory mechanics that led to better expansion of the chest which reflected as increase in the ventilatory function leading to early recovery of respiratory function impairment which consequently shortened the duration of postoperative hospitalization¹⁶.

Also the results of this study coincided with the results achieved by **Edwards and Cooke,2008**¹⁷ who concluded that the inspiratory muscle training device had a significant effect on maximum inspiratory pressure in patients underwent pulmonary lobectomy.

In agreement with the results of the current study **Hart et al., 2006**¹⁸ found that significant improvement in inspiratory muscle function and diaphragmatic excursion which were also associated with improved psychosocial status were noticed after eight weeks of high intensity IMT in patients with cystic fibrosis which suggested that increasing inspiratory muscle function may have significant long-term health implications for patients with respiratory disability.

The results of this study coincided with the results of trials with IMT. The finding revealed that IMT intervention improves exercise capacity and quality of life, particularly in patients with inspiratory muscle weakness. Some benefit from IMT may be accounted for by the attenuation of the inspiratory muscle metabolic reflex. Moreover, IMT results in improved cardiovascular responses to exercise and to those obtained with standard aerobic training¹⁹.

In agreement with the results of the current study **Ysayama et al., 2008²⁰** showed that IMT for 4 weeks before esophagectomy resulted in a significant increase of respiratory muscle strength and endurance.

In contrast **Mc Gill and Brown 2011²¹** reported a non-significant change in diaphragmatic excursion (DE) with inspiratory muscle training program for 4 weeks and might be explained by short duration of the study.

Moreover, the results of this study shows that the percentage of improvement of diaphragmatic excursion in women is better than in men as women percentage of improvement is 66.08% and men percentage of improvement is 65.29% although the excursion is more stronger in men as their rhythm of respiration is abdominothoracic while the rhythm of respiration in women is thoracoabdominal that may be attributed to the fact of reduction of the inspiratory muscles strength with age in men 25+ & women 35+ and that must be taken into consideration the presence of weakness already with hemodialysis. With IMT women with their smaller size of the lung experienced much better benefit of the mechanics of respiration.

Conclusion:-

Inspiratory muscle training by IMT device is an effective rehabilitation method that improves diaphragmatic excursion (DE), sense of breathlessness and functional capacity in hemodialysis patients.

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RESEARCH ARTICLE

STATUS OF INFORMATION AND COMMUNICATION TECHNOLOGY PROGRAM AND NATIONAL ACHIEVEMENT TEST PERFORMANCE OF PUBLIC SCHOOLS IN THE DIVISION OF LAGUNA, PHILIPPINES.

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Key words:-

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Abstract

The study sought to determine the status and indicators of teachers' ICT competency and National Achievement Test performance of pupils; relationship between the teachers' demographic profile and teachers' ICT competency, schools' ICT program and ICT infrastructure. The descriptive correlation survey was adopted and 1,012 teachers from 9 districts in the Division of Laguna were given questionnaire. The statements on competencies were based from the 4 domains of National ICT Competency Standards for Teachers which are social and ethical, technology and operation concepts, pedagogical, and professional domains. The findings revealed that competence on social and ethical, technology and operation concepts, pedagogical and professional domains obtained means were interpreted at basic level. In terms of NAT performance of 42 schools, 17 are big schools and 25 are small schools, only 4 schools obtained Moving-Towards-Mastery (MTM). The status of School's ICT program in selected clusters in terms of Teachers' ICT competence and Curriculum Integration with mean scores of 2.10, and ICT Infrastructure mean of 1.64, with an overall mean of 2.02 was interpreted at Progressing level. The relationship between the teachers' demographic profiles and teachers' ICT competency was found significant, however no significant associations were found between schools' ICT program and ICT infrastructure. All indicators of ICT competency skills of teacher respondents were not significant and did not affect significantly the NAT performance. The school administrators need to provide in-service trainings on ICT, improve schools' ICT facilities to enhance teachers' ICT competency skills and increase level of ICT integration to instruction.

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Introduction:-

The Department of Education (2009)³ of the Philippines points that Information and Communication Technology (ICT) must be an integral part of all learning areas, both hardware and software. Hence, according from Department

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of Education (DepEd), e-learning and the use and application of ICT must be employed in all subjects because this is now the most viable intervention in educational reform.

DepEd is giving its best to make Computerization Program one of the most functional, if not the greatest program, being implemented at present. The times call for education to aim towards advancement of technology. This can only happen with the installation and use of computers and other equipment which call forth for modern technology and advance learning.

The Human Development Report of the United Nations Development Program or UNDP (2001) states, "ICT is a pervasive input to almost all human activities. It breaks barriers to human development in at least three ways: by breaking barriers to knowledge, by breaking barriers to participation and by breaking barriers to economic opportunity." Even in the daily lives of teachers and school heads, the rapid advances in ICT can create new things such as storing information, retrieving information, handling, distributing and utilizing information. The advancement of ICT according to Ocampo (2002)⁹ is intertwined with globalization and together creates a new paradigm called the network age.

Despite the efforts of DepEd, various studies conducted on limited samples of teachers reflect alarmingly low levels of teacher competency. For instance, a study in 2004 revealed that only 1 out of 7 schools had teachers who were computer literate (Java, 2004)⁶. Two years back, a study found out that seventy-five percent of respondent schools had only 10 percent of their teachers with Internet skills (Tinio, 2002)¹⁰. These studies reveal a nuance that the teachers are deficient in ICT skills. This necessitates the conduct of a definitive study which seeks to measure the ICT competencies of Filipino teachers.

There is a growing challenge in the Philippines in the appropriate and efficient use of ICT in teaching and learning. With the multitudes of programs launched by both private and public sectors there are questions of what happened, present status, and where the program is heading. The resources poured into ICT in the supply of equipment, access to software, and availability of telecommunication infrastructures, capacity building for teachers, among others have reported to have been made in collaboration with the private sectors specifically Microsoft, Intel, and major telecommunications companies, organizations, and other governments with DepEd. (DBM, 2009).

It is imperative to examine the situation of learners and how information and communication tools help them prepare for the real life. This study aimed to analyze the different domains, causes of problems, and relationships that have bearing to the teaching and learning process. Hence, a research entitled Status of Information and Communication Technology Program and National Achievement Test Performance of Public Schools in the Division of Laguna, Philippines was conceived

Research Methods:-

Sampling Technique. This study was conducted in the Schools Division of Laguna, Philippines. It is composed of 2 selected clusters which are Cluster 2 and 3 with 9 selected districts with 42 selected elementary schools. Specifically, the 9 districts are Los Banos, Victoria, Bay, Magdalena-Majayjay, Cabuyao, Calauan, Alaminos and Cavinti Districts, all are located in the province of Laguna. Primarily, this study relied on the result of the quantitative data gathered. Thus, more number of schools were needed. Since the source of data is mainly quantitative, a large number of respondents were imperative. Purposive random sampling was done in the selection of schools. Central (big school) and non-central (small school) schools from each district were taken as samples. It was purposely chosen to make the findings generalizable to the population where schools from different categories are well-represented. Central and non-central schools were involved in this study to include instances chosen deliberately to reflect diversity on presumptively important dimensions from schools with varied predictors. Schools were not chosen randomly but rather purposively and conveniently.

Respondents of the Study. The 1,012 public elementary school teachers were selected from clustered 9 districts in the Division of Laguna. The teachers were taken as the primary respondents because they are the front liners in attaining Quality Education through the use of ICT in their teaching-learning process.

Research Instrument. The survey questionnaire for elementary school teachers on perceived ICT competencies and school's ICT program was used as the main instrument of the study. It was developed, validated and mainly utilized to answer the research questions and to find out the perceived competencies, extent of use and training needs on ICT

of public school teachers in selected districts in Division of Laguna. Their ICT competencies were measured using the questionnaire and the results of the survey contributed as bases in the National Achievement Test Performance of grade 6 pupils and to equip elementary school teachers with knowledge and skills on ICT as an appropriate tool in the teaching-learning process. Part I of the research questionnaire is the respondents' demographic profile. The questionnaire was patterned to several instruments. Part of which described the respondents' background information that includes; age, gender, civil status, family monthly income, highest educational attainment, length of service, position and number of ICT related trainings. Part II of the research questionnaire comprised of statements on competencies based on the 4 Domains of National ICT Competency Standards for Teachers (NICS for Teachers) which are the social and ethical, technology and operation concepts, pedagogical, and professional domains. The said domains and standards were customized to fit in the study. Schools' ICT inventory and checklist were also included to determine the status of schools' ICT program in the Division of Laguna. To gauge the capacity/capability of the Schools Division in terms of emerging, progressing, and advance, the researchers personally interviewed and gave the Division Planning Officer a questionnaire who is in charge of the physical facilities of the whole division. The questionnaire was validated by 3 experts from DepED Division of Laguna. After validating the questionnaire, it was pilot tested with 9 principals, 5 from central schools and 4 from non-central schools to ensure the reliability, validity and accuracy of the research instrument. It set an accepted level of proficiency by portraying demonstrable activities. These standards were concretized by specific indicators for the implemented questionnaire. These were measured by a Likert scale type of instrument where 1 shows a weak grasp of the indicator and 4 is the highest, showing high proficiency. The responses of the respondents were tabulated and analyzed using the Statistical Software (SPSS). The researcher used descriptive statistical technique to calculate the frequencies, percentage distribution and means and standard deviation of each response to present the details about the teacher profile, ICT competency of the teachers. Independent Sample's T-Test, Pearson, Regression were done to check the significant relationship between perceptions of the respondents in ICT competencies and school's ICT program.

Research Procedure. The researchers sought the permission of the Schools Division Superintendent, district supervisors and school heads of the Division of Laguna. Before the distribution of the questionnaires, the researchers met the administrators and explained to them the purpose of the study. Distribution of the questionnaires to the administrators and their teachers followed. The researchers then administered the questionnaires personally from one school to another in the identified public elementary schools. Documents from the Districts and Schools' Planning Offices were also analyzed to determine the status of schools' ICT program from school years 2011-2014. To answer the research problems, the instrument on ICT competencies, schools' ICT program, and NAT Performance of grade 6 were used. The weighted mean and qualitative descriptions were utilized to determine the ICT competencies and ICT capabilities of schools in the division of Laguna. Upon the retrieval of questionnaires, data from elementary school teacher-respondents were quantified. Data analysis and findings of this study hinge primarily upon the results of quantitative data gathered from the surveys. The quantitative data were analyzed using the mean, standard deviation, percentages and frequencies to find out the profile of elementary school teachers as to age, gender, civil status, family monthly income, highest educational attainment, length of service, position and number of ICT related trainings. Mean and SD were used in computing the perceived ICT competencies, and ICT program of schools.

Results and Discussion:-

Figure 1 shows that a small scale of the respondents are male, from 1,012 respondents only 103 are male or 10.2%, while the female comprised 89.8% with a total of 909 or 89.8%.

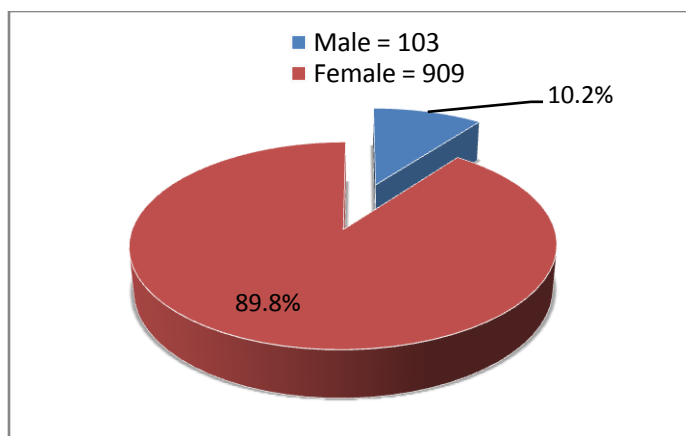


Figure 1:- Distribution of Gender of Teacher-Respondents

As presented in Figure 2, majority are married, comprising of 72.7% or 736, 21.8% are single, 4.3% are widow, and 1.1% or 11 are separated out of 1,012 teacher-respondents.

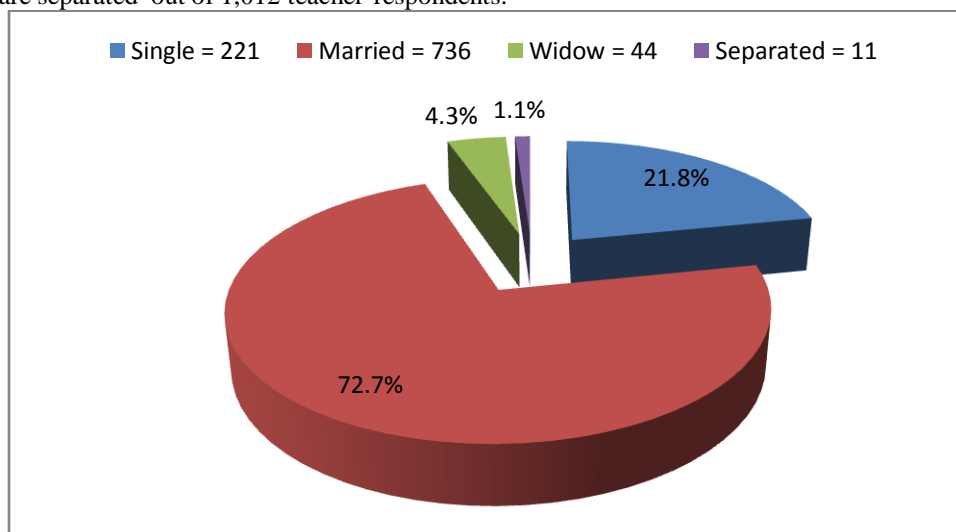


Figure 2:- Distribution of Civil Status of teacher-respondents

As revealed in Figure 3, among the 1,012 respondents, 558 or 55.1% are in the service for 10 years, while 28.4% or 287, 29 or 2.8% are in their 31 to 40 years in service and 1 is in the service for 40 years or more.

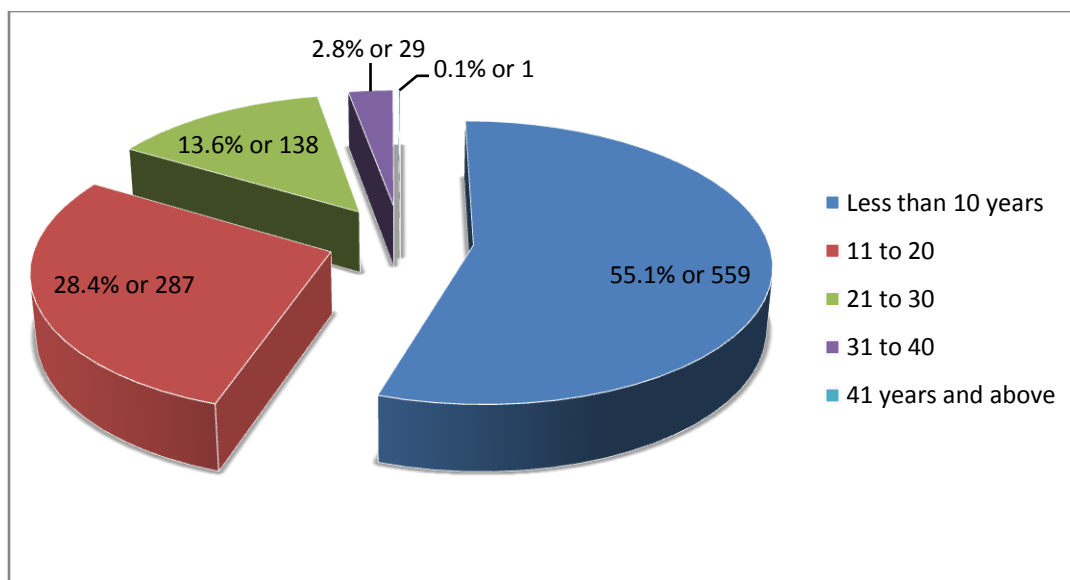


Figure 3:- Length of Service of the teacher-respondents

Table 1:- Level of Mean Perception of Respondents on ICT CompetencySkills

ICT COMPETENCY SKILLS	MEAN	SD	DESCRIPTIVE INTERPRETATION
SOCIAL AND ETHICAL	2.53	0.60	PROFICIENT
TECHNOLOGY AND OPERATION CONCEPTS	2.41	0.76	BASIC
PEDAGOGICAL	2.14	0.75	BASIC
PROFESSIONAL	2.2	0.72	BASIC
OVERALL	2.32	0.71	BASIC

Legend: 1.00-1.50-Below Basic, 1.51-2.50-Basic, 2.51-3.50-Proficient, 3.51-4.00-Highly Proficient

The respondents' ICT competence on social and ethical with a mean of 2.53 and interpreted as proficient, while technology and operation concepts, pedagogical and professional domains scored 2.41, 2.14 and 2.2, respectively were described as basic. The findings confirmed with the study of Kim and Baylor (2008)⁷ that the use of technology for teaching requires the development not only of knowledge, skills, and behaviors but also of appropriate attitude, social, and ethical foundation. Chang (2012)² posits that to be an experienced and capable technological leader, school administrators such as the principal, must be trained in vision, planning and management, how school reform will be affected by technology and ICT tools and infrastructure support, evaluation and research. Further, according from the study of

Table 2 shows that among 42 schools, only 4 schools have reached Moving-Towards-Mastery (MTM) in the NAT. The school with highest mean percentage score was obtained by C5 S4 (small school) with an MPS of 75.82; followed by C5 S3 (small school) with 69.05 mean percentage score and C5 B2 (big school) with 67.74, and C6 S1 (small school) with 66.29 mean percentages scores, respectively.

Table 2:- Mean Percentage Scores of National Achievement Test of Grade 6 in Selected Clustered Districts and Schools

Clustered Districts and Schools	NAT VI Mean Percentage Scores (MPS)	Mastery Level Descriptive Equivalent
C5 B1	60.91	Average
C5 B2	67.64	Moving-Towards-Mastery
C5 B3	54.70	Average
C5 B4	58.28	Average
C6 B1	60.06	Average
C6 B2	57.63	Average
C6 B3	43.35	Average
C6 B4	44.32	Average
C6 B5	45.97	Average
C7 B1	47.77	Average
C7 B2	50.92	Average
C7 B3	57.41	Average
C7 B4	51.08	Average
C8 B1	43.31	Average
C8 B2	49.72	Average
C8 B3	58.26	Average
C8 B4	52.42	Average
C5 S1	62.84	Average
C5 S2	50.36	Average
C5 S3	69.05	Moving-Towards-Mastery
C5 S4	75.82	Moving-Towards-Mastery
C5 S5	58.01	Average
C5 S6	65.91	Average
C6 S1	66.29	Moving-Towards-Mastery
C6 S2	64.55	Average
C6 S3	53.07	Average
C6 S4	53.70	Average
C6 S5	49.13	Average
C6 S6	50.67	Average
C6 S7	40.17	Average
C6 S8	40.72	Average
C7 S1	64.10	Average
C7 S2	54.21	Average
C7 S3	64.67	Average
C7 S4	53.86	Average
C7 S5	61.88	Average
C7 S6	58.20	Average
C7 S7	47.37	Average
C8 S1	52.38	Average
C8 S2	57.20	Average
C8 S3	45.97	Average
C8-S4	48.12	Average
Overall	55.05	AVERAGE

Mastery Level Descriptive Equivalent: 0%-4% Absolutely No Mastery (ANM), 5%-14% Very Low (VL), 15%-34% Low (L), 35%-65% Average (AVR), 66%-85% Moving Towards Mastery (MTM), 86%-95% Closely Approximating Mastery (CAM), 96%-100% Mastered (M)

With reference to the study of ELNORD(2006)⁴, ICT generally has a positive impact on learning but the expectations that ICT could in some ways revolutionize processes at school have not been realized. This goes beyond the use of computers by teachers since not only personal computers (PCs) and the Internet but also digital

cameras, mobile phones and other technologies can help to change teaching processes. The use of ICT is mostly focused on supporting the subject content, hence ICT-based activities by pupils are more time consuming than producing output. Moreover, the impact of integrating ICT in teaching can be measured in pupil engagement, differentiation, creativity and by the fact that less time is wasted, though the impact of ICT is very dependent on how it is used. Headmasters typically view ICT as a valuable tool for pedagogical development but very few of them actually experience this impact

Table 3:- Status of Schools' ICT Program

Components of Schools' ICT Program	SD	Mean	Descriptive Interpretation
Teachers' ICT Competence	0.66	2.10	Progressing
ICT Infrastructures	0.48	1.64	Progressing
Curriculum Integration	0.69	2.10	Progressing
Overall	0.49	2.02	Progressing

Legend: 1.00-1.50-Emerging 1.51-2.50-Progressing 2.51-3.00-Advanced

Table 3 reveals that the status of Schools' ICT Program in selected clusters are progressing in terms of Teachers' ICT Competence and Curriculum Integration with a mean of 2.10 and ICT Infrastructure with 1.64 mean score. Kozma (2008)⁸ acknowledges that ICT strategies in many countries cut across diverse fields. Infrastructure development is necessary in order to ensure access to schools, networks and resources for learning. Teacher training both initial and in-service is a prerequisite for the ability of education to use ICT in learning processes. Curricula and pedagogical approaches may have to be changed in order to cater for educational change with ICT.

Table 4 revealed significant associations between ICT School Program (in terms of teachers competence) and ICT Competency skills in social and ethical, and technology and operation concepts with p-value of less than 0.05. A study conducted by Ministry of Education of Norway (2006) states that the extent of teachers' education has implemented courses and strategies towards the increased competence of teachers in using ICT which is an important part of educational development and change. This could be seen as ICT literacy indicators for teacher education, and of how teachers are prepared to face the challenges in their practice as teachers.

Table 4:- Relationship between the Respondents' Perceptions on ICT Competency Skills and Schools' ICT Program in terms of Teachers' Competence

ICT Competency Skills	Correlation Coefficient	p-value
1 Social and Ethical	0.316	0.041*
2 Technology and Operations	0.293	0.059
3 Pedagogy	0.137	0.387
4 Professional	0.216	0.170

Table 5 shows that significant associations were not found between ICT School Program (in terms of Schools' ICT Program) and all Respondents' Perception ICT Competency Skills (p value is more than 0.05).

Table 5:- Relationship between the Respondents' Perception on the ICT Competency Skills and Overall Schools' ICT Program

ICT Competency Skills	Correlation Coefficient	p-value
1. Social and Ethical	0.227	0.149
2. Technology and Operations	0.249	0.111
3. Pedagogy	0.060	0.707
4. Professional	0.112	0.479

In accordance to the study of Baylor and Ritchie (2002)¹, which states that it is useful for school leaders as it underlines the importance of having a shared vision on the place of ICT in education and having a school based ICT policy plan. Such a plan acts as a blueprint for the sequence of goals a school hopes to achieve. It also outlines the overall philosophy of technology use and indicates how ICT will improve teaching and learning.

Table 6:- Relationship between Schools' ICT Program and NAT Performance

Schools' ICT Program	p-value
1. Teachers' Competence	0.466
2. Facilities and Infrastructure	0.426
3. Curriculum Integration	0.203

As could be gleaned in Table 6 , the Schools' ICT components were not significant and does not affect significantly the NAT performance. According to Honey (2005)⁵, efforts to integrate technology into schools and classroom practices must not only be acknowledged but also provide evidence that technology assists in meeting these accountability demands. School systems must prove the investments made in technology are improving student scores on standardized tests.

Conclusions:-

The respondents' ICT competence on social and ethical was described as Proficient, while technology and operation concepts, pedagogical and professional domains were described as Basic. It is therefore imperative that teachers need to be supported with ICT infrastructure for simulation and blended learning. Since only 4 schools have reached Moving-Towards-Mastery (MTM) level in the National Achievement Test, ICT integration in the basic education curriculum in all learning areas, to wit Mathematics, English, Science, Filipino and Social Studies is recommended. The interactive approaches in the learning process will enhance the performance of pupils in the standardized test administered by the Department of Education. The status of Schools' ICT Program in selected clusters are progressing in terms of Teachers' ICT Competence and Curriculum Integration. Significant associations were not found between ICT School Program (in terms of Schools' ICT Program) and all Respondents' Perception ICT Competency Skills. The Schools' ICT components were not significant and does not affect significantly the NAT performance. The school administrators need to provide in-service trainings on ICT, improve schools' ICT facilities to enhance teachers' ICT competency skills and increase level of ICT integration to instruction.

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REVIEW ARTICLE

LASERS FOR ESTHETIC REMOVAL OF GINGIVAL HYPERPIGMENTATION: A SYSTEMATIC REVIEW OF RANDOMIZED CLINICAL TRIALS.

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Abstract

Background: Being a science of beauty, beautiful smile is an integral part of contemporary dental practice. Gingival hyperpigmentation is considered an esthetic problem that needs a patient centered management. Dental operators are attracted to laser benefits not only during surgery but also for postoperative status. This review aimed to assess laser use in esthetic removal of gingival hyperpigmentation in comparison to conventional methods.

Materials and Methods: An electronic database search on PubMed and Cochrane library without date restriction was done and clinical studies in which laser was used for esthetic gingival depigmentation were selected and evaluated. Out of the initial search that yielded 183 studies, 21 were considered potentially relevant for the present review, out of which only 4 were finally selected. They studied the effect of laser on depigmentation, chair time, bleeding index, patient satisfaction and repigmentation.

Results: The analysis of the results shows that the assessed studies are too limited in number beside exhibiting small sample sizes. They are clinically heterogeneous so that a solid evidence based conclusion cannot be reached.

Conclusion: Researchers should be attracted to this research gap. Randomized clinical trials with a larger sample size, different laser parameters and longer follow up are strongly recommended.

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Introduction:-

Being a science of beauty, beautiful smile is an integral part of contemporary dental practice. Good smile involves not only teeth shape and color but also the gingival status. Pink gingival color is more favorable than dark one. Melanin hyperpigmentation or darker gingival color can be considered an aesthetic concern rather than a medical problem (Basha *et al.* 2015).

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Dentists should be prepared with the latest evidence based practice for management of such cases. The selection of technique should be based not only on clinical experience and doctor preferences but also on patient preferences (Thangavelu *et al.* 2012). Various methods can be used with varied degrees of success (Roshna and Nandakumar 2005)

Gingival hyperpigmentation can be defined as a darker gingival color beyond what is normally expected. The most common cause is racial pigmentation while other causes includes physiologic and/or pathologic factors (Dummett and Barens 1967). Gingival depigmentation is the treatment aiming to remove the melanin hyperpigmentation and it can be done by several techniques including mechanical, surgical, electrosurgical, cryosurgical, free gingival grafts, and lasers (Cicek and Ertas 2003, Hegde *et al.* 2013, Chandna and Kedige 2015). Surgical stripping was the old gold standard but now lasers have gained importance in the past years (Hegde *et al.* 2013, Basha *et al.* 2015).

A critical concern in the management of hyperpigmented gingiva is relapse or gingival repigmentation which can be defined as the reappearance of pigmentation after a period of no clinically hyperpigmentation. Recurrence usually varies according to the used methodology and the length of follow-up (Hegde *et al.* 2013).

The clinical rationale for comparing laser to conventional methods can be simply based upon the need for minimally invasive techniques for achieving a patient relevant outcomes (Ribeiro *et al.* 2014).

Based on this background, the aim of the present systematic review is to evaluate and compare the effects of conventional techniques versus lasers for gingival depigmentation as regards to removal of pigmentation, patient preference and time taken for repigmentation.

Materials and Methods:-

Protocol:-

The methods as well as inclusion/exclusion criteria used in the present study were determined in advance. The current systematic review was performed following the PRISMA guidelines for identification, screening, eligibility, and inclusion (Moher *et al.* 2010). The following focus question was developed: In patient requiring esthetic removal of gingival hyperpigmentation, is laser use recommended over conventional techniques, what is the evidence based recommendation for both patients and operators?

Information Sources:-

The electronic search was performed in two databases, MEDLINE (PubMed) and Cochrane library databases for articles with no date restrictions.

Search:-

The researched keywords were: (Black OR pigment OR melanin OR pigmentation OR hyperpigmentation OR pigmented OR dark OR Pigmentary OR pigmentations OR melanin) AND (Lased OR laser OR LLLT) AND (Gingiva OR mucosa OR gingival OR mucosal OR gum).

Selection of studies:-

Titles and abstracts resulted from the search were screened by authors considering the inclusion criteria. Authors decisions about choices and their qualification for further analysis was affirmed after discussion.

Inclusion and exclusion criteria:-

Inclusion criteria:-

- Randomized Clinical trials only
- Laser compared to conventional methods

Exclusion criteria:-

- Pigmentation due to amalgam tattoo, nevus or syndromes
- Case reports
- In vitro and animal studies
- Languages other than English

Assessment of methodological quality:-

The quality of all chosen randomized trials was investigated utilizing The Cochrane Collaboration's tool for evaluating risk of bias (Higgins *et al.* 2011).

According to Cochrane risk of bias tools, each RCT was assigned either; low risk of bias (if it is low for all key domains), high risk of bias (if it is high for one or more of key domains) and unclear risk of bias (if it is unclear for one or more of key domains). Because it was impossible to blind participants or personnel due to nature of intervention and control (i.e. Laser VS surgery), the “BLINDING OF PARTICIPANTS, PERSONNEL” item was not considered.

Results:-

Out of the initial search that yielded 183 studies, 21 were considered potentially relevant for the present study, out of which 4 were finally selected. Figure (1) represents the flow chart for the study. The excluded studies before final inclusion were uncontrolled or without conventional control (Trelles *et al.* 1993, Hanada *et al.* 1996, Nakamura *et al.* 1999, Ozbayrak *et al.* 2000, Tal *et al.* 2003, Esen *et al.* 2004, Azzeh 2007, Kaya *et al.* 2012, Giannelli *et al.* 2014, Kishore *et al.* 2014, Soliman *et al.* 2014, El Shenawy *et al.* 2015), review article (Karydis *et al.* 2011), uncontrolled case reports (Rosa *et al.* 2007, Bin Cho *et al.* 2010), non-randomized trial (Grover *et al.* 2014) and the study of Giannelli *et al.* which was a thermographic and fluorescent study for optimizing gingival laser photoablation (Giannelli *et al.* 2013).

The final included studies were four (Hegde *et al.* 2013, Ribeiro *et al.* 2014, Basha *et al.* 2015, Chandna and Kedige 2015). The heterogeneity between trials prevented meta-analysis. Rather, a descriptive analysis of the reported studies was performed. Table (1) represents summary of findings.

Risk of bias within studies:-

The review included 4 studies, 3 studies (Hegde *et al.* 2013, Ribeiro *et al.* 2014, Basha *et al.* 2015) used split mouth study design and 1 study (Chandna and Kedige 2015) used parallel design. Sex distribution was mentioned in 3 studies (Hegde *et al.* 2013, Ribeiro *et al.* 2014, Chandna and Kedige 2015) and not mentioned in 1 study (Basha *et al.* 2015).

For RCT studies, according to Cochrane Risk of Bias Tool, all studies were judged as low risk of bias for incomplete outcome data, selective reporting and other bias. Regarding random sequence generation, 3 studies (Hegde *et al.* 2013, Ribeiro *et al.* 2014, Basha *et al.* 2015) were judged as low risk of bias while it was judged as unclear for Chandna and Kedige 2015 study as it didn't mention type of randomization used. Allocation concealment was unclear for all included studies. Regarding Blinding of outcome assessment, 3 studies (Hegde *et al.* 2013, Basha *et al.* 2015, Chandna and Kedige 2015) were judged as unclear as there was no any mention about blinding of the assessor or statistician while it was low risk of bias for Ribeiro *et al.* 2014 study. All studies were considered overall as unclear risk of bias. Figure (2) represents risk of bias summary: review authors' judgements about each risk of bias item for each included RCT study while Figure (3) represents risk of bias graph: review authors' judgements about each risk of bias item presented as percentages across all included RCT studies.

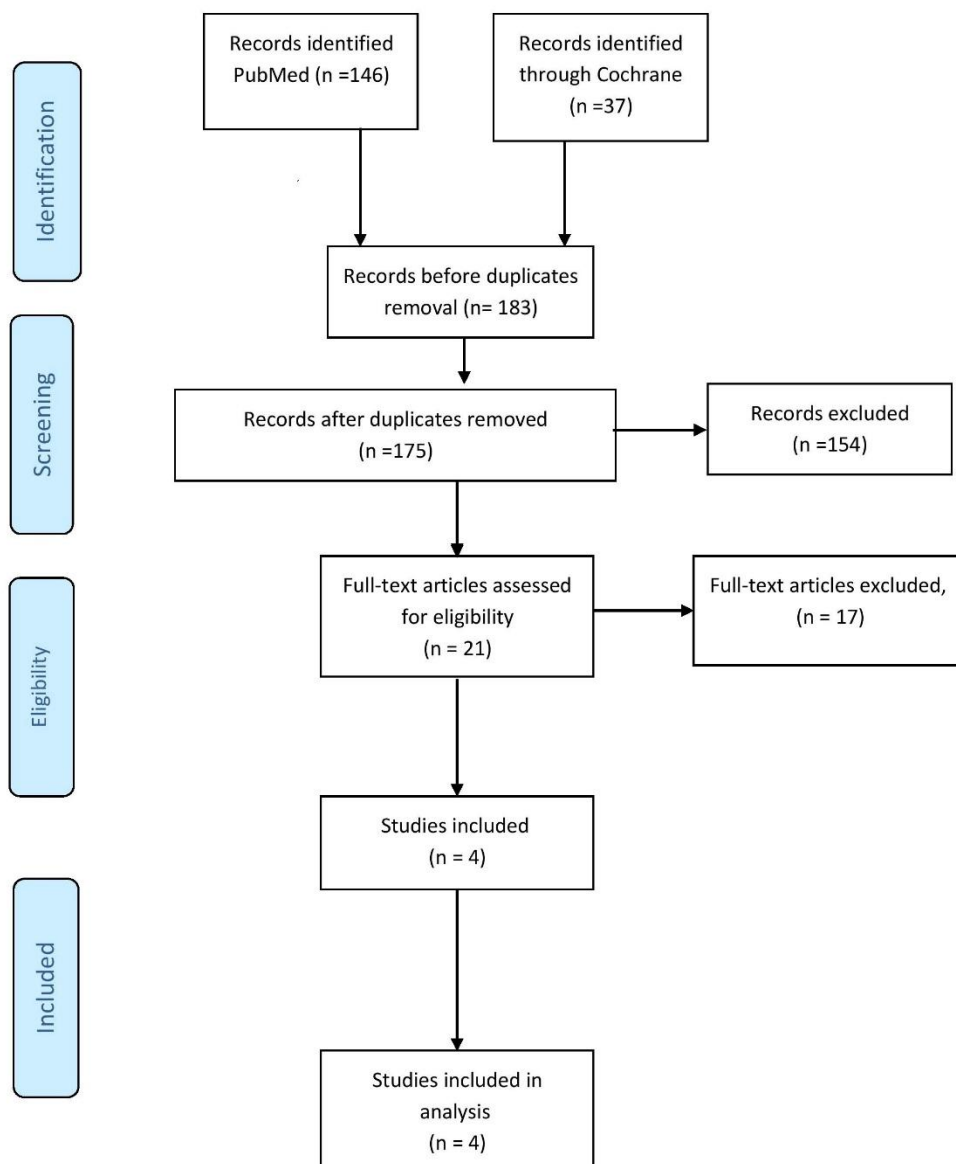


Figure 1:- Prisma flow chart for the study.

	Random sequence generation (selection bias)	Allocation concealment (selection bias)	Blinding of outcome assessment (detection bias)	Incomplete outcome data (attrition bias)	Selective reporting (reporting bias)	Other bias
Basha et al 2015	+	?	?	+	+	+
Chandna AND Kedige 2015	?	?	?	+	+	+
Hegde et al 2013	+	?	?	+	+	+
Ribeiro et al 2014	+	?	+	+	+	+

Figure 2:- Risk of bias summary: review authors' judgements about each risk of bias item for each included RCT study.



Figure 3:- Risk of bias graph: review authors' judgements about each risk of bias item presented as percentages across all included RCT studies.

Table 1:- Summary of findings table.

Authors and date	Population	Laser type <u>Vs</u> Control	Outcome
Basha et al 2015	20 patients with 40 bilateral maxillary sites	Nd: YAG laser <u>Vs</u> Scalpel surgery	Intergroup comparison for time taken, pain, and patient preference was statistically significant in favor of laser. Intragroup comparison for laser and surgical stripping at 6 months for Dummett Oral Pigmentation Index (DOPI), Hedin index, and size of pigmented area was statistically significant. Non-statistical difference in other results.
Hegde et al 2013	35 patients with 140 bilateral sites	CO2 and Er: YAG laser <u>Vs</u> Scalpel surgery	Thorough removal of pigments, pain reduction, and higher patient preference were associated with Er: YAG laser but more sites showed repigmentation. The CO2 laser was beneficial in terms of complete hemostasis intraoperatively and delaying the rate of repigmentation. Repigmentation was least after the surgical stripping procedure.
Ribeiro et al 2014	11 patients with bilateral anterior maxillary sites	Nd: YAG laser <u>Vs</u> Scalpel surgery	A statistically lesser chair time was observed in the laser group. A statistically lesser extent of discomfort/pain was in laser group during the first posttherapy week. Non-statistical difference in other results.
Chandna and Kedige 2015	20 patients (10 for each group)	<u>Diode laser</u> <u>Vs</u> Electro-surgery	A statistically highly significant difference in the pain levels between the two groups 24 h post-operatively in favor of laser group. Despite of lesser mean pain level for the laser group at other times, it was a non-significant difference.

Discussion:-

Lasers can be used as a contemporary alternative to the traditional scalpel in oral soft tissue surgeries because it offers simple manipulation as regards to absence of bleeding, reduced need for local anesthesia and increased patient comfort (Arnabat-Domínguez *et al.* 2010). From that points of view comes the research question, why not to recommend laser over conventional techniques.

Melanin is a word derived from the Greek one “melas” or black (Hegde *et al.* 2013). Melanin hyperpigmentation can affect all races and all ages with no gender predilection (Eisen 2000). Gingival hyperpigmentation is an esthetic problem for many patients, especially if visible when smiling. lasers have been suggested as an alternative to conventional procedures for providing a more patient centered outcome (Atsawasuwat *et al.* 2000). However, up to date, no evidence based recommendation for whether to go for the conventional strategy or replace it with the contemporary lasers.

The trials presented in this review agreed that both techniques are successfully used for the treatment of gingival hyperpigmentation but which one is recommended over the other, an issue that needs further discussion.

Blade surgery for gingival depigmentation is a definite precise and controlled procedure beside being the most economical procedure. Any depigmented areas are appreciated immediately and there is no chance for any residual pigments. On the other hands, this technique required the use of local anesthesia, associated with pain and hemorrhage and required care not to expose the bone or create gingival recession (Grover *et al.* 2014).

Also, periodontal dressing for at least a week is mandatory to guard against food debris, foreign irritants, thermal stimuli and infection (Gurumoorthy Kaarthikeyan *et al.* , Grover *et al.* 2014). The denuded tissues which heal by secondary intention after conventional surgery may promote bleeding after the procedure and is associated with some discomfort and pain for the patient (Ribeiro *et al.* 2014). The denuded connective tissue heals by secondary intention and is associated with pain and late repair (Roshna and Nandakumar 2005).

The expected complicated postoperative period, even if mild, may interfere with patient daily activities and well-being (**Ribeiro et al. 2014**). An experience that should be avoided by the modern patient centered care strategy.

Despite that surgical stripping has been the old gold standard for gingival depigmentation, lasers start to gain prominence in that field (**Coluzzi 2004**).

Using of lasers for oral soft tissues provides benefits not only to dental operators but also for the patients. Patient-centered outcomes are the main target for researches in the last decade (**Hujoel 2004, Ozcelik et al. 2007**).

Concerning the cosmetic outcomes, some aspects of using lasers may benefit the purpose as precise cutting and control and the application of delicate contact tips during the procedure which enhances trans-operative visualization (**Cobb 2006**).

One of the advantages of using laser is that patients felt no pain either during or after the treatment without requiring an additional anesthetic injection. Fear of pain during dental surgeries or even the needle used for anesthesia are among the causes of avoiding dental visits. Thanks to lasers that solve a part of that problem (**Matys and Dominiak 2016**).

The reduced pain in comparison with conventional surgery may be due to the protein coagulum formed on the surface of the wound that acts as a biologic dressing. Sealing of the ends of sensory nerves may be also considered which is not the case in surgical stripping where the nerve endings is left exposed (**Gurumoorthy Kaarthikeyan et al. , Hegde et al. 2013, Rathod and Mulay 2013, Basha et al. 2015**). A disrupted cell membrane $\text{Na}^+\text{-K}^+$ pump with the resulting loss of impulse conduction may be another cause of pain reduction associated with laser use (**Chandna and Kedige 2015**). The biologic dressing not only reduces pain but also protects the surgical area and provides a better postoperative repair with minimal damage to the tissue (**Ribeiro et al. 2014**).

Patients also appreciated the shorter operating time and minimal bleeding which is not the case in patients treated with the surgical scalpel technique (**Hegde et al. 2013, Grover et al. 2014**). The time taken for laser treatment was less than the surgical stripping may be attributed to better visibility due to less bleeding because of the coagulation of blood vessels (**Ko et al. 2010, Ribeiro et al. 2014, Basha et al. 2015**).

Several studies have compared different lasers with varying results, similar advantages includes a relatively bloodless surgical field, minimal swelling and scarring, reduction of surgical time and less postoperative pain (**Giannelli et al. 2014, Kishore et al. 2014, Soliman et al. 2014, Basha et al. 2015**).

It is the ablative property of laser that makes it possible to be used for gingival depigmentation by targeting the melanin in melanocytes in epithelium which absorbs and converts light energy into heat or what is known as photothermolysis (**Nakamura et al. 1999, Coluzzi 2004**).

A variety of laser systems can be used for gingival depigmentation, including carbon dioxide (CO_2) laser (**Nakamura et al. 1999**), diode laser (**Kaya et al. 2012**), Er:YAG laser (**Kaya et al. 2012**), and Nd:YAG laser (**Atsawasuwan et al. 2000**).

Neodymium–yttrium aluminum garnet (Nd:YAG) laser treatment has been considered as an alternative for gingival hyperpigmentation because of rapid healing and minimal bleeding during and after the procedures (**Ko et al. 2010**). Fortunately, Nd:YAG laser is selectively absorbed by hyperpigmented tissues (**Cobb 2006**).

It is the inherent ability of diode lasers to be absorbed within the chromophores that allows controlled cutting with a limited depth of necrosis or by other words a tissue-specific ablation. The other advantage of diode laser is that its energy is transmitted through water and fortunately it is poorly absorbed in hydroxyapatite (**Convissar 2004**).

Low-power setting (≤ 2.5) is recommended during the procedure to avoid discomfort and pain during the post-operative period and for faster healing time. The gated pulsed mode is also recommended to provide the necessary thermal relaxation (**Berk et al. 2005, Cercadillo-Ibarguren et al. 2010, Kaya et al. 2012, Romeo et al. 2012, Grover et al. 2014**).

Esthetic gingival depigmentation is considered successful if there is delayed, minimal, or no repigmentation (**Hegde et al. 2013**). According to **Bergamaschi et al.**, permanent results are non-achievable (**Bergamaschi et al. 1993**). Recurrence rate may differ not only with the used treatment modalities but also according to the duration of follow-up (**Ribeiro et al. 2014**).

Many studies reported recurrence after laser depigmentations (**Bergamaschi et al. 1993, Nakamura et al. 1999, Esen et al. 2004, Rosa et al. 2007, Kaur et al. 2010**) while others reported no repigmentation (**Atsawasuwana et al. 2000, Tal et al. 2003, Azzeh 2007**). Grover et al. reported that no statistical significance was seen in the comparison of conventional and laser techniques in terms of repigmentation (**Grover et al. 2014**).

It was noted that repigmentation appeared in the papillary areas and marginal gingiva which may be due to difficulty in treating these areas with lasers because of lying near tooth structure and the risk of thermal damage which recommends lower energy dose with inadequate depth of penetration of the laser beam, incomplete depigmentation, and faster recurrence (**Hegde et al. 2013**).

To avoid recurrence, the gingival tissue should be entirely cleared of melanin to avoid recurrence by melanocytes migration (**Sharon et al. 2000**). Also, patient race, environmental exposure and smoking may play a role in repigmentation (**Ünsal et al. 2001, Sridharan et al. 2011**).

Interestingly, another aspect that may affect repigmentation is the number of performed laser sessions. All the studies that used multiple sessions for laser ablation reported no recurrence (**Tal et al. 2003, Azzeh 2007, Kaya et al. 2012**).

Also, repigmentation was more common in the maxillary arch in comparison to the mandibular arch which may be due to more exposure to environmental factors during smiling and speech, factors that are not related to the technique used (**Hegde et al. 2013**).

Laser biomodulation may be a double-edged weapon as while accelerating healing, melanocytes from adjacent areas migrate faster, leading to faster repigmentation (**Hegde et al. 2013**).

Laser application requires that the operators are trained about safe and effective use of lasers (**Ribeiro et al. 2014**). The surgical stripping gives the benefits of tactile sensation beside no risk of thermal damage to the underlying structures (**Hegde et al. 2013**).

It is important to mention that care should be taken not to injure the gingival margin and interdental papilla during ablation of pigmented gingival tissue in these delicate areas. Also, higher doses can cause like delayed wound healing promoted by severe coagulation or pain caused by uncontrolled carbonization (**Ribeiro et al. 2014**).

Laser requires more advanced technology and armamentarium and it causes higher financial costs than more economical scalpel technique (**Ribeiro et al. 2014**). The patients' perceptions of costs and benefits balance interfere in the decision about choosing laser or scalpel surgical therapy.

The advantages of lasers over conventional surgical procedures are: bloodless field, reduced bacteremia and mechanical trauma, minimal post-operative swelling and scarring and minimal post-operative pain. On the other hand, loss of tactile sensation and expensive and sophisticated equipment that needs special training and precautions are considered disadvantages (**Chandna and Kedige 2015**).

Conclusion:-

The found clinical studies are not only too limited in number but also exhibit small sample sizes, besides being clinically heterogeneous so that a solid conclusion cannot be reached. Researchers should pay attention to this interesting field to work upon. Each kind of laser application should pull attention of researchers in oral and maxillofacial field to close obvious, yet important, research gaps of lack of enough randomized clinical trials that can be relied upon to get a standard evidence based clinical practice.

Within the limitations of this review, it can be concluded that lasers can be used as a safer and effective treatment modality to provide optimal esthetics and enhanced comfort with lesser pain to the patients during the treatment for

gingival hyperpigmentation. However, surgical stripping is a cost-effective procedure for achieving gingival depigmentation. Higher patient preference and pain reduction favor laser depigmentation, but more sites showed repigmentation. The lower cost of surgical stripping and the lower rate for repigmentation favor the old technique. Further studies with a larger study samples and a longer follow-up period would be desirable with special concern on multiple laser sessions to reach a solid evidence based conclusion regarding which technique is better keeping in mind the effect on repigmentation.

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REVIEW ARTICLE

AN INTRODUCTION TO OCEAN ACIDIFICATION AND TEMPERATURE CHANGES ON PHYSIOLOGICAL TRADE-OFF OF INTERTIDAL MARINE GASTROPODS

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Abstract

Due to an equilibration of partial pressure of CO₂ (pCO₂) in the atmosphere, 30% of its concentration has been absorbed by ocean and making it more acidic and generate potential global warming. Future changes of ocean pH and temperature are predicted to impact biodiversity of marine ecosystems, particularly those animals that rely on calcification process. Reduced pH will induce dissolution rates of calcium mineral particularly aragonites and calcites and that alter decalcification rates. The reduction of pH also disrupts acid base balance and metabolic rates that lead to metabolic depression whilst increase temperature affects organisms' thermo-tolerance capacity. Even though decreased metabolic rates were associated with metabolic depression, a strategy to match oxygen demand and availability, however prolong exposure to these stressors have affected growth, survival and reproduction rates. In addition, increase CO₂ and temperature have also magnitude end-product metabolites such as succinic and lactic acids and reduced energy nucleotides (adenosine 5-triphosphate, adenosine diphosphate and adenosine monophosphate) in the cells, indicating an increased reliance on anaerobic metabolism. Furthermore, anthropogenic alteration of CO₂ and temperature may also lead to plastic responses, a fundamental mechanism of many marine gastropods to cope environmental variability. Shells of marine gastropod were also more globular in order to defend desiccation rates, a primary threat to most intertidal organisms to elevated temperature particularly those that lack of mobilization aggregates (sessile). Although level of impacts may also vary from species to species as well as populations, however, physiologically, the result always came at cost.

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Introduction:-

Ocean acidification (OA) is the result of an ongoing process caused by an excessive increase of carbon dioxide (CO₂) into the world's oceans (Caldeira and Wickett 2003, Kleypas et al. 2006). This chemical change to seawater is driven by increased levels of atmospheric CO₂. For the past 650,000 years and prior to the Industrial Revolution, the concentration of atmospheric CO₂ was in the range of 180 to 300 ppm by volume (ppmv) (Augustin et al. 2004, Siegenthaler et al. 2005). However, Houghton et al. (2001) reported that CO₂ levels in the atmosphere had reached

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367 ppm in 1999 and will continue to increase by 1% per year over the next few decades. Current atmospheric CO₂ levels have increased to 380 ppm (Raven et al. 2005, Kleypas et al. 2006) with a resulting increased atmospheric partial pressure ($p\text{CO}_2$) of the gas (Jacobson 2005) (Fig. 1).

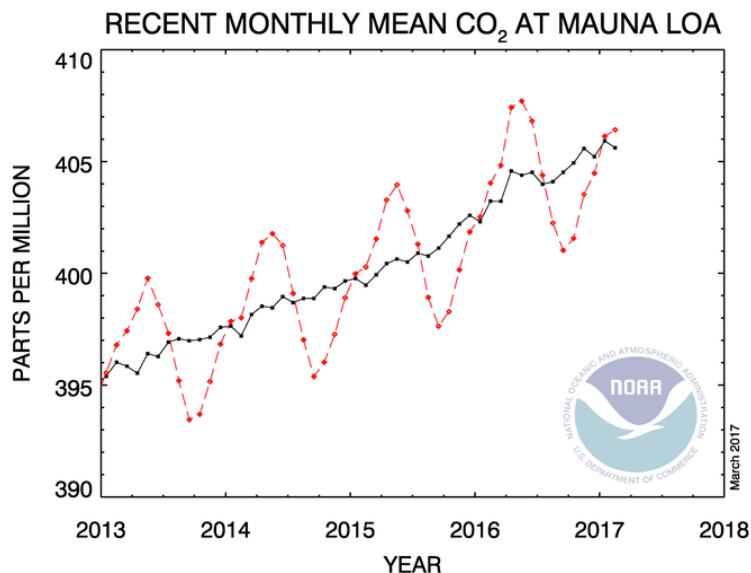


Figure 1:- The increase of mean atmospheric carbon dioxide (CO₂) at Mauna Loa Observatory, Hawaii. The CO₂ data constitutes the longest record of direct measurements of CO₂ in the atmosphere. The red line represents monthly mean values. The black curve represents seasonally corrected data.

(Source: <http://www.esrl.noaa.gov/gmd/ccgg/trends/>)

If CO₂ concentrations continue to increase in line with the current trend (as shown in Fig. 1) then it is predicted that atmospheric [CO₂] will increase by about 50% on the current concentrations by 2030 (McNeil and Matear 2008), exceeding 1000 ppmv by 2100 (Raven et al. 2005).

In general, the increase of CO₂ concentration in the atmosphere is caused by an imbalance of CO₂ exchange (input and output), for example, through the burning of fossil fuels, land-use changes and the industrial production of cement (Orr et al. 2005). Although CO₂ is also absorbed by terrestrial plants (Metaphytes), more than 40% of atmospheric CO₂ enters the oceans (Falkowski et al. 2000). There are several pathways in the carbon cycle which are particularly important for this process. Figure for the global carbon cycle (Fig.2) shows that there is a net flux of CO₂ into the oceans of 1.7×10^{15} g C y⁻¹.

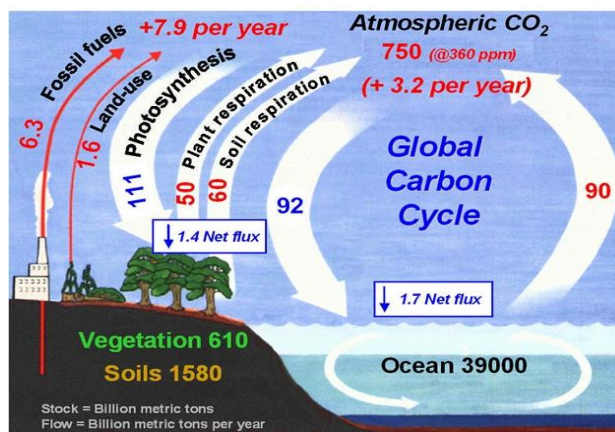


Figure 2:- The global carbon cycle pathways, pools and fluxes. About 3.2×10^{15} g C y⁻¹ of world production CO₂ enters the atmosphere. (Source: Kling et al. 2003).

Although CO_2 is not a chemically reactive gas in the atmosphere, its dissolution in seawater leads to several complex chemical, physical and biological changes (Raven et al. 2005). The initial reaction of CO_2 and water (H_2O) forms carbonic acid (H_2CO_3) which, in turn, forms bicarbonate (HCO_3^-) and, by dissociating H^+ ions, drives pH to lower values (Raven et al. 2005) (Fig. 3).

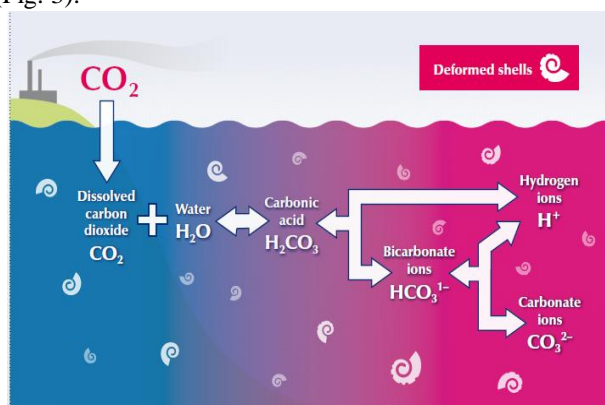


Figure 3:- Increased CO_2 in the atmosphere is being taken up by the ocean resulting in the sea water becoming less alkaline. As CO_2 dissolves in seawater, carbonate ions neutralise free hydrogen ions, causing carbonate levels to drop. (Source: University of Maryland in Andiman et al. 2012).

Therefore, increasing $[\text{CO}_2]$ ultimately leads to an increase in the concentration of carbonic acid (H_2CO_3) which will dissolve rapidly to bicarbonate (HCO_3^-), H^+ and carbonate ions (CO_3^{2-}) (Caldeira and Wickett 2003, Fabry et al. 2008). At present, ocean pH has been reduced by 0.1 units from ambient conditions, equivalent to a 30% increase in hydrogen ions (Sabine et al. 2004, Raven et al. 2005, Dashfield et al. 2008). Future predictions suggest that increased CO_2 levels will reduce seawater pH by 0.3 – 0.5 units by 2100 (Caldeira and Wickett 2003, 2005) and 0.77 units by 2300s if CO_2 release into the atmosphere continues to rise according to 'business as usual' scenario (IPCC 2007). In addition, Feely et al. (2008) have shown that corrosive water with lower aragonite saturation (Ω_{ara}) from the deep sea upwells to the continental shelf and coastal waters where it can affect intertidal regions. These coastal waters may already experience low aragonite saturation states (Feely et al. 2008, Wootton et al. 2008).

As well as altering the pH of the oceans, increased CO_2 has been suggested to be responsible for a 55% increase in radiative forcing (change in net solar radiation between different layers of the atmosphere in unit Wm^{-2}) (Jenkinson et al. 1991) and subsequent increases in air temperatures (McMullen 2009). Increased radiative forcing ultimately alters the incoming and outgoing energy balance between the Earth and its atmospheric system. When radiative forcing is positive, the Earth's surface tends to warm (IPCC 2007). In 2005 the mean value of radiative forcing was $+1.6$ indicating a rate of warming of $+1.6 \text{ Wm}^{-2}$ (IPCC 2007), which could increase to 8.0 Wm^{-2} by 2100 (Sokolov et al. 2009) (Fig. 4a).

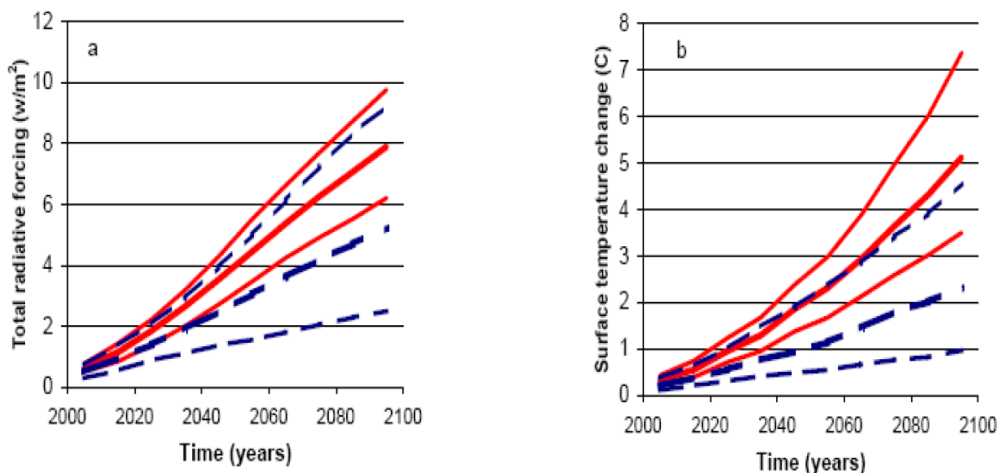


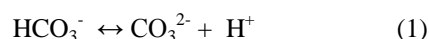
Figure 4:- Projected changes in climatic parameters between 2000 and 2100: a) decadal mean radiative force; b) sea surface temperature change. Red solid lines show median, 5% and 95% percentile values presented in the study by Sokolov et al. (2009) and dashed blue lines show median, 5% and 95% percentile predicted by Webster et al. (2003). (Source: Sokolov et al. 2009).

Jenkinson et al. (1991) also proposed that increased air temperatures would increase emissions of CO₂ gas from soils, through an increase in the turnover of organic matter (Jenkinson et al. 1991). Warming caused by increased atmospheric CO₂ levels will also increase ocean temperatures (Kleypas et al. 2006, Allison et al. 2008). At present, seawater temperature has warmed by approximately 0.7 °C compared with the pre-industrial era (Kleypas et al. 2006) and is predicted to rise by a further 2 °C by 2050 (Huesemann 2006, Guldborg et al. 2007).

Jacobson (2005) developed a model with uncertainty input parameters that predicted an increase of CO₂ to 375 ppmv there would be an increase in temperature of 3 °C. However this model has been corrected by Sokolov et al. (2009) with certainty input parameters (e.g. world volcanic eruptions and gross domestic product (GDP) growth) which predicts that sea surface temperature (SST) will increase between 3.5 and 7.4 °C with a median increase of 5.1 °C by 2100 (Fig. 4b).

Impact of Ocean Acidification and Temperature on Calcifying Organisms:-

Organisms that rely on calcified structures are thought to be particularly vulnerable to OA (Kleypas et al. 1999, Orr et al. 2005, Kleypas et al. 2006, Hendriks et al. 2010, Kroeker et al. 2010, Hale et al. 2011). The increased acidity of seawater reduces calcium carbonate saturation (Ω) and carbonate ion concentration (CO₃²⁻) according to the following reaction:



Therefore, increased H⁺ causes carbonate ions (CO₃²⁻) to react and form bicarbonate ions (HCO₃⁻). Decreases in carbonate ions will decrease the precipitation of CaCO₃ and the carbonate saturation state calculated as:

$$\Omega = \frac{[\text{Ca}^{2+}]_{\text{sw}} \times [\text{CO}_3^{2-}]_{\text{sw}}}{[\text{Ca}^{2+}]_{\text{sat}} \times [\text{CO}_3^{2-}]_{\text{sat}}} \quad (2)$$

In which the solution of saturation state of [Ca²⁺] and [CO₃²⁻] is calculated as:

$$k'_{\text{sp}} = [\text{Ca}^{2+}]_{\text{sat}} \times [\text{CO}_3^{2-}]_{\text{sat}} \quad (3)$$

where k'_{sp} is the stoichiometric solubility product for a particular mineral phase of CaCO₃ (i.e. calcite, aragonite, or high-magnesium calcite) (Kleypas et al. 1999, McNeil and Matear 2007, Gazeau et al. 2007, Boesch et al. 2010). Reduced calcium carbonate saturation can make the process of calcification more difficult (Sigler et al. 2008). Also with a carbonate saturation state less than 1 ($\Omega < 1$) CaCO₃ will start to dissolve, whereas at a value greater than 1 ($\Omega > 1$) CaCO₃ will spontaneously precipitate. Calcium carbonate is represented in seawater in two forms, aragonite and calcite (Raven et al. 2005, Gazeau et al. 2007), which are used in the construction of skeletons and shells in all calcifying organisms (e.g. Bowen and Tang 1996, Thompson et al. 2000, Schmidt 2005, Addadi et al. 2006, Gazeau et al. 2007). Both are soluble in acidified seawater and, although calcite is 35% less soluble than aragonite, under acidic conditions both minerals displayed no consistent pattern of dissolution rate; in other words, calcite does not have a lower dissolution rate than aragonite when exposed to acidified seawater at pH 7.4 (McClintock et al. 2009).

The alteration of CO₂ in seawater has been shown to cause a reduction in calcification rates, induce malformations, and impair growth of shells (Riebesell et al. 2000, O'Donnel et al. 2009, Nienhuis et al. 2010, Findlay et al. 2010a and b, Gaylord et al. 2011, Pistevos et al. 2011). Coccolithophores exhibited a reduction in the mineral content of their shells by about 10 - 30% when exposed to low pH condition (Muller et al. 2010) and reduced calcification rates by 66% when exposed to a CO₂ level three times that of pre-industrial levels (Zondervan et al. 2001). Coralline algae also showed an increased dissolution rate in seawater enriched by CO₂ (pH 7.7) (Martin et al. 2008). The destruction of the protective outer shell layer (periostracum) in gastropods has also been found to occur in the top shell *Osilinus turbinata* exposed to pH 7.2 and in the black foot limpet *Pattela caerulea* exposed to pH 7.4 in the

volcanic CO₂ vents (Hall-Spencer et al. 2008). Pacific Oyster (*Crassostrea gigas*) and edible mussel (*Mytilus edulis*) also reduced calcification between 10 and 25% after incubation in a mesocosm for a month under CO₂ levels ranging between 700 to 2000 ppmv (Gazeau et al. 2007). It has also been found that reef building organisms reduce calcification between 11- 46% (Langdon 2002). However, calcification rate can also increase under extreme CO₂ conditions as demonstrated in the mussel *M. edulis* in the Kiel Fjord (Thomsen et al. 2010).

Effects of OA on traits other than calcified structures have also been found in calcifying organisms. Larvae of the mussel *M. californianus* showed lower tissue growth when it was exposed to CO₂ of 900 ppmv for 8 d (Gaylord et al. 2011). The brittle star *Amphiura filiformis* showed muscle degeneration after being exposed to pH 6.8 in 40 d (Wood et al. 2008). Even more exposure to pH 7.7 after 8 d caused 50% mortality of a brittle star larvae *Ophiothrix fragilis* and after 25 d caused 100% mortality (Dupont et al. 2008). However, adult individuals of the velvet swimming crab *Necora puber* reached 100% mortality within 4 – 5 d when exposed to 6040 ppm CO₂ (pH 7.1) as a result of the ability to compensate for the changes in haemolymph pH (Spicer et al. 2007), contrasting with adult *N. puber* that can survive 30 d exposure to pH 6.69 or 21500 ppmv CO₂ (Small et al. 2010). An indirect effect of OA on the common periwinkle *Littorina littorea* was also reported by Bibby et al (2007); when exposed to low pH conditions (pH 6.6 for 15 d) this species had its ability to exhibit induced shell defences (shell thickness) disrupted.

As well there being clear evidence that OA affects calcifying organisms, temperature has also been shown to affect the process of calcification in marine organisms and can cause dramatic changes in shell plasticity. For example, Trussell and Smith (2000) demonstrated that shell thickness of the snail *Littorina obtusata* was positively correlated with temperature. The phenotypic variation of shell morphology in different temperatures may be related to the chemical properties of the different calcium carbonate shell materials (aragonite and calcite). For example decreased calcium carbonate availability at low temperatures is caused by increased solubility that makes shell deposition more difficult (Graus 1974, Vermeij 1978, Trussell and Etter 2001, Melatunan et al. 2013). Irie and Fischer (2009) also reported that temperature influenced shell size in the cowry *Monetaria annulus*, with this species demonstrating a reduced size during warm season compared with the cold season. In contrast, shell thickness in *Cypraea annulus* increased linearly with increasing temperature (Irie 2006), and in the marine mussel *M. edulis* (Nielsen 1988) increased temperature resulted in increased shell size (length).

Given that increased temperature enhances calcium carbonate precipitation (Kleypas et al. 1999) the physiological mechanism for reduced calcification rates at low temperatures may be linked to reduced metabolic rates and energy production (Whiteley and Faulkner 2005). In addition, increase temperature can also create systemic hypoxia, leading to an increase in ventilation rate and reduce energy production (Pörtner 2001). An overall, increase in temperature may disrupt energy production and a concomitant increases in *p*CO₂ would lead to the disruption in oxygen transport proteins (Seibel and Walsh 2003), which in turn would disrupt calcification.

Impact of Ocean Acidification and Temperature on Ecosystems:-

As well as affecting single organisms, it is now clear that OA (and temperature) can have significant effects at higher ecological levels. For example, OA has serious negative effects on several fundamental biogeochemical and ecosystem processes including key elemental cycles and biodiversity (Widdicombe and Spicer 2008, Blackford 2010) and nutrient fluxes (Widdicombe and Needham 2007). These processes included decalcification of planktonic organisms, carbon and nutrient assimilation, primary production and acid-base balance, all of which potentially affect the composition, size structure and successional processes of ecosystems and may lead to a modification of energy flow and resources (Blackford 2010). Although Blackford and Gilbert (2007) suggested that the alteration of ecological function by OA is still unclear with the potential for populations acclimating to altered ocean carbonate chemistry, Hall-Spencer et al. (2008) have showed that a shallow water benthic community in the vicinity of natural CO₂ seepage might change due to the vulnerability of important groups of organisms, with no evidence for adaptation.

In addition, raised ocean temperatures (IPCC 2007, Sokolov et al 2009) may disrupt the stratification of the upper-ocean, the availability of nutrients for phytoplankton growth and reduce primary production (Behrenfeld et al. 2006). Several studies have also revealed that climate warming has reduced ocean productivity in the past decade with potential implications for marine food webs (Kleypas et al. 2006, Behrenfeld et al. 2006, Guinotte et al. 2006, Guinotte and Fabry 2008). In addition, climate change has been predicted to cause phenological changes (Koeller et al. 2009) and shifts in species ranges and distributional patterns (Easterling et al. 2000, Roy et al. 2001, Thomas et al. 2004, Perry et al. 2005, Sekercioglu et al. 2008).

Impacts of Ocean Acidification and Temperature on Physiological Functions:-

Several studies have revealed negative effects of climate change and OA on the physiological function of various marine calcifying taxa (e.g. Pörtner et al. 2000, 2004, 2005, Michaelidis et al. 2005, Wood et al. 2008, 2010, Small et al. 2010, Melatunan et al. 2011). For example increased levels of $p\text{CO}_2$ in seawater leads to hypercapnia that causes metabolic rate depression in *Sipunculus nudus* (Reipschläger and Pörtner 1996, Pörtner et al. 1998) and brings about 31% metabolic rate reduction in jumbo squid, *Dosidicus gigas* (Rosa and Seibel 2008) and 23% reduced in *L. littorea* (Melatunan et al. 2011). Increased CO_2 levels has also been shown to decrease aerobic scope (Metzger et al. 2007, Walther et al. 2010, 2011), increase induced acidosis in extracellular fluid (Burnett 1997, Miles et al. 2006, Pane and Barry 2007, Spicer et al. 2007). Metabolic acidosis can also reduce protein synthesis, increase respiratory stress and induce metabolic depression (Seibel and Walsh 2002) which may be lethal to organisms unable to compensate for haemolymph acidosis (Burnett 1997, Spicer et al. 2007). Such lethal effects of high $p\text{CO}_2$ were shown in *Necora puber* exposed to highest levels of hypercapnia at 6040 ppm (pH 6.74) (Spicer et al. 2007). Lannig et al. (2010) reported that OA affects energy metabolism in the oyster *Crassostrea gigas*. Under high CO_2 conditions a substantial reduction of ATP was found in gill tissue following an increase of succinate at the same gill tissue. In addition Cumming et al. (2011) demonstrated that high CO_2 (735 ppm equal to pH 7.78) caused low expression of chitin synthase (a key enzyme involved in synthesis of bivalve shells) indicating that the bivalve *Laternula elliptica* is working harder to calcify in acidified seawater conditions.

Rates of calcification under low pH are not only determined by environmental conditions (e.g. Ω_{ara} and Ω_{calc}) but are also determined by physiological capacity and functional performance in terms of the regulation of extracellular fluid acid-base balance and maintenance of metabolic homeostasis (Michaelidis et al. 2005, Cumming et al. 2011, Findlay et al. 2011, Gaylord et al. 2011, Whiteley 2011). The interaction between high $p\text{CO}_2$ and elevated temperature could lead to oxygen limitations (Pörtner and Farrell 2008, Pörtner 2010) which will add to the metabolic acidosis caused by lowering of the environmental pH which is known to disrupt oxygen transport and reduce energy production (Seibel and Walsh 2003). Such effects could be critical for intertidal organisms that may experience hypoxia during emersion – an effect that may occur concomitantly with high temperatures (Sokolova and Pörtner 2003, Larade and Storey 2009). Species populations are likely to have different physiological strategies e.g. shifting metabolic pathways and decreasing energy budget allocation (Sokolova and Pörtner 2001, 2003, Findlay et al. 2010a and b), that would allow them to cope with local environmental conditions such as thermal stress. Such metabolic and energetic shifts are likely to have associated energetic costs. For example, the alteration of metabolism under anaerobic scope leads to a reduction in energy production and mitochondrial density (Pörtner 2002). Under such conditions, time-limited, passive survival is supported by increased synthesis of heat shock proteins (Hsp) as a cellular defence mechanism (Feder and Hofmann 1999) although prolonged exposure is likely to cause lethal effects (Gehring and Wehner 1995).

Increased temperature has also been shown to affect metabolic rates and energy metabolism of marine gastropods (Sokolova and Pörtner 2001, 2003, Melatunan et al. 2011), cause cardiac failure and reduce thermal windows (Stillman 2003, Tomanek and Helmut 2002, Denny et al. 2006). Pörtner (2001, 2002) and Frederich and Pörtner (2000) demonstrated that increasing temperature caused increased ventilation and heart rates of aerobic capacity. Progressive increases in temperature may cause excessive oxygen demand and decrease aerobic capacity (Pörtner 2001). In addition, temperature can also affect biochemical reactions for physiological homeostasis (Hochachka and Somero 1973, Prosser and Nelson 1981, Hoffman and Dionne 1983). Initially, organisms could adjust to such temperature extremes by shifting their thermal tolerance or narrowing thermal windows via the adjustment of mitochondrial densities (Pörtner 2002). However, Pörtner (2002) also suggested that the crucial process in shifting thermal tolerance is that the organisms should face unidirectional (higher or lower) thermal conditions which may disrupt molecular function. As an alternative to coping with extreme temperature conditions, animals may also shift metabolic scope under anaerobic conditions or shift to hypometabolic physiology (Storey and Storey 2004). This mechanism requires shifting metabolic pathways and biochemical mechanisms for regulatory reversible transition to and from anaerobic physiology (Storey and Storey 2004, Hochachka and Somero 2005).

Pörtner and Farrell (2008) suggested that under the optimum temperature conditions (T_{opt}), aerobic performance was high but an increase or decrease in temperature beyond this optimum (T_{pej}) will lead to lowering aerobic performance. Further progress on to the critical temperature (T_{crit}) will lead to a loss in aerobic scope and a transition to an anaerobic mode of mitochondrial metabolism (Fig. 5).

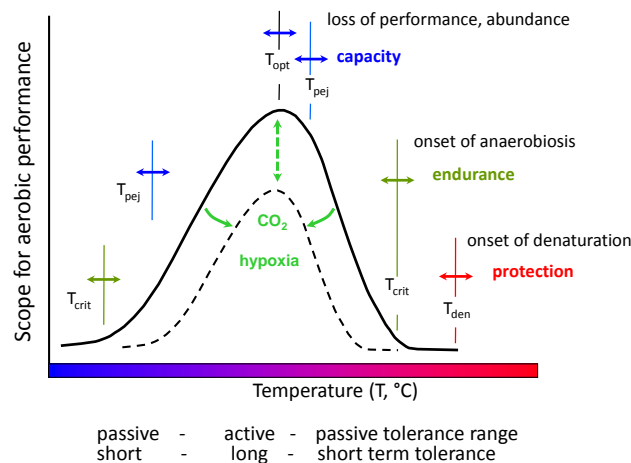


Figure 5:- A model indicating the hierarchies of functional limitation (beyond pejus temperatures, T_{pej}), hypoxemia, anaerobic metabolism and protection through metabolic depression (below and beyond critical temperatures, T_{crit}) and denaturation as well as reparation (beyond denaturation temperatures, T_{den}), (Source: Pörtner and Farrell 2008).

In this stage a progressive insufficiency of cellular energy levels occurs. At more extreme temperatures (T_{den}), only time limited passive survival is supported by anaerobic metabolism or the protection of molecular functions by heat shock proteins and antioxidative defense (Pörtner 2002, Pörtner and Farrell 2008). Pörtner and Knust (2007) have also suggested that increased temperature causes a mismatch between the demand for oxygen and the capacity of oxygen supply to tissues. Such a constraint could affect higher functions such as muscular activity, behavior, growth, and reproduction, ultimately leading to changes in species biogeography (abundance, occupancy, position of range edges and size of the geographical range of distribution).

It is highly likely that marine organisms will be subject to the combined effects of ocean acidification and temperature and recent studies have addressed these combined effects on physiological function in various organisms for example in jumbo squid *Dosidicus gigas* (Rosa and Seibel 2008), in the decapod *Metapenaeus joyneri* (Dissanayake and Ishimatsu 2011) and in brittle star *Ophiura ophiura* (Wood et al. 2010). Rosa and Seibel (2008) found that metabolic rate of jumbo squid *D. gigas* was depressed by 31% and activity levels by 45% under combined high CO_2 and temperature. Dissanayake and Ishimatsu (2011) demonstrated that in the decapod *M. joyneri*, reduced aerobic scope and swimming ability was reduced by 30% under high CO_2 (1000 ppm = pH 6.9) and temperature (20 °C). Even though Wood et al. (2010) found increased in brittle star *O. ophiura* under interaction of high CO_2 and temperature, there was an apparent trade off with increased arm degeneration. Also, increased metabolic rates in oysters *C. gigas* exposed to high CO_2 and temperature (25 °C) was associated with high accumulations of metabolic-end products (Lanning et al. 2010). Finally, Donohue et al. (2012) also found synergistic effects of OA and temperature on metabolic activity and heat tolerance in the intertidal crab *Porcellana platycheles*. These parameters were positively affected by temperature but exoskeleton calcification was negatively affected by high CO_2 of 2707 ppm. Clearly, more studies are needed if we are to understand more fully the synergistic effects of ocean acidification and warming on marine organisms.

Geographical variation in the impacts of Ocean Acidification and Climate Change:-

Impacts of ocean acidification are also suggested to vary across geographical and latitudinal boundaries (Walther et al. 2010, 2011, Findlay et al. 2010a, b). These differences may be linked to variations in interactive effects between CO_2 at each location. For example, annual sea surface temperatures (SSTs) and land surface temperatures (LSTs) in lower latitudes (southern ranges) are significantly different to those observed at higher latitudes (northern ranges) (e.g. Rastrick and Whiteley 2011, Whiteley et al. 2011 see also Whiteley et al. 1997). Such differences in responses to high CO_2 by organisms from different latitudes have been suggested in previous studies (e.g. Findlay et al. 2010a, b, Walther et al. 2010, 2011, Cumming et al. 2011). These studies are limited to comparisons between two populations.

Studies of the effects of elevated temperature caused by climate change in species populations and in different species at different latitudinal gradients have also been documented in various taxa (e.g. Sokolova and Pörtner 2001, 2003, Stillman 2002, 2003, Compton et al. 2007, Pörtner and Knust 2007, Rastrick and Whiteley 2011, Whiteley et al. 2011). Morphological variations in various taxa of calcified marine organisms were also found, for example, in the shallow water gastropod (Graus 1974), cowry the *Cypraea annulus* (Irie 2006), and in the zebra coral *Oulastrea crispata* (Chen et al. 2011). In general they found that morphological variations of these organisms were mainly caused by a change in CaCO_3 saturation state linked to temperature, however, causative factor such as salinity should also be considered.

Chemical effects of OA linked to carbonate saturation state (Ω) are likely to be linked to local seawater temperature regimes as the solubility of carbonate is temperature-dependent (Hill et al. 1999). Since the availability of carbonate depends on the saturation state of aragonite and calcite, which is temperature-dependent (see equations 2 and 3) (Kleypas et al. 1999, Azetsu-Scott et al. 2010), it does vary across latitudinal gradients (Bates et al. 2009, Yamamoto et al. 2009, Fabry et al. 2009, see also Riegl 2003 and Orr et al. 2005). Burton and Walter (1987) showed that the precipitation of aragonite increased rapidly with temperature. Aragonite precipitated four times faster than calcite at 25 and 37 °C respectively, whilst at 5 °C precipitation of aragonite decreased relative to calcite. This trend has also been shown in the ratio of calcite and aragonite in the shell of mussel *M. edulis*, which increased with decreasing water temperature along latitudinal gradient (Lowenstam 1954, Dodd 1963). Variation in biogenic calcification across latitudinal gradients also occurs with most calcified organisms in northern latitudes tending to be thinner shelled due to low temperature (Kuklinski and Taylor 2009, Trussell and Smith 2000, Findlay et al. 2010a). Carter (1980) revealed that such mineralogical differences may have functional consequences as calcite is softer and less strong than aragonite.

Increased dissolution rates may also vary along latitudinal gradients. For example Findlay et al. (2010a and b) showed that a population of the barnacle *S. balanoides* living at the northern range limit for this species was more sensitive to high CO_2 conditions, suggesting that the potential risk of climate change is likely to be greater in populations living at higher latitudes. Latitudinal gradients in pH, $[\text{CO}_3^{2-}]$ and Ω_{ara} and Ω_{calc} have been suggested to exist in both the Northern and Southern hemisphere (Orr et al 2005).

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RESEARCH ARTICLE

ESTIMATION OF HEART RATE USING SIGNAL FUSION OF ECG AND BP SIGNALS

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Abstract

The main aim of this paper is to estimate the heart rate and ECG pattern of a person as obtained from a polysomnographic record for detection of respiration related sleep disorders by fusing the data contained from the ECG and BP waveforms.. Polysomnography records the brain waves, the oxygen level in the blood, heart rate and breathing, as well as eye and leg movements during the study Data fusion can be defined as a combination of data from multiple sources to obtain improved information, meaning less expensive, higher quality, or more relevant information. For this study two specific signals from the polysomnogram is chosen- ECG and Blood pressure (BP). They are fused to obtain the improved heart rate of the subjects. Initially the individual heart rates are computed and compared against the heart rate of the fused signal. In case of noisy measurement environments, this method can be implemented.

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Introduction:-

Polysomnography is a sleep study. This test records certain body functions as we sleep, or try to sleep. Polysomnography is used to diagnose sleep disorders. A change in the cycle may make it hard for a person to sleep soundly. Multiple sleep latency test (MSLT) test measures how long it takes for us to fall asleep. It also determines whether you enter REM sleep [1]. Maintenance of wakefulness test (MWT) measures whether we can stay awake during a time when we are normally awake. If shift work sleep disorder or another problem with our body's internal clock (circadian rhythm) is suspected, then the test performed is called actigraphy.

Among the three main tests suggested above, polysomnography is perhaps the most elaborate and time consuming, due to the sheer amount of data collected and stored. But this is also the most valuable and reliable test in terms of the accuracy of sleep information obtained, provided they can be analyzed and interpreted in the clinically correct manner. Signal fusion involves the consolidation of all the various signals obtained in the polysomnogram in such a constructive manner so that it can be used constructively for diagnosis of sleep disorders.

Signal processing is an enabling technology that encompasses the fundamental theory, applications, algorithms, and implementations of processing or transferring information contained in many different physical, symbolic, or abstract formats broadly designated as signals. It uses mathematical, statistical, computational representations and techniques for representation, modeling, analysis, synthesis, discovery, recovery, sensing, acquisition, extraction, learning, security, or forensics. Dynamical mathematical model in this context is a mathematical description of the dynamic behavior of a system or process in either the time or frequency domain

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Biomedical signals are observations of physiological activities of organisms, ranging from genetic factor and protein sequences, to neural and cardiac rhythms, to tissue and organ images. Biomedical signal processing aims at extracting significant information from biomedical signals. With the help of biomedical signal processing, biologists can discover new methods of detection and physicians can monitor distinct illnesses. Initially the primary focus of biomedical signal processing was on filtering signals to remove noise. Sources of noise arise from imprecision of instruments to interference of power lines. Other sources are due to the biological systems themselves under study. The analysis framework derives its basics from statistical signal processing. This framework treats the data as random signals; the processing, e.g. Wiener filtering or Kalman filtering which utilize statistical characterizations of the signals to extract desired signal components.

Our bodies are constantly communicating information about our health. This information can be captured through physiological instruments that measure heart rate, blood pressure, oxygen saturation levels, blood glucose, nerve conduction, brain activity and so forth. Traditionally, such measurements are taken at specific points in time and noted on a patient's chart. Physicians actually see less than one percent of these values as they make their rounds—and treatment decisions are made based upon these readings.

Biomedical signal processing involves the analysis of these measurements to provide useful information upon which clinicians can make decisions. Engineers are discovering new ways to process these signals using a variety of mathematical formulae and algorithms. Working with traditional bio-measurement tools, the signals can be computed by software to provide physicians with real-time data and greater insights to aid in clinical assessments. By using more refined means to analyze what our bodies are saying, we can potentially determine the state of a patient's health through more noninvasive measures. Our proposed system works in this direction with further emphasis on processing the obtained data to a certain level so that it gives robust estimates of the patient's medical condition.

Sleep is defined as a reversible behavioral state, which involves a substantial form of disengagement and a decrease in the responsiveness to the environment. There are two types of sleep: Non rapid eye movement sleep (NREM) and rapid eye movement sleep (REM). NREM sleep is divided into four types based on the EEG signals. REM sleep is characterized by an increased activity in the EEG which could be a result of muscle atonia in the skeletal muscles or due to short episodic bursts of the REM sleep.

Polysomnography is used to diagnose sleep disorders. A change in the cycle may make it hard for a person to sleep soundly. Channels commonly recorded during a PSG are: Brain wave activity (EEG), Eye movement (EOG), Muscle tone (chin EMG), Airflow via thin catheters placed in front of nostrils and mouth, heart rhythm (EKG), Oxygen level (SpO₂), Leg muscle activity (PLM), Body position, Video recording. All the obtained information in this test can be used suitably for various purposes. This paper is one such application.

Materials and Methods:-

Estimation of respiratory rate [8] from waveforms recorded from passively breathing subjects is notoriously difficult, due in part to the non-stationary nature of the signals and in part the frequent non-stationary noise. Methods for recoding a time series of respiratory effort include impedance pneumography, impedance plethysmography and flow thermography which measures the changes in temperature of air flow as it moves in and out of the mouth and/or nose over a thermistor. Literature on deriving a respiratory signal from other related signals is dense in the case of the electrocardiogram, but relatively less for other signals such as the photo plethysmogram and blood pressure waveform [2]. The field of respiration rate estimation from respiratory signals (whether derived or not) is scantily covered in the public literature, particularly with respect to large data sets. Key works by O'Brien and Heneghan, de Chazal et al., Ishijima, Park et al., and Tarassenko [15] et al. highlights the importance of modeling noise and combining information from multiple ECG leads and sensor modalities to compensate for noisy measurements. To our knowledge, very little work has been public.

To evaluate our methods we have used polysomnographic data from a polysomnographic database obtained from www.physionet.org/cgi-bin/atm/ATM. It consists of the four most basic records namely- ECG, BP, EEG and respiratory rate. In this study we are concerned with the ECG [3] and BP [4] signals only. The records of five patients have been studied. The relation between the above mentioned signals is arrived at by the results of the correlation table presented below [5]. Out of the three signals, the most meaningfully correlated are the ECG and BP [6].

From these two signals the respective heart rates are obtained through a heart rate algorithm. This mainly counts the number of R peaks in an ECG. In the case of a BP signal [7], each peak corresponding to every cardiac cycle is counted. This provides the heart rate of the person. This procedure can also be followed for signals with noise. For a more realistic approximation we have added noise of different SNR values (10dB, 20dB, 30dB, 40dB and 50dB).

Table 1: Correlation between signals.

ECG	BP	EEG	RR
slp01	0.0736	0.1391	0.004
slp14	0.0983	0.1163	0.0098
slp37	-0.0962	0.3556	0.0237
slp41	0.1269	0.2456	-0.0134
slp59	0.0545	-0.0441	-0.0055
slp67x	0.0427	-0.2804	-0.0083

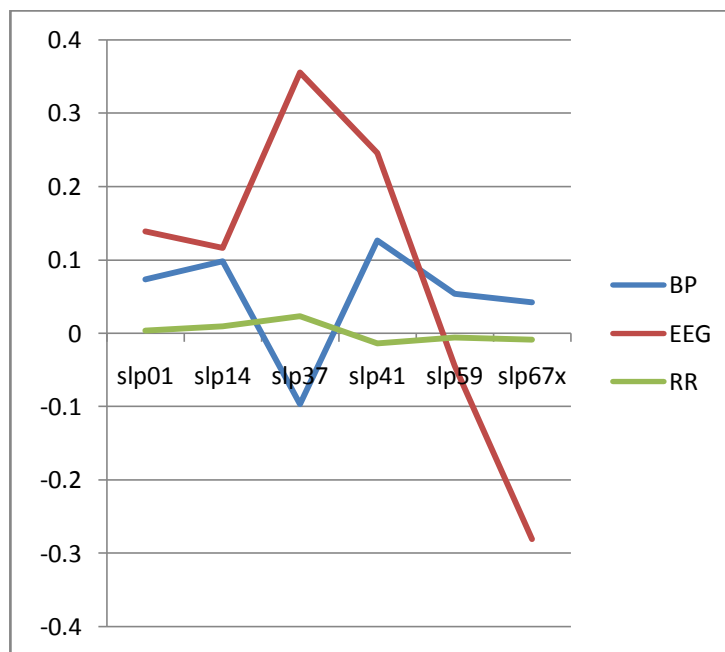


Fig 1: A plot of the correlation table

The main steps involved are presented in the block diagram given below. Both the required signals are obtained from the database. The signals in the database are clean and not corrupted by external noise. The heart rate for this is determined and this acts as the standard for comparison after fusion of the signals. The working data set is obtained by adding suitable noise to the signals. This noisy data is fused by the signal fusion algorithm and the output displayed is of a noiseless fused signal. The heart rate of this signal is computed by the beat detector algorithm and compared against the heart rate value of the reference signal [14].

The beat detection algorithm used is the Hooman Sedghamiz peak detector [9]. This algorithm requires the ECG as a signal vector along with the sampling frequency and the time period for the output window. As the output, it computes the indexes and amplitudes of each wave in the component ECG [8], the estimated heart rate and displays the processed signals. The function used is as given below

```
[R_i,R_amp,S_i,S_amp,T_i,T_amp,Q_i,Q_amp,heart_rate,buffer_plot]=peakdetect(ECG,fs,view)
```

Data fusion [10] is a multidisciplinary area that involves several fields, and it is difficult to establish a clear and strict classification. The fusion of these two signals is brought about by a fusion algorithm [11]. It is based on a kalman filter fusion that merges both the signals as one .this is further used for heart rate comparison after fusion.

The Kalman filter is the most popular estimation technique [9]. It was originally proposed by Kalman and has been widely studied and applied since then.

The Kalman filter estimates the state x of a discrete time process governed by the following space-time model:

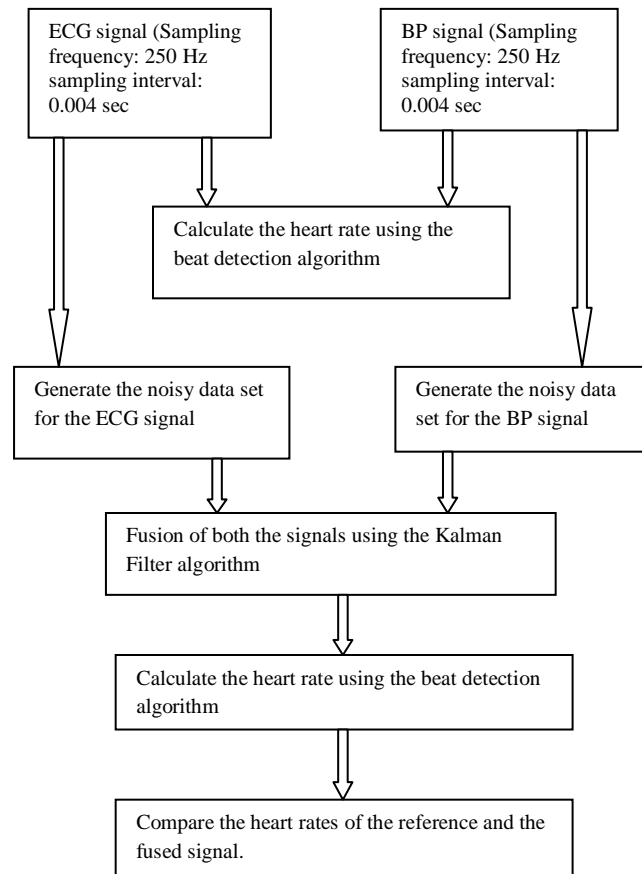


Fig 2: block diagram of the process

$$x(k+1) = \Phi(k)x(k) + G(k)u(k) + w(k) \quad (1)$$

With the observations or measurements z at time k of the state x represented by

$$Z(k) = H(k)x(k) + V(k) \quad (2)$$

Where $\Phi(k)$ is the state transition matrix, $G(k)$ is the input matrix transition, $u(k)$ is the input vector, $H(k)$ is the measurement matrix, and w and V are the random Gaussian variables with zero mean and covariance matrices of $Q(k)$ and $R(k)$, respectively.

The Kalman filter [12] is mainly employed to fuse low-level data. If the system could be described as a linear model and the error could be modeled as the Gaussian noise, then the recursive Kalman filter obtains optimal statistical estimations. However, other methods are required to address nonlinear dynamic models and non linear measurements .The filter as used in this method is given as:

For single signal:

$\hat{x} = \text{kalman}(z_1, A, C_1, R_1, Q);$

For two signals:

$\hat{x} = \text{kalman}([z_1; z_2], A, [C_1; C_2], [R_1 \ 0; 0 \ R_2], Q);$

Where:

\hat{x} = estimated signal

Z = input signal

A = state transition model

C = observation model

R = measurement noise covariance

Q = process noise covariance

Fig 3: A pure ECG signal

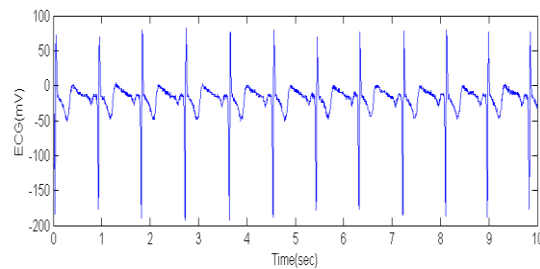


Fig 4: The same signal with a noise of 20 db added

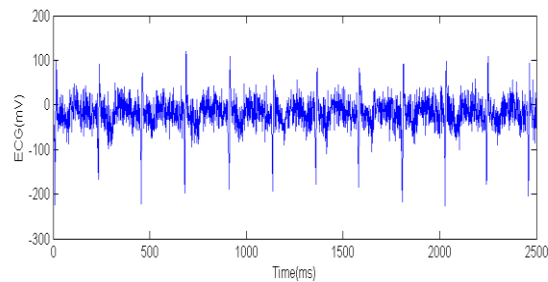


Fig 5: With 40db noise

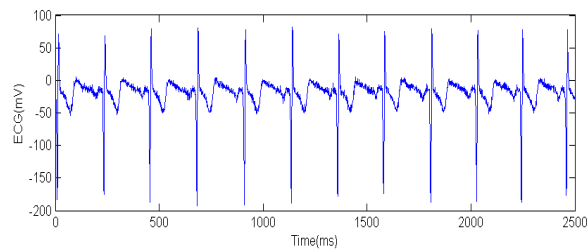


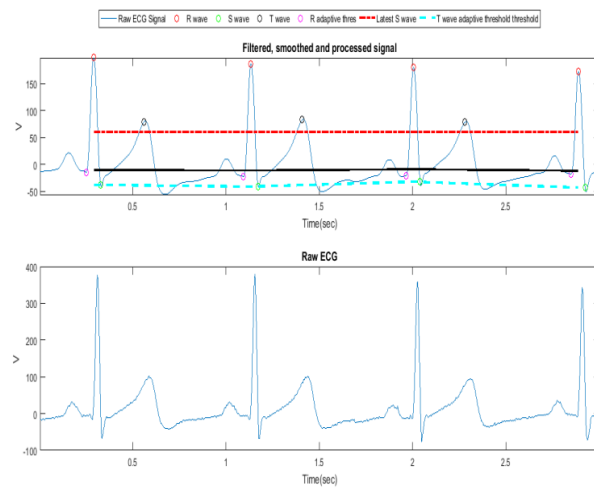
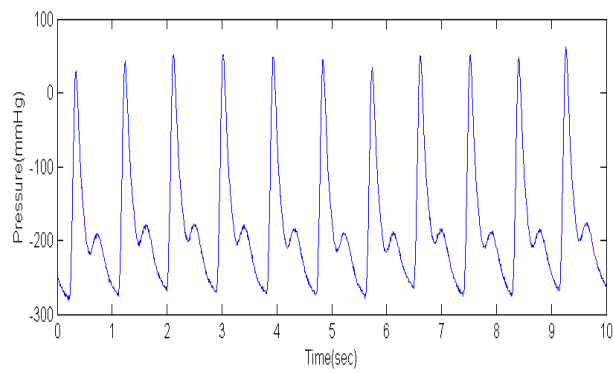
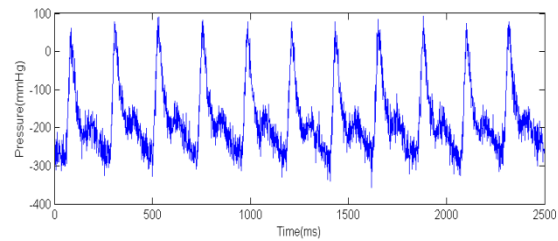
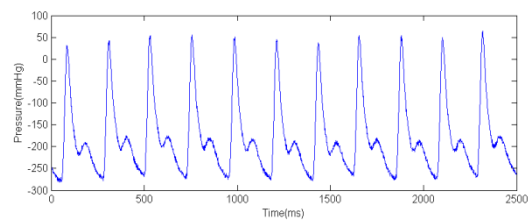
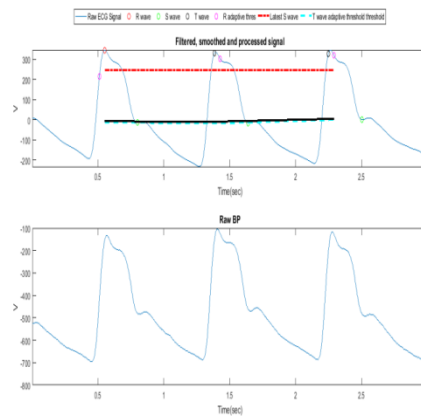
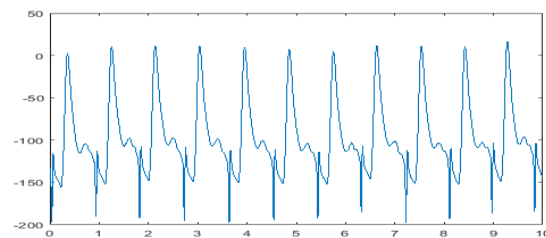
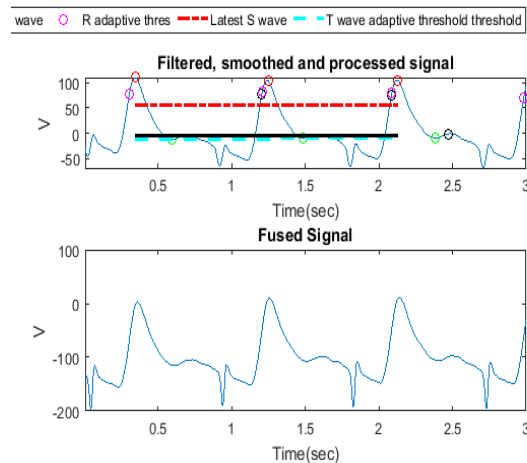
Fig 6: The heart rate obtained from the ECG signal**Fig 7: A pure BP signal****Fig 8: The same signal with 20db noise****Fig 9: With 40db noise**

Fig 10: The heart rate obtained from the BP signal**Fig 11: The fused output signal****Fig 12: The heart rate obtained from the fused signal**

Results and Discussion:-

Table 1 and Fig 1 shows the correlation of ECG signals with BP, EEG and RR signals separately. It is observed from the graph that there is a significant correlation between ECG and BP only. Though there is a wide variation in EEG, it cannot be chosen because brain waves are not only dependent on heart rate alone but have numerous parameters that affect it.

Fig 3 represents a pure ECG signal obtained from the sleep database. In order to process the signal as in a real time environment, noise was added represented by fig 4 and 5(SNR=20dB and 40dB respectively). Fig6 represents the heart rate obtained from the ECG signal.

Fig 7 represents a pure BP signal obtained from the sleep database. In order to process the signal as in a real time environment, noise was added represented by fig 8 and 9(SNR=20dB and 40dB respectively). Fig 10 represents the heart rate obtained from the BP signal.

Fig11 shows the output waveform of the fusion algorithm. The fusion algorithm estimates the combination of both the signals. Visually, it shows the merged form of the ECG and BP signals. Fig12 shows the heart rate of the fused signal in the processed form

Table 2: Comparison of HR between original and estimated signal.

Data/SNR	Actual HR from original ECG signal	10dB	20dB	30dB
		HR obtained by using Kalman fusion algorithm		
Slp01	66	66	66	66
Slp14	72	78	78	78
Slp37	60	66	66	66
Slp41	66	66	66	66
Slp59	78	84	84	84
Sp67x	72	72	72	72

Table 2 shows the outputs of our project that tabulates the improved heart rates of the various inputs considered. The standard or reference heart rate is the heart rate of the ECG alone. The tabulation shows the heart rates of the fused signals. It can be observed that in case of normal inputs; the estimated heart rate is the same as that for the reference signal. Only in the case of records with certain abnormalities, there is a difference.

Table 3: Comparison of error in estimating the HR values.

Data/SNR	Actual HR from original ECG signal	10dB	20dB	30dB
		% Error=(Original HR-Est HR)/Original HR *100 (obtained by using Kalman fusion algorithm)		
Slp01	66	0	0	0
Slp14	78	-7.69	-7.69	-7.69
Slp37	60	-10.01	-10.01	-10.01
Slp41	66	0	0	0
Slp59	78	-7.7	-7.7	-7.7
Sp67x	72	0	0	0

Table 3 displays the percentage error for all the signals considered. Percentage error is obtained as the ratio of the original heart rate to the difference between the original and the estimated heart rate. What we can infer from the table is that the maximum error is only 10%, which is quite a reasonable value considering the amount of noise added to the signals for evaluation purposes.

Conclusion:-

The main purpose of this paper is to obtain the heart rate from the fusion of ECG and BP signals. From the information obtained as the output of the peak detector for the fused signal the estimated heart rate is as shown in the above tables. This implies that fusion of signals gives the heart rate even on a noisy dataset. The added advantage of this approach is that the error value depends on the quality of the signals chosen for the study. In some cases the error is negligible or not present at all, which implies that this approach is headed in the right direction

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RESEARCH ARTICLE

PRENATAL DIAGNOSIS WITH GENETIC COUNSELING IN UHC SPLIT, CROATIA.

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 cytogenetic analysis, amniocentesis

Abstract

Purpose: The purpose of this study was to document the cytogenetic data obtained from amniotic fluid analysis, as a part of prenatal diagnosing.

Methods: 1.441 samples were analyzed by standard cytogenetic method.

Results: Indications for amniocentesis were divided into six groups. Of them advanced maternal age was the most common, and, as expected, it was associated with the largest number of pathological cytogenetic findings.

Conclusion: In comparison to other indicators for amniocentesis, advanced maternal age had a highest positive predictive value.

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Introduction:-

Prenatal diagnostics is a subdivision of clinical genetics; it enables early diagnosis of congenital anomalies and genetic disorders. This is very important because the population risk of having a child with some congenital abnormality varies between 3 and 5%. Although such malformations and genetic disorders can interfere with quality of the life of newborns and their families, sometimes they can also cause the spontaneous abortion.

There are varieties of methods that can be used for prenatal diagnostics. One of them, by which chromosomal abnormalities, neural tube defects and genetic disorders can be detected with high level of accuracy, is amniocentesis (AC). Although it is invasive test which carries a certain risk of miscarriage, it is strongly recommended (after genetic counseling) to a women at increased risk for chromosomal anomalies. So, AC in association with genetic counseling enables early diagnosis of congenital anomalies, which is essential for management the pregnancy and postnatal medical care. It is also crucial to making informed decisions about continuing or terminating the pregnancy. In a latter case it enables to counsel the couples in preparing for a next pregnancy (1, 2).

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The purpose of this study was to document the cytogenetic data obtained from amniotic fluid analysis, as a part of prenatal diagnosing.

Methods:-

During the period from 2007. to 2015. the amniotic fluid was collected from 1.441 pregnant women, who attended University Hospital Centre Split (UHC). All women were of European Caucasian origin. The analyses were done in the Department for Medical Genetics with Laboratory for Human Genetics and Genetic Counseling Unit, Paediatrics Clinic, UHC, Split, Croatia. Amniocentesis was performed between 13 and 25 weeks of gestation, with a peak at 17 week.

The study was approved by the Ethics Committee of the UHC Split. Informed consent to present the amniocentesis data was obtained from each couple.

Results:-

By the end of the march 2015. 1.441 amniocenteses and chromosomal analyses were carried out. Indications for amniocentesis were as follows: 1. maternal age, 2. family or personal history data (parental karyotype, syndrome Down in the family, previous child/children born with malformations, or spontaneous abortion or stillborn child with known or not known pathology), 3. results of prenatal tests (triple, double or combined test), 4. nuchal translucency (NT). 5. other fetal anomaly detected by ultrasound, 6. pregnant women demand.

Mother's age (>35) was the most frequent indicator for amniocentesis (1076; 74.67% cases). However, in 26 and 25 of those women indication for amniocentesis was combination of mothers age and anamnestic data and prenatal tests data, respectively. The second most frequent indicator for amniocentesis were the results of prenatal tests (156; 10.82% cases), followed by family or personal history data (129; 8.95 % cases) and fetal anomaly detected by ultrasound (56; 3.87% cases). Nuchal translucency was indication in 20 cases (1.39 %), while amniocentesis was performed on pregnant women demand in one case (0.07%) The majority of AC were conducted on woman between 35-40 years of age (49 %) and women older than 40 years (26 %) (data not shown).

The majority of samples were cytogenetically normal (1372 samples; 95.21%). In others (69 samples; 4.79%) some irregularities were found: balanced translocation (1), unbalanced translocation (2); aneuploidy (60), and mosaicism (6) (Table 1).

In majority of samples (66; 95.65 %) *de novo* chromosomal changes were present. In only three samples the chromosomal changes were either maternally or paternally inherited.

Maternal age was associated with the largest number of pathological findings (36 samples; 52.17%), followed by ultrasound findings (18 samples; 26.09%), results of prenatal tests (9 samples; 13.04%), NT (4 samples; 5.80%) and family or personal history data (2 samples; 2.90%). Sample analyzed on pregnant women demand turn out to be normal (Table 2).

Table 1. Cytogenetic results of amniotic fluid

	Number	Karyotype	
Amniocenteses total	1 441		
Normal	1372		No
Patologycal	69		
Translocation balance	1	46,XY,t(18;20)(18pter->20 q12)mat	No
-----	-----	-----	-----
--	-	46,XY,+21,rob(21;21)(q10;q10)	-
Translocation unbalanced	1	46,XY,add(12q)tr(8p;12q(8p12;12q24.3)(mat	Yes
	1)>parcial trisomy 8p	
Aneuploidy	60	45,X [5] 47,XXX [2] 47,XX+13 or 47,XY+13 [4] 47,XY+ 18 [10] 47,XX+21 or 47,XY+21 [35] 47,XXY [2]	Yes/No* Yes/No* Yes Yes Yes Yes/No*

		47,XY,+iso12p [1] 69,XXX [1]	Yes Yes
Mosaics	6	46,XX(22)/45,X(3) [1] 46,XX(18)/45,X(3) [1] 46,XX(50)/45,X(50) [1] 46,XX(10)/47,XXX(3) [1] 46,XX(8)/47,XXX(1) [1] 46,XY(10)/47,XXY(3) [1]	Yes/No* Yes/No* Yes/No* Yes/No* Yes/No* Yes

*TP (Termination of Pregnancy). In cases where changes in gonosomes were found, especially in mosaic form, the development of severe mental retardation is not expected, but the occurrence of sterility in the adulthood is possible, so the decision of TP should be done only by parents.

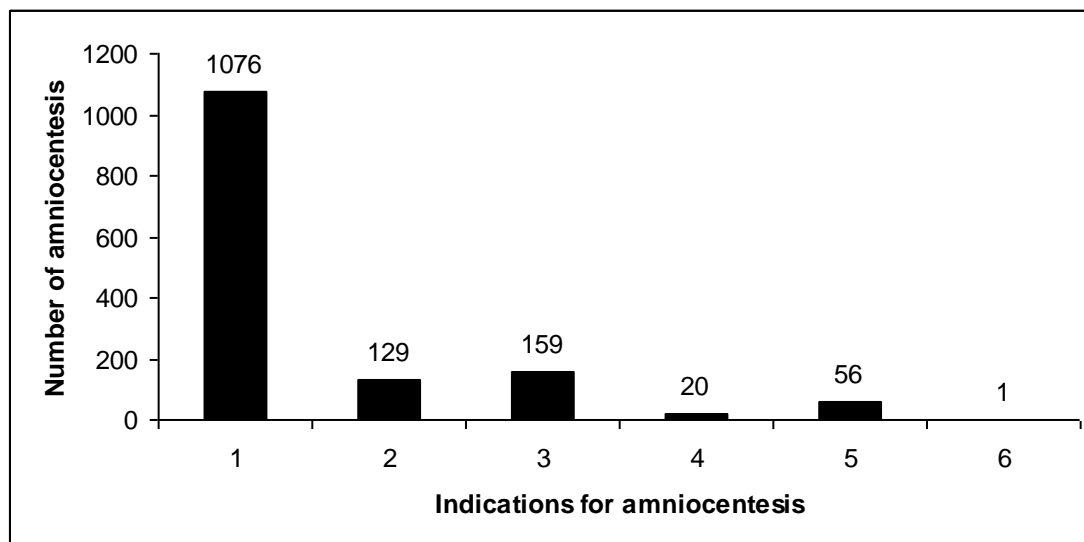


Figure 1. Indications for AC were divided into six groups: 1. maternal age, 2. family or personal history, 3. results of prenatal tests (triple, double or combined test), 4. nuchal translucency (NT), 5. fetal anomaly detected by ultrasound, 6. other (pregnant women demand). The numbers above the bars indicate total number of tested samples.

Table 2:- Number of pathological versus non-pathological findings in amniocentesis samples classified according to the indication for amniocentesis

	Non-pathological	Pathological			Mosaics	Total (pathological)
		Aneuploidy	Translocation balanced	Translocation unbalanced		
Maternal age	1040	31			5	36
Prenatal tests	150	8		1		9
Ultrasound	38	17	1			18
Nuchal translucency	16	3			1	4
Family history	127	1		1		2
Demend	1					0
Total	1372	60	1	2	6	69

Discussion:-

In thirty years (1983-2013) period in the Laboratory for Human Genetics and Genetic Counseling Unit more than 10,000 persons with different genetical as well as chromosomal problems were treated. These were children and adults with chromosomal abnormalities, sterile or infertile couples, couples with one or more spontaneous abortions, people with loads of personal or family history, people who were exposed to harmful factors, those with inherited neuromuscular diseases etc.

Special attention was also put on resolving possible cytogenetic cause of recurrent spontaneous abortions (RSA). By analyzing karyotypes of more than 350 couples (both women and men) who suffered from RSA, as well as karyotype of aborted material, some kind of abnormality was found in 17.6%, 11.5% and 25.9% of women, men, and aborted material, respectively. In a latter case, the majority of changes were *de novo* ones (3).

The methods of prenatal diagnostics can be divided into non-invasive (ultrasound and biochemical screening from maternal blood) and invasive ones (amniocentesis, chorionic villus sampling, cordocentesis, etc.) (4,5). In our study we paid attention to the women who were referred for amniocentesis. Indications for amniocentesis we divided into the six groups; similar divisions were also done in other studies (5,6). We found that the most prominent indicator for amniocentesis was the maternal age. This is in accordance with the findings of Yang et al (7), but in contrast to the finding of some other authors who found that the most prominent indicator for amniocentesis was an abnormal maternal serum-screening test or abnormal ultrasound findings (6,8,9).

In our study 4.55% women were pregnant with fetuses that had numerical chromosomal abnormalities. Some studies showed a similar results (4.61–4.85 %) (10,11), while some others showed lower incidence of chromosomal abnormalities (5,12).

The most common changes were autosomal trisomies. Similar to our results, Ocak et al. found that the most frequent numerical chromosomal abnormality was trisomy 21 (8).

After amniocentesis results are known, all women should be again subjected to genetic counseling. For those women where variation in population and balanced translocations were found we did not (we do not) counsel termination of pregnancy (TP). For the cases of gonosomes changes (yes/no), especially in mosaic form, decision about TP should be left to parents, because clinically, in the future, no development of severe mental retardation is expected, although the occurrence of sterility is possible. However, unbalanced translocations, mosaicism and aneuploidy were considered as indication for TP. For these women (couples) it is prerogative to carry out prenatal diagnostic during the next pregnancy as well as to repeat the genetic counseling (13).

In respect to the fact that invasive prenatal testing method bear certain risk of miscarriage, more and more women are choosing non-invasive prenatal testing methods such as analysis of circulating cell-free fetal DNA. Although detection of aneuploidy with this method is not 100% accurate, many couples, especially those with reassuring serum aneuploidy screening and normal ultrasound findings appear to favor a small risk of misdiagnosis of aneuploidy over the risk of procedure-related to pregnancy loss (14,15).

Conclusion:-

De novo chromosomal changes were most abundant in samples obtained from elderly woman. Therefore maternal age is the most prominent indicator for amniocentesis.

Disclosure Statement:-

The authors declare that they have no conflicts of interest.

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RESEARCH ARTICLE

PERSONAL HEALTH-CARE SYSTEM.

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Abstract

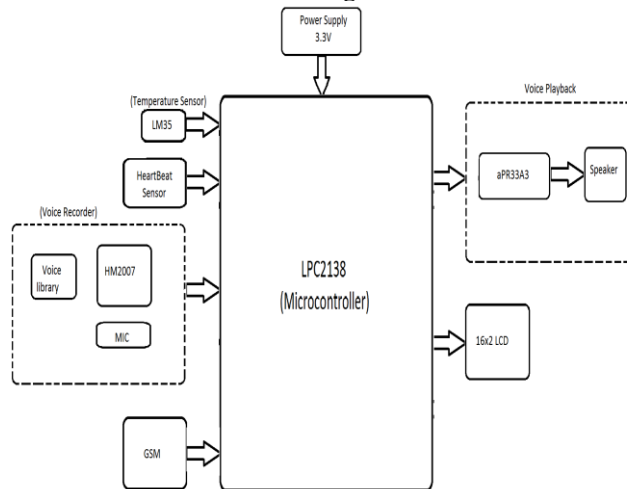
This document gives an overview about how “Personal Healthcare System” can extend a professional advice on what you might find useful to treat your illness. Following proper instructions and the medicine prescribed by the system may temporarily or permanently cure you. You may also consult your doctor if the illness persists or becomes more severe. “Personal Healthcare System” offers professional and free health advice at any time – you don’t need an appointment. From coughs and colds to aches and pains, it can give you expert help on everyday illnesses. Since there is no restriction for gender, you can talk in confidence. It may suggest you to visit your GP (General Practitioner) for more serious symptoms

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Introduction:-

The use of computers in health care evolved through a process driven by need. Over the last 30 years, health care has seen few real visionaries who have developed systems to aid in the clinical process. Only in the past five years has the emphasis shifted from applying computer technology to hospital administration, to applying technology to the clinical process. The use of computers in health care began to emerge in the early 1970s (through the use of shared systems) after Medicare reimbursement legislation was enacted, but it wasn’t until the late 1970s, when minicomputers began to become available, that computers began to be widely used in health care. Health care automation was quite limited for the first 10 years because of the insufficient file capacities of most systems.

The remote entry by patients of blood pressure, temperature, pulse, respiration, and conformance with treatment plans is already being accomplished through some home health systems. Innovation in clinical care would be sharply reduced were it not for the way the health care industry has flocked to the Internet. Probably no other technology since personal computers has gained such attention. Five years ago, the primary use of the Internet was for messages and promotional purposes. Today, the Internet has become an integral part of many vendors’ systems. Most important, from the perspective of clinical care, is the access to a myriad of disease and medical databases through the Internet. Such access provides medical data to even the most remote parts of the world. In fact, best-treatment plans that were developed in advanced countries can be made available to physicians in even the least-advanced countries over the Internet.

Block Diagram:-**Robots in Medical History:-**

Robots serving various tasks and purposes in the medical/health and social care sectors beyond the traditional scope of surgical and rehabilitation robots are poised to become one of the most important technological innovations of the 21st century. Traditional medical robots have focused mainly on highly specialized platforms for surgery or rehabilitation and low levels of autonomy, relying on tele-operation and/or the presence of qualified staff to enable and ensure appropriate conditions and use. A novel approach along these traditional lines is the magnetic microbots for vascular network intervention. A recent trend has been to move away from costly, task-specific platforms supporting well-defined medical, commonly surgical tasks, towards cheaper more generic platforms. These robots can commonly support a number of different and —softer human-robot interaction (HRI) tasks and have been used to improve the medical conditions of patients.

Robots have been applied in multiple ways to assist the logistics of health and social care. Non-tactile HRI has also been studied extensively in the health and social care domain. Verbal and gesture-based interaction provides opportunities for robots to support patients in two main ways. First, a robot can act as a conduit for socializing, enabling friends or family to engage with the user remotely, or for (remote) communication with a health professional, allowing professionals to reach a larger number of patients. Second, a robot can present itself as an autonomous entity, providing play scenarios that can reduce the effect of disabilities, or improving wellbeing through entertainment and companionship. The increasing numbers of people with long-term illness and an ageing population are putting an unsustainable demand on the already resource-constrained hospitals and healthcare/social care systems in developed countries. Tele-healthcare, home care and self-care have long been encouraged and pushed as strategic policies and directions in these countries, as the way forward for mitigating the increasing demands and burdens on conventional health/care and social care services. The goal is to shift part of the care burden from hospitals/clinics and healthcare professionals to patients and their informal care-givers, and to help individuals take responsibility of, and better manage, their own health.

Components Required:-

A) Hardware	B) Software
<ul style="list-style-type: none"> • LPC2138(Micro-controller) • HM2007p • LM35 • Heart-Beat Sensor • MAX232 • GSM • LCD (16x2) • aPR33A3(Voice Processor) 	<ul style="list-style-type: none"> • Orcad Capture • Express PCB • μVision IDE

Table I:- Components Used for Personal Health-care System**C) Working Principle:**

LM35 (temperature sensor) and heartbeat sensor are used to monitor physical parameters. Two modes of input are provided viz., voice and text modes. HM2007p is the voice processor used for learning and storing real-time voice inputs. APR33a3 is a voice playback used to give voice output while the LCD displays the same. GSM module receives inputs and reverts the remedy through SMS. LPC2138 is a 32-bit microcontroller which is responsible for all controlling actions.

LPC2138 has 32-bit timers, single or dual 10-bit 8-channel ADC(s), 10-bit DAC, PWM channels and 47 GPIO lines with up to nine edge or level sensitive external interrupt pins make these microcontrollers particularly suitable for industrial control and medical systems.

Advantages and Disadvantages:-**Advantages:-**

1. System has a low operating cost with one-time investment and the time of purchase. Its costing would make it affordable to every strata of the society.
2. Information regarding patients' ailments and treatment methods, and even emergency services can be received anywhere and anytime, regardless of the location. This makes it useful in remote areas lacking medical facilities for diagnosis and immediate treatment procedures.
3. A system that can be remotely accessed by user, even if the user isn't in the vicinity of the system. This makes it advantageous in any type of setting (domestic, industrial etc.).
4. No need of consulting a doctor for minor ailments which can be cured easily with the intake of proper medication.
5. Prognosis on par with professionals and first-aid for all kinds of problems provided.
6. The system can minimize medical errors which can occur due to human error or negligence and ensures safety of the user.

Disadvantages:-

1. Cannot provide treatment methods for major ailments that are life threatening or for which professional assistance is mandatory.
2. Flexibility of medication is restricted due to pre-fed database and hence cannot provide treatments to ailments that are not available in database.
3. Database has to be updated regularly to keep the system in touch with the updates and progress in medicine.

Application:-

1. A personal healthcare unit to provide a complete first-aid solution to user
2. Provides prognosis based on the input given by user
3. Healthcare companion to provide better treatment procedure based on user's medical history
4. Professional advice on par with that of doctors
5. Used in domestic and industrial medical systems for immediate first aid solutions.
6. Can be used as a helper at various critical stages in case of emergencies like stroke etc.

Future scope:-

- The system can be further modified to append an expansion to store the medical history of one or more patients. The database can furthermore be updated to include new treatment methods for existing ailments by doctors.
- The system can comprise of e-nursing and care-bots in addition to treatments.
- The system can be directly affiliated to medical institutions in case of requirement of professional help.
- The system can be made to directly contact nearest hospital in case of extreme emergencies.

Conclusion:-

The aim of this system is to provide professional medical advice for minor ailments and diseases for which a patient would not require to go to a doctor for its treatment. This would be beneficial in remote areas where medical help is sparse and not well facilitated for medical institutions. It would provide first-aid measures for proper treatment of injuries and minor accidents. Also in case of emergencies, the system can provide proper treatment method instead of waiting for emergency services for a solution.

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RESEARCH ARTICLE

SOME INVERSE AND SATURATION RESULTS ON CONVOLUTION OPERATORS

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Abstract

In this paper, firstly we have studied some inverse and saturation results for the family of linear positive convolution operators. We have used Bernstein inequality for proving inverse theorems. Then we have found some linear combinations which are not saturated by construction.

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Introduction:-

Consider a family of linear positive convolution operators [1],

$$(E_{(n,\eta)}f)(x) = (f * g_{(n)})(x) = \frac{1}{\pi} \int_{-\pi}^{\pi} f(t)g_{(n)}(x-t)dt, \quad n \in \mathbb{N} \text{ and } x \in \mathbb{R} \quad (1)$$

where $C_{2\pi}$ is the space of 2π -periodic functions with norm,

$$\|f\| := \max |f(x)|$$

with kernel $\eta = (g_n(x))_{n>0} \subset L^1_{2\pi}$

depending upon the parameters $n > 0$ and $n \rightarrow \infty$.

Here, kernel $\eta = \{g_{(n)}\}_{n=1}^{\infty}$ be a sequence of even trigonometric polynomials of degree at most $m(n) = O(n)$, which are normalized by,

$$\frac{1}{\pi} \int_{-\pi}^{\pi} g_{(n)}(t)dt = 1$$

$$g_{(n)}(x) = \frac{1}{2} + \sum_{k=1}^{m(n)} \rho_{(k,n)} \cos kx \quad (2)$$

Here, the operators are uniformly bounded,

$$\|(E_{(n,\eta)}f)\| \leq A\|f\| \quad (3)$$

and satisfy Bernstein type inequality,

$$\|(E_{(n,\eta)}f)'\| \leq A\varphi(n)^2 \|(E_{(n,\eta)}f)\| \quad (4)$$

Also, we have,

$$(E_{(n,\eta)}f)'(x) = (E_{(n,\eta)}f'(x)) \quad (5)$$

For all f with $\varphi(n) > 0$ monotonely increasing to infinity such that,

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$$\sup_{n > 0} \left\{ \frac{\varphi(n+1)}{\varphi(n)} \right\} = k < \infty$$

For $\delta > 0, 0 < \alpha \leq 2$, we have from [2],

$$\omega_2(f; \delta) = \sup_{0 < h \leq \delta} \|f(x+h) - 2f(x) + f(x-h)\|$$

$$= \sup_{0 < h \leq \delta} \|\Delta_h^2 f(x)\|$$

$$Lip_2 \alpha = \{f \in C_{2\pi}; \omega_2(f; \delta) = O(\delta^\alpha), \delta \rightarrow 0^+\}$$

By the monotonicity of the modulus of continuity,

$$\omega_2(f; h) \leq At^\alpha + Ah^2 t^{-2} \omega_2(f; t) \quad (6)$$

Inverse Results:-

Lemma 2.1. Let Ω be monotonically increasing on $[0, c]$. Then,
 $\Omega(t) = O(t^\alpha)$, $t \rightarrow 0^+$, if for some $0 < \alpha < r$ and all $h, t \in [0, c]$
 And $m \in N$, such that ,
 $h_m \leq t \leq h_{m-1}$
 And
 $\Omega(t) \leq \Omega(h_{m-1}) \leq Bh_{m-1}^\alpha = B(Mh_m)^\alpha \leq B(Mt)^\alpha$

Introducing the Steklov means for $\delta > 0$,

$$f_\delta(x) = \frac{1}{\delta^2} \int_{-\delta/2}^{\delta/2} \int_{-\delta/2}^{\delta/2} f(x+s+t) ds dt$$

Also, we have from [3],

$$\|f - f_\delta\| \leq \omega_2(f; \delta) \quad (7)$$

$$\|f_\delta''\| \leq \delta^{-2} \omega_2(f; \delta) \quad (8)$$

Theorem 2.1. [4] If $0 < \alpha < 2$, then we have,

$$\|E_{(n,\eta)} f - f\| \leq A\varphi(n)^{-\alpha}$$

This implies that, $f \in Lip_2 \alpha$

Proof. By the assumption and using (3), (4), (5), (7) and (8), we have for $h > 0$,

$$\begin{aligned} \|\Delta_h^2 f\| &\leq \|\Delta_h^2 (f - E_{(n,\eta)} f)\| + \int_{-h/2}^{h/2} \int_{-h/2}^{h/2} (E_{(n,\eta)} f)''(x+s+t) ds dt \\ &\leq 4\|f - E_{(n,\eta)} f\| + h^2 \left\{ \left\| [E_{(n,\eta)} (f - f_\delta)]'' \right\| + \|E_{(n,\eta)} f_\delta''\| \right\} \\ &\leq 4A\varphi(n)^{-\alpha} + Ah^2 \{ \varphi(n)^2 \|f - f_\delta\| + \|f_\delta''\| \} \\ &\leq 4A\varphi(n)^{-\alpha} + Ah^2 \left\{ \varphi(n)^2 + \frac{1}{\delta^2} \right\} \omega_2(f; \delta) \\ &\leq A\delta(n)^\alpha + A \left(\frac{h}{\delta(n)} \right)^2 \omega_2(f; \delta(n)) \end{aligned}$$

for $\delta = \delta(n) = \varphi(n)^{-1}$

If we choose, $\delta(n) \leq t \leq \delta(n-1) \leq k\delta(n)$,

then, $f \in Lip_2 \alpha$.

Theorem 2.2. [5] For $0 < \alpha < 2$, we have,

$$E_{(n,\eta)}^* f = O\left(\frac{1}{n^\alpha}\right), \text{ then, } f \in Lip_2 \alpha$$

where, for the polynomial $e_{(n,\eta)}^* f$ of best approximation, $E_{(n,\eta)}^* f$ is given by,

$$E_{(n,\eta)}^* f = \inf_{e_{(n,\eta)} \in \pi_n} \|f - e_{(n,\eta)} f\| = \|f - e_{(n,\eta)}^* f\|$$

π_n being the set of complex trigonometric polynomials of degree n .

Proof. Let $J_{(n,\eta)}$, for $n \in N$, be a sequence of convolution operators (1) satisfying (2),

$$J_{(n,\eta)}f \in \pi_n,$$

$$\text{and } \|J_{(n,\eta)}f - f\| \leq A\omega_2\left(f; \frac{1}{n}\right) \quad (9)$$

using Bernstein inequality for trigonometric polynomials,

$$\|e_{(n,\eta)}^*f\| \leq n^2\|e_{(n,\eta)}f\|$$

We have,

$$\begin{aligned} \|(J_{(n,\eta)}f)''\| &\leq \|J_{(n,\eta)}(f - f_{n-1})''\| + \|J_{(n,\eta)}f_{n-1}''\| \\ &\leq n^2\|J_{(n,\eta)}(f - f_{n-1})\| + A\|f_{n-1}''\| \\ &\leq An^2\omega_2\left(f; \frac{1}{n}\right) \quad (10) \end{aligned}$$

Using (9), (10) and theorem 2.1, we have,

$$\begin{aligned} \|\Delta_h^2 f\| &\leq \|\Delta_h^2(f - (e_{(n,\eta)}^*f))\| + \int_{-h/2}^{h/2} \|(e_{(n,\eta)}^*f)''(x+s+t)\| ds dt \\ &\leq 4\|E_{(n,\eta)}^*f\| + h^2 \|[e_{(n,\eta)}^*(f - J_{(n,\eta)}f)]''\| + h^2 \|[J_{(n,\eta)}f]''\| \\ &\leq 4A\left(\frac{1}{n^\alpha}\right) + n^2h^2\|e_{(n,\eta)}^*(f - J_{(n,\eta)}f)\| + n^2h^2A\omega_2\left(f; \frac{1}{n}\right) \\ &\leq A\left[\frac{1}{n^\alpha} + n^2h^2A\omega_2\left(f; \frac{1}{n}\right)\right] \\ &= A\delta_n^\alpha + h\delta_n^{-2}\omega_2(f; \delta_n) \end{aligned}$$

with $\delta_n = (1/n)$

Now, $\frac{\delta_n}{\delta_{n+1}} \leq 2$, and using (5), we have, $f \in Lip_2\alpha$.

Some Definitions:-

Definition 3.1.

Fejer type kernels.

Let $q \in L^1(R)$ be normalized by,

$$\int_{-\infty}^{\infty} q(t)dt = 2\pi \quad (11)$$

Now, for (2), we will consider even q with its fourier transform \hat{q} with compact support,

$$\hat{q}(v) = \frac{1}{2\pi} \int_{-\infty}^{\infty} q(t)e^{-ivt}dt = 0, \quad |v| > T, \quad (12)$$

for some $T > 0$.

Then $q^* = \{q_n^*\}_{n \in N}$ with,

$$q_n^*(x) = \frac{1}{2} \sum_{k=-\infty}^{\infty} nq[nx + 2nk\pi] \quad (13)$$

is called a kernel of Fejer type.

Closed representation is given by,

$$q_n^*(x) = \frac{1}{2} + \sum_{k=1}^{[nT]} \hat{q}kn^{-1} \cos kx, \quad x \in R, \quad (14)$$

Now with the Poisson formula [6], the singular convolution integral (1) with kernel q may be represented as convolution integral on real line as,

$$(E_{(n,q^*)}f)(x) = \frac{1}{\pi} \int_{-\pi}^{\pi} f(t)q_n^*(x-t)dt = (E_{(n,\eta)}f)(x) = \frac{1}{2\pi} \int_{-\infty}^{\infty} f(t)q_n\left(x - \frac{t}{n}\right)dt \quad (15)$$

The above condition placed on q guarantee that $(E_{(n,q^*)}f)(x)$ defines an approximation process on $C_{2\pi}$.

Definition 3.2.

Jackson Kernel. The positive Jackson kernels $j^{(p)} = \{J_{(n)}^{(p)}\}_{n \in N}$ are given by,

$$J_{(n)}^{(p)} = \frac{1}{\lambda_{(0,n)}(p)} \left\{ \frac{\sin(nx/2)}{\sin(x/2)} \right\}^{2p}, \quad \text{where, } x \in R \quad (16)$$

This is the closed representation of Jackson kernels.

Here, $\lambda_{(k,n)}(p) = \frac{2}{\pi} \int_0^\pi \left\{ \frac{\sin(nt/2)}{\sin(t/2)} \right\}^{2p} \cos kt \, dt$, where, $0 \leq k \leq (n-1)p$ (17)

Corresponding convergence factors are given by,

$$\rho_{(k,n)}(j^{(p)}) = \frac{\lambda_{(k,n)}(p)}{\lambda_{(0,n)}(p)}, \quad (18)$$

Where [1],

$$\rho_{(k,n)} = \begin{cases} (1/\pi) \int_{-\pi}^\pi g_{(n)}(t) \cos kt \, dt, & 0 \leq k \leq m(n) \\ 0, & k > m(n) \end{cases} \quad (19)$$

Here, kernel $\{g_{(n)}\}_{n=1}^\infty = \eta$ be a sequence of even trigonometric polynomials of degree atmost $m(n) = O(n)$, which are normalized by,

$$\frac{1}{\pi} \int_{-\pi}^\pi g_{(n)}(t) dt = 1$$

The Jackson kernels are not of Fejer type.

Also, $\lambda_{(k,n)}(p)$ can be represented as in [7] [8],

$$\lambda_{(k,n)}(p) = \sum_{j=0}^{2p} (-1)^j \binom{2p}{j} \binom{n(p-j) + p - k - 1}{2p-1}, \quad 0 \leq k \leq (n-1)p \quad (20)$$

Using property of central factorial numbers ,

$$\begin{aligned} \text{(i)} \quad & t_k^n = T_k^n = 0, \quad n < k \\ \text{(ii)} \quad & T_k^n = \frac{1}{k!} \sum_{j=0}^k (-1)^j \binom{k}{j} \left(\frac{k-2j}{2} \right)^n, \quad 0 \leq k \leq n \in N_0 \end{aligned} \quad (21)$$

Where, t_k^n is the central factorial numbers of first kind and is uniquely determined coefficients of the polynomials, $x^{[n]} = \sum_{k=0}^n t_k^n x^k$

Similarly, T_k^n is central factorial numbers of second kind and is uniquely determined coefficients of the polynomials, $x^n = \sum_{k=0}^n T_k^n x^{[k]}$

By putting $k = 0$ in (20) and using (21),

$$\lambda_{(0,n)}(p) = \frac{1}{(2p-1)!} \sum_{i=1}^p n^{2i-1} t_{2i}^{2p} \sum_{j=0}^{2p} (-1)^j \binom{2p}{j} (p-j)^{2i-1} \quad (22)$$

and for $1 \leq k \leq n-p$, we have,

$$\begin{aligned} \lambda_{(k,n)}(p) &= \frac{1}{(2p-1)!} \sum_{i=2}^p t_{2i}^{2p} \sum_{m=1}^{i-1} \binom{2i-m}{2m} n^{2i-2m-1} k^{2m} \\ &\times \sum_{j=0}^{2p} (-1)^j \binom{2p}{j} (p-j)^{2i-2m-1} \\ &+ \frac{(-1)^p}{(2p-1)!} \binom{2p}{p} \sum_{i=1}^p k^{2i-1} t_{2i}^{2p} + \lambda_{(0,n)}(p) \end{aligned} \quad (23)$$

For some, $C_{(i,j)} = C_{(i,j)}(j^{(p)})$ and $d_{(k,p)} \neq 0$, polynomial division of (23) by (22) gives,

$$1 - \rho_{(k,n)}(j^{(p)}) = \sum_{j=1}^{p-1} \left(\frac{1}{n^{2j}} \right) \sum_{i=1}^j C_{(i,j)} k^{2i} + d_{(k,p)} n^{-2p+1} + O(n^{-2p}) \quad (24)$$

We can see from (24),

$$j^{(p)} \in S^{(2,p-1)} \text{ and } j^{(p)} \notin S^{(2,p)}$$

For, $p \geq 3$, we have approximation rate higher than $O(n^{-2})$ for the linear combination $\chi = \{\chi_{(n)}\}_{n \in N}$ of even trigonometric polynomials of degree (na_v) , for Jackson kernels,

$$\chi_n(x) = \sum_{v=1}^s \gamma_v g_{(na_v)}(x), \quad x \in R, \quad (25)$$

Definition 3.3.

Central B-splines and Jackson De La Vallee Poussin kernel.

We can define central B-splines [9] as the Fourier transforms of the powers of the sinc function,

$$B_m(v) = \frac{1}{2\pi} \int_{-\infty}^\infty \left(\text{sinc} \frac{t}{2} \right)^m \cos v t dt \quad (26)$$

$$\operatorname{sinc} x = \frac{\sin x}{x}, \quad x \in \mathbb{R} \setminus \{0\}, \quad \operatorname{sinc} 0 = 1$$

Their closed form is given by,

$$B_m(v) = \begin{cases} \frac{1}{(m-1)!} \sum_{j=0}^{[(m/2)-|v|]} (-1)^j \binom{m}{j} \left(\frac{m}{2} - |v| - j\right)^{m-1}, & |v| \leq \frac{m}{2} \\ 0, & |v| > \frac{m}{2} \end{cases} \quad (27)$$

The main properties of B-Splines are,

$$B_m(v) \geq 0, \quad v \in \mathbb{R},$$

$$B_m(v) \in C^{m-2}(\mathbb{R}), \quad m \geq 2,$$

$$B_m(v) \in C^{m-1} \left[-\frac{m}{2} + i, -\frac{m}{2} + i + 1 \right], \quad 0 \leq i \leq (m-1), \quad i \in \mathbb{N} \quad (28)$$

The function, q is given by,

$$q(x) \equiv q_{(n)}(x) = \frac{1}{\left\{ B_{2p}(0) \left(\operatorname{sinc} \frac{x}{2} \right)^{2p} \right\}} \quad (29)$$

satisfies (11) and (12).

For $p \in \mathbb{N}$, the corresponding Fejer type kernel (13), namely, the kernel of Jackson and de la vallee Poussin $\hat{p}^{(p)} = \{P_{(n)}^{(p)}\}$, $n \in \mathbb{N}$, is given by, according to (2),

$$P_{(n)}^{(p)}(x) = \frac{1}{2} + \sum_{k=1}^{np-1} \frac{B_{2p}(k/n)}{B_{2p}(0)} \cos kx, \quad x \in \mathbb{R}, \quad (30)$$

For $p = 1$, we have well known Fejer kernel $f = \{F_n\}$, $n \in \mathbb{N}$ with,

$$F_n(x) = \frac{1}{2\pi} \left[\frac{\sin(nx/2)}{\sin(x/2)} \right]^2, \quad x \in \mathbb{R}, \quad (31)$$

and convergence factors,

$$\rho_{(k,n)}(f) = \frac{n-k}{n}, \quad 0 \leq k \leq (n-1), \quad (32)$$

For $p = 2$, we have classical kernel of Jackson de La Vallee Poussin,

$$P_{(n)}^{(2)}(x) = \left(\frac{1}{2n^3} + \frac{\cos x}{4n^3} \right) \left[\frac{\sin(nx/2)}{\sin(x/2)} \right]^4, \quad x \in \mathbb{R}, \quad (33)$$

Convergence factors for above kernel is,

$$\rho_{(k,n)}(\hat{p}^2) = \begin{cases} (4n^3 - 6nk^2 + 3k^3)/4n^3, & 0 \leq k \leq (n-1) \\ (2n-k)^3/4n^3, & 0 \leq k \leq (2n-1) \end{cases} \quad (34)$$

Saturation Results:-

Here, we will study saturation theorems that are not saturated.

Consider the best trigonometric approximation for $f \in C_{2\pi}$,

$$(I_{(n)}f)(x) = I_{(n)}f = \inf_{t_n \in \pi_n} \|f - t_n\| = \|f - t_n^*\| \quad (35)$$

For convergence factors of a kernel η , there holds,

$$\rho_{(k,n)}(\eta) = 1, \quad 1 \leq k \leq n, \quad (36)$$

We have, $E_{(n,\eta)}(t_n, x) = t_n(x)$, $t_n \in \pi_n$, (37)

$$\|E_{(n,\eta)}(f, \cdot) - f(\cdot)\| \leq \|E_{(n,\eta)}(f - t_n^*, \cdot)\| + \|f - t_n^*\|$$

$$\|E_{(n,\eta)}(f, \cdot) - f(\cdot)\| \leq I_{(n,\eta)} \|f - t_n^*\| + \|f - t_n^*\| \quad (38)$$

For example, the well-known linear combination, $\{\tau_n\}$, $n \in \mathbb{N}$, of the Fejer kernel f ,

$$\tau_n(x) = 2F_{2n-1}(x) - F_{n-1}(x) \quad (39)$$

satisfies (36), so that, we have,

$$\|E_{(n,\tau)}(f, \cdot) - f(\cdot)\| = O(1) (I_{(n)}f), \text{ for, } f \in C_{2\pi}$$

We can generalize above method in the following way.

Suppose the convergence factor of η admits an expansion for $\tau = 1$ or 2 , $\mu \in N$,

$$1 - \rho_{(k,n)}(\eta) = \sum_{j=1}^{\mu} (-1)^{j+1} \psi_j(k) \left(\frac{1}{n^{\tau j}}\right) + h_{(k,\mu)} \left(\frac{1}{n^{\tau \mu + (\tau/2)}}\right), \quad (40)$$

For $h_{(k,\mu)} \in R$, $1 \leq k \leq n$, thus in particular, $\eta \in S^{(\tau,\mu)}$.

We can now build a linear combination similar to (25) such that all terms on right hand side of (40) are cancelled. For example, kernels of Jackson and De La Vallee Poussin $\beta^{(p)}$ with its convergence factors.

For $p = 2$, we have,

$$\chi_n(x) = \gamma_1 P_{a_1 n}^2(x) + \gamma_2 P_{a_2 n}^2(x) + \gamma_3 P_{a_3 n}^3(x), \quad x \in R \quad (41)$$

Where, $a_i = i$, $1 \leq i \leq 3$, coefficients γ_i can be uniquely determined.

Here, for the linear combination,

$$\chi_n(x) = \sum_{v=1}^s \gamma_v g_{(na_v)}(x) \text{ holds (36).}$$

From this, we have the Following:-

Corollary 4.1. The unique linear combination χ of Jackson De La vallee Poussin kernels (41) satisfying (36) is given by,

$$\chi_n(x) = \frac{9}{4} P_{a_1 n}^2(x) - \frac{4}{3} P_{a_2 n}^2(x) + \frac{1}{12} P_{a_3 n}^3(x), \text{ for, } x \in R \quad (42)$$

Corresponding singular integral for $f \in C_{2\pi}$ is given by,

$$\|E_{(n,\chi)}(f, \cdot) - f(\cdot)\| = O(1) E_{(n)}(f)$$

The kernels of Jackson $j^{(p)}$ of (16), $p \geq 2$, admits no expansion of the form (40). Also, here linear combination are not saturated. This will be briefly outlined for $p = 2$. The convergence factors are then given by,

$$\rho_{(k,n)}(j^2) = \frac{1}{4n^3+2n} \begin{cases} 3k^2 - 6nk^2 - 3k + 4n^3 + 2n, & 0 \leq k \leq n \\ -k^3 + 6nk^2 - 12n^2k + k + 8n^3 - 2n, & n \leq k \leq 2n \end{cases} \quad (43)$$

Now for j^2 and $a_i = i$, $1 \leq i \leq 3$, leads to another corollary.

Corollary 4.2. [10] The unique linear combination $\tilde{\chi}$ of Jackson De la Vallee poussin kernels (42) satisfying (36), is given by,

$$\tilde{\chi}_{(n)}(x) = -\frac{1}{18} \left(1 + \frac{1}{n^2}\right) J_n^2(x) + \frac{1}{3} \left(4 + \frac{1}{2n^2}\right) J_{2n}^2(x) - \frac{1}{2} \left(1 + \frac{1}{2n^2}\right) J_{3n}^2(x) + \frac{1}{9} \left(32 + \frac{1}{n^2}\right) J_{4n}^2(x) \quad (44)$$

Corresponding singular integral for $f \in C_{2\pi}$ is given by,

$$\|E_{(n,\tilde{\chi})}(f, \cdot) - f(\cdot)\| = O(1) E_{(n)}(f)$$

Here, the coefficients depend on n , and are bounded by linear combination $\tilde{\chi} = \{\tilde{\chi}_{(n)}(x)\}$, $n \in N$, which defines an approximation process and is not saturated by construction.

Conclusion. Jackson de La Vallee Poussin kernels seems more suitable for these linear combinations since the calculation of coefficients is less elaborate.

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RESEARCH ARTICLE

LIMNOLOGICAL STUDY OF MADDIKUNTTA LAKE , DISTRICT: GADCHIROLI (MS)

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Abstract

Present paper Study with the physico-chemical parameters of Maddikunta lake, Tahsil :-Sironcha ,Dist:-Gadchiroli. The work was carried out during the period of Sep- 2015 to Aug 2016. This lake was established for irrigation, Drinking water and fish culture purpose .It was formed 300 years ago from migrant telugu peoples from Andhra Pradesh . It was in the out of 6 km distance from Tahsil:Sironcha . In rainy season it's receiving Village sewage, fertilizers , Pesticides . This typs of water injuries to the health of human other aquatic Fauna. So there is an urgent requirement for its extent of pollution which will help us in further management of conservation. During the study period examine the physico-chemical parameters such as: atmosphere, water temperature, pH, electrical conductivity, alkalinity, total hardness, TDS, Ca, Mg, chlorides, sulphates and phosphate, following stranded methods (APHA 1998). Now this lake is becoming eutrophic in nature.

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Introduction:-

Water is the most essential requirements of life. The survival and quality of human life depends up on the availability of fresh water. The aquatic animal's life directly or indirectly depends on water quality status (A. Bajpai, 1993). (A. Mishra et.al.1993). (sayeshwara,2010). Water quality study provide the current information about the suitability of water for designated uses and to improve existing condition (Lloyl 1992).

Now a day's most of the aquatic ecosystem receives million liters of village sewage, and agricultural runoff. Its cause to nutrient enrichment cause to the eutrophication in aquatic ecosystem. (A.A. Ansari,2006). Pollution of the aquatic environment by inorganic and organic chemicals is a major factor to the survival of aquatic organisms and including fish population (Saeed and Shaker 2008). The main object of this paper to examine the physic-chemical parameters of Maddikuntta Lake and to suggested to conservation methods for water for drinking and irrigation purposes.

Study area:-

The lake in Maddikuntta ,Tahsil:Sironcha ,District:Gadchiroli(MS). Maddikuntha is a Village in Sironcha Taluka in Gadchiroli District of Maharashtra State, India. It belongs to Vidarbha region . It belongs to Nagpur Division . It is located 179 KM towards South from District head quarters Gadchiroli. 6 KM from Sironcha.874KM from State capital Mumbai

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Ramanjapur (vl) (2 KM) , Arda (2 KM) , Nagram (4 KM) , Sironcha (ryt) (4 KM) , Sironcha (4 KM) are the nearby Villages to Maddikuntha. .This Place is in the border of the Gadchiroli District and Karimnagar District. Karimnagar District Mahadevpur is South towards this place . It is near to the Telangana State Border.

Material And Methods:-

The investigation was carried out to study the physico- chemical and biological aspect of the lake during September 2015 to August 2016, the Maddikuntta lake situated 6 km distance from Tahsil: Sironcha, District:Gadchiroli. Figure – 1. 160ht paddy field is irrigation under this lake. Water sample were collected in morning hours at 6-30am to 8-30 am.

Physical chemical parameters such as: temperature, pH, TDS, electrical conductivity, alkalinity, total hardness, TDS, Ca. Mg, chlorides, sulphates and phosphate, following were analyzed in the laboratory by using standard methods (APHA,2005).



Results:-

The result of physico-chemical characteristics of maddikunta lake water has been submitted in table. 1 and table 2.

Temperature:-

Water Temperature is an important role plays in determine the growth of organisms ultimately the water quality. Water is a vital parameter for growth of Organism. It also influences the metabolic rate of aquatic Organism. Excess amount of nutrients, high temperature cause to the eutrophication. The maximum temperature value was recorded to be 35.5 degree Celsius in the month of may at the sampling station B and minimum 19 degree Celsius January at sampling station A. In study period i.e. Sep-2015 to Aug-2016. .

pH:-

The high variation in pH of water is an indicate the highest productivity nature of lake water. According to Hora and Pillay the pH variation is mostly due to diurnal interplay of photosynthesis and community respiration of the biota and also is one of the most important single factors, which influences the aquatic production. The pH considered an important environmental factor. The maximum value of pH was recorded 8.5 in month of May at the sampling station B and minimum 7.8 was recorded in the month of September at sampling station A. In study period i.e. Sep-2015 to Aug-2016. .

Transparence:-

Intensity of light cause to the growth of planktons, it an indicator of productivity. The maximum value of transparency was recorded 25.5 cm in month of November at the sampling station A and minimum 14.5 was recorded in the month of August at the sampling station A. In study period i.e. Sep-2015 to Aug-2016. .

Electric Conductivity:-

Electric conductivity is a numerical expression ability of an aqueous solution to carry electric current. It's the best indicator of water pollution as conductivity is the indirect measure of TDS, nutrient. Conductivity increases with increasing amount and mobility of ions. These ions come from the break down of compounds. The maximum value of conductivity 1800 μm was recorded in month of July at the sampling station B and minimum 1150 μm was recorded in the month of April at the sampling station A. In study period i.e. Sep-2015 to Aug-2016. .

Total dissolve solids:-

Total dissolved solids are composed of mainly of chlorids, Sulphates, Phosphates, Nitrates, Calcium, Magnesium, Sodium, Potassium, Iron, Magnesium carbonates, bicarbonates (H.R. Esmaeli and M.S. Joshi 2005). Total dissolved solids denote mainly the various kinds of minerals available in the water. The maximum value of TDS 1330 mg/lit was recorded in month of September at the sampling station B and minimum 970 mg/lit was recorded in the onth of May at the sampling station A. In study period i.e. Sep-2015 to Aug-2016.

Total Hardness:-

The maximum value of total hardness 152 mg/lit was recorded in month of April at sampling station A and minimum 87 mg/lit was recorded in the month of September. Salve and Hiwre (2006), S.E. shinde et. Al (2011) reported that TH high in winter low in summer and monsoon season. In study period i.e. Sep-2015 to Aug-2016.

Table No. 1:- Physico-chemical parameters of Maddikuntta lake, District :Gadchiroli during Sep-2015 to Aug-2016 at Sampling Station A

	15-Sep	Oct	Nov	Dec	16-Jan	Feb	Mar	April	May	Jun	July	16-Aug
Water Temp.	28	26.5	25.2	22	18.5	23.6	29.5	32.2	36	32.5	29.2	26
pH	7.6	8.2	8.2	8.2	8.7	8.4	8.6	8.3	8.2	8.2	8.1	7.8
Transparence	16.6	22.2	25.5	22.5	23.5	24.5	19.5	22.5	22.6	20.5	17.6	14.5
Conductivity	1520	1800	1700	1520	1430	1300	1230	1150	1610	1750	1820	1720
TDS	1320	1164	1188	1205	1155	1255	1120	990	970	1040	1185	1210
Total hardness	87	118	124	127	134	135	145	151	145	127	115	97
Ca	64	66	72	73	82	88	94	102	112	94	84	78
Mg	17	54	52	53	54	41	57	51	32	28	29	19
Alkalinity	252	304	312	324	266	180	164	194	112	104	192	233
Cl	64	70	57	55	64	68	73	74	74.5	69.5	71.5	68.5
P	1.4	1.15	0.8	0.98	1.24	0.99	1.4	2.5	3.4	1.8	1.4	1.04
S	1.9	1.8	1.05	1.3	1.4	1.45	1.9	2.3	2.9	3.3	1.8	1.7

Table No. 2:- Physico-chemical parameters of Maddikuntta lake, District :Gadchiroli during Sep-2015 to Aug-2016 at Sampling Station B.

	15-Sep	Oct	Nov	Dec	16-Jan	Feb	Mar	April	May	Jun	July	16-Aug
Water Temp.	28.2	26.6	25.4	22.5	18.3	23.5	29.7	32.4	35.5	31.5	29.4	26.6
pH	7.6	7.9	8.0	8.2	8.3	8.2	8.4	8.5	8.6	8.1	8.1	7.8
Transparence	16.6	22.2	25.5	22.5	23.5	24.5	19.5	22.5	22.6	20.5	17.6	14.5
Conductivity	1550	1800	1700	1520	1430	1340	1230	1150	1620	1750	1800	1730
TDS	1330	1164	1188	1204	1150	1250	1125	992	970	1040	1185	1220
Total hardness	86	118	124	126	134	136	145	152	146	128	115	98
Ca	64.5	66	72	74	82	88	94	101	110	94	84	78
Mg	18	54	52	53	54	41	57	51	32	28	29.5	19.4
Alkalinity	253	305	312	325	266	180	164	194	115	105	192	233
Cl	64	70	57	56	64	68	73.5	74	74.5	69.5	71.5	68.5
P	1.3	1.15	0.8	0.98	1.25	0.99	1.5	2.5	3.4	1.8	1.5	1.05
S	1.8	1.9	1.05	1.2	1.5	1.46	1.8	2.5	2.9	3.3	1.9	1.8

Calcium:-

Calcium is one of the most abundant substances present in the natural water. In aquatic environment calcium serves as one of the micronutrients for most of the organisms. The maximum value of calcium 110 mg/lit was recorded in month of May at the sampling station A and minimum 64 mg/lit was recorded in the month of September at the station B. In study period i.e. Sep-2015 to Aug-2016.

Magnesium:-

Magnesium is essential for chlorophyll bearing organism, since it goes into composition of the pigments. Decrease the value of magnesium may be due to plankton and algae. The maximum value of magnesium 57 mg/lit was recorded in month of March at the sampling station B and minimum 17 mg/lit was recorded in the month of September at the sampling station A. In study period i.e. Sep-2015 to Aug-2016.

Alkalinity:-

Separation of alkalinity soft & hard water scale is 40 mg/lit. J.B. Moyle (1949). The maximum value of 325 mg/lit was recorded in month of December and at the sampling station B and minimum 104 mg/lit was recorded in the month of June at the sampling station A. In study period i.e. Sep-2015 to Aug-2016.

Chloride:-

Large amount of chloride in freshwater is an indicator of organic pollution. The maximum value of Chlorides 74.5 mg/lit was recorded in month of May at the sampling station B and minimum 55mg/lit was recorded in the month of December at the sampling station A. Chlorides increase in summer and decrease in winter (Singh 1960, Zafer A.R. 1964) in study period i.e. Sep-2015 to Aug-2016.

Phosphates:-

The maximum value of Phosphates 3.4mg/lit was recorded in month of May at the sampling station B and minimum 0.8 mg/lit was recorded in the month of November at the sampling station A. High value of phosphate during summer may be attributed to the decrease water level and release of phosphate due to decomposition of organic matter. (Seenyya and Jafar, 1979, and Saha 1984). In study period i.e. Sep-2015 to Aug-2016.

Sulphates:-

It's a present in fertilizer they contribute to the water body from agricultural runoff. Discharge of domestic sewage in water tends to increase its concentration. The high concentration of sulphate stimulates the action of sulphur reducing bacteria, which produce hydrogen sulphide, a gas highly toxic to fish life. The maximum value of Sulphates 3.3mg/lit was recorded in month of June at the sampling station B and minimum 1.2 mg/lit was recorded in the month of December at the sampling station A. Reddy et.al (2009) high value recorded in monsoon season. In study period i.e. Sep-2015 to Aug-2016.

Conclusion:-

The present study reveal that Naspur Lake was polluted by the sewage water and domestic uses that's why the lake was polluted.

To prevent the lake pollution some measures to be take.

1. Treatment of sewage water and avoid the Village sewage connection to lake.
2. Avid the washing of cloth.
3. Bathing of cattle.
4. Dumping of Agricultural waste.
5. Water quality comparision of these two sampling station of maddikuntta lake reveals that the situation is not too worst but it is alarming .
6. It needs prper conservation and managements plans,strategies for the restoration etc.

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RESEARCH ARTICLE

SMART CLOUD FILE RETRIEVAL USING SEARCHABLE CIPHERTEXT KEYWORDS & ACCESS CONTROL ON INDEX

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Cloud Computing, Data Security, Access Control, ABE & IBE

Abstract

Cloud Storage becomes more popular and many corporate, government organizations, share their data in public cloud which are semi-trusted cloud storage. Public cloud storages are hack able and data can be leak out, so we need a system where the files are encrypted and then stored in cloud. If the volume of the files is large then system need search engine to retrieve the file, in-turn search engine need keyword index for the fast performance. There may be a chance for the intruder to get the details of the files through unencrypted index keywords, So in proposed system index keywords are undergo hashing technique and cipher text are stored in index. This system will encrypt the file and encode the index keywords which give double production to the data in cloud. Further to the data security this system has access control system. At present ABE (Attribute Based Encryption) & IBE (Identity based Encryption) are used for the file access control in cloud which requires more processing time and calculations. Proposed system uses a new technique where Access Control is included in search index keywords, which require less computation and quick retrieval.

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Introduction:-

Cloud computing allows users to use enormous data storage and infinite computation capabilities at a very low price. Even though cloud storage gives lot of benefits, Data Owner is not having the full control over the outsourced data, it can be hacked. To avoid this problem, user should encrypt their data before outsourcing to the cloud. However, encryption can obstruct some useful functions such as searching over the encrypted data which is outsourced in cloud, while enforcing an access control policy. Moreover, it is natural to outsource the search operations to the cloud, while keeping the outsourced data private. There is a need to allow the data consumers to verify whether the cloud faithfully executed the search operations or not. To the best of our knowledge, existing solutions cannot achieve these objectives simultaneously.

Our Contributions:-

We propose a new concept, called *Smart Cloud File Retrieval using Searchable Cipher text Keywords & Access Control on Index*. This system allows a user to control the search, and retrieve its outsourced encrypted data according to a user access control policy, while allowing the authorized users to search and retrieve files through innovative index based access control system.

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In other words, a data user with proper authentication and attribute can access the encrypted files in the cloud with access control rules. This system provide followings (I) Data Owner can upload the files into the cloud in encrypted form, (ii) Data Owner can able to set Access Control Policies for the outsourced files (iii) Data User can able to search and retrieve the file through access control based cipher text index keywords.

We formally define the security properties of proposed system and present schemes that provably satisfy them. The scheme is constructed in a modular method, by using attribute-based encryption, bloom filter, digital signature, and a new building-block we call attribute-based Cipher text keyword search (ABCKS) that may be of independent value. Experimental evaluation shows that the system provides good result and performance.

Associated Works:-

To the best of our knowledge, no existing solution is sufficient for what we want to achieve. In what follows we briefly review the relevant techniques.

Attribute -Based Encryption (ABE). ABE is a popular method for enforcing access control policies via cryptographic means. Basically, this technique allows entities with proper credentials to decrypt a cipher text that was encrypted according to an access control policy [2]. Depending on how the right of entry control policy is enforced, there are two variants: KP-ABE (key-policy ABE) where the decryption key is associated to the right of entry control policy [2], and CP-ABE (cipher text-policy ABE) where the cipher text is associated to the access control policy [5]. ABE has been enriched with various features. In this paper, we use ABE to construct a new primitive called attribute-based keyword search (ABKS), by which keywords are encrypted according to an access control policy and data users with proper cryptographic credentials can generate tokens that can be used to search over the outsourced encrypted data. This effectively prevents a data owner from knowing the keywords a data user is searching for, while requiring no interactions between the data users and the data owners/trusted authorities. This is in contrast to [6], where the data users interact with the data owners/trusted authorities to obtain search tokens.

Keyword Search over Encrypted Data. This method allows a data owner to generate some tokens that can be used by a data user to search over the data owner's encrypted data. Existing solutions for keyword search over encrypted data can be classified into two categories: searchable encryption in the symmetric-key setting and searchable encryption in the public-key setting. Several variants have been proposed to support complex search operations. Moreover, searchable encryption in the multi-users setting has been investigated as well [9], [10], where the data owner can enforce an access control policy by distributing some (state full) secret keys to the authorized users. However, all these solutions do not solve the problem we study, because (i) some of these solutions require interactions between the data users and the data owners (or a trusted proxy, such as a trapdoor generation entity [6]) to grant search capabilities, and (ii) all these solutions assume that the server faithfully executed search operations. In contrast, our solution allows a data user with proper credentials to issue search tokens by which the cloud can perform keyword search operations on behalf of the user, without requiring any interaction with the data owner. Moreover, the data user can verify whether or not the cloud has faithfully executed the keyword search operations. This is true even for the powerful technique called predicate encryption, which does not offer the desired verifiability.

Verifiable Keyword Search. Recently, verifiable keyword search solutions have been proposed in [12], where each keyword is represented as a root of some polynomial. It is possible to check whether a keyword is present by evaluating the polynomial on the keyword and verifying whether the

Output is zero or not. However, these approaches work only when keywords are sent in plaintext to the cloud, and are not suitable for our purpose because the cloud should not learn anything about the keywords. It is worth mentioning that the secure verifiable keyword search in the symmetric-key setting can be insecure in the public-key setting because the attacker can infer keywords in question via an off-line keyword guessing attack (in lieu of the off-line dictionary attack against passwords).

Problem Statement:-

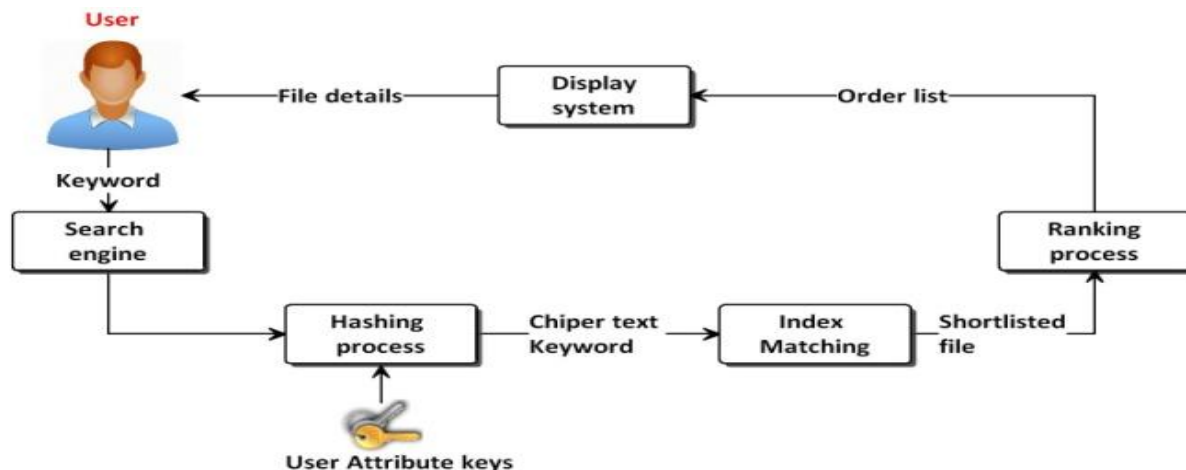
Consider a situation where a college has to store lot of e-books in cloud storage. That e-book has to be accessed by their students. If the e-books are less in number, the students can able to browse and download. Consider a situation the number of e-books is huge it is not possible to browse and download the file, so the students need a search engine. Whenever search engine come into picture, indexing technique have more weight age. The problem here is

to safeguard the file content and index keywords, index keywords are usually sensitive and which are linked with the file they are connected when a hacker get this index data he may guess the file content. Our aim is to safeguard the index keyword, file content and to implement access control system.

Access control is a big challenge, in current cloud storage for access control, attribute based encryption (ABE), identity based encryption (IBE) are used. Now in this system we are using Attribute Based Cipher text Index Keyword for access control.

The assumption in this system is each department in the college will have one Unique Attribute Key (UAK). At the time of uploading the file into the cloud the concern user has to provide the access policy details, like which are the department students and staff can able to access the file from the cloud.

Proposed system will automatically remove the unnecessary words from the file and create the keywords with weights using Term Frequency (TF)



algorithm. All the extracted keywords will processed with hashing technique of allowed department Attribute

Keys which will provide Attribute based cipher text Keywords; these cipher text keywords are stored in index array for searching process. For example there are I keywords are extracted from the e-book and the authority selected J departments in access control setting then in Attribute Based Cipher text Keywords generated will be $I * J$. All the ABCK are inserted into index array. The idea behind this is each keyword will undergo hashing process with accessible department unique attribute keys. Since department based attribute keys are unique and this will create the same Attribute Based Cipher text keywords when the users from the same department search in the search engine. This is show in fig.1.

In this paper, we propose the new approach of *Smart Cloud File Retrieval using Searchable Cipher text Keywords & Access Control on Index* as a better solution, as depicted, in Fig. 2, Data owner only needs to generate one department one attribute key instead of attribute key to all user, instead of $\{k_i\}_{i=1}^m$ for sharing m documents with User, and User only needs to submit a Login ID for accessing the file, from the Login ID user's department is identified and then the corresponding department attribute key is used to generate the attribute based cipher text keywords. The cloud server can use this aggregate trapdoor and some public information to perform keyword search and return the result to User. Therefore, in Smart Cloud File Retrieval, the delegation of keyword search right can be achieved by sharing the single aggregate key. We note that the delegation of decryption rights can be achieved using the key-aggregate encryption approach recently proposed in, but it remains an open problem to delegate the keyword search rights together with the decryption rights, which is the subject topic of this paper. To summarize, the problem of constructing a *Smart Cloud File Retrieval* scheme can be stated as:

"To design a Smart Cloud File Retrieval using Searchable Cipher text Keywords & Access Control on Index scheme under which any keyword cipher texts from any document is searchable with a constant-size cipher text keywords generated by a constant-size department attribute key."

Implementation:-

This system is developed in web technology with MVC architecture. The proxy server has crypto system process which encrypts and decrypts the file, hashing technique which is used to get the keyword from the user and produce user attribute based hash code. A powerful search engine with access control based indexing system. There, is a ranking system which short the shortlisted file based on search keyword weights. The proxy server provides software as a service, which is a private server and the encrypted files are stored in public cloud storage which provides storage as a service. This system combines private server and public server so it forms hybrid cloud approach.

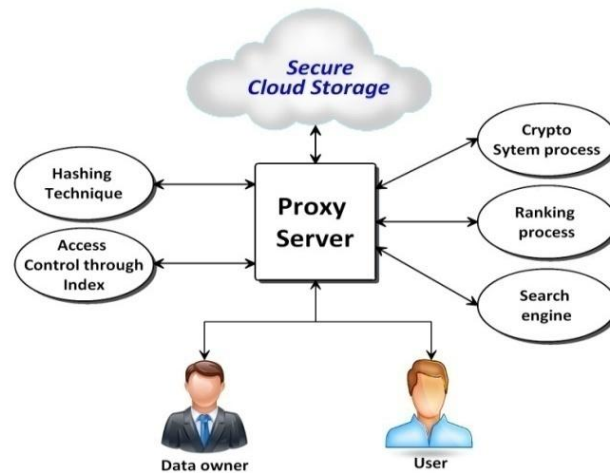


Fig. 2:- System Architecture

Security Analysis:-

The security system of proposed work is very strong. It gives security from outside hackers as well as security from insider (with access control) the registered user. The files which are stored in cloud storage are encrypted and the index keywords are in hash tag which gives double protection from the hackers. Since attribute based cipher text keywords are used even though the registered user can able to retrieve the files for which they have the access.

Conclusion:-

By considering the disadvantage of data outsourcing in public cloud storage we come up with a new idea attribute based cipher text keyword search (ABCK). In this the access control mechanism is built in search index. There is no need to provide attribute based key for each and every user. With the experimental result and evaluation technique our system result shows it is much better in performance and convince in secure file retrieval system from cloud storage for the further feature work. We consider multi owner document storage system and issues related to that.

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RESEARCH ARTICLE

FIRST-PRINCIPLES STUDY OF OLIVINE-TYPE NaMnPO_4 AS POSITIVE ELECTRODE MATERIALS FOR RECHARGEABLE SODIUM ION BATTERIES.

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Key words:-

First-principles calculations, Density functional theory, Na-ion batteries, Na ion diffusion, GGA+U Optimization.

Abstract

The diffusion mechanism of Na ions in the olivine-type NaMnPO_4 is investigated by first-principles calculations. Electron transport properties studied by first-principles density functional theory (DFT) GGA+U method and achieved Final Energy/Atom -7.03 eV. The calculated diffusion energy was 0.65 eV for Na to diffuse along the (0 1 0) direction. The large inter and intra-layer distance of Mn 3.156 Å° and 6.312 Å° offers feasible channels for fast Na ion diffusion in (0 1 0) direction. Implications of first-principles calculations useful strategy for better understanding the electrochemical properties of olivine-type NaMnPO_4 are discussed for designing better-performing Na ion rechargeable batteries.

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Introduction:-

Sodium-ion batteries have vast potential as an inexpensive, geopolitically-neutral alternative to Li-based rechargeable batteries, largely due to the global abundance and low cost of sodium-containing precursor materials. Na-ion electrode materials, while conceptually similar to their Li-ion counterparts, give rise to unique challenges and physical properties due to sodium's larger size and lower absolute standard electrode potential. Due to their larger size, Na ions can reversibly intercalate in more materials than Li-ions, giving a much broader chemical space in which electrode materials can be optimized. This, together with sodium's abundance, makes Na-ion batteries excellent candidates for large-scale energy storage [1-4]. In this computational studies with the goal of novel Na-ion electrode discovery, as well as material optimization and understanding of the underlying physics governing material function. First-principle computational efforts are used to obtain insights into phase transitions during electrochemical deintercalation of Na-ion cathodes, as well as diffusion mechanics and diffusivity. High throughput computation is utilized to search for the novel cathode, anode, and solid electrolyte materials. In this paper, we examine olivine-type NaMnPO_4 using first-principles calculations. It has been noted that LDA and GGA give relatively large errors for the sodium intercalation potentials. So we purpose is to extend such calculations to other compounds of the phospho-olivine family and to show that treating electronic correlations within the GGA+U method gives considerably better agreement with experiment and thereby provides a tool to accurately predict Na intercalation potentials. We first present the details of the GGA+U method as well as structural information on the Sodium-orthophosphate oxides and show how the electrochemical reactions take place in a rechargeable Sodium ion battery.

Computational methods:-

The First principle calculations have been performed using the Quantum ESPRESSO PWscf code [5] using a plane-wave basis to express the wave function of the valence electrons and ultrasoft pseudopotentials to describe the

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interactions of ionic cores and valence electrons [6]. QuantumWise VNL-ATK 2016 academic version was used as GUI for Quantum ESPRESSO. The exchange-correlation contribution was described by the generalized gradient approximation (GGA) of Perdew, Burke, and Ernzerhof [7]. Onsite Coulomb correction (GGA + U) was included to describe the localized electronic states of Mn 3d in mixed-valence rare earth metals. A spin polarization calculation was used. A kinetic cutoff energy of 520 eV, and $5 \times 5 \times 3$ k-point meshes in the NaMnPO₄ unit cell (28 atoms) were found by a convergence test (<3 meV/NaMnPO₄). Because the band gaps of both compounds were underestimated, which is typical of the GGA + U functional [8]. The relaxation of unit cell volume was done by total energy minimization, and atom positions were relaxed until the remaining force acting on the atoms was less than 10^{-3} eV/Å. The Na diffusion in NaMnPO₄ is calculated by Na jumps between neighboring M1 sites were obtained by means of the nudged elastic band (NEB) method [9]. The threshold for the total force that is acting on the NEB images of the interpolated reaction path was set to 0.05 eV/Å.

Results and discussion:-

Figure 1. Shows the bulk crystal structure of sodium Manganese phosphate [NaMnPO₄] with orthorhombic (ICSD - 201771) with space group Pnma. The olivine structure of the NaMnPO₄ is a hexagonal close-packed array of oxide ions containing isolated PO₄ tetrahedral and corner-sharing MnO₆ octahedra in the (0 1 1) plane and edge-sharing NaO₆ octahedral, stacked along the [0 1 0] direction. Because the oxygen atoms are strongly bonded to both Mn and P atoms.

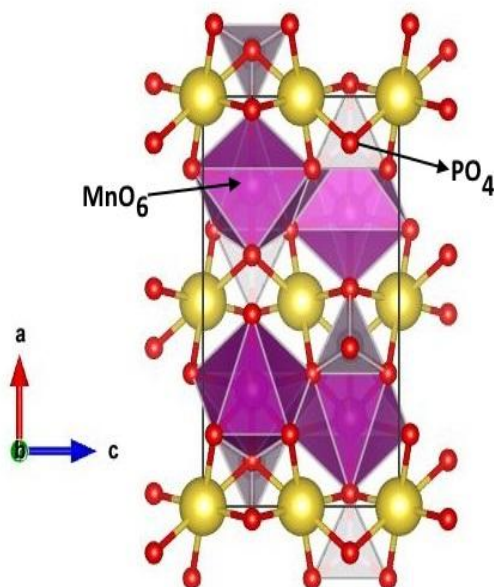


Figure 1:- Crystallographic structure of the bulk NaMnPO₄ (Yellow, red, gray and purple balls refer to Na, O, P and Mn atoms, respectively).

The optimized lattice constants for the NaMnPO₄ are summarized in Table 1, along with other experimental values. The calculated values are larger than the experimental ones, but the errors of the computed values compared with the experimental ones are within 0.04–0.33%. The underestimation of the lattice constant compared with the equilibrium values is due to the feature of GGA functional used.

Table 1:- Optimized lattice constants for olivine-type NaMnPO₄ (Å)

		a Å	b Å	c Å	reference
NaMnPO ₄	Exp.*	5.155	6.976	9.210	ICSD ID 201771
	Computed	5.172	6.981	9.214	This work

* J. Moring and E. Kostiner, J. Solid State Chem., 1986, 61, 379–383.

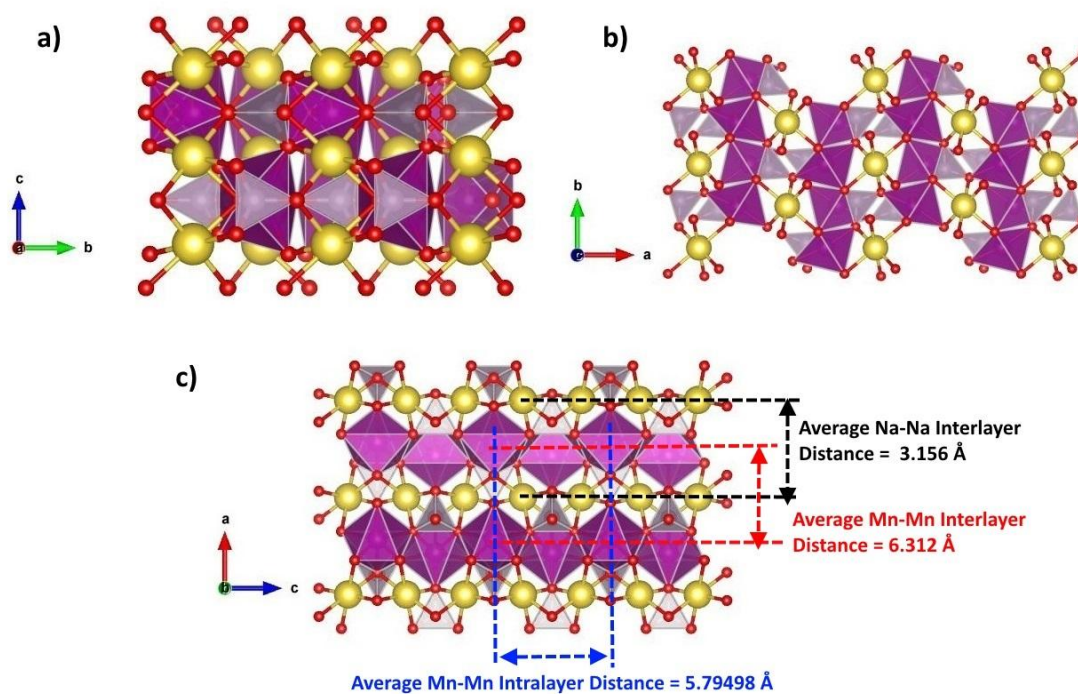


Figure 2:- Crystallographic structures of NaMnPO₄ a) 100 b) 001 c) 010 directions

Figure 2. shows structural illustration of olivine-type NaMnPO₄ (100), (001), (010) directions and Figure 2c shows the Na-diffusion channels in NaMnPO₄ in (010) directions. It offers more interlayer Mn–Mn distance (6.312 Å) than the inter-layer Mn–Mn distance (5.794 Å), which facilitates two-dimensional Na-ion diffusion pathways to achieve the fast intercalation and de-intercalation phenomena along the a and c directions. These free wider paths offer fast Na ion in NaMnPO₄.

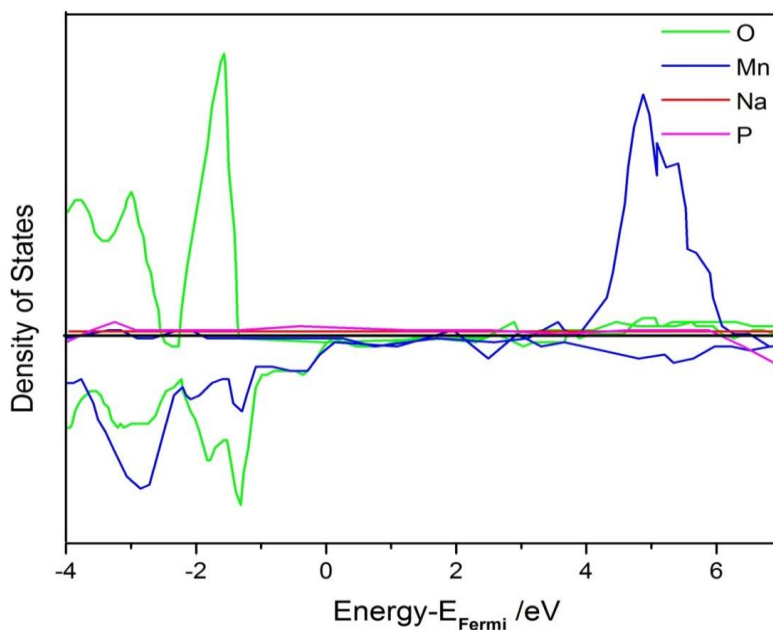


Figure 3:- Partial density of states (PDOS) for NaMnPO₄ (PDOS is aligned so that the Fermi energy is zero)

Figure 3. shows the partial density of states (PDOS) around the Fermi level for olivine-type NaMnPO_4 , respectively. The localized Mn 3d bands form a valence band maximum and conduction band minimum, and the band gaps are larger than 3.051 eV in NaMnPO_4 . Therefore, the intrinsic band conduction of electrons is negligible at room temperature because of the localization and wide band gap. Electronic conduction in NaMnPO_4 mainly arises from localized electron/hole (polaron) hopping. [10] Polarons are created by the redox reaction of $\text{Mn}^{2+/3+}$ to maintain charge neutrality during electrochemical Na^+ removal (uptake). In Figure. 3 the calculated energy profiles for localized hole (Mn^{3+}) jumps of ~ 0.2 eV for both compounds. The polaron migration energies are much smaller than the band gaps, and thus polaron migration is the dominant electron conduction mechanism. The similarity of the polaron migration energies of the two compounds indicates that the difference in rate performance does not stem from polaronic migration. Alkali ion migration is another factor affecting the rate performance in batteries.

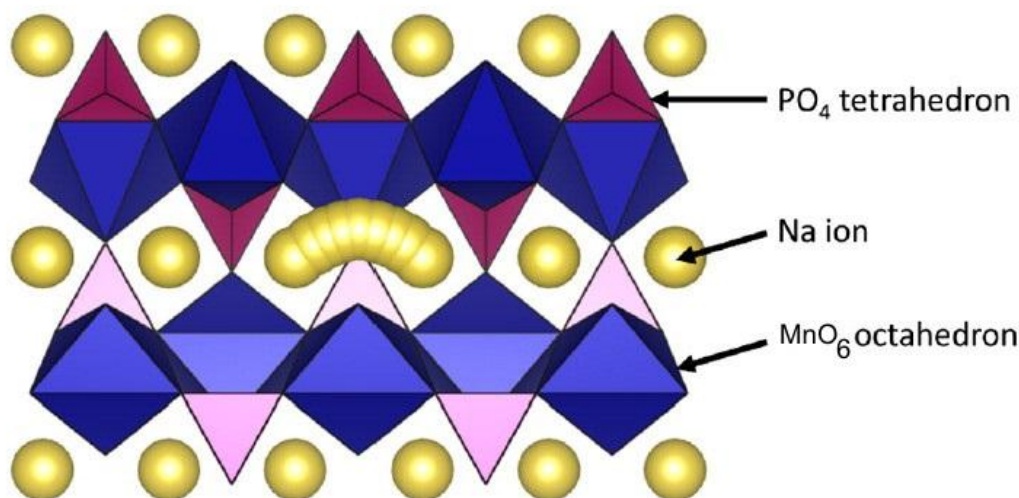


Figure 4:- Trajectory of Na ions in the NaMnPO_4 bulk structure calculated by the NEB method.

NEB calculations were performed to evaluate the migration energies of alkali ions NaMnPO_4 . The GGA functional was used to avoid the effect of complicated polaron interactions. Figure 4 shows the migration trajectory of Na^+ hopping in the NaMnPO_4 bulk structure. Na ions at the octahedral site jump to the neighboring site via the tetrahedral vacancy site along the (0 1 0) direction, forming a one-dimensional pathway. The migration energy profiles are shown in Figure 5, where the energy maximum is located at the center of migration path (tetrahedral vacancy).

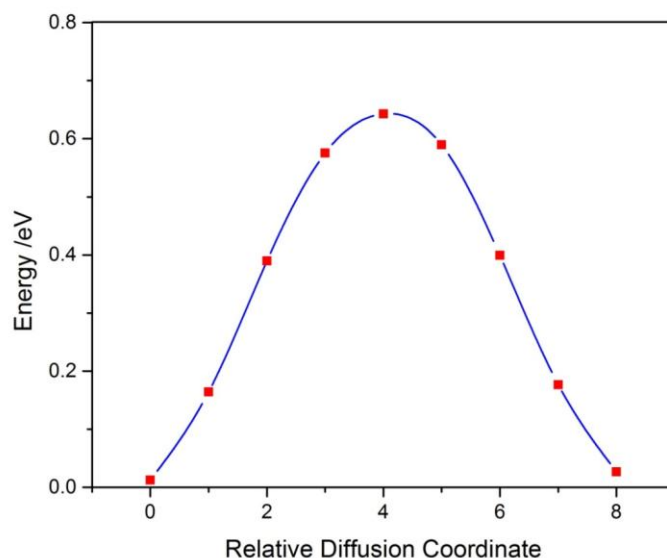


Figure 5:- Diffusion energy curves of Na in bulk NaMnPO_4

The migration energy 0.65 eV achieved in NaFePO₄. The migration energy 0.65 eV is quite higher than in LiMnPO₄ because Na⁺ is slightly larger than that of Li⁺, which may arise from the size effect of the larger ionic radius of 6-coordinate Na⁺ (1.02 Å) compared with Li⁺ (0.76 Å). the diffusion coefficient, D, can be expressed by migration energy, E_m, as follows,

$$D = a^2 v^* \exp\left(\frac{-E_m}{k_B T}\right) \dots\dots (1)$$

Where a, v*, k_B and T correspond to hopping distance, attempt frequency, Boltzmann constant, and absolute temperature, respectively.

Interfacial migration mechanism (Figure 6) indicate that a fast charge–discharge mechanism occurs in Na_{1-x}MnPO₄ electrodes owing to the interfacial Na ion migration mechanism (E_m ~ 0.15 eV) as proposed in the domino-cascade model, whereas a bulk conductivity mechanism is dominant in the Na_{1-x}MnPO₄ electrode (E_m ~ 0.65 eV) because of the miscibility in terms of Na/vacancy arrangements in the lattice.

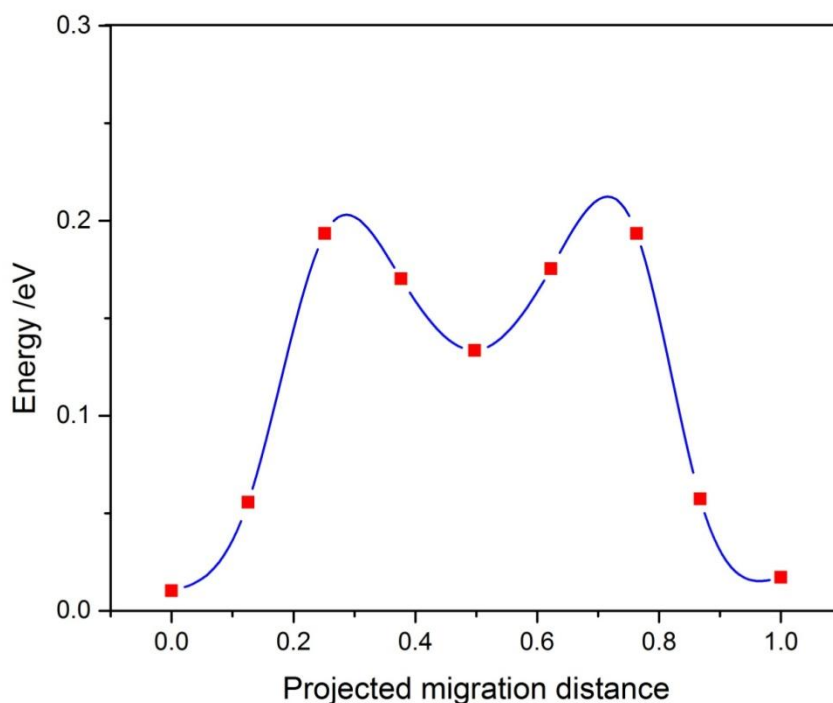


Figure 6:- Calculated migration energy of alkali ions at the (010) interface for NaMnPO₄/ MnPO₄

In addition, a phase transition will occur during the charge and discharge processes in NaMnPO₄ electrode material, thus a correction to the method will be required to calculate the voltages for Sodiation/deSodiation caused by the phase transition. Moreover, the energy of Sodiation/desodiation is very small in some electrode materials, much smaller than the activation energy of Na migration.

Conclusion:-

First Principle calculation of olivine-type NaMnPO₄ successfully studied by Density function theory. The calculated diffusion energy barriers of the bulk NaMnPO₄ and 0.65 eV. We Studied bulk polaron migration mechanism (Figure 5), and bulk alkali ion migration mechanism (Figure 6). Therefore, the difference in alkali ion size is not a major factor controlling the rate performance of the charge–discharge reaction. However, the phase stability of olivine-type compounds indicates a two-phase reaction mechanism for the NaMnPO₄ electrode Our results suggest that faster Na⁺ migration at the interface than in the bulk originates from the two-phase reaction mechanism. Accordingly, the formation of the interface in NaMnPO₄ may play an important role in the fast charge– discharge properties of olivine-type electrodes.

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RESEARCH ARTICLE

GENETIC DIVERSITY AMONG FOUR *MOMORDICA* SPECIES USING RAPD, SSR AND ISSR MARKERS

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Abstract

Background: Genus *Momordica*, is widely distributed over tropic and subtropical region. Besides the cultivated bitter melon (*M. charantia* L. var. *charantia*), many other species of the genus occur in the wild state have been found in India and neighboring countries. Among them, monoecious, *M. charantia* and *M. balsamina* and dioecious, *M.*

dioica and *M. cochinchinensis* exhibit divergence in morphological characters like growth habit, maturity, and fruit shape and size, sex expression, leaf, root and seed characters.

Methods: We studied the twenty-four accessions representing four *Momordica* species including two gynodioecious lines of *M. charantia* namely Gy-323, Gy-333 by morphological and molecular analysis through RAPD, SSR and ISSR markers.

Results: The analysis of study based on dendrogram obtained from the distance matrix from the mean value of the 15 quantitative traits for the genotypes grouped all the accessions into two major clusters. One cluster

separated two distinct groups each accession of *M. charantia* and *M. cochinchinensis* and *M. balsamina* formed distinct group within a major cluster while the second major cluster consisted of all accessions of *M. dioica*. For molecular diversity analysis, 101 primers including 50 RAPD, 16 SSR and 35 ISSR primers produced a total of 600 scorable amplicons across four species, of which 586 (97.08%) were polymorphic. Seventeen markers differentiating monoecious and dioecious and 85 amplicons specific to one of the four *Momordica* species were identified. The UPGMA dendrogram obtained from Jaccard's similarity coefficient (average similarity of 0.38) showed two major clusters clearly distinguishing monoecious and dioecious species with high bootstrap value (100) between nodes.

Conclusions: In the present study, morphological diversity

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based on quantitative traits and molecular diversity showed some

correspondence in the clustering pattern of accessions representing species. Molecular markers were more efficient in differentiating the accessions at and within species level than morphological markers.

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Introduction

Bitter melon (syn. bitter melon, *Momordica charantia* L.), an economically important member of the genus *Momordica* (Family: Cucurbitaceae) is widely cultivated in India, China, Malaysia, Africa and South America (Raj et al. 1993; Singh 1990) for its immature fruits as vegetable. Bitter melon is also used as a traditional medicine for diabetes in India, China and Central America (Grover et al. 2002; Ye et al. 2003), vaccine (Dutta et al. 1981) and other health related ailments due to its health promoting substances such as charantin (Ye et al. 2003). The genus *Momordica* comprising of 59 species is widely distributed all over tropic and sub-tropic region but the diversity of the species is more in tropical Africa including several species of economical and medicinal importance possessing different sex form and different basic chromosome number. About a third of the species are monoecious and remaining dioecious (Schaefer, 2010). The incompatibility among $2n=2x=22$, $2n=2x=28$ and $2n=4x=56$ species in this genus is indicated by the negative results of crossings between *M. charantia* L. and *M. muricata* L. and between *M. dioica* Roxb. *M. cochinchinensis* Spreng. (Trivedi and Roy 1972). Besides the cultivated bitter melon (*M. charantia* L. var *charantia*), several species are present in India, Sri Lanka, Bangladesh, Myanmar, Malay, etc. (Hooker 1879) occurring in the wild state and are gathered by tribal communities as vegetables. Seven species occur in India and among them *M. charantia* and *M. balsamina* are monoecious while *M. dioica*, and *M. cochinchinensis* are dioecious species and are propagated vegetatively through tuberous root (Rashid 1993). These species exhibit morphological divergence in characters like growth habit, maturity, fruit shape and size, sex expression, leaf, root and seed characters (Robinson and Decker-Walters 1999). The fruits are bitter in taste and are desirable for consumption. Thus, *M. charantia* is perhaps the only species in which bitterness has been considered as a trait for selection during domestication. Besides many culinary uses in south, southeast and east Asia, the genus is also cultivated for its use as folk medicines.

M. charantia provides for relatively broad phenotypic variation (Kundu 2012, Iqbal 2016), which produces large fusiform fruits, while *M. dioica* is an important vegetable in the Indian sub-continent. It has many advantages, like high market price, good nutritional value and longer shelf life (Rasul 2003, Singh 2013). Sweet melon (*M. cochinchinensis*) occurs in peninsular region, humid tropical forests and also in north-eastern region and balsam pear *M. balsamina* occurs in north-western, Indo-Gangetic plains adapted to dry and sandy soils. Sweet melon, tea melon and balsam pear being minor vegetables.

Application of molecular markers as a tool for identification and characterization of germplasm for their efficient management and use in plant breeding has gained importance (Karp & Edwards 1997). Among the different types of molecular markers, random amplified polymorphic DNA (RAPD) (Williams et al., 1990) is useful for the assessment of genetic diversity (Ulanowsky et al. 2001; Baranski et al. 2001; Pradeep Kumar et al. 2003; Cai et al. 2007; Tiwari et al. 2009;). The technique is ideally suited for fingerprinting applications because it is fast (Gwanama et al. 2000), requires little material (Lowe et al. 1996) and technically easy (Fenwick and Ward 2001). The wide availability of commercial primers makes this technique widespread (Gillies et al. 1997), inexpensive and yields large numbers of markers (Martin and Bermejo 2000).

Recently, some understanding of the genetic variations has been achieved using transcriptome analysis of within *M. charantia* species (Shukla et al. 2015) and molecular markers like RAPD (Dey et al. 2006; Behera et al. 2008; Bharti et al. 2012), Inter Simple Sequence Repeat (ISSR) (Singh et al. 2007; Bharti et al. 2012), AFLP (Gaikwad et al. 2008) and Simple Sequence Repeat (SSR) (Ji et al. 2012) in *M. charantia* and in related species (*M. dioica*) using RAPD (Rasul et al. 2007) and SSR markers (Ji et al. 2012). Transferability of SSR markers has also been reported in related

species (Jietal.2012). Although, most of these studies have focused on the efficiency of different molecular markers for assessing the diversity within one species, none aimed at assessing the diversity among some related *Momordica* species. The present efforts aim to get a comparative account of morphological and genetic diversity across the genus and unambiguously identify different species of *Momordica* using molecular and morphological markers.

Materials and Methods

Plant materials

Twenty four accessions of four *Momordica* species collected from different agro-climatic zones of India were selected based on morphological diversity and geographical distribution and were evaluated at the Research Farm, Indian Institute of Vegetable Research, Varanasi, (U.P.) during rainy-winter season of 2014. The list of accessions included in the study is detailed in Table 1.

Table 1: Salient Features of fused *Momordica* genotypes.

Species	Source	Salient Features
Spinegourd (<i>Momordica charantia</i>)		Monoecious, trailing vine with thin stems and tendrils, The leaves are heart-shaped, 5-10 cm in diameter, cut into 5-7 lobes, Flowers are borne singly in the leaf axils, fruits are small to very large, split open, revealing orange flesh and bright red placenta to which these seeds are attached, Seeds are tan and oval, with a rough etched surface.
GY-323	IIVR	Gynoecious plant, Large vines, large size leaves with heart shape,
GY333	IIVR	Gynoecious plant, medium vines, more branching, profuse bearing, green fruits
DRAR-1	TNAU	Monoecious, large vines, medium leaves, less branching, light green fruits
VRBT-1	ANGARU	Monoecious, more branching, medium size leaves, continuous fruiting for longer period, green fruits
MC-84	TNAU	Monoecious, medium size leaves, light green fruits, medium size fruits
DVBTG-7	IIVR	Monoecious, medium size leaves, more branching green fruits, medium size fruits,
DVBTG-5	IIVR	Monoecious, medium size leaves, more branching green fruits, medium size fruits,
PDM	IARI	Monoecious, medium size leaves, more branching green fruits, small size fruits,
Sweetgourd (<i>Momordica cochinchinensis</i>)		Dioecious, roots develop big gertubers, leaves are bigger, flowers are large, yellow, 3 small deep black or blue circular dots at the base of petal, Flowers large oblong fruits, fruits are light green to yellow in colour.
RCSG-1	ICAR Res. Complex Shilong	Dioecious, vigorous plant, light green fruits, more fruiting
RCSG-7	ICAR Res. Complex Shilong	Dioecious, medium plant, light yellow fruits, medium fruiting
RCSG-5	ICAR Res. Complex Shilong	Dioecious, vigorous plant, more spiny fruits, more fruiting, yellow fruits
RCSG-36	ICAR Res. Complex Shilong	Dioecious, vigorous plant, tough rind, less fruiting light green fruiting
DRMC-22	Kalayani (W.B.)	Dioecious, medium plant, tough rind, medium fruiting, green colour fruits
DRMC-25	Kalayani (W.B.)	Dioecious, vigorous plant, soft rind, more seed, medium fruiting, light yellow green fruits
Spinegourd (<i>Momordica dioica</i>)		Dioecious, roots develop small tubers, small leaves, flowers are small yellow colour and open in the evening, no circular dots at the base of petal, round to oval fruits, fruits are dark green to yellow in colour
VRMD-1	Varanasi	Dioecious, medium vine, more fruiting
RSR/DR-1	Mirzapur	Dioecious, long vine, medium fruiting
RSR/DR-2	Mirzapur	Dioecious, medium vine, more fruiting
RSR/DR-3	Sonbhadra	Dioecious, medium vine, less fruiting

VRMD-20-3	Satana	Dioecious, small viny, medium fruiting
VRMD-22-5	Satana	Dioecious, medium viny, less fruiting
VRMD-4	Patana	Dioecious, small viny, more fruiting
Balsampear (<i>Momordica balsamina</i>)		Monoecious, trailing vine with very thin stems and tendrils, The leaves are heart-shaped and 3-6 cm in diameter, cut into 3-4 lobes, Flowers are borne singly in the leaf axils, fruits are small having ridges throughout surface and in the end elongation take place, split open, revealing orange flesh and bright red placenta to which the seeds are attached, Seeds are small, light yellow and oval.
DRBS-1	Ranchi	Monoecious, thin and medium spread vines, bold small fruits with prominent distal end.
DRBS-2	ANGARU	Monoecious, thin and vigorous vines, medium prominent distal end, very small fruits
Sweetgourd (<i>Momordica cochinchinensis</i>)		
DRMC11	Bhagalpur (Bihar)	Dioecious, Dioecious, vigorous plant, soft rind, more seed, medium fruiting, light yellow green fruits

Field evaluation and data collection

For field evaluation, the accessions were planted following the recommended fertilizer dose and cultural practices to raise a good crop. Five plants were randomly chosen with in each accession for data recording, which was done for fifteen plant height, fruit diameter, fruit length, fruit/plant, fruit weight, seed/fruit, yield/plant, petiole length, pedicle length, node to first male flower, node to first female flower emergence and inter-nodal length.

Extraction of genomic DNA and quantification

Total genomic DNA was extracted from the fresh young leaves collected from field in the wee hours of the day following the procedure described by Doyle and Doyle (1990) and modification of Maguire et al. (1994). The DNA solution was purified and quantity of the DNA was estimated by recording OD of spectrophotometer (Gene Space-III, Hitachi, Japan) at 260 nm, and quality was checked on 0.8% agarose gel.

Polymerase chain reaction (PCR) assay and electrophoresis

Out of 120 random decamer primers from Operon Technologies, Inc., Alameda USA, a total of 50 primers were selected for RAPD analysis. Thirty-five commercially available universal ISSR primers of UBC series and 16 SSR primers reported in an earlier study (Wan et al. 2010) were used in the PCR assay. PCR reactions were performed in 96-well plates using Bio-rad thermo-cyclers system with the suitable amplification program using a total of 50 random decamer primers (Operon Technologies, USA). Each 25 µl reaction mixture contained about 50 ng of genomic DNA, 0.2 µM primer, 100 µM each of dATP, dGTP, dCTP and dTTP, 25 mM MgCl₂ and 0.5 U of *Taq* DNA polymerase along with suitable 10x buffer (10 mM Tris-Cl, pH 8.3, 50 mM KCl, 0.001% gelatin). The reaction was carried out at 94°C for 4 min as pre-denaturation step, then the reaction was cycled 40 times at 94°C for 1 min, 33°C for 1 min and extension at 72°C for 1 min. Additionally, a final cycle allowed extension at 72°C for 10 min. For ISSR and SSR assay, annealing temperature was standardized before performing the assay on complete set of accessions. Care was taken to ensure that a set of all accession to be compared were processed in the same machine.

The PCR amplified products from RAPD and ISSR assay were loaded in 1.5% agarose gel and separated by electrophoresis with 1x TAE buffer at 65 V for 1 hr and 30 minutes. The band sizes were estimated by comparing with bands of 1 Kb DNA ladder (MBI Fermentas), which was run along with the amplified products in a separate lane on the same gel. For products from SSR assay, the fragments were electrophoresed through 2.5% Metaphor agarose (FMC Bioproducts, USA) gel at a constant voltage of 90 volts for 5 hours. Fragment lengths were determined with the help of 50 bp ladder (MBI Fermentas, Germany) loaded separately in the gel along with the samples. The gels were stained with ethidium bromide (0.5 mg/ml) and visualized in a gel-documented system (Alfa-Imager 2200, Alfa-Innotech Corporation, California). The PCR amplification was repeated two times to ensure that the amplification obtained with the primers is consistent and reproducible.

Data analysis

Morphological analysis

The mean value of the data was considered for calculating the genetic distance among the accessions. DIST module of NTSYS-pc was used to achieve the same. The Nei and Li genetic distance matrix was used as input file for generating Unweighted Pair Group Method with Arithmetic Mean (UPGMA) based dendrogram using SAHN cluster module of NTSYS-pc.

Molecular analysis

PCR amplified fragments of the 24 accessions were scored as present (1) or absent (0) of bands. Only clear, unambiguous amplicons ranging from 300 bp to 3000 bp were scored. Unique presence or absence of single or group of bands was used for identification of species. Resolving power (Rp) and polymorphic information content (PIC) of primers were calculated to test the efficiency of these parameters in identifying primer that could best distinguish the cultivars. Resolving power is represented as $R_p = \sum I_b$, where $I_b = 1 - [2 \times (0.5 - p)]$, p being the proportion of cultivars containing the bands (Prevost and Wilkinson 1999). $PIC = 1 - (p_2 - q_2)$, where p_2 is proportion of accessions having an amplicon and q_2 is proportion of accessions not having the amplicon (Raina et al. 2001). PIC value of a primer was calculated by averaging the PIC values of all polymorphic fragments generated by the primer.

The genetic associations between accessions were evaluated by calculating the modified Nei and Li's coefficient $N_L = 1 - [2N_{11} / (2N_{11} + N_{10} + N_{01})]$ (Nei and Li, 1979) for pairwise comparisons based on the proportion of shared bands produced by the primers, where, N_{11} is the number of bands/alleles present in both individuals, N_{10} is the number of bands/alleles present only in individual A but absent in B and N_{01} is the number of bands/alleles present only in individual B but absent in A. Binary data based on presence (1) or absence (0) of bands was analyzed by pairwise comparison using Jaccard's coefficient (Jaccard 1908). The similarity matrix thus obtained was subjected to cluster analysis by UPGMA and dendrogram was generated to study the relatedness of the cultivars. The robustness of the nodes of the dendrogram was tested by bootstrap analysis using 1000 resamplings. These analyses were carried out using FreeTree software (Pavlicek et al. 1999). UPGMA dendrogram was drawn using Treeview (Page 1996).

Results

Morphological evaluation based on quantitative traits

The mean value of 15 quantitative traits recorded for 24 genotypes representing four different species of genus *Momordica* (Supplementary Table S1) and high morphological diversity among these accessions is not surprising. The day of flower anthesis and edible maturity is maximum (79.5 and 101.2) in DRMC-25 and minimum (32.5 and 40.5 respectively) is recorded in Gy-333. In addition, the fruit length is recorded highest (18.98) in DVBTG-5, whereas, it is minimum (4.15) in VRMD-20-3. The genotype DRMC-22 shows maximum fruit weight and seed per fruit (126.35 and 24, respectively), while it is minimum in VRMD-22-5 and DRBS-1 (10.87 and 9) and genotype RSR/DR-3 shows high seed number per fruit. The maximum yield per plant is recorded in RCSG-1 (2.255 kg) and minimum in VRMD-1 (0.365 kg). The genotype VRMD-20-3 shows highest fruits per plant (41.25) and is minimum (10.25) in DRBS-1.

Cluster analysis based on quantitative traits

Dendrogram obtained from cluster analysis of the distance matrix from the mean value of the 15 quantitative traits for the 24 genotypes (Fig. 1) groups all the accessions into two major clusters and separates at a distance of 29.00. Perusal of the dendrogram reveals two major clusters. All the accessions of *M. charantia* (Gy-323, Gy-333, DRAR1, MC84, PDM, VRBT1, DVBTG7 and DVBTG5); *M. cochinchinensis* (VRMD1, VRMD203, RSRDR3, RSRDR2, RSRDR1, VRMD225 and VRMD4) and *M. balsamina* (DRBS1 and DRBS2) constituted distinct groups within the major cluster. The second major cluster consisted of all accessions of *M. dioica*. However, RCSG-7 was distinct from other six genotypes and formed a distinct sub-cluster.

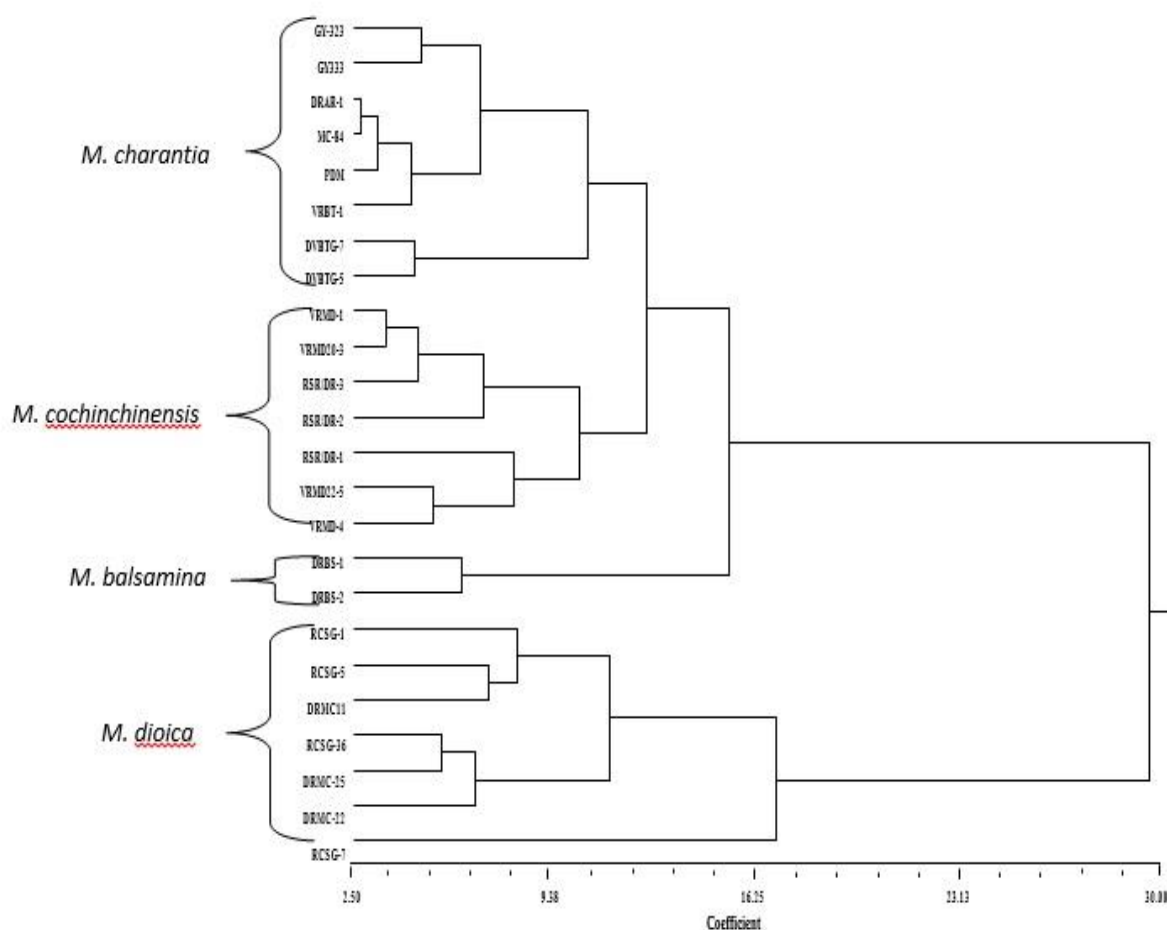


Figure 1: UPGMA dendrogram of *Momordica* accessions generated from mean values of 15 quantitative traits used for cluster analysis based on Nei and Ligenetic distance matrix.

Molecular variation and species relationships

The sequence of the RAPD, ISSR and SSR primers used for the molecular genetic fingerprinting of the four *Momordica* species and the basic bands produced by each primer, number of polymorphic bands and percentage of polymorphism produced by each primer are represented in supplementary Table S2, S3 and S4, respectively. The basic band statistics of obtained from the various set of primers in different combination is also given in table 2. The probability of chance identity of two accessions was 5.66×10^{-247} indicating the high efficiency of markers in differentiating the accessions.

Genetic diversity within and among species assessed using Nei's genetic diversity and Shannon information index is given in table 3 and supplementary Table S5.

This high level of polymorphism can easily be attributed to the accessions representing diverse species in the study. The results are not incongruent with earlier studies (Dey et al. 2006 and Behera et al. 2008) due to non-inclusion of accessions representing such diverse species. Rasu et al. (2007) included one accession of *M. dioica* as an out-group to study diversity in *M. charantia* using RAPD markers.

Table2: Analysis of banding pattern generated by the RAPD, SSR and ISSR marker and comparative analysis of different marker systems.

Component	RAPD	SSR	ISSR	RAPD +SSR	RAPD +ISSR	SSR+ISSR	RAPD+SR+ISSR
No.ofprimers	50	16	35	66	85	51	101
No.ofmarkers	264	63	273	327	537	336	600
No.ofmarkersperprimer	5.28	3.93	7.8	9.21	13.08	11.73	16.49
Polymorphicmarkers	255	61	270	316	525	331	586
Polymorphicmarkersper primer	5.10	3.81	7.71	4.80	6.18	6.50	5.80
Polymorphism(%)	96.6	95.62	99.03	96.11	97.81	97.32	97.08
Assayefficiencyindex	5.1	3.81	7.71	8.91	12.81	11.52	16.62
Markerindex	72.93	75.73	78.70	148.66	151.63	154.43	227.36
Averageresolvingpower	3.244	1.933	4.957	5.177	8.201	6.89	10.134
AveragePIC	0.755	0.792	0.794	1.547	1.549	1.586	2.341
CorrelationbetweenRP and PIC	0.053	0.521	0.083	0.574	0.136	0.604	0.657
SizeofPCRproducts(bp)	300-3000	150-500	300-3000	150-3000	300-3000	150-3000	150-3000

Table 3: Nei's genetic diversity and Shannon's information index values within the species.

Momordica sp. Name	Sample size	Number of polymorphic loci (%)	Nei's genetic diversity		Shannon's information index		Hs and Ht*	
			Mean	St.Dev	Mean	St.Dev	Mean	St.Dev
<i>M.charantia</i>	8	153 (25.37)	0.0999	0.1838	0.1454	0.2616	0.0999	0.0338
<i>M.cochinchinensis</i>	7	274 (45.44)	0.1572	0.1959	0.2357	0.2811	0.1572	0.0384
<i>M.dioica</i>	7	235 (38.97)	0.1448	0.1999	0.2136	0.2854	0.1448	0.0400
<i>M.balsamina</i>	2	108 (17.91)	0.0742	0.1590	0.1803	0.2321	0.0742	0.0253
<i>Momordica</i> sp.	24	589 (97.68)	0.3136	0.1274	0.4808	0.1622	0.3511	0.0186

Ht value is for complete set of 24 accessions depicting the diversity in the set of accessions included in the study.

The above results clearly distinguish both monoecious species from the dioecious ones and the species within both groups. Unique markers distinguishing both the groups and some species-specific markers could also be identified through the study (supplementary Table S6). Seventeen markers could be identified differentiating monoecious and dioecious species, while 85 amplicons specific to one of the four *Momordica* species were identified. These results highlight the potential of molecular markers in identifying accessions at species level and accessing the genetic diversity of accessions both within and across species.

Discussion

Morphological evaluation based on quantitative traits

Several species of genus *Momordica* occur in India. Among them *M.charantia*, *M.dioica*, *M.cochinchinensis* and *M.balsamina* are distributed across various agro-climatic zones of India. The accessions included in this study were collected from their respective zones, representing four species: two each of monoecious and dioecious nature (Table 1).

Cluster analysis based on quantitative traits

The dendrogram obtained from cluster analysis of the distinct matrix formed the mean value of fifteen quantitative traits clearly exhibits distinction among accession of four *Momordica* species represented in the study. Further, *M.dioica* forms distinct clusters while the other three species group in separate cluster forming different groups of each species within the cluster. This could probably be because the traits evaluated during the study did not involve those features of floral biology. This clustering pattern of species could not establish any correlation between the clustering patterns with the geographical distribution of the accessions. Dey et al. (2006) also could not establish any such correlation for *M.charantia* accessions. However, non-congruence of the results with previous study (Dey et al., 2006) could be due to inclusion of more number of species in the study and recording different quantitative traits for analysis. The results are in partial congruence with Bharathi et al. (2012) on relatedness of species demonstrated through crossability.

The morphological traits recorded in this study, could efficiently cluster all the accessions representing a species, these traits can be highly effective in diversity analysis of a set of germplasm and in developing core set of the germplasm representing maximum diversity across the species. Kumari et al. (2010) have advocated use of strategy of identifying highly variable descriptors for capturing maximum genetic diversity in the core set. Among *M. charantia* accessions, two gynoeious lines (Gy-323, Gy-333) recorded early days to flower anthesis and days to edible maturity indicating earliness and early harvest. These lines also exhibit maximum fruit per plant, fruit weight and yield per plant indicating their potential to develop high yielding gynoeious variety in future breeding programme (Rama et al., 2002, Dey et al., 2006, Behera et al., 2008 and Shukla et al., 2015). However, plants respond and adapt differently to selection pressures imposed by distinct agroclimatic zones (Singh et al., 1998). These results highlight the potential of quantitative traits in distinguishing accessions at species level. Rasu et al., 2007 also reported similar distinction of species using RAPD markers.

Molecular variation and species relationships

The genetic relationship among the four species is assessed based on Jaccard's similarity coefficient from the pooled data from different marker systems and dendrogram is generated using un-weighted pair group method with arithmetic averages (UPGMA). The robustness of nodes in the dendrogram is assessed by bootstrap analysis by analyzing the data with 1000 replications (Fig. 2) using FreeTree and TreeView softwares. The perusal of the dendrogram shows two distinct clusters of monoecious and dioecious species with bootstrap value of 100 at node. The first major cluster includes the accession from *M. cochinchinensis* with *M. dioica* species while the second consists of accession from *M. charantia* with *M. balsamina*. The result also supports the diversity analysis of plastids, mitochondria and nuclear DNA, where *M. dioica* and *M. cochinchinensis* are nested in a clade (Schaefer and Renner, 2010). The bootstrap values at the nodes differentiating species are high to a level of 100 for *M. cochinchinensis* and *M. dioica*, *M. charantia* and *M. balsamina*. The clustering pattern clearly divides 4 groups, each representing a single species included in this study. The bootstrap values range from 37 to 100 for *M. cochinchinensis*; 22 to 100 for *M. dioica* and 40 to 100 for *M. charantia* representing high level of diversity in accession of these species. The two accessions of *M. balsamina* included in this study were highly diverse from each other with bootstrap value of 100 for node connecting them.

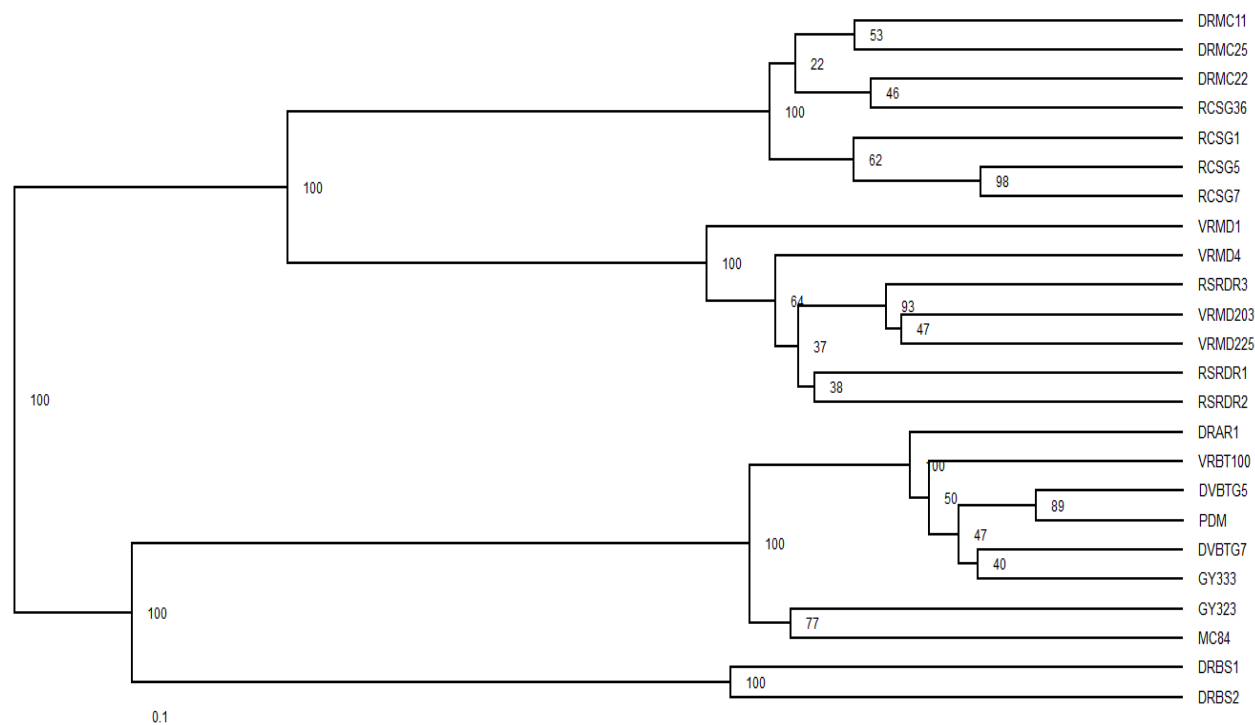


Figure 2: UPGMA dendrogram of *Momordica* accessions with bootstrap values from Jaccard's similarity coefficient obtained from molecular data analysis.

The perusal of dendrogram generated from the molecular data through 1000 replication for bootstrap reveals resolution of four distinct clusters each representing a species with intermediate to high bootstrap values at the nodes revealing high genetic heterogeneity among the accessions of the species. Similar results of high genetic diversity within species have been reported (Dey et al., 2006; Rasu et al., 2007; Behera et al., 2008; Dalam et al., 2012 and Jiet al., 2012).

Based on molecular data, 101 primers generated 600 scorable amplicons across four species, of which 586 polymorphic amplicons exhibiting 97.08% polymorphism among the samples. This high level of polymorphism can be attributed to the accessions representing diverse species in the study. The results are not incongruent with earlier studies (Dey et al., 2006; Behera et al., 2008 and Dalam et al., 2012) due to non-inclusion of accessions representing such diverse species. Rasu et al. (2007) included one accession of *M. dioica* as an out-group to study diversity in *M. charantia* using RAPD markers.

Seventeen markers could be identified differentiating monoecious and dioecious species, while 85 amplicons specific to one of the four *Momordica* species were identified. These results highlight the potential of molecular markers in identifying accessions at species level and accessing the genetic diversity of accessions both within and across species. The perusal of dendrogram generated from the molecular data through 1000 replication for bootstrap reveals resolution of four distinct clusters each representing a species with intermediate to high bootstrap values at the nodes revealing high genetic heterogeneity among the accessions of the species. Similar results of high genetic diversity within species have been reported (Dey et al., 2006; Rasu et al., 2007; Behera et al., 2008 and Jiet al., 2012).

In the present study, morphological diversity based on quantitative traits and molecular diversity showed some correspondence in the clustering pattern of accessions representing a species. All the species could be distinguished clearly, while more traits related to floral biology of the species were required to distinguish species morphologically in the study. The potentiality of gynoeious lines for earliness and yield attributes is evident and can be effectively exploited in future breeding programs. Though accessions of same species grouped together in both morphological and molecular analysis, molecular markers were more efficient than morphological parameters in not only differentiating the accessions at species level but also distinguishing monoecious and dioecious species.

Supplementary Table

Supplementary Table S1: Morphological performance of *Momordica* sp. genotypes measured for 15 quantitative traits

Accessions	Days to flower anthesis (Days)	Edible maturation	Branches /Plant	Plant height (cm)	Fruit diameter (cm)	Fruit length (cm)	Fruit /Plant	Fruit weight (g)	Seeds /Fruit	Petiole length (cm)	Pedicle length (cm)	Node to 1st male flower	Node to 1st female flower	Internodal length (cm)	Yield /Plant (kg)
<i>Mcharantia</i>															
GY-323	34.2	44.2	9	95.3	2.96	15.29	20.75	44.5	15	5.6	9.8	7.2	11.3	10.2	1.98
GY33	32.5	40.5	11	110.5	2.65	18.75	16.25	38.35	12	6.3	10.2	8.3	12.1	9.5	1.85
DRA-R-1	47.3	52.6	7	115.4	3.33	16.66	14.47	36.56	16	5.4	9.5	9.4	13.2	11.2	1.26
VRB-T-100	39.4	49.8	13	125.2	3.47	15.33	13.65	34.06	18	5.1	8.7	7.6	10.5	8.9	1.43
MC-84	41.7	52.2	10	113.4	3.12	10.25	15.23	35.55	14	3.2	9.1	8.8	12.5	7.9	1.66
DVB-TG-7	45.5	50.5	9	145.2	3.26	12.6	11.82	40.25	16	4.6	12.2	5.6	9.8	9.9	1.82

DVB TG- 5	42.3	54.1	15	15 3.6	2.62	18. 98	12. 65	30. 28	12	5.5	10. 5	7.8	10.2	11. 3	1.3 4
PDM	49.1	58.6	6	11 4.5	4.11	9.5 5	10. 52	35. 32	13	6.2	13. 9	9.4	13.7	10. 5	0.9 8
<i>M.cochinchinensis</i>															
RCS G-1	58.5	73.6	20	19 0.5	4.35	7.5 2	24. 55	112 .5 5	20	4.9	15. 6	18.3	25.4	15. 5	2.2 5
RCS G-7	55.5	70.2	18	21 0.5	3.98	6.5 3	36. 58	89. 54	19	3.7	13. 2	20.1	23.5	12. 3	1.9 8
RCS G-5	63.2	81.5	19	16 5.3	4.85	5.1 4	21. 35	96. 25	18	5.6	14. 6	21.4	27.4	15. 7	1.4 5
RCS G-36	75.8	90.5	19	14 4.5	4.65	4.9 5	18. 45	114 .5 6	16	5.9	15. 8	19.8	26.5	11. 2	2.1 0
DRM C- 22	68.9	88.6	14	13 5.6	3.25	5.1 1	16. 87	126 .3 5	24	4.5	15. 6	14.5	18.4	14. 7	1.5 6
DRM C- 25	79.5	101.2	21	15 4.8	3.88	4.6 8	21. 54	118 .4 7	19	4.7	12. 1	11.1	18.5	13. 6	1.3 4
<i>M.dioica</i>															
VRM D-1	50.5	65.4	11	11 9.8	2.64	4.5 4	32. 54	12. 54	19	3.6	13. 4	12.5	14.7	11. 2	0.3 6
RSR/ DR- 1	54.6	68.5	13	16 5.6	2.95	4.6 2	39. 21	19. 58	16	3.3	8.9	10.2	13.5	8.6	0.4 5
RSR/ DR- 2	60.2	75.6	9	11 4.5	2.54	4.3 5	18. 95	16. 54	18	3.2	9.5	9.8	12.4	9.5	0.5 4
RSR/ DR- 3	41.2	62.1	14	11 0.5	2.22	4.2 9	26. 56	14. 25	24	2.1	9.7	8.7	10.4	7.2	0.7 4
VRM D- 20-3	43.2	66.3	10	11 3.2	3.24	4.1 5	41. 25	11. 24	16	3.9	6.4	12.5	16.5	6.7	0.6 5
VRM D- 22-5	55.8	62.5	16	14 5.6	3.11	5.1 1	35. 89	10. 87	13	4.5	7.8	9.6	13.2	8.5	0.5 5
DRM C11	60.5	86.2	19	16 9.5	3.26	8.5 3	19. 55	115 .6 4	14	3.7	8.4	8.5	12.4	4.9	1.5 6
VRM D-4	55.4	68.5	12	12 9.6	2.98	3.6 5	40. 25	18. 11	11	4.9	7.8	9.1	11.2	5.5	0.4 8
<i>M.balsamina</i>															
DRB S-1	35	44.3	6	65. 8	1.98	5.8 2	10. 25	28. 51	9	3.6	6.8	6.5	8.5	5.8	0.5 8
DRB S-2	41	53.6	9	85. 3	1.85	6.8 9	13. 65	30. 55	12	2.4	9.4	5.4	6.7	3.5	0.8 5
Mean	51.3 1	65.05	12.9 2	13 3.0 7	3.22	8.4 7	22. 20	51. 27	16. 00	4.43	10. 78	10.92	14.68	9.7 4	1.2 5
S.D.	12.6 2	16.16	4.60	33. 19	0.78	5.0 2	10. 08	40. 48	3.8 0	1.16	2.8 6	4.62	5.74	3.2 5	0.6 0

Supplementary Table S2:DetailsofRAPDprimersselectedforthe study

Primers	Sequence5`to3`	AT(°C)	GC (%)	Total bands	Polymorphic bands(%)	Rp	PIC
OPAF-04	TTGCGGCTGA	33	60	8	8 (100)	5.667	0.749
OPAF-06	CCGCAGTCTG	33	70	4	3 (75)	2.167	0.651
OPAF-11	ACTGGGCCTC	34	70	4	4 (100)	2.917	0.613
OPAF-12	GACGCAGCTT	33	60	4	4 (100)	2.250	0.723
OPAF-15	CACGAACCTC	33	60	3	3 (100)	0.167	0.786
OPAG-03	TGCGGGAGTG	33	70	3	3 (100)	1.833	0.865
OPAG-04	GGAGCGTACT	33	60	3	3 (100)	2.167	0.860
OPAG-06	GGTGCCAAG	33	70	4	4 (100)	2.833	0.849
OPAG-10	GGTTGGAGAC	33	60	7	7 (100)	3.750	0.686
OPAG-11	TTACGGTGGG	34	60	4	4 (100)	2.417	0.749
OPAG-12	CTCCCAGGGT	34	70	4	4 (100)	3.667	0.706
OPAG-15	CCCACACGCA	34	70	4	4 (100)	2.500	0.715
OPC-06	GAACGGACTC	33	60	3	3 (100)	3.000	0.856
OPC-08	TGGACCGGTG	34	70	5	5 (100)	1.917	0.619
OPC-14	TGCGTGCTTG	33	60	3	3 (100)	5.167	0.760
OPC-16	CACACTCCAG	33	60	8	8 (100)	2.333	0.940
OPF-16	GGAGTACTGG	33	60	5	5 (100)	4.583	0.542
OPH-10	CCTACGTCAG	34	60	7	7 (100)	2.500	0.396
OPK-20	GTGTCGCGAG	33	70	5	5 (100)	4.167	0.843
OPL-07	AGGCGGGAAC	33	70	6	6 (100)	1.167	0.985
OPL-08	AGCAGGTGGA	33	60	5	4 (100)	4.306	0.839
OPL-14	GTGACAGGCT	33	60	6	6 (100)	3.500	0.793
OPL-16	AGGTTGCAGG	33	60	7	7 (100)	4.917	0.654
OPL-20	TGGTGGACCA	33	60	10	9 (90)	5.833	0.795
OPM-12	GGGACGTTGG	33	70	7	7 (100)	4.167	0.788
OPM-14	AGGGTCGTTC	34	60	6	6 (100)	1.083	0.237
OPN-08	ACCTCAGCTC	33	60	4	4 (100)	1.500	0.832
OPN-12	CACAGACACC	33	60	4	4 (100)	3.083	0.724
OPN-14	TCGTGCGGGT	33	70	4	4 (100)	3.250	0.870
OPO-06	CCACGGGAAG	33	70	6	6 (100)	4.167	0.792
OPO-10	TGTGCCCCGAA	33	60	6	6 (100)	1.750	0.947
OPY-03	ACAGCCTGCT	33	60	4	4 (100)	5.500	0.758
OPY-04	GGCTGCAATG	33	60	7	7 (100)	2.917	0.740
OPY-06	AAGGCTCACC	34	60	4	4 (100)	5.417	0.733
OPY-07	AGAGCCGTCA	33	60	7	7 (100)	5.250	0.770
OPY-08	AGGCAGAGCA	33	60	8	8 (100)	2.583	0.928
OPY-09	AGCAGCGCAC	34	70	5	5 (100)	2.833	0.701
OPY-14	GGTCGATGTG	33	60	4	4 (100)	2.333	0.592
OPY-15	AGTCGCCCTT	34	60	4	4 (100)	2.917	0.639
OPY-16	GGGCCAATGT	34	60	5	5 (100)	1.917	0.634
OPY-17	GACGTGGTGA	33	60	3	3 (100)	5.500	0.717
OPY-18	GTGGAGTCAG	33	60	8	8 (100)	2.750	0.857
OPZ-01	TCTGTGCCAC	33	60	5	5 (100)	4.250	0.767
OPZ-06	GTGCCGTTCA	33	60	4	1 (25)	2.583	0.590
OPZ-11	GGGTCTCGGT	33	70	6	6 (100)	3.417	0.724
OPZ-12	TCAACGGGAC	34	60	2	2 (100)	1.333	0.889
OPL-19	GAGTGGTGAC	33	60	5	5 (100)	3.917	0.841
OPZ-13	GACTAAGCCC	33	60	8	8 (100)	4.167	0.911
OPZ-14	TCGGAGGTTC	34	60	5	2 (100)	2.500	0.907
OPL-18	ACCACCCACC	33	70	11	11 (100)	5.417	0.895
				264	255 (96.59)		

Supplementary Table S3:DetailsofISSRprimersselectedforthe study

Primers	Sequence5'to3'	Tm (°C)	GC (%)	Total bands	Polymorphic band(%)	Rp	PIC
UBC880	GGAGAGGAGAGGAGA	55.6	60	14	14(100)	6.083	0.862
UBC834	AGAGAGAGAGAGAGACYT	54.5	47	10	10(100)	5.333	0.873
UBC856	ACACACACACACACACCTA	55.6	47	12	11(91.66)	5.833	0.651
UBC810	GAGAGAGAGAGAGAGAT	54.5	47	10	10(100)	4.167	0.727
UBC840	GAGAGAGAGAGAGAGAYT	57.5	47	9	9 (100)	5.333	0.881
UBC843	CTCTCTCTCTCTCTCTRA	53.2	47	9	9 (100)	4.083	0.906
UBC811	GAGAGAGAGAGAGAGAC	60.0	53	8	8 (100)	3.250	0.892
UBC888	BDBCACACACACACACA	60.0	50	12	12(100)	4.917	0.802
UBC861	ACCACCACCACCACCACC	60.0	67	10	10(100)	7.000	0.860
UBC835	AGAGAGAGAGAGAGAGYC	54.5	53	9	8 (88.88)	3.833	0.820
UBC825	ACACACACACACACACT	50.0	47	11	11(100)	3.667	0.825
UBC866	CTCCTCCTCCTCCTCCTC	54.5	67	10	10(100)	5.667	0.732
UBC841	GAGAGAGAGAGAGAGACTC	57.5	53	9	9 (100)	4.750	0.802
UBC813	CTCTCTCTCTCTCTCTT	54.5	47	8	8 (100)	5.833	0.883
UBC887	DVDTCTCTCTCTCTCTC	60.0	50	7	6 (85.71)	4.833	0.794
UBC822	TCTCTCTCTCTCTCTCA	54.5	47	9	9 (100)	4.917	0.824
UBC892	TAGATCTGATATCTGAATTCCC	53.2	36	9	9 (100)	1.667	0.534
UBC855	ACACACACACACACACCTT	57.5	47	9	9 (100)	6.417	0.865
UBC884	HBHAGAGAGAGAGAGAG	53.2	50	5	5 (100)	2.833	0.828
UBC848	CACACACACACACACAAGG	57.5	53	7	7 (100)	9.500	0.731
UBC842	GAGAGAGAGAGAGAGAYG	60.0	53	8	8 (100)	7.083	0.783
UBC890	VHVGTTGTGTGTGTGTGT	57.5	50	8	8 (100)	4.083	0.844
UBC824	TCTCTCTCTCTCTCTCG	54.5	53	6	6 (100)	5.083	0.787
UBC853	TCTCTCTCTCTCTCTCRT	55.6	47	7	7 (100)	6.250	0.781
BC854	TCTCTCTCTCTCTCTCAGG	57.5	53	6	6 (100)	3.500	0.869
UBC891	HVHTGTGTGTGTGTGTGTG	57.5	50	5	5 (100)	5.333	0.768
UBC809	AGAGAGAGAGAGAGAGAGG	57.5	53	5	5 (100)	5.250	0.712
UBC815	CTCTCTCTCTCTCTCTG	54.5	53	7	7 (100)	6.583	0.830
UBC836	AGAGAGAGAGAGAGACYA	54.5	47	7	7 (100)	5.500	0.867
BC850	GTGTGTGTGTGTGTGTCTC	60.0	53	5	5 (100)	4.500	0.720
UBC812	GAGAGAGAGAGAGAGAA	50.0	47	3	3 (100)	2.583	0.607
UBC814	CTCTCTCTCTCTCTCTA	54.5	47	6	6 (100)	5.417	0.562
UBC886	VDVCTCTCTCTCTCTCT	54.5	50	5	5 (100)	5.750	0.800
UBC844	CTCTCTCTCTCTCTCTRG	50.0	53	4	4 (100)	2.417	0.898
UBC895	AGAGTTGGTAGCTCTTGATC	57.5	45	4	4 (100)	4.250	0.898

R-Wobble(A+G),H-Wobble(A+C+T),Y-Wobble(C+T),Y~Q-3'Wobble(C+T),D-Wobble (A+G+T),V-Wobble(A+C+G),B-Wobble(C+G+T)

Supplementary Table S4: DetailsofSSRprimers(Wangetal.,2010)selectedforthe study

Primers	PrimerSequence(5'.....3')	Tm (°C)	GC (%)	Total bands	Polymorphic bands(%)	Rp	PIC
S24	F:GCTCTGCGTTTCATTCTTCA R:TGAACCCTCAGACTCAAATCTC	60	46	5	4 (80)	3.250	0.961
N6	F:GGGAATTCTCAAAGAGCCAGA R:TGGCACACTCTGCATGAAAT	57	46	4	4 (100)	0.750	0.786
N24	F:GCTCTGCGTTTCATTCTTCA R:TGAACCCTCAGACTCAAATCTC	57	46	6	6 (100)	1.333	0.581
S13	F:TTGGTTGTGGTGCTGAGTTC R:GATGTAGGGGTTGGGTTGAT	57	50	5	5 (100)	3.917	0.864
	F:GTCTTCCAGGTTGGGAACAG	59	53	10	10(100)	0.653	0.859

N1	R: ATCTGGTTCCTCGGGAGATT						
S15	F:GGGTAGTGGAATGATGGGTT R:TAGTGTTTTCTGTGAGGGAGG	57	50	5	5 (100)	1.917	0.886
S33	F: ATTTAGTGGGGCGGGTAGT R:TGGATGAGCATGTTAGGGATC	54	50	4	4 (100)	2.500	0.701
N9	F: ATCCATCCCCACAAGTTGAA R:CCATAAGGATATGTTTGCATGG	60	43	2	1 (50)	0.833	0.413
S18	F:GGGTAGTGGAATGATGGGTT R:TAGTGTTTTCTGTGAGGGAGG	58	50	3	3 (100)	1.667	0.825
S32	F:CTAAATCACGCAAACCCATC R:GAGCAAAAGACTGAGGAAAAC	54	43	4	4 (100)	3.250	0.891
S9	F:TTCCTTCTACAGATCACTCC R:CCACCAAATTCAAGAACCAC	57	48	2	2 (100)	1.500	0.734
N5	F:CGTCGCTCTCACAAGAGATAAG R:TTTGGTGGAATCCCCTATT	59	45	2	2 (100)	1.333	0.722
S20	F:CCCCTTCTAATCACAACCAA R:GGCCTAATTTCTGCCCTTT	58	46	3	3 (100)	1.917	0.886
N12	F:CAGAGGGGTGGTTCCTCTTT R:CCACATGGATGATCGAGAGA	59	53	3	3 (100)	2.250	0.920
S12	F:CAGAGGGGTGGTTCCTCTTT R:CCACATGGATGATCGAGAGA	57	53	3	3 (100)	2.083	0.814
S26	F:GAACGCCCTGTGACTTTAGC R:TTTCGTCTTCCAATGAGCC	58	51	2	2 (100)	2.750	0.829
				63	61(95.62)	1.993	0.792

Supplementary Table S5: Nei's coefficient for genetic diversity (above diagonal) and Nei and Licoefficient for genetic distance (below diagonal) among the species.

	<i>M.charantia</i>	<i>M.cochinchinensis</i>	<i>M.dioica</i>	<i>M.balsamina</i>
<i>M.charantia</i>	---	0.543	0.5728	0.7085
<i>M.cochinchinensis</i>	0.6107	---	0.7362	0.6924
<i>M.dioica</i>	0.5572	0.3111	---	0.7062
<i>M.balsamina</i>	0.3446	0.3675	0.3479	---

Supplementary Table S6: Primers with species-specific bands with their molecular size

Uniquemarkers	<i>Momordica</i> species identified with molecular size (bp)
RAPD Primers	
OPM-14	<i>M.dioica</i> (650), <i>M.cochinchinensis</i> (900)
OPL-07	<i>M.dioica</i> (950)
OPL-08	<i>M.cochinchinensis</i> (850)
OPAF-12	<i>M.charantia</i> (400)
OPH-10	<i>M.dioica</i> (750), <i>M.charantia</i> (400)
OPAG-012	<i>M.charantia</i> (500), <i>M.cochinchinensis</i> (950)
OPAF-04	<i>M.cochinchinensis</i> (890)
OPAF-11	<i>M.charantia</i> (1250), <i>M.dioica</i> (350)
OPC-16	<i>M.charantia</i> (900)
OPC-08	<i>M.charantia</i> (850)
OPZ-11	<i>M.balsamina</i> (1300)
SSR Primers	
N01	<i>M.charantia</i> (200, 250)
N24	<i>M.charantia</i> (320)
S13	<i>M.charantia</i> and <i>M.cochinchinensis</i> (220)
S32	<i>M.charantia</i> (500)
S18	<i>M.dioica</i> (240)

S33	<i>M.dioica</i> (425)
S33	<i>M.charantia</i> and <i>M.balsamina</i> (380)
S33	<i>M.cochinchinensis</i> and <i>M.dioica</i> (250)
ISSRMarkers	
UBC866	<i>M.charantia</i> (700,1300), <i>M.dioica</i> (800), <i>M.charantia</i> and <i>M.cochinchinensis</i> (550)
UBC822	<i>M.charantia</i> (700,900,1400), <i>M.dioica</i> (1100), <i>M.cochinchinensis</i> (330,400,800), <i>M.charantia</i> and <i>M.dioica</i> (550)
UBC834	<i>M.dioica</i> (1500), <i>M.charantia</i> (260,275,500), <i>M.cochinchinensis</i> (250)
UBC887	<i>M.charantia</i> (1500), <i>M.dioica</i> (700,750,1200), <i>M.cochinchinensis</i> (700), <i>M.dioica</i> and <i>M.balsamina</i> (500)
UBC842	<i>M.charantia</i> (500,680,1200,1500), <i>M.dioica</i> (500,750), <i>M.cochinchinensis</i> (700), <i>M.charantia</i> and <i>M.balsamina</i> (480)
UBC836	<i>M.dioica</i> (1000), <i>M.cochinchinensis</i> (540,800), <i>M.dioica</i> and <i>M.balsamina</i> (275,500), <i>M.balsamina</i> (260)
UBC886	<i>M.charantia</i> and <i>M.balsamina</i> (1000), <i>M.dioica</i> (750,900), <i>M.cochinchinensis</i> (600), <i>M.charantia</i> (500)
UBC888	<i>M.charantia</i> (2400), <i>M.cochinchinensis</i> (250,2000), <i>M.dioica</i> (250,1000,1700)
UBC890	<i>M.cochinchinensis</i> (500,1500), <i>M.dioica</i> (660), <i>M.balsamina</i> (430)
UBC840	<i>M.charantia</i> and <i>M.cochinchinensis</i> (250)
UBC855	<i>M.charantia</i> (2500), <i>M.cochinchinensis</i> (575)
UBC841	<i>M.cochinchinensis</i> (280), <i>M.dioica</i> (250)
UBC848	<i>M.charantia</i> and <i>M.dioica</i> (1500), <i>M.cochinchinensis</i> and <i>M.dioica</i> (900), <i>M.dioica</i> (500)
UBC812	<i>M.cochinchinensis</i> and <i>M.balsamina</i> (300)
UBC825	<i>M.balsamina</i> (590,1270), <i>M.charantia</i> (530,1100), <i>M.cochinchinensis</i> and <i>M.dioica</i> (645), <i>M.dioica</i> (600)
UBC861	<i>M.dioica</i> (1400), <i>M.cochinchinensis</i> (300,400,750,900), <i>M.dioica</i> (300,335)
UBC892	<i>M.cochinchinensis</i> and <i>M.dioica</i> (300,1600)
UBC843	<i>M.dioica</i> (800), <i>M.cochinchinensis</i> and <i>M.dioica</i> (400)
UBC884	<i>M.charantia</i> (200,600), <i>M.charantia</i> and <i>M.dioica</i> (530)
UBC855	<i>M.cochinchinensis</i> and <i>M.dioica</i> (200,700,750), <i>M.charantia</i> (450), <i>M.cochinchinensis</i> (300)
UBC891	<i>M.charantia</i> (250)
UBC815	<i>M.dioica</i> (800,1300), <i>M.charantia</i> (900,1200)
UBC809	<i>M.cochinchinensis</i> (600), <i>M.charantia</i> , <i>M.dioica</i> and <i>M.balsamina</i> (425,500), <i>M.dioica</i> (300)
UBC891	<i>M.cochinchinensis</i> and <i>M.dioica</i> (750), <i>M.cochinchinensis</i> (300)
UBC844	<i>M.charantia</i> (900), <i>M.cochinchinensis</i> and <i>M.dioica</i> (500)
UBC880	<i>M.dioica</i> (1300), <i>M.charantia</i> (335,750), <i>M.cochinchinensis</i> (573)
UBC856	<i>M.cochinchinensis</i> (300,1300), <i>M.dioica</i> (250), <i>M.charantia</i> and <i>M.balsamina</i> (240)
UBC853	<i>M.charantia</i> and <i>M.balsamina</i> (1500), <i>M.dioica</i> (750,1100), <i>M.cochinchinensis</i> (500)
UBC813	<i>M.dioica</i> (1300), <i>M.cochinchinensis</i> (1000), <i>M.charantia</i> (400)
UBC810	<i>M.cochinchinensis</i> (1200), <i>M.charantia</i> (250,275,730), <i>M.balsamina</i> (250)
UBC814	<i>M.cochinchinensis</i> and <i>M.dioica</i> (400)
UBC835	<i>M.charantia</i> and <i>M.balsamina</i> (200,1000), <i>M.charantia</i> (600), <i>M.dioica</i> (250)
UBC811	<i>M.charantia</i> (1600), <i>M.cochinchinensis</i> and <i>M.dioica</i> (1500), <i>M.charantia</i> and <i>M.dioica</i> (300,635), <i>M.dioica</i> (700)

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Conflict of Interest

The authors declared no conflict of interest.

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RESEARCH ARTICLE

Z-PLASTY TRANSPOSITION FLAPS A NEW TECHNIQUE IN CLOSURE OF SKIN DEFECTS AFTER LUMBOSACRAL MENINGOMYELOCELE RESECTION IN PEDIATRICS.

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Abstract

Background: Different closure techniques are available after lumbosacral meningomyelocele resection (MMC) resection for large defects reconstruction, but wound healing and tension-free closure in the midline remain major considerations. In this study we use Z-plasty technique in closure of MMC defects > 5 cm. Twenty newborns with large lumbosacral MMC were enrolled in this study. The mean age was 57.45 ± 59.8 days. Complete healing of the skin occurred within two weeks in 14 patients (70%). Superficial wound infection was seen in four patients (20%). Necrosis of the edges of the flap was showed in two patients (10%). No mortality or complete wound dehiscence were observed in this study. Almost skin wounds healed without any major complications.

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Introduction:-

Meningomyelocele (MMC) is the most common significant birth defect involving the spine ranging from less than one case per 1000 live births in North America to 7.7% of live births born in United Arab Emirates. MMC is associated with several CNS anomalies including hydrocephalus, varying degrees of caudal displacement of hindbrain and cerebellar hypoplasia.⁽¹⁾ Because MMC defects vary in size, shape and location no single procedure applies to all. A large number of techniques have been described for closing the back defects occurring after excision of the MMC sac, but new studies focus more on simple, reliable and versatile alternative. The neurosurgical repair of this defect is performed immediately after birth for those lesions with rupture dural sac and external CSF leakage and can be delayed for several days without additional morbidity or mortality.⁽²⁾

The goal of surgical back closure is to seal the spinal cord with multiple tissue layers, to guard against ascending infection and to prevent CSF leakage with preserving the neurological function and untethering of the spinal cord.⁽³⁾

The plastic surgeon draws and designs the Z plasty flap to close the skin defects without tension.⁽⁴⁾

The aim of this study:-

The aim of this study is to assess the Z-plasty transposition flap in closure of skin defects after lumbosacral (MMC) resection in pediatrics.

Patients and methods:-

Twenty (20) newborns with large MMC were enrolled in this study, presented to the neurosurgical department Damanhour Teaching Hospital with the collaboration of surgical unit, during the period of 1 year from Dec 2009 to Nov 2010. After obtaining informed consent from their parents, our initial step was a careful physical examination conducted by a pediatrician, neurosurgeon and plastic surgeon to reveal any associated anomalies including cardiac, renal and other cranial defects. Preoperative anesthetic consultation was done with the request of blood grouping with cross matching of one unit of packed RBC for all patients; three newborns only needed intraoperative blood transfusion. Premature babies and newborns with low birth weight, anemia, and upper respiratory tract infection were excluded from this study. Over hydrocephalus was associated in four newborns and CSF diversion was done in the same sitting for those cases.

This study included only large-sized MMC with an average diameter more than 5 cm which was impossible to close these defects by using simple closure surgery without tension especially in the presence of kyphosis.

The mean age of the infants was 57.45 ± 59.8 days (approximately 2 months), 11 (55%) were males while 9 (45%) were females. The mean operative time for flap elevation and closure of skin was 40 minutes.

The following steps were done:-

1. A thorough history was taken and clinical examination conducted by pediatrician and neurosurgeon was done.
2. MRI lumbosacral spine was done for those cases with motor defects and CT brain was done for all newborn with large skull diameter.
3. Blood laboratory tests including CBC, bleeding time, clotting time, prothrombin activity, serum albumin were done for all patients.

Surgical technique:-

Under general anesthesia, the newborn was placed in prone position with rolls under the chest and pelvis to allow abdomen to hang free.

Measurement of the expected raw area (skin loss) and designing of the Z-plasty flap and its landmark on the skin was done (Fig. 1). Also, decompression of bladder was done prior to placement in this position. The neurosurgical repair started with transverse or longitudinal elliptical skin incision extending through the subcutaneous tissue. The dural sac is undermined and reflected medially on each side. It is closed in a water-tight fashion using 4/0 non-absorbable suture the posterior layer of thoracolumbar fascia is then closed in a separate layer to reinforce the dural tube. The plastic surgeon made the defect as a rhombus shape placed vertically over the midline, equilateral Z-plasty were elevated at the sides of the rhombus and transposed across the defect. The skin lines of closure were chosen to be away from the dural suture, approximated and secured without tension.

Meticulous hemostasis with bipolar cautery was done, so suction drainage was not used in any patient.

Postoperative care:-

Daily dressing was done for all newborns to guard against fecal and urine contamination and also to detect any early signs of surgical wound complications including infection, subcutaneous seroma, CSF leakage, edge necrosis and wound dehiscence. All patients were discharged after removing the surgical sutures to be followed every week in the 1st month postoperative to secure and document the sound healing of the wound.

Results:-

The closure of the skin defect was possible in all patients, as a tension-free and in one stage. Figure 2, showed a newborn's back two weeks postoperative, the wound has healed without any major complications. No mortality has occurred among newborn in this study. Sound healing has occurred in 14 patients (70%), while total post-operative complications occurred in (30%) of patients. CFS leakage was observed in three patients (15%). Superficial wound infection was seen in four patients (20%). As regard partial wound dehiscence it was detected in two patients (10%). Complete wound dehiscence didn't occur in any patient. Necrosis of the edges of the flap was showed in two patients (10%). Subcutaneous seroma was found in five patients (25%), and it was aspirated under local anesthesia without impact on the flap vascularity. Necrosis of the whole flap did not happen in any patients. Table 2, showed the results of the Z-plasty technique after 1 month follow up in 20 patients with lumbosacral MMC.

Table 1: - Postoperative results of the Z-plasty technique in 20 new born with lumbosacral MMC.

Factor	Number of patients	%
Sound healing	14	70
CSF leak	3	15
Superficial wound infection	4	20
Partial wound dehiscence	2	10
Necrosis of edges of the flap	2	10
Subcutaneous seroma	5	25
Complete necrosis of the flap	-	-
Complete wound dehiscence	-	-
Mortality	-	-

**Fig. (1)****Fig. (2)**

Fig. 1:- showed designing and drawing of the flap and Fig. 2 the sound healing of the flap with sound healing two weeks postoperative.

Discussion:-

Unfortunately, MMC repair by skin direct closure cannot accomplish in nearly 25% of the neonates with large defects.⁽⁵⁾ So, for those with large defects, definitive operative management should be performed by a combined neurosurgical and plastic surgical team as to provide reconstruction and protection of the exposed spinal cord, elimination of any CSF leakage and stable soft tissue coverage.⁽⁶⁾

The suitable technique and skill of the managing of MMC defect have provoked substantial debate.⁽⁷⁾ Myriad plastic surgical techniques have been developed to address these issues.⁽⁸⁾ The literature suggests a number of convincing, effective procedures such as skin, subcutaneous and most frequently muscle flaps, bilateral sliding muscle flaps, and split thickness skin grafts.^(9,10,11) The decision to use a specific technique is dependent on the size of the defect, vascular supply and to some extent to the surgeon experience.⁽¹²⁾

The goal of using Z-plasty technique was not to replace the other techniques but to offer an effective option in the treatment of MMC with large cutaneous defects. Among the different methods, Z-plasty proved in our study to be one of the good options to cover large MMC defects. No major complications were encountered. In a study done by Gumus et al.,⁽¹³⁾ they found that Z-plasty flap is a safe alternative, rapid easy dissection, and short operation time with minimal blood loss. Also as regard this technique a low morbidity and mortality and satisfactory outcome have reported by Ozcelik et al.⁽¹⁴⁾

Conclusion:-

Surgical repair of large myelomeningocele (> 5 cm diameter) may be a challenging problem. The advantages of our method are simplicity, reliability and coverage of the defect with well-vascularized flaps, almost nil major flap complications and no donor site morbidity. Primary closure was possible in all patients. This procedure also provides a thick heavy duty skin and subcutaneous tissues over the dural sac; the flap can be taken 1:3 in diameter without compromising the blood circulation of the flap. No suture lines overlie the cord closure and no contour deviation occurred. The result of this technique is satisfactory as regard blood loss and time elapsed.

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RESEARCH ARTICLE

GOING GREEN IN LITERATURE: AN INSIGHT INTO NATURE AND ECOLOGY THROUGH LITERATURE.

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Radiotherapy, Collagenase gene, cancer and Swiss mice.

Abstract

Present paper focuses on relationship between human being and nature, man being the only literary creature on earth has personified and revered the nature in its varied form, and the ecological wisdom possessed by him speaks of divine status of Mother Nature in ancient times, due to advent of technologies and ignorance, man has called upon the wrath and fury of mother nature, present paper weaves itself around the journey of human being from ecological wisdom to ecological apocalypse.

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Introduction:-

Man is a part of ecosystem of earth and relies on it for its survival just like plants and animals but the superiority of man lies in the fact that he is the only literary creature of cosmos. He is unable to participate in small natural processes around the earth but with his brain he can produce enormous literature of different kind and this unique literary talent has kept him above all earthly beings. But his superiority is not independent he is closely related with other living beings for survival, this study we now refer to as ecology it studies interrelationship of living beings to one another and with their surroundings. Human ecology is the study of mankind and its response towards nature which covers all the earthly creatures. We cannot bind ecology with specific subject its appeal is universal its wisdom transcends the boundaries of various fields and manifests in any form. Ecological wisdom appeals to different cultures it manifests in romantic poetry pioneered by William Wordsworth, John Keats and PB Shelly, in pre-classical Greeks, in Zen Buddhism and many paintings.

Ecology has played a distinctive role and influenced Indian philosophy, mythology and literature. A notable feature of Indian thought towards universe is that all creation comes under one fundamental structure that is one universal spirit holds all creatures of this universe and everything has been originated from it. This is the central idea of most of the philosophical discourses of Indian literature, be it Vedas, Mahabharata, Ramayana. Apart from this, this philosophical treatise also teaches us compassion towards all life forms. The doctrine of Ahimsa and compassion towards all living being has been praised and propagated by Buddha in his teachings and sermons. It also held esteemed position in other religions. We can clearly realize that early man and nature were co-existed together in most comfortable way. Indian sages offered their reverences to the supreme which they believe existed in form of water, mountain, fire, trees etc. They revered them consider them as gods and unknowingly giving a way to sustainable development in a true sense. William Wordsworth an English Romantic poet has called the nature as 'Wisdom and spirit of Universe' he gave various stages of his love towards nature in many of his poems like *Tintern*

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Abbey, Exursion and The Prelude. Wordsworth two famous poems *The Tables Turned* and *Education of Nature* are wonderful example of his love for nature. And here I quote the very famous lines from *The Tables Turned*.

One Impulse from a vernal wood
May teach you more of man;
Of moral evil and of good
Than all the sages can.

According to the teachings of 'Atharva Veda' Nature is sacred it has to be understand as a friend, revered as mother and nurtured as a beloved child. Man is not separate from earth and universe the idea of dependent co-arising is the central theme of Buddhism, it propagates the idea of interconnectedness it suggests we are all interwoven thread in the complex structures of life. All things are interconnected according to Native American chief Seattle who says 'The earth is our mother. Whatever befalls the earth befalls the sons of earth. If man spits upon the ground they spits upon themselves. All the things are connected like blood which unites one family'. But unfortunately the human beings in this age became deaf and dumb and fail to understand such noble emotions. The brain through which human beings has mastered all other earthly creatures is now provoking him to evaluate plan and execute such manoeuvres which are disastrous for nature and natural existence. He is responsible for destruction and in turn invites the fury of nature. He is deeply engrossed in all the worldly pleasures and forgot about the management and utility of nature in a proper manner. A brilliant example of man's merciless destruction of nature is visible in Ted Hughes poem *Revenge Fable*.

There was a person
Could not get rid of his mother
As if he were her topmost twig
So he pounded and hacked at her
With numbers and equations and laws
He investigated incriminated
And penalized her, like Tolstoy,
Forbidding, screaming and condemning,
Going for her with a knife,
Obliterating her with disgusts
Bulldozers and detergents
Requisition and central heating
Rifles and whiskey and bored sleep
With all her babes in her arms, in ghostly weepings,
She died.

The need of hour is to practice unconditional love towards nature and this will help to change the world into a better place. The power and capacity to being in love is limitless and to fall in love with nature is a true blessing because nature always gives you back treasures of joy, bliss and happiness. Alice walker portrays this sentiment in her novel *The Color Purple*.

My first step away from the old white man was tree. Then air, then birds, then other people but one day when I was sitting quite and feeling like a motherless child, which I was, it come to me that feeling of being part of everything, not separate at all. I knew if I cut a tree, my arm would bleed. And I laughed and I cried and I ran all around the house. I knew just what it was. In fact when it happens you can't miss it... (167).

We should cultivate such emotions to feel the pain of nature. But the modern technologies and mechanization has blinded us and right from elite class every common man has held in its grip. And the greed to get more comfort has led to the serious damage of our biological environment leading to serious catastrophic consequences. Literary studies have played major role in dealing with these difficult issues related to environment. It was in mid eighties environmental literary studies came in existence. It was Joseph Meeker who introduced the term 'Literary Ecology' in the *Comedy of Survival* which is an attempt to analyze the role played by literature in ecology. It basically represents the man's relationship and responses towards nature. Literary theory in general examines relation between writer text with the culture and world. Ecology analyzes the interconnectedness sentiment therefore both ecology and literature emphasize on relationships interconnectedness. The sentiment of inter textuality in literature is similar with the interconnected sentiment in the ecology. And one of the major responsibilities of literature is to

inculcate the sense of compassion in human beings towards nature. Eco criticism refers to the study of relationship between literature and environment it shows how human culture is connected to the environment. Both literary theory and ecology criticize the stance of independent and separate meaning.

The love for the nature can be seen in the writings of many classical poets like Virgil, Horace, Theocritus they all found nature soothing comforting and peaceful. Milton found nature as a refreshing a panacea for city dweller. The rise of romantic age has changed the perspective of nature in the human eyes. Romantic poets and authors of this age personified the nature in various forms and revered its beauty sometimes as a goddess sometimes as a mother, child, lover etc. They redefine nature in new dimensions. Some contemporary poets describes the beauty of natural landscapes some yearns for the lost earthly paradise so these writings plays important role in establishing environmental forums and can help in conservation of nature by arousing human emotions and compelling them to take steps. "Lamenting on mankind Emerson a noted author in his essay *Nature* says 'We are as much strangers in nature as we are aliens from God. We do not understand the notes of the birds. The fox and the deer run away from us; the bear and the tiger rend us' (74).

The chain which bind us together that inter-connectedness between various creature men has lost the vision to appreciate it. And modern man does not find himself in harmony with nature but he found himself as an outside force meant to dominate and conquer the nature. The modern man has lost the sense of unity, oneness with the nature that has lost its meaning. The man's lost innocence has proved disastrous for nature and he is spreading on the globe destroying and burning it. The time demands immediate full stop on this mad dance of fury and exploitation of nature by man. A well planned methodology is needed for the preservation of nature otherwise within no time man will see the fury of nature casting its doom on civilization which he proud on the day of Apocalypse is not far away.

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RESEARCH ARTICLE

ACUTE EOSINOPHILIC APPENDICITIS PRESENTING BY LOWER GASTRO-INTESTINAL BLEEDING.

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Appendicitis, Eosinophil, Lower gastrointestinal bleeding.

Abstract

Acute eosinophilic appendicitis is a rare type of appendicitis where the inflamed appendix infiltrates by eosinophil rather than neutrophil in the muscularis propria accompanied by edema. Associated with type 1 hypersensitivity that may affect any part of the gastrointestinal track. Acute eosinophilic appendicitis similar to the classical appendicitis in symptoms. However, massive lower gastrointestinal bleeding with appendiceal origin considers a rare presentation of acute appendicitis. Different modalities are needed to reach the definitive diagnosis and identification of the bleeding site. We report a 51-year-old with appendiceal hemorrhage due to acute eosinophilic appendicitis. The Diagnosis was made by a multidisciplinary approach. The patient underwent upper and lower endoscopy, which was inconclusive. Therefore a computerized tomography scan was done to the patient to identify the source of bleeding. A right hemicolectomy was performed and the histopathology showed an acutely inflamed appendix with eosinophilic infiltration.

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Introduction:-

Acute appendicitis is one of the most common causes of acute abdomen that requires surgical intervention [1]. The lifetime risk of developing acute appendicitis in Western populations is 7 % [4]. It is a clinical-based condition where there is right iliac fossa pain and tenderness, vomiting, fever and high white blood cell (WBC) count [2,3]. The typical histopathologic finding of acute appendicitis is neutrophil infiltration of muscularis propria [2]. Eosinophil infiltration of the muscularis is considered a rare histopathologic cause of acute appendicitis [4]. Acute eosinophilic appendicitis (AEA) is still a not well-studied disease, and few cases were reported [5].

Lower Gastrointestinal Bleeding (LGIB) caused by acute appendicitis is extremely rare [6]. In this case report, the patient presented by severe LGIB as the main manifestation caused by acute eosinophilic appendicitis. The objective of this case report is to document two rare entities: Acute Eosinophilic Appendicitis and severe LGIB caused by appendicitis.

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Case Presentation:-

A 51-year-old Saudi male patient known case of hypothyroidism on Levothyroxine (250 mcg/day), He presented to the emergency department of a general hospital due to several attacks of diarrhea, passing dark colored stool, for one day. The last attack of diarrhea ended by passing bright red blood per rectum and was associated with loss of consciousness. In the hospital, he was vitally stable and afebrile; the abdomen was soft and lax with positive bowel sounds. The patient denied any history of fever, nausea, vomiting or abdominal pain.

The initial lab work revealed a hemoglobin (Hb) level of 12 g/dl, white blood cell (WBC) count of 15.3 and platelet count 265,000. The chemistry including liver function tests (LFTs) were within normal.

While in the emergency department, the patient had two episodes of per rectal bleeding. Repeated lab work revealed a severe drop of Hb level to 7.5 g/dl. The patient was shifted to ICU, received two units of Packed RBCs. Urgent upper GI endoscopy was performed without any abnormality detected. A lower GI endoscopy confirmed the presence of fresh blood in the colon; however, the source of the bleeding was not determined.

The patient was transferred to our center for further evaluation by CT angiography of the abdomen and pelvis (GI bleeding protocol) and for possible endovascular embolization.

The CT demonstrated no active bleeding from both upper and lower GI. However; the appendix was increased in diameter (12 mm) with mucosal hyper enhancement, diffuse mural thickening, peri-appendicular fat stranding (Fig. 1 and 2) and multiple ileocecal lymph nodes (Fig. 3). Near the tip of the cecum/the origin of the appendix, focal mural thickening was also noted (Fig. 4 and 5).

Another lower GI endoscopy was done in order to perform an endoscopic biopsy. The performing gastroenterologist was able to reach the ascending colon but was unable to reach the cecum. Again, blood clots were demonstrated in the ascending colon.

General surgery consultation was requested. Due to the focal thickening in the tip of the cecum, there was a serious concern for malignancy, which would explain the LGIB. Virtual CT colonoscopy was performed to further evaluate the cecum, as this was not fully evaluated by the conventional colonoscopy. The findings were more or less the same as the first CT study. A decision was made to perform a right hemicolectomy due to possible underlying malignancy. The histopathology of the specimen revealed an acutely inflamed appendix with transmural eosinophil cell infiltration. There were abundant luminal exudates and widespread mucosal ulceration. The included lymph nodes showed only reactive hyperplasia (Fig. 6).

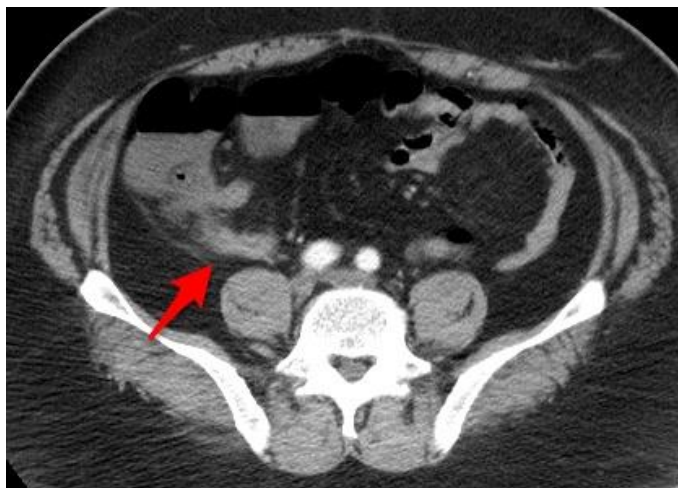


Fig (1):-CT abdomen and Pelvis, axial view, arterial phase: Circumferential mural thickening of the appendix with mucosal hyperenhancement (red arrow). There is also mild periappendicular fat stranding as shown. The maximum diameter of the appendix is 12 mm (measurement not shown).

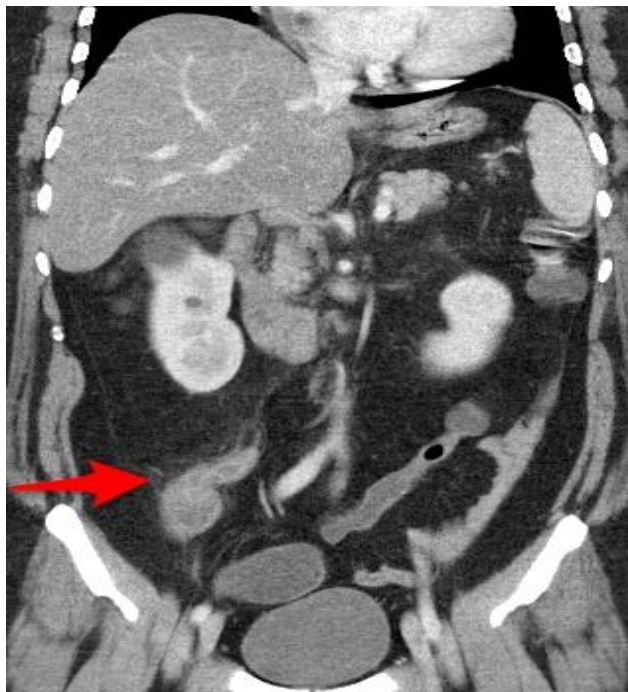


Fig (2):- CT abdomen and Pelvis, Coronal Reformats, portal venous phase: Circumferential mural thickening of the appendix with mucosal hyperenhancement (red arrow). There is also mild peri-appendicular fat stranding as shown. The maximum diameter of the appendix is 12 mm (measurement not shown).

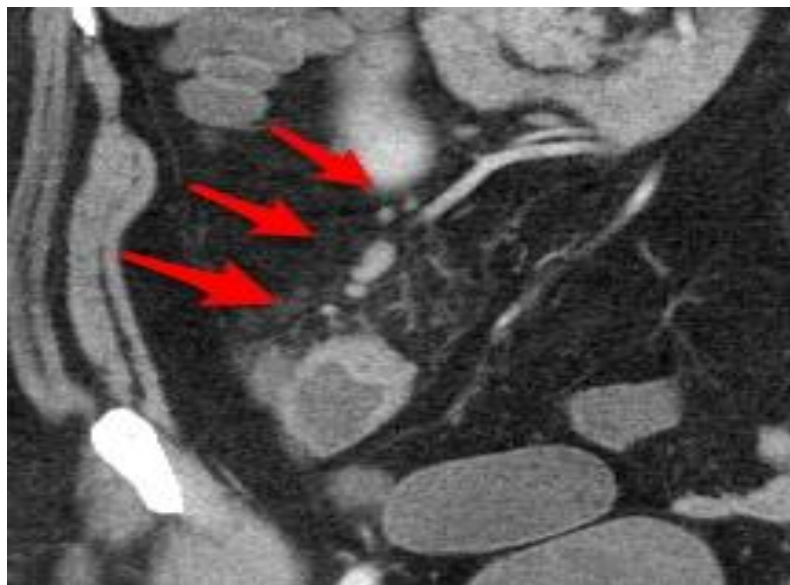


Fig (3):- CT abdomen and Pelvis, Coronal Reformats, portal venous phase, Focused on Ileocecal region: multiple small regional lymph nodes with meso-appendicular fat stranding (multiple red arrows)

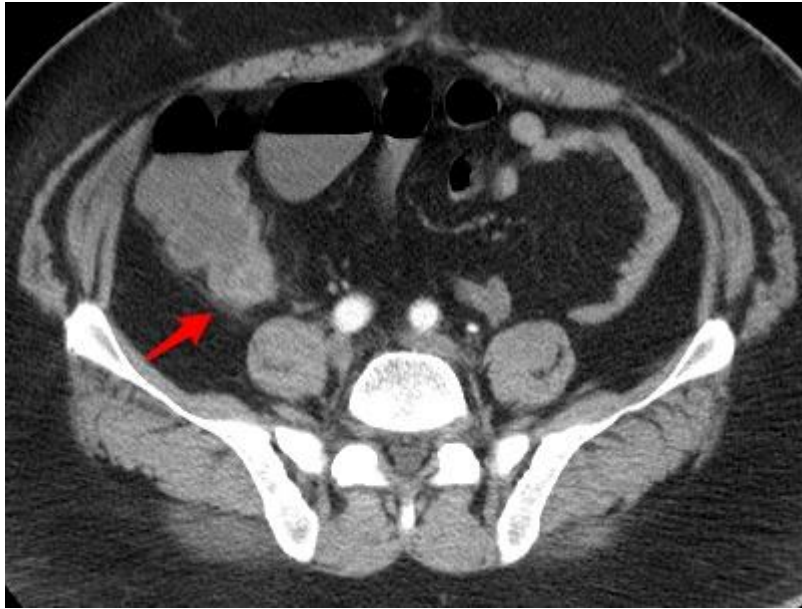


Fig (4):- CT abdomen and Pelvis, axial view, arterial phase: Focal mural thickening at the tip of the cecum/root of the appendix with mucosal hyperenhancement (red arrow).



Fig (5):- CT abdomen and Pelvis, Coronal Reformat, portal venous phase: Focal mural thickening at the tip of the cecum/root of the appendix with mucosal hyper enhancement (red arrow)

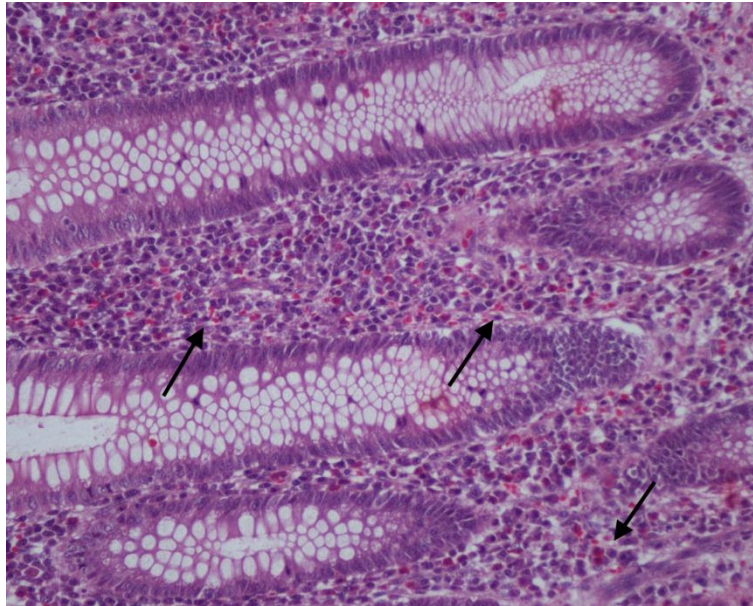


Fig (6):- Dense mucosal infiltration by eosinophilic (black arrows) H&E X20.

Discussion:-

Acute eosinophilic appendicitis was proposed in 1996 for the first time by Aravindan; then it was defined in 2010 by Aravindan et al. They reported that an allergic reaction type 1 hypersensitivity is a predisposing event, which triggers the acute inflammation of the appendix. This inflammation involves muscularispropria eosinophilic infiltration and edema [5], both are considered the hallmarks of this disease entity [8].

Type 1 hypersensitivity is not only developed in the appendix but also in other adjacent areas such as ileum and cecum, whereas the appendix considered the most affected organ by allergic reaction [4]. Although eosinophils are normally found in submucosal layer and lamina propria of the appendix, evidence of infiltration through the muscularispropria combined with edema should raise the suspicion of acute eosinophilic appendicitis [2].

The presentation of acute eosinophilic appendicitis is not different from the classic acute appendicitis, except in AEA, the symptoms are less in duration [2]. In our case report, the patient has presented mainly by acute LGIB, which is a rare presentation of acute appendicitis [6]. Magaz MM, et al, reported 30 cases that presented by bleeding from the appendix none were due to eosinophilic infiltration [10]. To our knowledge, this is the first case of acute eosinophilic appendicitis presenting with LGIB.

Many case reports have used different modalities such as colonoscopy, radionuclide scintigraphy, angiography, and multi-detector computed tomography (MDCT) to diagnose appendicular bleeding [11]. The combination of endoscopy with radiology may increase the accuracy of diagnosis during acute bleeding phase [6].

Management of appendiceal hemorrhage depends on patient's general condition and underlying etiology, and surgical intervention play a major role in treating appendiceal lesions and bleeding including laparoscopic versus open appendectomies, extensive resection of the bowel, and right hemicolectomy. In this case right hemicolectomy was done, as in most cases removal of the cecum is needed to control the bleeding [6].

In order, to reach definitive diagnosis histopathological features are needed which include: abundant eosinophilic infiltration in muscularispropria rather than neutrophil infiltration. This should be associated with edema [4].

According to this case, histopathology examination of the specimen revealed acutely inflamed appendix, mucosal ulceration, transmuraleosinophils infiltrate with no granulomatous formation or malignant changes.

Conclusion:-

Acute Eosinophilic Appendicitis (AEA) is a rare and not well-studied disease and it can be present with unusual symptom as LGIB. It is important to conduct further studies about AEA, for better diagnosis approach and management, as well as understanding the pathogenesis of this entity.

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RESEARCH ARTICLE

KINETICS AND MECHANISM OF THE HYDROLYSIS OF 4-HYDROXYBIPHENYL PHOSPHATE MONOESTER.

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Key words:-

Kinetics, Hydrolysis, Phosphate Monoester.

Abstract

Hydrolysis of 4-hydroxybiphenyl phosphate monoester has been carried out in acid (HCl) Solution at $97 \pm 0.5^\circ \text{C}$ and the inorganic phosphate obtained during hydrolysis has been estimated colorimetrically. Pseudo first order rate coefficients have been calculated neutral and conjugated acid species have been found to be reactive and contributed to the overall hydrolysis in the acid region. The calculated rate coefficients are in good agreement with the experimentally observed ones. The effect of temperature, solvent and substrate concentration on the rate of hydrolysis have been studied. The results suggest that the hydrolysis of monoester occurs by the cleavage of P-O bond fission.

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Introduction:-

The effect of the substituents, the hydrolysis of aryl phosphate esters having C-O-P¹⁻³ linkages have been studied exhaustively by Mhala et al⁴⁻⁶ and established that the substituents of graded polarity⁷⁻⁸ not only affect the reaction rates but also alter the course of the reaction path. Bunton et al⁹ found that acid catalyzed hydrolysis occurs only when an electron attracting substituent is present in the aryl moiety as in nitrophenyl phosphate monoester¹⁰, 4-chlorophenyl phosphate monoester¹¹ and 2,6-dimethylphenyl phosphate monoester¹² showed only feeble acid catalysis while 4-bromo-2,6-dimethylphenyl phosphate monoester showed acid catalysis.

Material and Methods:-

Preparation of mono-4-hydroxybiphenyl phosphate:-

Mono-4-hydroxybiphenyl phosphate ester have been prepared by general method¹³. Took 17.0 gm of 4-hydroxybiphenol in a round bottom flask with 200 ml of dry benzene. 9.0 ml of phosphorus oxytrichloride (POCl₃) was added drop by drop with constant stirring to the ice-cooled phenol with the help of separating funnel in about half an hour after the addition of POCl₃. It was refluxed for about 48 hours on the soxhlet heater at constant temperature 85°C in order to ensure complete reaction and then distilled at reduce pressure. The first fraction of benzene and unreacted POCl₃ was removed by distillation at b_{60} , $40-50^\circ \text{C}$.

The second fraction of a pungent smelling liquid, which was supposed to be 4-hydroxybiphenyl phosphate dichloride was distilled at b_{75} , $90-120^\circ \text{C}$. It was dissolved in 100 ml of ice cooled water and kept at low temperature overnight. The 4-hydroxybiphenyldihydrogen phosphate which was extracted with solvent ether. After removing the

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solvent ether a white coloured crystalline solid and it was identified to be mono-4-hydroxybiphenyl phosphate with the following physical characteristics.

1. Melting point (observed)=180°C
2. Theoretical percentage of "P"=12.40%
3. Observed percentage of "P"=13.40%
4. The Infrared spectrum of monoesters showed the appearance of absorption bands characteristics of-
 _OH = 3500-3265cm⁻¹
 _CH = 3050-2891cm⁻¹
 _C=C = 1640-1580cm⁻¹
 _P-O = 1385-1290cm⁻¹
 _C_O_P = 1200-1150cm⁻¹
 P_OH = 1040-976 cm⁻¹

Substituted aromatic ring =800-710cm⁻¹

Above characteristics confirm the structure of mono-4-hydroxybiphenyl phosphate.

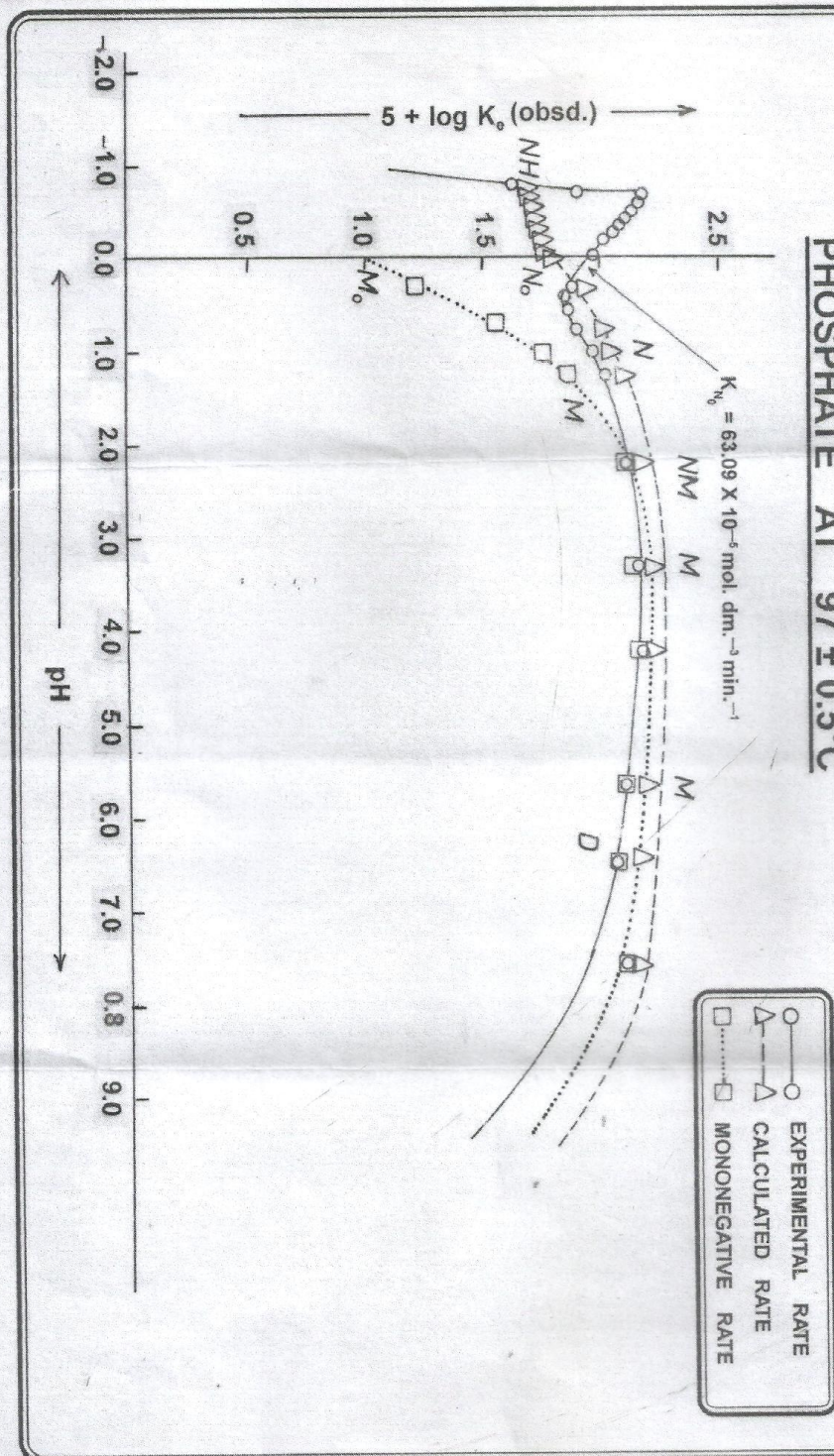
Result and Discussion:-

Hydrolysis of mono-4-hydroxybiphenyl phosphate was kinetically studied in 0.1 to 6.0 mol dm⁻³ HCl at 97 ± 0.5 °C Psuedo first order rate coefficients have been summarized in table-1

pH-log rate profile of mono -4-hydroxybiphenyl phosphate at 97± 0.5°C

HCl (mol.dm ⁻³)	P ^H	10 ⁵ .Ke(mol.dm ⁻³ min ⁻¹ Obsd)	5+log Ke
6.0	-0.778	42.61	1.63
5.0	-0.669	82.18	1.91
4.0	-0.602	154.54	2.19
3.5	-0.544	147.96	2.17
3.0	-0.544	147.96	2.17
3.0	-0.477	138.82	2.14
2.5	-0.397	126.34	2.10
2.0	-0.300	117.55	2.07
1.5	-0.176	104.84	2.02
1.0	0.000	92.80	1.97
0.5	0.301	74.36	1.87
0.4	0.400	72.21	1.86
0.3	0.520	69.66	1.84
0.2	0.700	79.02	1.90
0.1	1.000	93.60	1.97
Buffers-	1.24	102.38	2.01
Composition of buffers	2.20	127.58	2.11
	3.33	132.81	2.12
	4.17	138.74	2.14
	5.60	126.38	2.10
	6.43	111.51	2.05
	7.46	129.70	2.11

FIG. I
pH - log RATE PROFILE OF MONO - 4 - HYDROXYBIPHENYL
PHOSPHATE AT $97 \pm 0.5^\circ\text{C}$



A plot(fig-1) of pH-log rate of hydrolysis increases with increase in acid molarity. The rate maxima is at 4.0 mol dm⁻³ HCl and it gradually falls with further increase in acid molarity. The rate maxima or bend of pH log rate profile in acid region have also been found in some other cases. In organic amide system, similar rate maxima was observed and was supposed due to full conversion into their conjugate acid species the rate decrease is attributed to the lowering of concentration of attacking nucleophile taking part in the reaction, although it appears that there is no similarity in rate maxima with amides as it is not possible due to complete protonation because there is a small variation in temperature coefficient before and after rate maxima and it may be taken as an indication for the absence of maximum protonation. The rate maxima in mono-4-hydroxybiphenyl phosphate may be due to either ionic strength effect or acid catalyzed hydrolysis or due to both.

The initial decrease in rate hydrolysis may be attributed to the decrease in the concentration of more reactive mononegative species. Increase in rate in the region 1.0 to 4.0 mol dm⁻³ HCl may be due to either acid catalyzed hydrolysis or increase in the reactivity of neutral species by ionic strength effect. Therefore effect of ionic strength was studied by carrying out kinetic runs at different ionic strength maintained constant by adding appropriate mixtures of KCl and HCl. The plots of the rate constant of acid hydrolysis vs acid molarity at different ionic strengths are given in table-2.

Hydrolysis of mono-4-Hydroxybiphenyl phosphate at constant ionic strength at 97± 0.5°C

Composition

Ionic strength(u) (mol.dm ⁻³)	HCl (mol.dm ⁻³)	KCl (mol.dm ⁻³)	10 ⁻⁵ Ke (mol.dm ⁻³ .min ⁻¹)(Obsd)
1.0	0.2	0.8	61.87
1.0	0.4	0.6	66.16
1.0	0.6	0.4	71.66
1.0	0.8	0.2	77.68
1.0	0.0	0.0	92.80
2.0	0.2	1.8	56.59
2.0	0.5	1.5	64.91
2.0	1.0	1.0	76.80
2.0	1.5	0.5	87.05
2.0	1.8	0.2	93.84
2.0	2.0	0.0	117.55
3.0	0.5	2.5	58.67
3.0	1.0	2.0	65.20
3.0	1.5	1.5	73.23
3.0	2.0	1.0	84.04
3.0	2.5	0.5	92.18
3.0	3.0	0.0	138.82

Three linear plots show the acid catalyzed hydrolysis of mono-4-hydroxybiphenyl phosphate at three different ionic strengths (1.0u,2.0u,3.0u)

The intercepts on the rate axis are the neutral rates (K_N) which decrease with the increase in ionic strength showing ionic acceleration effect on the hydrolysis of monoester via its neutral species therefore the calculated rates were obtained by equ (1)

$$K_e = K_H \cdot C_H + K_N \text{ -----(1)}$$

Where K_e, K_H, C_H and K_N are experimental rates acid catalyzed rates and neutral rates respectively. The plot of mono 4-hydroxybiphenyl phosphate shows that three plots are intersecting the rate axis at different points (fig.2) hence this indicates in addition to conjugate acid species the presence of some other species.

Since it is established fact that mononegative species cannot be reactive at higher acidities i.e more than 1.0 mol.dm⁻³ HCl, so the neutral species participation may be expected together with conjugate acid species as it is evident that straight lines are intersecting the rate axis at different points which shows the contribution of neutral species at different acid concentration is varying definitely.

K_{H^+} is a slope of linear curve and its value decrease with the increase in ionic strength. It is an ionic retarding effect which indicates the mono-4-hydroxybiphenyl phosphate undergoes acid catalyzed hydrolysis with negative of ionic strength. The slope of these curves represents the specific acid catalyzed rates at the ionic strength which also decrease with the increase in ionic strength. Thus both specific neutral rates are subjected to negative effect of ionic strength. The neutral rates may be calculated using second empirical term of Debye –Huckel equation¹⁴⁻¹⁵ and modifying it logarithmically.

$$\log K_N = \log K_{NO} + b_N \cdot u \text{ -----(2)}$$

And its logarithmic form can be represented as

$$5 + \log K_N = 5 + \log K_{NO} + b_N \cdot u \text{ -----(3)}$$

$$\text{Where } b_N = b_N / 2.303$$

In equ (2) K_N , K_{NO} , b_N and u respectively are specific neutral rates at zero ionic strength and the slope of the linear curve and ionic strength (fig. not shown)

The intercept on the rate axis of such a plot i.e $\log K_N$ is found to be different thus the increase in specific neutral rate is due to($b_N \cdot u$) factor supporting ionic acceleration effect specific neutral rates calculated by equation 2 are almost different from the experimental rate upto 4.0 mol.dm⁻³ HCl and shown in Table-1. The rates above 4.0 mol.dm⁻³ HCl however deviate probably due to decrease in the reactivity of the neutral form as a result of decrease in water acidity therefore the rates beyond 4.0 mol.dm⁻³ HCl were calculated employing the Bronsted-Bjerrum equation¹⁶⁻¹⁷

$$K_e = K_H^+ C_H^+ \exp b_{H^+} \cdot u (a_{H_2O})^n + K_{NO} \cdot \exp b_N \cdot u (a_{H_2O})^n \text{ -----(4)}$$

And its logarithmic form can be represented as

$$5 + \log K_e + 5 + \log K_H^+ + \log C_H^+ + b_{H^+} \cdot u + n \log (a_{H_2O}) + 5 + \log K_{NO} + b_N \cdot u + n \log a_{H_2O}$$

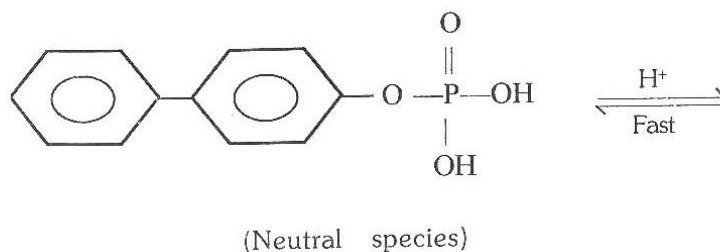
Where $(a_{H_2O})^n$ represents the water acidity parameter.

The values of $n=2,3$ were calculated for 5.0 and 6.0 mol dm⁻³ HCl respectively. The experimental rates thus calculated are in good agreement with the theoretical rates . The thermodynamic parameter for the reaction at 3.0 and 5.0 mol.dm⁻³ HCl were found to be $E=22.70$ and 22.06 K.cal.mol⁻¹ frequency factor (A)= 5.89×10^8 , 1.46×10^8 Sec⁻¹ and $S=-20.83$, -23.60 e.u. These values indicate the bimolecular nature of hydrolytic reaction.

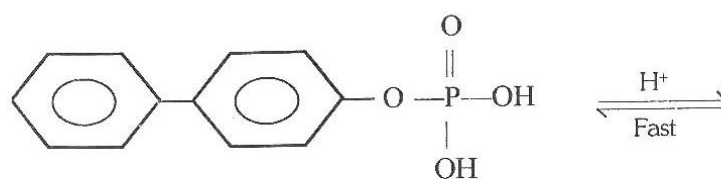
The acid hydrolysis of monoester, therefore, involves bimolecular attack of water on phosphorus of the neutral species as shown in schemes I and II.

Scheme- I

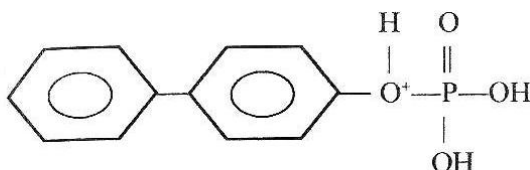
(a) **FORMATION OF CONJUGATE ACID SPECIES BY
FAST PRE-EQUILIBRIUM PROTON TRANSFER:**



(a) **FORMATION OF CONJUGATE ACID SPECIES BY
FAST PRE-EQUILIBRIUM PROTON TRANSFER:**

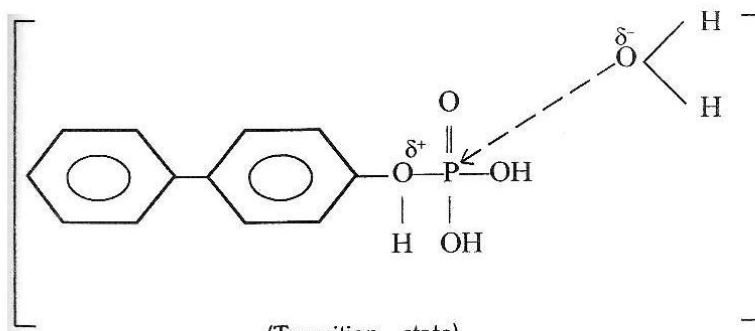
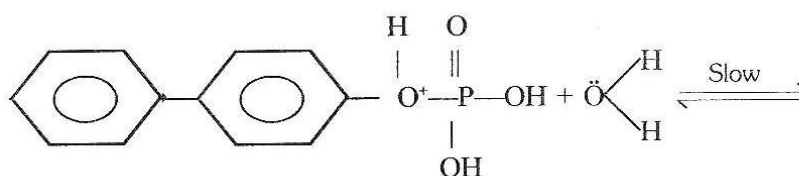


(Neutral species)



(Conjugate acid species)

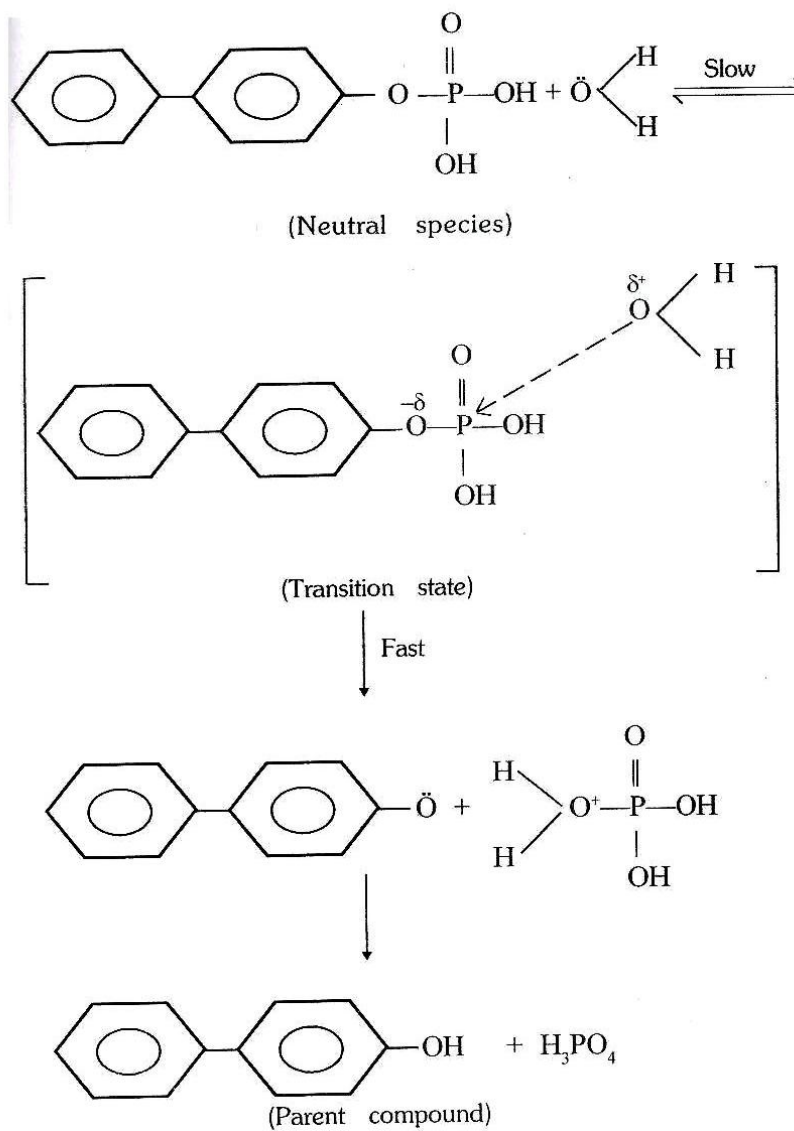
b) **BIMOLECULAR NUCLEOPHILIC ATTACK OF WATER
ON PHOSPHORUS OF CONJUGATE ACID SPECIES
INVOLVING P—O BOND FISSION $\text{S}_{\text{N}}2$ (P):**

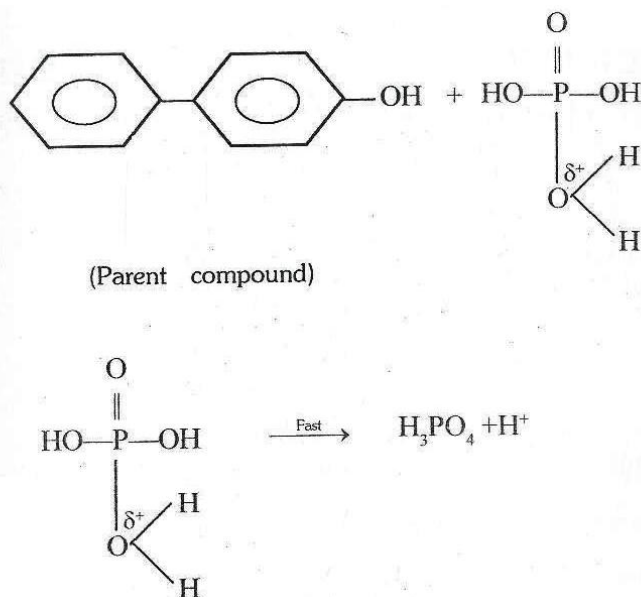


(Transition state)

Fast

BIMOLECULAR NUCLEOPHILIC ATTACK OF WATER
ON PHOSPHORUS ATOM WITH P—O BOND
FISSION:





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RESEARCH ARTICLE

A FOOD CHAIN STRUCTURES AND DYNAMICS ACROSS A REFUGE STAGE- STRUCTURE PREY-PREDATOR MODEL.

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Abstract

In the present paper the structures and dynamics of a food chain prey-predator model are proposed and studied. The two stages and refuge in prey and two predators (top and mid) are considered. Different types of functional responses have been proposed. The conditions, which guarantee the existence of equilibrium points, have been investigated. Uniqueness and boundedness of the solution of the system are proven. The local and global dynamical behaviors are discussed and analyzed. Finally, numerical simulations are carried out not only to confirm the theoretical results obtained, but also to show the effects of the refuge and the variation of each parameter on our proposed system.

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Introduction:-

The first pattern formation of predator-prey model was introduced in 1927 by the well-known Lotka[15] and Volterra[28] which starting from a simple and classical supposition, more complexity but factual predator-prey models have been constructed by mathematicians and ecologists. In 1992, Berryman [6] considered that the dynamic interaction between predators and preys has long taken and go on to be one of the most important and central subjects which play a great role in each of the mathematical ecology, natural, social, and technological sciences, especially in the research on biology and ecology [5, 17, 26, 27].

Many researchers working in these fields paying a great amount of attention to improve and generalize the pattern formation of prey-predator models to more involvement and realistic system, and merging this advantage into many applications that give the nature of existence and global asymptotic stability of predator-prey models a great interest.

On the other hand, the dynamical behavior of prey-predator models has been investigated by a lot of articles and peppers which results a significant expansion in the structural of the prey – predator models to involve multi-species; different functional responses, several types of food chain, refuge, harvesting, stage-structure, several types of diseases...etc.

Lately, Samantha[25] studied the existence and global asymptotic stability of a delay predator-prey model with disease in the prey, Paletal.[20–24]proposed prey-predator harvesting model under impreciseness by considering biological parameters as interval number, Naji and Mustafa [19] studied the dynamics of an eco-epidemiological model with nonlinear incidence rate. Majeed and Shawka studied prey-predator model involving SI and SIS infectious disease in prey population and the disease transmitted within the same species by contact and external

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source [4]. A lot of researches efforts have been made to investigating the interaction and coexistence mechanism of the harvested prey–predator.

In fact, the influence of refuge, stage-structure and the multiplicity of functional responses in the prey–predator ecosystem are the most important topics of interest. In recent years, stage structure models have been studied widely by a lot of researchers, Aiello and Freedman (1990) studied the single species model with stage structure [1, 2], several researchers studied prey–predator model with different functional responses. The prey–predator models with prey refuge have been investigated by Kar[14]. Maetal. [16]studied the effects of prey refuges on a prey–predator model with a class of function responses. Chenetal.[8] discussedaprey–predator model with Holling type II functional response in incorporating a constant prey refuge.

Moreover, someoftheexperimentalandtheoreticalworkhavestudiedtheeffectsofpreyrefuges and drawn a conclusion thatthepreyrefuges have a positively effectonthestabilityof the consideredinteractions,andprey extinction canbeconserved by theadditionofrefuges[9, 11, 12, 13, 17].Kadhim, Majeed and Naji [29] studied the stability analysis of food web stage structured prey-predator model with refuge involving Lotka-Volterra type of function response, Ali and Majeed discussed stability analysis of a food chain stage structured prey-predator model incorporating a preyrefuge and two types of functional responses such that the mid-predator consumed the mature prey individual only according to the Lotka-Volterra type of functional response and the top predator consumed the mid- predator individual only according to Holling type-II functional response.

In spite of the effect of prey refuges on the dynamical behavior of the system is very difficult in the reality, but it has been considered and analysis in this paper.

In this paper, a complicated four species food chain model with two functional responses has been proposed and analyzed. Two such stages (mature and immature) with refuge in the prey and two predators (top and mid) are also considered.

It is well known that the logistic function represents a mutualism interaction between the mature and immature prey, and due to the short of resources the typeIIfunctionalresponse is considered to describe the nature of the interaction between the mid predator and immature prey, while Lotka-Volterra type of functionalresponse considered due to unlimited resources and the linearly relationship between the mid predator and mature prey.

Furthermore, there is not any relationship between the top predator and the two stages of prey, while according to a food chain the interaction between the two predators has been considered by Lotka–Volterra type of functionalresponse.

Actually, due to the complexity of interactions in the proposed model especially among the top predator and the two stages of prey and the nature of interaction between the mid predator and the immature prey it was not easy to find an example identical of this model in the nature, but in any case is not impossible, so after more efforts and with the help of the biologist, we could find an example across two different environments represented by the aquatic and terrestrial environment such that the two aquatic species Blue Shark (*Prionaceglauca*) and Wels Catfishrepresent the top and mid predators respectively, and the terrestrial species pigeons (doves) represent the two stages of prey.

Finally, the behavior of the catfish when hunting pigeons is highlighted in the study, published on Dec. 5. The study says: “Among a total of 45 beaching behaviors observed and filmed, 28% were successful in bird capture... Since this extreme behavior has not been reported in the native range of the species, our results suggest that some individuals in introduced predator populations may adapt their behavior to forage on novel prey in new environments, leading to behavioral and trophic specialization to actively cross the water-land interface.” [10, 7].

The mathematical model:-

Consider the food chain model consisting of mid and top- predators, stage-structure prey in which the prey species growth logistically in the absence of predation, while the predators decay exponentially in the absence of prey species. It is assumed that the prey population divides into two compartments: immature prey population $X_1(t)$ that represents the population size at time t and mature prey population $X_2(t)$ which denotes to population size at time t . Furthermore the population size of the mid- predator at time t is denoted by $Y_1(t)$, while $Y_2(t)$ represents the population size of top predator at time t .

Now in order to formulate the dynamics of such system the following assumptions are considered:

1. The immature prey depends completely in its feeding on the mature prey that growth logistically with intrinsic growth rate $r > 0$ and carrying capacity $k > 0$. The immature prey individuals grown up and become mature prey individuals with grown up rate $s > 0$. However the immature and mature prey facing death with natural death rate $d_1 > 0$ and $d_2 > 0$ respectively.
2. There is type of protection of the prey species from facing predation by the mid- predator with refuge rate constant $m \in (0, 1)$.
3. The mid-predator consumed the immature prey individual according to Holling type-II functional response with predation rate $a_1 > 0$ and half saturation constant $b > 0$. And consumed the mature prey individual according to the Lotka-Volterra type of functional response with predation rate $a_2 > 0$ and contribute a portion of such food with conversion rates $0 < e_1 < 1$ and $0 < e_2 < 1$ respectively. Moreover, the top predator consumed the mid-predator individual only according to the Lotka-Volterra type of functional response with predation rate $a_3 > 0$ and contributes a portion of such food with conversion rate $0 < e_3 < 1$.

Finally, in the absence of food the mid and top predators facing death with natural death rate $d_3 > 0$ and $d_4 > 0$ respectively.

Therefore the dynamics of the above proposed model can be represented by the following set of first order nonlinear differential equations.

$$\begin{aligned}\frac{dX_1}{dT} &= rX_2 \left(1 - \frac{X_2}{k} \right) - \frac{a_1(1-m)X_1}{b+X_1}Y_1 - sX_1 - d_1X_1 \\ \frac{dX_2}{dT} &= sX_1 - a_2(1-m)X_2Y_1 - d_2X_2 \\ \frac{dY_1}{dT} &= \frac{e_1a_1(1-m)X_1}{b+X_1}Y_1 + e_2a_2(1-m)X_2Y_1 - a_3Y_1Y_2 - d_2Y_1 \\ \frac{dY_2}{dT} &= e_3a_3Y_1Y_2 - d_4Y_2.\end{aligned}\tag{1}$$

With initial conditions $X_i(0) \geq 0$ and $Y_i(0) \geq 0$, $i = 1, 2$.

Not that the above proposed model has fourteen parameters in all which make the analysis difficult. So in order to simplify the system, the number of parameters is reduced by using the following dimensionless variables and parameters:

$$\begin{aligned}t &= rT, u_1 = \frac{b}{k}, u_2 = \frac{s}{r}, u_3 = \frac{d_1}{r}, u_4 = \frac{a_2k}{a_1}, u_5 = \frac{d_2}{r}, a_6 = \frac{e_1a_1}{r}, u_7 = \frac{e_2a_2k}{r}, \\ u_8 &= \frac{d_3}{r}, u_9 = \frac{e_3a_3k}{r}, u_{10} = \frac{d_4}{r}, x = \frac{X_1}{k}, y = \frac{X_2}{k}, z = \frac{a_1Y_1}{rk}, w = \frac{a_3Y_2}{r}.\end{aligned}$$

Then the non-dimensional form of system (1) can be written as:

$$\begin{aligned}\frac{dx}{dt} &= x \left[\frac{y(1-y)}{x} - \frac{(1-m)z}{u_1+x} - (u_2 + u_3) \right] = f_1(x, y, z, w) \\ \frac{dy}{dt} &= y \left[\frac{u_2x}{y} - u_4(1-m)z - u_5 \right] = f_2(x, y, z, w) \\ \frac{dz}{dt} &= z \left[\frac{u_6(1-m)x}{u_1+x} + u_7(1-m)y - w - u_8 \right] = f_3(x, y, z, w) \\ \frac{dw}{dt} &= w[u_9z - u_{10}] = f_4(x, y, z, w).\end{aligned}\tag{2}$$

With $x(0) \geq 0, y(0) \geq 0, z(0) \geq 0$ and $w(0) \geq 0$.

It is observed that the number of parameters have been reduced from fourteen in the system (1) to eleven in the system (2).

Obviously the interaction functions of the system (2) are continuous and have continuous partial derivatives on the following positive four dimensional space.

$$R_+^4 = \{(x, y, z, w) \in R^4 : x(0) \geq 0, y(0) \geq 0, z(0) \geq 0, w(0) \geq 0\}.$$

Therefore these functions are Lipschitzian on R_+^4 , and hence the solution of the system (2) exists and is unique. Further, all the solutions of system (2) with non-negative initial conditions are uniformly bounded as shown in the following theorem.

Theorem (1): All the solutions of system (2) which initiate in R_+^4 are uniformly bounded.

Proof: Let $(x(t), y(t), z(t), w(t))$ be any solution of the system (2) with non-negative initial condition $(x_0, y_0, z_0, w_0) \in R_+^4$.

According to the first equation of system (2) we have

$$\frac{dx}{dt} = y(1-y) - \frac{(1-m)xz}{u_1+x} - (u_2+u_3)x.$$

$$\text{So, } \frac{dx}{dt} \leq \frac{1}{4} - (u_2+u_3)x.$$

Now, by using the comparison theorem on the above differential inequality with the initial point $x(0) = x_0$, we get:

$$x(t) \leq \frac{1}{4(u_2+u_3)} + \left(x_0 - \frac{1}{4(u_2+u_3)}\right)e^{-(u_2+u_3)t}.$$

Thus, $\lim_{t \rightarrow \infty} x(t) \leq \frac{1}{4(u_2+u_3)}$, and hence $\sup x(t) \leq \frac{1}{4(u_2+u_3)}$, $\forall t > 0$.

Now define the function: $N(t) = x(t) + y(t) + z(t) + w(t)$, and then taken the time derivative of $N(t)$ along the solution of the

$$\frac{dN}{dt} = y - y^2 - (1-u_6)\frac{(1-m)xz}{u_1+x} - (u_4-u_7)yz - (1-u_9)zw - u_3x - u_5y - u_8z - u_{10}w.$$

So, due to the fact that the conversion rate constant from prey population to predator population cannot exceeding the maximum predation rate constant from predator population to prey population, hence from the biological point of view, always $u_6 < 1$, $u_9 < 1$ and $u_7 < u_4$, we get,

$$\frac{dN}{dt} \leq y - sN, \quad \text{where } s = \min\{u_3, u_5, u_8, u_{10}\}, \quad \text{and hence, } \frac{dN}{dt} \leq \frac{u_2}{4a_5(u_2+u_3)} - sN.$$

$$\text{Then, } \frac{dN}{dt} + sN \leq G, \quad \text{where } G = \frac{u_2}{4a_5(u_2+u_3)}.$$

Again by solving this differential inequality for the initial value $N(0) = N_0$, we get:

$$N(t) \leq \frac{G}{s} + \left(N_0 - \frac{G}{s}\right)e^{-st}. \quad \text{Then, } \lim_{t \rightarrow \infty} N(t) \leq \frac{G}{s}. \quad \text{So, } 0 \leq N(t) \leq \frac{G}{s}, \quad \forall t > 0,$$

hence all the solutions of system (2) are uniformly bounded and the proof is complete.

The existence of equilibrium points:-

In this section, the existence of all possible equilibrium points of system (2) is discussed. It is observed that, system (2) has at most four equilibrium points, which are mentioned in the following:

● The equilibrium point $E_0 = (0, 0, 0, 0)$, which known as vanishing point is always exists.

● The free predators' equilibrium point $E_1 = (\bar{x}, \bar{y}, 0, 0)$, where:

$\bar{y} = 1 - u_5 \left(\frac{u_2+u_3}{u_2}\right)$, and $\bar{x} = \frac{u_5}{u_2} \left(1 - u_5 \left(\frac{u_2+u_3}{u_2}\right)\right)$, exists uniquely in $\text{Int. } R_+^2$ (Interior of R_+^2) of xy -plane if the necessary condition holds:

$$u_5 < \frac{u_2}{u_2+u_3}. \quad (3)$$

● The free top predator' equilibrium point $E_2 = (\hat{x}, \hat{y}, \hat{z}, 0)$, where:

$$\hat{z} = \frac{u_2x - u_5y}{u_4(1-m)y}. \quad (4a)$$

And,

$$\hat{y} = \frac{u_8(u_1+x) - u_6(1-m)x}{u_7(1-m)(u_1+x)}. \quad (4b)$$

While, \hat{x} is a positive root of the following fourth order polynomial:

$$f(x) = \gamma_1 x^4 + \gamma_2 x^3 + \gamma_3 x^2 + \gamma_4 x + \gamma_5 = 0, \quad (4c)$$

where:

$$\gamma_1 = u_7^2(1-m)^3[u_4(u_2+u_3)(u_6(1-m)-u_8) - u_2u_7(1-m)]$$

$$\gamma_2 = -u_4(1-m)(u_6(1-m)-u_8)[u_7(1-m) + (u_6(1-m)-u_8)^2]$$

$$-u_7^2(1-m)^3[u_5(u_6(1-m)-u_8) + u_1u_4u_8(u_2+u_3)] - 2u_1u_7^2(1-m)^3[u_2u_7(1-m) - u_4(u_6(1-m)-u_8)].$$

$$\gamma_3 = u_1 u_4 u_8 (1-m)(u_6(1-m)-u_8)[3(u_6(1-m)-u_8) - 2u_7(1-m)] + u_1 u_4 u_7 (1-m)^2 (u_6(-m)-u_8)^2 - u_1^2 u_4 u_7^2 (u_2 + u_3)(1-m)^3 (3u_8 - u_6(1-m)) + u_1 u_7^2 (1-m)^3 [u_5 u_8 - u_1 u_2 u_7 (1-m)] - u_1 u_5 u_7^2 (1-m)^3 (u_6(1-m)-u_8).$$

$$\gamma_4 = u_1^2 u_7 u_8 (1-m)^2 [u_4 u_8 + u_5 u_7 (1-m)] - u_1^2 u_4 u_8 (1-m)(u_6(1-m)-u_8)[3u_8 + 2u_7(1-m)] - u_1^3 u_7^2 u_4 u_8 (1-m)^3 (u_2 + u_3).$$

$$\gamma_5 = u_1^3 u_4 u_7 u_8^2 (1-m)^2 + u_1^3 u_4 u_8^3 (1-m) > 0.$$

Clearly, by using Descartes' rule of sign Eq. (4 c) has a unique positive root, denoted by \hat{x} , if

$$\gamma_i < 0, \quad i = 1, 2, 3 \quad \text{and} (\gamma_4 < 0 \text{ or } \gamma_4 > 0).$$

Therefore, the free top predator equilibrium point $E_2 = (\hat{x}, \hat{y}, \hat{z}, 0)$ exists uniquely in the interior R_+^3 of xyz -space if the following conditions hold:

$$\frac{u_6(1-m)}{3} < u_8 < u_6(1-m). \quad (4d)$$

$$u_2 > \max \left\{ \frac{u_5 \hat{y}}{\hat{x}}, \frac{u_5 u_8}{u_1 u_7 (1-m)} \right\}. \quad (4e)$$

$$(u_6(1-m)-u_8) < \min \left\{ \frac{u_1 u_8}{\hat{x}}, \frac{2u_7(1-m)}{3}, \frac{u_2 u_7(1-m)}{u_4}, \frac{u_2 u_7(1-m)}{u_4(u_2+u_3)}, \frac{u_2 u_7(1-m)}{u_1 u_7(u_2+u_3)(1-m)(3u_8-u_6(1-m))} \right\}. \quad (4f)$$

Finally, the positive (coexistence) equilibrium point $E_3(x^*, y^*, z^*, w^*)$, where:

$$z^* = \frac{u_{10}}{u_9}. \quad (5a)$$

$$w^* = \frac{u_6(1-m)x + u_7(1-m)(u_1+x)y - u_8(u_1+x)}{u_1+x}. \quad (5b)$$

$$x^* = \frac{(u_4 u_{10}(1-m) + u_5 u_9)y}{u_2 u_9}. \quad (5c)$$

While, y^* is a positive root of the following polynomial:

$$f(y) = y(\delta_1 y^2 + \delta_2 y + \delta_3) = 0, \quad (5d)$$

where:

$$\delta_1 = -u_2 u_9 (u_4 u_{10}(1-m) + u_5 u_9) < 0.$$

$$\delta_2 = u_2 u_9 (u_4 u_{10}(1-m) + u_5 u_9) - u_1 u_2^2 u_9^2 - (u_2 + u_3)(u_4(1-m) + u_5 u_9)^2.$$

$$\delta_3 = u_2 [u_1 u_2 u_9^2 - (u_4 u_{10}(1-m) + u_5 u_9)(u_{10}(1-m) + u_9(u_2 + u_3))].$$

Straightforward computation shows that the quadratic eq. (5 d), has a unique positive root, denoted by y^* , if and only if the following condition holds:

$$u_1 > \frac{(u_4 u_{10}(1-m) + u_5 u_9)(u_{10}(1-m) + u_9(u_2 + u_3))}{u_2 u_9^2}. \quad (5e)$$

Therefore, the positive (coexistence) equilibrium point $E_3(x^*, y^*, z^*, w^*)$ exists uniquely in the interior R_+^4 if in addition to the condition (5 e) the following condition holds:

$$y^* > \frac{u_8}{u_7(1-m)}. \quad (5g)$$

The local stability analysis of system (2)

In this section, the local stability analysis of system (2) around each of the above equilibrium points is discussed through computing the Jacobian matrix $J(x, y, z, w)$ of system (2) at each of them which is given by:

$$J = [a_{ij}]_{4 \times 4}, \quad \text{where} \quad (6)$$

$$a_{11} = -\frac{u_1(1-m)z}{(u_1+x)^2} - (u_2 + u_3), \quad a_{12} = 1 - 2y, \quad a_{13} = -\frac{(1-m)x}{u_1+x}, \quad a_{14} = 0, \quad a_{21} = u_2,$$

$$a_{22} = -u_4(1-m)z - u_5, \quad a_{23} = -u_4(1-m)y, \quad a_{24} = 0, \quad a_{31} = \frac{u_1 u_6(1-m)z}{(u_1+x)^2}, \quad a_{32} = u_7(1-m)z,$$

$$a_{33} = \frac{u_6(1-m)x}{u_1+x} + u_7(1-m)y - u_8 - w, \quad a_{34} = -z, \quad a_{41} = 0, \quad a_{42} = 0, \quad a_{43} = u_9 w, \quad a_{44} = u_9 z - u_{10}.$$

The local stability analysis at E_0 :

The Jacobian matrix of system (2) at E_0 can be written as:

$$J_0 = J(E_0) = \begin{bmatrix} -(u_2 + u_3) & 1 & 0 & 0 \\ u_2 & -u_5 & 0 & 0 \\ 0 & 0 & -u_8 & 0 \\ 0 & 0 & 0 & -a_{10} \end{bmatrix} \quad (7a)$$

Then the characteristic equation of $J(E_0)$ is given by:

$$[\lambda^2 + A\lambda + B](-u_8 - \lambda)(-u_{10} - \lambda) = 0, \quad (7b)$$

where:

$$A = u_2 + u_3 + u_5, \quad B = u_5(u_2 + u_3) - u_2,$$

so, either

$$(-u_8 - \lambda)(-u_{10} - \lambda) = 0, \quad (7c)$$

this gives two eigenvalues of J_0 by: $\lambda_{0z} = -a_3 < 0$, and $\lambda_{0w} = -a_8 < 0$.

Or

$$\lambda^2 + A\lambda + B = 0, \quad (7d)$$

where:

$tr(A) = \lambda_{0x} + \lambda_{0y} = -(u_2 + u_3 + u_5) < 0$, and $det(A) = \lambda_{0x} \cdot \lambda_{0y} = u_5(u_2 + u_3) - u_2$, which gives the other two eigenvalues of J_0 with negative real parts provided that the following condition holds:

$$u_5 > \frac{u_2}{(u_2 + u_3)}. \quad (7e)$$

Then E_0 is locally asymptotically stable in the R_+^4 . However, it is a saddle point otherwise.

The local stability analysis at E_1

The Jacobian matrix of system (2) at E_1 can be written as:

$$J_1 = J(E_1) = [b_{ij}]_{4 \times 4}, \quad (8a)$$

where:

$$\begin{aligned} b_{11} &= -(u_2 + u_3), \quad b_{12} = 1 - 2\bar{y}, \quad b_{13} = -\frac{(1-m)\bar{x}}{u_1 + \bar{x}}, \quad b_{14} = 0, \quad b_{21} = u_2, \quad b_{22} = -u_5, \\ b_{23} &= u_4(1-m)\bar{y}, \quad b_{24} = 0, \quad b_{31} = 0, \quad b_{32} = 0, \quad b_{33} = \frac{u_6(1-m)\bar{x}}{u_1 + \bar{x}} + u_7(1-m)\bar{y} - u_8, \\ b_{34} &= 0, \quad b_{41} = 0, \quad b_{42} = 0, \quad b_{43} = 0, \quad b_{44} = -u_{10}. \end{aligned}$$

Then the characteristic equation of J_1 is given by:

$$[\lambda^2 + \bar{A}\lambda + \bar{B}](b_{33} - \lambda)(-u_{10} - \lambda) = 0, \quad (8b)$$

where:

$$\bar{A} - tr(A) = u_2 + u_3 + u_5,$$

$$\bar{B} = det(A) = u_5(u_2 + u_3) - u_2 + u_2\bar{y},$$

so, either

$$(b_{33} - \lambda)(-u_{10} - \lambda) = 0, \quad (8c)$$

which gives two eigenvalues of J_1 by:

$$\lambda_{1z} = \frac{u_6(1-m)\bar{x}}{u_1 + \bar{x}} + u_7(1-m)\bar{y} - u_8, \quad \text{and} \quad \lambda_{1w} = -u_{10} < 0$$

$$\text{Or} \quad \lambda^2 + \bar{A}\lambda + \bar{B} = 0, \quad (8d)$$

where: $tr(A) = \lambda_{0x} + \lambda_{0y} = -(u_2 + u_3 + u_5) < 0$, and $det(A) = \lambda_{0x} \cdot \lambda_{0y} = u_5(u_2 + u_3) - u_2 + u_2\bar{y}$, which gives the other two eigenvalues of J_1 with negative real parts due to the condition (3).

Then, E_1 is locally asymptotically stable in R_+^4 if in addition to the condition (8e), the following condition holds:

$$u_8 > \frac{u_6(1-m)\bar{x}}{u_1 + \bar{x}} + u_7(1-m)\bar{y}. \quad (8e)$$

However, it is a saddle point otherwise.

The local stability analysis at E_2

The Jacobian matrix of system (2) at E_2 can be written as:

$$J_2 = J(E_2) = [c_{ij}]_{4 \times 4}, \quad (9a)$$

where:

$$\begin{aligned} c_{11} &= -\frac{u_1(1-m)\hat{z}}{(u_1 + \hat{x})^2} - (u_2 + u_3) < 0, \quad c_{12} = 1 - 2\hat{y}, \quad c_{13} = -\frac{(1-m)\hat{x}}{u_1 + \hat{x}} < 0, \quad c_{14} = 0, \quad a_{21} = u_2 > 0, \\ c_{22} &= \frac{-u_2\hat{x}}{\hat{y}} < 0, \quad c_{23} - u_4(1-m)\hat{y} < 0, \quad c_{24} = 0, \quad c_{31} = \frac{u_1 u_6(1-m)\hat{z}}{(u_1 + \hat{x})^2} > 0, \quad c_{32} = u_7(1-m)\hat{z} > 0, \\ c_{33} &= \frac{u_6(1-m)\hat{x}}{u_1 + \hat{x}} + u_7(1-m)\hat{y} - u_8, \quad c_{34} = -\hat{z} < 0, \quad c_{41} = 0, \quad c_{42} = 0, \quad c_{43} = 0, \quad c_{44} = u_9\hat{z} - u_{10}. \end{aligned}$$

Then the characteristic equation of J_2 is given by:

$$[\lambda^3 + A_1\lambda^2 + A_2\lambda + A_3](u_9\hat{z} - u_{10} - \lambda) = 0, \quad (9b)$$

where:

$$A_1 = -(c_{11} + c_{22}) > 0$$

$$A_2 = c_{11}c_{22} - c_{12}c_{21} - c_{13}c_{31} - c_{23}c_{32}$$

$$A_3 = c_{32}(c_{11}c_{23} - c_{13}c_{21}) + c_{22}c_{13}c_{31} - c_{12}c_{23}c_{31},$$

so, either

$$u_9\hat{z} - u_{10} - \lambda = 0. \quad (9c)$$

$$\text{Or } \lambda^3 + A_1\lambda^2 + A_2\lambda + A_3 = 0. \quad (9d)$$

Hence from equation (9c) we obtain that:

$\lambda_{2w} = u_9\hat{z} - u_{10}$, which is negative if the following condition holds:

$$\hat{z} < \frac{u_{10}}{u_9} \quad (9e)$$

Since $A_1 > 0$, then by using Routh-Hawirtiz criterion equation (9d) has roots (eigenvalues) with negative real parts if and only if

$$A_3 > 0 \text{ and } \Delta = A_1A_2 - A_3 > 0.$$

Straightforward computation shows that:

$$\Delta = -(c_{11} + c_{22})c_{11}c_{22} + c_{11}(c_{12}c_{21} + c_{13}c_{31}) + c_{22}c_{23}c_{32} + c_{12}c_{23}c_{31} + c_{21}(c_{22}c_{12} + c_{13}c_{32}).$$

Now, according to the form of A_3 and signs of the Jacobian elements all terms of A_3 will be positive under the following conditions:

$$\hat{y} > \frac{1}{2} \quad (9f)$$

$$u_7 > \frac{u_1 u_6(2\hat{y} - 1)}{u_1(1-m)\hat{z} + (u_2 + u_3)(u_1 + \hat{x})^2}. \quad (9g)$$

However Δ becomes positive, since the first four terms of Δ are positive, while the last one will be positive if and only if in addition to the condition (9f) the following condition holds:

$$u_7 < \frac{u_2(2\hat{y} - 1)(u_1 + \hat{x})}{(1-m)^2\hat{y}\hat{z}}. \quad (9h)$$

Therefore, all the eigenvalues of J_2 have negative real parts under the given conditions and hence E_2 is locally asymptotically stable. However, it is unstable otherwise.

The local stability analysis at E_3

The Jacobian matrix of system (2) at E_3 can be written as:

$$J_3 = [d_{ij}]_{4 \times 4}, \quad (10a)$$

where:

$$\begin{aligned} d_{11} &= -\frac{u_1(1-m)z^*}{(u_1 + x^*)^2} - (u_2 + u_3) < 0, \quad d_{12} = 1 - 2y^*, \quad d_{13} = -\frac{(1-m)x^*}{u_1 + x^*} < 0, \quad d_{14} = 0, \quad d_{21} = u_2 > 0, \\ d_{22} &= \frac{-u_2x^*}{\hat{y}} < 0, \quad d_{23} - u_4(1-m)y^* < 0, \quad d_{24} = 0, \quad d_{31} = \frac{u_1 u_6(1-m)z^*}{(u_1 + x^*)^2} > 0, \quad d_{32} = u_7(1-m)z^* > 0, \\ d_{33} &= \frac{u_6(1-m)x^*}{u_1 + x^*} + u_7(1-m)y^* - u_8 - w^*, \quad d_{34} = -z^* < 0, \quad d_{41} = 0, \quad d_{42} = 0, \quad d_{43} = u_9w^* > 0, \\ d_{44} &= u_9z^* - u_{10}. \end{aligned}$$

Then the characteristic equation of J_3 is given by:

$$[\lambda^4 + B_1\lambda^3 + B_2\lambda^2 + B_3\lambda + B_4] = 0, \quad (10b)$$

where:

$$\begin{aligned} B_1 &= -\alpha_0 > 0. \\ B_2 &= \alpha_1 - \alpha_2 - \alpha_3 - \alpha_4 - \alpha_5. \\ B_3 &= (\alpha_0\alpha_5 - \alpha_6) + d_{11}\alpha_4 + d_{22}\alpha_3 - \alpha_7. \\ B_4 &= \alpha_5(\alpha_2 - \alpha_1) > 0, \text{ with,} \\ \alpha_0 &= (d_{11} + d_{22}) < 0, \alpha_1 = d_{11}d_{22} > 0, \alpha_2 = d_{12}d_{21}, \\ \alpha_3 &= d_{13}d_{31} < 0, \alpha_4 = d_{23}d_{32} < 0, \alpha_5 = d_{34}d_{43} < 0, \\ \alpha_6 &= d_{12}d_{23}d_{31}, \alpha_7 = d_{13}d_{21}d_{32} < 0. \end{aligned}$$

Now by using Routh-Hawartiz criterion equation (10 b) has roots (eigenvalues) with negative real parts if and only if $B_i > 0$, $i = 1, 3, 4$ and $\Delta = (B_1B_2 - B_3)B_3 - B_1^2B_4 > 0$.

Clearly $B_1 > 0$ and $B_4 > 0$ provided that:

$$y^* > \frac{1}{2} \quad (10 c)$$

Straightforward computation shows that:

$$\Delta = [\alpha_0(\alpha_2 - \alpha_1) + (d_{11}\alpha_3 + \alpha_7) + d_{22}\alpha_4 + \alpha_6][(\alpha_0\alpha_5 - \alpha_6) + (d_{11}\alpha_4 - \alpha_6) + d_{22}\alpha_3 - \alpha_7].$$

Now, according to the form of B_3 and signs of the Jacobian elements the last three terms of B_3 are positive, while the first one will be positive in addition to the condition (10 c) the following condition holds:

$$w^* > \frac{u_1u_4u_6(1-m)^2(2y^* - 1)y^{*2}}{u_9[u_1(1-m)y^*z^* + [(u_2 + u_3)y^* + u_2x^*](u_1 + x^*)^2]} \quad (10 d)$$

On the other hand due to the conditions (10 c) and (10 d), all elements of Δ are positive and the two elements $(d_{11}\alpha_3 + \alpha_7)$, $(d_{11}\alpha_4 - \alpha_6)$ will be positive if the following condition holds:

$$L_1 < u_7 < L_2, \quad (10 e)$$

where:

$$L_1 = \frac{u_1u_6(2y^* - 1)}{u_1(1-m)z^* + (u_2 + u_3)(u_1 + x^*)^2}, \quad \text{and} \quad L_2 = \frac{u_1u_6[u_1(1-m)z^* + (u_2 + u_3)(u_1 + x^*)^2]}{u_2(u_1 + x^*)^4}.$$

Therefore, all the eigenvalues of J_3 have negative real parts under the given conditions and hence E_3 is locally asymptotically stable. However, it is unstable otherwise.

The global stability analysis of system (2)

In this section the global stability analysis for the equilibrium points, which are locally asymptotically stable, of system (2) is studied analytically with the help of Lyapunov method as shown in the following theorems.

Theorem (2): Assume that the vanishing equilibrium point $E_0 = (0, 0, 0, 0)$ of system (2) is locally asymptotically stable in R_+^4 . Then E_0 is globally asymptotically stable on the region $\omega_0 \subset R_+^4$, where $\omega_0 = \{(x, y, z, w) \in R_+^4 : y > 1\}$.

Proof: Consider the following function:

$$V_0(x, y, z, w) = x + y + z + w,$$

Clearly $V_0: R_+^4 \rightarrow R$ is a C^1 positive definite function.

Now by differentiating V_0 with respect to time t and doing some algebraic manipulation, gives that:

$$\frac{dV_0}{dt} = y(1-y) - (1-u_6)\frac{(1-m)xz}{u_1+x} - (u_4-u_7)yz - (1-u_9)zw - u_3x - u_5y - u_8z - u_{10}w.$$

Now, due to the facts that is mentioned in theorem (1), always $u_6 < 1$, $u_9 < 1$ and $u_7 < u_4$, we get,

$$\frac{dV_0}{dt} < y(1-y) - u_3x - u_5y - u_8z - u_{10}w.$$

Hence $\frac{dV_0}{dt} < 0$ in the region ω_0 and then V_0 is strictly Lyapunov function. Thus we obtain that E_0 is a globally asymptotically stable in the region ω_0 , and the proof is complete.

Theorem (3): Assume that the free predators equilibrium point $E_1 = (\bar{x}, \bar{y}, 0, 0)$ of system (2) is a locally asymptotically stable in R_+^4 . Then E_1 is a globally asymptotically stable on the region $\omega_1 \subset R_+^4$ that satisfies the following conditions:

$$\frac{1}{x} - \frac{(y + \bar{y})}{x} + \frac{u_2u_7}{u_4u_6y} \leq 2 \sqrt{\frac{u_2u_7(\bar{y} - \bar{y}^2)}{u_4u_6x y \bar{y}}} \quad (11 a)$$

$$\bar{y}^2 < \bar{y} \quad (11 b)$$

$$\bar{x} < x \quad (11c)$$

Proof: Consider the following function

$$V_1(x, y, z, w) = c_1 \left(x - \bar{x} - \bar{x} \ln \frac{x}{\bar{x}} \right) + c_2 \left(y - \bar{y} - \bar{y} \ln \frac{y}{\bar{y}} \right) + c_3 z + c_4 w,$$

where: c_1, c_2, c_3 and c_4 are positive constants to be determined.

Clearly $V_1: R_+^4 \rightarrow R$ is a C^1 positive definite function. Now by differentiating V_1 with respect to time t and doing some algebraic manipulation, gives that:

$$\begin{aligned} \frac{dV_1}{dt} = & -c_1 \frac{(\bar{y} - \bar{y}^2)}{x \bar{x}} (x - \bar{x})^2 - c_2 \frac{u_2 \bar{x}}{y \bar{y}} (y - \bar{y})^2 + \left[c_1 \left(\frac{1}{x} - \frac{(y + \bar{y})}{x} \right) + \frac{c_2 u_2}{y} \right] (x - \bar{x})(y - \bar{y}) \\ & - (c_1 - c_3 u_6) \frac{(1-m)xz}{u_1 + x} - (c_2 u_4 - c_3 u_7)(1-m)yz + \left[\frac{c_1(1-m)\bar{x}}{u_1 + x} + c_2 u_4(1-m)\bar{y} - c_3 u_8 \right] z \\ & - (c_3 - c_4 u_9) w z - c_4 u_{10} w. \end{aligned}$$

By chosen $c_1 = 1$, $c_2 = \frac{u_7}{u_4 u_6}$, $c_3 = \frac{1}{u_6}$, $c_4 = \frac{1}{u_6 u_9}$ we get:

$$\begin{aligned} \frac{dV_1}{dt} \leq & - \frac{(\bar{y} - \bar{y}^2)}{x \bar{x}} (x - \bar{x})^2 - \frac{u_7 u_2 \bar{x}}{u_6 u_4 y \bar{y}} (y - \bar{y})^2 + \left[\left(\frac{1}{x} - \frac{(y + \bar{y})}{x} \right) + \frac{u_7 u_2}{u_6 u_4 y} \right] (x - \bar{x})(y - \bar{y}) \\ & - \left[\frac{u_8}{u_6} - \frac{(1-m)\bar{x}}{u_1 + x} + \frac{u_7(1-m)\bar{y}}{u_6} \right] z - \frac{u_{10}}{u_6 u_9} w. \end{aligned}$$

Now by using the conditions (11a), (11b) and (11c) we obtain that:

$$\frac{dV_1}{dt} < - \left[\sqrt{\frac{(\bar{y} - \bar{y}^2)}{x \bar{x}}} (x - \bar{x}) - \sqrt{\frac{u_2 u_7 \bar{x}}{u_6 u_4 y \bar{y}}} (y - \bar{y}) \right] - \left[\frac{u_8}{u_6} - \frac{(1-m)\bar{x}}{u_1 + \bar{x}} + \frac{u_7(1-m)\bar{y}}{u_6} \right] z - \frac{u_{10}}{u_6 u_9} w^2.$$

Clearly, $\frac{dV_1}{dt}$ is negative definite on the region ω_1 due to the local stability condition (8e). Hence V_1 is strictly Lyapunov function. thus E_1 is a globally asymptotically stable on the region ω_1 and the proof is complete.

Theorem (4): Assume that the free top predators' equilibrium point $E_2 = (\hat{x}, \hat{y}, \hat{z}, 0)$ of system (2) is locally asymptotically stable in R_+^4 . Then E_2 is a globally asymptotically stable on any region $\omega_2 \subset R_+^4$ that satisfies the following conditions:

$$\mu_1 \leq 2 \sqrt{\frac{u_1 u_2 u_6 u_7 \hat{x}}{u_4 (u_1 + \hat{x}) y \hat{y}}} (\mu_2 - \mu_3) \quad (12a)$$

$$\hat{y}^2 < \hat{y} \quad (12b)$$

$$\mu_3 < \mu_2,$$

where:

$$\mu_1 = \frac{u_1 u_6}{u_1 + \hat{x}} \left(\frac{1}{x} - \frac{(y + \hat{y})}{x} \right) + \frac{u_2 u_7}{u_4 y}, \quad \mu_2 = \frac{(\hat{y} - \hat{y}^2)}{x \hat{x}}, \quad \mu_3 = \frac{(1-m)\hat{z}}{(u_1 + \hat{x})(u_1 + x)}.$$

Proof: Consider the following function:

$$V_2(x, y, z, w) = c_1 \left(x - \hat{x} - \hat{x} \ln \frac{x}{\hat{x}} \right) + c_2 \left(y - \hat{y} - \hat{y} \ln \frac{y}{\hat{y}} \right) + c_3 \left(z - \hat{z} - \hat{z} \ln \frac{z}{\hat{z}} \right) + c_4 w,$$

where: c_1, c_2, c_3 and c_4 are positive constants to be determined.

Clearly $V_2: R_+^4 \rightarrow R$ is a C^1 positive definite function. Now by differentiating V_2 with respect to time t and doing some algebraic manipulation, gives that:

$$\begin{aligned} \frac{dV_2}{dt} = & -c_1 \left(\frac{(\hat{y} - \hat{y}^2)}{x \hat{x}} - \frac{(1-m)\hat{z}}{(u_1 + \hat{x})(u_1 + x)} \right) (x - \hat{x})^2 - \left(c_1 - \frac{c_3 u_1 u_6}{u_1 + \hat{x}} \right) \frac{(1-m)}{(u_1 + x)} (x - \hat{x})(z - \hat{z}) \\ & + \left[c_1 \left(\frac{1}{x} - \frac{(y + \hat{y})}{x} \right) + \frac{c_2 u_2}{y} \right] (x - \hat{x})(y - \hat{y}) - c_2 \frac{u_2 \hat{x}}{y \hat{y}} (y - \hat{y})^2 \\ & - (c_2 u_4 - c_3 u_7)(1-m)(y - \hat{y})(z - \hat{z}) - (c_3 - c_4 u_9) w z - (c_4 u_{10} - c_3 \hat{z}) w. \end{aligned}$$

By chosen $c_1 = \frac{u_1 u_6}{u_1 + \hat{x}}$, $c_2 = \frac{u_7}{u_4}$, $c_3 = 1$, $c_4 = \frac{1}{u_9}$ we get:

$$\frac{dV_2}{dt} \leq -\frac{u_1 u_6}{u_1 + \hat{x}} \left(\frac{(\hat{y} - \hat{y}^2)}{x \hat{x}} - \frac{(1-m)\hat{z}}{(u_1 + \hat{x})(u_1 + x)} \right) (x - \hat{x})^2 - \frac{u_2 u_7 \hat{x}}{u_4 y \hat{y}} (y - \hat{y})^2 \\ + \left[\frac{u_1 u_6}{u_1 + \hat{x}} \left(\frac{1}{x} - \frac{(y + \hat{y})}{x} \right) + \frac{u_2 u_7}{u_4 y} \right] (x - \hat{x})(y - \hat{y}) - (u_{10} - u_9 \hat{z})w.$$

Now by using the conditions (12 a) and (12 b) we obtain that:

$$\frac{dV_2}{dt} < -\frac{u_1 u_6}{u_1 + \hat{x}} \left[\sqrt{\frac{(\hat{y} - \hat{y}^2)}{x \hat{x}} - \frac{(1-m)\hat{z}}{(u_1 + \hat{x})(u_1 + x)}} (x - \hat{x}) - \sqrt{\frac{u_2 u_7 \hat{x}}{u_4 y \hat{y}}} (y - \hat{y}) \right]^2 - (u_{10} - u_9 \hat{z})w.$$

Clearly, $\frac{dV_2}{dt}$ is negative definite on the region ω_2 due to the conditions (12 c) and the local stability condition (9 e).

Hence V_2 is strictly Lyapunov function. thus E_2 is a globally asymptotically stable on the region ω_2 and the proof is complete.

Theorem (5): Assume that the positive (coexistence) equilibrium point $E_3(x^*, y^*, z^*, w^*)$ of system (2) is locally asymptotically stable in the R_+^4 . Then E_3 is a globally asymptotically stable on any region $\omega_3 \subset R_+^4$ that satisfies the following conditions:

$$\frac{u_1 u_6}{u_1 + x^*} \left(\frac{1}{x} - \frac{(y + y^*)}{x} \right) + \frac{u_2 u_7}{u_4 y} \leq 2 \sqrt{\frac{u_1 u_2 u_6 u_7 x^* (\beta_1 - \beta_2)}{u_4 (u_1 + x^*) y y^*}} \quad (13 a)$$

$$y^{*2} < y^* \quad (13 b)$$

$$z < \frac{u_{10}}{u_9} \quad (13 c)$$

$$w^* < w \quad (13 d)$$

$$\beta_2 < \beta_1, \quad (13 e)$$

where:

$$\beta_1 = \frac{(y^* - y^{*2})}{x x^*}, \quad \beta_2 = \frac{(1-m)z^*}{(u_1 + x^*)(u_1 + x)}.$$

Proof: Consider the following function:

$$V_3(x, y, z, w) = c_1 \left(x - x^* - x^* \ln \frac{x}{x^*} \right) + c_2 \left(y - y^* - y^* \ln \frac{y}{y^*} \right) + c_3 \left(z - z^* - z^* \ln \frac{z}{z^*} \right) \\ + c_4 \left(w - w^* - w^* \ln \frac{w}{w^*} \right),$$

where: c_1, c_2, c_3 and c_4 are positive constants to be determined.

Clearly $V_3: R_+^4 \rightarrow R$ is a C^1 positive definite function. Now differentiating by V_3 with respect to time t and doing some algebraic manipulation, gives that:

$$\frac{dV_3}{dt} = -c_1 \left(\frac{(y^* - y^{*2})}{x x^*} - \frac{(1-m)z^*}{(u_1 + x^*)(u_1 + x)} \right) (x - x^*)^2 - \frac{c_2 u_2 x^*}{y y^*} (y - y^*)^2 \\ + \left[c_1 \left(\frac{1}{x} - \frac{(y + y^*)}{x} \right) + \frac{c_2 u_2}{y} \right] (x - x^*)(y - y^*) - \left(c_1 - \frac{c_3 u_1 u_6}{u_1 + x^*} \right) \frac{(1-m)}{(u_1 + x)} (x - x^*)(z - z^*) \\ - (c_2 u_4 - c_3 u_7)(1-m)(y - y^*)(z - z^*) - (c_3 - c_4 u_9)(z - z^*)(w - w^*).$$

By choosing $c_1 = \frac{u_1 u_6}{u_1 + x^*}$, $c_2 = \frac{u_7}{u_4}$, $c_3 = c_4 = 1$, we get:

$$\frac{dV_3}{dt} \leq -\frac{u_1 u_6}{u_1 + x^*} \left(\frac{(y^* - y^{*2})}{x x^*} - \frac{(1-m)z^*}{(u_1 + x^*)(u_1 + x)} \right) (x - x^*)^2 \\ + \left[\frac{u_1 u_6}{u_1 + x^*} \left(\frac{1}{x} - \frac{(y + y^*)}{x} \right) + \frac{u_2 u_7}{u_4 y} \right] (x - x^*)(y - y^*) \\ - \frac{u_2 u_7 x^*}{u_4 y y^*} (y - y^*)^2 - (1 - u_9)(z - z^*)(w - w^*)$$

Now by using the conditions (13 a) and (13 b) we obtain that:

$$\frac{dV_3}{dt} < - \left[\sqrt{\frac{u_1 u_6}{u_1 + x^*} (\beta_1 - \beta_2) (x - x^*)} - \sqrt{\frac{u_2 u_7 x^*}{u_4 y y^*} (y - y^*)} \right]^2 - (1 - u_9)(z - z^*)(w - w^*).$$

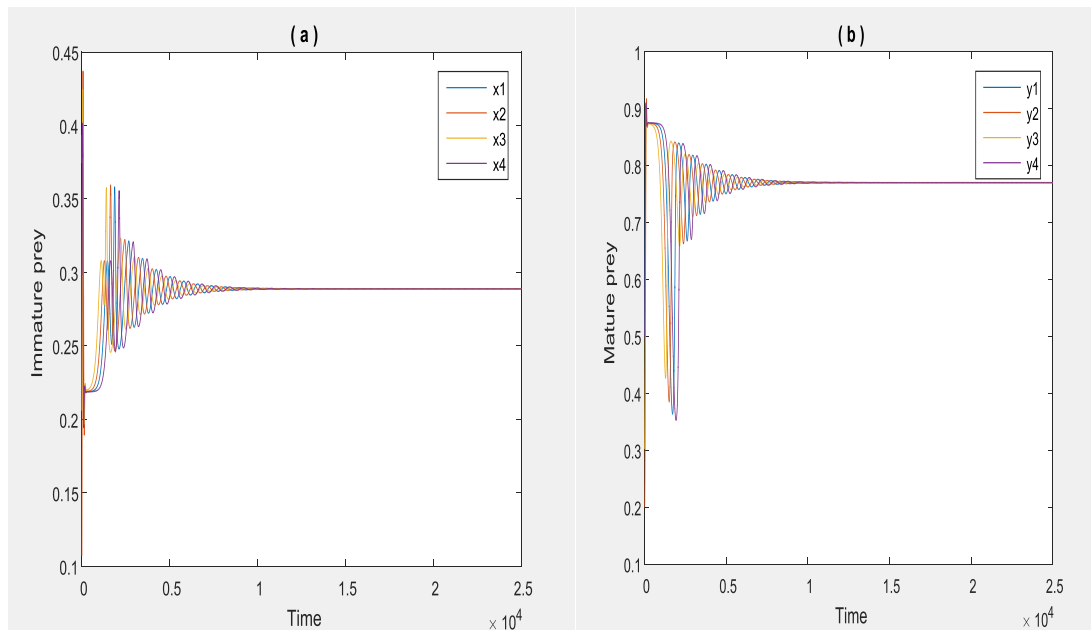
Clearly, $\frac{dV_3}{dt}$ is negative definite on the region ω_3 due to the conditions (13 c), (13 d), (13 e) and the fact that is mentioned in theorem (1), $u_9 < 1$.

Hence V_3 is strictly Lyapunov function. Thus E_3 is a globally asymptotically stable on the region ω_3 and the proof is complete.

Numerical analysis of system (2)

In this section, the dynamical behavior of system (2) is studied numerically for different sets of parameters and different sets of initial points. The objectives of this study are: first investigate the effect of varying the value of each parameter on the dynamical behavior of system (2) and second confirm our obtained analytical results. It is observed that, for the following set of hypothetical parameters that satisfies stability conditions of the positive equilibrium point, system (2) has a globally asymptotically stable positive equilibrium point as shown in Fig. (6.1).

$$u_1 = 0.6, u_2 = 0.4, u_3 = 0.1, u_4 = 0.5, u_5 = 0.1, u_6 = 0.3, \\ u_7 = 0.3, u_8 = 0.1, u_9 = 0.5, u_{10} = 0.1, m = 0.5.$$



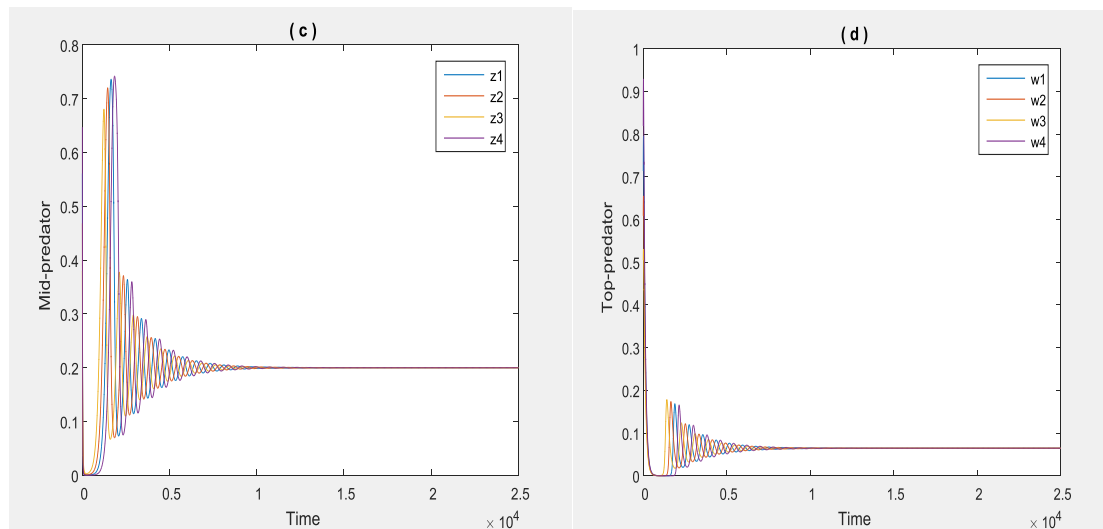


Fig.(6.1):-The time series of the solution of system(2) started from the four different initial points $(0.4, 0.5, 0.6, 0.7)$, $(0.1, 0.2, 0.5, 0.6)$, $(0.2, 0.3, 0.4, 0.5)$, and $(0.3, 0.5, 0.7, 0.8)$, for the data given by(6.1)(a), the trajectories of x as a function of time, (b) the trajectories of y as a function of time, (c) trajectories of z as a function of time, (d) the trajectories of w as a function of time.

Clearly, Fig.(6.1) shows that system(2) has a globally asymptotically stable as the solution of system(2) approaches asymptotically to the positive equilibrium point $E_3 = (0.28, 0.77, 0.2, 0.06)$ starting from four different initial points and this is confirming our obtained analytical results.

Now, in order to discuss the effect of the parameters values of system(2) on the dynamical behavior of the system, the system is solved numerically for the data given in(6.1) with varying one parameter at each time.

By varying the parameter u_1 which represents the half saturation rate of the mid-predator upon the immature prey and keeping the rest of parameters as data given in(6.1) in the range $0.01 \leq u_1 < 2$, it is observed that the solution of system(2) approaches asymptotically to the positive equilibrium point E_3 , as shown in Fig.(6.2), for typical value $u_1 = 0.4$.

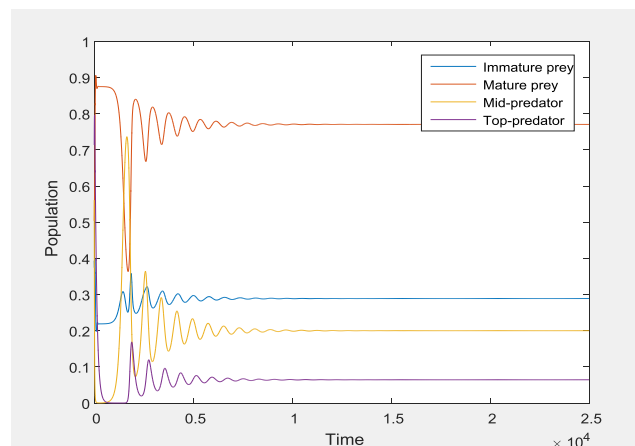


Fig. (6.2):-Time series of the solution of system (2) for the data given in(6.1) with $u_1 = 0.4$, which approaches to $E_3 = (0.28, 0.75, 0.2, 0.07)$ in the interior of R_+^4 .

Now, varying the growth rate parameter of immature prey u_2 and keeping the rest of parameters values as data given in (6.1), it is observed that for $0.1 \leq u_2 < 2$ the solution of system (2) approaches asymptotically to a positive equilibrium point E_3 .

On the other hand varying the natural death rate of immature prey parameter u_3 and keeping the rest of parameters values as data in (6.1), it is observed that for $0.01 \leq u_3 < 0.90$ the solution of system (2) approaches asymptotically to the positive equilibrium point E_3 , as shown in Fig. (6.3) a, for typical value $u_3 = 0.6$, while increasing this parameter for $0.90 \leq u_3 < 1$ causes extinction in the top-predator and the solution of system (2) approaches asymptotically to $E_2 = (\hat{x}, \hat{y}, \hat{z}, 0)$ in the interior of the positive quadrant of xyz – space, as shown in Fig. (6.3) b, for typical value $u_3 = 0.95$.

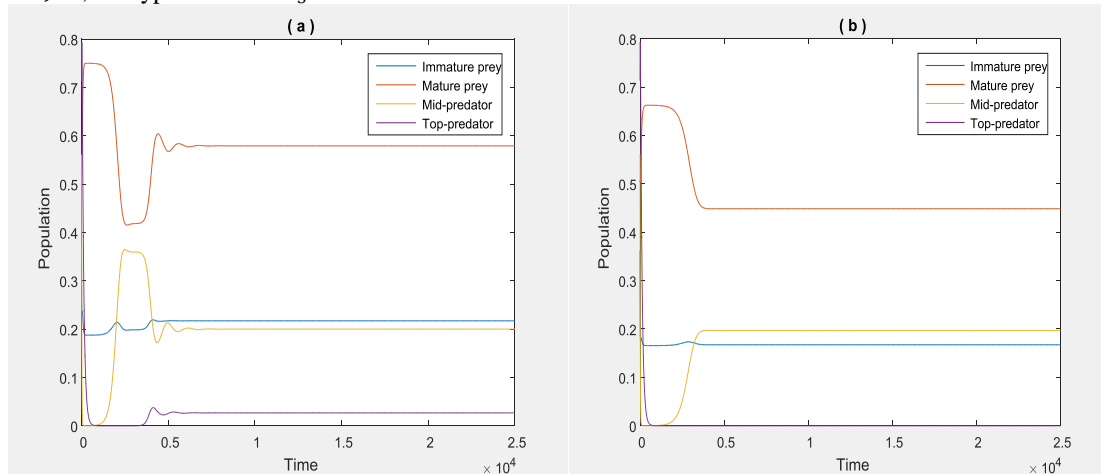
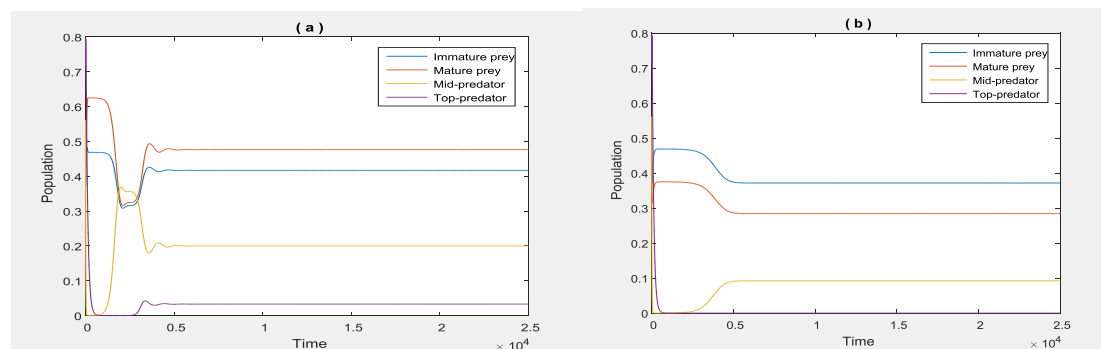
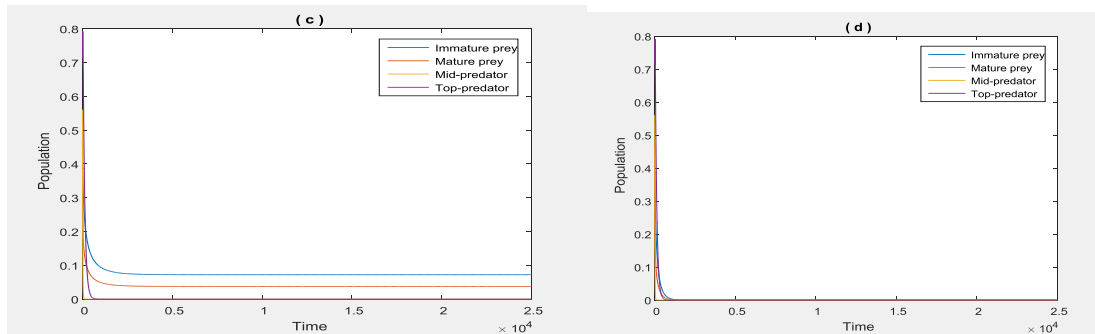


Fig (6.3) (a) :-Time series of the solution of system (2) for the data given by (6.1) with $u_3 = 0.6$, which approaches to $E_3 = (0.21, 0.57, 0.2, 0.02)$ in the interior of R_+^4 , and (b): Time series of the solution of system (2) for the data given by (6.1) with $u_3 = 0.95$, which approaches to $E_2 = (0.16, 0.44, 0.19, 0)$ in the interior of the positive quadrant of xyz – space.

Moreover, varying the parameter ru_4 which represents the predation rate of the mid-predator upon the immature prey, and keeping the rest of parameters values as data given in (6.1), it is observed that for $0.01 \leq u_4 < 1.5$ the solution of system (2) still approaches asymptotically to a positive equilibrium point E_3 .

The effect of varying the mature prey natural death rate parameter u_5 with $0.01 \leq u_5 < 0.41$ and keeping the rest parameters values as data given in (6.1), it is observed that the solution of system (2) approaches asymptotically to a positive equilibrium point E_3 , for typical value $u_5 = 0.3$ as shown in Fig. (6.4) a, however increasing this parameter in the range $0.41 \leq u_5 < 0.58$ causes extinction in the top-predator and the solution of system (2) approaches asymptotically to $E_2 = (\hat{x}, \hat{y}, \hat{z}, 0)$ in the interior of the positive quadrant of xyz – space, as shown in Fig. (6.4) b, for typical value $u_5 = 0.5$, further increasing in the range $0.58 \leq u_5 < 0.8$ causes extinction in the mid-predator and the solution of system (2) approaches asymptotically to the free predators equilibrium point $E_1 = (\bar{x}, \bar{y}, 0, 0)$ in the interior of the positive quadrant of xy – plane, as shown in Fig. (6.4) c, for typical value $u_5 = 0.77$, then more increasing of this parameter in the range $0.8 \leq u_5 < 1$ causes extinction in all species and the solution of system (2) approaches asymptotically to the vanishing equilibrium point $E_0 = (0, 0, 0, 0)$, as shown in Fig (6.4) d, for typical value $u_5 = 0.9$.

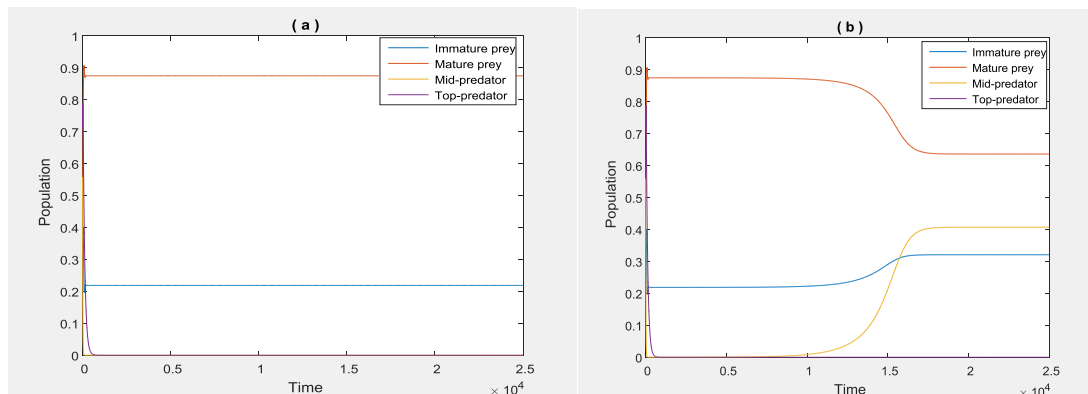


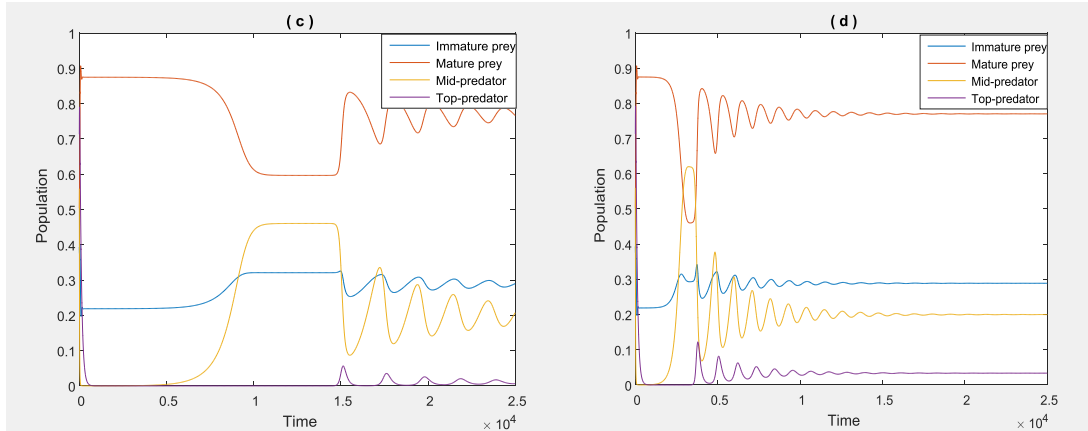


Fig(6.4) (a) :- Time series of the solution of system (2) for the data given by (6.1) with $u_5 = 0.3$, which approaches to $E_3 = (0.41, 0.47, 0.2, 0.03)$ in the interior of R_+^4 , (b) : Time series of the solution of system (2) for the data given by (6.1) with $u_5 = 0.5$, which approaches to $E_2 = (0.37, 0.28, 0.09, 0)$ in the positive quadrant of $xyz - space$, (c) : Time series of the solution of system (2) for the data given by (6.1) with $u_5 = 0.77$, which approaches to $E_1 = (0.07, 0.03, 0, 0)$ in the positive quadrant of $xy - plane$, (d) : Time series of the solution of system (2) for the data given by (6.1) with $u_5 = 0.9$, which approaches to $E_0 = (0, 0, 0, 0)$.

The varying of the parameter u_6 which represents the conversion rate from the mature prey to the mid-predator, and keeping the rest of parameters values as data given in (6.1), it is observed that for $0.01 \leq u_6 < 1$ the solution of system (2) still approaches asymptotically to a positive equilibrium point E_3 .

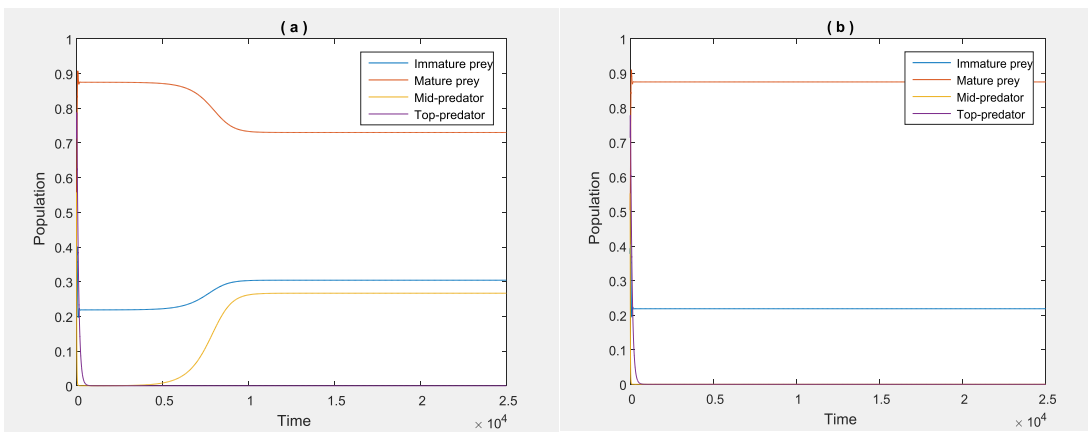
For varying the conversion rate parameter from the mature prey to the mid-predator u_7 , with $0.01 \leq u_7 < 0.15$ the solution of system (2) approaches asymptotically to the positive free predators equilibrium point $E_1 = (\bar{x}, \bar{y}, 0, 0)$ in the interior of the positive quadrant of $xy - plane$, as shown in Fig.(6.5) a , for typical value $u_7 = 0.1$, while for $u_7 = 0.15$ the solution of system (2) approaches asymptotically to $E_2 = (\hat{x}, \hat{y}, \hat{z}, 0)$ in the interior of the positive quadrant of $xyz - space$, as shown in Fig.(6.5) b , which means revival of the mid-predator population, then increasing this parameter in the range $0.15 \leq u_7 < 0.21$ leads revival of the top-predator and a small periodic attractor appears, as shown in Fig.(6.5) c , for typical value $u_7 = 0.16$ for more increasing in the range $0.21 \leq u_7 < 0.5$ the solution of system (2) approaches asymptotically to a positive equilibrium point E_3 , for typical value $u_7 = 0.22$, as shown in Fig.(6.5) d .





Fig(6.5):- (a) Time series of the solution of system (2) for the data given by (6.1) with different values of u_7 , (a) : $E_1 = (0.21, 0.87, 0, 0)$ is a asymptotically stable with $u_7 = 0.1$, (b) : $E_2 = (0.32, 0.63, 0.4, 0)$ is a asymptotically stable with $u_7 = 0.15$, (c) : periodic attractor with $u_7 = 0.16$, (d) : $E_3 = (0.28, 0.77, 0.19, 0.03)$ is a asymptotically stable with $u_7 = 0.22$.

The varying of the mid-predator natural death rate parameter u_8 , it is observed that for $0.01 \leq u_8 < 0.16$ the solution of system (2) approaches asymptotically to the positive equilibrium point E_3 , further increasing of this parameter with $u_8 = 0.16$ which causes extinction in the top-predator and the solution of system (2) approaches asymptotically to $E_2 = (\hat{x}, \hat{y}, \hat{z}, 0)$ in the interior of the positive quadrant of xyz - space, as shown in Fig.(6.6) a , for typical value $u_8 = 0.16$. While for $0.16 < u_8 < 1$ causes the extinction of the mid -predator the solution of system (2) approaches asymptotically $E_1 = (\bar{x}, \bar{y}, 0, 0)$ in the interior of the positive quadrant of xy - plane, as shown in Fig. (6.6) b , for typical value $u_8 = 0.25$.



Fig(6.6) (a):- Time series of the solution of system (2) for the data given by (6.1) with $u_8 = 0.16$, which approaches to $E_2 = (0.30, 0.73, 0.26, 0)$ in the interior of the positive quadrant of xyz - space , (b) : Time series of the solution of system (2) for the data given by (6.1) with $u_8 = 0.25$, which approaches to $E_1 = (0.21, 0.87, 0, 0)$ in the interior of the positive quadrant of xy - plane .

On the other hand, the varying of predation rate parameter of the top-predator upon the mid-predator, for $0.01 \leq u_9 < 0.15$ the solution of system (2) approaches asymptotically to $E_2 = (\hat{x}, \hat{y}, \hat{z}, 0)$ in the interior of the positive quadrant of xyz - space, as shown in Fig.(6.7) a , for typical value $u_9 = 0.1$, while for $0.15 \leq u_9 < 1$ the top-predator population revives and the solution of system (2) approaches asymptotically to a positive equilibrium point E_3 , as shown in Fig.(6.7) b , for typical value $u_9 = 0.5$.

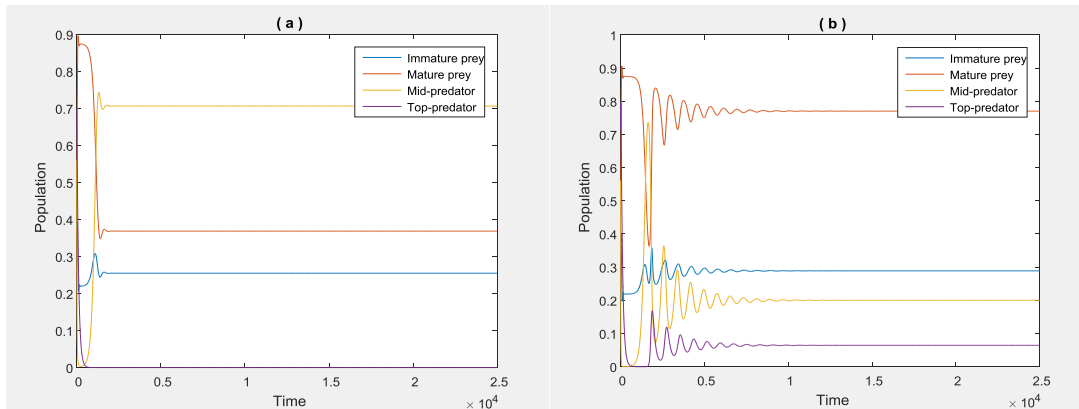


Fig (6.7):- (a) Time series of the solution of system (2) for the data given by(6.1) with $u_9 = 0.1$, which approaches to $E_2 = (0.25, 0.36, 0.7, 0)$ in the interior of the positive quadrant of $xyz - space$, (b):Time series of the solution of system (2) for the data given in(6.1) with $u_9 = 0.5$, which approaches to $E_3 = (0.28, 0.77, 0.2, 0.06)$ in the interior of R_+^4 .

Moreover, increasing the natural death rate of top-predator parameter u_{10} in the range $0.1 \leq u_{10} < 0.35$, the solution of system (2) approaches asymptotically to a positive equilibrium point E_3 , as shown in Fig.(6.8) a , for typical value $u_{10} = 0.3$, while the increasing of this parameter for $0.35 \leq u_{10} < 1$ causes extinction of the top-predator population and the solution of system (2) approaches asymptotically to $E_2 = (\hat{x}, \hat{y}, \hat{z}, 0)$ in the interior of the positive quadrant of $xyz - space$, as shown in Fig.(6.8) b , for typical value $u_{10} = 0.5$.

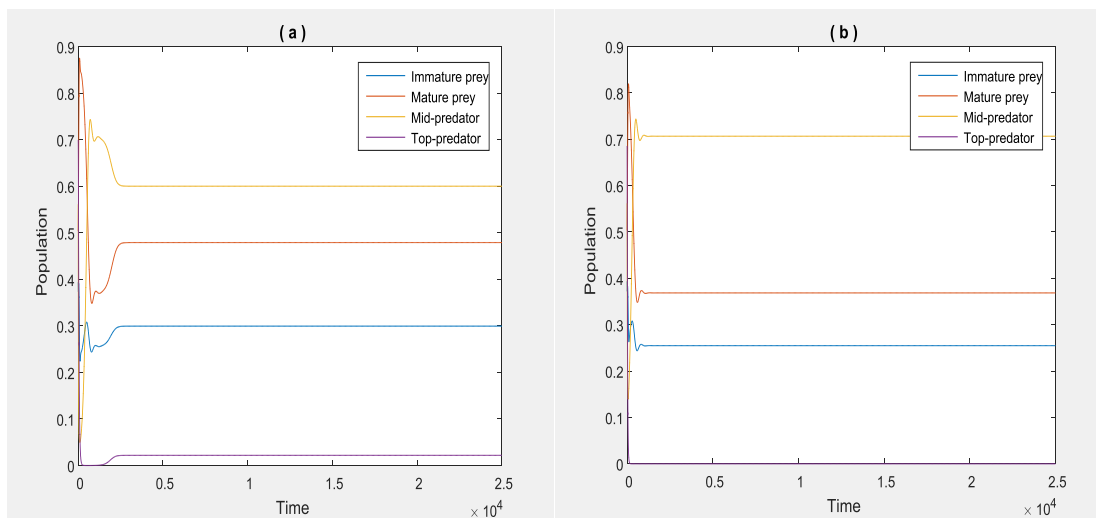
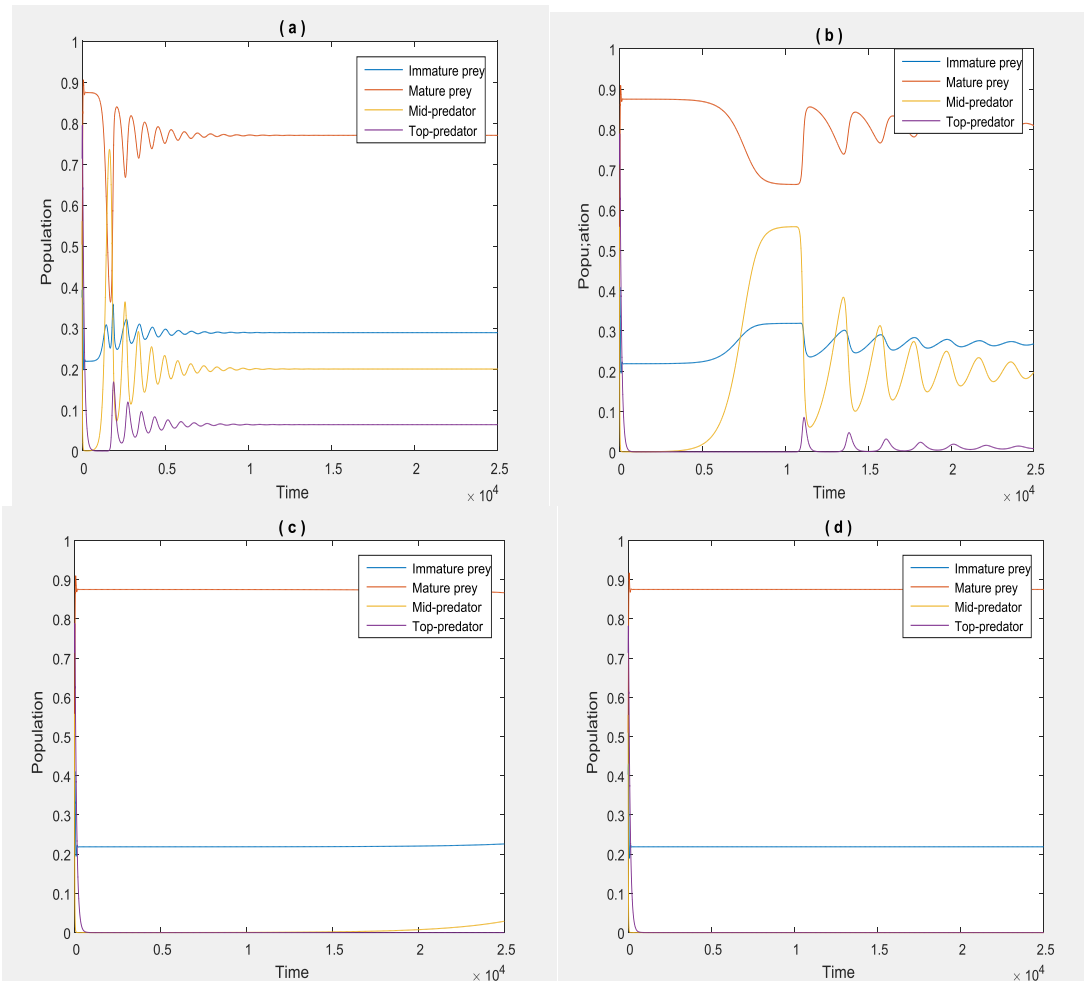


Fig.(6.8):- (a): Time series of the solution of system (2) for the data given by(6.1) with $u_{10} = 0.3$, which approaches to $E_3 = (0.29, 0.47, 0.6, 0.02)$ in the interior of R_+^4 . (b): Time series of the solution of system (2) for the data given by(6.1) with $u_{10} = 0.5$, which approaches to $E_2 = (0.25, 0.36, 0.7, 0)$ in the interior of the positive quadrant of $xyz - space$.

Finally, varying the number of prey inside the refuge parameter m and keeping the rest of parameters values as data given in(6.1) , it is observed that for $0.01 \leq m < 0.63$ the solution of system (2) approaches asymptotically to the positive equilibrium point E_3 , as shown in Fig.(6.9) a , for typical value $m = 0.5$, while increasing this parameter in the range $0.63 \leq m < 0.69$ leads that the solution of system (2) approaches asymptotically to a periodic dynamics in $Int. R_+^4$, as shown in Fig.(6.9) b , for typical value $m = 0.67$, more increasing of this parameter in the range $0.69 \leq m < 0.71$ causes extinction of the top-predator population and the solution of system (2) approaches asymptotically to the free top-predator equilibrium point $E_2 = (\hat{x}, \hat{y}, \hat{z}, 0)$ in the interior of the positive quadrant of $xyz - space$. as shown in Fig.(6.9) c , for typical value $m = 0.7$.and for $0.71 \leq m < 1$ the solution of system (2)

approaches asymptotically to the free predators' equilibrium point $E_1 = (\bar{x}, \bar{y}, 0, 0)$ in the interior of the positive quadrant of xy – plane, as shown in Fig.(6.9)d , for typical value $m = 0.9$.



Fig(6.9): – Time series of the solution of system (2) for the data given by(6.1) with different values of m , (a): $E_3 = (0.28, 0.77, 0.02, 0.06)$ is a asymptotically stable with $m = 0.5$, (b): A small periodic attractor with $m = 0.67$, (c): $E_2 = (0.22, 0.86, 0.02, 0)$ is a asymptotically stable with $m = 0.7$, (d): $E_1 = (0.21, 0.87, 0, 0)$ is a asymptotically stable with $m = 0.9$.

Conclusions and discussion:-

In this chapter, we proposed and analyzed an ecological model that described the dynamical behavior of the food chain real system. The model included four non-linear autonomous differential equations that describe the dynamics of four different population, namely first immature prey (X_1), mature prey (X_2), mid-predator (Y_1) and (Y_2) which is represent the top predator. The boundedness of system (2) has been discussed. The existence conditions of all possible equilibrium points are obtain. The local as well as global stability analyses of these points are carried out. Finally, numerical simulation is used to specific the control set of parameters that affect the dynamics of the system and confirm our obtained analytical results. Therefore system (2) has been solved numerically for different sets of initial points and different sets of parameters starting with the hypothetical set of data given by Eq. (6.1) and the following observations are obtained.

1-System (2) has two types of attractor in $Int. R_+^4$ either a stable point or a periodic attractor.

2-For the set hypothetical parameters value given in Eq. (6.1), the system (2) approaches asymptotically to globally stable positive point $E_3 = (0.28, 0.77, 0.2, 0.06)$. Further, with varying one parameter each time, it is observed

that varying the parameter values, $u_i, i = 1, 2, 4$ and 6 do not have any effect on the dynamical behavior of system (2) and the solution of the system still approaches to positive equilibrium point $E_3 = (x^*, y^*, z^*, w^*)$.

3- As the natural death rate of immature prey u_3 increasing to 0.89 keeping the rest of parameters as in Eq. (6.1), the solution of system (2) approaches to positive equilibrium point E_3 . However if $0.90 \leq u_3 < 1$, then the top predator will face extinction then the trajectory transferred from positive equilibrium point to the equilibrium point $E_2 = (\hat{x}, \hat{y}, \hat{z}, 0)$, thus, the $u_3 = 0.9$ parameter is a bifurcation point.

4- As the natural death rate of mature prey u_5 increasing to 0.40 keeping the rest of parameters as in Eq. (6.1), the solution of system (2) approaches to positive equilibrium E_3 , however if $0.41 \leq u_5 < 0.58$, then the top predator will face extinction then the trajectory transferred from positive equilibrium point to the equilibrium point $E_2 = (\hat{x}, \hat{y}, \hat{z}, 0)$, further increasing in the range $0.58 \leq u_5 < 0.8$ causes the mid-predator faced extinction in and then the trajectory transferred from the free top predator equilibrium point $E_2 = (\hat{x}, \hat{y}, \hat{z}, 0)$, to the free predators equilibrium point $E_1 = (\bar{x}, \bar{y}, 0, 0)$, then more increasing of this parameter in the range $0.8 \leq u_5 < 1$ causes extinction in all species and then the trajectory transferred from equilibrium point $E_1 = (\bar{x}, \bar{y}, 0, 0)$, to the vanishing equilibrium point $E_0 = (0, 0, 0, 0)$, thus, the u_5 parameter when $u_5 = 0.41, u_5 = 0.58$ and $u_5 = 0.8$ is a bifurcation point.

5- As the parameter u_7 which represents the conversion rate from the mature prey to the mid-predator decreasing to 0.15 keeping the rest of parameters as in Eq. (6.1), the solution of system (2) approaches to the positive free predators equilibrium point $E_1 = (\bar{x}, \bar{y}, 0, 0)$, while for the $u_7 = 0.15$, then the trajectory transferred from the free predators equilibrium point $E_1 = (\bar{x}, \bar{y}, 0, 0)$ to $E_2 = (\hat{x}, \hat{y}, \hat{z}, 0)$, which means revival of the mid-predator population, then increasing this parameter in the range $0.15 \leq u_7 < 0.21$ leads revival of the top-predator and the trajectory approaches asymptotically to a periodic dynamics in $\text{Int. } R_+^4$, for more increasing in the range $0.21 \leq u_7 < 0.5$, the trajectory will transferred asymptotically from a periodic dynamics in $\text{Int. } R_+^4$ and then approaches asymptotically stable to a positive equilibrium point $E_3(x^*, y^*, z^*, w^*)$, thus, the parameter u_7 when $u_7 = 0.15$ and $u_7 = 0.21$ is a bifurcation point.

6- As the natural death rate of the mid predator u_8 increasing to 0.15 keeping the rest of parameters as in Eq. (6.1), the solution of system (2) approaches to the positive equilibrium point E_3 , further increasing in the range $0.16 \leq u_8 < 0.18$ causes the top-predator faced extinction and the trajectory transferred from the positive equilibrium point E_3 to the free top predator equilibrium point $E_2 = (\hat{x}, \hat{y}, \hat{z}, 0)$, but for $0.18 \leq u_8 < 1$ causes the mid-predator faced extinction and the trajectory transferred from $E_2 = (\hat{x}, \hat{y}, \hat{z}, 0)$ to $E_1 = (\bar{x}, \bar{y}, 0, 0)$, thus, the parameter u_8 when $u_8 = 0.16$ and $u_8 = 0.18$ is a bifurcation point.

7- As the predation rate parameter of the top-predator upon the mid-predator u_9 increasing to 0.14 keeping the rest of parameters as in Eq. (6.1), the solution of system (2) approaches the free top predator equilibrium point $E_2 = (\hat{x}, \hat{y}, \hat{z}, 0)$, while for $0.15 \leq u_9 < 1$ the top-predator population revives and then the trajectory transferred from the point $E_2 = (\hat{x}, \hat{y}, \hat{z}, 0)$ to the positive equilibrium point E_3 , thus, the parameter $u_9 = 0.15$ is a bifurcation point.

8- As the natural death rate of the top-predator parameter u_{10} increasing in the range $0.1 \leq u_{10} < 0.35$ keeping the rest of parameters as in Eq. (6.1) the solution of system (2) approaches asymptotically to a positive equilibrium point E_3 , while increasing this parameter in the range $0.35 \leq u_{10} < 1$ causes extinction of the top-predator population and then the trajectory transferred from positive equilibrium point to $E_2 = (\hat{x}, \hat{y}, \hat{z}, 0)$, thus, the parameter $u_{10} = 0.35$ is a bifurcation point.

9- As the number of prey inside the refuge parameter m varying in the range $0.01 \leq m < 0.63$ and keeping the rest of parameters values as data given in Eq. (6.1), the solution of system (2) approaches asymptotically to the positive equilibrium point E_3 , while increasing this parameter in the range $0.63 \leq m < 0.69$ leads that the trajectory approaches asymptotically to a periodic dynamics in $\text{Int. } R_+^4$, while increasing this parameter for $0.69 \leq m < 0.71$ causes extinction of the top-predator population and restore the stability and then the trajectory transferred asymptotically from a periodic dynamics in $\text{Int. } R_+^4$ to the stable free top-predator equilibrium point $E_2 = (\hat{x}, \hat{y}, \hat{z}, 0)$, then more increasing of this parameter for $0.71 \leq m$ causes extinction of the mid-predator population and the trajectory transferred from $E_2 = (\hat{x}, \hat{y}, \hat{z}, 0)$, to $E_1 = (\bar{x}, \bar{y}, 0, 0)$, thus, the parameter when $m = 0.63, m = 0.69$ and $m = 0.71$ is a bifurcation point.

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RESEARCH ARTICLE

COMPARATIVE STUDY ON TUNING METHODS FOR SISO SYSTEM.

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IMC-PID, Direct Synthesis, SIMC, Set-point tracking and Disturbance Rejection.

Abstract

This paper presents various tuning methods for a first order process with time delay. The performance of IMC based PID controller is compared with other tuning methods like Modification of IMC-PID for disturbance rejection, Direct synthesis, Direct Synthesis with disturbance rejection (DS-d) and Simple Internal Model Control (SIMC). This work analyses the various parameters of the system. The comparative study of set point tracking and disturbance rejection of a first order process is evaluated using performance criteria like ISE and IAE. From the comparison it was found that IMC-PID provides better results for set-point tracking as well as for the disturbance rejection.

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Introduction:-

The Proportional-integral-derivative (PID) controllers are extensively used in the industrial applications at the regulatory level. The fundamental reason for choosing PID controllers is their relatively simple structure, which can be readily understood and allows them to easily resolve in the real world. However, it is observed that many PID controllers are not properly tuned and exertion has been made to systematically resolve this problem. Therefore, the goal of this work is , to develop a direct approach tuning method of controller from closed loop set point and comparisons has been made for different tuning methods [1].

There are certain goals for tuning methods. The tuning rules should be transparent and easy to memorize. The tuning rules should work on ample range of process. The tuning rules should be well motivated, and preferably model-based and analytically derived.

There are different types of PID controller tuning techniques and they are based on open-loop or closed-loop plant tests and most of these techniques utilize the process gain (k), time constant (τ) and time delay (θ). The vast article on PID controllers includes a wide variety of design and tuning methods based on the performance criteria. The two most oldest and common method of tuning are Zeigler-Nichols (ZN) [2] and Cohen and Coon[3]. These methods were developed to provide a closed-loop response with a quarter decay ratio. The ZN tuning method yields a very good disturbance response for integrating processes[4], but in another way, it results in rather aggressive settings and also give poor performance for process with a dominant delay.

The popular tuning methods for PID controllers which have been used for different kind of processes are Direct Synthesis (DS) [5] and Internal Model Control [6]. The output response based on both the approaches has satisfactory performance and robustness. The main purpose of using IMC-PID is that, it results in a controller with a

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single tuning parameter (λ). For a system which is “minimum phase”, λ is equivalent to a closed loop time constant the “speed of response” of the closed-loop system. IMC-PID provides satisfactory response for set point tracking but it provides poor disturbance rejection for higher order processes in order to overcome this various disturbance rejection techniques were proposed. In the Direct Synthesis (DS) method, the controller design is based on a desired closed-loop transfer function. The next step in the controller is calculated analytically so that the closed-loop set-point matches the desired response. The apparent benefit of using direct synthesis is that the performance requirements are fused directly through the closed loop transfer function. Therefore, the resulting direct synthesis controllers contribute to achieve well for set-point changes. The disturbance rejection is considered to be important so different modification techniques were proposed.

The modification proposed by Dan Chen and Dale E. Seborget *al* for disturbance rejection of Direct synthesis(DS-d)[7] does not provide satisfactory results. The modification for IMC-PID[8] proposed by Shamsuzzoha for disturbance rejection[9] could not produce the expected results.

SKOGESTAD [10] has altered the integral time in SIMC method which is an exceptional remedy for processes with a large time constant [11] in order to improve load disturbance rejection.

Although the PID tuning rule on the basis of IMC and direct synthesis methods gives excellent performance for set-point changes, it shows slow output responses to input (load) disturbances for lag-dominant as well as integrating processes.

Therefore the goal of this paper is to find the IMC-PID procedure for set-point tracking and disturbance rejection to the processes. In this paper, first-order with time delay has been considered and comparisons have been made against Direct Synthesis method, IMC-PID, direct synthesis disturbance rejection (DS-d), modified IMC(Shamsuzzoha) and SIMC method. The various parameters are analysed to obtain the satisfactory results for set point tracking and disturbance rejection.

IMC-PID:-

Internal Model Control based PID controller has the advantage of good set point tracking when it is compared with other methods. The fig 1 shows the basic IMC structure

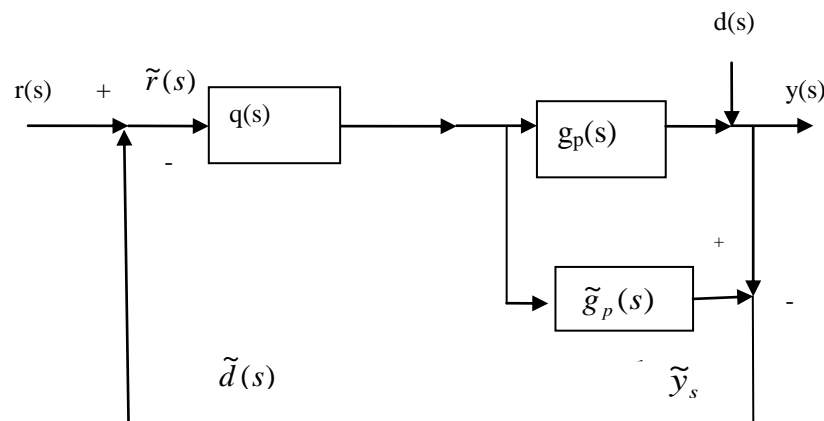


Fig 1:- IMC Structure

It is done by padé approximation technique or using Taylor’s series. In this paper, we have used only the padé approximation technique. IMC-PID provides poor disturbance rejection in order to improve this SIMC have been developed because it provides a better disturbance rejection.

IMC-Based PID Design For A First-Order+ Deadtime Process:-

Consider a first order process with dead time

$$g_p(s) = \frac{ke^{-\theta s}}{\tau s + 1}$$

Where k , τ and θ are the process gain, time constant and time delay.

Step 1: Use a first order padé approximation for the dead time.

$$e^{-\theta s} = \frac{-0.5\theta s + 1}{0.5\theta s + 1}$$

The main reason for using padé approximation is to obtain the simple PID control structure with enhanced performance.

$$\tilde{g}_p(s) = k \left(\frac{-0.5s + 1}{(0.5s + 1)(\tau s + 1)} \right)$$

Separate $\tilde{g}_p(s)$ into invertible and non-invertible parts

$$g_{p-}(s) = k \left(\frac{1}{(0.5s + 1)(\tau s + 1)} \right)$$

$$g_{p+}(s) = (-0.5s + 1)$$

Where $g_{p+}(s)$ and $g_{p-}(s)$ are the non-inverted and inverted parts of the model, respectively.

Step 2: Find the feedback controller

$$q(s) = g_{p-}^{-1}(s) * f(s)$$

Where $f(s) = \frac{1}{(\lambda s + 1)^n}$; $n=1$ (for first order)

$$q(s) = \frac{[(0.5s + 1)(\tau s + 1)]}{k(\lambda s + 1)}$$

Where λ is the filter tuning parameter.

Step 3: The filter $f(s)$ is added and the numerator term to find the PID equivalent,

$$g_c(s) = \frac{[0.5\tau\theta s^2 + (\tau + 0.5\theta)s + 1]}{k(\lambda + 0.5\theta)}$$

Comparing with the standard expression $g_c(s) = k_c \frac{\tau_I \tau_D s^2 + \tau_I s + 1}{\tau_I s + 1}$ we can calculate the values of k_c , τ_I and τ_D .

$$k_c = \frac{2\tau + \theta}{2k(\lambda + \theta)}$$

$$\tau_I = \tau + 0.5 * \theta \quad \tau_D = \frac{\tau\theta}{2\tau + \theta}$$

Rivera et al. (1986) recommend that $\lambda > 0.8\theta$ because of the model uncertainty due to the padé approximation.

MODIFIED IMC-PID (Shamsuzzoha):-

The modified IMC proposed by Shamsuzzoha has the same way of solving as IMC-PID. The variation is only in terms of the tuning parameter. Here we substitute the tuning parameter by 0.6 times of θ .

$$\lambda = 0.6\theta$$

$$k_c = \frac{2\tau + \theta}{3.2k\theta}$$

$$\tau_I = \min\{(\tau + 0.5\theta), 4.8\theta\}$$

$$\tau_D = \frac{\tau\theta}{2\tau + \theta}$$

Direct Synthesis:-

In the direct synthesis method, the design of the controller is based on the closed loop transfer function. It can be done by either padé approximation or Taylor series

Consider a first order system with the dead time

$$\tilde{G} = \frac{k_p e^{-\theta_p s}}{\tau_p s + 1}$$

The exponential term can be approximated using padé approximation and Comparing with the standard form we get the desired parameters as for PID controller as:

$$K_c = \frac{\tau_p + 0.5\theta_p}{k_p(\tau_c + \theta_p)}$$

$$\tau_I = \tau_p + 0.5\theta_p$$

$$\tau_D = \frac{\tau_p \theta_p}{2\tau_p + \theta_p}$$

Direct Synthesis Disturbance Rejection (Ds-d):-

Consider a first order system with delay

$$G_p(s) = \frac{ke^{-\theta s}}{\tau s + 1}$$

The standard form for direct synthesis for a first order system with delay is given by

$$G_p(s) = \frac{\frac{ke^{-\theta s}}{\tau s + 1}}{1 + \frac{ke^{-\theta s}}{\tau s + 1} k_c \left(1 + \frac{1}{\tau_I s} + \tau_D s\right)}$$

The time delay term in the denominator can be approximated by using a first order padé approximation,

$$e^{-\theta s} \approx \frac{1 - 0.5\theta s}{1 + 0.5\theta s}$$

We use padé approximation techniques to obtain the $G_p(s)$, it is then compared with the standard expression to obtain the various parameters k_c, τ_I and τ_D .

By comparing with the standard form, the Parameters obtained are,

$$K_c = \frac{1}{k} \frac{(2\tau\theta + \frac{\theta^2}{2})(3\tau_c + \frac{\theta}{2}) - 2\tau_c^3 - 3\tau_c^2\theta}{2\tau_c^3 + 3\tau_c^2\theta + \frac{\theta^2}{2}(3\tau_c + \frac{\theta}{2})}$$

$$\tau_I = \frac{(2\tau\theta + \frac{\theta^2}{2})(3\tau_c + \frac{\theta}{2}) - 2\tau_c^3 - 3\tau_c^2\theta}{(2\tau + \theta)\theta}$$

$$\tau_D = \frac{3\tau_c^2\tau\theta + \frac{\tau\theta^2}{2}(3\tau_c + \frac{\theta}{2}) - 2(\tau + \theta)\tau_c^3}{(2\tau\theta + \frac{\theta^2}{2})(3\tau_c + \frac{\theta}{2}) - 2\tau_c^3 - 3\tau_c^2\theta}$$

Simple Internal Model Control (Skogestad):-

Skogestad Internal Model Control is the modified method of IMC-PID for improved disturbance rejection than IMC-PID. Here the only parameter used is τ_c . For improved disturbance rejection and good robustness the tuning parameter should be made equal to the value of time delay (θ). Here we consider a first order process with delay and the parameters are obtained using the Half reduction rule[9]

The largest neglected time constant in the denominator is evenly distributed to the effective delay and the smallest retained time constant.

$$G(S) = \frac{\pi_j(-T_{j0}s + 1)}{\pi_i(\tau_{i0}s + 1)} e^{-\theta s}$$

Let the original model be

$$G(S) = \frac{1}{\tau_1 s + 1} e^{-\theta s}$$

Where

$$\tau_1 = \tau_{10} + \tau_{20}/2$$

$$\theta = \theta_0 + \tau_{20}/2 + \sum_{i \geq 3} \tau_{i0} + \sum_j T_{j0} + \frac{h}{2}$$

The delay term is approximated by using Half Rule and from the reduced order of the system the various parameters can be calculated and the PID controller is designed.

$$K_C = \frac{1}{K} \frac{1}{\tau_c + \theta}$$

$$\tau_I = \min(\tau, 4(\tau_c + \theta))$$

Results and Discussion:-

The transfer function considered for our work is $g_p(s) = \frac{e^{-s}}{0.2s + 1}$

Set point tracking:-

Table 1 shows the results obtained for set point tracking for various methods for the above transfer function:

By analyzing the table1, we can see that the settling time is less for IMC-PID when compared with modified IMC, Direct synthesis, Direct synthesis-d and SIMC. Therefore the IMC-PID shows a good set point tracking when compared to other methods. This can also be found from the fig 2.

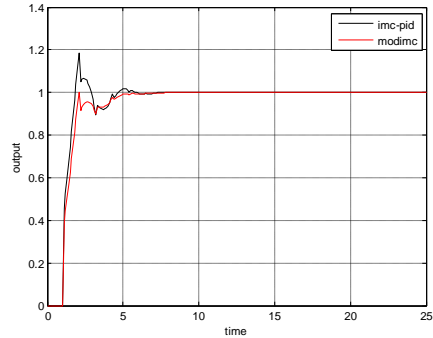
The fig1 shows the response obtained for the transfer function $g_p(s) = \frac{e^{-s}}{0.2s + 1}$ using various methods to track the

set point. The response shows that IMC settles faster when compared to modified IMC, Direct synthesis, direct synthesis-d and SIMC. The settling time varies depending on the tuning parameter λ or τ_c . The errors were found to be less in IMC-PID method compared to modified IMC, Direct synthesis, direct synthesis-d and SIMC.

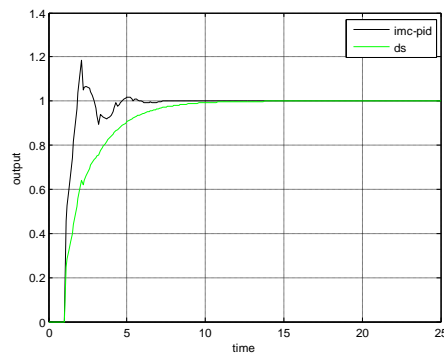
Table 1:-Comparison of various tuning method for Set point tracking.

	Settling Time	K_c	τ_I	τ_D	ISE	IAE	τ_c or λ
IMC	8	0.5185	0.700	0.1429	1.12	1.378	0.850
IMC-Mod	10	0.4375	0.700	0.1429	1.193	1.601	0.600
DS	15	0.280	0.700	0.1429	1.614	2.501	1.500
DS-d	13	0.3179	0.665	0.1095	1.46	2.097	0.380
SIMC	10	0.100	0.200	0.00	3.471	4.486	1.00

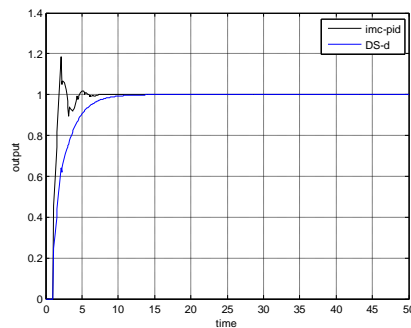
In Fig 1, The time taken for the IMC-PID to settle is less when compared to the modified IMC-PID. The errors are also found to be lesser.

**Fig 1:-**

In Fig 2, Direct Synthesis provides good set point tracking but when compared with IMC-PID for first order system with dead time performance

**Fig 2:-**

In Fig 3, the performance is evaluated by comparing Direct Synthesis for disturbance In rejection with IMC-PID. It is clear that IMC-PID provides good set point tracking.

**Fig 3:-**

In Fig 4, It is clear from the graph that setting time is less for IMC-PID than SIMC.

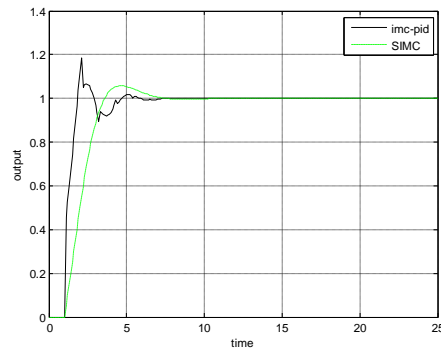


Fig 4:-

Disturbance Rejection:-

The table 2 shows the improved disturbance rejection for the transfer function $g_p(s) = \frac{e^{-s}}{0.2s+1}$ using various methods to obtain the improved disturbance rejection.

The fig 5-8 shows the response obtained for the transfer function $g_p(s) = \frac{e^{-s}}{0.2s+1}$ using various methods for disturbance rejection. The response shows that IMC-PID shows a good set point tracking and provides better disturbance rejection compared to other methods.

Table 2:- Comparison of various tuning methods for Disturbance Rejection.

	Settling Time	K_c	τ_I	τ_D	ISE	IAE	τ_c or λ
IMC	9	0.5185	0.700	0.1429	2.408	3.033	0.850
IMC-Mod	8	0.4375	0.700	0.1429	2.510	3.200	0.600
DS	14	0.280	0.700	0.1429	3.218	4.808	1.500
DS-d	12	0.3179	0.665	0.1095	2.920	4.194	0.380
SIMC	8	0.100	0.200	0.00	1.736	2.243	1.00

In Fig 5, It is clear from the graph that disturbance rejection is better for IMC-PID than modified IMC-PID.

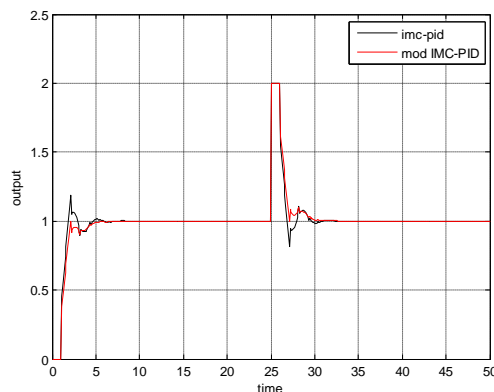


Fig 5:-

In Fig 6, The performance is evaluated by comparing Direct Synthesis with IMC-PID. It is clear that IMC-PID provides good disturbance rejection.

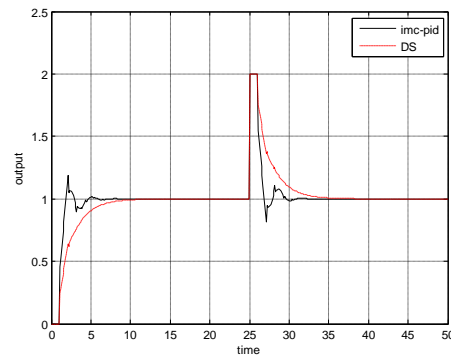


Fig6:-

In Fig 7, The graph depicts that the IMC-PID shows better result for disturbance rejection compared to DS-d.

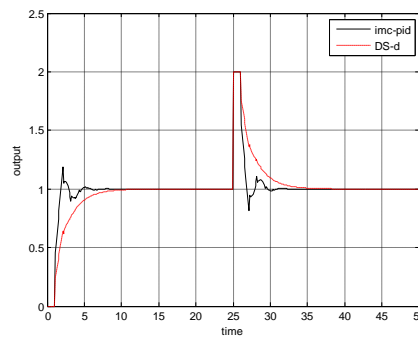


Fig 7:-

In Fig 8, For the first order with time delay process, the IMC-PID shows better result for disturbance rejection compared to SIMC.

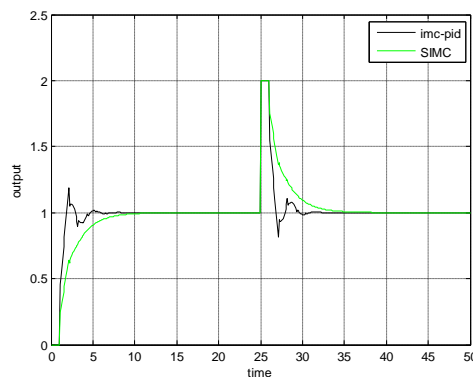


Fig 8:-

Conclusion:-

The IMC-PID provides good set point tracking and disturbance rejection considering the above first order with time delay process when compared to other methods. The DS methods are generally dependent upon the desired closed-loop transfer function for the set-point changes. Subsequently, the resulting Direct Synthesis controllers tend to perform well for the set-point changes in comparison to the disturbance rejection where the result is not satisfactory. Hence, controller design that produces satisfactory results for the disturbance rejection compared to the set-point tracking is considered as a major design problem. Therefore, it is withal to develop a modified direct synthesis

approach based on disturbance rejection. The new design method will be denoted by “DS-d”. Another method for disturbance rejection of IMC-PID was proposed by Mohammad Shamsuzzoha. This method does not produce satisfactory results. Another method was proposed by Sigurd Skogestad known as SIMC. This method provides improved disturbance rejection for the higher order process.

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RESEARCH ARTICLE

ASSESSMENT OF ENDOTHELIAL HEMOSTASIS; SERUM NITRIC OXIDE AND ENDOTHELIN-1 LEVELS IN ISCHEMIC CEREBROVASCULAR STROKE WITH OR WITHOUT TYPE 2 DIABETES.

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endothelial dysfunction; ischemic stroke; endothelin-1; nitric oxide.

Abstract

Background: stroke is the most common cause of disability as well as the second leading cause of mortality worldwide. The role of endothelial dysfunction in stroke is critical. Nitric oxide (NO) and endothelin-1 (ET-1) which are produced in endothelial cells are leading molecules regulating vascular function. **Aim of the work:** the aim of our study was to estimate serum nitric oxide and endothelin-1 levels as biomarkers of endothelial function in ischemic cerebrovascular stroke with or without type 2 diabetes mellitus (T2DM).

Methods: A cross-section study included 100 patients with ischemic stroke who were stratified into two groups according to their fasting blood glucose into non-diabetic group (n=45) and diabetic group (n=55). The levels of serum NO were determined calorimetrically. Serum ET-1 concentrations were also estimated by enzyme immunoassay technique.

Results: Our results showed that non diabetic patients with ischemic stroke had significantly higher values of serum NO compared to diabetic group. On the contrary; there was highly significant elevated serum ET-1 levels in diabetic group compared to non-diabetic group. After adjusted for the traditional risk factors, logistic regression analysis test demonstrated that both serum NO and ET-1 levels were statistically significant predictors of insulin resistance among patients with ischemic stroke. Linear regression analysis test showed that serum NO levels were independently correlated with high density cholesterol (HDL.C) values and systolic blood pressure. Regarding serum ET-1 levels, they were independently correlated with homeostasis model assessment of insulin resistance index (HOMA-IR) and waist/hip ratio (WHR) in patients with ischemic stroke.

Conclusions: Both serum endothelin-1 and nitric oxide levels could be useful diagnostic biomarkers predicting insulin resistance among patients with ischemic stroke.

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Introduction:-

Stroke is defined as rapidly developing clinical symptoms and/or signs of focal, and at times global loss of brain function, with symptoms lasting more than 24 hours or leading to death, with no apparent cause other than that of vascular origin [1]. There is much evidence suggesting that stroke is the most common cause of disability as well as the second leading cause of mortality in the world [2,3].

Remarkably, diabetes is increasing in epidemic proportions globally in the third world countries. In Egypt, the prevalence of diabetes in adults is 14.9%, and there are over 7.8 million cases of diabetes in Egypt in 2015. Moreover, the number of undiagnosed diabetic patients is more than 3.2 million according to International Diabetes Federation (IDF), 2015 [4]. Patients with diabetes always show impaired endothelial function, this is somewhat due to the frequent association of the disease with other cardiovascular risk factors, such as hypertension, obesity, and dyslipidemia as well [5].

Endothelial dysfunction is characterized by a shift in the actions of the endothelium toward reduced vasodilatation, a pro-inflammatory state, and prothrombotic properties as well. There is much evidence suggesting that endothelial dysfunction can play a role in the pathogenesis of ischemic stroke [6]. It is associated with most forms of cardiovascular diseases. Free radicals can damage the endothelium, and leave it overly permeable, allowing toxins to pass into body tissues [7].

NO is a pluripotent regulatory gas in the vascular system. Endothelial derived NOS (eNOS) plays an important role in maintenance of vascular homeostasis, including regulation of cerebral circulation [8].

Endothelin (ET) is a bioactive peptide produced by endothelial cells that can constrict vessels vigorously. It can also enhance the constriction of myocardium and smooth muscles, as well as promote neuroendocrine role [9]. ET is a powerful pro-differentiation agent and a cell-growth factor that can promote cell mitosis, participate in tumor growth and induce mitosis in tumor growth as well [10]. Three types of ET have been identified, ET-1, ET-2 and ET-3, of which ET-1 is the most potent biomolecule. It has been also shown that ET-1 plays a major role in the regulation of the pathogenesis of malignant tumors [11].

The burden of stroke and other cardiovascular diseases is raising in low and middle income countries, where especially, prevention remains the most cost-effective means of intervention. Noteworthy, there are few studies about the correlations between endothelial hemostasis and ischemic cerebrovascular stroke. Thus the aim of our study was to estimate serum nitric oxide and endothelin-1 as biomarkers of endothelial hemostasis in ischemic cerebrovascular stroke with or without type 2 diabetes mellitus (T2DM) and to assess the correlation between them and clinical-laboratory features of stroke.

Subjects and Methods:-

A cross-section study included 100 patients with ischemic stroke recruited from Internal Medicine and Neurology Departments; Zagazig University Hospitals. Patients were stratified into two subgroups according to their fasting blood glucose levels based on the American Diabetes Association (ADA), criteria reported in 2015 (American Diabetes Association, 2015). Those without T2DM (n= 45), and 55 patients with T2DM. All subjects were matched, as regard age, gender, and ethnic origin.

The patients were chosen with the following inclusion and exclusion criteria:

[I] Inclusion criteria: Focal or global neurological deficit lasting > 24 hours on initial neurological evaluation, CT scan of the brain showed evidence of cerebral ischemia.

[II] Exclusion criteria: Non-ischemic etiology such as hemorrhagic stroke (patients with intracerebral hemorrhage, subarachnoid hemorrhage), patients who had received drugs known to affect the level of ET-1 or NO, such as glucocorticoids, NSAID, nitrate, beta-blockers or heparin, cases with history of respiratory disease, cancer, severe hepatic, renal diseases, acute illness, hormonal therapy, any active inflammatory diseases, alcoholism, carotid artery surgery and chronic heart failure were excluded from the study.

All patients in the study were subjected to the following: thorough history taking, full clinical assessment including general and neurological examination. Stroke severity within 72 hours of onset of symptoms was assessed using the National Institute of Health Stroke Scale (NIHSS) [12]. CT scan of the brain was performed for each patient to exclude intracranial hemorrhage and to diagnose cerebral infarction including its site and size. If CT scan was negative, it was repeated after 72 h. The size of the lesion was calculated according to the formula $0.5 \times A \times B \times C$ [where A & B are the largest perpendicular diameters measured on CT and C is the slice thickness (10 mm)] [13]. All scans were performed on Siemens Somatom Balance scanner (Siemens Company, Germany).

Full laboratory investigations were carried out including (Complete blood count, erythrocyte sedimentation rate, blood glucose level, lipid profile, liver and kidney function tests. In addition, ECG, transthoracic echocardiography, and carotid duplex were performed as part of stroke workup.

Blood sampling:-

Blood samples were drawn from all subjects after an overnight fast and divided into 3 portions: 1 ml of whole blood was collected into evacuated tubes containing EDTA, for hematocrit, HbA1c. The second ml of whole blood was collected into evacuated tubes containing potassium oxalate and sodium fluoride (2:1) for fasting blood glucose. Sera were separated immediately from remaining part of the sample and stored at -20°C until analysis.

Biochemical assays:-

We measured fasting blood glucose using the glucose oxidase method (Spinreact, Girona, Spain). Total cholesterol, HDL cholesterol, and triglycerides were measured by routine enzymatic methods (Spinreact, Girona, Spain). LDL cholesterol was calculated by Friedewald formula [14].

Fasting serum insulin concentrations were measured using high-sensitivity enzyme-linked immunosorbant assay (ELISA). The homeostasis model assessment of insulin resistance (HOMA-IR) index was calculated.

Estimation of serum NO and serum ET-1 levels:-

Serum NO levels were measured using colorimetric method of Montgomery and Dymock, by kit purchased from Biodiagnostic (Egypt). Serum ET-1 levels were estimated using a quantitative sandwich ELISA method according to manufacturer's instructions (R&D Minneapolis, MN, USA) ELISA kit.

Ethical consideration:

The ethical committee of Faculty of Medicine, Zagazig University approved our study protocol, and all participants assigned written informed consent.

Statistical analysis:-

Statistical analyses were performed using the Statistical Package for the Social Sciences for Windows (version 19; SPSS Inc., Chicago, IL, USA). Continuous data were expressed using (mean \pm standard deviation) and were analyzed using t test. One-way analysis of variance (ANOVA) test was done to compare different parameters between more than two groups. Receiver operating characteristic (ROC) analysis was performed to assess sensitivities, specificities, area under the curve (AUC), and the cutoff values of NO as well as ET-1 for diagnosis of T2DM among patients with ischemic stroke. Linear regression analysis was done to detect the main predictors of NO and ET-1 in patients with ischemic stroke. Logistic regression analysis was performed to determine the predictor markers associated with T2DM among patients with ischemic stroke. We considered P to be significant at <0.05 .

Results:-

Clinical and biochemical characteristics of the studied groups as summarized in Table 1

There were significant higher values of LDL-cholesterol in female group compared to male group, however, there were non-significant differences regarding other parameters ($p > 0.05$).

Table 1:- clinical, anthropometric and laboratory characteristics of studied subjects.

	Male group (mean \pm SD),(n=60)	Female group (mean \pm SD) ,(n=40)	P
Age (years)	59.45 \pm 14.012	57.13 \pm 17.3	0.686
Systolic blood pressure (mm Hg)	131.76 \pm 7.82	134.03 \pm 11.2	0.239
Diastolic blood pressure (mm Hg)	86.36 \pm 4.58	87.70 \pm 5.33	0.185
Body mass index (kg/m ²)	29.2 \pm 11.15	30.1 \pm 10.60	0.697
Waist/hip ratio	0.962 \pm 0.234	0.98 \pm 0.17	0.569
Total cholesterol (mg/dL)	191.75 \pm 36.6	200.67 \pm 43.9	0.274
Triglycerides (mg/dL)	173.6 \pm 41.1	181.4 \pm 43.47	0.365
LDL -C (mg/dL)	109.85 \pm 26.4	120.4 \pm 24.3	$<0.05^*$
HDL -C (mg/dL)	47.96 \pm 8.47	47.19 \pm 10.3	0.681
Fasting blood glucose (mg/dL)	135.5 \pm 62.2	118.1 \pm 46.7	0.135
Fasting serum insulin	25.26 \pm 22.2	29.9 \pm 27.2	0.352
HOMA-IR	9.24 \pm 8.72	9.24 \pm 8.72	0.467
HbA1c (%)	6.378 \pm 1.55	6.1 \pm 1.32	0.373

HDL-C, high-density lipoprotein-cholesterol; LDL-C, low-density lipoprotein-cholesterol; HOMA-IR, homeostasis model assessments of insulin resistance; HbA1c, hemoglobin A1c cholesterol,* $P < 0.05$.

Clinical anthropometric and laboratory parameters in ischemic stroke subgroups:-

As shown in Table 2, diabetic cases group had significantly higher mean values of systolic blood pressure, fasting blood glucose, fasting serum insulin, HbA1c, HOMA-IR and TG than in non-diabetic group. Moreover, type 2 diabetic patients had significant higher values of body mass index and waist/hip ratio ($p < 0.001$) compared to non-diabetic group. On the contrary, there was a significant lower value of HDL-cholesterol in non-diabetic group compared to diabetic group ($p < 0.001$). There were non-significant differences regarding other parameters ($p > 0.05$).

Table 2:- Laboratory and anthropometric parameters in patients with ischemic stroke stratified according to fasting blood glucose.

	Non-diabetic group (mean \pm SD) (n=45)	Diabetic group (mean \pm SD) (n=55)	P
Age (years)	59.45 \pm 14.01	57.13 \pm 17.3	0.342
Body mass index (kg/m ²)	24.8 \pm 11.7	33.5 \pm 8.42	<0.001*
Waist/hip ratio	0.89 \pm 0.29	1.03 \pm 0.06	<0.001*
Systolic blood pressure (mm Hg)	128.2 \pm 7.176	136.3 \pm 9.4	<0.001*
Diastolic blood pressure (mm Hg)	87.58 \pm 4.92	86.06 \pm 4.8	0.126
Total cholesterol (mg/dL)	206.1 \pm 40.6	182.1 \pm 34.6	<0.001*
Triglycerides (mg/dL)	187.2 \pm 41.1	206.1 \pm 40.7	<0.001*
LDL .C (mg/dL)	112.3 \pm 29.2	115.5 \pm 23.2	0.534
HDL .C (mg/dL)	53.7 \pm 4.33	42.6 \pm 9.14	<0.001*
Fasting blood glucose (mg/dL)	84.6 \pm 7.6	164.4 \pm 54.5	<0.001*
Fasting serum insulin (mg/dL)	131 \pm 5.27	38.6 \pm 27.58	<0.001*
HbA1c (%)	5.4 \pm 1.47	6.9 \pm 1.02	<0.001*
HOMA-IR	2.92 \pm 0.984	13.22 \pm 7.02	<0.001*

HDL-C, high-density lipoprotein-cholesterol; LDL-C, low-density lipoprotein; HOMA-IR, homeostasis model assessments of insulin resistance; HbA1c, hemoglobin A1c cholesterol,* $P < 0.05$.

Comparison of serum NO (μ mol/l) and plasma ET-1(pg/ml) levels among the studied groups (Fig.1, 2)

Non diabetic patient with ischemic stroke had significantly higher values of serum NO ($2.47 \pm 0.19 \mu$ mol/l) compared to diabetic group ($1.94 \pm 0.41 \mu$ mol/l) (**Fig .1**). On the contrary, there was highly significant elevated serum ET-1 level in diabetic group (9.5 ± 2.16 pg/ml) compared to non-diabetic group (6.41 ± 1.98 pg/ml) (**Fig .2**).

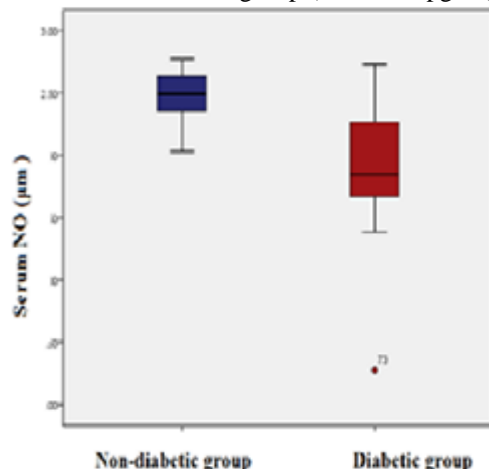


Figure 1

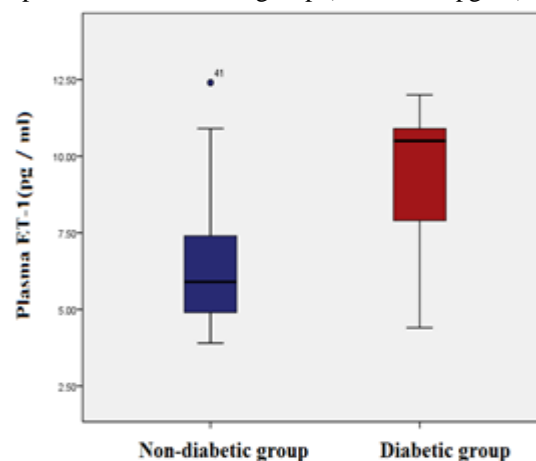


Figure 2

Linear regression analyses in patients with ischemic stroke

Linear regression analysis was done to assess the main independent parameters associated with serum ET-1. Our results showed that, plasma endothelin-1 levels were independently correlated with HOMA-IR and Waist/hip ratio ($p < 0.001$) (**Table 3**).

Table 3:- Linear regression analysis to test the influence of the main independent variables against endothelin-1 pg/ml (dependent variable). * P < 0.05

Model		Unstandardized Coefficients		Standardized Coefficients	t	p	95% C. I	
		β	SE	Beta			Lower Bound	Upper Bound
	(Constant)	2.184	5.107		0.428	0.670	7.939	12.240
	LDL.C	0.002	0.008	0.021	0.259	0.796	0.018	0.013
	HDL.C	-0.030	0.037	0.107	0.810	0.420	0.101	0.044
	Fasting blood glucose	0.001	0.004	0.029	0.357	0.722	0.005	0.009
	HOMA-IR	0.205	0.048	0.580	4.246	<0.001*	0.109	0.293
	Waist/hip ratio	7.103	2.893	0.203	2.455	<0.001*	1.359	12.383
	Body mass index	0.008	0.028	0.030	0.302	0.764	0.064	0.045
	Systolic blood pressure	0.009	0.028	0.034	0.340	0.735	7.939	12.240

Linear regression analyses in patients with ischemic stroke:-

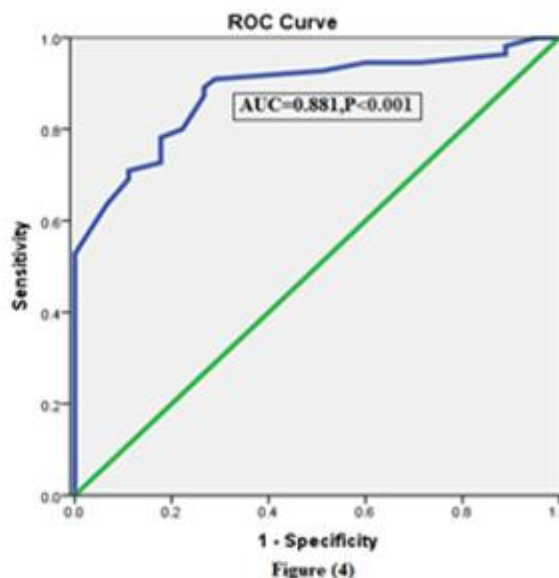
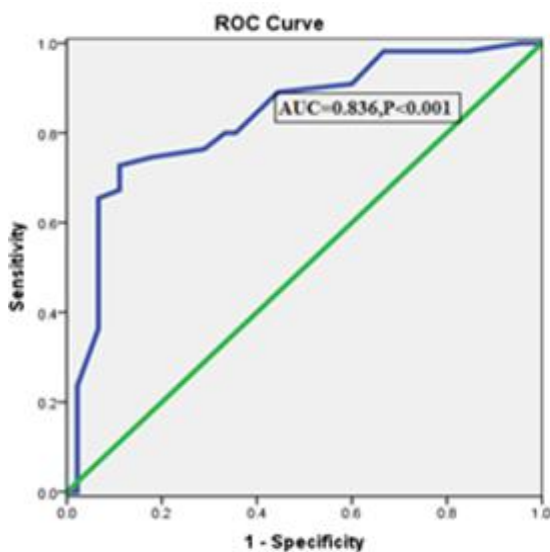
Linear regression analysis was done to assess the main independent parameters associated with serum nitric oxide. Our results showed that serum nitric oxide levels were independently correlated with HDL-cholesterol and systolic blood pressure ($p < 0.001$) (**Table 4**).

Table 4:- Linear regression analysis to test the influence of the main independent variables against serum nitric oxide (μm) (dependent variable).

Model		Unstandardized Coefficients		Standardized Coefficients	t	p	95% C. I	
		β	SE	Beta			Lower Bound	Upper Bound
	(Constant)	0.118	0.077		1.522	0.131	0.272	0.036
	LDL.C	0.000	0.000	0.043	1.919	0.058	0.000	0.000
	HDL.C	0.016	0.001	1.007	27.74	<0.001*	0.015	0.017
	Fasting blood glucose	7.408	0.000	0.029	1.339	0.184	0.000	0.000
	HOMA-IR	0.000	0.001	0.013	0.355	0.723	0.002	0.001
	Waist/hip ratio	0.058	0.044	0.030	1.326	0.188	0.045	0.029
	Body mass index	0.000	0.000	0.024	0.887	0.378	0.001	0.000
	Systolic blood pressure	0.001	0.000	0.067	2.472	<0.001*	0.000	0.002

Accuracy of plasma endothelin-1 and serum nitric oxide for discriminating diabetic from non-diabetic patients with ischemic stroke by ROC Analyses:-

The cut-off values of plasma ET-1 and serum nitric oxide levels were determined by ROC to discriminate diabetic from non-diabetic patients with ischemic stroke; they were 8.65 and 2.3, and the AUC were 0.836(95% CI 7.45–9.17, $P < 0.001$) and 0.881 (95% CI 0.814–0.948, $p < 0.001$, respectively). The sensitivities and the specificities of endothelin-1 were 72.9 % and 99.8 % and nitric oxide serum levels were 80.1% and 77.8%, respectively (Fig. 3, 4).



Logistic regression analysis evaluating the association of serum NO and ET-1 with T2DM among patients with ischemic stroke (Table 5)

After adjusted for the traditional risk factors, logistic regression analysis test was done to evaluate the predictors of insulin resistance among patients with ischemic stroke, serum NO and serum ET-1 were statistically significant predictors of insulin resistance among patients with ischemic stroke ($p < 0.001$).

Table 5:- Logistic regression analysis of the main clinical and biochemical predictors of diabetic patients with ischemic stroke.

		B	S.E.	Wald	P	odds	95% C.I	
							Lower	Upper
	Constant	5.246	5.757	0.830	0.362	189.890		
	Serum ET-1(pg/ml)	.275	.136	4.114	0.043	1.317	1.009	1.718
	Serum NO ($\mu\text{mol/l}$)	13.075	3.710	12.419	0.000	0.000	0.000	0.003
	Body mass index (kg/m^2)	0.030-	0.043	0.484	0.487	0.970	0.891	1.056
	LDL-Cholesterol (mg/dl)	0.006-	0.012	0.274	0.601	0.994	0.971	1.017

ET-1, endothelin-1 ; NO, nitric oxide levels ; LDL-C, low-density lipoprotein* $P < 0.05$.

Discussion:-

There was great evidence that the etiology of ischemic stroke is known to be a multifactorial disorder in addition to the commonly accepted risk factors [15]. Additionally, increasing evidences suggested endothelial dysfunction role in the pathophysiology of ischemic stroke. Endothelial dysfunction can be found to be both macro- and microvascular, which leads to an imbalance between vasodilation and vasoconstriction capability in homeostasis regulation [16-19]. ET-1 being a potent vasoconstrictor, increases, while NO, as vasodilator substance, decreases in endothelial dysfunction. The vasoconstrictive effect of ET-1 is more prominent on microvascular wall including subendocardial compared to macrovascular. Moreover, an increase in ET-1 causes an endothelial dysfunction [20-21].

Patients with diabetes mellitus are at markedly increased risk of death due to cerebrovascular disease, and this is true of both T1DM and T2DM [22]. In fact, T2DM patients make up the vast majority of diabetic stroke (97% in the Nurses' Health Study) [23].

Early diagnosis and management of endothelial dysfunction in ischemic stroke especially in diabetics is very important. To address this need, we have focused on biomarker of endothelial dysfunction, especially NO and ET-1. Thus the aim of our study was to investigate the role NO and ET-1 as biomarkers of endothelial hemostasis in ischemic cerebrovascular stroke with or without T2DM and to assess their correlation with clinical-laboratory features of stroke.

Our results showed that, among patients with ischemic stroke, 60 % were males and 40% were females. In comparison between male and female group, our findings confirmed that female group had significantly higher values of LDL-cholesterol. Otherwise all other parameters showed non-significant difference between both groups.

In accordance to our finding, a prospective population-based study, showed an increased relative risk for developing stroke of 1.5 to 2 fold in men and 2 to 6.5 fold in women. This increased risk is seen even early after diagnosis of stroke in newly treated T2DM patients [23].

In our research, we found significant higher levels of systolic blood pressure, fasting serum insulin, fasting blood glucose, HOMA-IR, HbA1c, total cholesterol, triglycerides, body mass index, and waist/hip ratio in T2DM group compared to non-diabetic group.

Similar finding was observed also by **Jeerakathil and his colleagues**; who found higher relative risk of stroke in the T2DM group compared to general population [25]. This can be explained as both diabetic as well as obese patients usually consume a high-calorie diet rich in macronutrients which induce vascular abnormalities [26]. Indeed, protein, lipid, and glucose loads are associated with a marked production of reactive oxygen species (ROS) and high-fat meals, with impaired endothelium-dependent vasodilation [27]. Similar to our finding, several studies detected the impairment of endothelial function in relation to blood glucose levels fluctuation, HbA1c, and insulin resistance [28,29]. In contrast to our results, the study of **Zampetaki et al.**, revealed significantly higher value of low-density cholesterol levels in association with endothelial function in [30].

Increasing evidence suggests that diabetes mellitus, hypercholesterolemia, hypertension, and smoking lead to atherosclerosis as well as endothelial dysfunction [31, 32]. The main finding in the current study that non-diabetic patient with ischemic stroke had significantly higher values of serum NO compared to diabetic group. On the contrary; there were highly significant elevated serum ET-1 levels in diabetic group as compared to non-diabetic group. In agreement with our results, **Manrique et al.** observed that patients with diabetes had endothelial dysfunction. There is a general consensus that hyperglycemia and diabetes lead to impair NO production and damaged vasodilatory activity [33]. **Piconi et al.**, found that chronic hyperglycemia leads to weak integrity and apoptosis of endothelial cell [34]. In light of this fact, several clinical studies have demonstrated an impaired endothelium-dependent vasodilation in conduit or resistance vessels of T2DM patients [35]. ROS produced in diabetic patients also contributes to endothelial injury and impair endothelial repair. They are directly cytotoxic for endothelial cells, react with NO, decrease NO bioavailability, and form peroxynitrite anions which act as powerful oxidants. Lifestyle modification has a potential to increase the number of endothelial progenitor cells and improve their migratory capacity, helping to repair the damaged endothelium [36-38].

The results presented herein are innovative; as this study performs a robust evaluation of NO and ET-1 as diagnostic biomarker of endothelial dysfunction. Both serum ET-1 and nitric oxide could be useful diagnostic biomarkers detect T2DM among patients with ischemic stroke. Interestingly; the power of NO was sensitive and specific parallel to ET-1. Our study explored that after adjusted for the traditional risk factors, serum NO and ET-1 levels were statistically significant predictor of ischemic stroke among patients with ischemic stroke. Endothelial dysfunction seems to precede the development of diabetes, as impaired endothelium-dependent vasodilation was observed in healthy non-diabetic subjects who have a first degree relative with T2DM [32], as well as in subjects with impaired glucose tolerance [31].

Our results showed that, serum NO levels were independently correlated with HDL-cholesterol and systolic blood pressure. In agreement with our results, **Boden and Shulman** found the progression of insulin resistance to T2DM parallels to the progression of endothelial dysfunction to atherosclerosis. Moreover, insulin resistance is closely linked with visceral adiposity, and early data suggested that free fatty acids were responsible for this association [37]. The eNOS acts as a protective role in ischemic stroke by inhibiting platelet aggregation and leukocyte adhesion to vascular endothelium, and thus protects against vascular pathological changes such as vascular muscle cell growth and proliferation [38]. Most importantly, eNOS also protects from atherosclerosis, which is an independent risk factor for ischemic stroke incidence [39-41].

Our results showed that ET-1 concentrations were independently correlated with HOMA-IR and Waist/hip ratio., findings which were similar to results of **Romero et al.** Who observed that ET-1 may contribute to the development of endothelial dysfunction, and consequently insulin resistance by increasing the production of ROS, mainly superoxide anion, in the vasculature. This is mainly dependent upon the activation of NADPH oxidase

protein expression and activity [42]. Similar to our finding, **Lee and Poh** reported in their study that ET-1 values were positively correlated with insulin resistance as well as endothelial dysfunction [43].

Conclusion:-

The results of this study reached to a conclusion that serum ET-1 levels were significantly elevated in T2DM patients with ischemic stroke. While serum NO levels were decreased, hence Serum NO and ET-1 could be early predictors of insulin resistance in patients with ischemic stroke.

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RESEARCH ARTICLE

EFFECT OF MUSIC AND PERSONALITY ON SEMANTIC MEMORY RETRIEVAL IN YOUNG ADULTS.

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Semantic, Memory, retrieval, music, introvert, extrovert.

Abstract

Background -Listening to music while studying/working has been a commonly observed trait amongst youngsters. Effect of music and personality on short term memory and episodic memory has been studied but there is less evidence regarding the recall of semantic memory. Hence the purpose of the study was to explore the effect of different types of music and personality type on Semantic Memory Retrieval. **Methods**-In this cross-sectional study, asymptotic adults aged between 18-24yrs were administered a questionnaire to determine their personality type. Later, they were assessed on questions related to semantic memory recall in three different conditions namely; Silence, with Lyrical music and with Instrumental music. Total Time taken and the Total Score in all three conditions were analyzed. **Conclusion**- The mean of the Total Time and score was significantly more for Silence as compared to music condition. However they performed the fastest while instrumental music was played. There was no significant difference in Total Time as well as Total Score for both introverts and extroverts

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Introduction:-

Memory gives us the ability to encode, store and recall information from our brain that helps us to function optimally. Semantic memory which is a part of explicit long term memory constitutes the knowledge of basic concepts or facts. It differs from the episodic memory related to the events (Tulving, 1972). Psychologists describe semantic memory as the general knowledge/information one has acquired over a period of time. (eg. Mumbai is in India, milk is white in colour etc.) Semantic memory is necessary while using language and a variety of cognitive activities. Recognition and use of objects includes association and combination of concepts, all of which are a part of the semantic memory. Thus it plays a crucial role in learning.

Review of literature suggests that music and type of personality are two important factors affecting memory recall in cognitive tasks. But the results are conflicting. Review of effect of background music on learning showed inconsistent findings (Scholz D.,2011).A autobiographical memory recall in patients with dementia concluded that both familiar and non familiar music facilitated memory retrieval. (Foster NA and Valentine ER, 2001) But a study on Recall of word list suggested that encoding stage was influenced by music but not recall (Konantz E.,2012).Later another study stated that music has a negative effect on recall of words and not digits. (Jameson C., 2013).Similarly personality traits also showed influence on memory recall. Music was found distracting for introverts and extroverts

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performing cognitive tasks and introverts performed the cognitive tasks worse than extroverts in presence of music. (Furnham & Bradley, 1997, Furnham 2002) But complexity of music had not much effect on word memory task in introverts and extroverts (Matias L, 2014). Thus the evidence is more on short term memory and episodic (autobiographical) memory. However there is lack of clear understanding of effect of music and personality on semantic memory retrieval.

Today, music has become an integral part of the youngsters' lives in metropolitan city like Mumbai in India. Even while studying/working, people tend to listen to music. Some of them listen to lyrical music while others prefer instrumental music. Hence the objective of our study was to assess the effect of lyrical and instrumental music and personality on semantic memory retrieval in young adults.

Material & Methods:-

Approval of the study was obtained from the Institutional Research Committee. This cross-sectional study was conducted during 4 months (from November 2015 to February 2016) at various colleges in Mumbai and Gujarat. Asymptomatic adults, with level of education above Grade 10, fluency in English language and with intact vision and hearing ability were eligible to participate. Any person with known musculoskeletal, neurological or psychiatric illness or with absence of vision and hearing ability were excluded from this study. A total of 111 participants were a part of this study. The study included the individuals in the age group of 18-24 yrs.

After taking informed consent, the basic information was collected including participant's preference to listening to music, their mode of listening to music and the type of music they preferred to hear. Short-scale Eysenck Personality Questionnaire – Revised (Short-scale EPQ-R) was administered and the psychologist determined the personality type of each participant.

The participants were made to answer 3 sets of 15 questions each, in 3 different conditions, namely in silence, when lyrical music was played and when instrumental music was played. High-tempo lyrical music was chosen as one of the condition. The low-tempo instrumental music was chosen as the second condition. All the questions in the test were validated by a psychologist. The participants were instructed to write the answers on blank sheets which were provided to them. The questions were based on information stored in semantic memory. Although each question was different in all 3 sets, the basic criteria remained the same. For example, if one question was 'Which is the 5th month of the year', in the other set the question would be 'Which is the 7th month of the year?'.

The questions were displayed on a computer/laptop screen. Music was played over a headphone. For the lyrical music, a high tempo popular hindi movie song (Sooraj dooba hain from movie Roy) was chosen. For the instrumental music a low tempo song (relaxdaily N°062) was chosen. The same set of songs was played for each subject. The same headphone was used for everyone. The volume of the songs was also constant for every subject. The sequences of all 3 set of questions were also shuffled amongst all the participants.

For each subject, the time taken for them to complete each of the questionnaire and their score in all three conditions were analysed.

Result:-

Table I:- Demographic Characteristics.

Characteristics	Total participants (n=111)
Age (year)	20.73(mean)
Gender (M: F)	41:70
Dominance (R:L)	91:9
Personality traits	Extroverts 72 Introverts 39
Preference to listening to music while studying/working	Yes : 65 No : 46
Mode of listening (%)*	headphones/earphones : 66 and loudspeaker : 34
Type of music preference* (%)	Lyrical : 80 Instrumental : 20
*For the ones who preferred listening to music while studying/working (n=65)	

Table II:- Comparison of time and score of all participants across 3 conditions(silence,lyrical &instrumental) .

	Mean±SD	Mean±SD	p value
	Silence	Lyrical	
Time (sec)	404.43 ± 108.74	365.55 ± 92.36	0.01*
Score (Out of 38)	32.66 ± 3.00	31.65 ± 3.34	0.06
	Silence	Instrumental	p value
Time (sec)	404.43 ± 108.74	346.04 ± 90.99	0.001*
Score (Out of 38)	32.66 ± 3.00	31.28 ± 3.60	0.0083*
	Lyrical	Instrumental	p value
Time (sec)	365.55 ± 92.362	346.04 ± 90.99	0.11
Score (Out of 38)	31.65 ± 3.34	31.28 ± 3.60	0.42
SD, Standard Deviation; Statistical calculation and comparison between the groups by Kruskal-Wallis Test (Nonparametric ANOVA) and Dunn's Multiple Comparisons Test;*p value <0.05, considered significant			

Statistical analysis shows that the mean of the Total Time was significantly more for Silence as compared to Instrumental music condition as well as for the Lyrical music condition. For the Total Score, mean in Silence was significantly more as compared to Instrumental music condition.

Table III:- Comparison of time and score between Introverts and Extroverts.

	Introverts (n=39)	Extroverts (n=72)	p value
	Mean±SD	Mean±SD	
Silence			
Time (sec)	379.07 ± 78.86	418.22 ± 120.29	0.16
Score (Out of 38)	32.64 ± 2.82	32.68 ± 3.11	0.71
Lyrical			
Time (sec)	350.61 ± 76.21	373.65 ± 99.59	0.46
Score (Out of 38)	31.74 ± 3.30	31.61 ± 3.38	0.76
Instrumental			
Time (sec)	330.07 ± 79.58	354.69 ± 96.03	0.15
Score (Out of 38)	31.41 ± 2.87	31.22 ± 3.95	0.95
SD, Standard Deviation; Statistical calculation and comparison between the groups by Mann-Whitney Test; *p value <0.05, considered significant			

On comparing the mean values of Total Time and Total Score between introverts and extroverts in each condition, the two-tailed P value was considered not significant.

Discussion:-

On comparing the semantic memory recalling capacity in three different conditions, it was found that participants took the longest to complete the questionnaire in silence but scored the best in it. Thus the result of the present study showed that silence is the best environment for studying/working. We observed that the participants were able to focus more readily as there was least amount of distraction around while they performed in silence. This is in agreement with the previous studies on cognitive tasks where participants performed best in silence(Furnham & Bradley, 1997, Konantz E, 2012,Jameson C, 2013).

In both the music conditions,time taken to complete the test was shorter compared to silence.But the scores dropped when music was played as compared to the scores in silence. When comparison was made between lyrical music condition and instrumental music condition, participants performed better when lyrical music was played. A previous study also showed that word recall was better when vocal music was played as compared to when instrumental music was played(Jameson C,2013).

In the current study, we did not test just the word recall but tested the conceptual knowledge. The test included various domains like knowledge about living, non living things as well as their visual and non visual attributes. It has been found that there are different areas in brain that get activated in memory retrieval(Martin A. et al.1995,2001, Thompson-Schill et al, 1999,2003 Martin A. 2007). There is a structured system along a posteroanterior axis in the temporal cortex, anterior temporal lobe being responsible for abstract knowledge and generalisation.(Roger T.T.et al 2006 ,Yee E.et al 2013) Memory and music possess a unique connection as the

auditory processing area is also present in the temporal lobe. A PET study revealed that the verbal and musical semantic processes activated a common network extending throughout the left temporal neocortex. (Groussard et al., 2010) Hence it is difficult for one to perform the memory recall to their optimum level with music.

Based on the Short-scale Eysenck Personality Questionnaire – Revised, each participant was classified as being an introvert or an extrovert (Eysenck, S. B. 1985). In the participants, 35% were introverts and 65% were extroverts. Introverts performed the best during silence and worst when instrumental music was played which is in agreement with previous studies. But in our study, extroverts also performed the best during silence and worst when instrumental music was played, thereby differing with the results of previous studies. (Furnham & Bradley, 1997, Mistry H. 2015) The reason behind this varied result may be because the tasks in the previous studies involved working memory whereas this study demanded recall from long term part of the memory.

The study suggests that silence is the best kind of environment one can study/work in as it aids in maximum semantic memory retrieval for majority of the individuals. For both introverts and extroverts as well, silence is the best environment to be preferred while studying/working. Even if the time required was more in silence in the present study, the difference was very less in terms of minutes.

Further research should be done by investigating semantic memory recall on the basis of the preference of the participants to listening to music while they study/work as well as when music is played in the background versus when it is played over headphones.

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RESEARCH ARTICLE

THE ROLE OF HIP HOP IN ADDRESSING SOCIO-POLITICAL SITUATIONS IN INDIA- A PERCEPTION STUDY.

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Abstract

Music has always occupied a central place in the imagination of Indians. Over the years, the audience started welcoming different genres of music. Hip Hop, a popular form of music which evolved itself from oppression has its share of audience in India. The freedom to "express" oneself without class barriers and conventions popularise this genre. In India, non-commercial Hip Hop has a serious take on social and political issues through strong lyrics. Hip Hop artists are addressing the issues that are more relevant to the times rather than blindly rapping about living in a place with gun fights and racism which imitates the scenario in the West. Issues of poverty, lack of employment, environmental problems, drugs, Kashmir, corporate influence, caste barriers, child care etcetera which are severe issues to be considered in India, are approached through Hip Hop which acts as protest music. This paper analyses the Role of Hip Hop in addressing the social and political issues in India. By analysing the lyrical content qualitatively and by using an online this paper examines how the audience perceives and respond to non-commercial Hip Hop which has socially and politically charged lyrics.

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Introduction:-

Music plays a central role in people's lives. Lull (1985) suggested that people select music for particular reasons and are personally involved with their favourite types of music on physical and cognitive levels. Musical content (e.g., rhythm, tempo, melodies) amplifies the message of songs. It "is designed to invoke the emotional state, the state of consciousness, or mental state as it is experienced with the self and others" (Chesebro, Foulger, Nachman, & Yannelli, 1985, page 118).

Rap music is considered as one of the most controversial forms of popular music to find its way into the mainstream popular culture as far as texts go. However, rap has an interesting history. It is said to have evolved from "Reggae", one of the popular music genres in the Rastafarian subculture attributed to the Jamaicans. The Rastafarian subculture had its religion which included music and lifestyle. As far as the music goes, it sounded like a "fusion of rock, soul and calypso underscored by a heavy syncopated bass guitar line and drums" (Neil.J.Savishinsky). This island music espoused a lot of social, political and religious ideologies which expanded the repertoire of transnational music culture. While the music created ripples, the Rastafarian culture was known for its 'set-against-the-flow' style. The music started at an off-beat, which itself is a convention broken in the first place. The idea was to convey the

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disagreement to rules and conventions. From there, different types of subcultures were born, and rap is one among them.

The early 2000s saw a sudden surge in Indian rapping population who mainly rapped in English. The year 2011 saw many new underground artists sprouting up with what they call independent music. These rappers from all corners of the country rapped in their regional language and delivered several hits occasionally which proved rap to be futuristic for fresh talents.

When these rappers finally started developing their lyrics, it still had the influence of the American rappers they adored.

However, in recent years, more youngsters have emerged with non-commercial Hip Hop with lyrics that addresses issues that are relevant to the society that they are familiar with, rather than plainly aping the American rap which has stories of gunfights and gang wars.

What draws many musicians to Hip Hop is the lyrical freedom it offers. Many artists raise sensitive issues through their songs which otherwise remain grounded due to political or social pressure. Rappers do not hesitate to sing about the governmental policies and programs and the politics behind everything.

Usually, such take on government is considered as an “attack” to defame the government when other methods, apart from music are used to express one's opinions. In the case of Hip Hop, no such restrictions have been imposed so far which makes it a choice for many to raise an issue.

Literature Review:-

Content analysis and use of language were the focus of previous researchers about rap music. Started out in the form of protest music, rap soon got the attention of many because of its lyrical content. Initially, the musicians came up with responsible protests which were predominantly relevant to the times and represented a larger community. Some research findings show that this genre indeed relates to some communities which can identify the problems as their own and form a system of themselves. Studies suggest that "the core essence of hip-hop is shared by marginalised groups" Motley, Carol M. and Geraldine Rosa Henderson. "The Global Hip-Hop Diaspora: Understanding The Culture". Journal of Business Research 61.3 (2008): 243-253. Web.

Later on, it shrank into a thin form of expressing the artists' frustrations which usually had explicit content. Rap, considered as “intensely violent, both sexually explicit and sexually violent, misogynistic, drug and gang oriented, and politically radical”(Derek Bushton,1986) is said to reflect on the artists as well.

Rap music contained themes depicting gang fights, drive-by shootings, and widespread gang violence. Johnson, Jackson, and Gatto (1995) suggested that those attitudes extend to using violence against women. They observed that those who viewed a violent rap video were more likely than those in nonviolent-music and control conditions to accept the use of violence against women, and to intend to use violence to resolve a decision making conflict.

Conceptualization of Hip-hop:-

Psychological studies provide a different approach to the content of hip hop. There are many conceptualizations of hip-hop as it was not solely a musical phenomenon; it embodied an entire way of existence for those immersed in or touched by its culture. One factor that distinguishes hip-hop from other genres of music is its primary focus on the spoken word”(Kobin and Tyson 343-356)

Cultivation Theory:-

The influence on audience perception was studied by applying Cultivation Theory which proposes that continuous exposure to media content over time can subtly cultivate or instil beliefs about the world that the audience has been part of.

Research Questions:-

1. What is the perception of the audience to socially and politically charged lyrics of Hip Hop?
2. Do the lyrics have any influence on audiences' decision-making?

Method:-

This study incorporated cultivation theory to examine the effect of rap/hip-hop music on shaping the audience attitudes and influencing decision making. By using a qualitative content analysis and an online survey, this study analysed the lyrical content of 10 rap/hip-hop songs released during the year 2010-2016.

The sample set was chosen based on the level of socially and politically charged lyrics that were present in the songs.

Qualitative Content Analysis:-

The study incorporates qualitative content analysis guided by the cultivation theory. The study examined the presence of protest lyrics found in 10 popular rap and hip-hop songs released during the year 2010 to 2016. The lyrics of the songs were obtained from various internet sources and the portion which had the presence of protest was coded. The level of protest was coded based on the presence of words which points towards political and social issues: inequality, discrimination, poverty, hunger, scam, shooting, political injustice, invasion of corporates in public matters, illegal trade and business, drugs and sexual assault.

Coding Descriptions:-

- Songs which had reference to political content was coded as such if they contained words that included: political parties, government, military, scams, etc.
- Songs which had reference to social content was coded as such if they contained words that included: poverty, inequality, hunger, corporates.
- Songs which indicated sexual conquest was coded as such if they contained phrases like: feel the pain, don't have to fight back.

Frequency or Emphasis of Protest Lyrics:-

The songs which had portions indicating protest terminology was first coded, and the amount or emphasis of content was labelled accordingly to determine the protest levels.

- Lyrics which had one or two references to protest- Low level
- Lyrics which had three to four reference to protest- Medium level
- Lyrics which had reference to five or more reference to protest- High level

Survey:-

In addition to a qualitative content analysis, the study performed an online survey administered through Zoho. The respondents were asked to respond to the survey which consisted of 10 questions by clicking on the link provided through email or social media. The survey considered a total population of 100 participants of women and men in equal numbers (N=100).

The first five questions asked the respondents general issues to determine the age, gender and listening habits.

The remaining questions tested the attitude of the interviewees towards non-commercial Indian Hip-Hop. It involved a series of multiple-choice questions graded on Likert Scale.

The questions also tested if the lyrics had any influence on the respondents' decision making.

Findings:-**Content Analysis:-**

Following are the list of popular Hip Hop songs with their year of release and the issues they address. By using a content analysis method, this study analysed the lyrics of 10 rap/hip-hop songs from 2010-2016. The sampling set of music was selected based on the strong political and social themes appearing within the lyrics of these popular hits.

Table 1:- List of songs taken for content analysis.

S. No	Song	Artist/Album	Release Year	Remarks	Level of Protest
1.	Haq Hai	Naezy	2016	Talks about the inequality in India and addresses government frauds.	Medium
2.	Takkaru Takkaru	Hip Hop Tamizha	2016	Addressed the need for Jallikkattu in Tamil Nadu. Became the Anthem of pro-jallikkattu protesters.	High
3.	Tragedy Mein Comedy	Naezy	2016	An attempt to mock the system which is corrupt and hollow.	Low
4.	Kodaikanal Wont	Sofia Ashraf	2015	Issue of mercury poisoning by Unilever Limited in Kodaikanal	High
5.	Sunlo Gall	Krull172	2014	Drug abuse in Punjab	Low
6.	Listen, My Brother	MC Kash	2012	Insurgency in Kashmir	Medium
7.	My People	MC Kash (Rebel Republic)	2012	Military attack on civilians in Kashmir	Medium
8.	Why We Rebel	MC Kash	2012	Explains the torture by Indian military troops on young Kashmiri youth in Indian jails.	Medium
9.	Re-introduction	Cryptographik Street Poets	2012	The battle between Uranium Corporation of India and environmental groups in Shillong.	Low
10.	I protest	MC Kash	2010	Protest demonstrations in Kashmir and remembering every youth who lost his life in mass military firing.	high

Inequality and poverty in the fastest growing metro:-

India is a developing country. However, if one needs to fill the stretched chasm about ultra-rich and ultra-poor, Mumbai is the place to be. The songs by artist Naezy, who hails from Mumbai, reflects the issues that the common man is familiar with. The level of protest is medium. The song involves criticising the government and its unsuccessful programs and policies. The song also talks about how government looted a humongous amount in the name of setting up a telecom network in the country. The artist calls the politicians "thief" and vents out his frustrations about how his city still has kids who go to sleep on an empty stomach. Thirty of India's sixty-eight billionaires live in Mumbai. Though it is a commercially strong city, more than 40 percent of Mumbai's population lives in the slums. The inequality is evident.

The following lyrics refer to the 2G scam in this verse which took the country by storm. The scam involved the teaming up of government officials and politicians to illegally allocate frequencies to telecom companies. This act resulted in the missing of a massive sum of rupees 1,76,645 crores. This scam was ranked second by the TIME magazine's list of top 10 cases of abuse of power.

" Sarkaar apni so ri hai, Paiso ki padi bori hai
 2 g 3 g kyu ji kitni inki chori hai?
 Chori toh chori uppar se seena zori muh par jhooti choli hai
 ye raakshaso ki toli hai."
 "Habastiq Hai
 n Prod By Sez Lyrics Meaning | Artist- Naezy /
 Dohaz.Com". Doha
 z.com. N.p., 2017. Web. 10 Mar. 2017.

Lyric Translation:-

2 g 3 g, how much was the whole theft worth?
 They steal and lie about it; they wear masks and cheat
 They are an evil gang
 What a neighbourhood this is? Tell me

The State of Tamil Nadu and “Jallikkattu” Protest:-

South India is a culturally-rich region. Though westernisation is a part of development, people from Tamil Nadu are well rooted in their culture. The recent peaceful protest by the youngsters demanding permission to conduct "Jallikkattu" which is a traditional bull-taming sport of Tamil Nadu is a proof of it. One of the reasons which initiated this protest was the album Takkaru Takkaru by Hip Hop Tamizha, released during early 2016. The lyrics of the song was found to have a profound impact on youngsters. The sport was banned in 2014 upon petitions filed by PETA which stated that animals were mistreated in the name of tradition. However, it was in the year 2016, the release of the song slowly started creating ripples. The song went viral on social media which attracted supporters who demanded a lift of the ban. The song did not plainly talk about the sport. The artist rapped about how large corporations are intruding into the culture and practice of the farmers and grass-root level population.

The artist did not hesitate to lash out on corporations, and PETA whom the artist says has vested interested in this matter. The artist talks about how these non-governmental bodies are so powerful and influential that they are trying to eradicate the culture of a state which puts its population in question.

The song created large-scale protest across Tamil Nadu which was supported by movie artists and was known as the most non-violent protest by youngsters in India. The protest finally resulted in the lifting of the ban on the sport. The song falls under the High-level protest category. The song soon became an anthem for the protesters.

*"idhan pinne ulla sarva desa arasiyal,
 viyabharathirkkaaga nadanthidum veri seyal.
 Indha vilaiyaattai thadai seidhaal, naatu maadu azhiyum, veli naatu niruvanangal,
 viyabaaram perugum Ariyaatha tamizha, un ariyaamai pizhaiyaal,
 un adayalaam izhanthaal, nee methuvaaga azhivaai, un adayalaam izhanthaal,
 un thaai naatil neeyum, oor agathiyai maariduvaai.
 Idhu maatta pathina prechana illa, un naatta pathina prechana da,
 naatin porulaathaaram veezhum, naamum edukanum pichchayada."
 Takkaru Takkaru Lyrics: Hip hop Tamizha". LyricsRaag.Com. N.p., 2017. Web. 10 Mar. 2017.*

Lyric Translation:-

Behind this ruthless act for trading is international politics. Banning this sport will only result in the extinction of our native breeds, increase the profit of foreign corporations.
 Tamizha! Because of your ignorance, you will lose your own identity in your native land.
 If you lose your identity, you will end up being a refugee in your homeland.
 This is not just about the bulls, it is about the downfall of our economy
 which will result in us begging to the foreign countries.

Environmental Issues:-

Sofia Ashraf, in her protest song “Kodaikanal Won't” addresses the illegal dumping of toxic waste into the Kodaikanal river. The song openly slams the company Unilever by mentioning the name of its products that the consumers are familiar with it. The song calls for support to sign the petition demanding the company to take up immediate remedial measures. The artist raps about how the company, being a big corporation is influential and powerful, bends the laws and regulations. Illegal dumping of toxic mercury into the Kodaikanal River and exposure such toxins resulted in children being born with deformities. However, the company was not held accountable for this. The song brought the issue to light. Unilever finally agreed for an out-of-court settlement with the people affected.

*"They dumped their waste in the local shrubbery
 Now that's some toxic shit
 By the way, what'd they say?
 That their factory was safe as day*

*They don't trust a word of what their workers say
 What about the polluting, the mercury poisoning?
 It's life threatening. There's kids suffering.
 Poison in the soil you don't have us foiled.
 Your clean was a sham, there's poison in the air.
 You ain't done done-da done done done
 Unilever has been hiding behind
 Their fake PSAs and Pepsodent smiles
 They washed their hands off Kodai with Lifebuoy
 There's nothing Fair or Lovely about this trial."
 "Kodaikanal Won't". prezi.com. N.p., 2017. Web. 10 Mar. 2017.*

Kashmir Insurgency:-

Jammu and Kashmir, a state in the extreme north, is an integral part of the Indian Union. Since 1990, the state is under Presidential rule due to militancy. The Governor runs the administration with the help of Advisors appointed by the Centre. The Constitution of India applies to the State of Jammu and Kashmir as well. The territories of Jammu, Kashmir, Ladakh and Gilgit form the state of Jammu and Kashmir. In 1846, it was the biggest princely state in India before the 1947-partition. At that time, the total area of the state was 2,22,236sq.km. In 1949, due to a cease-fire line between India and Pakistan, 78932 sq.km of the area(Gilgit, Mirpur, Kotli and part of Poonch) came under the possession of Pakistan.

Again, in 1962, 64000 sq.km was occupied by China, now known as Aksai Chin. Later, Pakistan again made illegal possession over Chhamb, Deva, Chakla and Manawar gaining an area of 3999 sq.km. So finally, India is left with the remaining 12850 sq.kms.

After the defeat of the U.S.S.R, which was a superpower by Afghan mujahideen, General Zia, who was the Pakistani dictator initiated a similar move to bleed India. He sent his trained troops of Islamic militants to Kashmir in the late 1980s. During that time, Pakistan was well aware that it cannot declare a direct war with India.

It began expanding as ISIS, Indian Mujahideen, Al-Qaeda planting their personnel in the soil they want to attack. The tension between India and Pakistan hiked with a five-man suicide squad attacking the Indian parliament in December 2001, and the insurgency still continues.

MC Kash, a youth who hails from Kashmir, talks about everything in Kashmir through his songs as his land remained disconnected to the remaining parts of the country. His songs gained international recognitions. In his song, *I protest*, he speaks about the protest demonstrations in Kashmir and lists the names of every youth who lost his life in the military attack. He refers to his homeland Kashmir as a paradise which is under fire due to the military strike. The murder and rape by Indian troops are unspoken details of insurgency in Kashmir which is covered up by the Indian government.

*My paradise is burning
 With troops left loose with ammo
 Who murder and rape
 Then hide behind a political shadow*
 Other lyrics which relate to such issues are in his song *Why We Rebel* and *Listen My Brother*
*I come from the land of bloody crackdowns
 where silhouettes of life rush to the playgrounds
 you sit your ass down, and don't make a sound
 take off that Pheran, you motherfuckin' clown
 whose son is he? whispers ask
 doesn't matter really 'cause it ain't the last
 faint hearts of men beat a little slow
 doesn't matter when there's a gun down your throat*

Act of torture and violence on Kashmiri kids who are forcibly taken and locked up in Indian jails. They are raped and mistreated. The artist tries to empathise with the victims. The following lyrics imitate the act of rape and sodomy on Kashmiri kids in Indian Jails.

*Pressure Up, You hold the hands
Show this motherfucker how to bleed in this land
Push It Push, let him scream and crawl
make his blood ricochet of these walls
thirteen, and you holdin' a stone?
lemme see those scars and yo broken bones
Brother, allow me to hold you
knowing even the Lord can't console you*

Results:-

Views on Non-Commercial Rap/Hip-Hop and its Interpretations from the Survey:-

91 out of 100 respondents listen to Indian Hip-Hop as compared to the remaining nine who do not. Over 63% of respondents agree that non-commercial Indian Hip-Hop is addressing many social and political issues. Out of this 60% of respondents agree that they are also able to relate to the issues while 31% responded that they could sometimes relate to the issues.

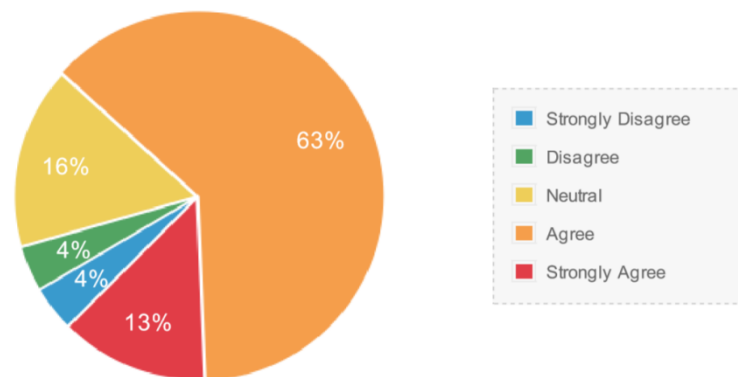


Figure 1:- Population Indicating Agreement/Non-agreement to the Ability to Relate to Societal Issues After Listening to Socially or Politically Charged Rap/Hip-Hop Lyrics.

- When questioned about if the lyrics had any influence on the respondents' decision making, 65% respondents agree that they have been part of/supported a cause/campaign after being influenced by the lyrics

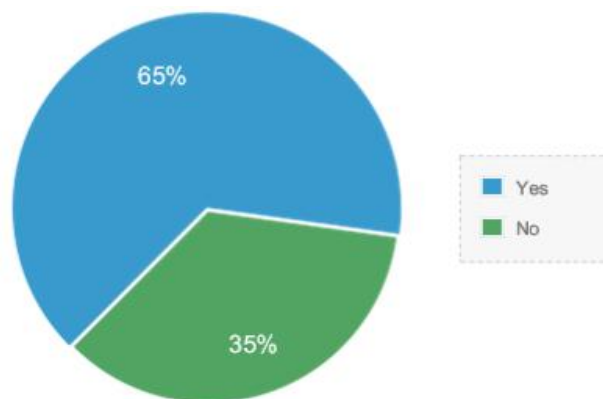


Figure 2:- The population of Respondents who have been a Part of/ Supported a Cause/Campaign After Being Influenced by the Lyrics.

- 68% responded positively to when questioned if they would consider raising an issue through Rap/ Hip-Hop as opposed to 13% who chose not to consider and 18% who may/may not consider.

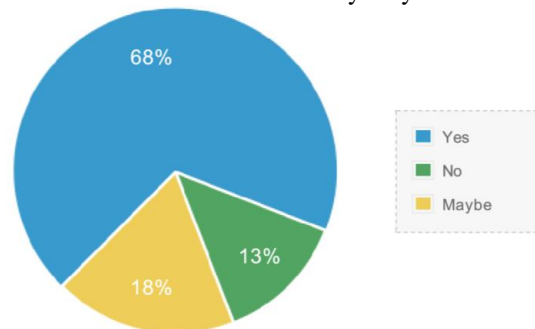


Figure 3:- Population of Respondents who would Consider Raising an Issue Through Rap/Hip-Hop.

Results of Content Analysis:-

Out of the ten songs analysed, three songs were labelled as High-level protest songs. The songs are found to have influenced the audience in decision making and also to support/be a part of the issue raised. Three songs were labelled as low-level protest songs because of the repetition of subjects and poorly influential lyrics. The songs were not successful in creating a mass perspective. They were restricted to a small group or a place that the artist had a personal connection to.

The remaining four songs were labelled as medium-level protest songs as they were successful in bringing out more global perspective and also got attention for the same. Some of them openly criticised the system and threw light upon issues that were kept under folds.

Comparing Results:-

To consensus between content analysis and survey results, both the methods were tested for differences and similarities. While the content analysis dealt with the explanation of lyrics and its context, a similar analysis was drawn from the survey results which described the audience perception about social issues being raised by Rap/Hip-Hop.

The survey results also indicate a positive response to lyrics having influenced the respondent in decision-making. This is similar to the analysis made about 'Kodaikanal Won't' and 'Takkaru Takkaru' albums which had large scale supporters for the issue that these songs addressed.

Conclusion:-

Over the years, protest songs, which were once used as a medium to voice the voiceless lost its significance as people began venting out the anger and frustration on whatever they found annoyed them. A community's view shrunk to an individual's view and it was tagged as violent and inappropriate. The lyrics lost quality and artists were rapping about anything and everything.

In India, non-commercial rap/Hip-Hop scene is better with the artists sticking to its 'Rasta' roots. Socio-political protest constitute the major feature of this "counter-culture movement" (Derek Bishton(1986:2))

The study, with the help of cultivation theory proves that exposure to media content for a prolonged period has an effect on the perception and decision making of the audience. The role of Rap/Hip-Hop in addressing social and political content has an influence on listeners who may develop "similar attitudes in their own lives, including accepting the objectification of women" (Dixon, Zhang, & Conrad, 2009, p. 348).

The study thus proves that non-commercial Indian Hip-Hop has an influence on the perception of the audience.

Scope Of Study:-

Previous studies on Hip-Hop were mainly about the deteriorating content and excessive use of unaccepted language. From this research results, a positive side of protest music can be inferred. With new artists using rap/hip-hop as a tool to voice serious issues, this could probably be a new music movement in India.

The perception study can be performed on a larger scale which can be used in clinical psychology studies to understand the attitudes and behavioural patterns. Rap therapy is another field of study which is gaining popularity.

Limitations:-

The survey used a convenience sample of 100 respondents who were exposed to this survey through emails and social media. As a result, the final findings cannot be applied to a general population. Additionally, the survey was administered to a population of rap listeners and non-rap listeners. A case of such may have affected the evaluation process. This might have resulted in obtaining skewed data which may otherwise have been different if only rap listeners were considered.

The lyrics may be interpreted differently and individual opinions may vary.

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RESEARCH ARTICLE

EXPRESSION OF PROPER NOUNS AND PRONOUNS BY DEAF SIGNERS OF INDIAN SIGN LANGUAGE (MUMBAI-DELHI DIALECT).

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Key words:-

Proper nouns, Pronouns, Mumbai-Delhi dialect of Indian Sign Language.

Abstract

The aim of the study was to investigate the expression of proper nouns and pronouns by deaf signers of Indian Sign Language (ISL) (Mumbai-Delhi dialect) in narration. Ten fluent Deaf signers of Indian Sign Language (5 Males & 5 Females), in the age range of 18-40 years using signs of Mumbai-Delhi dialect were included in the study. The picture cards of the story "The snake and the mongoose" was used for the narration task. The participants were presented with the picture cards and instructed to narrate the story using sign language. The signed expressions of the participants were video recorded and later analyzed by two judges individually. The judges transcribed and coded the signed expression of the participants for proper nouns and pronouns types using ELAN software and the frequency of occurrence was calculated for the proper nouns and pronoun types. The results showed that the deaf signers of ISL used noun sign or fingerspelling to express the proper nouns; and used a combination of 'manual pointing and eye gaze' as a predominant strategy to indicate pronouns. Use of other strategies such as a) manual pointing/indexing alone and b) manual pointing/indexing using combination of non-manual markers were also used to express the pronoun types.

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Introduction:-

Indian Sign language (ISL) is used in Nepal, Bangladesh, Sri Lanka and border regions of Pakistan (Vasishta, Woodward and Wilson, 1978; Woodward, 1993; Zeshan, 2000 & Zeshan, Vasishta & Sethna, 2004). Gordon Jr., (2005) identified three dialects of ISL: Mumbai-Delhi Sign Language, Kolkata Sign Language and Bengaluru-Chennai Sign Language.

The 'Signing space' in Sign languages extends from the top of the head to just below the waist vertically; and from the signer's extreme right to extreme left horizontally/laterally. The signing space is used to express phonological contrasts in signs (Brentari, 1998; Sandler, 1989) and express pronouns, number and verb arguments in the morphological structure of signs (Janis, 1992; Mathur, 2000 & Padden, 1988). At the discourse level, however, spatial locations are associated with discourse referents (Klima & Bellugi, 1979).

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Pointing gestures typically co-occur with speech, and are used to draw the attention of the listener (Bangerter, 2004; Clark, 2003 & Haviland, 2000b). Kendon (2004) & Kita (2003a, 2003b, 2003c & 2009) reported that pointing signs in sign language are not different from that of pointing gestures used by non-signers. In pointing gestures, an extended index finger is used to point at the object, location, person etc. In sign languages, pronouns are signed by pointing to a person or thing that is “present” in the space or by pointing to a specific location in the signing space which represents a person or thing that is “not present” in the communication area. Kendon (2004) and Kita (2003a, 2003b, 2003c & 2009) observed that the pointing signs are used to express pronominal reference in sign languages and they do not look different from that of the pointing gestures that are used by the non-signers. The association between the referent and a spatial locus is established by articulating the sign for the referent in space, by producing the sign and then pointing to the locus or by gazing in the direction of the locus being established while making the sign (Lillo-Martin & Klima, 1990; Padden, 1983). Once the locus for a referent is established, the locus is maintained throughout the discourse. This phenomenon of establishing loci in space in sign languages to indicate pronouns is known as ‘Pronominalization’. Bangerter (2004) observed that in sign languages, the signs used for pointing refer to precise, unambiguous, pronominal reference.

The use of pronominalization to express pronouns is well reported in several sign languages such as American Sign Language (ASL) (Hatzopoulou, 2008 & McBurney, 2002), British Sign Language (BSL) (Kyle & Woll, 1985; Meier, 1990), Israel Sign Language (ISL) (Meier, 2003), Swedish Sign Language (SSL) (Bergman, 1980), and Brazilian Sign Language (LSB) (Berenz, 1996). Three distinct signs are often reported across many sign languages in the signed expression of pronouns which include: (a) pointing towards the signer to refer to first person pronoun ‘I/Me’, (b) pointing towards the person to indicate second person pronoun ‘You’ and (c) pointing to another person to indicate ‘He/Him’, or ‘She/Her’ (For example, in American sign language (ASL) [Berenz (1996); Cormier (2005, 2007); McBurney, (2002); Hatzopoulou (2008); Liddell (2000b, 2003)] in British Sign Language (BSL) [Cormier, Fenlon, Rentelis, & Schembri (2011)] in Sign Language of Netherlands [Bos (1995)] in Swedish Sign Language (SSL) [Bergman (1980)] in Israel Sign Language (ISL) [Meier (2003)] in Brazilian Sign language (LSB), [Berenz, 1996]). Further, there are no stand alone signs reported to express pronouns (lexicalized pronouns) in ASL (Friedman, 1975); the pronouns in ASL are signed by establishing a frame of reference in front of the signer’s body, wherein the reference points for objects, persons, and locations are established.

The pointing (with an extended index finger)/ indexing in sign languages convey several functions including the expression of pronominals, adverbials (locative) and determiners. Locative (adverbial) pointing signs are those signs that point to the signing space to refer other location which have adverbial function. E.g., ‘here’ ‘there’ etc. Demonstrative pointing signs are those signs used to express demonstrative pronouns such as ‘this’, ‘that’ ‘here’ and ‘there’ etc. The distinction between proximal demonstrative pronouns (this, these & here) and distal demonstrative pronouns (that, those & there) are made based on the distance of the locus being established by the speaker to indicate object, place etc (Lyons, 1979). Diessel (1999) reported that demonstrative pronouns are the very first words that typical children learn and that demonstrative pronouns are frequently used by typical adults in conversation (Wu, 2004).

Possessive pronouns in sign languages are expressed in the same way as that of Non-possessive pronouns/Personal pronouns, but the latter have a distinct handshape. Non-possessive pronouns/personal pronouns are generally expressed with pointing handshape whereas possessive pronouns assume different handshapes i.e., palm directed towards the referent in several sign languages such as ASL (Cormier & Fenlon, 2009), Australian Sign Language (OGS) (Johnston & Schembri, 2007), and Hong Kong Sign Language (HKSL) (Tang & Sze, 2002). The pointing function is also reported to indicate plural pronominal signs. In ASL and BSL, Cormier (2005 & 2007) found that, the plural pronominal signs have less of a pointing function than that of the singular pronoun signs i.e., plural pronominal signs are less indexic in nature when compared to singular pronominal signs. IPSL has a transnumeral pronominal sign (an extended index finger pointing sign) which is used for singular and plural; pointing towards the signer’s chest refers to the signer or the signer plus others, depending on the context. Most sign languages use spatial inflections to mark person and number inflections (Zeshan et al., 2004).

The expression of pronouns in sign languages also involves signing of a manual sign along with the non-manual markers. The non-manual expressions used to express pronouns include eye gaze, head turn in American Sign Language (ASL) (Baker & Cokely, 1980; Bellugi & Fischer, 1972; Liddell & Metzger, 1998), wherein, the direction of eye gaze follows direction of hand; the direction in which pointing sign is made. Neidle, Kegl, MacLaughlin,

Bahan and Lee (2000) reported that in ASL, in addition to eye gaze, pronouns are often accompanied by other non-manual markers such as wrinkled nose, furrowed brows, and a slight rapid head shake.

Several other studies on ASL have recorded the role of 'body shift' to indicate the third-person reference with a pointing sign directed towards the signer; the shift indicated by the change in orientation of signer's shoulders, head and/or eyes in ASL (Lillo-Martin & Klima, 1990). On the other hand, Meier (1990) reported that head position and the direction of eye gaze in Israel Sign Language (ISL) distinguishes third-person pronoun from non-third person pronoun. MacLaughlin (1997) noted that pointing signs in Hong Kong Sign Language (HKSL) could be interpreted as a pronoun when signed alone. The pronouns in HKSL are optionally signed by using eye gaze directed at the location of referent in signing space. Liddell and Metzger (1998) reported that the use of eye gaze to express pronouns in ASL was consistently directed toward the addressee during the production of both second person pronouns and third-person pronouns and no distinction in eye gaze was found between second and third person referents.

Studies addressing the linguistic attributes of Indian Sign language (ISL) when compared to other sign languages are very scarce (Prajapati & Asai, 2010). There is need to address the sign language structure of Indian Sign language, in order to provide a base for formalizing the approaches towards teaching and learning the Indian sign language by the hearing impaired individuals as well as their communication partners. It will also facilitate and open opportunities to research and develop sign language recognition systems. This study aimed to analyze and report on expression of pronouns used by the deaf signers of Indian Sign language (ISL) using Mumbai-Delhi dialect, including the use of non-manual markers in a story narration task.

Method:-

Participants:-

Ten fluent Deaf Signers (5 Males & 5 Females) using Indian Sign Language (ISL) signs of Mumbai-Delhi dialect were included. The participants were in the age range of 18-40 years. Informed written consent was obtained from each participant prior to the recording of their samples. The participants met the following criteria:

They were pre-lingually deaf born to hearing parents.

They were proficient in using ISL in day to day conversation. They were using ISL for a minimum of 10 years, as ascertained using a checklist prepared for the purpose by the investigator.

They interacted with their parents, family members and other communication partners using a combination of sign language and few verbal expressions for day to day communication.

They had a minimum educational qualification of secondary education in a school for Persons with Hearing Impairment with the medium of instruction for curricular work being English.

They did not have any other disabilities in motor, visual and neurological domains (as ruled out using Quick neurological screening test (QNST) by Mutti, Sterling and Spalding, 1978), and cognitive deficit (as ruled out using Mini-Mental State Examination (MMSE) by Folstein, Folstein, and McHugh, 1975).

All the participants used Mumbai-Delhi dialect of Indian Sign Language (ISL) as ascertained through information collected in the demographic data sheet by the investigator.

Task and procedure:-

A story on the topic 'The Snake and the Mongoose' was selected and depicted as sequential picture event on 6 cards with the help of an artist. This story was selected as it provided opportunities to use pronouns repeatedly when the story was narrated. The pictures depicting the story sequence were verified for disambiguity and order of continuity in the sequence by two qualified speech-language pathologists on a three-point rating scale. Video recording of each participant at a time was carried out at the educational centre or work place of the participant by the principal investigator. The video recording of the signed expression of the participants for narration of story was carried out in a room which was well illuminated with minimal visual and sound distraction. The sequential story cards were initially given to the participant (from whom the data was collected individually) and was asked to understand the concept represented in each card and the summary of the story. The participant was then asked to narrate the story using sign language as naturally as possible and the signed expressions of the participants were video-recorded by

the principal investigator using a hand held Sony Digital Video Camera Recorder (Model no: DCR-SR47E) with the capacity of 800K pixel, 60*optical zooming, and storage capacity of 45 hours of video footage. The camera was mounted in front of the participant on a tripod stand at a distance of minimum 5 to 7 feet and zoomed to capture the sign space from head to trunk and arms extended on both the sides of the participant. Each participant was given two trials each and the same was recorded.

Analyses:-

Of the two video samples recorded, the sample which had less visual distracters/repetitions/good visibility of signing was considered for the analyses. The sample was fed into a computer. ELAN (EUDICO Linguistic Annotator) software (Hellwig & Van Uytvanck, 2007) was used for coding the signed expressions. A set of annotations sharing the same characteristics can be coded on a single tier using ELAN software (Johnston & Crasborn, 2006). Few other investigators have provided annotation guidelines for coding sign language video samples (Crasborn et. al., 2007; Johnston & Crasborn, 2006; Johnston & Schembri, 2006; Neidle, 2002; Zwitterlood, Ozyurek & Perniss, 2008). Based on these guidelines of annotations and transcription conventions, the signed expression of each participant was coded by the principal investigator. The coding of the signed expressions of the participants was carried out to record the use of (a) different pronoun types, (b) strategies used in the expression of the pronouns and (c) the use of non-manual markers to sign various pronoun types.

The data was analyzed and coded under three tiers in ELAN software as follows:-

1st tier - Sign Gloss: The data was transcribed and annotated for expression of lexical signs i.e., glossing for each sign of each participant was carried out by selecting the respective portion in video sample under this tier. The glosses in this tier were transcribed in upper case as per the guidelines.

2nd tier – Identification of types of pronouns: In this tier, data was checked for the presence of pronouns in signed expression. Instances where pronouns were expressed by the participant were noted and coded for types of pronouns such as personal pronouns, demonstrative pronouns, possessive pronouns etc.

3rd tier – Identification of non-manual markers used in the expression of pronouns: In this tier, the data was coded for use of different non-manual markers in signed expression of pronouns. This was carried out as per the operational definition of Non-Manual Markers defined by the investigators (provided in Annexure 3). The function of each of the 3 tiers as defined in this study is presented in Annexure 1.

The annotations used to code different types of pronouns and non manual markers expressed by participants are shown in Table 1 and 2 respectively.

Table 1:- Annotations used in the coding of different pronoun types.

Sl no.	Annotations	Pronoun Types
1.	IX-1P (pe pr)	1 st Person (personal pronoun)
2.	IX-2P (pe pr)	2 nd Person (personal pronoun)
3.	IX-3P: i (pe pr)	3 rd Person (personal pronoun) associated with location 'i'.
4.	POSS-1P (po pr)	1 st Person (possessive pronoun)
5.	POSS-2P (po pr)	2 nd Person (possessive pronoun)
6.	POSS-3P (po pr)	3 rd Person (possessive pronoun)
7.	Dem	Demonstrative Pronoun

Table 2:- Annotations used for various non-manual markers.

Sl no.	Annotations	Non-manual markers
1.	Eg	Eye Gaze
2.	Psp	Pointing to space
3.	Pse	Pointing to self
4.	hm: bd	Head movement: bent down
5.	Hp:ti fr	Head position: tilt front
6.	Hp:ti side	Head position: tilt side
7.	Htu	Head turn
8.	hm:Jut	Head movement: jut
9.	BL: for	Body leaned forward
10.	EBr	Eye Brow: raised

Reliability check:-

10% of the video samples of each participant were subjected to reliability check. Another coder/judge with post graduate degree in speech- language pathology and knowledge of sign language, having normal hearing and vision, and familiar with the operation of a computer was recruited for the reliability check. The 2nd coder was trained by the first coder (principal investigator) using samples of deaf signers that were not included in this study, on the following aspects: 1) identification and selection of signed expressions 2) referring to operational keys and annotating data on ELAN software and 3) coding the pronoun types as per the operational definition of the pronoun types (Annexure 2) and reviewing the annotated samples to confirm the expression of pronouns as per the definitions. Once the data was coded by the 2nd coder, the inter and intra judge reliability was established.

Inter judge reliability: This was established between the two coders, based on item by item verification for agreement/disagreement in various domains of the data. In instances of disagreement between coders for any of the items, analyses were repeated by coders independently and parameters mutually agreed upon by two coders were noted as final. The reliability coefficient was calculated

Intra-judge reliability: Reanalyses of 10% of the data per participant was carried out within a month from the day of first analyses by the 1st coder (investigator) and the 2nd coder also in order to check for intra judge reliability. In instances of disagreement between the two instances of coding, it was resolved by reviewing and confirming the parameter analyzed. The reliability coefficient for the samples analyzed in two instances by investigator and other coder was determined.

The raw data obtained by the coders was tabulated and subjected to statistical analysis.

Results and Discussion:-

The coded data was used to tabulate and record frequency of occurrence of different pronoun types, strategies used to express pronouns and use of non-manual markers/facial expressions by participants to express the pronoun types. The signed expressions of the story narrated by most participants majorly included use of proper nouns (woman, snake, mongoose, baby, and cradle) that occurred in the story and several pronoun types such as demonstrative pronouns (here, there, that, this), 1st person pronoun (I, me), 2nd person pronoun (you, she), 3rd person pronoun (it) and 1st person possessive pronoun (my, mine) that substituted the proper nouns.

The proper nouns were expressed by participants using following strategies: a) Signing the noun signs/lexical signs and b) Finger spelling the nouns. The expression of pronoun types involved use of strategies such as a) manual pointing/indexing alone and b) manual pointing/indexing used along with non-manual markers to mark pronoun types. The frequencies of occurrence of the features under study were calculated as follows:

The frequency of occurrence of proper nouns:

The number of occurrences of proper nouns
The observation period of the sample.

The frequency of occurrence of different pronoun types:

The number of occurrences of a pronoun type
The observation period of the sample

The frequency of occurrence of different strategies:

The number of occurrences of a strategy type
The observation period of the sample

The expression of various features studied as computed by the ELAN software were noted and subjected to statistical analysis. The data was subjected to statistical analysis using SPSS (version 16). Shapiro-Wilk test of normality revealed that the data was not normally distributed ($p > 0.05$). Hence non-parametric tests were used to find significant difference if any in the frequency of occurrence of various pronoun types and proper nouns.

1. Frequency of occurrence of proper nouns and pronoun types

The frequency of occurrence of proper nouns and different pronoun types is represented in table 3.

Table 3:- Frequency of occurrence of proper nouns and various pronoun types

Participants	Proper nouns	Demonstrative pronouns	1 st person pronouns	2 nd person pronouns	3 rd person pronouns	1 st person possessive pronouns
1	0.4395	0.0845	0	0	0	0.0338
2	0.4572	0.0795	0	0	0.0198	0.0397
3	0.2844	0.0890	0	0.0098	0	0.0196
4	0.4333	0.1083	0	0	0.0154	0
5	0.2309	0.0355	0.0355	0.0355	0.0177	0.0177
6	0.3110	0.1072	0.0214	0	0.0214	0.0214
7	0.6068	0.1213	0.0242	0	0	0
8	0.4373	0.2460	0.1093	0.1913	0.0546	0.0000
9	0.4655	0.1643	0.0273	0	0	0
10	0.4939	0.0790	0.0592	0.0197	0	0

The data was subjected to Friedman test and the results revealed a significant difference in frequencies [$\chi^2(5) = 37.997$, $p < 0.01$] across proper noun and various pronoun types. There was a significant difference in the frequency of occurrence of proper nouns, demonstrative nouns, 1st person pronouns, 2nd person pronouns, 3rd person pronouns and 1st person possessive pronouns. Hence, Wilcoxon Signed Rank test was run to make a pair-wise comparison of frequencies of occurrence of proper nouns and pronoun types. Significant differences between the frequency of occurrence of proper nouns and demonstrative pronouns ($Z = -2.805$, $p < 0.01$), proper nouns and 1st person pronouns ($Z = -2.803$, $p < 0.01$), proper nouns and 2nd person pronouns ($Z = -2.803$, $p < 0.01$), proper nouns and 3rd person pronouns ($Z = -2.803$, $p < 0.01$), and proper nouns and 1st person possessive pronouns ($Z = -2.803$, $p < 0.01$) were found. The proper nouns were more frequently signed when compared to demonstrative pronouns, person pronouns (1st, 2nd and 3rd person pronouns) and possessive pronouns (1st person), as represented in Figure 1.

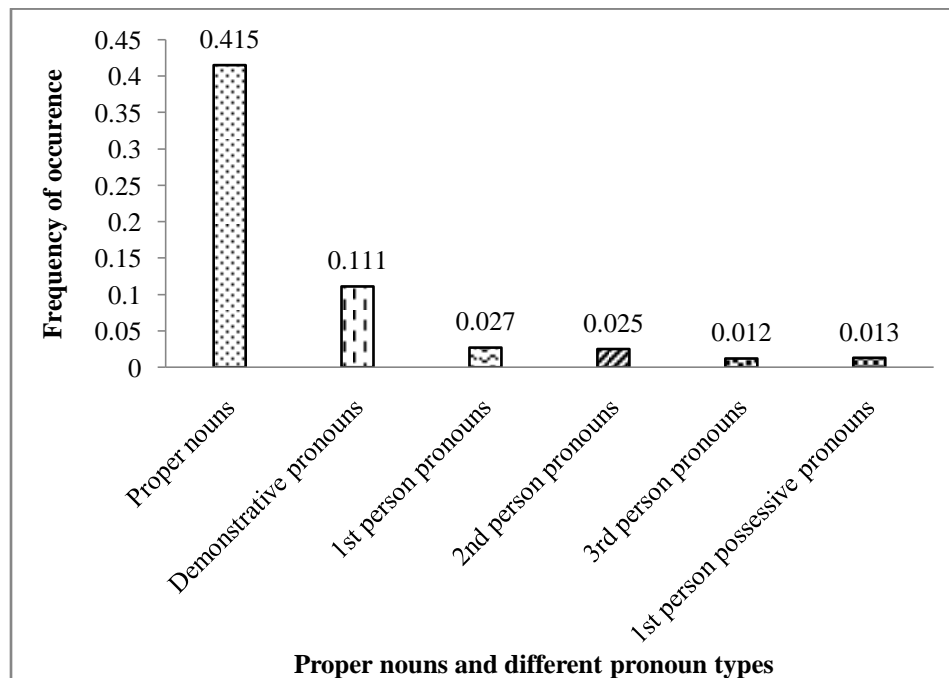


Figure 1. Mean frequency of occurrence of proper nouns and different pronoun types used by the participants.

The participants signed proper nouns using lexical signs representing them and also by using finger spelling. The use of pronouns such as demonstrative pronouns, person pronouns (1st, 2nd and 3rd person pronoun) and possessive

pronouns (1st person) required the participant to establish a loci in space, whereas use of proper nouns by the deaf signers involved signing the noun signs or using fingerspelling, which could probably be the reason for more frequent use of proper nouns when compared to the pronouns.

There was a significant difference in the frequency of occurrence of demonstrative pronouns demonstrative pronouns and 1st person pronouns ($Z = -2.666$, $p < 0.01$), demonstrative pronouns and 2nd person pronouns ($Z = -2.666$, $p < 0.01$), demonstrative pronouns and 3rd person pronouns ($Z = -2.803$, $p < 0.01$), and demonstrative pronouns and 1st person possessive pronouns ($Z = -2.803$, $p < 0.01$). Amongst the pronoun categories, the demonstrative pronouns occurred more frequently compared to 1st, 2nd and 3rd person pronouns and 1st person possessive pronouns. However, difference between the frequencies of 1st, 2nd, 3rd person pronouns and 1st person possessive pronoun were not significant.

Overall, the participants used proper nouns and different types of pronouns in their signed expression of the story. Amongst the pronoun types that represented nouns, demonstrative pronouns were used more frequently compared to that of other pronoun types (1st, 2nd, 3rd person pronouns and 1st possessive pronoun).

2. Frequency of occurrence of demonstrative pronouns

It was observed that the participants used demonstrative pronouns such as 'this', 'that', 'here', 'there' more frequently compared to person pronouns and possessive pronouns while narrating the story. Diessel (1999) reported that demonstrative pronouns are the very first words that typical children learn. Wu (2004) also reported that demonstrative pronouns are frequently used by typical adults in conversation. This seems to be true amongst the deaf signers also as seen in the results of this study. The demonstrative pronouns were expressed using the following strategies by the participants:

Using manual pointing signs/Indexing alone: this involved the signers extending their index finger towards the referent in signing space to establish reference/loci in space.

Using manual pointing/indexing accompanied by various combinations of non-manual markers such as Eye gaze, raising of eyebrows, body leaning forward in the direction towards the loci, head movements such as head bent down towards the loci, head turn towards the loci, head movements such as head bent down towards the loci, head turn towards the loci /referent etc in conjunction with pointing sign to mark the demonstrative pronouns.

Similar findings have also been reported in sign languages such as ASL (Baker & Cokely, 1980; Bellugi & Fischer, 1972; Liddell & Metzger, 1998; Neidle, Kegl, MacLaughlin, Bahan & Lee, 2000) and Israel Sign Language (Meier, 1990), where eye gaze, head turn, body shifts, were observed in addition to manual pointing to mark pronouns.

Several groups of strategies were used by the deaf signers of Mumbai-Delhi dialect of ISL to express the demonstrative pronouns during the story narration task and they are as follows:

Group 1: Pointing to space, eye gaze, head bent down and Body leaned forward

Group 2: Pointing to space, eye gaze, and head bent down

Group 3: Pointing to space, eye gaze

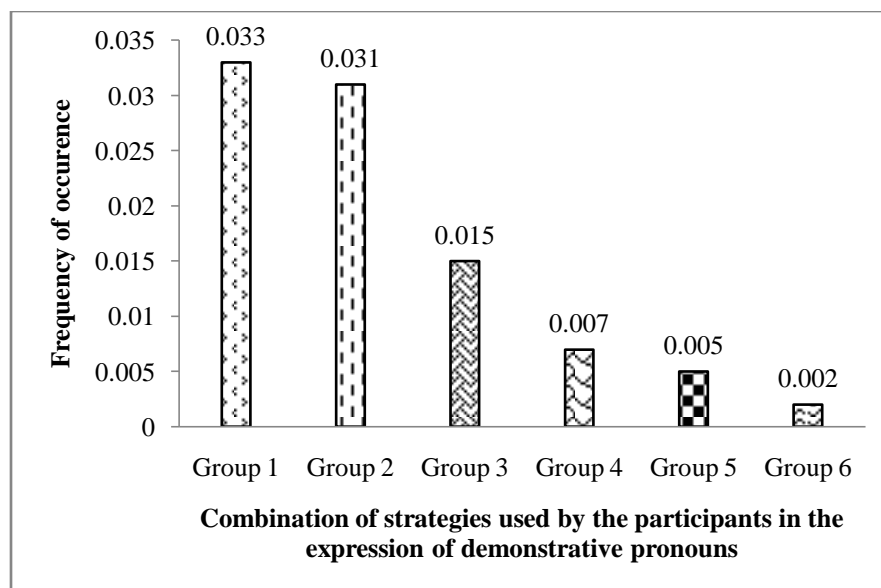
Group 4: Pointing to space, eye gaze and head turn towards the referent

Group 5: Pointing to space

Group 6: Pointing to space, eye gaze, and eyebrows-raised

Shapiro-Wilk test for normality of distribution revealed that the data was not normally distributed ($p > 0.05$). Hence non-parametric tests were administered to verify for significant difference if any in the frequency of occurrence of combination of strategies used in the expression of demonstrative pronouns. Friedman test revealed a significant difference in frequencies of occurrences [$\chi^2(5) = 13.612$, $p < 0.05$] of combination of strategies used to express the demonstrative pronoun.

Several combinations of non-manual markers along with manual pointing were used by participants to indicate demonstrative pronouns in story narration task and the same is represented in Figure 2.



[Combination 1: Pointing to space, eye gaze, head bent down and Body leaned forward; Combination 2: Pointing to space, eye gaze, head bent down; Combination 3: Pointing to space, eye gaze; Combination 4: Pointing to space, eye gaze, head turn towards the referent; Combination 5: Pointing to space; Combination 6: Pointing to space, eye gaze, eyebrows-raised]

Figure 2. Mean frequency of occurrence of different group of strategies used by the participants in the expression of demonstrative pronouns.

A pair –wise comparisons of frequencies of occurrence of combination of strategies was carried out using Wilcoxon Signed Rank test. The results revealed a significant difference in frequency of occurrence ($Z = 2.521$, $p < 0.01$) between combination 1 and combination 4. A significant difference in frequency of occurrence between the combination 1 versus combination 6 ($Z = -2.547$, $p < 0.01$), and combination 3 versus combination 6 ($Z = 1.992$, $p < 0.01$) was also observed.

The predominant strategy used to sign demonstrative pronouns by participants in story narration task involved a) manual pointing sign accompanied by non-manual markers such as Eye gaze, Head bent down, Body leaned forward toward the referent (combination 1) followed by a combination of b) manual pointing sign and Eye gaze (combination 3) when compared to other combination of strategies (Figure 2). Observation of the data also revealed that although various combinations of non-manual markers and pointing were used to express the demonstrative pronouns by participants, manual pointing sign and eye gaze were invariably used as a common strategy by the participants.

Qualitative observation of the data revealed that the proximal demonstrative pronouns ('this' & 'here') were differentiated from that of distal demonstrative pronouns ('that' & 'there') based on the distance of locus being established by the signer in signing space. The demonstrative pronouns 'this' & 'here', were expressed with manual pointing sign in space that was proximal to the signer's body whereas, for pronouns 'that' & 'there', manual pointing signs were far away from the signer's body. This observation is similar to the report by Lyons (1979) for American Sign Language (ASL).

The phenomenon of establishing loci in space through use of manual pointing sign and use of non-manual markers to express the pronouns in sign language is known as pronominalization. Use of 'pronominalization' to express pronouns have also been documented in several sign languages such as American Sign Language (ASL) (Cormier, 2005, 2007; Hatzopoulou, 2008; McBurney, 2002), British Sign Language (BSL) (Kyle & Woll, 1985; Meier, 1990), Israel Sign Language (ISL) (Meier, 2003), Sign Language of Netherlands, (SLN) (Bos, 1995), Swedish Sign Language (SSL) (Bergman, 1980), and Brazilian Sign Language (LSB) (Berenz, 1996).

As reported by several researchers, pointing is also used in spoken language as a co-speech gesture (Clark, 2003; Haviland, 2000b) and it is used to indicate the direction, location, objects and/or people (Kita, 2003b). The sign languages also makes use of pointing gesture to mark person and number inflections (Zeshan et al., 2004). In this study, the deaf signers of Mumbai-Delhi dialect also relied on manual pointing sign to establish the pronominal reference in space. They also used non-manual markers such as raised eyebrows, body leaned forward (towards the referent), head bent down (towards the referent), head turn (towards the referent) in addition to pointing sign. This can be reasoned based on the fact that these were used by the deaf signers to augment semantic meaning for the expression of the demonstrative pronoun.

3. Frequency of occurrence of 1st person pronouns

The participants used 1st person pronouns forms such as 'I', 'Me' in the narration of the story. The expression of 1st person pronouns involved the use of following strategies:

a) Pointing to oneself i.e., usually towards the signer's chest with extended index finger or with open hand touching the signer's chest or

b) Use of non-manual markers along with pointing to oneself

The combination of strategies that were used by the deaf signers of Mumbai-Delhi dialect of ISL to express the 1st person pronouns during the story narration task are as follows:

Combination 1: Pointing to oneself

Combination 2: Pointing to oneself and body leaned forward

Combination 3: Pointing to oneself, head tilted to side, and body leaned forward

Combination 4: Pointing to oneself and head movement in jut position i.e., the chin touching the neck.

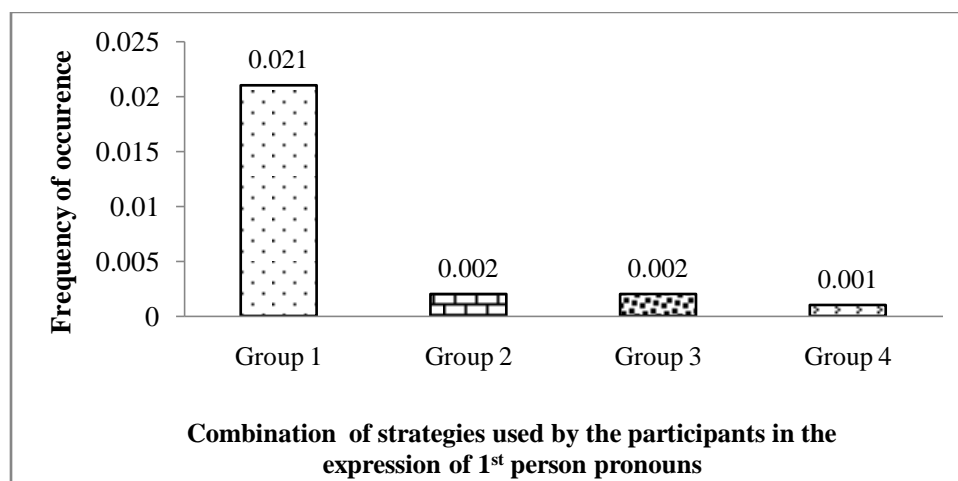
Friedman test revealed a significant difference in frequencies of occurrence [$\chi^2(3) = 14.122$, $p < 0.05$] across various combination of strategies used to express 1st person pronouns. Pair-wise comparisons of different combination of strategies were carried out using Wilcoxon Signed Rank test and results revealed a significant difference between the following combinations:

a) Combination 1 versus Combination 2

b) Combination 1 versus Combination 3 and

c) Combination 1 versus Combination 4.

The mean frequency of occurrence of combination of strategies used by deaf signers to express the 1st person pronouns are represented in figure 3.



[Combination 1: Pointing to oneself; Combination 2: Pointing to oneself and body leaned forward; Combination 3: Pointing to oneself, head tilted to side, and body leaned forward; Combination 4: Pointing to oneself and head movement in jut position i.e., the chin touching the neck]

Figure 3. Mean frequency of occurrence of different group of strategies used by the participants in the expression of 1st person pronouns.

It was observed that pointing to oneself was the predominant strategy used to indicate 1st person pronoun. Various combinations of non-manual markers and pointing were also used to express the 1st person pronouns by the participants. This confirms the use of 'pronominalization' phenomenon by the participants. Similar observations have also been reported in sign languages such as American Sign Language (ASL) (Friedman, 1975; Padden, 1983), British Sign language (BSL) (Kyle & Woll, 1985; Meier, 1990), Swedish Sign language (Bergman, 1980), Brazilian Sign Language (LSB) (Berenz, 2002) and Croatian Sign Language (HZJ) (Alibasic Ciciliani & Wilbur 2006). It was also observed that the direction of pointing sign in the expression of 1st person pronoun differed from that of pointing sign used in the expression of demonstrative pronouns. The 1st person pronoun was expressed with index finger pointing and moving towards the signer's chest. On the contrary, demonstrative pronouns were expressed with index finger moving away from the signer to a locus in signing space. Thus, in Mumbai-Delhi dialect of ISL, the deaf signers used 'pointing to oneself' as a main strategy to express 1st person pronouns

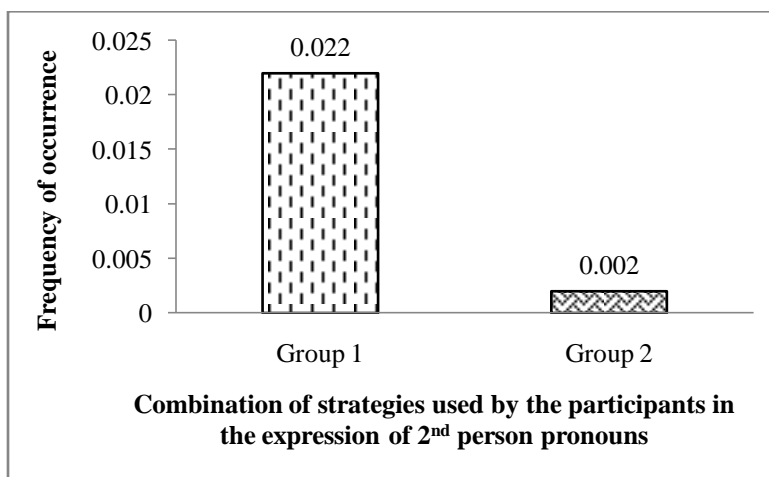
4. Frequency of occurrence of 2nd person pronouns

The frequency of occurrence of 2nd person pronouns was less when compared to frequency of occurrence of proper nouns and demonstrative pronouns. Few instances of 2nd person pronoun form such as 'You' were observed during narration of the story. It was observed that pronominal reference equivalent to 2nd person pronouns were carried out using few strategies such as pointing to space and use of non-manual markers. Various combinations characterizing pronominalization such as manual pointing to space accompanied by non-manual markers such as eye gaze, head bent down, body leaned forward towards referent were used and the mean frequency of occurrence of these is shown in figure 3. Similar observations are reported in several sign languages such as American Sign Language (ASL) (Berenz, 2002; Cormier, 2005, 2007; Hatzopoulou, 2008; McBurney, 2002), British Sign Language (BSL) (Kyle & Woll, 1985; Meier, 1990), Israel Sign Language (ISL) (Meier, 2003), Sign Language of Netherlands, (SLN) (Bos, 1995), Swedish Sign Language (SSL) (Bergman, 1980), and Brazilian Sign Language (LSB) (Berenz, 1996). The group of strategies that were used by deaf signers of Mumbai-Delhi dialect of ISL to express the 2nd person pronouns during the story narration task are as follows:

Combination1: Pointing to space, head bent down and body leaned forward

Combination 2: Pointing to space and eye gaze

The mean frequency of occurrence of combination of strategies used by the deaf signers to express the 2nd person pronouns are represented in figure 4.



[Combination 1: Pointing to space, head bent down & body leaned forward; Combination 2: Pointing to space and eye gaze]

Figure 4. Mean frequency of occurrence of different combination of strategies used by the participants in the expression of 2nd person pronouns.

Friedman test revealed a significant difference in frequencies [$\chi^2 (1) = 4.000, p < 0.05$] across strategies to express 2nd person pronouns. Further, pair-wise comparisons were made using Wilcoxon Signed Rank test to compare

frequencies between combination of strategies used to indicate 2nd person pronouns. There was no significant difference observed between combination 1 and combination 2. The use of other non-manual markers such as Eye gaze, head bent down and body leaning forward in the direction of the locus accompanied by manual pointing were used to augment the semanticity of the 2nd person pronouns. Similar findings have also been reported in other sign languages such as ASL (Bahan & Lee, 2000; Baker & Cokely, 1980; Bellugi & Fischer, 1972; Liddell & Metzger, 1998; Neidle, Kegl, MacLaughlin,), Israel Sign Language (Meier, 2003), Brazilian Sign Language (LSB) (Berenz, 2002) and Croatian Sign Language (HZJ) (Alibasic Ciciliani & Wilbur, 2006).

5. Frequency of occurrence of 3rd person pronouns

The participants used 3rd person pronouns such as 'It', 'He' and 'she' during the narration task. The 3rd person pronoun form 'it' was expressed by pointing towards the referent in space (Psp); the signed expression of 'He' involved signing the noun 'Boy'/'Girl' (as per the situation) followed by pointing towards the referent in space (Psp). These 3rd person pronouns (it, He & she) were also signed using non-manual markers such as eye gaze, head bent down, body leaned forward towards the referent, and head turn towards the referent. Similar findings have also been reported in several sign languages such as American Sign Language (ASL) (Berenz, 2002; Cormier, 2005, 2007; Hatzopoulou, 2008; McBurney, 2002), British Sign Language (BSL) (Kyle & Woll, 1985; Meier, 1990), Israel Sign Language (ISL) (Meier, 2003), Sign Language of Netherlands, (SLN) (Bos, 1995), Swedish Sign Language (SSL) (Bergman, 1980), and Brazilian Sign Language (LSB) (Berenz, 1996). The following combinations of strategies were observed in the signed expression of the deaf signers to express the 3rd person pronouns and the same is represented in figure 5.

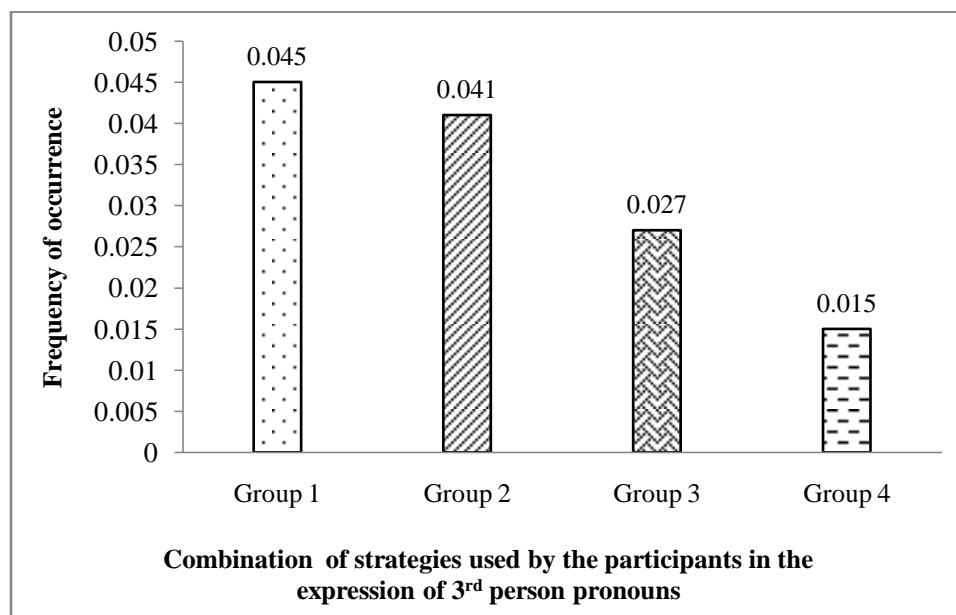
Combination 1:- Pointing to space, eye gaze, Head bent down and body leaned forward towards the referent in combination

Combination 2: Pointing to space and eye gaze in combination

Combination 3: Pointing to space, eye gaze, Head turn towards the referent and body leaned forward towards the referent

Combination 4: Pointing to space, Head bent down towards the referent in combination.

The mean frequency of occurrence of group of strategies used by the deaf signers to express the 3rd person pronouns are represented in figure 5.



[Combination 1: Pointing to space, eye gaze, Head bent down and body leaned forward towards the referent; Combination 2: Pointing to space and eye gaze; Combination 3: Pointing to space, eye gaze, Head turn towards the referent and body leaned forward towards the referent; Combination 4: Pointing to space, Head bent down towards the referent in combination]

Figure 5. Mean frequency of occurrence of different group of strategies used by the participants in the expression of 3rd person pronouns.

The use of non-manual markers in association with the manual pointing sign thereby provided additional semantic cue to the pronoun and the use of these non-manual cues in addition to manual pointing have also been documented in other sign languages such as ASL (Baker & Cokely, 1980; Bellugi & Fischer, 1972; Liddell & Metzger, 1998, Neidle, Kegl, MacLaughlin, Bahan & Lee, 2000), Israel Sign Language (Mier, 2003) Brazilian Sign Language (LSB) (Berenz, 2002) and Croatian Sign Language (HZJ) (Alibasic Ciciliani & Wilbur, 2006). The frequency of occurrence of various combinations of non-manual markers in association with manual pointing used by the deaf signers in the narration of the story to mark the 3rd person pronouns were compared.

The data of 3rd person pronouns was subjected to Friedman's test and the results did not reveal any significant difference in frequencies of occurrence [$\chi^2(1) = 4.000, p > 0.05$] between combination of strategies used to express the 3rd person pronouns. In other words, all these strategies were used equivalently by the deaf signers to refer to 3rd person pronouns.

6. Expression of 1st person possessive pronoun

The 1st person possessive pronouns like 'MY' & 'MINE' were used by the deaf signers while narrating the story. The participants expressed first person pronouns with open hand/palm directed towards the signer's chest (PSe). Similar findings have also been reported in sign languages such as ASL (Cormier and Fenlon 2009), Australian Sign Language (OGS) (Johnston & Schembri, 2007), and Hong Kong Sign Language (HKSL) (Tang & Sze, 2002).

Zeshan et al., (2004) reported that in most sign languages spatial inflections to mark person and number inflections are used; i.e., the pronouns are indicated by pointing to different locations in space to differentiate across person pronoun (first, second and third person pronouns) and possessive pronouns (first, second and third possessive pronouns).

Summary and Conclusions:-

The study aimed to analyze the expression of pronouns by deaf signers using Mumbai-Delhi dialect of Indian Sign language in a story narration task. The results revealed that the signed expression of the deaf signers in the story narration task involved use of proper nouns and different pronoun types such as demonstrative pronouns, 1st person pronoun, 2nd person pronoun, 3rd person pronoun and 1st person possessive pronoun. The proper nouns were expressed by the deaf signers using finger spelling of the noun sign or signing the noun sign. The expression of demonstrative pronoun by the deaf signers involved making a pointing sign in space accompanied by combinations of various non-manual markers such as eye gaze, raising of eye brows, body leaned forward in the direction of the referent, head movements such as head bent down, head turn toward the referent, head tilt to the side etc. The expression of the 1st person pronoun and 1st person possessive pronoun also involved manual pointing but, the pointing sign was directed towards the signers' chest accompanied by non-manual markers. The 2nd person pronoun and 3rd person pronoun also involved making a pointing sign in space accompanied by combinations of various non-manual markers. The expression of pronouns thus confirms the use of 'pronominalization' as a phenomenon in Mumbai-Delhi dialect of Indian Sign language (ISL).

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Annexure 1:-

The functions defined for the three tiers of analysis and coding of the signed expressions for the use of plurals by the participants using ELAN software.

Tiers		Functions
Tier 1	Sign Gloss	Identification of signs representing concepts is transcribed using glosses in 'Sign Gloss' tier.
Tier 2	Identifying types of pronouns	Identify the instances of signed expression of pronouns from the tier one, and the type of pronoun (1 st person pronoun, 2 nd person pronoun, 3 rd person pronoun, demonstrative pronouns, possessive pronouns etc) were identified and coded this tier
Tier 3	Identifying non-manual markers used in the expression of pronouns	Identification of the non-manual markers such as eye gaze, pointing, body and head positions etc as evidenced in the signed expressions of the participants for pronouns was carried out and coded across the tier.

Annexure 2:-

Operational Definition of different types of Pronouns.

	Definition
Pronouns	The part of speech that substitutes for nouns or noun phrases.
Personal pronouns	Pronouns which represents particular person, group, or thing in a phrase or a sentence. E.g. I, Me, You, We, Us, He, She, Him, Her, it, they, them.
1 st Person Pronouns	Pronouns which represents first person in a phrase/sentence. E.g. I, Me.
2 nd Person Pronouns	Pronouns which represents second person in a phrase/sentence. E.g. You.
3 rd Person Pronouns	Pronouns which represents person or thing spoken about. E.g. He, She, Him, Her.
Possessive Pronouns	Pronouns that represents the ownership of the noun in a phrase/sentence. E.g. mine (1 st person possessive pronoun), yours (2 nd person possessive pronoun), hers, his, theirs (3 rd person possessive pronoun).
Demonstrative pronouns	Pronouns that takes the place of particular objects or people. E.g. This, That, These, Those, Here, There.
Indexing/Establishing loci in space to indicate many	The participant points to each loci in the sign space and indicates several loci to indicate several referents.

Annexure 3:-

Operational Definition of Non-Manual Markers

Non-Manual Markers	Definition
Pointing	This involves making an action with an extended index finger or with the open hand to show the referent which can be a person, place or thing.
Pointing to self	Pointing to oneself/Pointing to one's own chest.
Pointing to space	Pointing towards the space to indicate the referent (present/absent) which can be a person, place or thing.
Eye Gaze	It is direction in which one looks at the person, thing, place etc steadily and intently.
Head positions and movement	Includes change in the position and movement of the head, such as head tilt (front/back), head turn (left/right), head movement (nod, side to side headshake, raised and bent down).
Body positions	Includes change in the orientation of the body which includes, body leaned forward/backward, body leaned sideways (right/left) etc.

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RESEARCH ARTICLE

LINSEED: A VALUABLE CROP PLANT

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Abstract

This article reviews the different aspects of economic botany of linseed /flax (*Linum usitatissimum* L.), namely, history and origin, morphology, area of cultivation, production and productivity, economic importance and cultural methods.

Key words

Economic Botany, Flax, Linseed, Uses.

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Introduction

Linseed or flax (*Linum usitatissimum* L., 2n= 30, X = 15) belongs to the order Malpighiales, the family Linaceae, and the tribe Lineae. It is the second most important *rabi* oilseed crop and stands next to rapeseed-mustard in area of cultivation and seed production in India. The genus *Linum* is composed of approximately 230 species but cultivated linseed / flax is the only species of economic importance in the genus (Rowland *et al.*, 1995; Tadesse *et al.*, 2010) and is one of the oldest plants cultivated for fibre and oil. Linseed is popularly known as *Atasi*, *Pesi*, *Phesi* or *Tisi* in Odia. Although linseed plants have several utilities, it is cultivated commercially for its seed, which is processed into oil and after extraction of oil, a high protein stock feed is left (Sankari, 2000; Kurt and Bozkurt, 2006). Linseed oil has been used for centuries as a drying oil whose oil content varies from 33-45% (Gill, 1987). About 20% of the total linseed oil produced in India is used by farmers and rest about 80% goes to industries for the manufacture of paints, varnish, oilcloth, linoleum and printing ink etc. Fibres obtained from the stem are known for their length and strength and are two to three times as strong as those of cotton (Taylor, 2012). The fibre is lustrous and blends very well with wool, silk and cotton etc. Linseed has an important position in Indian economy due to its wide industrial utility. But the national average productivity of linseed seed is quite low as compared to other countries. In India, linseed is grown mostly under rainfed (63%), *utera* (25%), irrigated (17%) and in input starved conditions in major linseed producing states i.e. Madhya Pradesh, Chhattisgarh, Maharashtra, Jharkhand, Uttar Pradesh and Odisha (Srivastava, 2009).

Due to the availability of other cheaper petroleum products with equally good or some times better drying properties, the widespread use of linseed oil is restricted during recent times in the paints and varnish industries. However, linseed oil continues to be the base stock in medicinal, chemical, pharmaceutical and cosmetic industries as they have renewable, biodegradable properties and are non-allergic in nature along with enriched phytochemical contents.

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In order to make linseed crop more remunerative and to generate employment, product diversification for medicinal and other industrial purposes of linseed needs to be improved by concerted research efforts. One of the major lines of researches is crop improvement programme through recombination breeding using elite germplasm for development of double purpose (oil and fibre) crops having high yield and stress resistance (both biotic and abiotic) qualities.

History and origin

The use of flax for the production of lilen dates back to 3000 B.C. as evidenced from the pictures on tombs and temple walls at Thebes of flowering flax plants. The use of flax fibre in the manufacturing of cloth in northern Europe dates back to Neolithic times (New Stone Age). The origin of flax (*Linum usitatissimum* L.), which is one of the oldest of cultivated plants is uncertain. However, it is generally accepted that linseed has originated from "Fertile Crescent"- an area east to Mediterranean Sea towards India (Zeven and Zhukovsky, 1975; Anonymous, 2010) and was probably first domesticated there. The generic name "*Linum*" comes from Celtic word *lin* means thread and the species name "*usitatissimum*" given by Carl Linnaeus, means very useful. So it directly refers to its multiple applications and their importance in ancient times.

Linseed is supposed to have originated in the four centres of origin, viz. Central Asiatic Centre, Near Eastern Centre, Mediterranean Centre and Abyssinian Centre. It spread northward to Europe and other parts of Asia and southward to India. But the botanical origin of linseed has not yet been determined with certainty. However, two theories such as polyphyletic origin and origin from the wild species of *Linum angustifolium* Huds. (2n=30) have been postulated (Gill, 1987).

Theory of polyphyletic origin regarding the probable progenitor of *Linum usitatissimum* L. is based on the postulation on the varying etymology of the names (*lin*, *llion*, *linu*, *linon*, *lien*, *lan*, *linseed* etc.), which existed in the European language and the antiquity of its cultivation in Egypt, Europe and the north India. According to this theory, the origin of the Indo-Gangetic types of cultivated flax (tall plants, small seeded and low oil content) originated through natural crossing of *Linum usitatissimum* with *Linum strictum* and the Peninsular types (shorter plants, bold seeded and more oil content) were derived through natural crossing of *Linum usitatissimum* with *Linum perene* or *Linum mysorensense*. However, experimental evidences to support this hypothesis are still lacking, as it seems to have been based on observation rather than experimentation.

The view of origin of *Linum usitatissimum* L. from a single wild species *Linum angustifolium* Huds. is from Mediterranean region, which might be the probable primary centre of origin of flax. It seems apparent that the species, which might have been cultivated by the lake-dwellers of Switzerland and Italy, was *Linum angustifolium* Huds. This theory is based on the archaeological remains of early civilization and enough experimental evidences. Both species are annual and grow in winter and have long slender stem with profusely branching system. They have small deep blue flowers and dehiscent capsules. The chromosome number in both the species is same (2x=30). However, available evidences indicate that *Linum bienne* Mill. a small seeded flax, might have contributed some germplasm to be the probable progenitor of *Linum usitatissimum* L. The species might have originated from Kurdistan and Iran. However, experiment by Muravenko *et al.* (2003) showed that three species viz. *Linum usitatissimum* L., *Linum bienne* Mill. and *Linum angustifolium* Huds. were assumed to have originated from a common ancestor, *Linum angustifolium* Huds. being closest to it.

Morphology

Linseed (*Linum usitatissimum* L.) is an erect annual herbaceous plant 30-120 cm, in height with slender glabrous, grayish green stem. The linseed types are grown for extraction of oil from the seeds. The flax types are grown for fibre extraction from the stems. The flax types are relatively taller (80-120 cm) in height with straight culms, less number of secondary branches towards the top of the stem (Gill, 1987). These plants generally produce fewer capsules and smaller seeds. The former (linseed) type is 60-80 cm in height and the plant has a short tap root system with fibrous branches. The shoot is profusely branched and bushy in character (Plate 1). Leaves are without stipules, 20-40 mm long and 3 mm broad, simple, narrow, alternate, lanceolate, acute or acuminate with a smooth upper surface and grayish green in colour. Flowers are showy, variously shaped (Plate 2), regular, hermaphrodite, pentamerous, hypogynous and borne in loose terminal raceme or open cyme with blue, white or pink colour. The calyx consists of five ovate acuminate persistent sepals. The corolla consists of five free petals, imbricate or twisted in the bud, fugacious, often clawed, bluish or white, deciduous which fall before noon. The androecium has ten

stamens present in two whorls. But the outer whorl consists of five stamens being reduced to staminodes. The inner five fertile stamens are widened to form a fused ring which surrounds the base of the gynoecium (Plate 3). The petals are narrow at their bases and insert into this ring. Nectar secreting glands are present at the widened bases of the stamens. In most of the flowers of *Linum usitatissimum* L., the anthers encircle and reach over top of the stigma, but in some varieties, the stigma extends beyond the anthers slightly. The anthers are two celled, introrse and dehisce longitudinally. The superior gynoecium consists of five united carpels. The ovary is ovoid and ten celled due to the presence of false septum in each carpel and producing up to two ovules in each carpel. Ovary has axile placentation and ovules are pendulous and anatropous with five styles, which are filiform, free or united below and twisted together forming slightly club-shaped stigmatic surface. Fruit is small round smooth globular capsule of 5-9 mm diameter. *Linum usitatissimum* L. is the only species with non-dehiscent or semi-dehiscent capsules for modern cultivation of the family Linaceae (Getinet and Nigussie, 1997). It contains up to 10 smooth, glossy apple pip shaped, light brown colour seeds which is 4-7 mm long. In most of the varieties, the capsule is of indehiscent type. Linseed is predominantly self pollinating. Cross pollination can occur at a very low level (less than 2%) by insects (Tadesse *et al.*, 2010). Wind pollination is not seen because the pollens are relatively heavy and sticky (Anonymous, 2010).

Area of cultivation, production and productivity

Linseed is being cultivated in Egypt, Europe and India since pre-historic times. The important linseed growing countries are India, Russia, Canada, Argentina, and the U.S.A. From the 21.12 lakh hectares global area, 41.62% i.e. 8.79 lakh ha. belongs to the Asian region with 5.35 lakh tonnes contribution to the total world production (21.23 lakh tonnes). Productivity of this region (608 kg/ha) is approximately 60% of the world productivity of 1006 kg/ha (Anonymous, 2011a). India is the second largest (21.21%) linseed growing country in the world in terms of area of cultivation after Canada. Production wise, India ranks 4th (8.20%) in the world after Canada (40.51%), China (18.68%) and the USA (10.89%) (Srivastava, 2009). But as per Food and Agricultural Organization Statistical data (FAO-STAT, 2007), India ranks 3rd (9%) in the world's top 20 linseed producing countries. However, in terms of productivity, India (449 kg/ha) is far below than Canada (1492 kg/ha), USA (1484 kg/ha), Egypt (1469 kg/ha), Russia (1272 kg/ha) and China (944 kg/ha). Although, the area of cultivation of linseed is decreasing, the productivity is increasing not only in India (+17%), but also all over the world (+18%).

Now, linseed is under cultivation in as many as 13 states of India, viz., Madhya Pradesh, Maharashtra, Chhattisgarh, Uttar Pradesh, Jharkhand, Bihar, Odisha, Karnataka, Nagaland, Assam, West Bengal, Himachal Pradesh, and Rajasthan. At present, linseed is cultivated in about 3.420 lakh ha with the contribution of 1.537 lakh tonnes to the annual oilseed production of our country and the yield being 449 kg/ha is far below the world production of 21.23 lakh tonnes from 21.12 lakh ha with productivity of 1006 kg/ha (Anonymous, 2011a). However, there is significant increase in the productivity of the states like Rajasthan (2006 kg/ha), Bihar (846 kg/ha), Nagaland (689 kg/ha) and Assam (517 kg/ha) is at a par with Asia (608 kg/ha) and the world (1006 kg/ha).

Economic Importance

Linum usitatissimum L. occupies an important position in world market because of its multiple trade use. It is a valuable crop and every part of the plant has specific economic importance.

Uses of flax fibre

Flax fibre is one of the most natural and eco-friendly fibres among all the textile fibres. The characteristic features of flax fibres are their strength, fineness and durability. They are lustrous, stronger, less stretchy, more durable and more resistant to environmental fluctuation than cotton and jute. This fibre blends very well with wool, silk, cotton etc. Bundles of fibres look like blonde hair. The threads are very strong and so are used for shoe making, manufacture of fishing lines and nets etc. The fibre is extensively used in the manufacture of canvas, twine, carpets, blankets and mats. Rough and coarser grade fibres are used in the manufacture of strong ropes, shipping cord, twines and cordage, which are very indispensable for aeronautical and defense purposes. Flax fibre textiles which are called 'lilen' or 'linso-fabrics', in which best grades of flax fibres are used for manufacturing suiting, shirting, bedsheets, cloth laces, damasks, curtains etc. Flax-jute (Linju) and Flax-cotton (Linco) blended fabrics show better quality than cent-percent jute or cotton fabrics (Pandey and Dayal, 2003).

Pulp sweeteners

After extraction of fibre from stalk, the woody core of the stem and short fibres are used as raw pulp for making high grade paper. To increase the quality of the paper more than 20% strong virgin wood fibre must be added to the pulp. This extra strong fibre which is to be added to the pulp is called as "pulp sweeteners". As flax fibres are stronger and longer than any other virgin wood fibre, a small quantity of flax fibre can be used in place of virgin wood fibres. This pulp can be utilized for the manufacture of paper used for currency notes, air mail, parchment paper, good writing paper, cigarette paper and straw boards of all grades of economic value.

Geotextiles (Insulation)

Coarse and fine flax fibres are blended and processed to produce insulation batts (fibre wadded into sheets) having similar insulation properties to fibre glass batts in order to insulate walls and ceilings (Jacobsz and Vander Merwe, 2012).

Plastic composites

As flax fibres are cheaper and lighter in weight, these can be used in place of fibre glass in the manufacture of plastic composite applications (car dashboards, fencing materials, septic tanks etc.). Flax fibre is better known as plastic crop (Vittal *et al.*, 2005), due to their importance in the use of paper and plastic in an eco-friendly condition.

Wax

Commercial wax is extracted from the cortical tissues by different organic solvents. This wax is used in shoe polishes (Gill, 1987).

Commercial industrial grade oil

Linseed oil is a versatile "drying oil" as it undergoes polymerization due to the presence of di- and tri-unsaturated fatty acids. So it is converted into a solid form when exposed to oxygen in air. The polymer forming properties facilitate linseed oil to use by its own or blended with other oils, solvents and resins etc. Raw linseed oil dries in a slow rate. It has tendency of yellowing with poor colour retention. In order to improve the drying and colour retention properties of linseed oil, some processes have been developed by heat modification (heating raw linseed oil at different high temperature range). Linseed oil is also blending or combining with dryers (compounds like linoleates or resinates of lead, manganese, cobalt, zinc etc) or with other compounds/chemicals to meet the demand for special application in dyeing industry. These oils are called modified oils and named as boiled oils (raw linseed oil heated at 90°C-150°C with dryers), blown oil (blowing air through raw linseed oil at 130°C), stand oil (alkali refined), enamel oil (mixing of linseed oil with tung oil), sulphurized oil (linseed oil treatment with sulphur chloride), styrenated oil (reacting with styrene), esskol and solinox oil (hydrogen treated oil) and polymerized oil (treatment with chlorinated hydrocarbon with aluminium trichloride, boron trifluoride, silicon tetrachloride or titanium chloride). These oils have high industrial values as these types of oils are used in the manufacture of paints, varnish, waterproof materials, patent leather (Japanned leather) and special finishes for cotton and silk fabrics etc.

Commercial edible grade oil

The linseed oil is not edible because it contains high linolenic acid, a poly-unsaturated fatty acid, which makes the oil highly susceptible to oxidation as a result of which its drying property increases and gives a pungent flavour and rancidity. Further, the laxative properties of the mucilage in seed coat of the seed make the oil unsuitable for edible purpose. A number of new flax seed varieties, which are low in alpha-linolenic acid content (25-30%) have been identified in India. It produces high quality poly unsaturated edible oil developed through plant breeding techniques. Linola[™] is the trademark for a new form of linseed in Australia developed in 1984 (Srivastava, 2009). These were specifically bred to produce edible oil, rather than for the traditional industrial use (Budwig, 1995). 'Solin' in Canada is another edible variety of linseed developed in 1990/91 (Srivastava, 2009). There are some farmers who are socially and economically forced to cultivate only linseed for years together. Some populations are there, who use only linseed oil for edible purpose. It is a basic need to develop linseed varieties with edible grade oil for these people. By the joint research efforts of Bhaba Atomic Research Centre (BARC) and All India Coordinated Research Project (AICRP) on linseed, five genotypes *viz.*, TL11, TL 26, TL 27, TL36 and TL43 have been developed having less than 1% linolenic acid content (Srivastava, 2009). Further, a number of attempts have been made by induced mutation techniques to develop low linolenic acid types of seeds for edible purpose (Budwig, 1995). When the seeds were treated with chemical mutagens, varieties with less than 2% linolenic acid were developed. Linola oil is better than other edible oils such as sunflower, safflower and corn oil with regard to its composition of low saturated fatty acids (palmitic acid, 6%; stearic acid, 4%; oleic acid, 16% and high unsaturated fatty acid content linoleic acid,

72% and low unsaturated fatty acid content alpha linolenic acid, 2%). However, as edible oil, its prospects may not be progressive in comparison to traditional edible oils like Sun flower, Safflower and Soybean oil etc. (Nagaraj, 1995). Again, the residue (cake) left after extraction of oil from Linola seed is a valuable source of protein and energy for animal feeding. But with respect to its protein and carbohydrate composition, Linola cake is high in linoleic acid and low in linolenic acid. So linola cake can be used for cattle and horses wherever, linseed cake is currently used in animal feeding (Budwig, 1995).

Human consumption

Due to the presence of higher concentration of health promoting omega-3 fatty acids (alpha-linolenic acid), which lowers cholesterol level and imparts cardio vascular benefits, many linseed based recipes have been standardized. The crushed seeds/flour is used for value addition and for making various nutritious food preparations (Chauhan *et al.*, 2009). But the linseed oil is not edible due to the laxative properties of the mucilage in the seed coat and presence of higher level of linolenic acid which causes rancidity and emits pungent flavors on oxidation. So on a very small scale, it is used for edible purpose as flax seed breads, bagels and fried food stuff by a small segment of people (Anonymous, 2006). In Karnataka and parts of Maharashtra, linseed is used as a traditional food adjunct-chutney powder. It is also used as a medium of frying certain foods in Himachal Pradesh (Preethi, 2009).

Nutritional value

Linseed possesses many vital nutrients and nutraceuticals with promising health benefit for both human beings and animals. It contains eight essential amino acids, *viz.*, isoleucine, leucine, lysine, methionine, phenylalanine, threonine, tryptophan and valine. It also contains carbohydrates, vitamins, minerals, crude fibre and this is the best herbal source of omega -3 and omega -6 fatty acids (Anonymous, 2006). Health experts prescribe these nutrients and other compounds for better health. One hundred thirty grams (one cup) of ground flax seeds give about 585 kcal energy, 26.0g protein, 53g total fat, high levels of dietary fibres (36.0g), total carbohydrate (38.0g) and all necessary vitamins, minerals for human body (Anonymous, 2003). So in recent years, it has attracted considerable interest due to its nutritional composition which has positive effects on disease prevention providing health beneficial components such as alpha linolenic acid, lignan and polysaccharides (Bozan and Temelli, 2008). Omega-3 fatty acid (alpha-linolenic acid) is a substrate for the synthesis of longer chain poly-unsaturated fatty acids mainly eicosapentaenoic and docosahexaenoic acids, which influence the biophysical properties of the cell membrane and are required for normal cell functions (Preethi, 2009).

As fodder

After the extraction of oil from the linseed seed, the residue left behind is called cake, which is brown in colour. Prior to defatting (to remove all or almost all), this cake contains 21.78% of non nitrogenous extract, 29.37% lipids and 27.78% protein, 7.02% fibre, 3.40% ash and 10.65% total humidity (Gutierrez *et al.*, 2010). So it is a protein rich palatable feed for livestock. It is fed to the cattle to improve their health condition and to develop the gloss of their coat, although linseed meal is used as an additive in baking products (Coskuner and Karababa, 2007). However, linseed contain phytic acid, cyanogenicglucoside and goiterogen which limit the linseed cake as animal feed as these are anti nutritional factors. So meal or seeds should be properly processed to remove the toxic constituents.

Mucilage

Linseed mucilage which resides in seed coat is obtained by precipitation process from aqueous extract of seed (soaking in water for 24 hours) and gives high viscosity solution. It is a white fibrous mass and friable when fully dry and gives high viscosity solution when gets dissolved in water. It is a heterogeneous mixture of polysaccharides made up of xylose, glucose, galactose, arabinose, ramnoseand galacturonic acid. Taking the advantage of the physical properties of linseed mucilage as a water soluble emulsifier, thickener and binder, it is used as a thickener and emulsifier in food products. The potential application of linseed in foods as functional food stuff developed a great interest. The mucilage obtained from flax seed is very similar to gum arabic in its emulsifying properties (Coskuner and Karababa, 2007) and is comparable to guar gum in its capacity to bind water. Mucilage is used in cosmetic and pharmaceutical industries as ademulcent and as a useful base for eye ointment.

As fertilizer

Linseed oil cake is one of the best nitrogenous fertilizer among oilcakes with respect to nitrogen, phosphorus and potassium (4.7% N, 11.7% P₂O₅, 1.3% K₂O) contribution to soil (Anonymous, 2011b). It provides slow and steady nourishment, stimulation, protection from soil nematodes (particularly *Meloidogyne javanica*) and insects. So it also

improves yields and quality of product like taste, flavour and amino acid composition etc. Moreover, it can also be used as a manure to prevent soil from unwanted microbes due to its germicidal properties by improving plant health and thereby gives greater resistance to infection (Anonymous, 2011b).

Composition of linseed seed

Seed is small, flat, oval, brown or yellow or deep brown coloured. The seed composition varies with variety, size, climate and maturity. The seed contains oil (36-48%), which is high in unsaturated fatty acid especially linolenic acid (Khan *et al.*, 2010 and Rahimi *et al.*, 2011) and about 6% mucilage, which resides in the seed coat. The seed has all essential components like crude fibre (5-10%), proteins (20.3%), fats (37.1%), minerals (2.4%), carbohydrates (28.9%), moisture (6.5%), calcium (170 mg/100 g), iron (370 mg/100g), carotene (2.7mg/100 g), thiamine (0.23 mg/100g), riboflavin (0.07 mg/100g), niacin (1.0mg/100g) together with wax, resin, phosphorus, sterols and small quantity of cyanogenic glucoside-linamarin (Nagaraj, 1995) and all essential amino acids such as isoleucine, leucine, lysine, methionine, phenylalanine, threonine, tryptophan and valine (Weiss, 1983). Though the amino acid composition is similar to that of any other oilseed, it is rich in lysine and poor in methionine and the quality of linseed protein is better than that of other rapeseed proteins (Nagaraj, 2009). Flax seed contains at least three types of phenolics such as phenolic acids (8-10g/kg), flavonoids (35-70mg/100g) and lignans (secoisolariciresinol diglucoside, SDG) about (1-26mg/g). Due to the presence of all vital elements of proteins, sugars, fats, dietary fibres to a sufficient level now a days, fried and powdered seed is utilized in the preparation of day to day foods.

Linseed oil

The chief constituent of linseed seed is 30-45% oil. The oil is obtained by cold pressing (mechanical extraction with reduced heat, which is produced by rotational friction) has a golden yellow colour whereas a hot pressed oil (heat supply up to 200°C) is yellowish brown. Freshly extracted oil is a viscous, distinct odour and bland taste.

Fatty acid composition of linseed oil

In the aliphatic chain of fatty acid, carboxyl end is called alpha and the methyl end is called omega. From the omega end there is double bond at 3rd carbon called omega-3 and double bond at 6th carbon called omega-6. Body of the human being cannot produce its own omega-3 and omega-6 fatty acids. So there is requirement of omega-3 and omega-6 rich food in the daily diet (Brenna, 2002). Linseed oil is a mixture of triglycerides. The different fatty acid constituents of triglyceride are triple unsaturated α -linolenic acid (an omega-3 fatty acid) (35-67%), saturated palmitic acid (4-16%), saturated stearic acid (0.3-10.0%), mono unsaturated oleic acid (13-38%) and double unsaturated linoleic acid (an omega 6- fatty acid) (7.0-18.0%).

Tocopherols, phytosterols (plant sterols) and campesterol

Tocopherols are a group of closely related fat soluble alcohols which behave like vitamin E. Natural tocopherols exist as a mixture of four homologous series such as α , β , γ and δ tocopherols. Linseed oil contains higher levels of β tocopherols (200 ppm) than α tocopherols (15-20 ppm) and γ tocopherols (5-7 ppm). Phytosterols (plant sterols) are the group of steroid alcohols present in linseed oil (about 70 ppm). In addition to above, linseed oil also contains campesterols, which are the group of phytosterols and their chemical structure is similar to that of cholesterol (23-27 ppm). Due to the presence of higher levels of unsaturated fatty acids (more than 80% of the total fatty acids) and lower levels of α and γ tocopherol in linseed oil, the oil is highly unstable with limited shelf-life. So the oil gives bad frying smell (Nagaraj, 2009).

Medicinal value

Linseed seed contains omega-3 fatty acid in the form of alpha linolenic acid (ALA) which gives cardiovascular benefits by affecting prostaglandins and leukotrienes which are related to blood clotting and inflammatory disorder like rheumatoid arthritis (Anonymous, 2005). Gamma linolenic acid found in flax seed showed tremendous effect in diabetics by normalizing the faulty fatty acid metabolism for diabetics. Linseed contains antibiotics called "Linatine" in its seed, which cures diseases for which no other medicine is effective (Anonymous, 2006). Flax seed contains both soluble and insoluble fibre. The soluble fibre can lower the blood cholesterol and insoluble fibre have laxative property which help to treat varieties of gastrointestinal conditions and provide health benefits for diarrhoea, constipation, irritable bowel syndrome (IBS) and inflammation in the lining of intestines. Linseed is one of the richest sources of lignans which are phytoestrogens, provide protection against certain forms of cancer (Morton *et al.*, 1994). Flax seed sprouts induce apoptosis and inhibit cancer cell growth by demonstrating their anti proliferative effects in breast cancer cell (Lee, 2012). Recent reports (Adugna and Labuschagne, 2003 and Tripathi *et al.*, 2013) indicate that consumption of linseed imparts numerous health benefits, including anti-

hypercholesterolemic and anti-carcinogenic effects and is also beneficial in the development of brain and retinal tissues. Linseed oil may alleviate some cases of Alopecia Areata (hair loss) and facilitates weight loss in persons suffering from obesity. It is also useful in the treatment of some cases of Edema. Linseed oil is the only dietary oil allowed in the orthomolecular treatment (use of naturally occurring nutrients in maintaining health and treating disease) of Acquired Immune Deficiency Syndrome (AIDS) patients. Further, this oil is useful in the alleviation of some cases of Pre Menstrual Syndrome (PMS) and makes pregnancy less eventful, makes deliveries easier and produces healthier offsprings (Tripathi *et al.*, 2013). Linseed tea, which is prepared by boiling 1 part of linseed seed with 20 parts of water till grains are soft, is used as a demulcent in respiratory irritation and in intestinal or urinary catarrhs (Vajpeyi *et al.*, 2005). The anti-microbial activity of linseed oil and its therapeutic efficacy in bovine mastitis (inflammatory disorder) has been reported recently (Kaithwas *et al.*, 2011). Linseed oil is recommended for the treatment of piles. It is also used as a base for different cosmetic and pharmaceutical products. Linseed oil is traditionally used in various skin infections externally as a soothing application for burns, wounds, acne, eczema, psoriasis and boils. This oil is available in the form of *Arectal oil* prepared by equal quantity of lime water with linseed oil (David and Toms, 2006) for various skin treatments. Linseed oil is used as a carrier of vitamins and irritant drugs. Crushed linseed is used in the form of poultice, which is prepared by boiling 28g linseed seeds in 72ml of water. Before its application it may be sprinkled with boric acid. Cough syrup in the name of 'Mucilage lini' is prepared from the mucilage of linseed taking 1 part of mucilage with 8 parts of water.

Linseed oil used as surface treatment materials

Boiled linseed oil is one of the widely used surface treatment materials for protection of concrete structures (Pfeifer and Scali, 1981). It can be used either in the form of solution or emulsion. The solution is prepared by taking linseed oil and mineral spirit with the proportional ratio ranging from 90:10 to 30:70, but commonly used ratio is 50:50. In order to accelerate the dryness of the oil, some dryers such as cobalt, manganese, lead salts or naphthenic acids are often added to the solution. Another type of linseed solution is available taking 50% linseed oil with 50% kerosene. On the other hand, in a linseed oil emulsion equal volume proportions of oil phase is added with water phase (Xie *et al.*, 1995).

Development of linseed varieties with good fibre quality

Now-a-days, eight double purpose linseed varieties (oil and fibre) have been developed. These are Gaurav, Jeevan, Nagarkot, Shikha, Rashmi, Meera, Parvati and Pratap Als-1 (Srivastava, 2009). The productivity of oil (1200-1600kg/ha) and fibre (860-1200kg/ha) of these developed varieties are better than traditional varieties. The disease resistance of these varieties are high but the quality of fibres collected from the available varieties are inferior in comparison to imported fibres from European countries which are used in our country for defense and industrial purposes (Srivastava, 2009). So, more research work is to be initiated to improve the quality of fibre by breeding/agronomic manipulations, which would save 35 crores of rupees foreign exchange and will generate more employment.

Characteristics of flax fibre

Flax fibres are an interesting alternative to mineral fibres due to its low cost, low density, high specific stiffness and recyclability nature. These constitute for their use as rigid composite materials (Baley, 2002).

Composition of flax fibre

According to Batra (1998), flax fibre is composed of cellulose (64.1%), hemicellulose (16.7%), pectin (1.8%), lignin (2.0%), water soluble compounds (3.9%), wax (1.5%) and water (10.0%). However, these compositions are relative to the variety of plant, soil quality, weathering conditions, the level of plant maturity and the quality of retting process etc.

Structure of flax fibre

The detailed structure of flax fibre cell reveals that it consists of two cell walls viz. primary cell wall (0.2µm thick) and secondary cell wall. They are arranged in concentric cylinders with a small channel in the middle called lumen, through which uptake of water continues. The main core of the fibre is contributed by secondary wall, which is designed as S₁, S₂ and S₃, from which S₂ is the main bulk of the fibre (Fig.1 and Fig. 2). Again it has been found that the flax fibre is strengthened by the highly crystalline cellulose fibrils which are spirally arranged in a matrix of amorphous hemicellulose and lignin (Fig.3). Each fibre is made up of macrofibrils, microfibrils and elementary fibrils. Macro fibrils are 0.5mm in diameter and are an assembly of microfibrils, which are 10 to 20 nm in diameter, which in turn are an assembly of up to 20 elementary fibrils called micelles. Each micelle is made up of 50 to 100

molecules of cellulose. The distance between two micelles is about 01 nm and about 10nm between two microfibrils (Baley,2002).

Cultural methods

Linseed is purely a *rabi* and cool season crop, which requires moderate to cool temperature during the growing season. This is generally confined to low elevation, and the plains. It can grow where annual precipitation ranges from 48-76 cm. If the crop is exposed to drought and high temperature (about 32°C) during and after the flowering stage, the yield, size and oil content of seeds are greatly reduced. Linseed also grows on marginal and sub-marginal rainfed soil, which may vary from light to heavy soil. When the plants become golden yellow the capsules turn brown and seeds in the capsules rattle (Plate 4). This is the appropriate time to harvest. In an irrigated field, the crop can produce seed yield of 12-15 q/ha.

Diseases with causal organisms and symptoms

Disease is one of the major constraints that limit productivity of linseed. The linseed crop is infected by all classes of pathogens *viz.* virus, bacteria, fungi and nematodes. There are a number of diseases to which linseed is exposed. But rust, wilt, *Alternaria* blight and powdery mildew are the major ones causing losses up to 80-100%, 80%, 27-60% and 60% respectively (Anonymous, 2005).

Rust (*Melampsora lini*)

Rust disease appears in the form of bright- yellow or orange coloured uredia on the leaves, stems and capsules. Uredia are generally circular on leaves and elongated on the stems and converted to pustules. Later the entire plant becomes covered by yellow-orange pustules. The disease commonly affects stem but more severely leaves, which die prematurely.

Wilt (*Fusarium oxysporum f. lini*)

Wilt is one of the most serious diseases in linseed causing a great loss. It develops in all the stages of their growth of the plants. Infection in seedling stage mostly causes falling off cotyledons. In the growth stage, dark green or brownish spots appear and these leaves shrivel. Infected plants ripen prematurely and develop black colouration of the vascular tissue in the stem and root of the plant. The dead and defoliated plants conspicuously appear in the field.

Alternaria blight (*Alternaria lini* and *Alternaria linicola*):-

All aerial parts of the plants are severely infected which causes heavy damage to the crop. However, disease appears on the floral parts particularly near the calyx in the form of small dark brown lesions. But on the young leaves, these lesions are large and extend to the stem. Later such leaves dry up and get twisted. Severely infected plants dry up by giving a burnt appearance in the field.

Powdery mildew (*Oidium lini*):-

The major symptom of this disease is the appearance of superficial whitish colonies of pathogen on the aerial parts including floral buds.

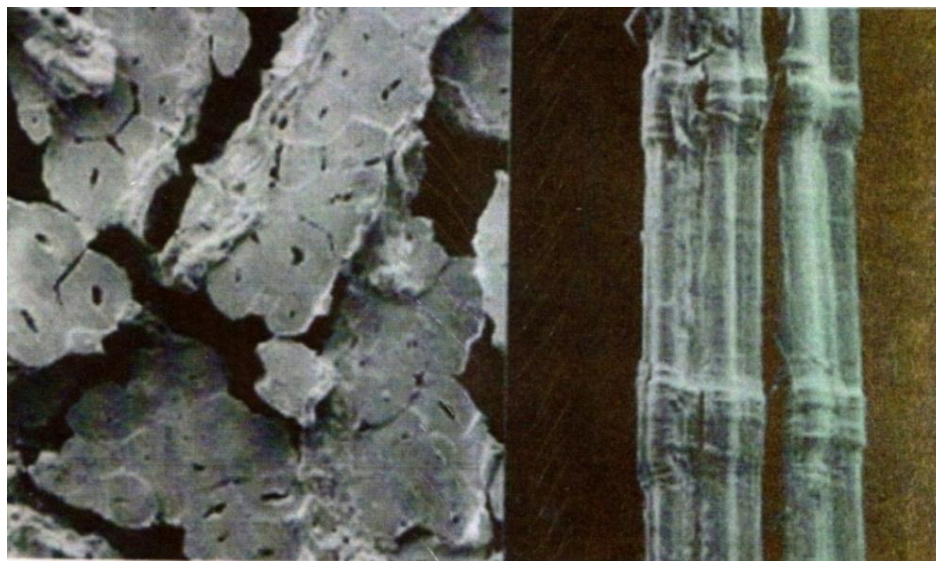


Fig.1: Structure of flax fibre cell under microscope.

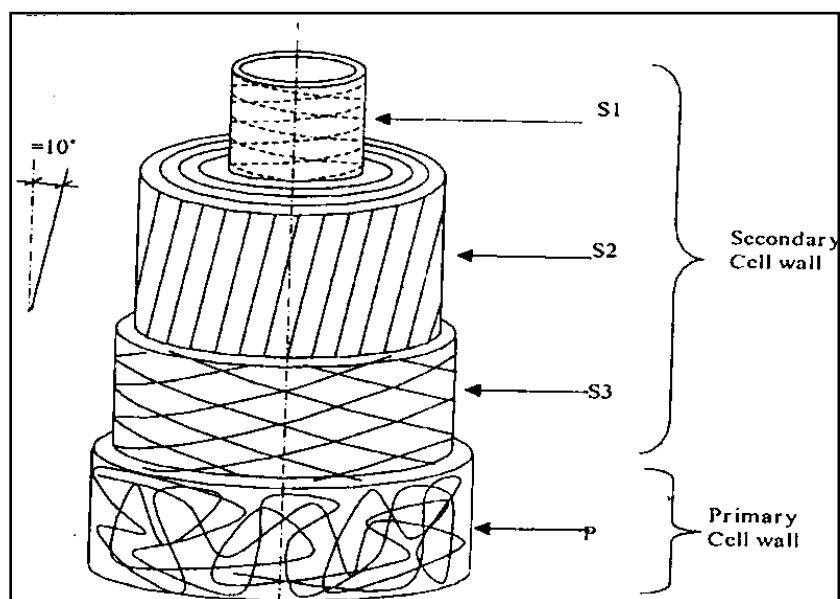


Fig.2: Characteristics of flax fibre.

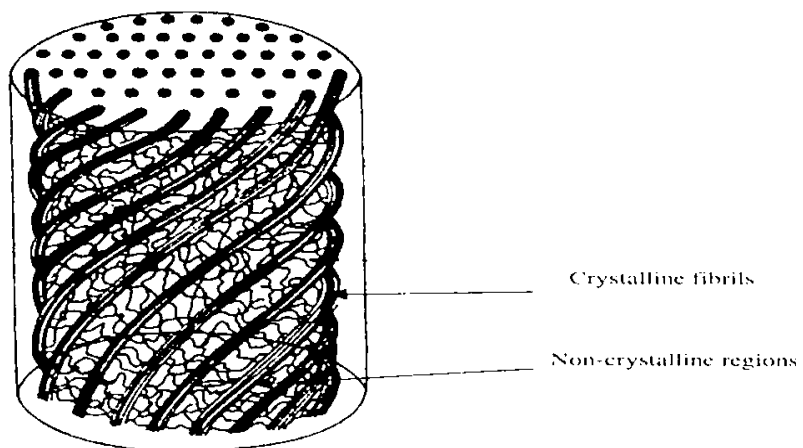


Fig. 3: Matrix of amorphous hemicellulose and lignin.



Plate 1: Plant types in respect to height and colour of flower; **a.** Bushy (Bushy branching); **b.** Semi erect (Lateral Branching); **c.** Red colour flower of linseed with Semi erect (Lateral Branching); **d.** Long erect (top branching)

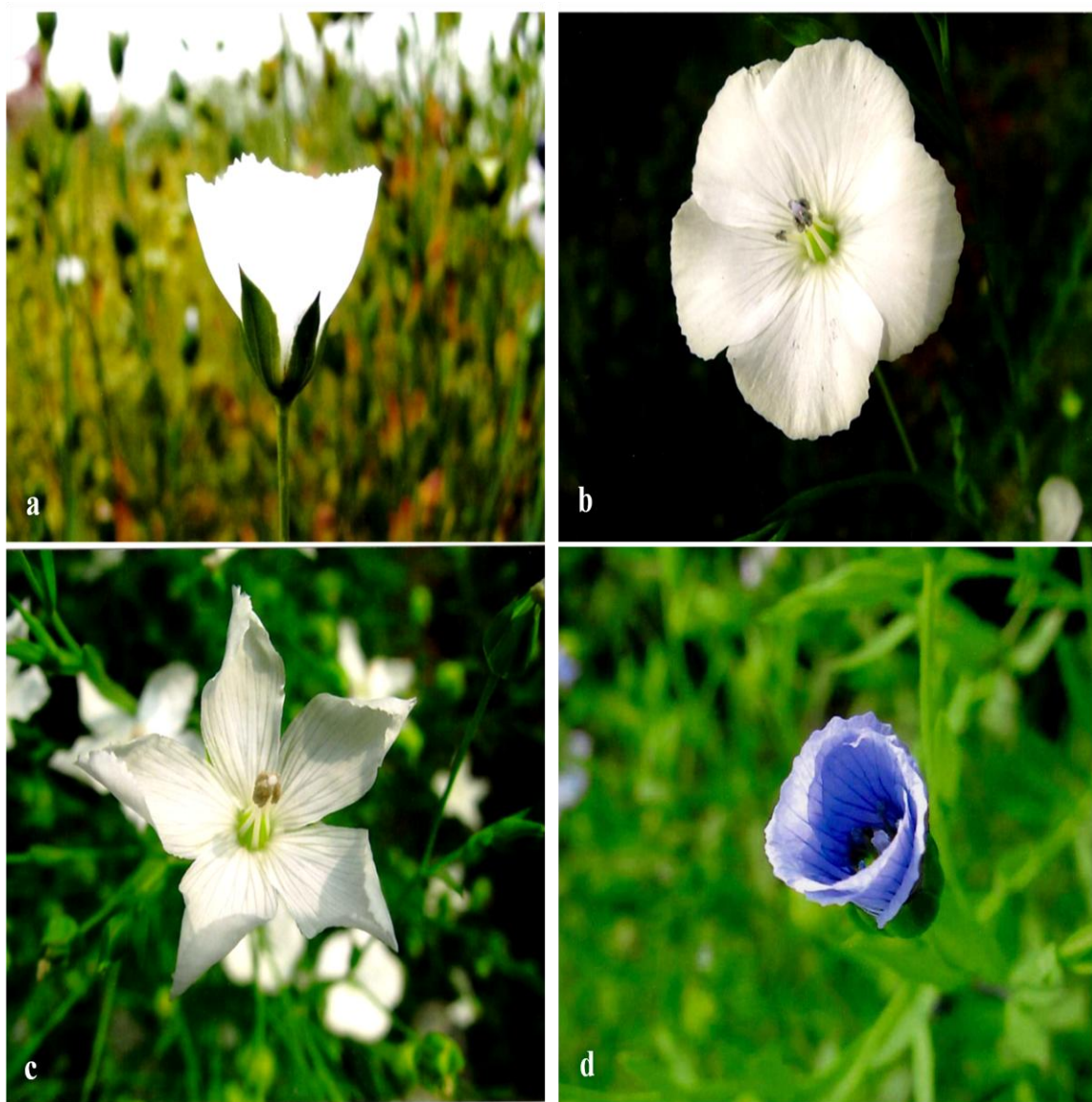


Plate 2: Shape of flower; **a.** Funnel shape; **b.** Disc shape; **c.** Star shape; **d.** Tubular shape



Plate 3: Flowering and fruiting of linseed plants; **a.** Individual flower with five petals; **b.** Fused stamens; **c.** Number of flowers in a plant; **d.** Capsules bearing seeds of *Linum usitatissimum* L. plant

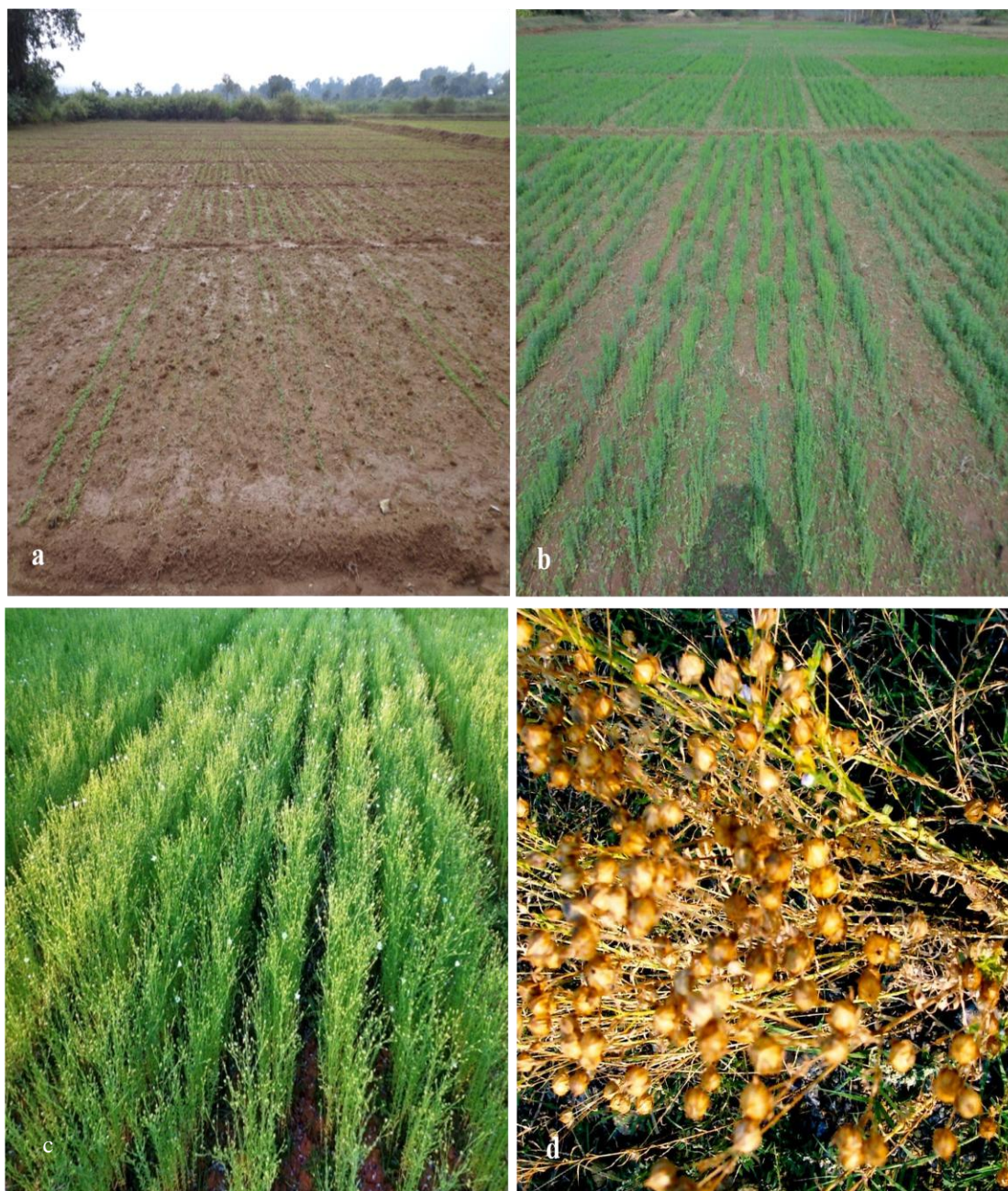


Plate 4: Field preparation and cultivation; **a.** Field shows pre-seed germination/starting of seed germination; **b.** Seedling stage observed from field; **c.** Linseed plants bearing a number of flowers in the erect branches. **d.** Matured capsules ready to harvest

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RESEARCH ARTICLE

RHEUMATOID ARTHRITIS SEVERITY INDEX AND ITS RELATION TO COMORBIDITY IN EGYPTIAN RHEUMATOID ARTHRITIS PATIENTS.

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Abstract

Objective: This study aimed to apply the previously validated Rheumatoid arthritis medical records-based index of severity index (RARBIS) to RA patients in Egyptians and its relation to Comorbidity.

Method: Data of RA patients retrospectively collected from medical records in the follow up unit of the Rheumatology and Rehabilitation Department of Zagazig University hospital. The data were collected as regarding socio-demographic and clinical characteristics of the studied RA patients, the collected data were computerized and statistically analyzed using SPSS program version 20.0

Results: A total of 1142 patients files were reviewed and included in the study, their ages ranged from (20-73) years and the duration of the RA disease ranged from (1-20) years.

We found that the total (RARBIS) score ranged from 3-16 with a mean of 8.09 ± 2.41 . Also, the most frequent comorbidities in our patients are osteoporosis, hypertension, and peptic ulcer. RARBIS total score was high significant statistically increased in RA patients with comorbidities compared to those without comorbidities. In addition, the mean age of RA patients, rural area residency and illiteracy, increasing disease duration, prescribed NSAIDs or prednisolone were associated with high frequency increase in RA comorbidities. It was found that no significant difference when comparing frequency of comorbidities with sex, smoking and working status.

Conclusion: Our study demonstrated that the Egyptian RA patients have relatively high scores of severity index, and most of patients with severe RA suffer from a number of associated comorbidities with significant relationship with the severity index.

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Introduction:-

Rheumatoid arthritis (RA) is the most common chronic inflammatory joint disease with the average prevalence of 0.5–1% in developed population¹, and of 0.3–0.5% in developing countries². This disease results in long-term disability and premature mortality in the patients³.

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Comorbid diseases are the medical conditions associated with RA. The mechanisms of this association may be due to the pathogenesis of RA itself, the effects of medications used for treating RA, or only a coincidence⁴. Substantial evidence indicates that the continuous systemic inflammation and immune dysfunction characteristic for RA plays a critical role in the development and acceleration of comorbidities⁵.

Comorbidities most frequently seen in patients with RA include cardiovascular disease, lung disease, malignancies, osteoporosis, changes in body composition and neuropsychiatric disease. Most of these comorbidities occur more frequently than expected in RA patients as compared to the general population. The average patient with RA has two or more comorbid disorder⁶.

RA disease severity is a major predictor of development of comorbidities. Surveys indicate that although clinical outcomes have improved in patients with RA, mainly due to access to more efficient pharmacotherapy, comorbidity remains a major issue in many patients. As a part of strategies to improve further the management of patients with RA, multidisciplinary collaboration for prevention and early detection of comorbidities is of major importance⁷. Because RA severity may affect both choice of treatment and incidence of comorbid conditions, RA severity is an important potential confounder in pharmacoepidemiologic studies⁸.

The patient may have severe RA but been unable to take more potent therapy due to contraindications, including comorbid conditions⁹. As the comorbid conditions may impact treatment regimens of RA, or the prescribed drugs may worsen the comorbidity. Additionally, in many instances, rheumatologist may be forced to prescribe RA medications that exacerbate the comorbid conditions. Therefore, to successfully manage RA, comorbidities should be carefully considered and they should be treated in addition to prescribing anti-rheumatoid medication regimens. As RA patients with more comorbidities experience greater functional impairment¹⁰. Rheumatoid arthritis medical records-based index of severity index (RARBIS) was developed through an expert Delphi panel of six rheumatologists, and convergent validity was assessed by comparing it with intensity of the actual RA treatments that patients received. The index includes indicators from five categories: prior surgical history (C1–C2 fusion and joint surgeries), radiologic findings (C1–C2 subluxation and erosions), laboratory findings (rheumatoid factor (RF), erythrocyte sedimentation rate (ESR), C-reactive protein (CRP), and platelet counts), clinical and functional status (arthritis flares, morning stiffness, physician global rating, and functional status), and extra-articular manifestations (vasculitis and pulmonary nodule)⁸.

This study used a previously validated (RARBIS) for rheumatoid arthritis to estimate the distribution of disease severity and the association between disease severity and comorbidity in Egyptian RA patients from Sharkia Governorate, Egypt

Methods:-

Subjects:-

Data of RA retrospectively collected from medical records in the follow up unit of the Rheumatology and Rehabilitation Department of Zagazig University hospital (Monocentric study). The Zagazig University Hospitals is the biggest hospital in eastern Egypt that provides health care to the population in that part of Egypt, as it full of highly qualified professors and doctors of various specialties.

The data were collected as regarding socio-demographic and clinical characteristics of the studied RA patients, We obtained information on surgical history (C1–C2 fusion and joint surgery), laboratory values (RF, ESR, CRP and platelet counts), extra-articular manifestations (subcutaneous nodules and vasculitis) and X-rays (C1–C2 subluxation, erosions), out-patient medications prescribed, comorbid conditions, and clinics attended were retrieved from the patients' physical file. Because the RARBIS does not require a physician's assessment for its calculation, it is easily applied retrospectively to medical records. All of the data for the RARBIS are accessible from medical records.

A total of 1142 patients files were reviewed, the diagnoses of RA based on the 1987 American College of Rheumatology criteria were included in the study, their ages ranged from (20-73) years and the duration of the RA disease ranged from (1-20) years.

Objective:-

This study aimed to apply the previously validated severity index for rheumatoid arthritis (RARBIS score) to RA groups in Egyptians and its relation to Comorbidity.

Statistical analysis:-

The collected data were computerized and statistically analyzed using SPSS program (Statistical Package for Social Science) version 20.0. Qualitative data were represented as frequencies and percentages. Chi-square test (X²) was carried out for comparing the qualitative data. Quantitative data were compared using student's t-test. The test results were considered significant when p-value <0.05 and highly significant when p-value <0.01.

Results:-

A total of 1142 RA patients were included in the study. Most of them were females (79.6%) with mean age of 43.86 ± 11.40 . Only (14.6%) of them were from urban areas and had higher education (15.1%). Most of patients were non-working (82.3%) but only (7.7%) were current smokers, with mean disease duration of about 9.24 ± 4.76 . As regarding treatment, more than half of patients (68.1%) were on non-steroidal anti-inflammatory drug NSAIDs, and (87.1%) on prednisolone (**Table 1**).

Table (1): Socio-demographic and Characteristics data of the studied RA patients:

Variables	No. (%) (n. =1142)
Age (years)	
Mean± SD	43.86 ± 11.40
Range	20 – 73
<50 years	402 (35.2)
≥50 years	740 (64.8)
Gender	
Female	909(79.6)
Male	233 (20.4)
Residence	
Rural	975 (85.4)
Urban	167 (14.6)
Level of education	
Illiterate	735 (64.4)
Basic education	235 (20.6)
Higher education	172 (15.1)
Current Smokers	88 (7.7)
Working status	
Non-working	940 (82.3)
Working	202 (17.7)
Disease duration (years)	
Mean ± SD	9.24 ± 4.76
Range	1 – 20
<5 years	179 (15.7)
5 – 10 years	616 (53.9)
>10 years	347 (30.4)
RA-related medications	
NSAIDs	778 (68.1)
Prednisolone	995 (87.1)

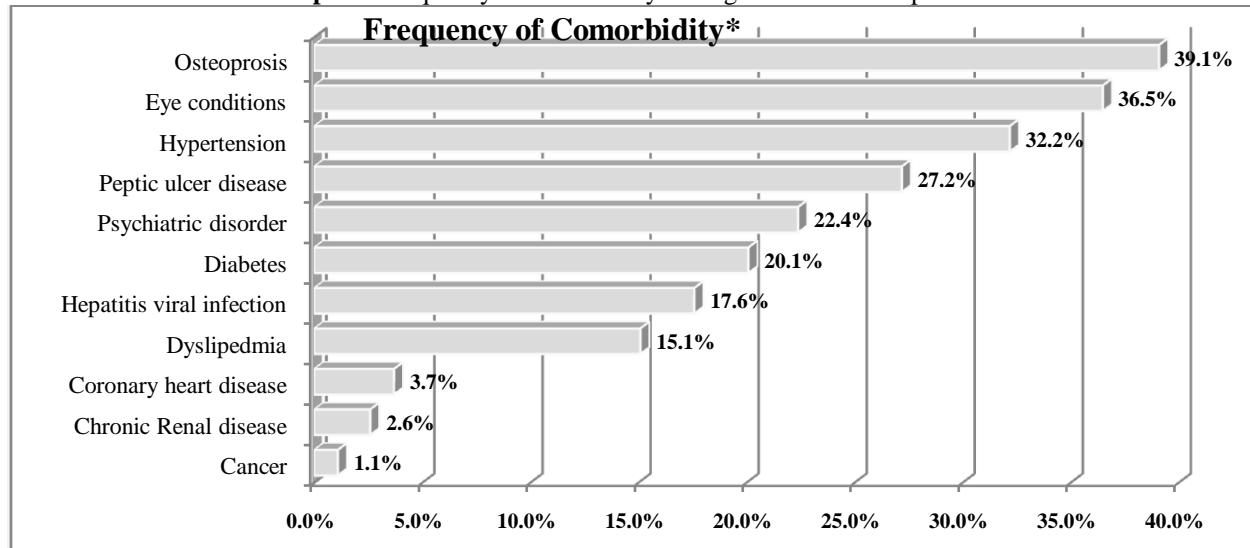
As regard to RARBIS of RA patients (**Table 2**), only 17 patients (1.5%) had prior surgical histories, while 473 patients (41.4%) had radiologic joint destructions. Extra-articular manifestations (vasculitis and pulmonary nodules) were evident in 61 (5.4 %) of RA patients, (RF) were positive in about half of them (53.3%), where the other Laboratory sub-scale (Erythrocyte sedimentation rate > age/2 or C-reactive protein > upper limit normal or platelets > 450 K) were observed in most RA patients (87.5 %). The optional RARBIS score with medication use was also calculated with the addition of data on medication use. As regarding treatment, more than half of patients were on

DMARDS (Disease-modifying Anti-rheumatic Drugs). The Summary score for extended index was calculated by summing the five subscales ranged from 3-16 with a mean of 8.09 ± 2.41 .

Table 2:- Rheumatoid arthritis medical records-based index of severity (RARBIS):

Sub-scale Points	No. (%) (n. =1142)
1. Surgery sub-scale: <ul style="list-style-type: none"> C1–C2 fusion Any hand joint Any foot joint Major joints (hips, knees, shoulder, elbow, wrist, ankle) 	5 (0.4) 0 (0.0) 2 (0.2) 10(0.9)
2. X-ray sub-scale: <ul style="list-style-type: none"> C1–C2 subluxation Any erosions 	21 (1.8) 452 (39.6)
3. Extra-articular manifestations sub-scale: <ul style="list-style-type: none"> Vasculitis Pulmonary nodule 	51 (4.5) 10 (0.9)
4. Clinical status sub-scale: <ul style="list-style-type: none"> Arthritis flares 1 2 to 4 5 + <ul style="list-style-type: none"> Worst physician global rating: "doing poor" Functional status Unable to do hobbies Unable to work Unable to care for self <ul style="list-style-type: none"> Hours of morning stiffness <1 1 to 4 >4	53 (4.6) 442 (38.7) 647 (56.7) 601 (52.6) 798 (69.9) 604 (52.9) 103 (9.0) 443 (38.8) 597 (52.3) 102 (8.9)
5. Laboratory sub-scale: <ul style="list-style-type: none"> Rheumatoid factor titre > upper limit normal Erythrocyte sedimentation rate > age/2 or C-reactive protein > upper limit normal or platelets > 450 K 	609 (53.3) 897 (78.5)
6. Optional medication sub-scale: <ul style="list-style-type: none"> Any of the following medications: hydroxychloroquine, gold, sulfasalazine Any of the following medications: methotrexate, leflunomide Any of the following medications: cyclophosphamide, azathioprine, cyclosporin, anakinra, adalimumab, etanercept, infliximab 	804 (70.4) 853 (74.7) 52 (4.6)
Summary score for extended index Mean \pm SD Range	8.09 \pm 2.41 3 - 16

Graph (1) showed that the frequency of comorbidity among the studied RA patient was (62.2%). Higher frequencies of comorbidities were noticed for osteoporosis (39.1%), eye (36.5%), hypertension (32.2%) and peptic ulcer (27.2%). (22.4%) of studied RA patients had psychiatric problems (mostly depression), (20.1%) were Diabetic and (17.6) had hepatitis viral infection. (15.1%) of the patients had abnormal serum levels of lipids. While cancer, chronic renal disease, and coronary heart disease, represented lower frequencies (1.1%, 2.6%, 3.7%; respectively).

Graph 1:-Frequency of comorbidity among the studied RA patients

* Patients can be counted more than twice if they have more than one comorbid disease, therefore the total will not equal (the number of patients in the cohort)

Relationship between socio-demographic and characteristics data of the studied RA patients and comorbidity is demonstrated in **Table 3**. Mean age of RA patients with comorbidity was highly significantly older than RA patients without comorbidities ($p < 0.000$). Also, rural area residency and illiteracy ($p < 0.000$) were significant predictors of comorbidity among RA patients. There was a highly significant increase in the frequency comorbidity with increasing disease duration ($p < 0.000$). As regarding treatment, patients with prescribed NSAIDs, or prednisolone showed significantly higher prevalent comorbidities ($p < 0.0001$).

It was found that no significant difference when comparing frequency of comorbidities with sex, smoking and working status ($P > 0.05$).

As regard RA severity index total score analyses, we found that RARBIS total score was high statistically increased in RA patients with comorbidities compared to those without comorbidities (**Table 3**).

Table 3:-Relationship between socio-demographic and severity index score of the studied RA patients and comorbidity:

Variables	RA patients with comorbidity (n=710)	RA patients without comorbidity (n=432)	Test	p
Age (years)				
<50 years	187 (26.3)	215 (49.8)	64.6	0.000*
≥50 years	523 (73.7)	217 (50.2)		
Mean± SD	54.33±8.77	35.22±12.43	30.38	0.000*
Gender				
Female	568 (80.0)	341 (78.9)	0.19	0.665
Male	142 (20.0)	91 (21.1)		
Residence				
Rural	660 (93.0)	315 (72.9)	86.4	0.000*
Urban	50 (7.0)	117 (27.1)		
Education				
Illiterate	588 (82.8)	147 (34.0)	279.0	0.000*
Literate	122 (17.2)	285 (66.0)		
Current Smokers	57 (8.0)	31 (7.2)	0.274	0.600
Working status				
Non-working	588 (82.8)	352 (81.5)	0.329	0.566

Working	122 (17.2)	80 (18.5)		
Disease duration (years):				
<5 years	22(3.1)	157 (36.3)		
≥5 – <10 years	353(49.7)	263(60.9)	393.0	0.000*
≥10 years	342(48.2)	5(1.2)		
Mean ± SD	12.33±4.26	7.54±2.37	21.44	0.000*
RA-related medications				
NSAIDs	590 (83.1)	188 (43.5)	194.0	0.000*
Prednisolone	645 (90.8)	350 (81.0)	23.1	0.000*
RA severity index total score (RARBIS)				
	9.54±3.01	5.79±2.38	22.04	0.000*

* Statistically highly significant.

Discussion:-

Our study demonstrated that the most patients with severe RA suffer from a number of associated comorbidities. These comorbid conditions may impact treatment regimens of RA, or the prescribed drugs may worsen the comorbidity. Therefore, to successfully manage RA, comorbidities should be carefully considered and they should be treated in addition to prescribing RA regimens medication.

Similar to previous studies like **Ikuyama et al.¹¹**, our study showed that more women (79.6%) than men (20.4%) suffer from RA. Also, in our study, the average patient age was 43.86 ± 11.4 years, which is similar to the average age of patients with RA in **Sineglazova et al. study¹²**.

Personal characteristics of Egyptian patients showed lower educational level and smoking status with common non-working status, these findings are may be related to cultural and economic background¹³. Which would have as a consequence poor compliance to treatment, and therefore, poor control of the inflammatory activity of RA. In addition, biologic therapy is expensive and not covered or reimbursed governmentally or routinely by insurance. So, As regarding treatment, more than half of patients (68.1%) were on NSAIDs, and (87.1%) on prednisolone, which were comparable with that from a study on established RA study done by **Osiri & Sattayasomboon⁴**. The rate of prednisolone used in our patients was 87.1%, which was comparable with that from **Briggs et al. study¹⁴** and **Al-Bishri et al. study¹⁵**.

RA severity is considered complex measure determined by objective components such as disease activity (for example, tender/swollen joints and acute-phase reactants) and physical damage (for example, radiologic damage, and functional disability) and subjective components (for example, global health assessment, pain, grip strength, fatigue, and costs). The RARBIS does not require a new physician's assessment and the information on indicators can be routinely collected from typical medical records¹⁶.

To the best of our knowledge, this is the first study discuss the RARBIS among Egyptian RA patients. We found that the total score ranged from 3-16 with a mean of 8.09 ± 2.41 and this relatively consider high score, although our patients haven't large percent of extra articular manifestations like vasculitis (4.5%) or pulmonary nodules (0.9%) or cervical subluxation (1.8%), but our patient have higher disease activity clinically and laboratory with more flares 2-4 (38.7%), with 5 arthritis flare (56.7%), high inflammatory markers as ESR and CRP (78.5%) high RF (53.3%), high DMARDs usage like; hydroxychloroquine, gold, sulfasalazine (70.4%) & methotrexate, leflunomide (74.7%) and this uncontrolled disease may be due to educational and sociocultural aspects, which would have as a consequence poor compliance to treatment, and therefore, poor control of the inflammatory activity of RA, as well as a lower use of biological agents; drugs that show an important ability to induce remission in RA; due to the economic problems in our country.

We studied also the comorbid conditions associated with our RA patients and found that 62.2% patients had at least one comorbidity. These results were consistent with several results where study done by **Al-Bishri et al.¹⁵** found that (66.0%) patients of the 340 investigated were found to suffer from at least one comorbidity. In addition, **Osiri & Sattayasomboon⁴** study revealed that the prevalence comorbidities in RA patients ranged from 54.0 to 56.8%. The higher frequencies of comorbidities were noticed for osteoporosis (39.1%) which is similar to the study

done by **Briggset al. study**¹⁴ which revealed that the frequency of osteoporosis in the studied RA was (31.0%) and this match can reflect that the use of steroid is a major factor in susceptibility to osteoporosis in some RA patients.

As regarding eye problems frequencies (36.5%), our study was in agreement with the study done by **Osiri&Sattayasomboon**⁴, they found that the frequency of eye conditions (34.6%) (KCS, cataract, glaucoma).

Also, our results matched with the study done by **Al-Bishriet al.**¹⁵ among Saudi RA patients, we found that (36%) of RA patients had hypertension in comparison of the studied RA (32.2%). Further, it seems that Egyptian patients had also highest ever prevalence of gastric ulcers (27.2%) which is similar to **El-Zorkany et al**¹³ which revealed the prevalence of gastric ulcer (22.0%) as compared to prevalence in all other participating countries (10.0%). So, the management strategy which concentrates on the use of available NSAIDs, steroids, and synthetic DMARDs would be responsible¹⁷.

We found (22.4%) of studied RA patients had psychiatric problems mainly depression, this finding consistent with previous studies in a cohort study of more than 33000 patients with RA, depressive symptoms were reported by 11.7% of patients. There was association RA and depression and the coexistence of depression and RA is associated with increased pain, fatigue, reduced health-related quality of life, increased physical disability and healthcare costs¹⁸. And also matched the meta-analysis that included 13.189 patients with RA from 72 studies, the prevalence of a major depressive disorder was found to be 16.8 %¹⁹.

In this study we observed that the frequency of diabetes mellitus was (20.1%) which were in agreement with the study done by **Espiño-Lorenzo et al.**²⁰ and revealed that (27.0%) of RA patients were diabetic.

Our results not matched with the study done by **El-Zorkany et al**¹³ which was done on 308 Egyptian patients with RA and found that RA with hepatitis viral infection were (7.1%) but in our study (17.6%) had hepatitis viral infection and these differences may be related to different sample size, and residency and sociocultural aspect.

We observed (15.1%) of the RA patients had abnormal serum levels of lipids and this finding consistent with previous study done by **Al-Bishri et al.**¹⁵ who found that (19.4%) of RA patients had dyslipidemia.

While; cancer, chronic renal disease, and coronary heart disease, represented lower frequencies (1.1%, 2.6%, and 3.7%; respectively). Moreover, these finding matched **El-Zorkany et al.**¹³ study as they found that cancer, chronic renal disease, and coronary heart disease, represented lower frequencies (0.3%, 1.3%, and 3.2%; respectively).

In our study RA patients with comorbid conditions were significantly older and had longer duration of DMARDs treatment than those with-out comorbid diseases. Rural area residency and Illiteracy were significantly associated with comorbidity among RA patients. As regarding treatment, patients with prescribed NSAIDs, or prednisolone showed significantly higher prevalent comorbidities ($p < 0.0001$). And when we comparing RA severity index, we found highly statistically significant differences between RA patients with comorbidity than RA patients without comorbidities

Our study was in agreement with the study done by **Osiri&Sattayasomboon**⁴ they found that RA patients with comorbid conditions were significantly older and had longer duration of DMARDs treatment than those without comorbidity.

Moreover, **Espiño-Lorenzo et al.**²⁰ observed that age is the main predictive factor of comorbidity in these RA patients. They identified four factors associated with comorbidity in the univariate analysis, two related to age and the rest related to the severity of RA, however, age was the only predictive factor for both comorbidity and multi-morbidity in the multivariate analyses. Because many health problems are known to increase with age, this demographic trend may lead to an increase in the absolute number of chronic conditions in RA patients. For this reason, age always appears as the strongest predictor of comorbidity in addition to pre-existing comorbidity in RA patients.

In study done by **El-Zorkany et al.**¹³, they observed that the degree of RA disease activity found among Egyptian patients (highest ever among different COMORApopulation cohort) might reflect that the reason for patients visiting the rheumatologist is a flare of disease activity. In some countries, patients are evaluated routinely even when their

RA is under good control. So, uncontrolled RA disease contributes to the increased risk of comorbidity in RA patients.

In addition, **Navarro-Cano et al.²¹**, concluded that both comorbidity and RA severity scales displayed strong bivariate associations with the probability of survival, and these associations were statistically significant.

Conclusion:-

This is the first time to apply the severity index for RA among Egyptian RA patients and we found that the Egyptian patients have relatively high scores of severity but may not reflect the actual severity nature of the disease as they have the higher score in the clinical activity subscale and this continuously active disease may be due to socioeconomic problems in our country. We found also high percent of comorbid conditions associated with the disease with significant relationship with the severity index.

Limitation of our study is the retrospective nature, since some data may be overlooked; additionally, data regarding the impact of comorbidities on disease activity and quality of health were not available. New researches should focus on the complex interdependencies between RA severity and comorbidity. The RARBIS may have great utility for researchers using medical record.

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Abbreviations:-

RA: Rheumatoid Arthritis

RARBIS: Rheumatoid arthritis medical records-based index of severity index

NSAID:Non-steroidal anti-inflammatory drug

ESR: Erythrocyte sedimentation rate

CRP: C-reactive protein

RF: Rheumatoid factor

DMARDS :Disease-modifying Anti-rheumatic Drugs

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RESEARCH ARTICLE

THERMO ACOUSTIC STUDY OF BINARY LIQUID MIXTURES OF DIMETHYL SULFOXIDE AND TRIETHYLAMINE BY ULTRASONIC INTERFEROMETRIC TECHNIQUES.

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Abstract

The ultrasonic velocity, density and viscosity have been measured in the binary systems of dimethyl sulfoxide (DMSO) with triethylamine (TEA). From the experimental data, various acoustical parameters such as adiabatic compressibility (β), intermolecular free length (L_f), free volume (V_f), internal pressure (π_i), Relaxation time (τ), Acoustic impedance (Z), Gibb's free energy (ΔG) and Vander Waal's constant (b) were calculated. The results are interpreted in terms of molecular interaction between the components of the mixtures.

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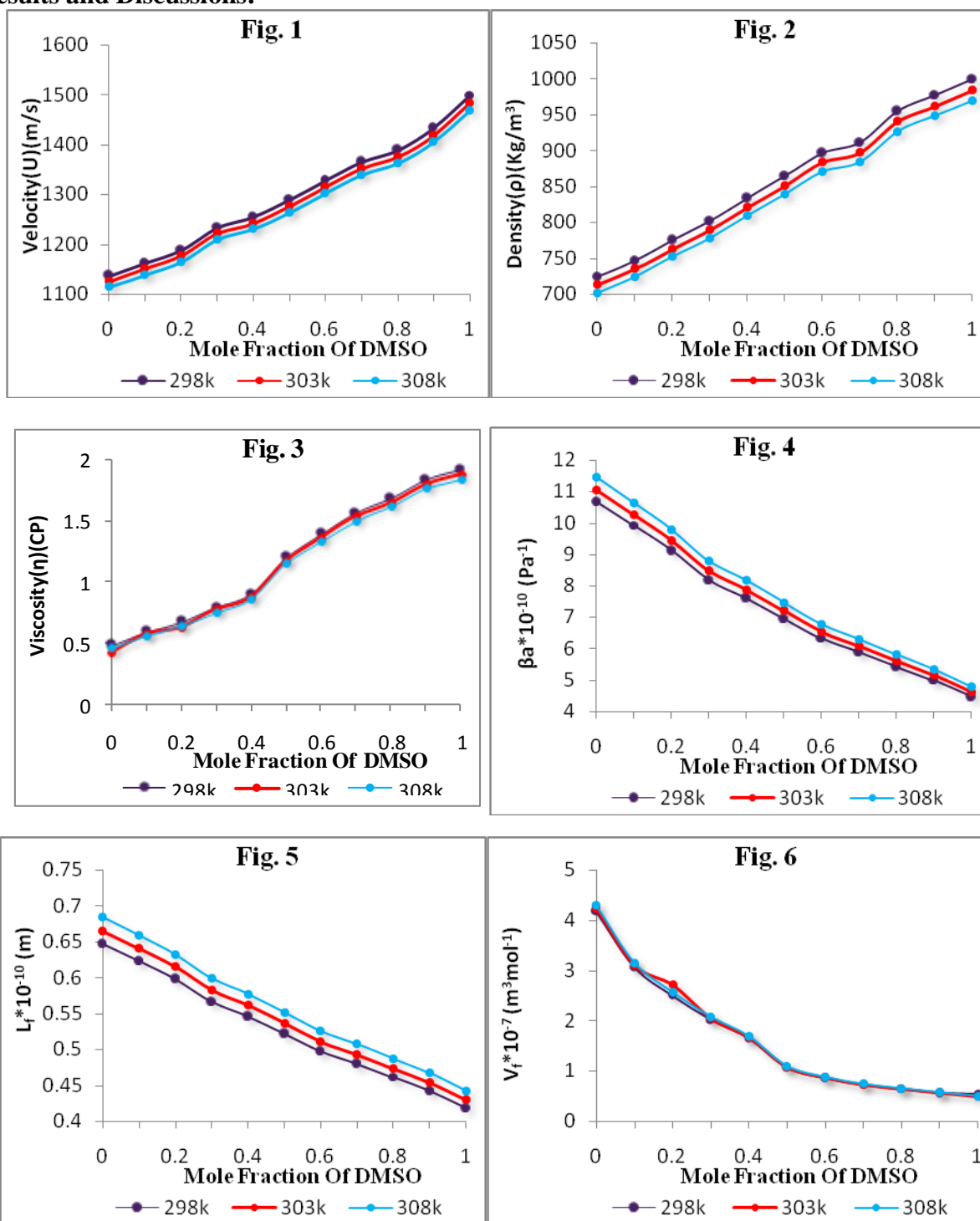
Introduction:-

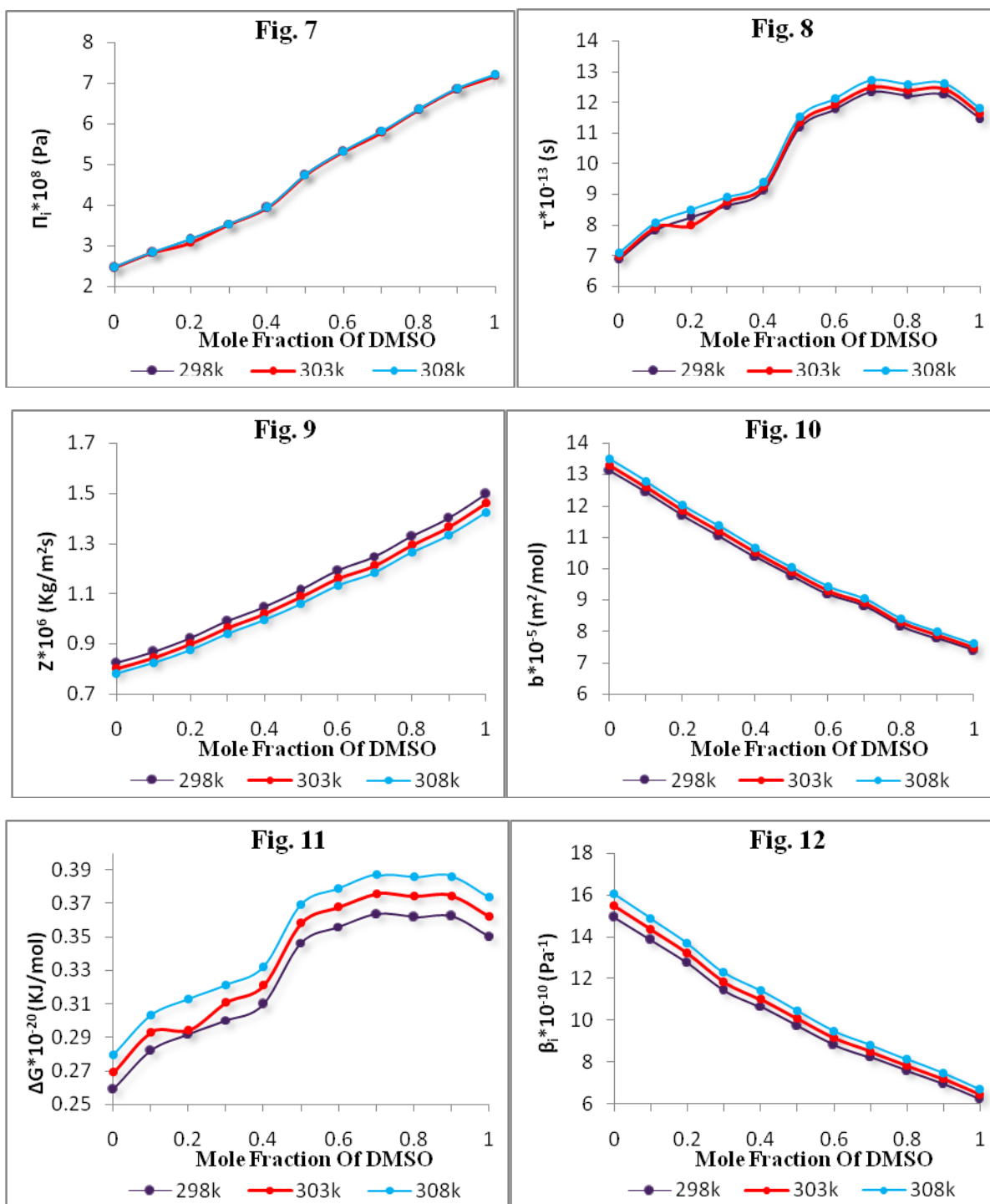
In recent years, ultrasonic technique has become a powerful tool for studying the molecular behavior of liquid mixtures [1-3]. The measurements of ultrasonic velocity have been adequately employed in understanding the molecular interactions in liquid mixtures. Molecular interaction studies can be carried out by both spectroscopic [4-5] and non-spectroscopic [6-7] techniques. A large number of studies have been made on the molecular interaction in liquid systems by various physical methods like, Raman Effect, Nuclear Magnetic Resonance, Ultra Violet and ultrasonic method [8-10]. However, ultrasonic velocity and viscosity measurements have been widely used in the field of interactions and structural aspect evaluation studies. In recent years ultrasonic technique has become powerful tool in providing information regarding the molecular behavior of liquids and solids, owing to its ability of characterizing Physico-chemical behavior of the medium. The ultrasonic velocity data for binary and ternary liquid mixtures have been used for by many researchers [11-14].

In order to have a clear understanding of the intermolecular interactions between the component molecules, the authors have performed thorough study on the liquid mixtures using ultrasonic velocity data. The present work deals with the ultrasonic velocity and computation of related parameters in binary system of dimethyl sulfoxide + triethylamine at 298.15K, 303.15K and 308.15K at frequency 7MHz. For preparing various concentration mixtures, mole fractions of component liquids were varied from 0.0 to 1.0. Dimethyl sulfoxide, as a polar solvent, is certainly too some extent associated by dipole-dipole interactions, and is of particular interest because of the absence of any significant structural effects due to the lack of hydrogen bonds; therefore, it may work as an aprotic, protophilic solvent with a large dipole moment and high dielectric constant.

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Results and Discussions:-



The variation of density, viscosity and ultrasonic velocity Vs mole fraction for the binary system dimethyl sulfoxide +triethylamine at 298.15K, 303.15K and 308.15K are shown in **fig.-1, 2 and 3 respectively**. While other thermodynamics parameters such as adiabatic compressibility (β_a), free length (L_f) free volume (V_f) internal pressure (π_i), relaxation time (τ), and acoustic impedance (Z), Gibb's free energy (ΔG), Vander Waal's constant (b), Isothermal compressibility(β_i) at temperature 303K are shown in **fig.-4,5,6,7,8 9,10,11 and 12 respectively**.

Fig. 1 shows the graph of ultrasonic velocity versus concentration. It is observed that ultrasonic velocity increase with increase in the concentration of DMSO in TEA, indicating association in constituent's molecules. The association may involve due to hydrogen bonding or dipole-dipole interaction between DMSO in TEA.

Fig.2 shows the graph of density versus concentration. It is observed that density increases with increase in concentration of DMSO in TEA. Increase in density decrease in volume, indicating association in component molecules. The density of binary liquid mixtures may be increase due to structural reorganization indicating the closed packed structure of the molecules cluster. This makes the liquid medium less compressive. Increasing temperature of the mixture decreases its density (ρ). The decrease in density (ρ) with temperature indicates decrease in cohesive force. Thus increase of temperature favors increase of kinetic energy and volume expansion and hence decrease of density.

Fig. 3 consists of viscosity (η) versus concentration. It is observed that viscosity increases with increase in the concentration of DMSO in TEA, indicating strong molecular interaction. The viscosity gives the strength of molecular interaction between interacting molecules. Increase in temperature of the mixture, increasing disorders of the medium and hence entropy increases. As entropy increases, viscosity of binary mixture decreases.

Free Length, Increase in concentration of DMSO thus results in decrease in adiabatic compressibility (β_a) and free length (L_f) as shown in **Fig. 3 and Fig 4**. The regular fall in free length with increase in concentration of DMSO causes a rise in sound velocity [16]. This trend is an indication of clustering together of the molecules as the associative effect of the polar group dominates over the other type of interaction [17]. Velocity shows a reverse trend as temperature is increased. This happens as the spacing between the molecules increases leading to a less ordered structure.

The adiabatic compressibility (β_a) and Isothermal compressibility (β_i) exhibit similar trend. From **Fig.4 and 12**, it is observed that adiabatic compressibility (β_a) and isothermal compressibility (β_i) decreases with increase in molar concentration of DMSO in TEA indicating strong intermolecular interaction in the component molecules in binary mixtures shows associating tendency of the component molecules.

Fig. 6, it is observed that free volume decreases (V_f) with increase in mole concentration of DMSO in TEA, indicating association in the molecules of the component liquids. It was observed that the variation in the free volume values show exactly the reverse trend as that of internal pressure as shown in **Fig. 7**, gradually increases indicating weak molecular interaction amongst DMSO in TEA [18]. In the present paper, it is observed that free volume decreases and internal pressure increases.

Increase in internal pressure (Π_i) may be due to strengthening of cohesive force. Since the interaction in our case is weak, ' Π_i ' increases slowly with concentration of DMSO, but decreases rapidly with increase of temperature.

Relaxation time (τ) increases with increase in mole fraction of DMSO as presented in **Fig. 8**, but increases as temperature increases. The former indicates weak molecular interaction between the components of the mixture, and the latter is true because of instantaneous conversion of excitation energy to translational energy. . The relaxation time which is in the order of 10^{-13} sec is due to structural relaxation process [19] and in such a situation, it is suggested that, the molecules get rearranged due to co-operative process [20]. Relaxation time is the time taken for the excitation energy to appear as translational energy and it depends on temperature and impurities.

Acoustic impedance (Z) is the ratio of the effective sound pressure at a point to the effective particular velocity at that point. The pressure is measured by the totality of the force of dispersion, repulsion, ionic and dipolar. In our present investigation, **From Fig. 9**, acoustic impedance increases with increase in concentration of DMSO, showing weak molecular interaction. Since $Z = U\rho$, as temperature increases, Z decreases. Acoustic impedance is also used for assessing the absorption of sound in the medium.

Fig. 10 represents the Vander Waal's Constant (b). Vander Waal's constant (b) is also known as the co-volume in in Vander Waal's equation and it varies in similar way to that of the available volume. The co-volume decrease with increasing concentration of DMSO in TEA indicating strong interaction between the component molecules. Also the linear variation of these acoustical properties indicating absence of complex formation in the mixture. These

parameters help for the study of range of Repulsive [21] and attractive forces in the molecules of the components. The change in Vander Waal's Constant (b) would be due to a change in intermolecular geometry (micro geometry).

Gibb's free energy measures mobility of the medium. Higher the mobility of the medium, higher will be the entropy; lower will be the free energy. **Fig. 11** represents the variation of Gibb's free energy with concentration. It is observed that Gibb's free energy increase with increase in concentration DMSO in TEA, indicating decrease in mobility of the mixture hence disorder decreases. This leads to the low entropy and hence salvation increase. This is because of the fact that salvation is directly proportional to size of the molecule.

Conclusions:-

Density, viscosity and ultrasonic velocity increases with increase in mole fraction of dimethyl sulfoxide in triethylamine. This is due to the dipole - dipole interaction between dimethyl sulfoxide and triethylamine components. The decrease in adiabatic compressibility and free length for the system- dimethyl sulfoxide in triethylamine is due to strong association. The dipole-dipole interactions between dimethyl sulfoxide and triethylamine are found to be responsible for association in the binary liquid mixture.

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RESEARCH ARTICLE

CORROSION INHIBITION OF MILD STEEL USING *ALOE BARBEDENSIS MILLER* SKIN EXTRACT IN 0.5 MHCL.

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Manuscript Info

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Mild Steel Aloe barbedensis
Miller, EDX, EIS

Abstract

Mild steel is employed widely in most industries due to its low cost and availability in ease for the fabrication of various reaction vessels such as cooling tower tanks, pipelines etc. It can be subjected to aggressive and unfavorable environmental conditions, making it susceptible to corrosion. From the economic and environmental view, plant extracts are an excellent alternative as inhibitors because of their availability, biodegradability, non toxicity and environmental friendly. The rich source of anthraquinone and other chemical composition of Aloe Vera has been resulted a wide range uses in different sector. The present study deals with the inhibition effect of biodegradable, nontoxic and eco-friendly. Aloe Vera leaf extract on corrosion of mild steel in HCl. The corrosion inhibition of mild steel in the presence of *Aloe Barbedensis Miller* in 0.5 MHCl was studied by weight loss measurement; electrochemical techniques (Potentiodynamic polarization, AC Impedance). The maximum efficiency was found to be 81% at 150 ppm. From the result of weight loss studies, the mode of adsorption is found to be physisorption. Kinetic and thermodynamic parameters were calculated and discussed. Potentiodynamic polarization studies indicate that *Aloe Barbedensis Miller* acts as a mixed type of inhibitor. The surface morphology has been analyzed.

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Introduction:-

Corrosion is the destruction of material resulting from an exposure and interaction with the environment. It is a major problem that must be confronted for safety, environmental and economic reasons in various chemicals, mechanical, metallurgical, biomedical and medical engineering applications and more specifically in the design of a much more varied number of mechanical parts. Several efforts have been made using corrosion preventive practices and the use of green corrosion inhibitors is one of them. Natural products have been studied extensively as corrosion inhibitors both in product mixtures extracted from natural sources such as plants or essentially pure products derived from animals or plants (i.e., vitamins and amino acids)[1]. From the economic and environmental view points, plant extracts are an excellent alternative as inhibitors because of their availability and biodegradability. These extracts contain a variety of natural products such as essential oils, tannins, pigments, steroids, terpenes, flavones and flavonoids, among other well-known active substances used as CIs. In general, these compounds contain conjugated aromatic structures, long aliphatic chains such as nitrogen, sulfur, and oxygen heteroatoms with free electron pairs that are available to form bonds with the metal surface; in most cases, they act synergistically to exhibit good efficiency regarding the corrosion protection. Aloe Vera is an important medicinal plant which

belongs to the family of Liliacea. A more recent review concludes that the cumulative evidence supports the use of Aloe barbadensis miller for the healing of first to second degree burns. Aloe Vera plant is used for corrosion protection of metal in acidic medium since it was found for environmental eco-friendly and available in affordable price. The Aloe Vera leaves contain several free anthraquinones and phenolic compounds that aid in absorptive processes in metal surface. The rich source of anthraquinone and other chemical composition of Aloe Vera has been resulted a wide range uses in different sector. In most of the studies related to Aloe Vera gel alone was taken and the outer part of the skin was considered as a waste material. In the present study the outer part of Aloe Vera alone was taken to investigate the inhibitive effect for the corrosion of mild steel in 0.5M HCl.

Materials and Methods:-

Preparation of Mild Steel Specimens:-

The mild steel samples were obtained from a locally available industrial Fe-C steel with very low concentration of carbon. A large sheet of cold rolled mild steel coupons have the following chemical composition 0.026% Si, 0.006% P, 0.4% Mn, 0.1% C, 99.4% Fe. The mild steel samples, with an active surface of $1.2 \times 4.0 \times 0.3$ cm were used for Mass loss measurements and $1\text{cm} \times 1\text{cm}$ specimen for electrochemical measurements. The mild steel samples were mechanically polished, washed in double distilled water and degreased with acetone and used for the weight loss method and surface examination studies.

Weight – loss method:-

Determination of Corrosion Rate:-

Weight loss measurements were carried out using an ACCULAB Electronic balance with readability/sensitivity of 0.1 mg in 210g range. The specimens were immersed in beaker containing 100ml acid solutions without and with different concentration of Aloe Barbedensis Miller leaves extract using hooks. At the end of exposure period, specimens were cleaned according to ASTM G-81 and the weight recorded. The average mass loss of two parallel mild steel specimens was obtained. The test specimens were removed and then washed with de-ionised water, dried and reweighed [2]. From the change in weight of specimens the corrosion rate was calculated using the following relationship,

$$\text{Corrosion Rate} = 87.6 \times W / A \times T \times D (\text{mpy})$$

$$\text{Corrosion Inhibition Efficiency (IE)} = 100[1 - (W_2/W_1)] \%$$

Electrochemical Impedance Spectroscopy:-

Electrochemical measurements were run using a Potentiostat/galvanostat (Electrochemical system Model Vertex.100mA.D) and a personal computer was used. IVIUM software was used for Electrochemical Impedance Spectroscopy (EIS) and potentiodynamic polarization (PDP) analysis. The EIS measurements were carried out using AC signals of amplitude 10 mV peaks-to-peaks at the open circuit potential in the frequency range of 10 MHz to 1 Hz. The charge transfer Resistance (R_{ct}) values have been calculated from the difference in the impedance at low and high frequencies. The capacitances of the double layer (C_{dl}) values are estimated from the frequency (f) at which the imaginary component of the impedance ($-Z''$) is maximum and the double layer capacitance (C_{dl}) was calculated by using following equation:

$$\text{Obtained from the equation: } C_{dl} = \frac{1}{2} \times 3.14 \times R_{ct} \times f_{\max}$$

Potentiodynamic Polarization:-

After impedance spectrum was obtained, the potentiodynamic current potential curves was recorded immediately by changing the electrode potential automatically taken from OCP value with scan rate of 5 mV / S [3]. Tafel lines extrapolation method was used for detecting I_{corr} and E_{corr} values for the studied systems.

Scanning Electron Microscope and Energy Dispersive Spectroscopy Analysis:-

For surface morphological study of the uninhibited and inhibited mild steel samples were sent to SEM and EDX analysis [4][5].

Results and Discussion:-

Mass Loss Measurements:-

Effect of Inhibitor Concentration:-

Inhibition efficiency of mild steel with different concentration of *Aloe Barbadensis Miller* extract in 0.5M HCl at room temperature are presented in Table 1. From the table, it is clear that the corrosion rate decreases with an increase in inhibitor concentration, i.e. the corrosion inhibition enhances with the inhibitor concentration. This behavior is due to the fact that the adsorption and coverage of the inhibitor on the mild steel surface increase with the inhibitor concentration. Maximum inhibition efficiency of 81% was shown at 150ppm in 0.5M HCl at 3 hour immersion, period beyond 200 ppm the system remains constant. The high inhibitive performance of *Aloe Barbadensis Miller* suggests a higher bonding ability of inhibitor on mild steel surface[6].

Table 1:- Corrosion rate of mild steel in 0.5M HCl.

S.no	Concentration(ppm)	Corrosion rate(mpy)	Surface coverage(θ)	% I.E
1	Blank	18	-	-
2	50	6	0.66	66
3	100	5	0.72	72
4	150	3.4	0.81	81
5	200	3.7	0.79	79
6	250	3.6	0.80	80
7	300	3.6	0.80	80

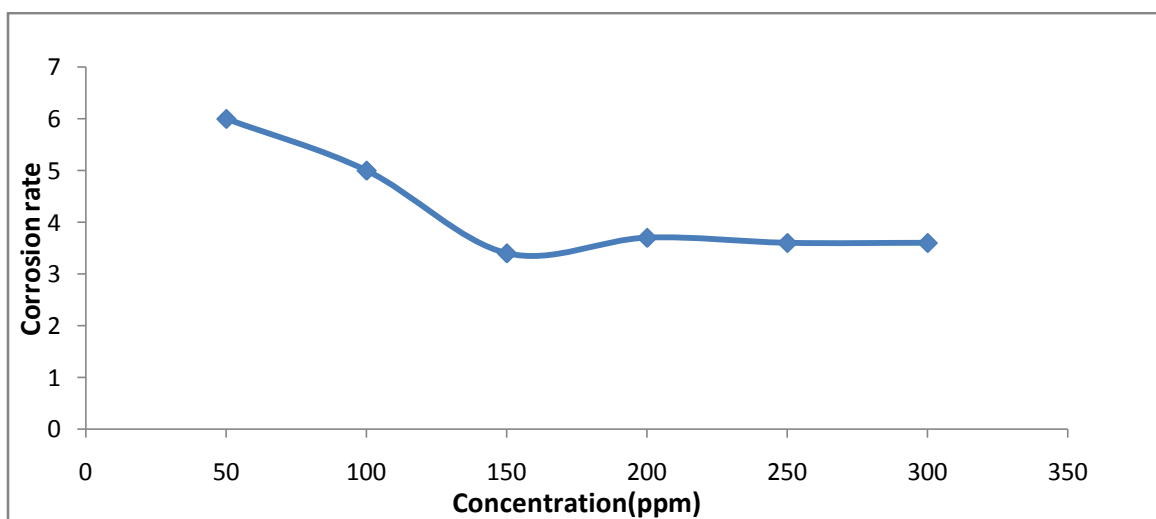


Fig 1:- Corrosion rate of *Aloe Barbadensis miller* at different concentration.

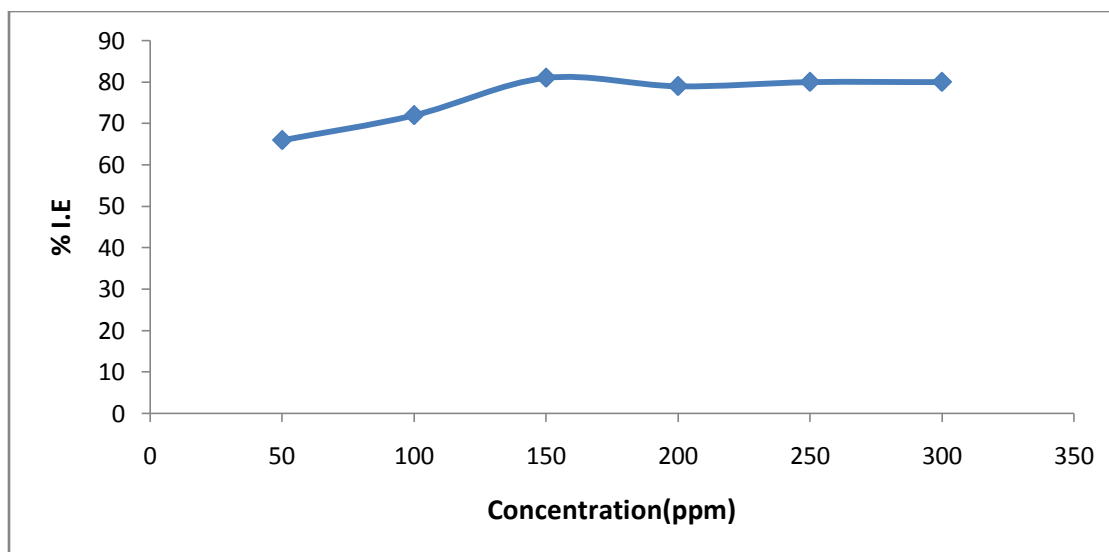


Fig 2:- Inhibition efficiency of *Aloe Barbadensis miller* at different concentration.

Effect of temperature:-

To show the effect of temperature on inhibition efficiency of Aloe Barbedensis Miller, Weight loss experiment was performed in the temperature range of 308 to 328 K at optimum concentration. The variation of inhibition efficiency with temperature at optimum concentration are listed in Table 2. From table 2, it is clear that corrosion rate is temperature dependent and increases with increase in temperature [7], [8].

Table 2:- Effect of temperature on the CR of mild in 0.5 M HCl.

S.no	Temperature (K)	Corrosion rate(Blank)	Corrosion rate(Inhibitor)	Surface coverage	% I.E
1	308	22	5.3	0.76	76
2	313	31	9.1	0.71	71
3	318	41	10.8	0.73	73
4	323	50	15	0.70	70
5	328	75	26	0.65	65

Kinetic and Thermodynamic parameter for corrosion rate:-

The apparent activation energy (E_a), the enthalpy of activation (ΔH^*) and entropy of activation (ΔS^*) for the corrosion of mild steel in 0.5M HCl solution with and without inhibitor at different temperatures are calculated from Arrhenius equation:

$$CR = A \exp(-E_a/RT) \quad (1)$$

And from Transition state equation:

$$CR = RT/h \exp(\Delta S^*/R) \exp(-\Delta H^*/RT) \quad (2)$$

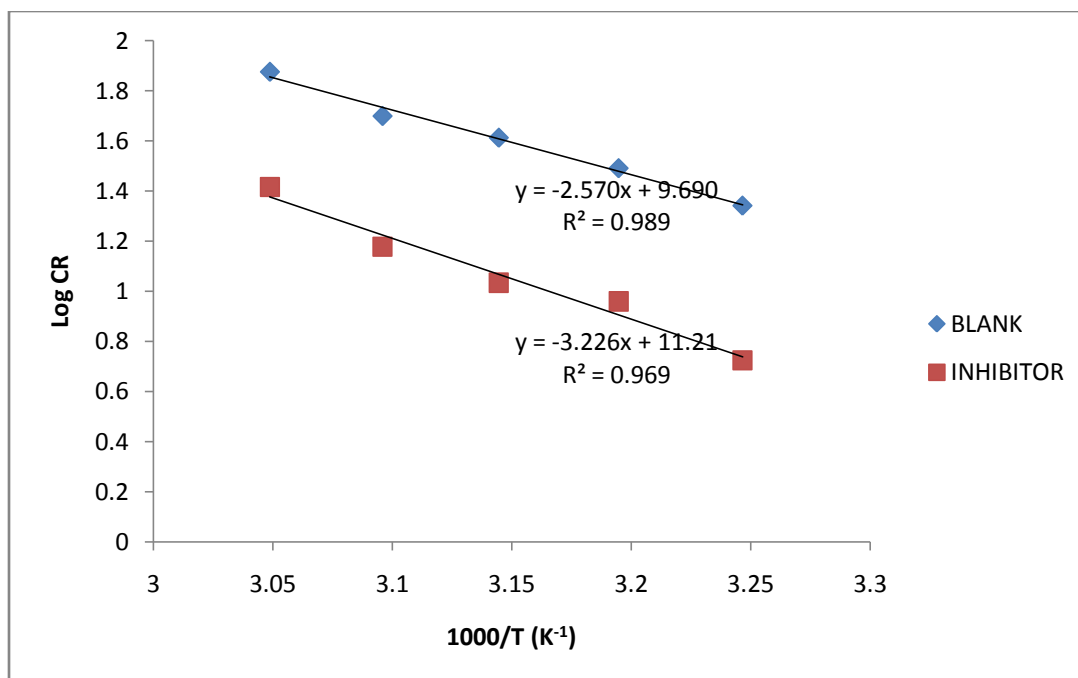


Figure 3:- Arrhenius plots of log CR Vs $1000/T(K^{-1})$

The Arrhenius plot for mild steel immersed in 0.5M HCl with optimum concentration and blank solution is depicted in fig.3. The Activation energy (E_a) is obtained from the slope ($-E_a/2.303RT$) and the values are listed in table 3. It is evident from the increase in activation energy value in the presence of inhibitor that the reaction undergoes through different mechanism which has a high E_a value compared to uninhibited system. Hence the rate of corrosion is decreases, as less reactants have that activation energy to reach the activation state.

Table 3:- Thermodynamic parameters for Mild steel in 0.5M HCl in the absence and presence of optimum concentration of *Aloe Barbadensis Miller* leaves.

	E_a (KJmol ⁻¹)	ΔH^\ddagger	ΔS^\ddagger
Blank	49.21	46.5	-68.18
Inhibitor	61.7	59.12	-39.07

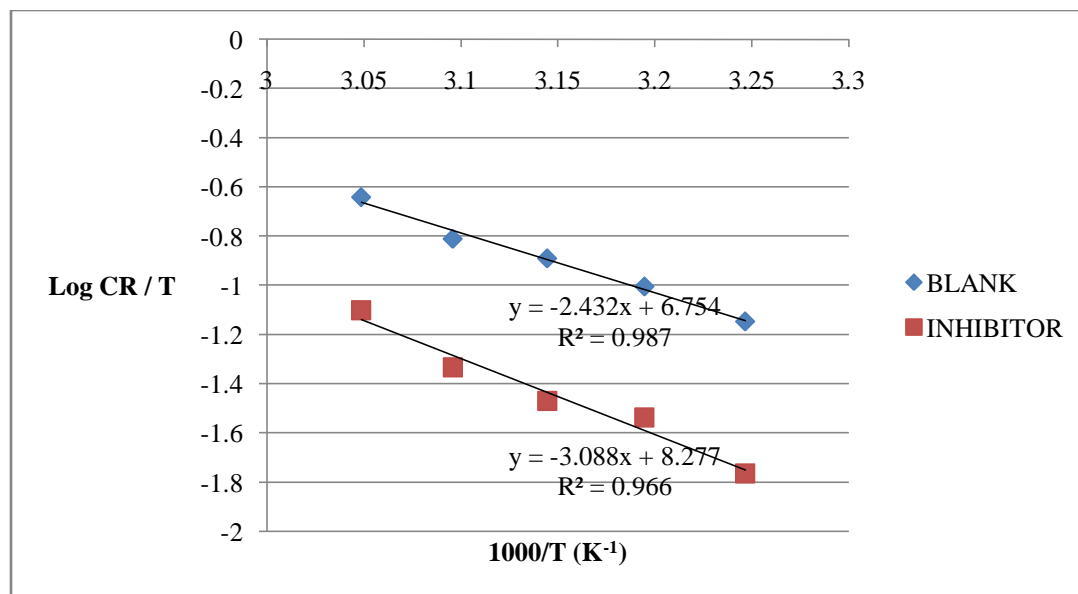


Figure 4:- Transition state plots of Log CR/T Vs $1000/T(K^{-1})$.

A plot of $\log (CR/T)$ Versus $1000/T$ is shown in fig.4. Straight lines are obtained with slope $(-\Delta H^*/2.303R)$ and intercept of $[\log(R/Nh) + (\Delta S^*/R)]$. The positive value of enthalpy of activation (ΔH^*) in the absence and presence of inhibitor at different temperatures reflects the endothermic nature of mild steel dissolution process, which indicates that the dissolution of steel is difficult. It is evident from the table that the value of ΔH^* increases in the presence of the inhibitor than in the absence. This shows the higher protection efficiency of the inhibitor. This may be attributed to the presence of high energy barrier for the reaction and hence there is rise in enthalpy of the corrosion process.

From Table 3, it is clear that there is an increase in the entropy of activation in the presence of inhibitor system compared to its absence. The large negative value of entropy of activation in the absence of inhibitor shows that it has more ordered arrangement in the transition state whereas in the presence of inhibitor system the entropy of activation becomes less negative showing that the surface of the metal is covered with inhibitor molecules thus moving the system towards less ordered arrangement.

Adsorption Parameter:-

The primary step in the action of inhibitors in solution is generally agreed to be adsorption on the metal surface. This involves the assumption that the corrosion reactions are prevented from occurring over the area (or active sites) of the metal or alloy surface covered by adsorbed inhibitor species, whereas these corrosion reaction occurred normally on the inhibitor free area. Accordingly, the fraction of surface covered with inhibitor species ($\theta = \%IE/100$) can be followed as a function of inhibitor concentration and solution temperature. The surface coverage (θ) data are very useful while discussing the adsorption characteristics. When the fraction of surface covered is determined as a function of the concentration at constant temperature, adsorption isotherm could be evaluated at equilibrium condition. The dependence of the fraction of the surface covered θ on the concentration C of the inhibitor was tested graphically by fitting it to Langmuir's isotherm, which assumes that the solid surface contains a fixed number of adsorption sites and each site holds one adsorbed species.

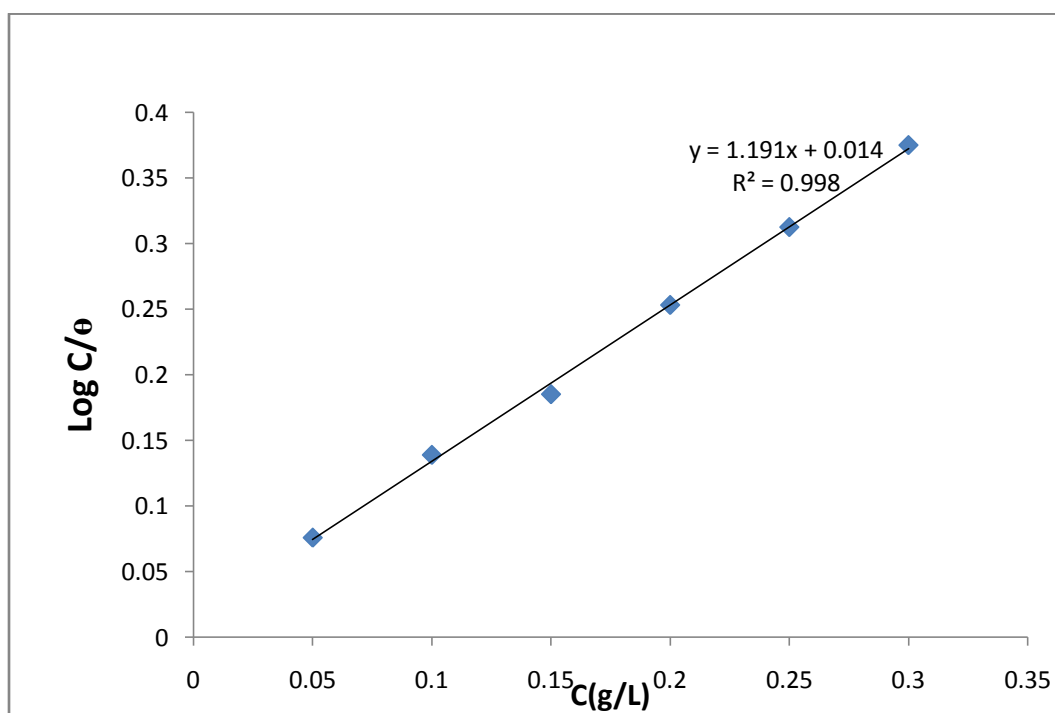


Figure 5:- Langmuir adsorption plot of $\log C/\theta$ Versus C .

Table 4:- Langmuir adsorption of *Aloe Barbadensis* Miller in 0.5M HCl on the mild steel.

Inhibitor	Intercept	K_{ads}	R^2	ΔG_{ads}° (KJ/mol)
<i>Aloe Barbadensis miller</i>	0.014	71.428	0.998	-10.75

The equilibrium constant for the adsorption process is related to the standard free energy of adsorption by the equation:

$$\Delta G_{\text{ads}}^{\circ} = -RT \ln K \quad (3)$$

The value of K can be obtained by the following equation

$$C/\theta = 1/K + C, \quad (4)$$

where C is the concentration of inhibitor (g/lit), θ is the surface coverage which is given as $\theta = \%IE/100$. A plot of C/θ VS C gives a straight line, from the intercept of the straight line the value of K can be calculated and the value is substituted in above equation and hence the $\Delta G_{\text{ads}}^{\circ}$ value is obtained. The negative value of $\Delta G_{\text{ads}}^{\circ}$ from Table 5 shows that the adsorption process is spontaneous. Normally, the magnitude of $\Delta G_{\text{ads}}^{\circ}$ around -20 KJ/mol or less negative is assumed for electrostatic interactions exist between inhibitor and the charge metal surface (i.e. physisorption). Those around -40 KJ/mol or more negative are indication of charge sharing or transferring from organic species to the metal surface to form a coordinate type of metal bond (i.e. chemisorption). The calculated $\Delta G_{\text{ads}}^{\circ}$ value is -10.75 KJ/mol which is less than -20 KJ/mol indicates that the adsorption is physical adsorption. The data are also used to study the El-Awady isotherm. The characteristic of the isotherm is given by:

$$\text{Log } (\theta/1-\theta) = \text{log } K + y \text{ log } C \quad (5)$$

Where C is the concentration of the inhibitor, θ is the degree of surface coverage, K_{ad} is the equilibrium constant of adsorption process and $K_{\text{ad}} = K^{1/y}$.

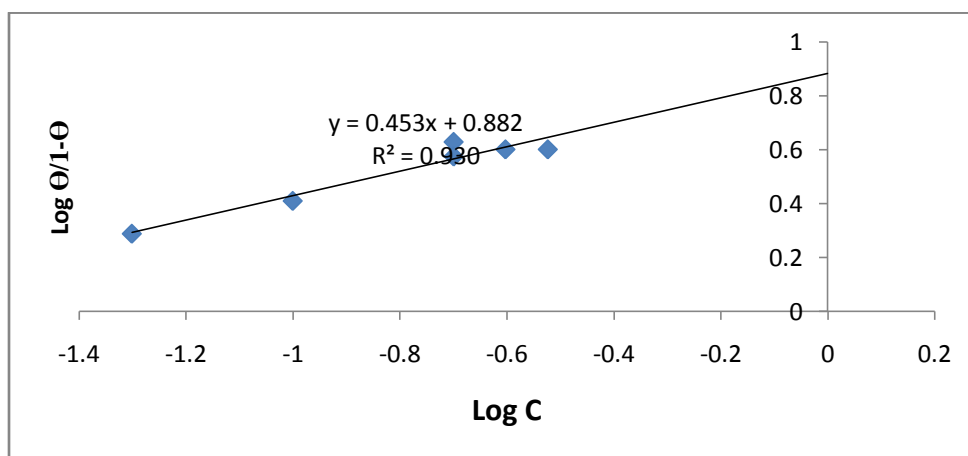


Figure 6:- El-Awady adsorption plot of $\text{Log}(\theta/1-\theta)$ Versus $\text{Log } C$.

Table 5:- El-Awady adsorption of *Aloe Barbadensis Miller* in 0.5M HCl on the mild steel.

Inhibitor	K_{ads}	$1/y$	R^2	$\Delta G_{\text{ads}}^{\circ}$ (KJ/mol)
<i>Aloe Barbadensis miller</i>	87.09	2.207	0.930	-11.25

In this model, the number of active sites y is included. Values of $1/y$ is less than one implies multilayer adsorption, while $1/y$ greater than one suggests that a given inhibitor molecule occupies more than one active site. Curve fitting of the data to the thermodynamic-kinetic model is shown in fig 6. The plot gives straight line which clearly show that the data fitted well to the isotherm. The value of $1/y$ and K_{ad} calculated from the El.Awady et.al model curve is given in table 5. It is evident from table.7 that the value of $1/y$ is greater than unity showing that the inhibitor molecule occupies more than one active site. The $\Delta G_{\text{ads}}^{\circ}$ value can be obtained by substituting the value of K_{ad} in equation.3. The negative value of $\Delta G_{\text{ads}}^{\circ}$ from Table.3 shows that the adsorption process is spontaneous. The calculated $\Delta G_{\text{ads}}^{\circ}$ value is -11.25 KJ/mol which is less than -20 KJ/mol indicates that the adsorption is physical adsorption.

SEM Analysis:-

The SEM mages were recorded to establish the interaction of inhibitor molecule with metal surface .Figure 7.represents the SEM images of (a) mild steel immersed in 0.5 M HCl, (b) mild steel immersed in the presence of Aloe Barbedensis Miller in 0.5 M HCl covered with the inhibitors. Result shows that the phytochemical constituents present in the *Aloe leaves* form a protective layer of the mild steel specimen and thereby reduce the corrosion rate.

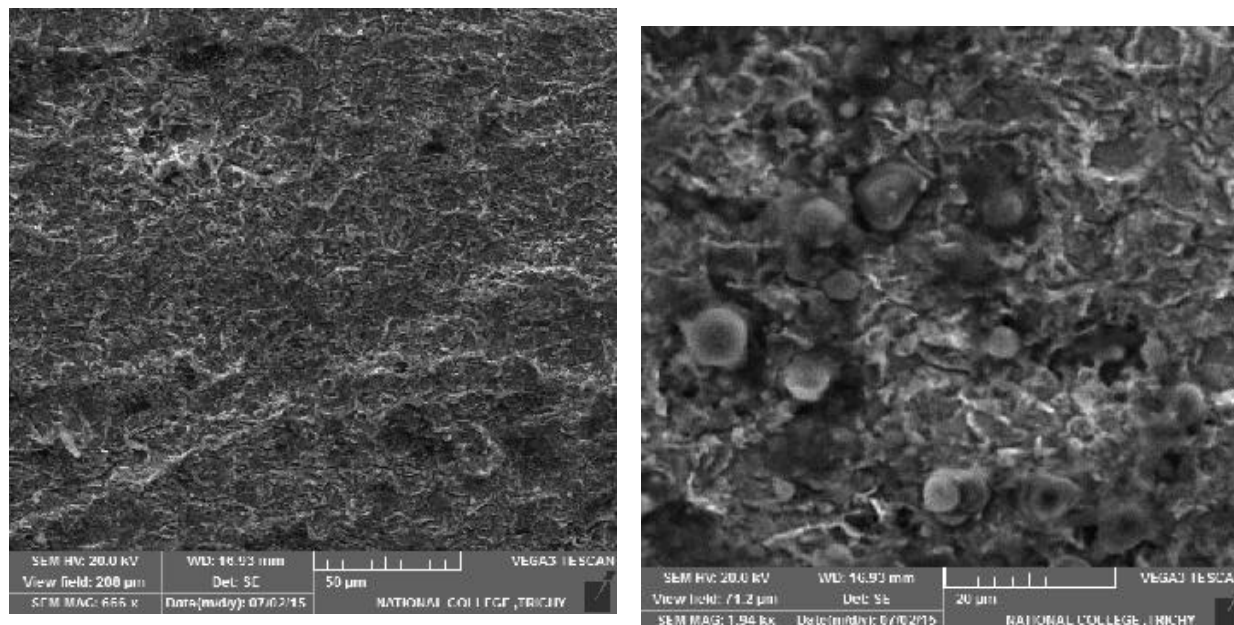


Fig.7:- SEM micrograph of (a) Mild steel immersed in acid (b) Mild steel immersed in the presence of ABMLE in 0.5M HCl).

Energy Dispersive Spectroscopy:-

Energy dispersive x-ray analysis (EDX) technique was employed in order to get information about the composition of the surface of the mild steel sample in the absence and presence of the inhibitor in 0.5 M HCl solution. The EDX spectra of uninhibited and inhibited mild steel samples are shown in fig.8.

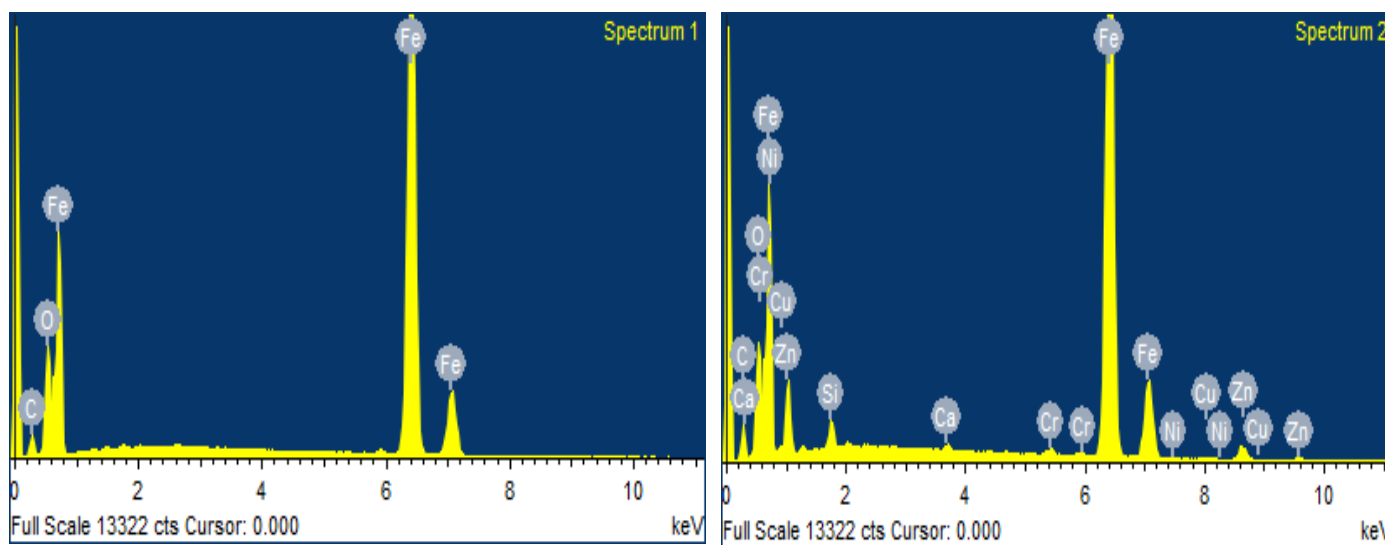


Fig.8:- EDX spectra of mild steel specimens (a) After immersion without inhibitor (b) After immersion with inhibitor.

The percentage atomic content of various elements of the uninhibited and inhibited mild steel surface determined by EDX is listed in table 6. The percentage atomic content of Fe for mild steel immersed in 0.5 M HCl solution is 79.69 % and those for mild steel dipped in an optimum concentration of Aloe Barbedensis Miller are 71.96 respectively. From figure 8, the spectra of inhibited samples show that the Fe peaks are considerably suppressed, When compared with the uninhibited mild steel sample. This suppression of Fe lines is due to inhibitory film formed on the mild steel surface.

Table 6:- .Percentage atomic contents of elements obtained from EDX spectra.

Mild steel	Fe	C	O	Si	Ca	Cr	Ni	Cu	Zn
Blank	79.69	8.46	11.85	-	-	-	-	-	-
Inhibitor	71.96	10.52	10.61	1.18	0.22	0.47	0.34	0.54	4.15

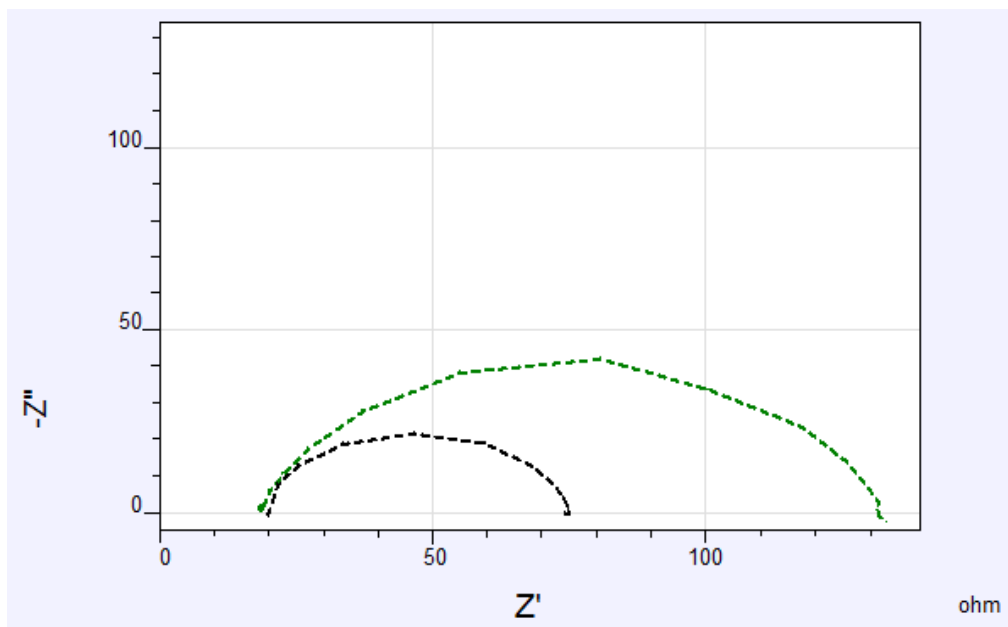
Electrochemical Impedance Spectroscopy:-

In general, as impedance diagrams for solutions examined have almost a semi circular appearance, it indicates that the corrosion of mild steel is mainly controlled by a charge transfer process. The impedance data such as R_s , R_p and C_{dl} were estimated by assuming Randles circuit. The values of charge transfer resistance (R_t) and double layer capacitance (C_{dl}) were evaluated from Nyquist and Bode plots. The values of R_t , C_{dl} and %IE derived from these investigations are given in table 7. The existence of single semicircle showed that single charge transfer process occurred during dissolution of mild steel which is unaffected by the presence of extract. It was found that addition of extract increases the value of R_t and reduces the C_{dl} value[9]. The decrease in C_{dl} value attributed to increase in thickness of electrical double layer. The increase in R_t value is attributed to the formation of protective film on the metal-solution interface.

Table 7:- The Electrochemical Impedance Parameters of Aloe Barbadensis Miller in 0.5 M HCl at optimum concentration.

System	$R_s(\Omega)$	$R_{ct}(\Omega \text{ cm}^2)$	$C_{dl}(\text{F cm}^{-2})$
Blank	20.9	31.19	6.39×10^{-5}
Inhibitor	20.31	108.3	5.780×10^{-5}

The impedance behavior of mild steel in 0.5 M HCl in the absence and presence of inhibitor at optimum concentration of Aloe Barbadensis Miller is shown as Nyquist plot in fig.10 and EIS parameter such as R_s , R_{ct} and C_{dl} were derived from the Nyquist plot are given Table.7 The ω_{max} represents the frequency at which the imaginary component reaches the maximum. It is the frequency at which the real part (Z_r) is midway between the low and high frequency X-axis intercepts. It is clear from the result that the value of R_{ct} increases from $31.19 \Omega \text{ cm}^2$ (Blank) to $108.3 \Omega \text{ cm}^2$ on the addition of 150 ppm of inhibitor. The value of C_{dl} decreases from $6.39 \times 10^{-5} \text{ F cm}^{-2}$ (Blank) to $5.780 \times 10^{-5} \text{ F cm}^{-2}$. The decrease in capacitance (C_{dl}) on the addition of the inhibitor may be due to increase in the local dielectric constant and /or may be due to increase in the thickness of the double layer, showing that *Aloe Barbadensis Miller* leaves inhibited Iron metal corrosion by adsorbing at metal/acid interfaces.

**Fig 9:-** Nyquist plot in absence and presence of optimum concentration of Aloe Barbadensis Miller.

Linear Polarization Measurement:-

The inhibition behavior of Aloe Barbadensis Miller in 0.5M HCl in the presence and absence of the inhibitor at optimum concentration are calculated by linear polarization Parameters and are given in Table.8.The efficiency found by linear polarization shows good agreement with efficiency obtained from Tafel and EIS data.

Potentiodynamic Polarization Measurements:-

The potentiodynamic polarization behavior of mild steel in 0.5 M HCl in the absence and in the presence of Aloe Barbadensis Miller is shown as Tafel plot in fig.10 .The various electrochemical potentiodynamic parameters such as corrosion potential(E_{corr}),corrosion current density (I_{corr}),anodic and cathodic slope (β_a and β_c) are calculated from Tafel plots and the values are listed in Table.8.It is seen that addition of Aloe Barbadensis Miller decreases the corrosion current (I_{corr}) density from 420.9(Blank) to 101.8 $\mu\text{A cm}^{-2}$ and shifts the E_{corr} from -426.8 to -396.8 m V SCE. Even though the slight shift of E_{corr} to less negative side makes one to think it as anodic inhibitor, but the large shift in the cathodic slope points to the control of cathodic process. Hence it is a mixed inhibitor controlling both cathodic and anodic process.

Table 8:- The potentiodynamic polarization and linear polarization parameters of Aloe Barbadensis Miller.

System	E_{corr} (m V /SCE)	I_{corr} ($\mu\text{A cm}^{-2}$)	β_a (m V /dec)	β_c	R_p
Blank	-426.8	420	987	1023	51.6
Inhibitor	-396.8	101	985	629	213.2

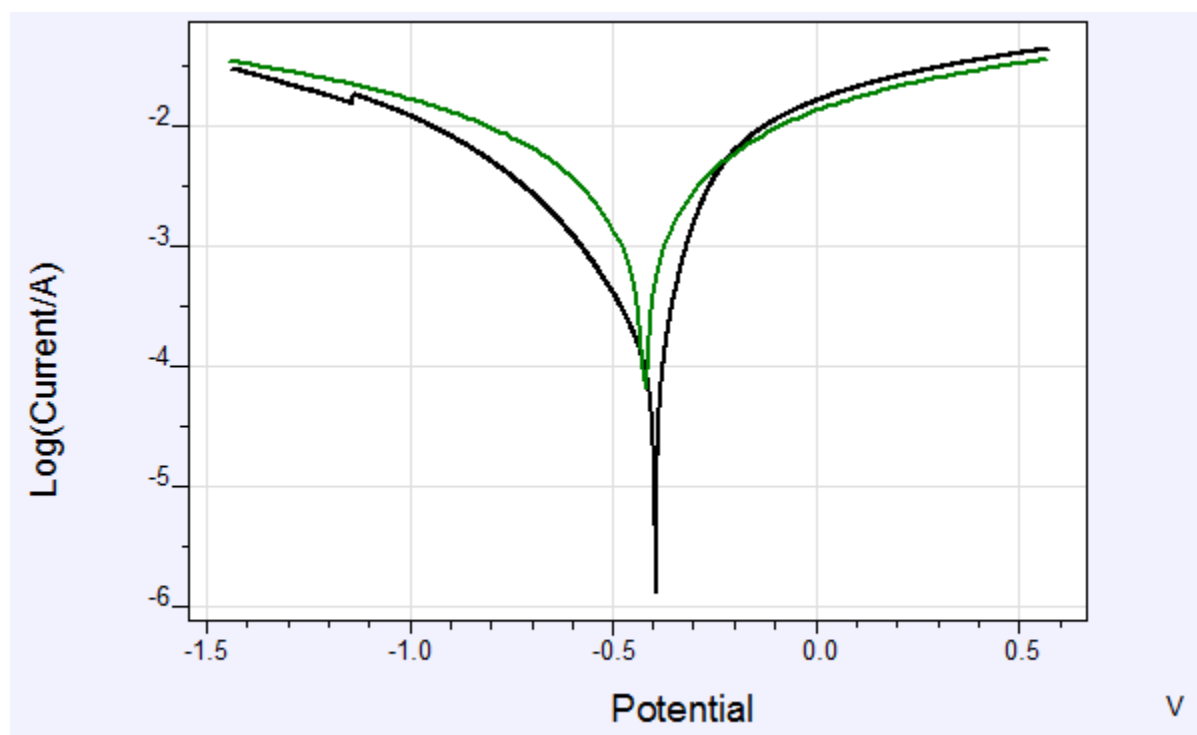


Fig10:-Tafel polarization curves for corrosion product of mild steel in 0.5M HCl in the absence and presence of Aloe Barbadensis Miller.

Conclusion:-

Aloe Barbadensis Miller skin is good corrosion inhibitors for corrosion of mild steel in 0.5 M HCl solution. The maximum efficiency was found to be 81 % at 150 ppm. The adsorption of *Aloe Barbadensis Miller* on mild steel surface obeyed the Langmuir and Al-Ewady Isotherm. The potentiodynamic studies reveal that Aloe Barbadensis Miller is a mixed type inhibitor. The negative value of ΔG° shows that adsorption of *Aloe Barbadensis Miller* on mild steel is a spontaneous process. The increase in E_a value is proportional

to the inhibitor concentration, indicating that the energy barrier for the corrosion interaction is also increased. The results obtained from the Weight loss, electrochemical methods, SEM, EDX suggested that the mechanism of corrosion inhibition is occurring mainly through adsorption process. EIS measurement results indicate that the charge transfer resistance of the mild steel electrode increases and double layer capacitance decreases by increasing the inhibitor concentration, showing the formation of the film. The SEM proves the formation of a film on the surface of the metal.

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RESEARCH ARTICLE

SELECTED UNDERUTILIZED EDIBLE WILD FOOD PLANTS; THEIR ASSOCIATION WITH LEPIDOPTERON FAUNA AND ROLE IN TRIBAL LIVELIHOOD OF JAMBOORI PANCHAYAT SAMITI, ABU ROAD BLOCK IN SIROHI DISTRICT OF RAJASTHAN.

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, lepidopteran fauna.

Abstract

Some plants (underutilized) are lesser-known plant species in terms of marketing and research, but well adapted to marginal and stress conditions. Their indigenous potential and ethno-botanical data are well known to people, whereas, commercial importance and market value is unknown to the public. A socio-economic survey was conducted in Jamboori Panchayat samiti of Abu Road area in Sirohi district of Rajasthan to assess the role of four edible underutilized food plants in tribal livelihood of Jamboori Panchayat Samiti. Findings reveals that the Garasia tribes inhabiting the area are unique in their ethno cultural heritage, far from the modern civilization and mostly depend on the forest and forest produce for their livelihood. These tribes are most backward and live in the interior forest. Livelihood systems in the study area are complex, based on primitive mode of agricultural practices. Common species in natural forest include *Butea monosperma*, *Anogeissus latifolia*, *zizyphus* spp., *Azadirachta indica*, *Madhuca longifolia*, *Boswellia serrata*, *Manilkara hexandra*, *Diospyros melanoxylon*, *Phonix* spp., *Pithocellobium dulce*, *Annona squamosa*, etc. and the main forest product exploitable commercially are *Diospyros melanoxylon* (leaves), bamboo, firewood, gum and barks etc. but naturally occurring and underutilized species includes- *M. hexandra*, *D. melanoxylon*, *M. dioica* and *P. dulce* which are good source of vitamins proteins carbohydrates and minerals. Due to good source of proteins they are also found associated or infested by many insects and also serves as their host plants. In the present paper the association of lepidopteron fauna (Butterflies & Moth) with some selected underutilized plant species with their role in tribal livelihood has been studied. The fruits were nutritionally rich and could be utilized for making by-products. Survey data reveals that if managed properly, these underutilized species could easily compete with commercial fruit-yielding species. Therefore, such fruit trees deserve priority action for conservation in natural forest stands and domestication in farmers' fields as a source of income generating activity.

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Introduction:-

Plants have been a good source of nutrition since the time immemorial and can be categorized as underutilized or neglected crops that occur in wild or are grown in local scale with their poorly addressed economic potential, confining them for mostly traditional and local use only. A considerable proportion of tribal population is still undernourished and the people living in remote areas cannot produce enough food grains to meet the yearly requirement. Therefore, a large share of such population meets its food requirement through non conventional means by consuming various wild plants and animal resources especially in periods of food scarcity (FAO, 2004; Balemie & Kebebew, 2006). Wild food plants serve as alternative to staple food during periods of food deficit and are the valuable supplements for a nutritional balanced diet; one of the primary alternative source of income for many resource poor communities, and the source of species for domestication (Shrestha & Dhillon, 2006). Wild fruits are important for the potential resources which could be utilized at the time of scarcity or during normal days or cultivated as a source of food material for an ever increasing population (Rashid et al, 2008). Besides, these trees and shrubs harbor butterflies, honey bee and other insects of economic importance. Being food source for many insects and animals; these underutilized plants serve as a host for caterpillars whereas other plants provide sustenance for butterflies. In turn, butterflies help to pollinate plants. Insects visit flowers to obtain food, usually in the form of pollen or nectar. (Sharma & Sharma, 2013)

Materials and Methods:-

A preliminary survey was conducted to gather information on geographic area of villages, occupation pattern and socio-economic status of households. The interaction was held with State Forest Department, Sirohi, Zila Parishad and District Statistical Officer, Sirohi and Revenue Department, Abu Road. Similarly Key informant interviews were conducted with selected community leaders, traders and collectors at various study sites. For detailed socio-economic survey 10% families in each study village was surveyed. Total 160 households were surveyed in four study villages of Jamboori Panchayat Samiti through semi-structured and pre-tested questionnaire. Secondary data were collected from State Forest Department, Sirohi, Zila Parishad and District Statistical Officer, Sirohi and Revenue Department, Abu Road. Quantitative data was analyzed using SPSS where as qualitative data through coding and memoing (Punch, 2005). Results were analyzed for livelihood approach through strategic intervention for further implication. To find out the food resources of butterflies & Moth in Jamboori Panchayat Samiti, Abu Road, a systematic study was carried out. The butterflies and moth were observed and identified with the help of various field guide (Evans, 1932).

Result and Discussion:-

Jamboori Panchayat Samiti of Abu Road area of Sirohi district in Rajasthan situated in 24.4641° N, 72.7718° E and consists of four tribal dominated villages viz. Jamboori, Meen, Taleti and Bosa. The main tribes of the region are Garasia but small population of Bhils- Gameti are also found. Their average literacy rate is 55.25% with male and female literacy as 69.98 % and 39.73 % respectively. Ratio of female per 1000 male is 940. There are practically no basic amenities like safe drinking water, electricity, health services, roads and schools etc. These tribes reside in the interior parts of the forest and depend on the forest as their main source of livelihood. They also undertake agricultural activities during the monsoon season. 65% of the total area consists of forestland and only 6 % land is under cultivation (Tripathi, 2016).

Socio-economic status:-

Most of the tribals of the region belongs to BPL category and percentage poverty ratio and intensity ranges from 54.49 -130.70. The Average distance of a village from Block Head quarter is 21 Km and from the district head quarters is 72 Km. Hinduism is the main religion followed in the villages. Hand pump and wells are the most important sources of domestic use of water. Almost all the villages reported to have a primary school at the village level. The average distance of a secondary school from these villages is reported to be around 6 kms. Friends and relatives are the sources for availing credit by the households at the time of the need of credit. Ground water is available at a depth of around 120 feet and the quality of water is good and non brackish.

Average family size is 5.75 in Jamboori, 5.87 in Meen, 6.17 in Bosa and 9.51 in Taleti. Average land holdings per family is 1.39 beegha in Bosa, 1.61 beegha in Jamboori, 1.90 beegha in Taleti, and 2.00 beegha in Meen. Agriculture is mostly rainfed. *Zea mays* (Maize), *Triticum aestivum* (Wheat), *Cajanus cajan* (Tuar), *Phaseolus vulgaris* (Chanwla/Lobia), *Cicer arietinum* (Chana/gram), Vegetables, *Ricinus Communis* (Arandi/castor), *Cyamopsis tetragonoloba* (Gawar), *Brassica juncea* (Raida), *Brassica campestris* (Mustard) are some of the

important crops that are cultivated in the study villages. Animal husbandry is also one of the important source of livelihood. Every family has one or two cows/buffaloes and 5-10 goats but the milk production from cows is very low i.e. 1-1.5 litres per day. Average Cattle owning families per village is 95.49% in Bosa, 96.51% in Jamboori, 98.03% in Taleti and 100% in Meen. People usually do not sell milk and they use it for household consumption. However, the sale of goat and poultry fetches some money (about 1000- 1500) every year. Women equally share the economy of a household and in some instances earns up to 75% of the income (Tripathi, 2016).

Fuelwood is the only source of cooking and is being extracted from nearby forest. On an average minimum and maximum amount of fuel-wood used by a household ranged 50- 70 kg/household respectively and average collection of fuelwood from natural forest ranges between 50-70% percent and remaining requirement is fulfilled by crop residue and cowdung. The distance travelled by tribals for collection of fuelwood is less than 5 km in all the villages. The preferred species for fuelwood includes *Manilkara hexandra*, *Butea monosperma*, *Anogeissus latifolia*, *Acacia nilotica*, *Tamarindus indica* etc. whereas firewood of *Diospyros melanoxylon*, *Madhuca indica*, *Annona squamosa*, *Feronia limonia* etc. are generally not preferred as firewood by tribals.

The average income of households ranges from Rs. 20,000-25,000/- per annum. On average NTFPs contributes about 17% of the total income. However, the contribution ranges from 10-30% to different families. Among underutilized species maximum collection/family/annum is done by *P. dulce* followed by *D. melanoxylon*, *M. dioica* and *M. hexandra* (Table-2; Fig.1) whereas *P. dulce* is maximum underutilized species followed by *M. hexandra*, *D. melanoxylon* and *M. dioica* respectively (Table-2, Fig.-2). Honey collection is also done in Bosa. Tribals generally destroy the bee hive completely during honey collection. Intervention by NGO (Samarthak Samiti, Udaipur) / Agriculture Department etc. may help in non-destructive honey collection. Traders play an important role in NTFP selling. Most of them are local traders operating as middlemen simply collecting NTFP from village agents/shopkeepers and deliver in the nearby town. The tribals barter the daily NTFP collection for their daily use food items viz., pulses, jaggary, soap etc. All the underutilized NTFP species are nutritionally rich and some of them are good source of Vitamin A, B & C (Misra & Misra, 2016).

Table 1:- Socioeconomic status of the people in the study area.

S. No.	Study area and villages →	Bosa	Jamboori	Meen	Taleti
	Variables ↓				
1	Total number of households	222	402	385	357
2	Number of households sampled	40	40	40	40
3	Total population	1261	2419	2175	2032
4	Average family size (persons per sampled household)	6.17	5.75	5.87	9.51
5	Number of males	677	1230	1094	1015
6	Number of females	584	1189	1081	1017*
7	Sex ratio (females per thousand males)	863	967	988	1002
8	Av. Child sex ratio	753	895	934	971
9	Landholding per family (ha)	1.39	1.61	2.00	1.90
10	Annual income per family (Indian Rupees)				
11	**% Poverty ratio and intensity	70.09	54.49	76.50	130.70
12	Average number of animals/household	10	10	7	7
13	Livestock-owning families	212	388	385	350
14	Average fuel wood consumed/household/day (kg)	50.00	70.50	60.00	64.50
15	Families extracting fuelwood from forest (%)	50	70	55	60
16	Distance traveled for fuel wood collection (km)	<5	<5	<5	<5

**Poverty Ratio and Intensity=No of BPL households/ No of APL households x 100

Table 2:- Average fruit yield/annum/family of four selected wild edible species and their utilization by tribals.

S. No.	Species	Local name	Family	Fruit yield/annum (kg) (Mean \pm S.E.)	Prevailing Market Rate(Rs.)	% use by tribals	% remaining unutilized
1.	<i>Diospyros melanoxylon</i>	Tendu	Ebenaceae	44.29 \pm 4.69	15-30	45	55
2.	<i>Momordica dioica</i>	Kankoda	Cucurbitaceae	42.02 \pm 6.90	40-60	60	40
3.	<i>Manilkara hexandra</i>	Khirmi	Sapotaceae	36.75 \pm 3.36	30-60	35	65
4.	<i>Pithocellobium dulce</i>	Jungle jalebi	Fabaceae	50.98 \pm 6.69	15-30	25	75

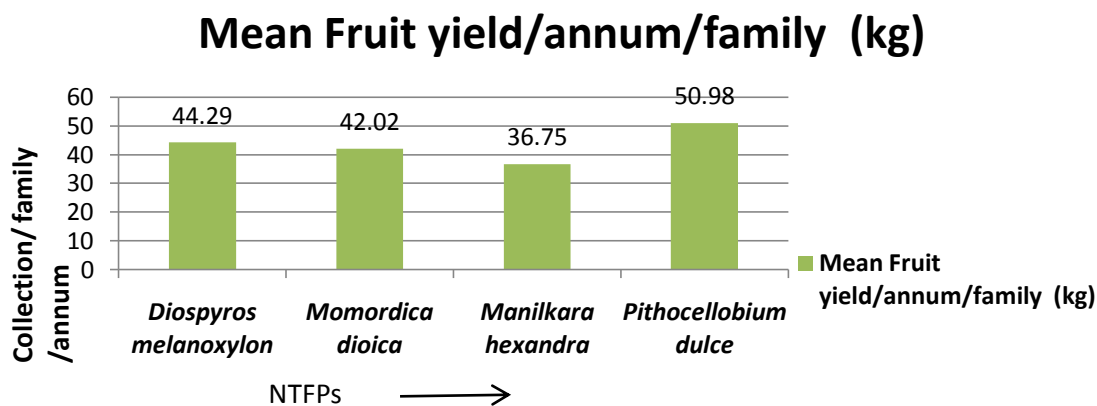
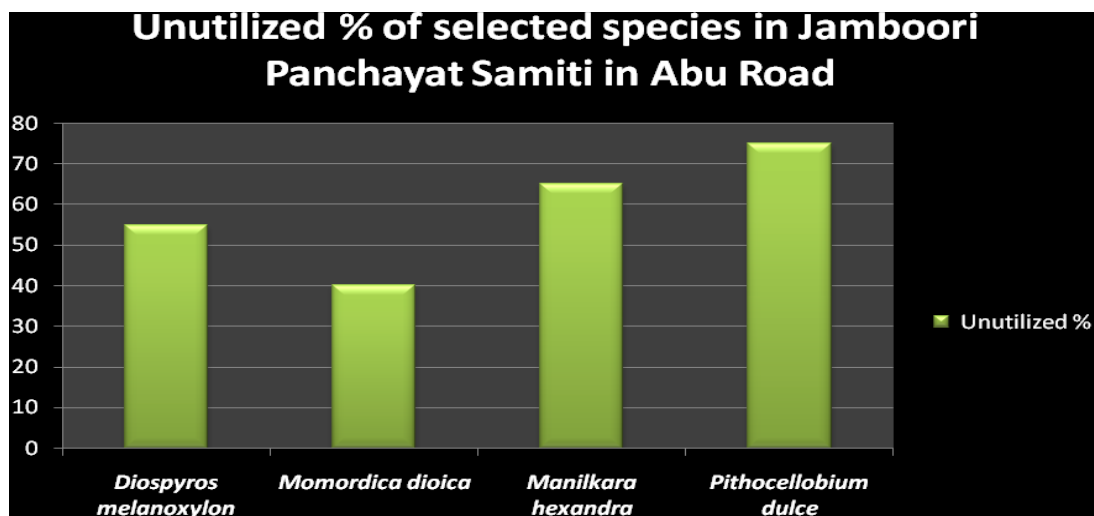
**Fig 1:-** Average fruit yield/annum/family of four selected wild edible species and their utilization.**Fig 2:-** Unutilized % of selected species in study area

Table 3:- Nutritional value of Important edible fruits collected wild by tribals.

NTFP	Moisture content %	Crude protein	Total sugar (mg/g)	Total lipid (mg/g)	Vit. B1 (mg/g)	Vit. C (mg/g)	Ref.
<i>Diospyros melanoxylon</i>	69.6	8.64 (mg/g)	266 (mg/g)	.002 (mg/g)	.004 (mg/g)	.02 (mg/g)	(Misra and Misra, 2016)
<i>Pithocellobium dulce</i>	78.6	28.83 (mg/g)	161.83 (mg/g)	.006 (mg/g)	.0022 (mg/g)	1.08 (mg/g)	
<i>Momordica dioica</i>	87.0	52.06 (g/100 gDW)	14.58 (g/100 gDW)	4 (g/100 gDW)	-----	-----	(Salvi & Ketwa, 2015)
<i>Manilkara hexandra</i>	-----	0.48 (g/100g)	-----	-----	-----	15.67 (mg/100g)	(Jhaver,2014)

Lepidopteron fauna associated with selected underutilized edible wild plant species**Plant species:-** *Diospyros melanoxylon****Grammodes geometrica* Fabricius, 1775 (Lepidoptera: Noctuidae):-**

This is a beautiful moth colored in brown, black and cream. Fore wing consists of a distinct white band flanked on either side by broader black bands. The more distal one is also edged narrowly with white. Beyond this narrow white lining, an orangish-brown band is seen. Again an irregular black lining is present in discal area that goes upto apex. Costa is broadly grey-brown and none of the bands reach upto it. A black lined boundary in wavy manner is present beyond which fringes are clear in brown. The wild host of the moth were found to be *Phyllanthus* (Euphorbiaceae); *Diospyros* (Ebenaceae); *Ricinus* (Euphorbiaceae); ‘grasses’ (Gramineae); *Polygonum* (Polygonaceae); *Zizyphus* (Rhamnaceae).

***Grammodes stolidia* Fabricius, 1775 (Lepidoptera: Noctuidae):-**

This is a beautiful moth colored in brown, black and cream. Forewing consists of a distinct white band flanked on either side by broader black bands. The more distal one is also edged narrowly with white. Beyond this narrow white lining, an orangish-brown band is seen. Again an irregular black lining is present in discal area that goes upto apex. The moth was found to be polyphagous. The larvae feed on Rosaceae, Rhamnaceae (*Zizyphus jujuba*), and Zygophyllaceae (*Tribulus*).

***Euthalia nais* Forster, 1771 ((Lepidoptera: Nymphalidae):-**

This butterfly species is generally seen in dry forested areas. It is beautiful orange colored with black boundary on terminals of both wings with hind wing boundary being broader and diffuse. The edges of wings are wavy, more clearly in HW. FW is shaped like a right angled triangle. The butterfly is fond of sunshine and is attracted by sap, over-ripe fruit and toddy. It feeds on Ebony, *Diospyros melanoxylon* (Ebenaceae) which grows in regions of light rainfall

Plant species:- *Momordica dioica* Roxb.***Pelopidas mathias* Fabricius, 1798 (Lepidoptera:Hesperiidae):-**

It is commonly known as small branded swift. Presence of brand has given it the common name *Small branded swift*. White spots are present on forewing. 2 separate cell spots, 1 in 1b may/may not be present. 3 large quadrangular spots are present in discal area. 3 small spots lie in subapical region. Hind wing shows 2-3 inconspicuous spots. Ventral side shows similar spots on forewing while hind wing shows spots from 1 to 7 in a curved manner. 1 spot is visible in cell area. The common food plants are *Momordica dioica* and monocots.

***Pelopidas conjuncta* Herrich-Schaffer, 1869 (Lepidoptera:Hesperiidae):-**

This is a small brown colored butterfly with white spots. Most of these spots appear to merge with each other. Their flight is very smooth and powerful, therefore the name *Conjoined swift* is given to it. Ventral side is similar. Antennae are small, reaching upto end cell spots. Male has no brand. This butterfly for food depend on *Rottboellia exaltata* (Poaceae), *Tridax procumbens* (Asteraceae), *Aerva lanata*, *Amaranthus gracilis* (Amaranthaceae).

Plant species:- *Manilkara hexandra*

***Graphium sarpedon sarpedon* Linnaeus, 1758 (Lepidoptera: Papilionidae):-**

The blue bands on wings give it the common name *Common bluebottle*. Ground color of wings is brownish-black and the species is generally seen mud puddling. Hind wing is produced to end in a short tail or tooth. A pale blue or greenish blue discal band runs from inside apex of forewing and through cell of hind wing nearly to its dorsum. The adult Common Bluebottle feeds on nectar from a variety of flowering herbs. The larvae feed primarily on the leaves of trees in the families *Myrtaceae*, *Sapotaceae* and *Rutaceae*. Some other favorites are *Polyalthia longifolia* and *Miliusa tomentosa* (Annonaceae).

***Graphium sarpedon teredon* Felder and Felder, 1864 (Lepidoptera: Papilionidae):-**

The blue bands on wings give it the common name *Common bluebottle*. Ground color of wings is brownish-black and the species is generally seen mud puddling. Hind wing is produced more compared to *sarpedon* to end in a short tail or elongated tooth. The discal bands are narrower compared to *sarpedon* on both the wings. The adult Common Bluebottle feeds on nectar from a variety of flowering herbs. The larvae feed primarily on the leaves of trees in the families *Myrtaceae*, *Sapotaceae* and *Rutaceae*. Some other favorites are *Polyalthia longifolia* and *Miliusa tomentosa* (Annonaceae).

***Graphium agamemnon* Agamemnon Linnaeus, 1758 (Lepidoptera: Papilionidae):-**

The triangular shaped wings and green spots gives it the common name *Green spotted triangle*. The synonymous name is *Zetides*. It is a large tailed butterfly with green colored spots or bands all over the wings. The ground color is black with spots towards the base in band form, especially on hind wing. These large butterflies are very fond of *Lantana camara* (Verbenaceae), *Ixora* and *Mussaenda*.

***Graphium agamemnon menides* Fruhstorfer, 1904 (Lepidoptera: Papilionidae):-**

The synonymous name is *Zetides*. It is a large tailed butterfly with green colored spots or bands all over the wings. The ground color is black with spots towards the base in band form, especially on hind wing. Females are similar but with a streak of greenish white along the dorsal margin on both dorsal and ventral sides. They are rarer compared to males. Thorax and abdomen on sides streaked with greenish grey. These large butterflies are very fond of *Lantana camara* (Verbenaceae), *Ixora* and *Mussaenda*.

***Graphium nomius* Esper, 1793 (Lepidoptera: Papilionidae):-**

The presence of prominent spots on forewing and pointed tail on hindwing gives it the common name *Spot swordtail*. Wings are bluish white. Dorsal side shows black while ventral side shows brown bands. Forewing cell with 5 broad transverse black bands, a terminal black band occupies a terminal third of the wing and is traversed by a subterminal series of rounded spots of the ground color. The lower discal area is blackish. This butterfly also feed on *Gmelina arborea*.

Plant species:- *Pithocellobium dulce*

***Charaxes athamas* Drury, 1773 (Lepidoptera: Nymphalidae):-**

Their extremely swift and powerful flight on high trees gives it the common name *Nawab*. It is also known as *Ergolis athamas*. The ground color of wings is black characteristic to the genus. Broad yellowish or slightly green bands are present. They feed on various Fabaceae plants like *Acacia*, *Albizia lebbek*, *Caesalpinia* sp., *Delonix regia*, *Leucaena leucocephala*, *Grewia* sp.

***Charaxes solon* Fabricius, 1793 (Lepidoptera: Nymphalidae):-**

The royal black color of wings gives it the common name *Black Rajah*. It is also known as *Charaxes fabius*. It is a large sized butterfly with basal area and costal margin tinged blue.

***Hypolimnias bolina* Linnaeus, 1758 (Lepidoptera: Nymphalidae):-**

The prominent egg shaped marking on forewing and hindwing gives it the common name *Great Eggfly*. Wings are completely black with forewing having a rounded white patch shaded blue outside covering interspaces 3,4,5. Another small similar patch lies in pre-apical area and a series of white dots in curvy fashion is visible in post discal area. These are attracted to *Lantana camara* (Verbenaceae), *Bauhinia purpurea* (Fabaceae) and *Celosia argentea* (Amaranthaceae). The caterpillar feeds on various tropical plants. Food plants of female are low growing species of *Urticaceae*- *Pouzolzia zeylanica*.

***Danaus chrysippus* Linnaeus, 1758 (Lepidoptera: Danaidae):-**

It is the most common butterfly seen flying at low heights with low speed. It displays warning colors of black and orange which resembles like that of a tiger's body. Abdomen is yellowish while head and thorax are black. These butterflies could be seen at any time of the day. The female form of Danaid eggfly mimics this species.

***Eurema blanda* Linnaeus, 1758 (Lepidoptera: Pieridae):-**

As the name suggests, three spots are present in the cell area of forewing on ventral side, therefore the name derived as *Three Spot Yellow Grass*. The adult butterfly are distinct yellow, small butterflies, with black marginal marks on the wings similar to *Eurema hecabe*. During the five larval instars, the yellow, hairless caterpillars successively defoliate the host tree. Pupae exuviae are often abundant on infested trees. The symptoms of *Eurema blanda* infestation are: skeletonized leaflets, during severe infestation host can be completely defoliated, often pupal exuviae abundant on skeletonized leaves, mostly young trees are affected.

***Ascalapha odorata* Linnaeus, 1758 (Lepidoptera: Noctuidae):-**

Ascalapha odorata is a large bat-shaped, dark-colored nocturnal moth. The forewings are shaped like an obtuse triangle with a fairly long apex. Males can attain a wingspan of 16 cm. The wings are mottled brown with hints of iridescent purple and pink, and, in females, crossed by a white bar. The forewing reniform spot is comma shaped, with dark filling except lighter bluish in the "tail," producing a convincing eyespot. *Ascalapha odorata* is a large bat-shaped, dark-colored nocturnal moth. The forewings are shaped like an obtuse triangle with a fairly long apex. Males can attain a wingspan of 16 cm. The wings are mottled brown with hints of iridescent purple and pink, and, in females, crossed by a white bar. The forewing reniform spot is comma shaped, with dark filling except lighter bluish in the "tail," producing a convincing eyespot.

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Conclusion:-

The underutilized crops are species are used traditionally for food, fibers, fodder, oil or medicinal properties. They also have an underexploited potential to contribute to food security, nutrition, health, income generation and environmental services. Considering the importance of the crops as food, medicine and for industries, these underutilized crops can be exploited at the commercial level and their commercial potential will also provide important environmental services, as they are adapted to marginal soil and climatic conditions. Therefore, these underutilized crops have been included in world-wide plans of action after having successfully raised the interest of decision makers. The high nutritional qualities indicate that the cultivation and consumption of these crops may be helpful in overcoming the nutritional deficiencies predominant in many rural areas of the country and boost the socio-economic condition of the society.

On the other hand, butterflies are known to be the most fascinating creatures existing on this earth and have a great aesthetic value too. They are in close relation with these underutilized species in Jamboori Panchayat Samiti as they procure nectar from flowering plants to meet their energy requirements as well as they serve to be pollinators for various plant species which is the only means of their propagation. Hence, induction of these underutilized crops in agro-ecosystem for value addition products and their commercial exploitation through Village Forest Protection Committees /Self Help Groups as income generating activity will be beneficial for conservation of lepidopteron fauna of the region.

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RESEARCH ARTICLE

BIO DEGRADATION AND CYTOTOXIC POTENTIAL OF BIOSURFACTANT FROM MARINE BACTERIA ASSOCIATED WITH ALGAE *ULVA LACTUCA*.

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Ulva lactuca, antimicrobial activity, TLC, hemolytic activity, metal removal, anticancer activity.

Abstract

Background: The use of biosurfactants to protect the marine environment seems possible since a number of marine bacterial strains can produce biosurfactants during growth on hydrocarbons (Bertrand *et al.*, 1993). For the sake of the environment, the use of biosurfactants is preferable to those of synthetic surfactants. However, little information on either biosurfactant produced by marine microorganisms or biosurfactants active in saline has been reported so far.

Objective: The isolated colonies were identified, potential biosurfactant producer was screened out. Biosurfactant extraction was done by lowering pH using 5M HCl followed by the detection and purification. The extracted biosurfactants were also used to study the effect on metal removal and anticancer activities.

Materials and method: The antibacterial activity was carried out by disc diffusion method, screening of potential biosurfactant producers by hemolytic assay, drop collapsing test, oil displacement test and emulsification activity, detected and purified by silica gel plate (TLC). Anticancer activity was done by *Dimethyl thiazolyl diphenyl tetrazolium bromide*, (MTT). Effects on metal removal were studies using the media with salts of CrSO₄ and ZnSO₄.

Result: Antimicrobial activity of SP2 showed highest activity against *Pseudomonas aeruginosa* (1cm) and *Bacillus megaterium* (2.2cm). Species 2 showed highest activity in the screening methods. TLC showed the presence of lipopeptide and rhamnolipid. The biodegradation using same concentration of bio surfactants produced by SP2, increased with increased concentration of the salts. SP2 microbial extracts showed cytotoxic activity in L929 cell line. These findings suggest that the identified sponges are source of pharmaceutical important compounds.

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Introduction:-

The number of natural products, discovered from several organisms that include plants, animals, and microorganisms, overcomes millions of compounds. Forty to sixty percent derives from terrestrial plants, from

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which twenty to twenty-five possess bioactive properties such as antibacterial, antiviral, anticancer and anti-inflammatory activity (Berdy, 2005). Bio surfactants are categorized mainly by their chemical composition and microbial origin. Major classification includes glycolipids, phospholipids, fatty acids, polymeric bio surfactants and particulate bio surfactants (Desai and Banat, 1997). They have many advantages, such as biodegradability, low toxicity (Poremba *et al.*, 1991) and environmental compatibility (Georgiou *et al.*, 1990). The use of biosurfactants to protect the marine environment seems possible since a number of marine bacterial strains can produce biosurfactants during growth on hydrocarbons (Bertrand *et al.*, 1993). They have the same physical and chemical properties as the synthetic surfactants. However, little information on either biosurfactant produced by marine microorganisms or biosurfactants active in saline has been reported so far. This article aims to provide an overview on the production of biosurfactants by marine microorganisms and to discuss their potential use in bioremediation.

Materials And Methods:-

Collection of algae:-

The algae were collected from Muttom Beach, Kanyakumari District, Tamilnadu. They were found attached to the rocks. Unwanted components were removed from the algae by washing with sea water and then preserved in refrigerator to carry out further studies.

Isolation and Enrichment of Bacterial Species from Marine Algae:-

Frozen algae were cut into small pieces. Mixed colonies of bacterial species were isolated from the algae by growing one of the small pieces of algae in Zobell Marine Broth for 24-48hrs at 37°C. The bacterial colonies were then isolated by spread plate technique (Shirling and Gottlieb, 1966). Characterization of bacterial strains was performed based on Bergey's manual of determinative bacteriology (Bergey *et al.*, 1994).

Screening Methods for Potential Biosurfactant Producers:-

The potential biosurfactant producer was screened by different method such as Hemolytic assay (Carillo *et al.*, 1996), Drop collapsing test by Bodour and Maier (1998) and Oil displacement test (Morikawa *et al.*, 1992). Maximum biosurfactant producing marine bacterial species was maintained on Zobell Marine Agar medium for further study. Those species with highest activity is selected for further studies.

Extraction of Biosurfactant:-

The extraction procedure was carried out based on the method described by (Kingsley *et al.*, 2004). After 96 hrs cultivation, each culture was centrifuged at 8000 rpm, 4°C for 10 min to harvest the cells. The culture supernatant was taken and pH of the culture supernatant was lowered to 2 with 5M HCl and incubating at 4°C for 24 hrs. The precipitate was separated by centrifugation at 8000 rpm for 20min. White precipitate formed culture was selected for further experimentation

Detection and Purification of Bioactive Bio surfactant:-

Thin layer chromatography was carried based on the procedure described by Rashedi (2005), using silica gel plate. A small quantity of crude extract was dissolved in chloroform: methanol (15:35 v/v). 10µl of the crude extract was added onto a TLC plate (silica gel 60) and apply at point of origin near the bottom of the plate. Once dried, develop plate in solvent system of chloroform: methanol: water (65:25:4). When developed remove plate and allow air drying in a fume cupboard. Spray the plate evenly with water or a solution of 5% sulphuric acid and place in an oven at 110°C for 20 min to visualize spots. At preparative scale, remove lipopeptide spots by scraping the silica from plate into a flask. The extract was dissolved in chloroform: methanol (2:1), incubated overnight.

Effect of Bio surfactant on Metal Removal:-

The extracted bio surfactants were used for the removal of metal such as chromium and zinc ions. The nutrient broth medium containing the salts of Chromium sulphate and Zinc sulphate was prepared and sterilized. The salts of Chromium and Zinc were added to the medium at a concentration of 60mg, 80mg, 100mg at 50mL of broth respectively. The pH of the medium was adjusted to 7 and sterilized in an autoclave at 15Lbs pressure for 15 minute. Then the extracted bio surfactants (about 50µL) was inoculated into the medium and incubated for 24hrs, 48hrs, 72hrs at 30°C to study the degradation effects. The tubes were analyzed for the concentration of metals present after treatment using a spectrophotometer. The absorbance can be read at 650nm (Perez *et al.*, 2007)

Antimicrobial Activity:-

The crude bio surfactant was tested for antimicrobial activity using Kirby Bauer disc diffusion method (Bauer *et al.*, 1996). The filter paper disc of uniform size 6mm were impregnated with the bioactive compound and then placed on the surface of the agar plate seeded with the microorganism to be tested. All the plate culture was incubated in an inverted position at 26-28°C for 24-48 hrs. The disc which showed maximum zone of inhibition was selected for further studies.

Invitro cytotoxic assay:-

MCF cells were purchased from NCCS Pune was maintained in Dulbecco's modified eagle's media (HIMEDIA) and grown to confluency at 37°C and 5% CO₂ in a humidified atmosphere in a CO₂ incubator. The cells were trypsinized (500µl of 0.025% Trypsin in PBS/ EDTA solution) for 2 minutes and passaged to T flasks in complete aseptic condition. Extracts were added to grown cells at a concentration of 100µg, 500µg and 1000µg from a stock of 100mg/ml and incubated for 24 hours.

MTT assay:-

MTT assay was performed based on the method followed by (Arung *et al.*, 2000). MTT is a colorimetric assay that measures the reduction of yellow 3-(4, 5dimethylthiazol-2-yl)-2, 5-diphenyl tetrazolium bromide (MTT) by mitochondrial succinate dehydrogenase. The MTT enters the cells and passes into the mitochondria where it is reduced to an insoluble, coloured (dark purple) formazan product. The cells are then solubilised with an organic solvent Dimethyl sulfoxide (Himedia) and the released, solubilised formazan product was measured at 540nm. Since reduction of MTT can only occur in metabolically active cells the level of activity is a measure of the viability of the cells.

The cell culture suspension was washed with 1x PBS and then added 30 µl of MTT solution to the culture (MTT - 5mg/ml dissolved in PBS). It was then incubated at 37°C for 3 hours. MTT was removed by washing with 1x PBS and 200µl of DMSO was added to the culture. Incubation was done at room temperature for 30 minutes until the cell got lysed and colour was obtained. The solution was transferred to centrifuge tubes and centrifuged at top speed for 2 minutes to precipitate cell debris. OD was read at 540 nm using DMSO as blank.

Results:-**Isolation and Identification of Marine Bacteria associated with Ulva lactuca:-**

Four individual marine bacteria have been isolated by Zobell Marine Agar and were named as SP1, SP2, SP3 and SP4. The marine bacteria were identified as *Bacillus* species 1, *Bacillus* species 2, *Pseudomonas* species and *Staphylococcus* species based on Bergey's manual of determinative bacteriology (Bergey *et al.*, 2004).

Screening of biosurfactant:-

Hemolytic test was done for all the four biosurfactants produced from marine bacteria. The highest activity is shown by SP2 which is represented by a clear zone around the colony. In Drop collapsing test, the culture supernatant was added to the surface of oil droplet placed in a 96 well micro titer plate lid. The flat drops shown by the bio surfactant producing culture were scored as positive. More positive results were shown in vegetable oil and petrol by SP1 and SP2 (TABLE-1). Oil biodegradation activity was checked by oil displacement test. Of the four-culture species SP1, SP3, SP4 showed degradation against vegetable oil. SP2, SP3, SP4 showed degradation against kerosene. SP3 and SP4 showed degradation against petrol. SP1, SP3, SP4 shows degradation against diesel (TABLE-1).

Table 1:- Screening By Drop Collapsing And Oil Displacement Test.

Isolates	Drop collapsing test				Oil displacement test (cm)			
	Vegetable oil	Kerosene	Petrol	Diesel	Vegetable oil	Kerosene	Petrol	Diesel
SP1	+	+	+	-	1.3	-	-	1.2
SP2	+	-	+	+	-	1.5	1.9	-
SP3	+	-	-	-	1.5	2.1	1.3	1.6
SP4	+	+	-	-	1.4	1.6	1.5	1.8

Screening of antimicrobial activity:-

The antimicrobial activity was done against pathogenic bacteria *Salmonella typhi* (MTCC-733), *Klebsiella pneumoniae* (MTCC-4030), *Enterobacter aerogenes* (MTCC-111), *Mycobacterium tuberculosis* (MTCC-300), *Streptococcus pyogenes* (MTCC-928), *Pseudomonas aeruginosa* (MTCC-4676), *Bacillus megaterium* (MTCC-453). SP1 showed activity against *Mycobacterium tuberculosis*, *Streptococcus pyogenes*, and *Bacillus megaterium*. SP2 showed high activity against *Pseudomonas aeruginosa* and *Bacillus megaterium*. SP3 showed high activity against *Mycobacterium tuberculosis* and also have activity against *Salmonella typhi*, *Streptococcus pyogenes* and *Bacillus megaterium*. SP4 showed activity against *Klebsiella pneumoniae*, *Streptococcus pyogenes* and *Bacillus megaterium* (GRAPH-1)

Graph 1:- Antimicrobial activity of biosurfactants isolated from the algae.**Separation of Biosurfactant:-****Thin Layer Chromatography:-**

The biosurfactant produced by SP2 were characterized by using TLC plates. The sediments obtained was placed in the TLC plate and the plates when sprayed with 5% sulphuric acid reagent it showed pinkish red spots and yellowish spots in the plates. This shows the lipopeptide and rhamnolipid biosurfactants production by the organism (TABLE-2)

RF Value =

Table 2:- rf value of the compounds separated by tlc.

Species	Rf value		Name of compound
SP1	Pink	0.93	Rhamnolipid
	yellow	0.73	Lipopeptide
SP2	Pink	0.86	Rhamnolipid
	Yellow	0.76	Lipopeptide
SP3	Pink	0.95	Rhamnolipid
	Yellow	0.75	Lipopeptide
SP4	Pink	0.91	Rhamnolipid
	Yellow	0.82	Rhamnolipid

Effect of Biosurfactant on Metal Removal:-

The biodegradation of heavy metal salts ZnSO_4 and CrSO_4 using same concentration of biosurfactants produced by SP2 and different concentration of salts at pH 7 revealed that there was significant degradation of the heavy metals by the surfactant. The degradation increased with the increased concentration of the salts which is illustrated in the following tables (TABLE-3)

Table 3:- Efficiency Of Biosurfactant On Reduction Of Zinc And Chromium.

Concentration of ZnSO_4 (mg)	OD at 650nm				Concentration of CrSO_4 (mg)	OD at 650nm			
	0 hrs	24 hrs	48 hrs	72 hrs		0 hrs	24 hrs	48 hrs	72 hrs
60	0.172	0.143	0.121	0.101	60	0.013	0.018	0.041	0.245
80	0.172	0.148	0.111	0.087	80	0.018	0.029	0.170	0.576
100	0.211	0.011	0.009	0.005	100	0.029	0.035	0.219	0.707

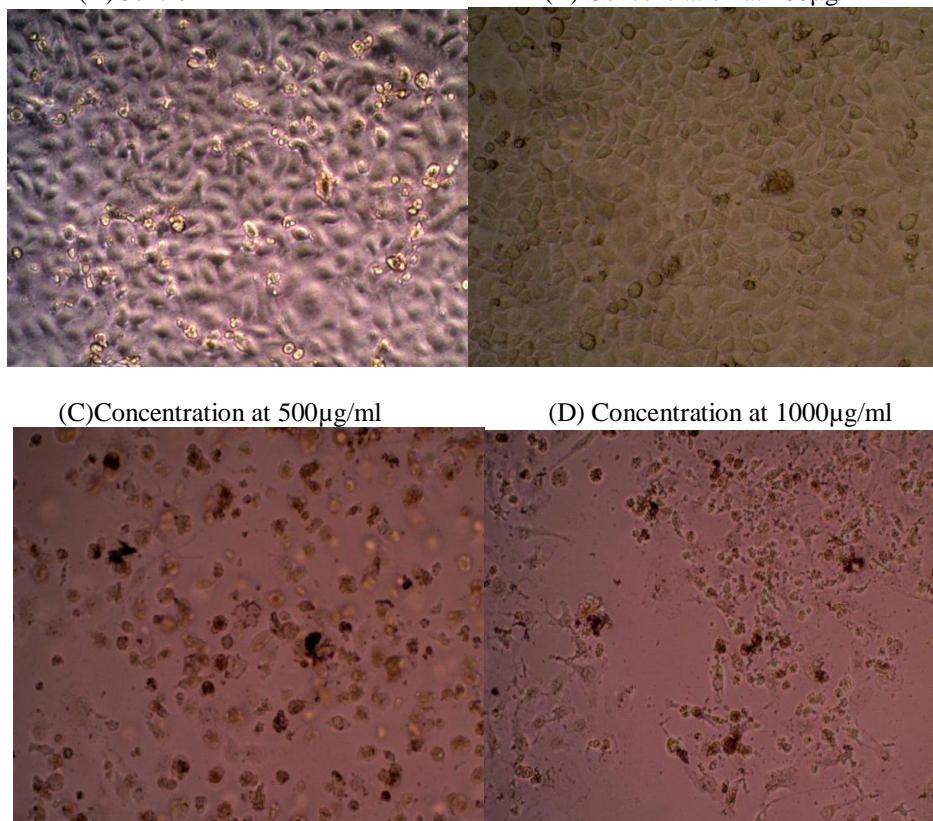
Anticancer activity of bioactive compound produced from algae on MCF cell line:-

The cytotoxic effect of SP2 microbial extract on MCF (Breast cancer) cell line was tested by MTT cell viability assay. The percentage of viability of MCF cell line by 1000 μg of microbial extract showed 54.9% viable cell. Results were showed in (TABLE-4, PLATE-1)

Table 4:- Anticancer Activity Against Cell Line Mcf.

Sample concentration	OD at 540nm	% viability
Control	1.2	100
100 $\mu\text{g}/\text{ml}$	0.7844	65.36
500 $\mu\text{g}/\text{ml}$	0.6840	57.0
1000 $\mu\text{g}/\text{ml}$	0.6588	54.9

Plate 1:- Anticancer Activity Of Microbial Extract On Mcf Cell Line.
 (A)Control (B) Concentration at 100µg/ml



Discussion:-

During the last decade, they have been under investigation as potential replacements for synthetic surfactants and are expected to have many potential industrial and environmental applications (Banat *et al.*, 2000). Among them lipopeptides represent a class of microbial surfactants with increasing scientific therapeutic and biotechnological interests. Currently there is a limited offer of commercially available biosurfactants e.g., surfactin, sophorolipids and rhamnolipids. A variety of new biosurfactants respectively producing strains are the key issue in overcoming the economic obstacles of the production of biosurfactants. Therefore, increased efforts in the discovery of new biosurfactant producing microbes must be made by applying a broad range of different screening methods.

Biosurfactants were first discovered as extracellular amphiphilic compounds of fermentation bacteria (Kitamoto *et al.*, 2009). Initially they were seen interesting due to their ability to increase solubility of insoluble or poorly soluble hydrocarbons. However, the more and more popular trend of using renewable resources in industry (especially in food and pharmaceutical industries) have led to relentless interesting in gaining and application of natural surfactants, mainly biosurfactants. Biological degradation of light crude dispersed in sea water by a surfactant produced by a hydrocarbon degrading microorganism has been monitored in laboratory tests and it showed that oil dispersed by a biosurfactin was more easily degraded than chemically dispersed oil (Francesco Crescenzi *et al.*, 1971) Many bacterial strains have been isolated from coastal and oceanic environments; these bacteria, including the genera *Pseudomonas*, *Vibrio*, and *Flavobacterium*, has been considered to be representative of marine bacteria (Harayama *et al.*, 2004). In this work, four different species of microorganisms have been isolated namely *Bacillus sp. 1 and 2*, *Pseudomonas* and *Staphylococcus*.

In the past few decades, biosurfactants have gained attention because they exhibit some advantages over chemically synthesized surfactants, such advantages include bio degradability, low toxicity, ecological acceptability and ability to be produced from renewable and cheaper substrates and effectiveness at extreme temperature and pH values (Cameotra and Makkar., 1998). The range of industrial applications of biosurfactants includes enhanced oil recovery, crude oil drilling, lubricants, bio remediation of pollutants, health care and food processing. The

antibacterial, antifungal and antiviral activities of biosurfactants make them relevant molecules for applications in combating many diseases and as therapeutic agents (Rodrigues *et al.*, 2006).

Biological methods for the removal of heavy metals from industrial waste may provide an attractive alternative to the physico-chemical process; bio surfactants are one of the compounds that aid in alleviating the heavy metals. Several microbes such as *Bacillus* sp., *Pseudomonas* sp., *Acinetobacter* sp., and *arthrobacter* sp., are reported to carry out the process of bioremediation. In this work, it has been proved that the lipopeptide bio surfactants isolated from the fungal species are also capable of degrading heavy metals like chromium and zinc salts. Hemolysis may occur in vivo or in vitro (inside/outside the body). In vivo hemolysis can be caused by a large number of medical conditions including many gram-positive bacteria (e.g., *Streptococcus*, *Enterococcus* and *Staphylococcus*). There are at present several methods for the detection of hemolytic activity (Arimi *et al.*, 1990; Fricker *et al* 1985). It is the preliminary test for anticancer activity studies. Anticancer activity of the bioactive compound from algae had been performed which results in 54.9% viability in 1000g/ml concentration of SP2 microbial extract. This shows that the compound produced by SP2 cause destruction of half of the total viable cells. The results of the present study thus suggest that isolated biosurfactant may prove to be a promising agent and requires further investigation of its potential cytotoxic activity.

Conclusion:-

Biodiversity of marine algae and its associated microorganisms have been reported to produce several pharmaceutically and industrially important compounds. This work has been done concentrating mainly on the antimicrobial screening and heavy metal degradation of the lipopeptide bio surfactant. It was found that the bacterial species isolated from marine algae *Ulva lactuca* have antimicrobial activity. It is thus concluded that the algal associated bacterial strains are a good source for the treatment of infectious diseases and also it is a good candidate for the treatment of breast cancer (MCF cell line). It was also confirmed that the bio surfactant produced from the antagonistic bacterial strains are a good vector for bioremediation. In future, these properties can lead to the production of novel drugs for the treatment of various diseases including cancer treatment.

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RESEARCH ARTICLE

PERIODONTAL AWARENESS AMONG MEDICAL INTERNS AND POSTGRADUATES IN ANDHRA PRADESH.

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Abstract

Aim: Chronic periodontitis may act as a source for systemic inflammation that impacts overall health. It is associated with an increased risk of systemic illness such as cardiovascular diseases, rheumatoid arthritis, adverse outcomes in pregnancy and diabetes. So it triggered the need of a close association between physician and dentist. Hence the present study was conducted to know the awareness of medical interns and post graduates towards periodontal diseases.

Materials and Methods: A cross sectional study was conducted from June to November 2015. Total 300 participants are selected, in that 260 were responded to participate in the study. A 14-Question survey was distributed to participants chosen randomly from various medical colleges in Andhra Pradesh, India. Questions are designed to assess the knowledge of participants about periodontitis and its association with systemic diseases. The study was approved by institutional ethical committee. Data were analysed by using commercially available statistical software SPSS(P <0.05).

Results: Of the 260 participants, 65% are male and 35% are female. About 52.4% don't know that periodontal infection may cause preterm low birth weight child. Only 2% have rated their knowledge regarding the association of periodontal disease with systemic diseases was excellent but 51.4% know that periodontal disease is a risk factor for systemic diseases. About 25% never screened their patients for periodontal disease i.e. they were not prepared nor trained to screen periodontal diseases.

Conclusion: In this study medical interns and post graduates had limited awareness towards periodontal diseases. Oral health training as part of the medical curriculum is strongly recommended.

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Introduction:-

Periodontal disease is a multifactorial with a complex interaction between microbial infection and host responses. Several risk factors and susceptibilities include age, sex, socioeconomic status, tobacco usage, genetic factors and systemic diseases¹.

There is growing evidence that periodontal disease may act as a source for systemic inflammation that affects the overall health status². Literature stated that systemic inflammatory markers like blood leukocytes and plasma level of C-reactive proteins are highly elevated in persons with periodontal disease^{3,4}. The chronic elevation of inflammatory markers due to periodontitis might be associated with an increased risk of systemic diseases like cardiovascular disease, stroke, peripheral vascular disease⁵, diabetes mellitus⁶ and adverse pregnancy outcomes^{7,8}. This association poses a compelling reason for physician to increase their role in screening oral problems and led to a call for greater collaboration between dentist and physicians⁹.

Periodontal disease may be associated with cardiovascular diseases (CVD) due to mutual risk factors for atherogenesis and periodontal disease. One of such risk factors is pathogens like *T. denticola*, *P. gingivalis*, *F. nucleatum*, *P. intermedia* etc. which are associated with periodontal infection should be localized in serum or atheromatous plaques. Chronic presence of these periodontal microbes can lead to atherogenesis via direct invasion of the arterial wall and the release of systemic inflammatory mediators with atherogenic effects in response to infection. Serum C-reactive protein (CRP) plays a role in endothelial dysfunction and elevated levels of CRP provide insight into the linking of periodontal disease and CVD^{10,11,12,13}.

The bidirectional inter-relationship between diabetes and periodontal disease has been described¹⁴. In other way systemic inflammation play a major role in insulin sensitivity and glucose dynamics. Evidence suggests that periodontal disease can induce an elevated chronic inflammatory state which can exacerbate the insulin resistance by preventing autophosphorylation of insulin receptor and inhibiting second messenger signalling via inhibition of the enzyme tyrosine kinase there by increasing the risk of diabetes^{15,16}. Diabetes results in impairment of neutrophil adherence, chemotaxis and phagocytosis, which may facilitate bacterial resistance in the periodontal pocket and significantly increase the periodontal destruction.

The growing evidence that infection remote from the fetal-placental unit may have a role in the preterm delivery of low-birth-weight infants has led to an increased awareness of the potential role of chronic bacterial infections in the body. Periodontal disease with chronic gram-negative infections results in local and systemic elevations of pro-inflammatory prostaglandins and cytokines¹⁷. Up-regulation of pro-inflammatory cytokines resulting from the normal host response to an infectious agent may represent the key mechanism linking periodontal disease to pre-term low birth weight (PLBW). Microbiological products such as endotoxin will trigger a host immune response, causing both local inflammation and activation of soluble pro-inflammatory mediators such as IL-1, TNF- α and matrix metallo proteinases (MMPs). These inflammatory markers have been shown to cross the placental barrier and to cause fetal toxicity, resulting in preterm delivery and low-birth-weight babies¹⁸.

All these evidence clearly provides an outline, the influence of periodontal disease on systemic health. About 60% of Indian population and half of the American population have been reported with periodontal diseases¹⁹.

Hence the present study was conducted to evaluating periodontal awareness among medical interns and post graduates towards periodontal disease in Andhra Pradesh.

Materials and methods:-

A Cross sectional Study was designed and conducted by department of periodontics, Lenora institute of dental sciences, Rajahmundry, India. Source of the data was collected from the medical interns and post graduates from various medical colleges in Andhra Pradesh, during June to November 2015. Total of 300 participants were selected for the study, in that 260 were responded to participate in the study. A self-administered, structured questionnaire was distributed to all participants. The questionnaire had 14- questions. These questions were developed to assess the knowledge of medical interns and postgraduates regarding the etiopathogenesis of periodontitis and its relationship with systemic diseases. Participants were asked to circle the single best answer. The study was approved

by the Ethical Committee, Lenora Institute of Dental Sciences, Rajahmundry, Andhra Pradesh. Informed consent was taken from all the participants.

Analyses were performed using commercially available statistical software (SPSS). Chi-square analysis was done to compare male and female subgroups. The spearman correlation co-efficient was used to correlate the self-assessed knowledge /orientation of medical interns and post graduates.

Results:-

The survey was conducted with a sample of 300 participants, in that 260 were responded. The 260 responders were medical interns and post graduates from various medical colleges in Andhra Pradesh. Mean age of the participant's ranges from 26-34 years. All participants answered all questions.

Table 1 & Graph 1 summarize the true or false knowledge items with correct answers and percentage of subjects who answered correctly. Among the participants 39.8% of the people don't know the signs and symptoms of the periodontal diseases and nearly 50.8% didn't know the effect of diabetes increasing the severity of periodontal diseases. Most of the participants i.e. 75% of medical interns and post graduates knows that poor oral health may increase the risk of cardiovascular diseases but its percentage is less when compared to the results of Farah Asaad et al. study in medical interns of Saudi Arabia i.e. 92.8%.

Graph 2 shows that percentage of medical interns and post graduates exploring clinical practice behaviours/ orientations. 73.8% participants believed that the smoking is an etiological agent for periodontal disease, only 15.8 % of participants knows that the periodontal disease act as a risk factor for systemic diseases. About 52.4% people don't know that the periodontal diseases associated with low birth weight baby and 33.5% of people marked that genetics may have a role in periodontal diseases.

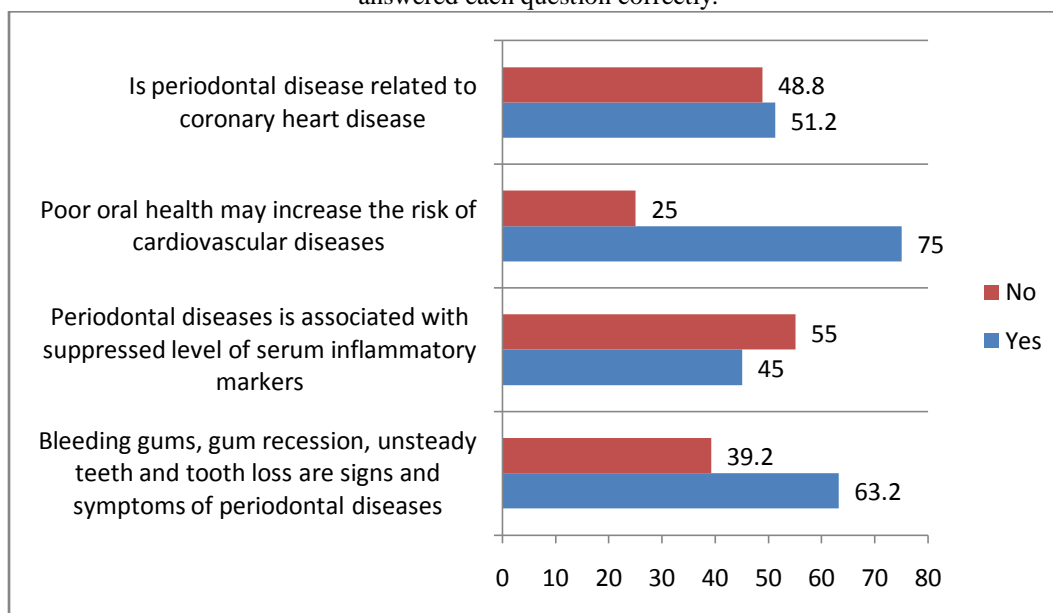
Graph 3 shows that the percentage of attitudes towards periodontal disease and perception that may influence the clinical practices. More than half of the people i.e. 44.6% never asked their patients whether they were diagnosed with periodontal diseases nor screened for it. About one fourth of the people only often referred their patients to a dentist for evaluation and care.

About 40% of the respondents disagree/ strongly disagree that discussing or evaluating periodontal status of their patients was peripheral to their role as physician. Half of the study population (53.1%) rate their knowledge about periodontal diseases and its association with systemic disease was moderate whereas only 2.3% rated as excellent and about half of the people known that periodontal disease is a risk factor for systemic diseases.

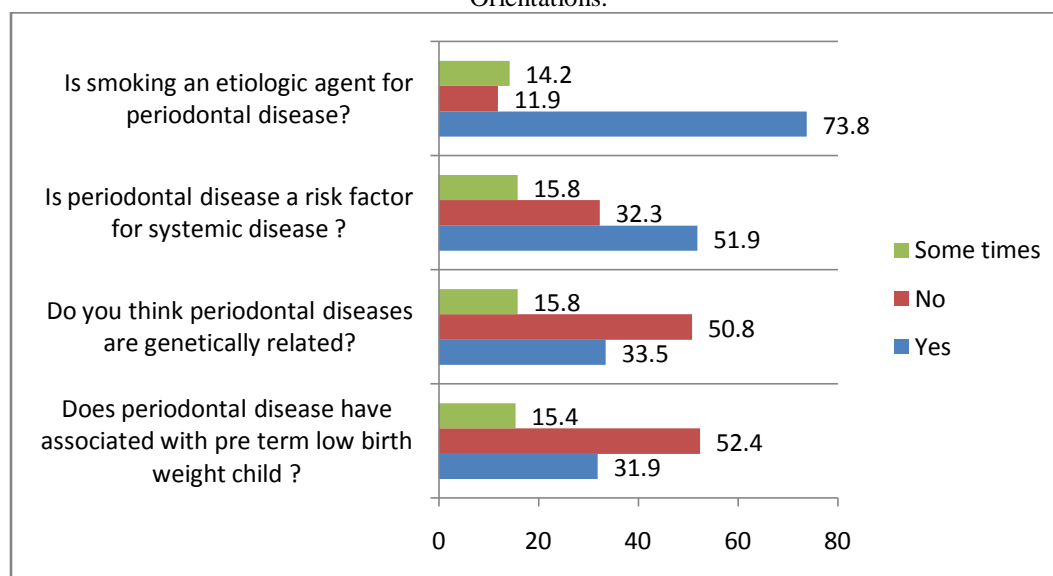
Table 1:- True/ false knowledge items with correct answer percentage of subjects who answered correctly

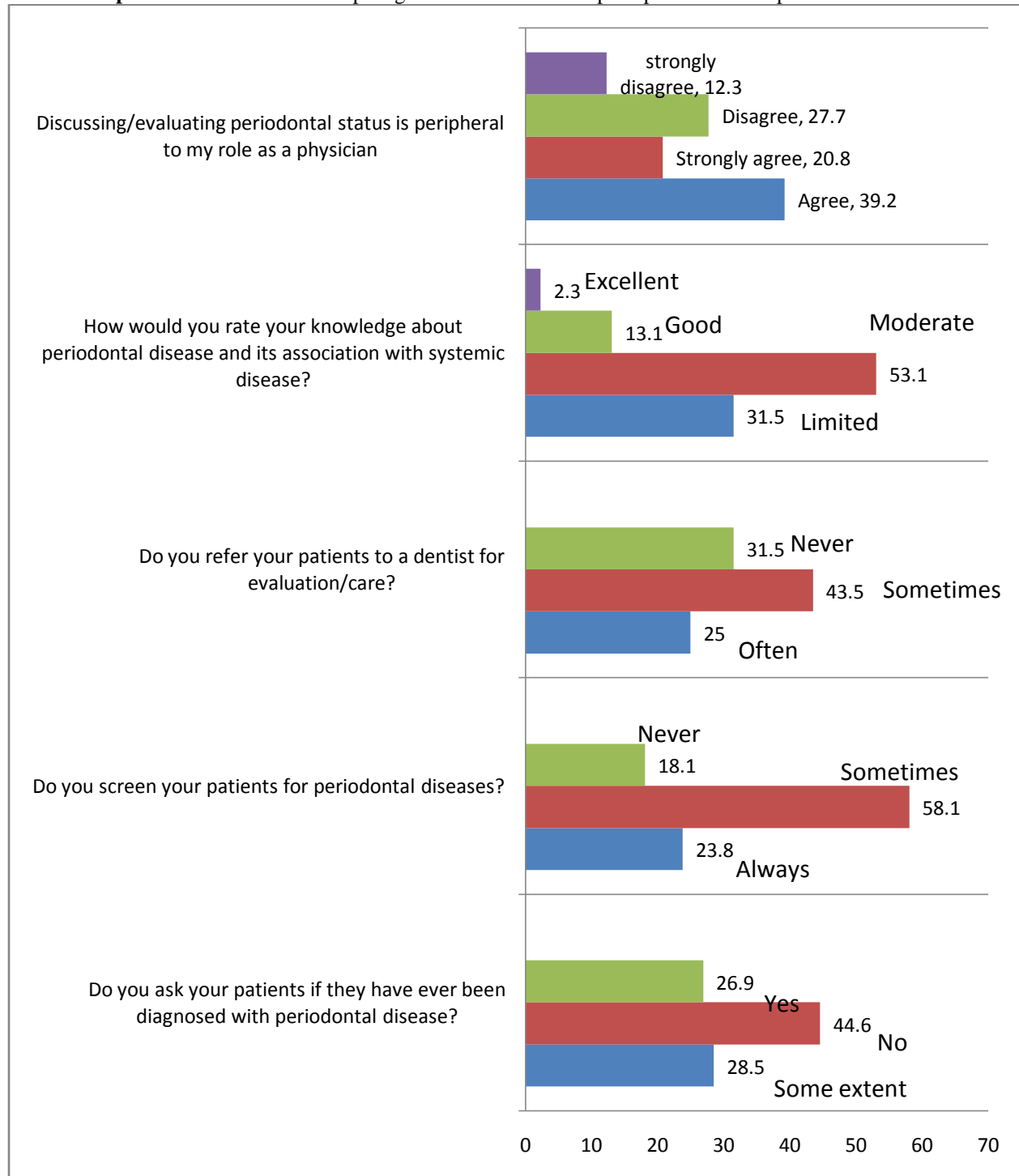
S.No	Question	Correct answer	Yes	No
1.	Bleeding gums, gum recession, unsteady teeth and tooth loss are signs and symptoms of periodontal diseases	True	63.2	39.8
2.	Periodontal diseases is associated with suppressed level of serum inflammatory markers	False	45	54.5
3.	Poor oral health may increases the risk of cardiovascular diseases	True	75	25
4	Is periodontal disease related to coronary heart disease	Yes	51.2	48.8
5.	Periodontal diseases are less prevalent/ severe in patients with diabetes	False	49.2	50.8

Graph 1:- shows the true/ false knowledge items with the correct answer and the percentage of subjects who answered each question correctly.



Graph 2:- Percentage of medical interns and post graduates questions exploring clinical practice behaviours/ Orientations.



Graph 3:- Medical interns and post graduates attitudes and perceptions towards periodontal disease**Discussion:-**

Periodontal diseases are the most common diseases of the oral cavity caused due to the microbial interactions and host immune response. More than 500 different bacterial species are capable of colonizing the oral cavity²⁰. The wide spread of bacterial products like lipopolysaccharides throughout the body may cause interaction between periodontal and systemic diseases^{21,22,23,24}. Hence periodontal diseases may act as an independent risk factor for

systemic diseases. In this study we found that awareness of medical interns about periodontal diseases is limited. Medical interns and post graduates rarely asked about periodontal diseases when taking medical history and they were not comfortable in performing a simple periodontal examination.

Most of the medical interns and postgraduates did not have enough knowledge about the relationship between periodontal diseases and diabetes. Only 49.2% know that the periodontal disease is more prevalent in patients with diabetes.

In our study 31% of medical interns and post graduates agreed that periodontal diseases might increase the risk of premature foetal death but Farah asaadet al²⁵. showed 60% of medical interns know about the risk of periodontal diseases in pregnancy.

Our study explored the attitudes of medical interns and post graduates towards periodontal disease. 23.8% of the participants never screened for periodontal diseases in their patients and 44.6% people never asked their patients if they ever diagnosed with periodontal diseases. Nearly 40% of the participants disagree/ strongly disagree that discussing/ evaluating periodontal status is peripheral to my role as a physician. In a similar study done by Quijano et al²⁶ in medical trainees and found that 30% strongly disagree that evaluating/ discussing periodontal status is peripheral to their role. Sometimes patients require a physician opinion regarding the dental treatment; therefore there is a crucial need of a knowledge regarding the periodontal diseases and its effects on systemic health.

Our results stated that medical interns and post graduates are not prepared or nor trained to screen their patients for periodontal diseases and they had limited knowledge about association between periodontal health and general health. Since there is a strong correlation between the periodontal and systemic health; it is wise to implement oral health education training in medical curriculum. This will result in relevant and more efficient collaboration between medical and dental professionals which will improve the overall health of a patient.

Limitations of the study include limited population size and area. Larger population size is indicated for precise observations. It is a self-reported attitudes and practices; it might be biased to what participants believe.

Our study concluded that medical interns and post graduates have limited knowledge regarding the periodontal diseases and its influence on systemic diseases. Hence the oral health education and training as a part of medical school curriculum is strongly recommended to medical council of India.

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RESEARCH ARTICLE

A STUDY ON FACTORS INFLUENCING EMOTIONAL INTELLIGENCE OF TEACHERS OF MANAGEMENT EDUCATION IN COIMBATORE CITY.

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Emotional Intelligence, Influencing factors of Emotional Intelligence, Management education, Teachers.

Abstract

This study aimed at studying and understanding the factors influencing the Emotional Intelligence of teachers of management education in Coimbatore City and studying the relationship between the demographic variables and factors of the Emotional Intelligence. Present study is descriptive in nature and convenience sampling technique was adopted. Both primary and secondary data were collected for the study. Primary data were collected through questionnaire and the secondary data were collected from magazines, journals, online databases, websites etc. The collected were analysed using percentage analysis and ANOVA. From the analysis, certain findings were made. Based on the findings some suggestions were given and certain conclusions are drawn.

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Introduction:-

The relationship between teachers and students was oriented towards just imparting knowledge in the past. This concept is gradually changing though not in a radical manner; education is not restricted with knowledge but formation of students. Developing students in courage and maturity to encounter the complexity of life is capturing the visionary educators of present day.

Current theories - The Multiple Intelligences Theory (Gardner, 1983) & The Emotional Intelligence Theory (Mayer & Salovey, 1995 Goleman, 1995) - point out the important of emotional intelligence for the students of higher learning. IQ alone does not guarantee the required success in the modern organizations; in fact it only counts for 20%, and the rest goes for Emotional and Social Intelligences, and luck (Goleman, 1995).

Emotional intelligence is increasingly becoming an important concept in the workplaces, workshops and conferences are organized to help employees and managers to understand the important of Emotional Intelligence and to implement it in their work place (Abraham, 1999). If Emotional Intelligence is considered nowadays vital for success, then it becomes imperative that students of higher learning must pay more attention to the components of Emotional Intelligence.

The purpose of this study is to understand the factors influencing Emotional Intelligence of teachers of management education in various colleges.

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Emotional Intelligence

Emotional Intelligence: It is being able to monitor our own and others' feelings and emotions, to discriminate among them, and to use this to guide our thinking and actions (Salovey & Mayer, 1990). The emotionally intelligent person is skilled in four areas: Identifying, using, understanding, and regulating emotions (Salovey & Mayer, 1990).

According to Goleman (1995) emotional intelligence consists of five components: Knowing our emotions (self-awareness), managing them, motivating ourselves, recognizing emotions in others (empathy), and handling relationships.

Teaching emotional and social skills is very important at school, it can affect academic achievement positively not only during the year they are taught, but during the years that follow as well. Teaching these skills has a long-term effect on achievement. (Elias et al, 1991).

Richardson and Evans (1997) explored some methods for teaching social and emotional competence within a culturally diverse society. Their purpose was to help students connect with each other, in order to assist them in developing interpersonal, intrapersonal, and emotional intelligences, arguing that these intelligences are essential for personal accomplishment.

At La Salle Academy, a private school in providence, Rhode Island, students are given lessons in emotional intelligence across the curriculum. This is part of an exhaustive program in social and emotional education called "Success for Life."

Finnegan (1998) argues that schools should help students learn the abilities underlying emotional intelligence. Possessing those abilities, or even some of them, "can lead to achievement from the formal education years of the child and adolescent to the adult's competency in being effective in the workplace and in society".

People with emotional intelligence have a confidence in themselves and they know that their personal happiness is up to them and no one else. They check their emotions before labeling other people and their actions. Such people can inspire and lead and make others feel good about them, while maintaining their own integrity and sense of personal worth. People with emotional intelligence look out for their own well being, as well as that of others as they understand that life is not just about them, it's about balance.

There are two types of intelligence: rational and emotional. Deliberate, analytic reflection is the hallmark of the thinking mind, but the emotional mind is a moment quicker, springing into action on impulse unless tempered by the rational mind.

There should be a balance between emotional and rational thinking, with the emotional brain feeding into and informing the operations of the rational brain, and the rational mind refining the inputs of the emotional mind. Developing a harmony between the head and the heart can foster happiness and promote success in life.

EI is defined as the ability to process emotional information, particularly as it involves the perception, assimilation, understanding, and management of emotions (Mayer & Cobb, 2000). Baron (1997) considers EI as an array of non-cognitive capabilities, competencies, and skills that influences one's abilities in succeeding and coping with the environmental demands and pressures.

According to Hein (2005), EI is the innate potential to feel, use, communicate, recognise, remember, learn, manage and understand emotions. Goleman (1998) considers EI as the capacity for recognising our own feelings and those of others, for motivating ourselves, and for managing emotions well in ourselves and in our relationships. It describes abilities distinct from, but complementary to, academic intelligence, the purely cognitive capacities measured by IQ. Daniel Goleman's theory is one of the most widely accepted concepts of EI.

According to Goleman's emotional competence framework, EI can be viewed within a set of personal and social competencies. Table 1 illustrates the Goleman's EI competency framework. Within this framework, Goleman has indicated the factors that influence the EI levels of an individual. Goleman (2001) has also suggested that these factors could be measured and improved. Henceforth, begins the concept of learning EI and improving leadership qualities of an individual and thereby the organisation.

Table-1:- Goleman's EI competency framework.

	Self Personal Competence	Other Social Competence
Recognition	Self-Awareness <ul style="list-style-type: none"> Emotional self-awareness Accurate self-assessment Self-Confidence 	Social Awareness <ul style="list-style-type: none"> Empathy Service Orientation Organisational awareness
Regulation	Self-Management <ul style="list-style-type: none"> Self-control Trustworthiness Conscientiousness Adaptability Achievement drive Initiative 	Relationship Management <ul style="list-style-type: none"> Developing others Influence Communication Conflict management Leadership Change catalyst Building blocks Teamwork and collaboration

History of Emotional Intelligence:-

1930 – Edward Thorndike describes the concept “social intelligence” as the ability to get along with other people.

1940 – David Wechsler suggests that affective components of intelligence may be essential to success in life.

1950 – Humanistic psychologists such as Abraham Maslow describe how people can build emotional strength.

1975 – Howard Gardner publishes the Shattered Mind, which introduces the concept of multiple intelligence.

1985 – Wayne Payne introduces the term emotional intelligence in his doctoral dissertation entitled “A study of emotion: developing emotional intelligence; self- integration; relating to fear, pain and desire”.

1987 – In an article published in Mensa Magazine, Keith Beasley uses the term “emotional quotient.” It has been suggested that this is the first published use of the term, although the Reuven Bar-On claims to have used the term in an unpublished version of his graduate thesis.

1990 – Psychologists Peter Salovey and John Mayer publish their landmark article, “Emotional intelligence,” in the journal *Imagination, Cognition, and Personality*.

1995 – The concept of emotional intelligence is popularized after publication of psychologist and New York Times science writer Daniel Goleman's book *Emotional Intelligence: Why it can matter more than IQ*.

Significance of the study:-

“More students leave college because of disillusionment, discouragement, or reduced motivation than because of lack of ability or dismissal by school administration”.

- The use of Emotional Intelligence to aid the student development process can address these non- academic challenges.
- Investing in the emotional development of students also impacts leadership effectiveness, both on campus and in the future.

Finally, emotional competency development benefits the career development process, promoting a successful transition from college into the workplace.

Why EI is important for teachers?

- It influences what we do
- It affects who we meet
- It determines how we look
- It decides students and teachers course of life
- It helps to express emotions appropriately instead ignore them

Statement of the problem:-

Emotional intelligence of teachers plays an important role in determining the educational institution success. Now a days, students are more sensitive and very reactive towards the action of their teachers. Emotional intelligence of teachers not only affects the students but also affect their peer group. So it is one of the important duties of every teacher to control and deliver his/her Emotional Intelligence in a positive way.

- Teachers can't focus on their work fully when their family problem interrupts their mind. So normally they deliver negative emotions.
- Usually most of the teachers show the negative emotions when they receive negative comments from the higher authority in front of their peer group.
- Most of the times misbehaving students are subject to receive negative emotions from the teachers.

These are the major problems that every institution and the students were facing in the current scenario. This study helps to understand the factors influencing their Emotional Intelligence.

Scope of the study:-

The study of Emotional Intelligence focuses on handling of emotions by oneself and with others. There is no relation between the qualification and the intelligence level. The study is based on the evaluation of Emotional Quotient among the teachers. The reason for the study of Emotional Intelligence is that nowadays students are more sensitive and inactive. So the teachers should have a positive Emotional Intelligence that may help the students' growth as well as the institution growth because the teachers are the foundation for the students' life and for the institution. This study helps to understand the factors that influence the Emotional Intelligence of teachers of Management education.

Objectives of the Study:-

Core Objective:-

To study and understand the factors influencing the Emotional Intelligence of teachers of management education in Coimbatore City.

Secondary Objectives:-

1. To trace the theoretical perspective of Emotional Intelligence.
2. To study the relationship between the demographic variables and factors of the Emotional Intelligence.

Hypotheses:-

1. The influencing factors of Emotional Intelligence have a positive relationship when compared with Age.
2. The influencing factors of Emotional Intelligence have a positive relationship when compared with Gender.
3. The influencing factors of Emotional Intelligence have a positive relationship when compared with Working Experience.
4. The influencing factors of Emotional Intelligence have a positive relationship when compared with Marital status.

Methodology:-

Present study is descriptive in nature and convenient sampling technique was adopted. Both primary and secondary data were collected for the study. The major sources of the data used to carry out the analysis were primary data. First-hand information was collected through a questionnaire from 250 management education teachers working in various colleges in Coimbatore city. The secondary data namely literature relating to the study was gathered from the national and international journals, newspaper, magazines, articles, other records and also from web sources on the internet. Pilot study was done for 50 among the sample respondents. The data were first evaluated for their validity and Reliability. Incomplete and ambiguous responses were rejected. The tools of analysis like Percentage Analysis, ANOVA were used in the study. The Statistical Package for Social Sciences (SPSS) was used to analyse the data and draw the inference.

Table – 2:- Gender

S.No	Gender	Frequency	Percent
1	Male	117	46.8
2	Female	133	53.2
	Total	250	100.0

From the table it could be understood that a 53.2 percent of the teachers were female and a remaining 46.8 percent of the teachers were Male. It is understood that the Female respondents were higher in the management education appointments.

Chart-1:- Gender

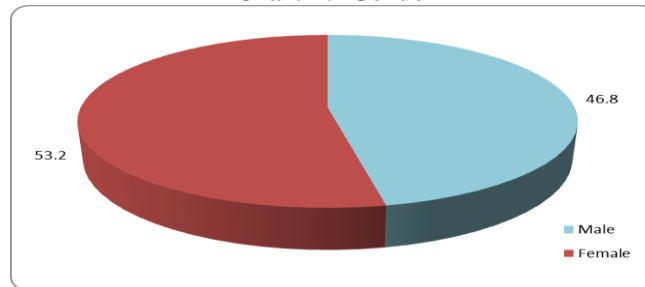


Table – 3:-Age

S.No	Age	Frequency	Percent
1	20-25yrs	36	14.4
2	25-30yrs	71	28.4
3	30-35yrs	57	22.8
4	35-40yrs	54	21.6
5	>40yrs	32	12.8
	Total	250	100.0

It is clear from the analysis that 28.4 percent of the teachers age ranges from 25 – 30 years, 22.8 percent of the respondents age ranges between 30 – 35 years, 21.6 percent of the respondents age ranges between 35 – 40 years, 14.4 percent of the respondents age ranges between 20 – 25 years, and a least of 12.8 percent of the respondents age was above 40 years.

Chart-2:- Age

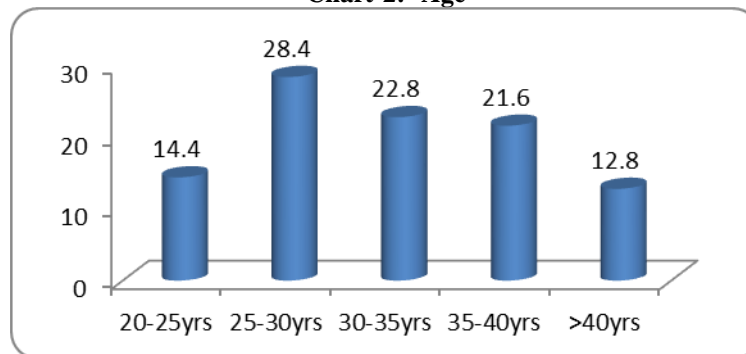


Table – 4:- Marital status

S.No	Marital Status	Frequency	Percent
1	Single	39	15.6
2	Married	102	40.8
3	Widow	52	20.8
4	Divorcee	57	22.8
	Total	250	100.0

From the table it could be understood that 40.8 percent of the respondents were married, 22.8 percent of the respondents were Divorcees, 20.8 percent of them were Widow, and a least of 15.6 percent of them were Single. It is well understood from the table that a 40.8 percent of the respondents were married.

Chart-3:- Marital Status

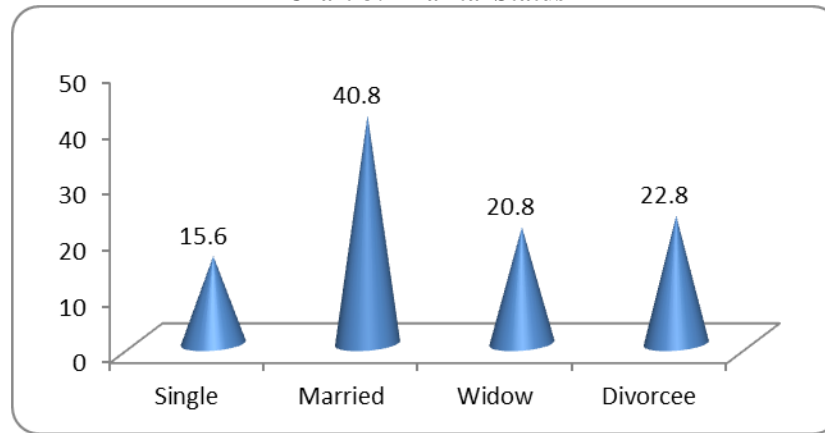


Table – 5:- Work Experience

S.No	Work Experience	Frequency	Percent
1	Less than 5 yrs	33	13.2
2	5-10yrs	72	28.8
3	10-15yrs	53	21.2
4	15-20yrs	52	20.8
5	Above 20yrs	40	16.0
	Total	250	100.0

It could be learnt from the table that 28.8 percent of the respondents have an experience of 5- 10 years, 21.2 percent of the respondents have an experience of 10 -15 years, 20.8 percent of them have an experience of 15 – 20 years, 16 percent of the respondents have an experience of above 20 years, 13.2 percent of the respondents have an experience of less than 5 years.

Chart-4:- Work Experience

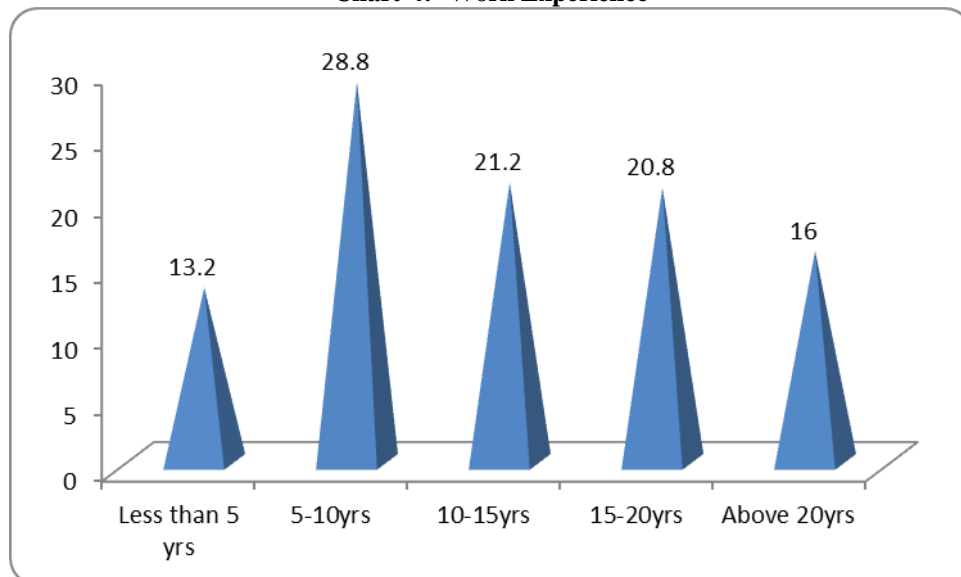


Table – 6:- Influencing Factors of Emotional Intelligence

S.No	Statements	Agree	Disagree	Total
1	I enjoy establishing a good working relationship with a group	110 (44)	140 (56)	250
2	They often seem just not motivated to join in	134 (53.6)	116 (46.4)	250
3	I am always worried I am going to be asked the question that will catch me out.	135 (53.6)	116 (46.4)	250
4	When teaching I see myself as making the best of a bad situation	73 (29.2)	177 (70.8)	250
5	Although I get anxious, I feel confident and relaxed in my role.	124 (49.6)	126 (50.4)	250
6	There always seem to be learners who are simply lazy.	128 (51.2)	122 (48.8)	250
7	Sometimes I wonder what is the point	142 (51.2)	108 (43.2)	250
8	I do worry I am going to be found out as knowing as much as they think.	111 (44.4)	139 (55.6)	250
9	It is really like casting pearls before swine.	139 (55.6)	111 (44.4)	250
10	I sense that nothing really changes.	123 (49.2)	127 (50.8)	250
11	I often feel that I have let at least some of the learners down .	125 (50.0)	125 (50.0)	250
12	When I approach a new session I expect that both they and i will enjoy ourselves and learn something useful.	111 (44.4)	139 (55.6)	250
13	No matter how much effort I put in there will always be some who are just not going to get it.	128 (51.2)	122 (48.8)	250
14	I am always conscious of some members not getting much from it.	128 (51.2)	122 (48.8)	250
15	I enjoy corresponding to the challenges each new session brings	122 (48.8)	128 (51.2)	250
16	Sometimes I think they look as despondent as I feel.	124 (49.6)	126 (50.4)	250

The above table shows how many percent of respondents agree and disagree to the statements regarding the factors influencing the Emotional Intelligence. The numbers within the brackets indicate the respective percentage value. These statements are clustered into four life positions and those are given below.

Table-6.1 Get Rid of Position

S.NO	GET RID OF POSITION	Agree	Disagree	Total
2	They often seem just not motivated to join in	134 (53.6)	116 (46.4)	250
6	There always seem to be learners who are simply lazy.	128 (51.2)	122 (48.8)	250
9	It is really like casting pearls before swine.	139 (55.6)	111 (44.4)	250
13	No matter how much effort I put in there will always be some who are just not going to get it.	128 (51.2)	122 (48.8)	250

The agreeability towards the statements 2,6,9 ,13 indicates that the teaching professionals spends time and on the idea on “**Defensive or Get Rid of Position**”. These statements responds to the life position that “I am ok, You are not ok. A defensive position where generally see the teaching professionals as blameless but good at seeing failings in others.

Table-6.2 Get Away Position

S.NO	GET AWAY POSITION	Agree	Disagree	Total
3	I am always worried I am going to be asked the question that will catch me out.	135 (53.6)	116 (46.4)	250
8	I do worry I am going to be found out as knowing as much as they think.	139 (55.6)	111 (44.4)	250
11	I often feel that I have let at least some of the learners down .	125 (50.0)	125 (50.0)	250
14	I am always conscious of some members not getting much from it.	128 (51.2)	122 (48.8)	250

The agreeability towards the statements 3,8,11 and 14 indicates that the teaching professionals spends time on the idea on “**Get Away Position**”. These statements indicate to the life position that “I am not ok, you are ok”. Where in the teaching professionals find fault with themselves and feels that others are capable and as “Potential prosecutors”.

Table-6.3 Get –Nowhere- With Position

S.NO	GET- NOWHERE-WITH POSITION	Agree	Disagree	Total
4	When teaching I see myself as making the best of a bad situation	73 (29.2)	177 (70.8)	250
7	Sometimes I wonder what is the point	142 (51.2)	108 (43.2)	250
10	I sense that nothing really changes.	123 (49.2)	127 (50.8)	250
16	Sometimes I think they look as despondent as I feel.	124 (49.6)	126 (50.4)	250

The agreeability towards the statements 4,7,10 and 16 indicates that the teaching professionals spends time on the idea on “**Get nowhere with**” position. These statements indicate to the life position that “I am not ok, you are not ok”. And can be acknowledged as the teaching professionals are having negative view of yourself and everyone else and feel “Despair”.

Table-6.4

S.NO	GET ON WITH POSITION	Agree	Disagree	Total
1	I enjoy establishing a good working relationship with a group	110 (44)	140 (56)	250
5	Although I get anxious, I feel confident and relaxed in my role.	124 (49.6)	126 (50.4)	250
12	When I approach a new session I expect that both they and I will enjoy ourselves and learn something useful.	111 (44.4)	139 (55.6)	250
15	I enjoy corresponding to the challenges each new session brings	122 (48.8)	128 (51.2)	250

The agreeability towards the statements on 1,5,12 and 15 indicates that the teaching professionals spends time and on the idea on “**Get on with**” position. These statements indicate to the life position that “I am ok, you are ok”. These statements link to the life position I am ok and you are ok. The teaching professionals are open minded and tackle problems.

Analysis of Variance:-

H1 : The influencing factors of emotional intelligence have a positive relationship when compared with Age.

Table – 7:- The influencing factors of emotional intelligence have a positive relationship when compared with Age.

Source of variance		Sum of Squares	Df	Mean Square	F	Sig.	Result
Defensive position	Between Groups	78.389	4	19.597	3.067	.017*	S
	Within Groups	1565.547	245	6.390			
	Total	1643.936	249				
Get rid of position	Between Groups	66.758	4	16.690	2.915	.022*	S
	Within Groups	1402.558	245	5.725			
	Total	1469.316	249				
Get away position	Between Groups	557.483	4	139.371	29.001	.000**	S
	Within Groups	1177.401	245	4.806			
	Total	1734.884	249				
Get on with position	Between Groups	193.555	4	48.389	7.485	.000***	S
	Within Groups	1583.809	245	6.465			
	Total	1777.364	249				

*significant at 5%, **Significant at 1%

H2 : The influencing factors of emotional intelligence have a positive relationship when compared with Gender.

Table – 8:- The influencing factors of emotional intelligence have a positive relationship when compared with Gender

Source of variance		Sum of Squares	Df	Mean Square	F	Sig.	Result
Defensive position	Between Groups	.109	1	.109	.016	.898	NS
	Within Groups	1643.827	248	6.628			
	Total	1643.936	249				
Get rid of position	Between Groups	.295	1	.295	.050	.824	NS
	Within Groups	1469.021	248	5.923			
	Total	1469.316	249				
Get away position	Between Groups	32.989	1	32.989	4.807	.029*	S
	Within Groups	1701.895	248	6.862			
	Total	1734.884	249				
Get on with position	Between Groups	36.961	1	36.961	5.267	.023*	S
	Within Groups	1740.403	248	7.018			
	Total	1777.364	249				

*significant at 5%, **Significant at 1%

H3 : The influencing factors of emotional intelligence have a positive relationship when compared with Working experience.

Table – 9:- The influencing factors of emotional intelligence have a positive relationship when compared with Working experience

Source of variance		Sum of Squares	Df	Mean Square	F	Sig.	Result
Defensive position	Between Groups	126.067	4	31.517	5.087	.001**	S
	Within Groups	1517.869	245	6.195			
	Total	1643.936	249				
Get rid of position	Between Groups	23.218	4	5.805	.983	.417	NS
	Within Groups	1446.098	245	5.902			
	Total	1469.316	249				
Get away position	Between Groups	379.379	4	94.845	17.143	.000**	S
	Within Groups	1355.505	245	5.533			
	Total	1734.884	249				
Get on with position	Between Groups	569.189	4	142.297	28.856	.000**	S
	Within Groups	1208.175	245	4.931			
	Total	1777.364	249				

*significant at 5%, **Significant at 1%

H4 : The influencing factors of emotional intelligence have a positive relationship when compared with Marital status.

Table – 10:- The influencing factors of emotional intelligence have a positive relationship when compared with Marital status.

Source of variance		Sum Squares	Df	Mean Square	F	Sig.	Result
Defensive position	Between Groups	15.122	3	5.041	.761	.517	NS
	Within Groups	1628.814	246	6.621			
	Total	1643.936	249				
Get rid of position	Between Groups	27.181	3	9.060	1.546	.203	NS
	Within Groups	1442.135	246	5.862			
	Total	1469.316	249				
Get away position	Between Groups	36.349	3	12.116	1.755	.156	NS
	Within Groups	1698.535	246	6.905			
	Total	1734.884	249				
Get on with position	Between Groups	54.110	3	18.037	2.575	.055*	S
	Within Groups	1723.254	246	7.005			
	Total	1777.364	249				

*significant at 5%, **Significant at 1%

Findings:-

- 53.2 percent of the teachers were female and a remaining 46.8 percent of the teachers were Male. It is understood that the Female respondents were higher in the B- school appointments.
- 28.4 percent of the teachers age ranges from 25 – 30 years, 22.8 percent of the respondents age ranges between 30 – 35 years, 21.6 percent of the respondents age ranges between 35 – 40 years, 14.4 percent of the respondents age ranges between 20 – 25 years, and a least of 12.8 percent of the respondents age was above 40 years.
- 40.8 percent of the respondents were married, 22.8 percent of the respondents were Divorcees, 20.8 percent of them were Widow, and a least of 15.6 percent of them were Single. It is well understood from the table that a 40.8 percent of the respondents were married.
- 28.8 percent of the respondents have an experience of 5- 10 years, 21.2 percent of the respondents have an experience of 10 -15 years, 20.8 percent of them have an experience of 15 – 20 years, 16 percent of the respondents have an experience of above 20 years, 13.2 percent of the respondents have an experience of less than 5 years.
- The agreeability towards the statements on They often seem just not motivated to join in, There always seem to be learners who are simply lazy, It is really like casting pearls before swine, No matter how much effort I put in there will always be some who are just not going to get it, indicates that the teaching professionals spends time and on the idea on **“Get rid of Position”**. These statements respond to the life position that “I am ok, you are not ok. A defensive position where generally see the teaching professionals as blameless but well at seeing failings in others.
- The agreeability towards the statements on, I am always worried I am going to be asked the question that will catch me out, I do worry I am going to be found out as knowing as much as they think, I often feel that I have let at least some of the learners down, I am always conscious of some members not getting much from it. Indicates that the teaching professionals spends time and on the idea on **“Get away from Position”**. These statements indicate to the life position that “I am not ok, you are ok”. Where in the teaching professionals find fault with themselves and feels that others are capable and as “Potential prosecutors”.
- The agreeability towards the statements on when teaching I see myself as making the best of a bad situation, Sometimes I wonder what is the point, I sense that nothing really changes, Sometimes I think they look as despondent as i feel, indicates that the teaching professionals spends time and on the idea on **“Get nowhere with position”**. These statements indicate to the life position that I am ok, you are ok”. And can be acknowledged as the teaching professionals are having negative view of yourself and everyone else and feel “Despair”.
- The agreeability towards the statements on I enjoy establishing a good working relationship with a group, Although I get anxious, I feel confident and relaxed in my role, When I approach a new session i expect that both they and i will enjoy ourselves and learn something useful, I enjoy corresponding to the challenges each new session brings, indicates that the teaching professionals spends time and on the idea on **“Get on with”**

position. These statements indicate to the life position that “I am ok, you are ok”. These statements link to the life position I am ok and you are ok. The teaching professionals are opening minded and tackle problems.

Results of hypothesis:-

1. The influencing factors of emotional intelligence have a positive relationship when compared with Age.
2. The influencing factors of emotional intelligence have a positive relationship when compared with Gender.
3. The influencing factors of emotional intelligence have a positive relationship when compared with Working Experience.
4. The influencing factors of emotional intelligence have a positive relationship when compared with Marital status.

Suggestions:-

1. Emotional literacy can be introduced into the standard curriculum of Faculty Development Programmes.
2. Teachers can be given Affirmation training which helps build in their self- confidence and self –esteem not only in their teaching profession but in their personality too.
3. Students can be given orientations on Emotional Intelligence before even they step in to their curriculum which may help in better co-operation with the teachers.
4. Provision of good organization climate from the institutions will induce positive emotions in teachers.
5. Relaxation and recreation programmes can be organized for the teachers.

Conclusion:-

It is found that the teachers are aware of overt behavior which helps them to identify the emotions. When they are capable of identifying the emotions then they can understand and manage their emotions. This means that they are emotionally intelligent. The study reveals that the respondents have a high level of emotional awareness. This is good sign. In their reaction they seem to be wiser and everyone wants to have healthy relationship with in defense of their position, power and other factor. And in handling grievances and problems they adopt a smooth way of expressing their feelings, redressing grievances and solving problem. The study emphasizes the importance and the value of imparting emotional intelligence through continuous Faculty Development Programs by claiming that teachers with emotional intelligence skills will be able to maintain better classroom management. Thus the study will help the teachers to identify the situations in which the negative emotions arise and help them to overcome those through various coping strategies like relaxation and recreation programmes. This will directly or indirectly create positive emotions in them.

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RESEARCH ARTICLE

AN ARDUINO BASED WSN TO CONTROL AND MONITOR THE GREENHOUSE PARAMETERS.

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Key words:-

Actuator, agriculture, Environment parameters, ESP 8266 precision Arduino based system, Wi-Fi module, soil wetness.

Abstract

Agriculture, being the backbone of India, needs huge improvement for economic growth of India. So an effective technology is needed which helps in the expansion of agriculture, productivity and profit gain. The green house and its parameters play a very important role for the production of good quality crops. In this paper an automatic control system is discussed which throws light on the monitoring and controlling of the green house ecosystem parameters. The proposed system targets on the development of a hardware which is capable of measuring temperature, humidity, soil wetness using an Arduino based system which can make decision for turning the actuators on and off for balancing the green house parameters. In present greenhouses, various points of measurement are required to track the local ambience parameters, for making the automation system of the greenhouse, work properly. Wired system will make the system expensive. Moreover, the location sensor measurement nodes are hard to change once they are positioned. So, a Wireless Sensor Network (WSN) having wireless nodes with sensor on an Arduino based system with a communication device is a very attractive and economic option for building the require measurement system.

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Introduction:-

The greenhouse vegetable production is managing the growing environment to produce an ideal ecosystem for the crop. Agriculture is the worldwide prime occupation of human beings. A greenhouse farmer cannot detect the level of greenhouse parameter accurately, they only know the parameters manually. So they cannot produce the ideal eco system for crop. The global warming due to climate change has effect on temperature and water availability which are critical for crop production. The climate change will increase temperature, reduce the water availability for irrigation and other domestic uses. Plants need water for growth and cooling. Plants extract water from the bottom of the soil and transport the water to the plant leaves. Small apertures (stomata) located on the upper and lower surfaces of the leaves allow for the intake of CO₂ required for photosynthesis and plant growth. Water vapor is gone from the plant leaves by evaporation in the stomatal cavity and the flow of the vaporized water through the stomata into the atmosphere. A considerable amount of energy is required to dissolve the water in the stomatal cavity. If the water did not evaporate, the energy would be used to heat the plant. Without transpiration, plants could reach lethal temperatures. So, good irrigation water management improves crop quality. Humidity, or air moisture content, may also play a role. High humidity can create high temperatures and heavy dew at night, which can be beneficial in reducing moisture stress, but which can create the development of certain diseases, such as leaf rust and leaf spots,

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on some crops. A system proposed in this paper helps to monitor and control the microclimate parameters of a Greenhouse atmosphere. Use of this technique, off season production of crops is also possible. Control on weather condition like temperature, humidity, soil moisture can improve the crop production & quality both [1]. Precision irrigation [2] is one of the most important practice maximizing the productivity while saving water.

Wireless Sensor Network (WSN) is a useful part of the automation system in modern greenhouses [3, 4]. Wireless communication system can be used for the collection of measurement values and for communication between the central control system and the actuators situated at various parts of the greenhouse. Compared to the wired systems, the installation of WSN is fast, cheap and easy. Moreover, it is easy to relocate when needed by just moving sensor nodes from one location to another within a range of the operating device. If the greenhouse flora is high and dense, the small and light weight nodes can be held up to the plants' branches. WSN maintenance is also relatively cheap and easy. The only extra costs occur when the sensor nodes run out of batteries life and the batteries need to be charged or replaced, but the ability to give power supply can be several years if an efficient power saving algorithm is applied. Related work in this field is discussed in next section and third section gives an outline of the proposed system.

Related Work:-

Nagesh K.D.N. proposed an ARM based remote monitoring and Control System for environmental parameters in greenhouse [5]. They discussed about Greenhouse environment parameter monitoring and control system based on GSM communication using ARM7 microcontroller. It will regulate the environmental parameters in every greenhouse automatically.

Ahonen, T. Virrankoski, R.; Elmusrati, M., have discussed on Greenhouse Monitoring with Wireless Sensor Network [6]. In this article greenhouse monitoring, control and sensor network feasibility and reliability is given. Tiantian Guo, Weizhu Zhong, presented the new structure integrated data acquisition system and intelligent control system on agricultural facilities [7], in order to promote the information-based and intelligent level of comprehensive agricultural zone and to improve the production efficiency and effectiveness.

Purna Prakash Dondapati proposed an Automated Multi Sensor Green House Management [8], which gives the explanation on how to overcome the disadvantages in the normal cultivation without any observation of human being. It also explains the effective working of sensors which help the project to become automated to yield more useful results in cultivation.

Hybrid wired/wireless approach for green house was given by Orazio Mirabella and Michele Brischett; where Controller Area Network & ZigBee protocol are used to find its flexibility and ability to operate in different environments [9].

Vijay S. Kale and Rohit Kulkarni have discussed on "Real Time Remote Temperature & Humidity Monitoring Using Arduino and Xbee S2" [10]. An Arduino Uno based system that measures and monitors the temperature and relative humidity of a remote place and passes data through network to the main location that the base location.

Proposed System:-

Greenhouse atmosphere monitoring and controlling system is used to measure the various parameters like temperature, humidity, light, soil moisture and air moisture. Sensor is a device that receives and responds to a signal and generates corresponding electrical signal. It is an element that senses a variation in input energy to produce a variation in another or same signal.

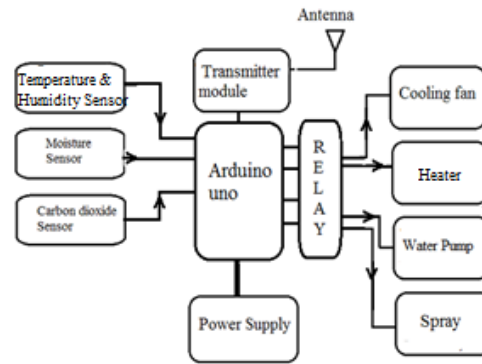


Figure 1:- Functional block diagram

The block diagram of the system is shown in Fig.1 which is composed of remote sensor nodes and the sensor nodes are implemented with an ESP 8266 Wi-Fi module and analog sensors. These radios support ESP 8266 topologies which are configured to read analog signals directly from sensors and generate digital data to be transmitted within a data packet. Each node is enabled to read temperature, humidity, moisturizer, CO₂ level, light level. The measured data are sent periodically to the central computer.

Arduino:-

In this project we use Arduino Uno [11] which has ATmega328P i.e. 32 bits AVR microcontroller uses power supply with a regulated voltage ranging from 5-9 volts with 14 digital input output pins, 6 analog inputs that may be interface with the various expansion boards and also with the other circuit.

Arduino Uno has 16MHz crystal oscillator and a USB serial communications port. This is an open source hardware. The microcontrollers are programmed using the programming language C and C++. And also the Arduino provides an integrated development environment (IDE) which is cross platform application written in Java, allows users to write program for Arduino in C or C++.



Figure 2:- Arduino Uno

Temperature-Humidity Sensor:-

The DHT11 is temperature and humidity sensor with digital signal output. This sensor is relatively a cheap sensor for any kind of project. The digital output of the sensor gives accuracy and it ensures highly reliable and long term stable system. This module includes resistive type humidity measurement subpart and temperature measurement subpart and offers excellent quality with fast response. It is small in size and low power consumption.



Figure 3:- DHT11 sensor

DHT11 works on 5V and gives the both output i.e. temperature and humidity in the clocked out values in handshaking mode. Its working temperature range is from 0 to +50 degree Celsius with accuracy of +/- 2 degree Celsius and humidity range 20 -95% RH with +/- 5.0% RH humidity accuracy and response time is less than 5 sec. DHT11 has four lines: GND, +5V, NC and a single data line.

Soil moisture sensor:-

Water is needed for the basic growth of plants. When enough quantity of water is not present at the time of plant desires, then eventually the plant can prompt lessened quality or dying.

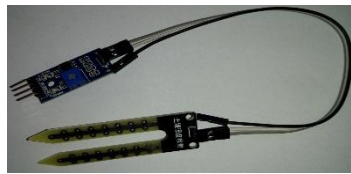


Figure 4:- Soil moisture sensor

Since it is very agitated for human to appear when plants all the time, engineers designed soil moisture sensors to reduce the burden. Now using the sensing element system designer will build any sorts of system that may take care of the water desires of plant.

Carbon dioxide sensor:-

To gardening the green house we might control carbon dioxide that critical to plant growth and development. Photosynthesis, the process through which plants use to create food, requires CO_2 . So, at the time of plants growing in greenhouse, the CO_2 levels will be reduced as the plant use it up during photosynthesis. Increase the CO_2 levels in this environment is essential for good results. Here we use the K30 CO_2 level measuring sensor.

Transmitter/Receiver:-

For the transmission of the data from node to base station and to receive the data here we use ESP8266 Wi-Fi SOC module which is low cost with full TCP/IP stack and microcontroller unit capable. This module has on-board processing and storage capability and its on-chip integration allows us to minimal external circuitry. It has 32 bit RISC processor with 64 KB of instruction RAM and 96 KB of data RAM, follow IEEE 802.11 b/g/n Wi-Fi. It has more features like integrated PLLs, power management units. +19.5dBm power output in 802.11b mode, flash memory of 1MB, wake up and transmit packets in less than 2ms and standby power consumption is less than 1mW. Figure-5 shows the control station which consists of the same ESP 8266 module as a coordinator connected base station PC with RS232 serial communication.

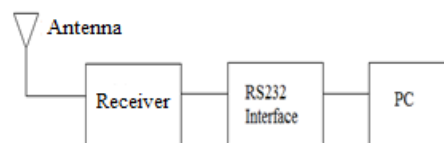


Figure 5:- Block diagram of components in base station.

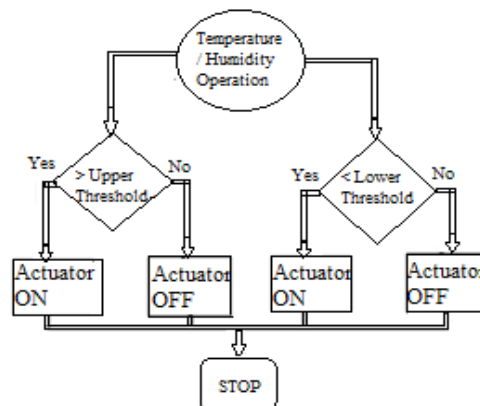


Figure 6:- Flowchart of Processing Data.

A flowchart of the proposed automatic atmosphere temperature, humidity and soil moisture control system is shown in figure-7. Each node calculates these parameters and compare with the preset values. The process flow of this automatic control system is as follows: Initialize the system and read the sensors data. Next store these data. After storing these data, first compare the temperature data with preset value, if the value crosses the upper threshold then cooling actuator will be turned on, if the temperature is less than lower threshold then heating actuator will be turned on till the surrounding temperature maintained at the required level. Similarly perform and control the humidity which is shown in figure-6. The current sensors values are send to the base station and permanently stored. And halt the process for a minute and repeat again and continue.

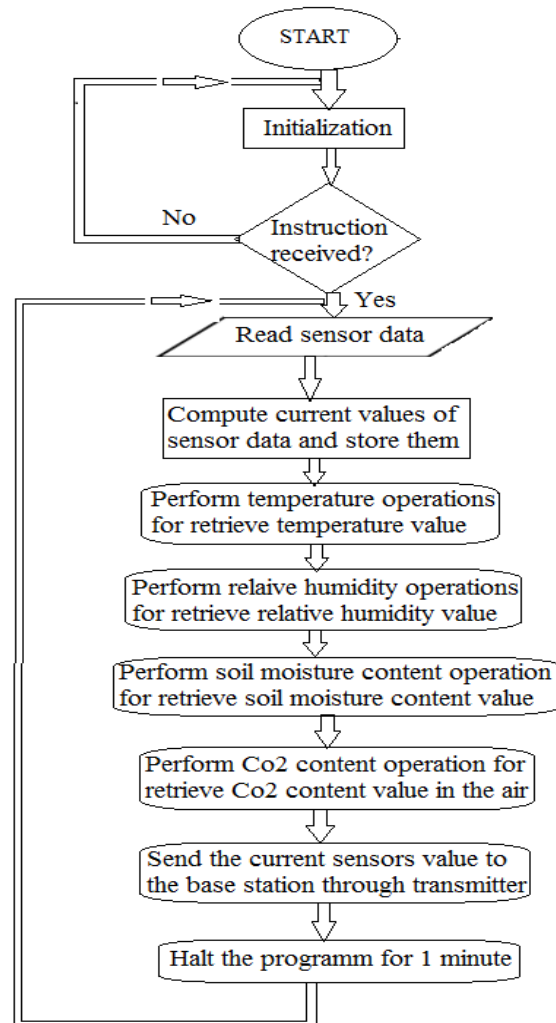


Figure 7:- flowchart of Processing Data

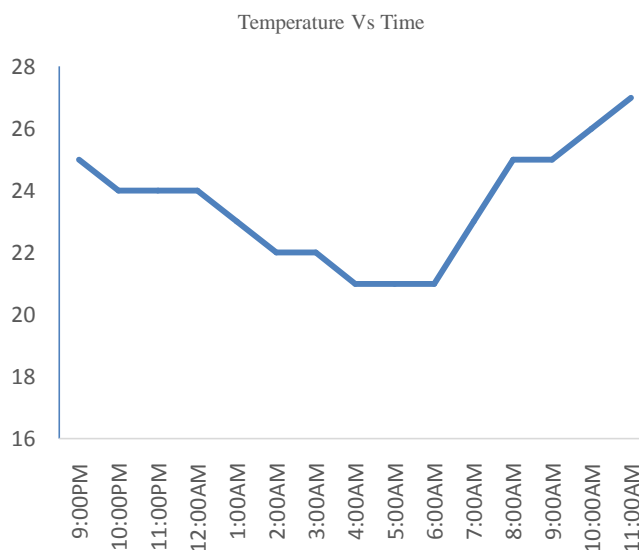
Result and Discussion:-

Recording of variation of temperature and humidity along with time during February and March is given in table 1. Field observations for various crops have been discussed in past by some researchers[12, 13]. Optimum temperature range is very important for agriculture and is to be maintained for maximum dry matter accumulation. It is well known that for wheat and some other important crops the optimum temperature required is 25 °C. We set the upper threshold value of temperature to be 27°C and lower threshold value to be 22°C. As discussed above the actuators are controlled by the threshold value of the temperature and maintain the required optimum temperature.

Table 1:- Observations for the crop in duration February- March

Time	Temperature(°C)	Humidity(RH)
9:00PM	25	34%
10:00PM	24	37%
11:00PM	24	39%
12:00AM	24	40%
1:00AM	23	41%
2:00AM	22	44%
3:00AM	22	44%
4:00AM	21	46%
5:00AM	21	45%
6:00AM	21	43%
7:00AM	23	41%
8:00AM	25	39%
9:00AM	25	39%
10:00AM	26	38%
11:00AM	27	36%

The graphical representation of variation between temperature and time with the controlling action of actuator is shown in figure 8 whereas figure 9 relates the variation of relative humidity at that temperature. We can notice the maximum temperature of 27°C and minimum humidity at 11 am morning. Cooling fan turns on at this temperature till it reaches 25°C i.e. the optimum temperature required. Temperature dip and humidity peak can be seen between 4 am to 6 am in the morning which is maintained to the required level by the controlling action of heater. Heater turns on as soon as the lower cutoff is reached at 2 am night. Heater is turned off, once the temperature returns to its optimal value.

**Figure 8:-** Temperature variation curve

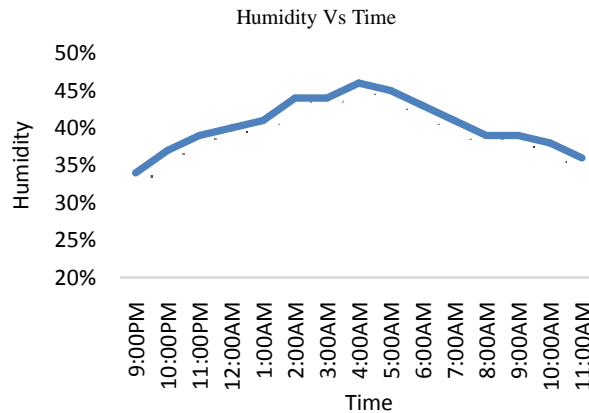


Figure 9:- Humidity variation curve

Future Aspect:-

The proposed implementation of ESP 8266 based WSN for greenhouse parameters monitoring system offers less energy consumption and long battery life is presented. The use of high power WSN is good for various works in industries such as manufacturing, constructing and also can be used to monitor pollution level of a city. Another important fact about the system is the easy installation because of wireless mobility where the database center can be positioned at the local center near the target area and the monitoring task can be done and can generate alarm message to user mobile using GSM/GPRS technology.

Conclusion:-

In future various parameters can be controlled as required by the huge variety of crops. The varieties of crop are increasing day by day because of the development in agriculture technology. In this situation, the wireless sensor network and additional hardware with software is a better solution for green house control. This flexibility in precision agriculture through Wireless Sensor Network in greenhouse control is very useful.

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RESEARCH ARTICLE

SCREENING OF INFERTILE FEMALES FOR VAGINAL CANDIDIASIS.

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Infertility, Lower genital tract infection, *Candida* spp., Fluconazole resistance

Abstract

Introduction: Infertility is a major public health problem across worldwide and in India. As per WHO, 60 to 80 million couples worldwide currently suffer from infertility. The role of female reproductive tract infection, symptomatic or asymptomatic, is well recognized. Studies reveal that *Candida albicans* have sperm-immobilizing property which might lead to infertility. Keeping this in mind the present study was done. **Aim:** To screen infertile females for vaginal candidiasis. **Material & Methods:** 100 infertile women were screened for vaginal candidiasis. High Vaginal swab collected from them were processed for fungal culture using conventional methods. Anti-Fungal susceptibility testing was done for all the isolates of *Candida* spp. as per guidelines. **Results:** 17% of the patients suffered from vaginal candidiasis. *Candida albicans* followed by *Candida tropicalis* were the most common isolates. None of the isolates were resistant to Amphotericin B and Nystatin while maximum resistance was shown for Fluconazole. **Conclusion:** There was more predominance of non albicans *Candida* in the study thereby showing a change in causative agent of vaginal candidiasis. Moreover since studies show sperm-immobilising property of *Candida* spp., infertile women should be routinely screened for vaginal candidiasis.

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Introduction:-

Infertility is a major public health problem across worldwide and in India. World Health Organization (WHO) defines infertility as “the inability of a sexually active, non-conceiving couple to achieve pregnancy in one year” (WHO laboratory manual for the examination and processing of human semen, 2010). As per WHO, 60 to 80 million couples worldwide currently suffer from infertility (Infecundity, infertility and childlessness in developing countries. Demographic and Health Surveys (DHS) Comparative reports No. 9, 2004).

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Besides other causes of female infertility, the role of female reproductive tract infection is well recognized. Infections may be in the form of Pelvic Inflammatory diseases (PID) or lower genital tract infection, either of which can be asymptomatic or symptomatic. These infections need to be diagnosed and treated properly to set the patient free from the grasp of infertility. *Candida* infection plays an important role in this regard. As per studies, *Candida albicans* have sperm-immobilizing property (Harpreet & Vijay, 2015; and Tian et al., 2007). When the sperm-immobilizing strains of *Candida albicans* were intravaginally inoculated into female mice, it showed 100% decrease in fertility in all groups as compared with control mice. Moreover, since no clinical or histopathological changes were observed in the reproductive organs, colonization of the genital tract with sperm-impairing micro-organisms could be the reason behind female infertility (Harpreet & Vijay, 2015). As per Mathur et al. (Mathur et al., 1980), patients with chronic vaginal candidiasis of at least 5 years duration showed high titres of auto-antibodies to ovary which may be due to the presence of one or more cross-reactive antigens on ovarian follicle and *Candida* spp. Thus chronic vaginal candidiasis may be associated with decreased ovarian function, thereby affecting the fertility outcome.

In view of this, our study was carried out to evaluate vaginal candidiasis in infertile females.

Material and Methods:-

It was a cross sectional study carried out for a period of one year (2014-2015) in a Tertiary care teaching hospital in Western India. The study was approved by the institutional ethics committee. A total of 100 married reproductive age group females (18 – 49 years) attending infertility clinic, with the exception of those receiving antibiotic and antifungal therapy in previous one month, were included in the study. A questionnaire covering demographic data, menstrual history, contraceptive use, recent antibiotic use, medical history & history of vaginitis and infertility was completed for each of the participants. After taking consent, High Vaginal swab (HVS) was collected under speculum examination using sterile swab.

KOH (10%) mount was prepared to detect presence of *Candida*. A gram stained smear was examined for the composition of bacterial morphology and presence of yeasts and leukocytes. All the specimens were inoculated on Sabouraud Dextrose Agar (SDA) which was incubated at 37°C. In case of growth, discrete colonies were picked and processed further for identification using conventional methods like Germ Tube Test (GTT), Chrome agar, Corn Meal Morphology.

Antifungal susceptibility testing was done by disc diffusion method as per guidelines (NCCLS May 2004). Antifungal agents used for susceptibility testing (HIMEDIA Hexa Antimyc-01) are mentioned in Table 1. Antifungal susceptibility pattern was interpreted as per NCCLS (NCCLS May 2004) and manufacturer's guidelines.

Table 1:- Antifungals used for susceptibility testing

Sr. No.	Antifungal agent	Dose
1.	Fluconazole	25 µg
2.	Ketoconazole	10 µg
3.	Itraconazole	10 µg
4.	Nystatin	100 units
5.	Amphotericin B	100 units
6.	Clotrimazole	10 µg

Results:-

The various findings of our study were analysed as follows. Out of 100 patients, *Candida* spp. was isolated from 17 of them. Among the fungal isolates of our study, *Candida albicans* (35.3%) was the most predominant fungal isolate followed by *Candida tropicalis* (29.4%). The distribution of various *Candida* spp. was as shown in Fig 1. The resistance pattern of the isolates was as shown in Fig 2.

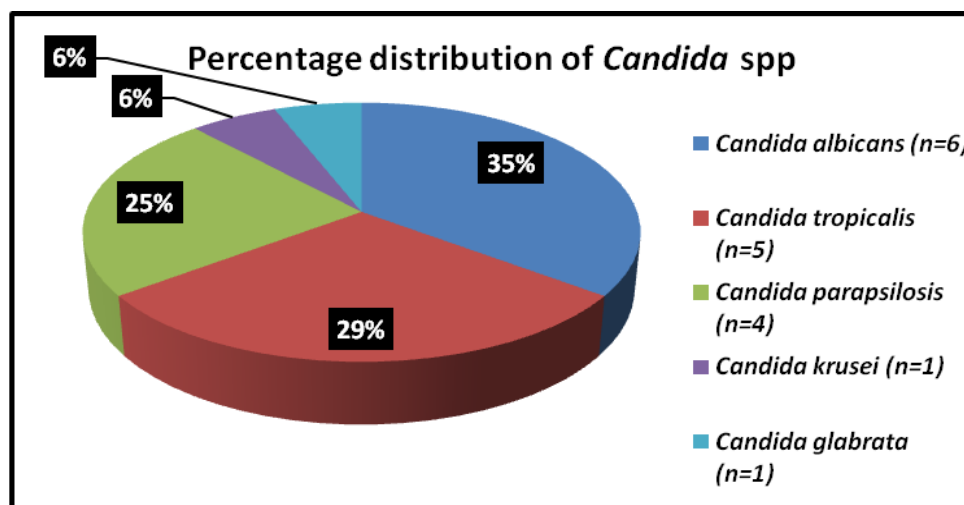
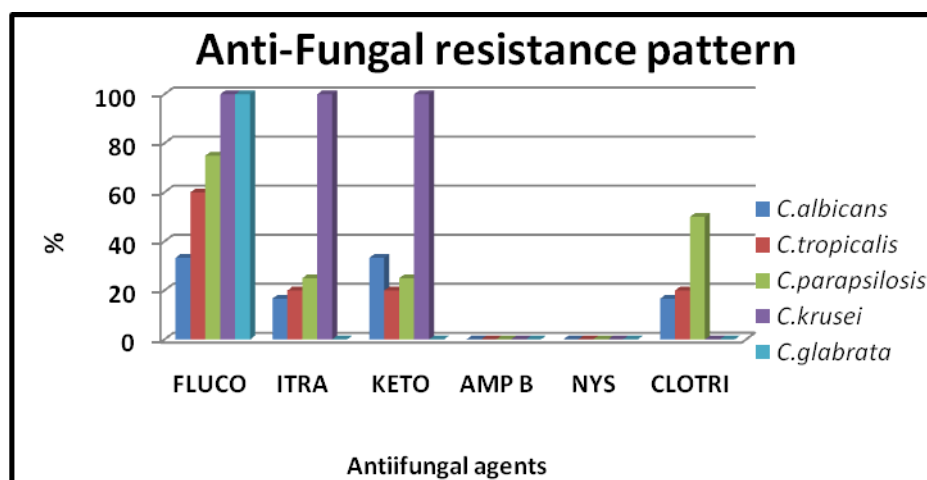
Fig 1:- % distribution of *Candida* spp

Fig 2:- Anti-fungal resistance pattern

Discussion:-

In our study, *Candida* spp was isolated from 17% of the cases which is bit higher than that reported by Rodriguez et al. (Rodriguez et al., 2001). Ghiasi et al. (Ghiasi et al., 2014) and Gupta et al. (Gupta et al., 2014) had shown prevalence of *Candida* spp. as low as 1.4% and 5.9% respectively. Unlike our study, other studies showed prevalence of *Candida* spp. as high as 22%, 25%, 30.8% and 43.5% (Saleh, 2010; Okonofua et al., 1995; Paul et al., 2011; Ogiogwa et al., 2012).

Candida albicans (35.3%) was the most common fungal isolate which is similar to results of the study done by Verghese et al. (Verghese et al., 2001). In our study, *Candida tropicalis* (29.4%) was the second most predominant fungal isolate, unlike Verghese et al. (Verghese et al., 2001) where the second most common isolate was *Candida glabrata*. *Candida krusei* constituted 5.9% of the fungal isolates in our study which is similar to Verghese et al. (Verghese et al., 2001).

Broadly speaking, none of *Candida* spp. in our study showed resistance to Amphotericin B and Nystatin. Maximum resistance was shown for Fluconazole followed by Ketoconazole, Itraconazole and Clotrimazole. Unlike our study, Fluconazole was the drug of choice for vaginal candidiasis in another study (Khan & Rakshanda, 2010). As per Gandhi et al. (Gandhi et al., 2015), antifungal susceptibility pattern showed that *Candida* isolates were more sensitive to Amphotericin-B (98.36%) and Nystatin (100%), compared to that of Clotrimazole (79.5%) and Azoles like Fluconazole (79%), Itraconazole (52%) and Ketoconazole (76%). As per Verghese et al. (Verghese et al., 2001),

16.7% *Candida* strains showed resistance to Fluconazole, 45.23% of the strains showed resistance to Itraconazole, 9.5% strains showed resistance to Nystatin while there was no resistance to Amphotericin B.

A comparative analysis of the resistance pattern of *Candida* species isolated in our study, for Fluconazole, Itraconazole, Ketoconazole and Clotrimazole are given Table 2-5 respectively.

Table 2:- Comparison of Resistance pattern (%) for Fluconazole

<i>Candida</i> species	Our study	Gandhi et al., 2015	Adeshji et al., 2010	Sachin & Santosh, 2013	Salehei et al., 2012
<i>C. albicans</i>	33.33	17	84.6	-	81.13
<i>C. tropicalis</i>	60	17	66.7	29.5	100
<i>C. parapsilosis</i>	75	0	-	10	-
<i>C. krusei</i>	100	33	100	18.8	100
<i>C. glabrata</i>	100	15.70	50	27.3	100

Table 3:- Comparison of Resistance pattern (%) for Itraconazole

<i>Candida</i> species	Our study	Gandhi et al., 2015	Sachin & Santosh, 2013	Salehei et al., 2012
<i>C. albicans</i>	16.67	13.50	-	15.09
<i>C. tropicalis</i>	20	8.34	9.8	0
<i>C. parapsilosis</i>	25	16.66	10	-
<i>C. krusei</i>	100	33.33	21.9	0
<i>C. glabrata</i>	0	15.78	12.7	0

Table 4:- Comparison of Resistance pattern (%) for Ketoconazole

<i>Candida</i> species	Our study	Gandhi et al., 2015	Sachin & Santosh, 2013	Salehei et al., 2012
<i>C. albicans</i>	33.33	11	-	43.39
<i>C. tropicalis</i>	20	17	39.4	25
<i>C. parapsilosis</i>	25	0	10	-
<i>C. krusei</i>	100	33.5	25	0
<i>C. glabrata</i>	0	5	13.7	12.25

Table 5:- Comparison of Resistance pattern (%) for Clotrimazole

<i>Candida</i> species	Our study	Gandhi et al., 2015	Ajitha & Maimoona, 2014	Salehei et al., 2012
<i>C. albicans</i>	16.67	9	20	1.88
<i>C. tropicalis</i>	20	17	45.5	50
<i>C. parapsilosis</i>	50	0	50	-
<i>C. krusei</i>	0	67	-	0
<i>C. glabrata</i>	0	5	33.3	0

Conclusion:-

There was more predominance of non albicans *Candida* in our study thereby showing a change in causative agent of vaginal candidiasis. *Candida* spp. isolated in our study were least resistant to Amphotericin B and Nystatin while showed maximum resistance to Fluconazole. As per reports from other studies, *Candida albicans* plays an important role in female infertility; hence infertile females should be routinely screened for vaginal candidiasis.

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RESEARCH ARTICLE

THE PERSPECTIVE OF FIRST YEAR DENTAL STUDENTS TO CASE-BASED LEARNING AS AN EFFECTIVE TOOL IN LEARNING PHYSIOLOGY: A CASE STUDY.

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Case-based learning, PBL, Small group teaching, personal skills, professionalism

Abstract

Background: Understanding and recall of human physiology is much easier when it is linked to the real life patient cases. Case-based learning (CBL) is a teaching method where the students are motivated towards self-directed learning and to develop analytic and problem-solving skills.

Aim: The present study aimed to measure dental students' perceptions of introducing small group case-based learning in teaching cardiorespiratory physiology.

Methodology: Data were collected from semester one dental students at MAHSA University, Malaysia. Cardiorespiratory case-based questions were given to the students after finishing their didactic lectures of cardiovascular and respiratory physiology. 74 students were divided into 9 groups to learn and discuss the case. At the end of the session, a 23 items perception scale was given to the students to measure their acceptance of CBL, impact of CBL on personal skills and impact of CBL on professionalism.

Results: The results showed that the students enjoyed CBL and reported that CBL improved their personal skills and professionalism. There was a positive correlation between CBL acceptance and impact of CBL on personal skills as well as professionalism. The study showed a significant difference in between the small groups in their acceptance of CBL and their opinions on the impact of CBL on the professionalism with insignificant difference in their opinions about the impact of CBL on their personal skills.

In conclusion: CBL helped in understanding and integrating cardiovascular and respiratory physiology, improved the student's personal skills and professionalism.

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Introduction:-

Human physiology is seen as an important core component of any medical curriculum [1]. The main objective of the physiology teacher is to make sure that the subject of physiology is easily understood by diverse groups of students belonging to different courses [2]. Learning and remembering human physiology is much easier when you can link it to real life patient cases. A genuine feel for how essential it is to clinical practice is thereby obtained which makes it not only interesting but also understandable. From this point of view emerged the importance of the new learning

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strategies such as Problem-Based-Learning (PBL) [3] and Case-Based-Learning (CBL) [4] in teaching physiology for medical students. Conventionally, teaching of undergraduate students is done with the help of didactic lectures, practical's, tutorials, and clinics, which are mostly used as passive teaching and learning methods and mostly they lack in the development of problem solving or reasoning skills of the students. Furthermore, there is hardly any involvement of students in the teaching-learning process [4].

MAHSA University college dental program started in 2007 uses a traditional curriculum where Anatomy, Physiology and Biochemistry are taught along with dental anatomy in the first year of a 5 year program. The teaching learning modalities followed are didactic lectures, tutorials, question based discussions and practical's- both hands on and demonstration. Since this is a traditional subject based curriculum, introducing problem based learning seemed to be difficult with students in their first semester. The physiology department decided to introduce CBL instead of PBL to stress on physiological principles and facilitate proper learning amongst the students.

CBL is a teaching method where students are motivated to learn on their own so as to inculcate the habit of self-learning and integrating knowledge from different subjects [5]. CBL is similar to PBL in the sense that both of the methods are based on the use of a case, problem, or inquiry to stimulate and motivate the students to acquire knowledge [6]. However CBL is different from PBL in the following: first, CBL uses a guided inquiry method in which the facilitator plays an active role however PBL uses an open inquiry method in which the facilitator plays a passive role, second, in CBL, the students are already had the knowledge and CBL motivates them through learning the application of their knowledge, however in PBL the students are motivated to identify what they need to know and learn themselves [7].

The objective of this study was to assess the attitude of the dental students in their first semester towards using an active learning strategy in the form of case based learning and whether it was fruitful in improving their knowledge, skills and attitude.

Methods:-

Design and Procedure:-

This is an observational retrospective study involving 74 students who were admitted in the Doctor of Dental Surgery (DDS) program in 2013.

Institutional Setting:-

MAHSA University College, a relatively new private institution in Kuala Lumpur, Malaysia offers a 5 year Doctor of Dental Surgery [DDS] course. The curriculum requires basic sciences namely Anatomy, Physiology, Biochemistry along with Dental Anatomy to be taught in the first year, followed by preclinical subjects of Microbiology, Pharmacology, Pathology in the second year and subsequently three years of clinical training.

Study format:-

The study was conducted among year one DDS students at MAHSA University College. After students had finished the didactic lectures, tutorials, practical's of the physiology of cardiovascular and respiratory systems, a cardiorespiratory case (Appendix 1) and physiology based questions on the case were prepared by a team of physiologists in the physiology department. The case was formulated in a brief format and framed in a way that matches the student's acquired knowledge that they have been taught in their didactic lecture classes. The questions were discussed in a systematic way and each member of the group had a chance to participate [8].

The 74 students were divided into 9 groups each group contained 8 students and the whole groups were facilitated by two facilitators from the physiology department. The students were given the important instructions of how to handle the CBL session (read the case & questions, discussing on between themselves, using their learning resources and keep their findings ready to be presented at the end of the session).

The role of facilitators was to guide the students, discuss with them the answers, clarifying any doubt and remained as facilitators and not instructors. The total time for supervision was 60 min, and 45 min was the wrap-up discussion presented by all groups; 5 minutes for each group. The facilitators divided the questions in between the groups and asked each group to present their answers for one to two questions.

At the end of the session, the students were given a questionnaire (Appendix 2) to check for their acceptance of the CBL and the ability of this TLA in helping them to improve their knowledge & understanding of the cardiorespiratory physiology, improving their cooperativeness, communication skills and presentation skills.

Research material and Statistics:-

A 23 items- questionnaire was given to the students after the intervention with a response scale of 5 points rated from (strongly disagree, disagree, neither agree nor disagree, agree, strongly agree) were framed in such a way to cover the opinions of students regarding;

- Acceptance of CBL.
- Impact of CBL on their interpersonal, communication and oral presentation skills.
- Impact of CBL on their attitude and professionalism.

The data was analyzed through the Social Sciences (SPSS) program in its version 22 using descriptive statistics for the means and standard deviation for the responses for each item. Statistical significance was decided whenever $p < 0.05$. Simple regression analysis test was used to determine the relationship between the three factors of the CBL scale, Tukey's range test in conjunction with ANOVA were used to find any significant differences in means of the responses of the different groups.

Reliability and factor analysis of the questionnaire- items:-

Reliability which is the best single measure of test accuracy is the extent to which test results are consistent, stable, and free of error variance. It is the extent to which a test provides the same ranking of students when it is readministered, and it is measured by Cronbach's α . Factor analysis, which is the first step in the calculation of α -coefficient, is a data reduction technique used to group various items having homogeneity/unidimensionality [9]. This reduced the twenty three items into three meaningful factorial groups, which were grouped as F1, F2, and F3 under the following headings: acceptance of CBL F1 items 1- 11, perception of the impact of CBL on interpersonal, communication and oral presentation skills F2 items 12- 18, and perception of the impact of CBL on their attitude and professionalism F3 items 19-23. Spearman's rank correlation was calculated to check the interrelation of each item's score with the total score (item interrelatedness) [10]. Cronbach's α reliability coefficient was then calculated. The value normally ranges between 0 and 1. The closer Cronbach's α coefficient to 1.0, the greater the internal consistency of the items in the scale. Interpretation of the value varies according to the number of items. Reliabilities as low as 0.50 are satisfactory for short tests of 10–15 items, but tests with >50 items should have reliabilities of 0.80 or higher. If reliability is <0.8 , a single test score should not be used to make important decisions about individuals [11]. The results for factor analysis of the items are shown in Table 1.

Results:-

The results of the study showed high acceptance of CBL among first year dental students (M of 3.37 and SD of 0.66) and the mostly appreciated was that CBL helped the students to demonstrate constructive and critical thinking process (M of 3.91 and SD of 0.78). The results showed a positive impact of CBL on the skills (Mean of 3.62 with SD of 0.65) especially small group working dynamics (M of 3.79 and SD of 0.83), then searching skills-locating and finding information skills (M of 3.69 and SD of 0.86). Also the figure showed a positive impact of CBL on the student's professionalism (M of 4.07 with SD of 0.75) especially punctuality and attending the discussion (M of 4.19 and SD of 0.93) followed by respect of other students opinions (M of 4.14 and SD of 0.78) (Fig. 1).

The results showed a significant difference between the groups in their acceptance of CBL ($p = 0.020$) and the impact of CBL on attitude & professionalism ($p = 0.021$) done by ANOVA test (Fig. 2). Both were higher in group 6 (TukeyHSD^{a,b} test) with insignificant difference in the impact of CBL on the personal skills ($p = 0.543$). ANOVA also showed insignificant differences within the groups in CBL acceptance, impact of CBL on personal skills and impact of CBL on professionalism (Fig. 2).

CBL acceptance was found to be positively correlated with the impact of CBL on personal skills and professionalism, p -value = 0.00. The simple regression test between the acceptance of CBL as an independent variable and impact of CBL on personal skills, attitude and professionalism as dependent variables showed that acceptance of CBL could predict 52% change in the impact of CBL on the personal skills, adjusted; $R^2 = 0.525$ ($F = 27.458$; $p < 0.001$) and 45% of the impact of CBL on attitude and professionalism with adjusted $R^2 = 0.456$ ($F = 18.865$; $p < 0.001$) (Table. 2)

Discussion:-

Understanding of cardiorespiratory physiology is one of the important human body systems required for dentists. Depending only on a traditional way of teaching through lectures lacks the advantages of motivation, active interaction from the students, triggering of thinking and empowering them with the ability of integrating the knowledge and their application [12]. Learning human physiology through relevant cases has been shown a positive impact on understanding physiology among medical students [3] and to promote a deep learning in dental students [13]. The aim of the current study was to explore the perception of dental students to CBL and whether the method helped in understanding the knowledge about cardiorespiratory physiology and improved the ability to apply these knowledge's and to assess the perception of the students about the effectiveness of CBL in improving the personal skills and professionalism.

The results of this study showed that CBL was interesting, motivated the students to learn by themselves, and helped them in gathering knowledge and application of this knowledge in the understanding the current case. This is in agreement with Yooet *al. who* examined the effects of CBL on learning motivation in nursing students and found that it was significantly higher in the CBL group than in the non-CBL one [14]. The results showed that CBL helped students to demonstrate constructive and critical thinking process which is in line with Kaddoura who found same effect of CBL in nursing students [15]. This enforces the aim of introducing this type of teaching for dental students.

The results also showed that CBL improved the student's personal skills specially their communication skills which are known to be essential for medical as well as dental doctors [16]. In line with our findings, it has been reported that student communication skills and ability to work within a team were significantly improved due to CBL implementation in a microbiology course [17-18] and pharmacology course [19]. Our data indicated that this innovative pedagogical approach not only improved students' personal skills but also improved students' professionalism in the form of punctuality and respect of others opinions in the group.

The results of this study showed a positive correlation between the acceptances of CBL and the student's opinions about CBL impact on their personal skills and professionalism with higher correlation found with the personal skills. Although the study showed insignificant difference between the individual groups in the impact of CBL on the personal skills there was a significant difference in the acceptance of CBL and their perception about impact of CBL on the professionalism. This can be due to the different personality traits of the students which was found to affect the preference of medical students to problem based learning [20] or any other factors which affect the acceptance of the students of any teaching methods.

Conclusion:-

Teaching human physiology becomes much easier if the students understand its application in their clinical life. This can be achieved by using a relevant case- based learning which will not only help in understanding but empower the students with the critical thinking skills, problem solving skills, communication skills as well as proper attitude and professionalism.

Recommendations:-

Further studies on the effectiveness of CBL in teaching human physiology to be joined with pre and post assessment of the students, personality scales, quality of life and other factors which can affect the effectiveness of CBL as a teaching method

Appendix A: Case – Scenario:-

A 45-year-old man presented at the dental clinic with bleeding gums. After checking the dentist found the gums were healthy but his blood pressure was found to be 160/110 and was equal on both arms and legs. The dentist referred the patient to a physician for assessment. The patient was alert and cooperative but appeared to be anxious. The patient reported that he had been having frequent coughs with sputum production and feels uncomfortable while breathing, which worsen with exertion. He reported that he is a social drinker and smokes 2 packs of cigarettes a day for the past 20 years. He also indicated that he was too busy to exercise. There was a family history of heart disease but no family history of diabetes.

Physical examination showed:-

Vital signs:

Pulse	80/min
Blood pressure	160/110
Temperature	37 ⁰ C
Respiratory rate	15/min
BMI	32

Cardiovascular examination:-

ECG: normal

Heart sounds: normal

Respiratory examination:-

Wheezes were heard on both sides of the chest

The physician referred him to get a pulmonary function test done

Appendix B: Feedback questionnaire about CBL:-

Using 5-point Likert scale (Strongly disagree- Disagree- neither agree or disagree- Agree- Strongly agree)

- Acceptance of CBL:-**

- The case was interesting to me
- It helped me to demonstrate constructive and critical thinking process
- CBL helped me to develop better skills of gathering information, organizing it and storing for future use.
- I believe that CBL will make me capable of applying knowledge to new situations to solve problems and reach decisions
- CBL method has motivated me to learn
- CBL method has instilled self-learning skills in me
- I think CBL method will change my way of learning
- My overall experience with case-based-learning is satisfactory
- Though this was the first case study, I understood the pathogenesis of the condition
- Though this was the first case study, I was able to interpret the investigations done for this patient
- Though this was the first case study, I was able to understand the case and answer the questions given

- CBL and personal skills:-**

- I was able to participate in the group discussions and activities
- I liked the small group working dynamics
- I find that I am discussing with other students more than usual
- I feel CBL will help me improve my communication skills
- I think CBL will improve my oral presentation skills
- I think CBL will improve my searching skills-locating and finding information
- I think CBL will improve my skill of comprehension, express my views in my own words

- CBL and professionalism:-**

- I was punctual and attended the discussion
- While doing this case study, I showed responsibility and commitment in the group activity
- I learnt to respect other student's opinions
- CBL method has developed positive attitudes providing and accepting constructive feedback
- I became more perceptive and sensitive to the needs of others during group work-contribute to group harmony, listening to conflicting opinions, tolerating shortcomings of others.

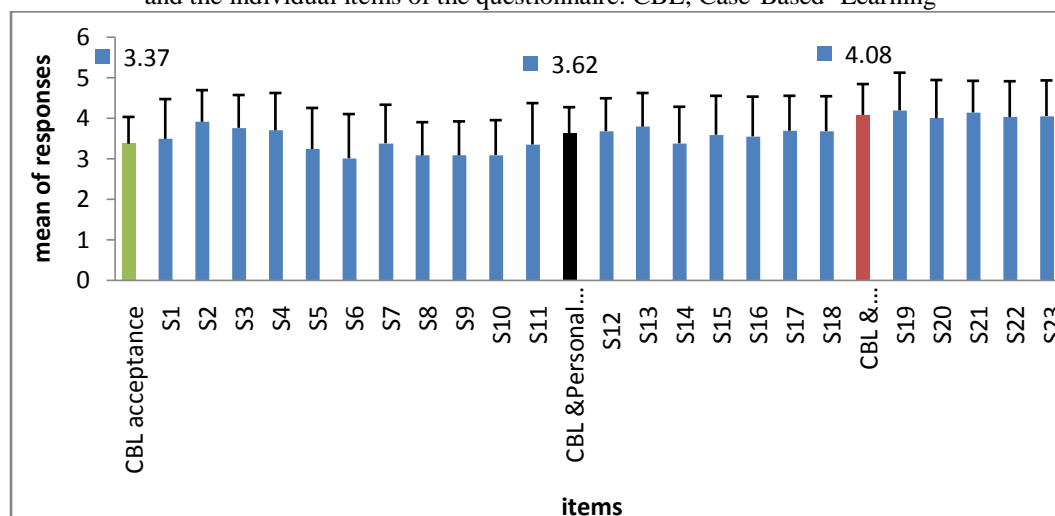
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Disclosures:-

The authors declare no conflicts of interest

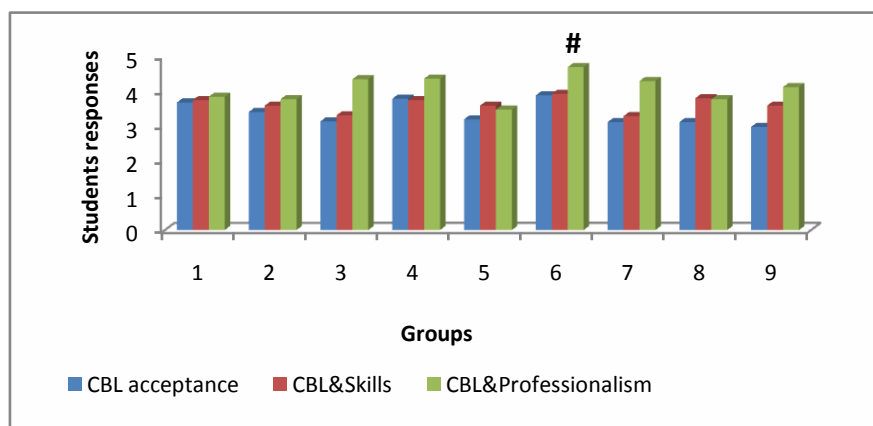
Figure 1:-Bar chart showing means with error bars of the responses of 74 students for the three factors of CBL scale and the individual items of the questionnaire. CBL, Case-Based- Learning



CBL. Case Based Learning, S1. The case was interesting to me, S2. CBL helped me to demonstrate constructive and critical thinking process, S3.CBL helped me to develop better skills of gathering information, organizing it and storing for future use., S4.I believe that CBL will make me capable of applying knowledge to new situations to solve problems and reach decisions,S5.CBL method has motivated me to learn, S6. CBL method has instilled self-learning skills in me, S7.I think CBL method will change my way of learning, S8. My overall experience with case-based-learning is satisfactory, S9.I understood the pathogenesis of the condition S10.I was able to interpret the investigations done for this patient, S11.I was able to understand the case and answer the questions given, S12.I was able to participate in the group discussions and activities, S13.I liked the small group working dynamics, S14. I find that I am discussing with other students more than usual, S15. I feel CBL will help me improve my communication skills; S16.I think CBL will improve my oral presentation skills, S17. I think CBL will improve my searching skills-locating and finding information, S18. I think CBL will improve my skill of comprehension, express my views in my own words, S19.I was punctual and attended the discussion, S20. While doing this case study, I showed responsibility and commitment in the group activity, S21. I learnt to respect other students opinions, S22.CBL method has developed positive attitudes providing and accepting constructive feedback and S23.I became more perceptive and sensitive to the needs of others during group work-contribute to group harmony, listening to conflicting opinions, tolerating shortcomings of others.

Data are presented as mean \pm SD

Figure 2:-Bar chart showing the means of the responses of individual groups of the students to CBL acceptance, impact of CBL on personal skills and impact of CBL on attitude and professionalism. CBL, Case-Based- Learning



CBL, Case-Based- Learning

Significant difference from other groups with p value < 0.05

Table 1:-Factor analysis of items related to CBL from survey*(N=74)

		F1	F2	F3
1.	The case was interesting to me	.675		
2.	It helped me to demonstrate constructive and critical thinking process	.663		
3.	CBL helped me to develop better skills of gathering information, organizing it and storing for future use.	.735		
4.	I believe that CBL will make me capable of applying knowledge to new situations to solve problems and reach decisions	.608		
5.	CBL method has motivated me to learn	.757		
6.	CBL method has instilled self-learning skills in me	.571		
7.	I think CBL method will change my way of learning	.660		
8.	My overall experience with case-based-learning is satisfactory	.663		
9.	Though this was the first case study, I understood the pathogenesis of the condition	.737		
10.	Though this was the first case study, I was able to interpret the investigations done for this patient	.768		
11.	Though this was the first case study, I was able to understand the case and answer the questions given	.506		
12.	I was able to participate in the group discussions and activities		.664	
13.	I liked the small group working dynamics		.723	
14.	I find that I am discussing with other students more than usual		.554	
15.	I feel CBL will help me improve my communication skills		.762	
16.	I think CBL will improve my oral presentation skills		.817	
17.	I think CBL will improve my searching skills-locating and finding information		.590	
18.	I think CBL will improve my skill of comprehension, express my views in my own words		.664	
19.	I was punctual and attended the discussion			.764
20.	While doing this case study, I showed responsibility and commitment in the group activity			.799
21.	I learnt to respect other students opinions			.842
22.	CBL method has developed positive attitudes providing and accepting constructive feedback			.817
23.	I became more perceptive and sensitive to the needs of others during group work-contribute to group harmony, listening to conflicting opinions, tolerating shortcomings of others.			.830
Kaiser-Meyer-Olkin Measure of Sampling Adequacy		.861	.777	.834
Sig		.000	.000	.000

*Factor structure with Principal Component Extraction with Varimax rotation: 23 items of CBL scale. F1 interpreted as CBL acceptance items emerged as the first extracted factor. F2 interpreted as CBL impact on the personal skills items was the second extracted factor. F3 interpreted as CBL impact on the professionalism and attitude items was the third extracted factor. Correlations with the latent extracted factors below 0.45 were not reported. The total scale captured 80.7% of total variance.

Table 2:-Impact of CBL on personal skills and professionalism as dependent variables in simple linear regression analysis with CBL acceptance as one block of independent variable

One block of independent variable	Personal skills			Professionalism		
	B (Standardized Coefficients)	t	P-value	B (Standardized Coefficients)	t	P-value
CBL acceptance	.525	5.479	.000	.456	5.602	.000
		5.240	.000		4.343	.000
R ²	.525 ^a			.456 ^a		
F	27.458		.000	18.865		.000
df	1			1		

CBL: Case based learning; R² is square R which means the strength of relationship between the independent and dependent variables.

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RESEARCH ARTICLE

ESTABLISHMENT OF REFERENCE INTERVAL FOR SOME BONE TURNOVER MARKERS IN HEALTHY, YOUNG, PREMENOPAUSAL EGYPTIAN FEMALES.

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Abstract

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Introduction:-

The appropriateness of Laboratory tests selection and reliability of reference interval is crucial for healthcare delivery and clinical diagnosis. This is one of the main functions of clinical laboratory and test manufacturers.¹

With the increasing life expectancy in Middle East countries, osteoporosis should be set as one of the health priorities; addressed by the International Osteoporosis Foundation (IOF). According to the Egyptian national nutrition institute, osteoporosis is a major health problem in Egypt. Among adult females, 12.2% to 15.3% have osteoporosis. This percent reaches 28.4% to third of postmenopausal women.^{2,3,4,5} Moreover, it has been calculated that more than one-quarter of females aged 10-19 years and up to 53.9% of postmenopausal were relatively osteopenic. In the 40 to 50 years age group, 42% of females have low bone mineral density.^{3,5} Besides, almost 50% suffer vitamin D deficiency.⁶

Bone remodeling is a physiological process that occurs continuously in healthy adult bone tissue with coupling of both resorption and formation.⁷ Every year around 20% of total bone tissue is replaced.⁸ This process begins in the intra uterine life and continues all through life.⁸ Bone turnover is monitored either by dynamic histomorphometry or bone turnover markers.⁹ The dynamic histomorphometry had some impracticability and imprecision,⁹ Thus the need for development of highly specific, accurate and fully automated noninvasive bone turnover markers was essentially needed.¹⁰

Bone turnover markers (BTMs) are classified as either bone formation, including peptides and enzymes secreted by osteoblasts during bone formation, or resorption markers, which are the degradation products of bone collagen or enzymes secreted by osteoclasts.¹¹ Their level in blood or urine reflect bone formation and resorption rates, respectively, this is affected by several variables either physiological or pathological.^{12,13} These changes in BTM proves the coupling of formation and resorption.

Biochemical markers of bone turnover are used in the initial assessment of osteoporosis, including the suspicion of secondary causes and the identification of patients with rapid bone loss, but the most important function is to monitor the response to treatment.^{14,15,16}

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The biochemical markers levels varies rapidly and correspondingly to bone turnover, thus could be used to assess response and efficacy of pharmacological agents used in treatment of bone diseases such as osteoporosis.¹⁷ There are so many causes of high BTM levels other than osteoporosis, such as hyperparathyroidism, myeloma or thyrotoxicosis.^{18,19} Vitamin D insufficiency as defined by 25(OH)D levels between 30 and 50 nmol/L is associated with an increase in parathyroid hormone (PTH) levels that is attributed to decreased calcium absorption. This may indicate secondary hyperparathyroidism in postmenopausal women.¹⁵

The aim of antiresorptive treatments is to reduce BTMs to the lower part of the healthy premenopausal reference interval.²⁰ Thus the lower limits of BTMs became more important than previously, as not only has it been questioned if decreases of BTMs below the reference interval could harm microdamage repair of the bone, but they also may be helpful in taking decisions in clinical practice, such as whether or not to give the next dose of a potent antiresorptive drug.^{21,22}

Therefore accurate and robust reference intervals for bone turn over markers become mandatory.²³ Unfortunately the available reference ranges are released through commercial laboratory kits, which lack standardization and don't take in consideration preanalytical variables.²⁴ As a result establishment of population based reference intervals that takes in consideration the preanalytical variables including age is considered a priority.

Several BTM reference intervals for healthy premenopausal women have previously been established.^{17, 25,26, 27} In the last few years, reference intervals in premenopausal women from different countries in Europe (UK, France, Belgium and Denmark)^{27,17} and the US²⁸, have been reported. In addition, data from Germany on men and women have been recently published.²⁹ In Spain, there are data on Procollagen I intact N-Terminal (PINP) and Carboxy-terminal collagen cross links (CTX-I) in older men and in postmenopausal women^{30,31,32} as well as on CTX-I in a subset of 50 premenopausal women.³³

To our knowledge up till this moment there is no published data regarding reference values in premenopausal Egyptian females. Thus establishment of a valid and robust reference interval will allow comparing BTMs among different studied groups, differentiate between normal and osteoporotic patients³⁴ also predict the likelihood of fracture in osteoporotic patients and monitoring the suggested effective treatment response which cannot be done by bone mineral density as the latter can't detect minor and acute changes caused by therapeutic agents.^{17,35}

It is reported that nearly 70-80% of adult BMD is attained by the age of 18 years. One study indicated that there was no significant bone loss in 30- 45 year-old women who were calcium balanced and of optimal bone health; we chose women aged 30-45 yrs for reference interval calculations because these women have achieved peak bone mass and are not yet perimenopausal.³⁶ This study aims to establish robust reference intervals for some bone turnover markers in healthy premenopausal Egyptian females between the age of 30-45 yrs.

Material and method:-

Study population:-

This study protocol was approved by the Medical Research Institute, Alexandria University Ethics committee in accordance with the Code of Ethics of the World Medical Association (Declaration of Helsinki) for experiments involving humans.

This cross sectional study was conducted in Alexandria governorate. The specimens used in this study were obtained from an age stratified random sample. All subjects provided written consent.

The clinical and Laboratory Standards Institute (CLSI) released the EP28-A3c document that recommends 120 reference values as an adequate sample size for each reference population or subclass. This study included one hundred and fifty one healthy, non pregnant, regularly menstruating (10 or more periods per year), premenopausal females who were recruited as volunteers from the outpatient clinic of the MRI, Alexandria university, family planning clinics as well as volunteers from the medical, nursing, or secretarial staff of the units or their relatives from April 2015 till November 2015. The sample of this study was age-stratified random sample, the participants where 30-45 years of age; as women in this age have their bone turnover markers stabilized, making them suitable for calculation of clinically useful reference ranges for BTMs.²¹ (mean age 36.6 ± 4.5), with a body mass index (ranging from 17-27 Kg/m²) (mean 22.8), each having regular cyclic menses (12/year) , Weighed between (55-85Kg)

Women under this range were excluded as they might have elevated BTMs due to skeletal immaturity, while those above 45 were excluded as they tend to have increased markers in the perimenausal period with normal estrogen levels.²⁵

Exclusion criteria:-

None of the subjects taking part in the study had cancer, chronic disease, or any medical condition known to affect bone metabolism. All subjects were not receiving, within a year of starting the study, any bone metabolism affecting medications such as : selective estrogen receptor modulators , phytoestrogens , anticonvulsants , calcitonin , anabolic agents , steroids , vitamin D supplements , hormonal treatment for endocrinopathies , NSAID and depo-provera as well as calcium supplements, serotonin uptake inhibitors, L-troxin , antihypertensive drug and antiresorptive treatments . Subjects with history of alcoholism, smoking, recent clinical fracture, pregnancy, lactation, renal and endocrinal disorders were excluded. All participants were subjected to a detailed questionnaire concerning demographic data, medical history, lifestyle, etc.

Sample collection:-

Venous blood were taken from all subjects on a gel separator tube, as well as urine sample after overnight fasting from 8am till 10 am in the second week after menstruation . Venous samples were kept for 10 minutes at room temperature and centrifuged under standard guidelines (2500 g) for 10 min and serum was separated and stored at - 80 C , no freezing and thawing is allowed only once upon analysis.

CTX has significant diurnal rhythm with a peak in the morning and a nadir in the afternoon. Moreover the circulating levels are influenced by food intake. In non fasting conditions, the peak is reached at night, followed by a significant decrease and nadir from 1100 to 1500 h. Therefore all the samples were collected after overnight fast in the morning within a standardized time period ideally from 8 and 10 a.m. thus reducing the day to day variation^{12,37}

Analytical standardization and biochemical analysis:-

Standard Operating Procedures were followed during pre-analytical and analytical phases of the study with two levels of internal quality control included in each run. Corrective actions were done based on Westgard rules . The following biochemical markers were measured at the same time point using a single lot of reagents and the same batch.

Alkaline phosphatase , 25 (OH) vitamin D , PTH and CTX were measured in serum by immunoenzymatic assay on Cobas 411e (Roche diagnostics) with an intra-assay CV of 0.6-0.9 % , 1.7-7.8% , 1.4-7.4% and 1.0-4.6% the interassay CV was 0.9- 2.4 % , 2.2- 10.7% , 3.1-9.4% and 1.6-4.7% while the limit of detection (LOD) was 5 U/L , 3.00 ng/ml (7.5 nmol/L), 5.5-2300 pg/ml (0.583-244 pmol/L) , 0.01 ng/ml (10 pg/ml) respectively.

While Calcium and Phosphorus were carried out on Beckman coulter Olympus with an intra-assay CV of 0.46- 0.55% and 0.6-1.9%, the interassay CV of 0.68-1.34% and 0.9-2.1% respectively. The limit of detection (LOD) was 0.13mg/dl and 1mg/dl respectively.

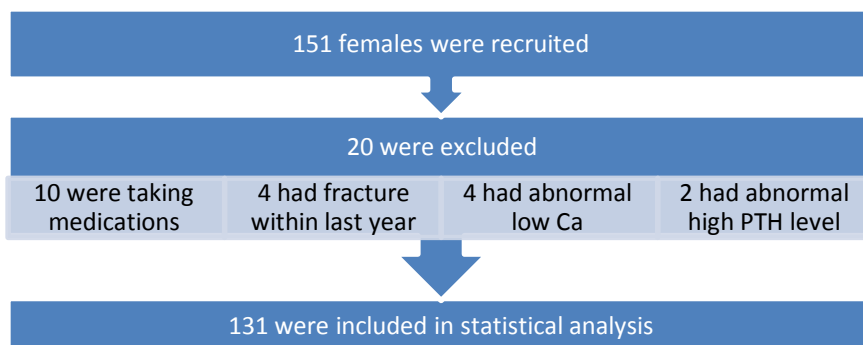
Statistical analysis:-

Statistical analyses were performed using SPSS version 22.0. The data were analyzed using nonparametric statistical techniques.

The initially collected 151 values were tested for outliers (outliers were considered for data point below $Q1 - 1.5 \times IQR$ or above $Q3 + 1.5 \times IQR$), outliers were investigated and overall, 20 cases were excluded from all the parameters for the reasons mentioned above.

Results:-

In this study one hundred fifty one females participated, of these women; four were excluded for having a fracture within one year, ten were excluded for medication intake (vitamin D, calcium and oral contraceptive pills), 4 were excluded for having abnormally low calcium levels and 2 were excluded for having abnormally high PTH level. 131 women completed all lab tests and the questionnaire fig(1)

**Fig (1)**

The age selected was between 30-45, the sample was restricted to this age group and our results showed no statistically significant correlation between age and any of the studied parameters except phosphorus (weak indirect correlation $r_s = -0.189$) and Vit D (intermediate correlation $r_s = -0.58$); thus the norms were established from the whole participants for each of the 6 parameters, furthermore VitD values were presented for the age categories 30-<35yrs ($n=45$), 35-<40yrs ($n=42$) and 40-45 yrs ($n=44$) as the values of VitD varied between the 3 age categories using Kruskal Wallis test. And phosphorous was presented by the age groups <40 ($n=87$) and 40+ years ($n=44$) as values differed using Mannwhitney U test.

Guided by the Clinical and Laboratory Standards Institute (CLSI) analysis of reference values¹, and Adult Reference Intervals on the Basis of the Canadian Health Measures³⁸, values were presented as median (90%CL) and the Survey reference values were constructed using 2.5th and 97.5th percentiles as lower and upper reference limits. The 90% confidence intervals around the estimates of the limits of the reference intervals were constructed by bootstrapping of 500 random samples of the same size.³⁵

Table (1) displays the range and Median (90%CI) values of the 6 parameters (Ca, Phosphorous, Alkaline phosphatase, PTH, CTX and Vit D) for the studied sample ($n=131$). The Coefficient of Variation (CV) of all the studied parameters lay in the accepted values of the Canadian quality standards except CTX which exceeded 33.3%. In the present study the premenopausal women had a median value for calcium 9.4 mg/dl (9.3-9.4), phosphorous 3.5mg/dl (3.4- 3.6), alkaline phosphatase 66 U/L (64-68), PTH 44 pg/ml (39.1- 45.6), CTX of 188 pg/ml (178 – 191pg/ml) and vitamin D 16 (15-16) ng/ml.

Table (1):- Reference intervals for 6 biochemical BTM

Parameter	Range	CV	Median (90%CI)	Bootstrap no. of samples	Lower value 90% CI	Higher value 90% CI	Mean \pm SD	Normal values set by manufacturer
Calcium (mg/dl)	8.5-10.5	2.7%	9.4 (9.3-9.4)	500	8.5-8.7	9.9-10.41	9.34 \pm 0.36	(min-max)8.6-10
Phosphorous(mg/dl)	2.5-4.7	10.0%	3.5 (3.4-3.6)	500	2.5-2.73	4.37-4.64	3.54 \pm 0.49	2.5-4.8
Age(yrs)								
30-<40(n=87)	2.5-4.7	8.3%	3.6 (3.5-3.7)	500	2.5-2.82	4.38-4.7		
40+(n=44)	2.5-4.4	10.6%	3.3 (3.2-3.5)	500	2.5-2.8	4.1-4.4		
Alkaline phosphatase (U/L)	41-99	13.6%	66 (64-68)	500	41.0-44.3	95.0-99.0	67.89 \pm 14.6	35-104
PTH (pg/ml)	21-77	20.5%	44 (39.1-45.6)	500	21.78-24.2	71.35-75.5	45.00 \pm 14.3	(mean) 31.3
CTX(pg/ml)	91-430	42.8%	188 (178-191)	500	91.6-97.95	396.1-428.8	213.9 \pm 95.34	299
Vitamin D (ng/ml)	10-24	15.6%	16 (15-16)	500	10.0-10.0	21-23.7	15.70 \pm 3.40	21.6
Age (yrs)								
30-<35(n=45)	10-24	9.7%	18 (17-19)	500	10.0-13.34	22.8-24		
35-<40(n=42)	10-23	15.4%	17 (16-17.5)	500	10.0-11.0	20.9-23		
40-45(n=44)	10-16	14.4%	13 (12-14)	500	10.0-10.0	15-16		

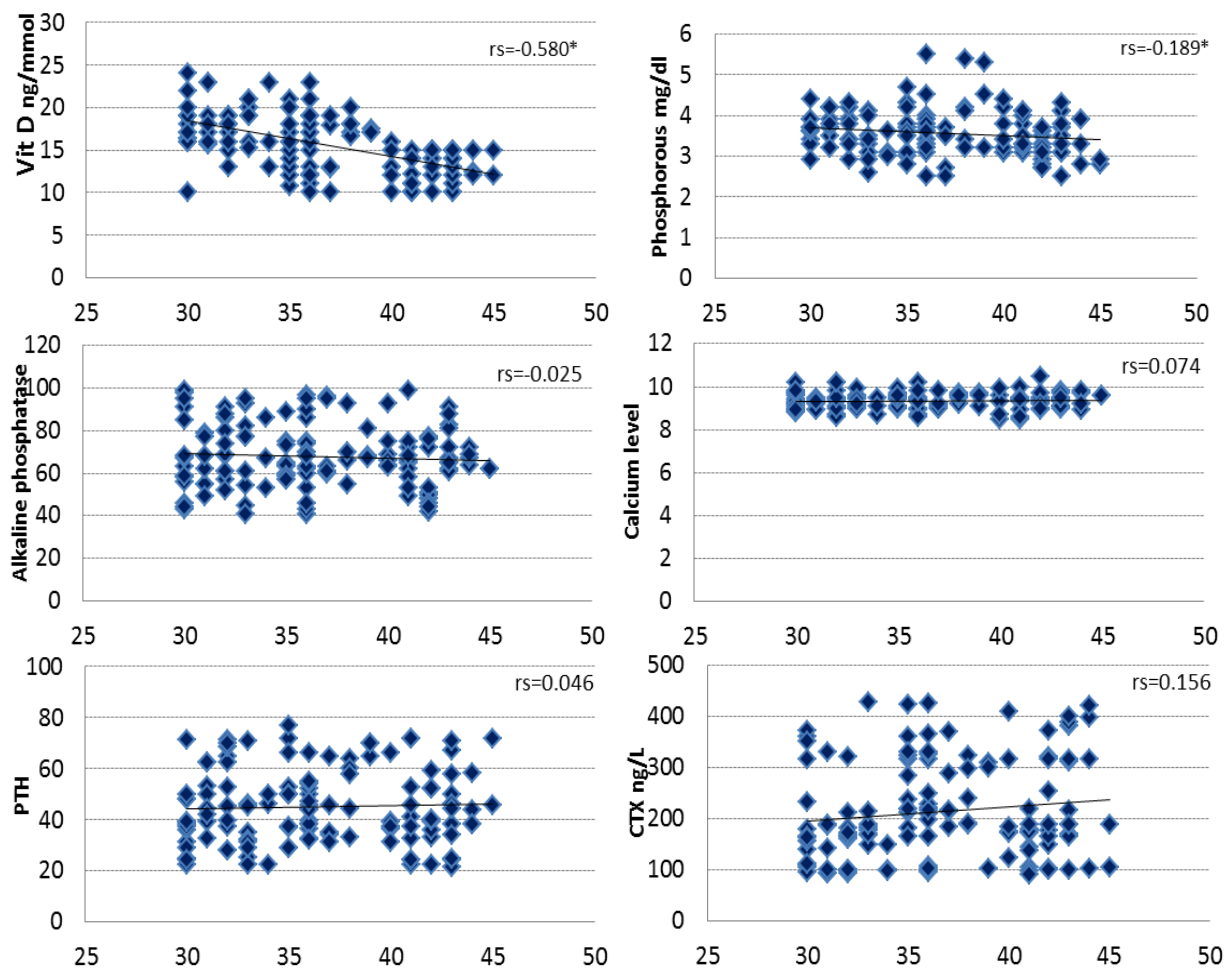


Figure 2:- Correlation between age and each of the 6 biochemical BTM

The new values for Ca and phosphorous did not statistically differ from the manufacturer though accepted higher upper limit (10.4) for Ca and a lower upper limit for phosphorous (4.64).

The new values for ALP as well did not statistically differ from the manufacturer. Concerning CTX and Vit D the new averages were lower than that set by the manufacturer, the differences were statistically significant ($t=-10.2$, $p=0.0001$ for CTX, and $t=-19.7$, $p=0.0001$ for Vit D). While, the new average for PTH was higher than that of the manufacturer, the difference was statistically significant ($t=10.9$, $p=0.0001$).

Discussion:-

The importance of establishment of a reliable reference interval is because most medically important decisions are based on comparing the laboratory tests with their reference intervals.

In fact most reference intervals used in many laboratories are those of the manufacturer. These reference intervals don't represent the actual population and thus needs to be verified or established for the relevant population.

Bone turnover markers are convenient tool used to assess systemic bone turnover rates, both in clinical practice and in clinical trials.

It has been recommended that normal reference ranges for BTMs shall be established in large samples of healthy premenopausal women.¹⁷ However, the reference ranges currently reported by commercial laboratories are often not rigorously established and fail to account for relevant preanalytical variables.¹² Some of these variables including fracture, metabolic bone disease, chronic disease (including; malignancies, rheumatoid arthritis, diabetes), abnormal laboratory results of serum calcium, phosphorous.¹⁷

In the last few years many reference intervals in premenopausal women in different countries like UK, France, Belgium, Denmark, US and Germany have been established and to our knowledge this is the first for Egyptian population.^{17,27,28,29}

The present study aimed to establish robust reference interval of some bone turn over markers in healthy premenopausal females aged from 30 to 45 years with mean age 36.6 ± 4.5 years. This age range was taken because BTM levels were considered to be stable by Glover et al. in their study assessing the reference interval in premenopausal British women which is beneficial in the assessment of response to antiresorptive therapy in postmenopausal osteoporosis.⁽¹⁷⁾

One of the main limiting factors was to include larger number of healthy individuals based on the literature exclusion criteria.¹⁷ The CLSI released EP28-A3c document to help researchers by trying to unify reference interval studies. However bone turn over markers required detailed history due to many factors influencing their level.

The women who participated in the current study fulfilled the detailed inclusion criteria and provided additional information that was used to characterize the population as a whole.

The exclusion criteria to the studied group included: vitamin D supplements, history of fracture, smoking, consumption of alcohol, metabolic bone disease and chronic diseases.¹⁷

All these preanalytical variables contribute to the BTM markers in premenopausal females.

This study includes 151 premenopausal females who reached 131 after application of the above exclusion criteria. Calcium, phosphorous, Alkaline phosphatase, 25 (OH) vitamin D, PTH and CTX were done to all the candidates of this study.

As shown in table (1) the Coefficient of Variation (CV) of all the studied parameters lay in the accepted values of the Canadian quality standards except CTX which exceeded 33.3%. In the present study the premenopausal women had a median CTX value of 188 ng/L (178 - 191 ng/L). This is in accordance with Gossielet al 2014²⁰ who reported a median CTX value of 190 ng/L.

Although these values were lower than those reported by Jenkins et al^{xxxix}, De Papp et al¹⁰, Glover et al²⁵, Martinez et al³⁰ and Lenora et al^{xi} where the median CTx values were 289, 280, 300, 387, 312 ng/L respectively. These discrepancies could be attributed to inter-assay variability of BTM, which represents a significant problem in their clinical application. Variability of collection as well as diurnal variability and intra individual variability have been described and lead to wider reference intervals and less diagnostic certainty. It is known that circadian variation is greater for bone resorption than bone formation. For example serum CTX exhibits a circadian rhythm with an amplitude of 80% of the 24 hour mean.^{xli}

Also Figure (2) shows scatter plots for the age range 30-45 years, showing that VitD values decrease by age (intermediate inverse correlation) and phosphorous values as well decrease by age but with a weak inverse correlation. Otherwise the correlation between age and the other 4 parameters was not statistically significant.

Other studies shows negative correlation between BTM and age, but these studies included women aged from 21-54, so they included women with skeletal immaturity.^{35,xlii} Other study included the same age group as ours and reported higher CTX between the age of 30-34 suggesting they haven't yet reached skeletal maturity.²⁵ This highlights the need for determination of the age at which each bone mass is reached. In our study this problem was unlikely and therefore the selected age group was the optimum for bone turnover markers reference interval establishment.

Comparing the norms resulted from the current research with those set by the manufacturer it was found that:

The new values for Ca and phosphorous did not statistically differ from the manufacturer though accepted higher upper limit (10.4) for Ca and a lower upper limit for phosphorous (4.64).

The new values for ALP as well did not statistically differ from the manufacturer. Concerning CTX and Vit D the new averages were lower than that set by the manufacturer, the differences were statistically significant ($t=-10.2$, $p=0.0001$ for CTX, and $t=-19.7$, $p=0.0001$ for Vit D).

While, the new average for PTH was higher than that of the manufacturer, the difference was statistically significant ($t=10.9$, $p=0.0001$).

The low level of vit D in the Egyptian population may be due to lack of direct sun exposure and reduced intake of vit D supplements as well as reduced calcium, this decreased vit D stimulates the parathyroid gland to secrete more parathyroid hormone and this explains the increased level of PTH which exceeds the manufacturer reference range.

Conclusion:-

BTM are widely used in therapeutic monitoring of osteoporosis or other bone diseases as well as having a significant impact in research. Our trial for establishment of reference interval in Egyptian population may help in harmonization of reporting by Egyptian laboratories. The strength points of this study are the standardization of the preanalytical and analytical conditions with adherence to the CLSI guidelines recommendations together with considering seasonal variation. The small sample size and the absence of the bone mineral density data with relatively low level of 25OHD were the limitations of this study. For a new or improved assay for BTM it is important to include a larger sample size with for a clinical reliable reference range to be used in assessment of the efficacy of the antiresorptive therapy.

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RESEARCH ARTICLE

NUMERICAL SOLUTION FOR MATHEMATICAL MODEL OF EBOLA VIRUS.

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Abstract

Mathematical Modeling has emerged as an important tool for understanding dynamics of many infectious diseases, one of which is the Ebola virus. The main focus of the presented work was to model mathematically the transmission dynamics of Ebola virus, for this purpose, the basic Susceptible-Infected-Recovery (SIR) model of Ebola was reviewed. The basic concept was underpinning the implementation of different numerical techniques like Euler, RK-2, and RK-4 of SIR model. Most optimistic estimates for each group of individuals were obtained like the susceptible group of individuals did not change their values and remained 460, but the infected group of individuals gradually decreased their values, and only nominal increase in case of recovered group of individuals were observed due to the high mortality rate of infected group in case of Ebola.

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Introduction:-

Ebola virus is negative stranded RNA virus causing Ebola haemorrhagic fever. It is an infectious and fatal disease. Its signs and symptoms appear in two days and after two or three weeks fever, sore throat, and muscular pain initiates by reducing the function of the liver and kidneys following the diarrhea and rashes. Bleeding may start from the body in certain cases. Ebola virus spreads by direct contact as well as through blood transfusion. It is so dangerous that after the recovery, the breast milk and semen of the male may carry this virus for several weeks to months as per World Health Organization report (WHO, 2016).

Ebola was first identified at two different places, one is Yambuku and the other is Nazara. It was mostly confined to Central Africa, but recently was also identified in West Africa [1]. A village near the Ebola River, was first affected by this disease, so the disease was named as Ebola Virus Disease (EVD). World Health Organization (WHO) reported 4656 cases of Ebola virus deaths in October 8, 2014, with most cases occurring in Liberia [9]. Natural environment of Ebola consists of 3 hosts which are from the natural host, intermediate animal host and then to humans. There are four sub types of Ebola virus: Ebola Sudan, Ebola Zaire, Ebola Ivory, and Ebola Reston [10].

Mathematical modeling is a significant and powerful tool that can be employed in analyzing the spread and control of infectious diseases such as Ebola. Mathematical models are assumed to provide understanding of methods and suggest prevention, and control strategies [11].

The basic concept in mathematical modeling is to understand the transmission dynamics of diseases. The current study mainly deals with mathematical model of Ebola Virus and implication of three numerical methods Euler,

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Runge- Kutta-2 and Runge-Kutta-4. Finally the numerical results obtained are very optimistic for each group of individuals.

Materials and Method:-

The motivation of this study was to solve the well-known Mathematical model called Susceptible-Infected-Recovery (SIR) model. The real physical problems in the world usually exhibited nonlinear mathematical models which includes biological issues. It was extremely challenging to obtain the exact solutions for such problems that actually represented such phenomena. It was a big task for scientific community to search for appropriate methods such as numerical or perturbation method to solve nonlinear problems [4], but the numerical methods were considered to best for such problems. Therefore numerical techniques Euler, Runge-Kutta 2 and Runge-Kutta 4 were applied to solve the proposed Susceptible-Infected-Recovery(SIR) model in this study.

Description of Model:-

The total population in a specific place was divided into three groups, the susceptible group $S(t)$, the infected group $I(t)$ and the recovered group $R(t)$, "t" was anytime interval ([2], [3], [14]).

The total population was represented by "N" and was taken to be constant for a short time interval and was given by $N=S(t)+I(t)+R(t)$.

Population of the susceptible group reduced as the infected peoples come into contact with them by the rate of infection β . Therefore the change in population of susceptible group was equal to the negative product of $-\beta$ with $S(t)$ and $I(t)$:

$$\frac{dS(t)}{dt} = -\beta \frac{S(t)I(t)}{N} \quad (1)$$

Population of the infected group was changed according to two different ways:

(a) Susceptible group who joined the infected group by adding the total population of infected group with the term $\beta S(t)I(t)$

(b) Infected group who joined the recovered group in any time interval, in such way that the total population of infected group was decreased by a term $\mu I(t)$. Therefore the differential equation of infected group was written as:

$$\frac{dI(t)}{dt} = \beta \frac{S(t)I(t)}{N} - \mu I(t) \quad (2)$$

Finally, the differential equation of recovered group that based on those peoples who recovered from the Ebola virus by a rate μ .

$$\frac{dR(t)}{dt} = \mu I(t) \quad (3)$$

Euler Method:-

Values of parameters used were $\beta = 0.000318$ and $\mu = 0.0175$ and the initial conditions were $S(0) = 460$, $I(0) = 12$, $R(0) = 0$

The Euler method

$$\begin{cases} w_0 = \alpha \\ w_{i+1} = w_i + hf(t_i, w_i) \end{cases}, \quad i = 0, 1, 2, \dots, N-1$$

The time interval was $[0, 90]$ and $N=10$ so

$$h = \frac{b-a}{N}$$

$$h = \frac{90-0}{10} = 9$$

$$t_0 = 0, t_1 = 9, t_2 = 18, t_3 = 27, \dots, t_{10} = 90$$

$$w_0 = \begin{pmatrix} S(0) \\ I(0) \\ R(0) \end{pmatrix} = \begin{pmatrix} 460 \\ 12 \\ 0 \end{pmatrix} = \begin{pmatrix} w_{1,0} \\ w_{2,0} \\ w_{3,0} \end{pmatrix}$$

Iteration-1 for $i = 0$

$$w_1 = w_0 + hf(t_0, w_0)$$

$$w_1 = \begin{pmatrix} w_{1,0} \\ w_{2,0} \\ w_{3,0} \end{pmatrix} + hf(0, w_0)$$

$$w_1 = \begin{pmatrix} 460 \\ 12 \\ 0 \end{pmatrix} + 9 \begin{pmatrix} -0.00038 \\ -0.20962 \\ 0.21 \end{pmatrix}$$

$$w_1 = \begin{pmatrix} 459.99658 \\ 10.11342 \\ 1.89 \end{pmatrix}$$

The successive iterations of Euler method were given in Table-1.

Table 1:- Solution of SIR Model by Euler method up to 10 iterations.

i	t_i	$S(t_i)$	$w_{1,i}$	$I(t_i)$	$w_{2,i}$	$R(t_i)$	$w_{3,i}$
0	0	460	460	12	12	0	0
1	9	460	459.9965	10.2513	10.1134	1.7486	1.8900
2	18	460	459.9937	8.75747	8.52339	3.2425	3.4828
3	27	460	459.9912	7.48130	7.1833	4.5187	4.8252
4	36	460	459.9892	6.39110	6.0540	5.6089	5.9566
5	45	460	459.9874	5.45977	5.1022	6.5402	6.9102
6	54	460	459.9860	4.66415	4.3000	7.3358	7.7138
7	63	460	459.9847	3.98448	3.6239	8.0155	8.3910
8	72	460	459.9838	3.40385	3.0541	8.5961	8.9618
9	81	460	459.9829	2.90783	2.5740	9.0921	9.4429
10	90	460	459.9821	2.48409	2.1693	9.5159	9.8483

Runge-Kutta-2 Method

Solution of the SIR model by RK-2 Heun's method is presented as.

$$\begin{matrix} w_0 = \alpha \\ \left\{ w_{i+1} = w_i + \frac{h}{4} \left[f(t_i, w_i) + 3f\left(t_i + \frac{2h}{3}, w_i + \frac{2h}{3}f(t_i, w_i)\right) \right] \right\}, \quad i = 0, 1, 2, \dots, N-1 \end{matrix}$$

The time interval was [0 90] and N=10

so

$$h = \frac{b-a}{N}$$

$$h = \frac{90-0}{10} = 9$$

$$t_0 = 0, t_1 = 9, t_2 = 18, t_3 = 27, \dots, t_{10} = 90$$

$$w_0 = \begin{pmatrix} S(0) \\ I(0) \\ R(0) \end{pmatrix} = \begin{pmatrix} 460 \\ 12 \\ 0 \end{pmatrix} = \begin{pmatrix} w_{1,0} \\ w_{2,0} \\ w_{3,0} \end{pmatrix}$$

Iteration-1 for $i = 0$

$$\begin{aligned} w_1 &= w_0 + \frac{h}{4} \left[f(t_0, w_0) + 3f\left(t_0 + \frac{2h}{3}, w_0 + \frac{2h}{3}f(t_0, w_0)\right) \right] \\ w_1 &= \begin{pmatrix} 460 \\ 12 \\ 0 \end{pmatrix} + \frac{9}{4} \left[f\left(0, \begin{pmatrix} 460 \\ 12 \\ 0 \end{pmatrix}\right) + 3f\left(0 + \frac{2(9)}{3}, \begin{pmatrix} 460 \\ 12 \\ 0 \end{pmatrix} + \frac{2(9)}{3}f\left(0, \begin{pmatrix} 460 \\ 12 \\ 0 \end{pmatrix}\right)\right) \right] \end{aligned}$$

$$\begin{aligned}
 w_1 &= \begin{pmatrix} 460 \\ 12 \\ 0 \end{pmatrix} + 2.25 \left[\begin{pmatrix} -0.00038 \\ -0.20962 \\ 0.21 \end{pmatrix} + 3f \left(6, \begin{pmatrix} 460 \\ 12 \\ 0 \end{pmatrix} + 6 \begin{pmatrix} -0.00038 \\ -0.20962 \\ 0.21 \end{pmatrix} \right) \right] \\
 w_1 &= \begin{pmatrix} 460 \\ 12 \\ 0 \end{pmatrix} + 2.25 \left[\begin{pmatrix} -0.00038 \\ -0.20962 \\ 0.21 \end{pmatrix} + 3f \left(6, \begin{pmatrix} 459.99772 \\ 10.74228 \\ 1.26 \end{pmatrix} \right) \right] \\
 w_1 &= \begin{pmatrix} 460 \\ 12 \\ 0 \end{pmatrix} + 2.25 \left[\begin{pmatrix} -0.00038 \\ -0.20962 \\ 0.21 \end{pmatrix} + \begin{pmatrix} -0.00101 \\ -0.56295 \\ 0.56397 \end{pmatrix} \right] \\
 w_1 &= \begin{pmatrix} 460 \\ 12 \\ 0 \end{pmatrix} + \begin{pmatrix} -0.00313 \\ -1.73828 \\ 1.74143 \end{pmatrix} \\
 w_1 &= \begin{pmatrix} 459.99687 \\ 10.26172 \\ 1.74143 \end{pmatrix} = \begin{pmatrix} w_{1,1} \\ w_{2,1} \\ w_{3,1} \end{pmatrix}
 \end{aligned}$$

The successive iterations of RK-2 Heun's method are given in Table-2.

Table 2:- Solution of SIR Model by RK-2 method up to 10 iterations.

i	t_i	$S(t_i)$	$w_{1,i}$	$I(t_i)$	$w_{2,i}$	$R(t_i)$	$w_{3,i}$
0	0	460	460	12	12	0	0
1	9	460	459.99687	10.25132	10.26172	1.74868	1.74143
2	18	460	459.99419	8.75747	8.77523	3.24253	3.23059
3	27	460	459.99189	7.48130	7.50112	4.51870	4.50470
4	36	460	459.98993	6.39110	6.41453	5.60890	5.59325
5	45	460	459.98871	5.45977	5.46631	6.54023	6.54718
6	54	460	459.98731	4.66415	4.67447	7.33585	7.34044
7	63	460	459.98601	3.98448	3.99733	8.01552	8.01879
8	72	460	459.98497	3.40385	3.41827	8.59615	8.59886
9	81	460	459.98405	2.90783	2.92311	9.09217	9.09492
10	90	460	459.98331	2.48409	2.49968	9.51591	9.51591

Runge-Kutta-4 Method

The approximate solution by RK-4 method was

$$\begin{cases} w_0 = \alpha \\ w_{i+1} = w_i + \frac{1}{6}[k_1 + 2k_2 + 2k_3 + k_4] \end{cases}, \quad i = 0, 1, 2, \dots, N-1$$

Where

$$\begin{aligned}
 k_1 &= hf(t_i, w_i) \\
 k_2 &= hf\left(t_i + \frac{h}{2}, w_i + \frac{k_1}{2}\right) \\
 k_3 &= hf\left(t_i + \frac{h}{2}, w_i + \frac{k_2}{2}\right) \\
 k_4 &= hf(t_i + h, w_i + k_3)
 \end{aligned}$$

Iteration-1 for $i=0$

$$\begin{aligned}
 k_1 &= 9f(t_0, w_0) \\
 k_1 &= 9f\left(0, \begin{pmatrix} 460 \\ 12 \\ 0 \end{pmatrix}\right) = \begin{pmatrix} -0.00342 \\ -1.88658 \\ 1.89 \end{pmatrix} \\
 k_2 &= 9f\left(t_0 + \frac{9}{2}, w_0 + \frac{k_1}{2}\right)
 \end{aligned}$$

$$\begin{aligned}
 k_2 &= 9f\left(4.5, \begin{pmatrix} 459.99829 \\ 11.05671 \\ 0.945 \end{pmatrix}\right) = \begin{pmatrix} -0.00315 \\ -1.73835 \\ 1.74143 \end{pmatrix} \\
 k_3 &= 9f\left(t_0 + \frac{9}{2}, w_0 + \frac{k_2}{2}\right) \\
 k_3 &= 9f\left(4.5, \begin{pmatrix} 459.99842 \\ 11.13082 \\ 0.87072 \end{pmatrix}\right) = \begin{pmatrix} -0.00315 \\ -1.74996 \\ 1.75310 \end{pmatrix} \\
 k_4 &= 9f(t_0 + 9, w_0 + k_3) \\
 k_4 &= 9f\left(9, \begin{pmatrix} 459.99685 \\ 10.25004 \\ 1.75310 \end{pmatrix}\right) = \begin{pmatrix} -0.00288 \\ -1.61145 \\ 1.61438 \end{pmatrix}
 \end{aligned}$$

The obtained results were

$$\begin{aligned}
 w_1 &= \begin{pmatrix} 460 \\ 12 \\ 0 \end{pmatrix} + \frac{1}{6} \left[\begin{pmatrix} -0.00342 \\ -1.88658 \\ 1.89 \end{pmatrix} + 2 \begin{pmatrix} -0.00315 \\ -1.73835 \\ 1.74143 \end{pmatrix} + 2 \begin{pmatrix} -0.00315 \\ -1.74996 \\ 1.75310 \end{pmatrix} + \begin{pmatrix} -0.00288 \\ -1.61145 \\ 1.61438 \end{pmatrix} \right] \\
 w_1 &= \begin{pmatrix} 459.99685 \\ 10.25422 \\ 1.74891 \end{pmatrix} = \begin{pmatrix} w_{1,1} \\ w_{2,1} \\ w_{3,1} \end{pmatrix}
 \end{aligned}$$

Similarly, the successive iterations of the RK-4 method were presented in Table-3.

Table 3:- Solution of SIR Model by RK-4 method up to 10 iterations.

i	t_i	$S(t_i)$	$w_{1,i}$	$I(t_i)$	$w_{2,i}$	$R(t_i)$	$w_{3,i}$
0	0	460	460	12	12	0	0
1	9	460	459.99685	10.25132	10.25422	1.74868	1.74891
2	18	460	459.99416	8.75747	8.76241	3.24253	3.24338
3	27	460	459.99185	7.48130	7.48765	4.51870	4.52043
4	36	460	459.98987	6.39110	6.39833	5.60890	5.61169
5	45	460	459.98817	5.45977	5.59877	6.54023	6.41293
6	54	460	459.98671	4.66415	4.78427	7.33585	7.22891
7	63	460	459.98546	3.98448	4.09809	8.01552	7.91631
8	72	460	459.98441	3.40385	3.51744	8.59615	8.49799
9	81	460	459.98351	2.90783	3.00576	9.09217	9.01060
10	90	460	459.98271	2.48409	2.56848	9.51591	9.44866

Results and Discussion:-

Results obtained in case of all three numerical techniques were found to be exactly the same as were found by homtopyperturbation method in [4]. Solution of the model is presented graphically in Fig-1. Moreover we solve the model analytically by linearization as follows:

The system of equations 1-3 was nonlinear which was linearized as:

Let

$$\begin{aligned}
 f_1 &= \frac{dS(t)}{dt} = -\beta \frac{S(t)I(t)}{N} \\
 f_2 &= \frac{dI(t)}{dt} = \beta \frac{S(t)I(t)}{N} - \mu I(t) \\
 f_3 &= \frac{dR(t)}{dt} = \mu I(t)
 \end{aligned}$$

$$J = \begin{pmatrix} \frac{\partial f_1}{\partial S} & \frac{\partial f_1}{\partial I} & \frac{\partial f_1}{\partial R} \\ \frac{\partial f_2}{\partial S} & \frac{\partial f_2}{\partial I} & \frac{\partial f_2}{\partial R} \\ \frac{\partial f_3}{\partial S} & \frac{\partial f_3}{\partial I} & \frac{\partial f_3}{\partial R} \end{pmatrix}_{(0,0,0)}$$

$$J = \begin{pmatrix} 0 & 0 & 0 \\ 0 & -\mu & 0 \\ 0 & \mu & 0 \end{pmatrix}$$

For Eigenvalues

$$|\lambda I - J| = 0$$

$$\begin{vmatrix} \lambda & 0 & 0 \\ 0 & \lambda & 0 \\ 0 & 0 & \lambda \end{vmatrix} - \begin{vmatrix} 0 & 0 & 0 \\ 0 & -\mu & 0 \\ 0 & \mu & 0 \end{vmatrix} = 0$$

$$\lambda(\lambda(\lambda + \mu) + 0) = 0$$

$$\lambda = 0, 0, -\mu$$

Eigen vectors were found as

$$(\lambda I - J)x = 0$$

For $\lambda = 0, 0, -\mu$, Eigenvectors obtained were

$$\begin{pmatrix} 0 \\ 0 \\ 1 \end{pmatrix}, \begin{pmatrix} 1 \\ 0 \\ 0 \end{pmatrix}, \begin{pmatrix} 0 \\ -1 \\ 1 \end{pmatrix}$$

And the solution of SIR Model was

$$\begin{aligned} X &= C_1 e^{\lambda_1 t} V_1 + C_2 e^{\lambda_2 t} V_2 + C_3 e^{\lambda_3 t} V_3 \\ \begin{pmatrix} S(t) \\ I(t) \\ R(t) \end{pmatrix} &= C_1 e^{(0)t} \begin{pmatrix} 0 \\ 0 \\ 1 \end{pmatrix} + C_2 e^{(0)t} \begin{pmatrix} 1 \\ 0 \\ 0 \end{pmatrix} + C_3 e^{-\mu t} \begin{pmatrix} 0 \\ -1 \\ 1 \end{pmatrix} \\ S(t) &= C_1 e^0(0) + C_2 e^0(1) + C_3 e^{-\mu t}(0) \\ S(t) &= C_2 \\ I(t) &= C_1 e^0(0) + C_2 e^0(0) + C_3 e^{-\mu t}(-1) \\ I(t) &= -C_3 e^{-\mu t} \\ R(t) &= C_1 e^0(1) + C_2 e^0(0) + C_3 e^{-\mu t}(1) \\ R(t) &= C_1 + C_3 e^{-\mu t} \end{aligned}$$

By the use of initial conditions $S(0) = 460$, $I(0) = 12$, $R(0) = 0$ at $t = 0$ obtained values were

$$C_1 = 12, C_2 = 460 \text{ and } C_3 = -12$$

Final analytical solution for nonlinear SIR model was:

$$S(t) = 460 \quad (4)$$

$$I(t) = 12e^{-\mu t} = 2.4841 \quad (5)$$

$$R(t) = 12 - 12e^{-\mu t} = 12(1 - e^{-\mu t}) = 9.5159 \quad (6)$$

These results are presented in Fig-1 for justification purpose. Each group of individuals i.e., Susceptible, Infected, and recovered were taken along y-axis versus time along x-axis.

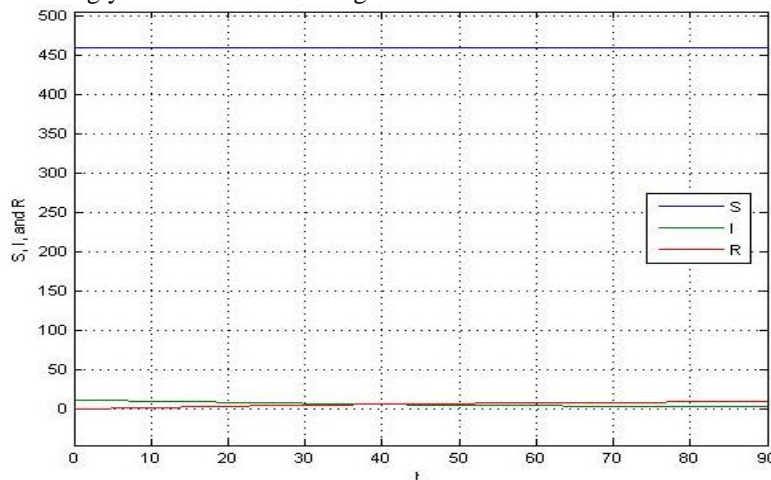


Figure1:-Solution of SIR Model (Time along X-axis & S, I,R along Y-axis).

Different numerical methods like Euler, Runge- Kutta 2, RK-2 and RK-4 were used to solve the model numerically, the results were presented in Tables 1, 2 and 3 were exactly the same as in Equations 4, 5 and 6 obtained by analytical method .

Conclusion and Future Work:-

Solution of model by three numerical methods Euler, Runge-Kutta-2 (Heuns) and Runge-Kutta-4 were presented in Tables 1, 2 and 3. Conclusion was drawn that most optimistic estimates for each group of individuals were obtained like the susceptible group did not change and remained 460, but the infected group gradually decreased its values, due to the high mortality rate of infected group, and the recovered group slightly increased its values. This study can be extended further for parameter estimation and sensitivity analysis of the model.

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RESEARCH ARTICLE

THEORETICAL STUDY OF THE EFFECT OF LINEAR DEFORMATION ON BULK MODULUS AND COMPRESSIBILITY OF METALS.

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Abstract

The effects of deformation on the bulk modulus and compressibility of different elemental metals were computed and studied based on pseudopotential formalism. The electron density parameters of deformed metals under the application of different strains were obtained for different metals. The poisson ratio relating the transversal compression to elongation in the direction of applied deformation for different elemental metals were computed using elastic moduli for homogeneous isotropic material and used in this work. The results obtained revealed that there is a good agreement between the computed and experimental value of the bulk modulus and compressibility of metals. There is high concentration of electron in the high density region than in the low density region for the bulk modulus and compressibility of metals these seems to suggest that the bulk modulus and compressibility of metals depend on the density of valence electron in metals. The bulk modulus of all the metals investigated decreases as deformation increases. These could be due to reduction in electron compressible rate, fracture density and increase in inter atomic distance between the electrons in the metals. The effect of deformation is more pronounced on the polyvalent metals than in alkaline metals these could be due to the high electronic concentration and high electronic energy level of the free atom in the alkaline metal. Compressibility increases with an increase in deformation for all the metals investigated. These could be due to an increase in the collision between the interacting electrons in metals which forces the compressibility of the electron in metals to increase as deformation increases.

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Introduction:-

The simplest model for describing electrons in solid is to assume that the valence electrons of an atom in solids are free to move anywhere throughout the volume of the material but are not allowed to leave (Pillai, 2010). The electric and magnetic properties of solids are mainly determined by the properties of electrons in them, the energy levels of electrons hold the key to the properties of solids (Solymar and Walsh, 1984). The electrons in solid behave as a delocalized gas of free electrons (Elliot, 1999).

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Pseudopotential approach in a way gives organizing experimental and computational information about conduction electrons in solid in a compact and physical form (Marder, 2000). The goal of pseudopotential theory is to obtain the key physical properties of solids by dealing with only the valence electrons (Kiejna, 1993). Transport processes in solids leads to electrical conductivity, thermal conductivity, thermal resistivity etc. These are affected by electron-electron, electron-impurity and electron-phonon scattering. Carriers in conductors move in a definite direction under applied fields and temperature gradients (Duan and Guojun, J.2005)

The bulk modulus of solid is a measure of its resistance to uniform compression and is defined as the ratio of the infinitesimal pressure increase to the resulting relative decrease in volume (Animalu, 1977). Bulk modulus is a thermodynamic quantity, and is specify by how the temperature varies during compression (Kittel, 1976). Compressibility is determined by the second derivative of the total energy with respect to the volume and can also be express as the reciprocal of the bulk modulus of solids (Kukkonen and Wilkins, 1979). The compressibility sum rule expresses the self-consistency condition that the static response of the system to the long-wavelength perturbation is equal to the compressibility where the density dependent ground state energy obtained via the Monte Carlo method is used to determine the compressibility via thermodynamic relations. (Iwamoto, 1999).

Deformation is the change in shape or size of solid (Backofen, 1972). A solid body is deform when a stress is applied to it, all solid materials exhibit nearly Hookean behaviour for small enough strain or stress (Borg,1990). When a material is subjected to external forces its behaviour depends not only on the magnitudes of the forces and the inherent strength of the material itself but also on the way the forces are applied and combined. The particular combination of forces may cause the material to deform elastically or plastically. The amount of deformation depends on knowing the intensity of the forces at all points throughout the material (Hugh Ford and Alexander, 1977). Metals can be deformed by compressive, tensile and torsion force. During deformation, atoms at the surface and interior of metals changes together with their atomic distance depending on the metallic surface area that is subjected to different deformation. The contact potential difference on the metal surface also changes during deformation (Borg, 1990).

Over the years, the bulk modulus and the electron compressibility of solids has been studied both theoretically and experimentally. Osiele and Edenma, (2009) developed a model for computing the bulk modulus of metals based on the theory of structureless pseudopotential. The results obtained revealed that the calculated and experimental bulk modulus of metals varies in the same manner with experimental values. Ling and Gelatt(1981) used a local pseudopotential constructed from the bulk stability condition alone to calculate the shear and bulk moduli of 19 simple metals. They also studied the chemical trends in the elastic modulus of the simple metals and calculated the ratio of shear modulus to bulk modulus, the result obtained was in good agreement with experimental value. Vackar et al, (1998) developed the all-electron pseudopotential model and used it to calculate the lattice constant and bulk modulus of silicon, diamond, non-magnetic cobalt, cubic TiC and hexagonal TiS. The results they got were in satisfactory agreement with experimental values. Osiele et al (2011) develop a model for computing thermal resistivity, compressibility ratio and screening parameter of metals based on free electron theory. The result obtained revealed that thermal resistivity of metals increases with an increase in the electron gas parameter. While the compressibility ratio of metals decreases with an increase in the electron gas parameter. On the screening parameter, the thermal resistivity also increases with an increase in the screening parameter while the compressibility ratio decreases with an increase in the screening parameter. Keijna and Pogosov, (2000) experimentally investigate the effect of deformation on electronic properties of some metals by measuring directly a sample of deformed metal using kelvin method. They observed that the contact potential difference of metals decreases when tensed and increases when compressed. Adeshakin and Osiele (2012) computed the surface energy and surface stress of deformed metals based on the structureless pseudopotential formalism. The results obtained revealed that deformation causes a reduction of surface energy and this reduction is more pronounced in simple and alkaline metals. Tensile stress is present in most metallic surfaces whose surface stress were computed, although a few metals possess compressive stress on their surfaces. In the presence of deformation, the surface stress of some metals decreases, while deformation causes an increase in the surface stress of some metals. Adesakin, (2016) develop a model to compute the electrical conductivity of different elemental metals based on pseudopotential formalism. The results obtained revealed that there is a good agreement between the computed and experimental value of the electrical conductivity of metals. There is high concentration of electron in the high density region than the low density region. The electrical conductivity of metals decreases as deformation (strains) increases for all the metals investigated. The effect of deformation is more pronounced on the electrical conductivity of noble and transition metals than in alkaline metals.

In this work the structureless pseudopotential is extended to the study of the effects of linear deformation on the Bulk modulus and compressibility of some metals. This will give an insight into how the Bulk modulus and compressibility of metals changes with deformation. The metals were chosen based on their technological and industrial applications and availability of some physical constants of metals that is required for computation.

Theory And Calculations:-

Electrons are quantum not classical particles and so the allowed electronic states must be the solutions of the Schrodinger equation which in the general (non-relativistic) case where the potential energy V is a function of both space and time is:

$$\frac{\hbar^2}{2m_e} \nabla^2 \psi(r, t) + V(r, t) \psi(r, t) = i\hbar \frac{\partial}{\partial t} [\psi(r, t)] \quad (2.1)$$

Where m_e is the electron mass, \hbar is the normalised Planck's constant and $\psi(r, t)$ is the electron wavefunction. The energy functional of the system of interacting electron in an external potential arising from the interaction with ions represented by a local pseudopotential ϕ can be expressed as a functional of the valence electronic density as

$$E[n] = T_s[n] + E_{xc}[n] + \frac{1}{2} \int d^3r \int d^3r' \frac{n(r)n(r')}{|r' - r|} + \int d^3r \phi(|r - R_i|)n(r) + \frac{1}{2} \sum_i \frac{Z^2}{|R_i - R_j|} \quad (2.2)$$

where the first three terms represents the kinetic, exchange-correlation and electrostatic energy of the interacting system. The two last terms describes the interaction of electrons with the ions at sites R_i via a pseudopotential, ϕ and the Coulomb interaction between the ions.

The total energy required to assemble the valence electrons and ions to form the solid which is the binding energy is

$$E = T_s(n) + E_{xc} + W_R + E_m \quad (2.3)$$

Where T_s is the kinetic energy, E_{xc} is the sum of the exchange and correlation energies, W_R is the average value of the non-Coulombic part of the pseudopotential and E_m is the Madelung energy of points ions embedded in a uniform negative background.

Based on the density functional theory in the low density limit, the kinetic and exchange energies are given as (Kiejna and Wojciechowski, 1996)

$$T_s = \frac{1.105}{r_s^2} \text{ and } E_{xc} = \frac{-0.458}{r_s} \quad (2.4)$$

Where r_s is the electron density parameter, which is describe as the radius of a sphere containing one electron on the average.

The correlation energy used in this work is that of Carperly and Alder as parameterised by Perdew and Zunger and is given as

$$E_c = \frac{-0.1423}{1 + 1.0529r_s^2 + 0.3334r_s} \quad (2.5)$$

From equation (2.3), the binding energy is

$$E = \frac{1.105}{r_s^2} - \frac{0.458}{r_s} - \frac{0.1423}{1 + 1.0529r_s^2 + 0.3334r_s} + W_R + E_m \quad (2.6)$$

The average value of the repulsive part of the pseudopotential is

$$W_R = \frac{n}{2} \int d^3r W_R(r) = \frac{n_{av}}{2} \int 4\pi r^2 \frac{Z}{r} dr = 2\pi n_{av} r_c \quad (2.7)$$

Where n_{av} is the average density of the electron.

The self-energy of the electrostatic interaction average over the whole Wigner-Seitz sphere is

$$E_{es} = \frac{\frac{1}{2} \int V(r) 4\pi r^2 dr}{\frac{4\pi r_0^3}{3}} = \frac{3Z}{5r_0} \quad (2.8)$$

The factor of half is to avoid double counting. The positive contributions are compensated for by the negative energy of the electrons interacting with the uniform positive background.

The Medelung energy for a jellium system is given by (Perdew and Zunger, 1981) as

$$E_m = \frac{-3Z}{2r_0} + E_{es} = \frac{-9Z}{10r_0} \quad (2.9)$$

Where $r_0 = Z^{1/3}r_s$. Substituting equation (2.7) and (2.9) into (2.8), the binding energy according to the structureless pseudopotential model is obtain as

$$E = \frac{1.105}{r_s^2} - \frac{0.458}{r_s} - \frac{0.1423}{1 + 1.0529r_s^2 + 0.3334r_s} + 2\pi n r_c - \frac{9Z}{10r_0} \quad (2.10)$$

But $r_0 = Z^{1/3}r_s$, and $n = 4\pi r_s^3/3$, hence

$$E = \frac{1.105}{r_s^2} - \frac{0.458}{r_s} - \frac{0.1423}{1 + 1.0529r_s^2 + 0.3334r_s} + \frac{3r_c^2}{2r_s^3} - \frac{9Z^{2/3}}{10r_s} \quad (2.11)$$

Where r_s is the electron density parameter of metals and r_c is the Ashcroft core radius given as (Perdew, et, al, 1990)

$$r_c = \left[\frac{-2}{15} \left(\frac{9\pi}{4} \right)^{2/3} r_s + \frac{1}{6\pi} \left(\frac{9\pi}{4} \right)^{1/3} r_s^2 + \frac{1}{5} Z^{2/3} r_s^2 + \frac{2}{9} r_s^4 \frac{dE_c}{dr_s} \right]^{1/2} \quad (2.12)$$

Where E_c is the correlation energy given by equation (2.5)

Since the conduction electrons in a solid is being treated as a gas of particles, albeit a quantum gas, they will exert a pressure given at zero kelvin by (Pillai, 2010)

$$P = - \left(\frac{\partial E}{\partial V} \right)_N \quad (2.13)$$

Since it is assumed that the potential energy is zero, the origin of the pressure is regarded as being due to the repulsion experienced by electrons caused by the Pauli Exclusion Principle, when they are compressed and tend to occupy the same region space.

The bulk modulus which measures resistance to structure preserving volume-changing for an underformed metal is (Ling and Gelatt, 1981)

$$B = -V \left(\frac{\partial p}{\partial v} \right)_{T,N} = \frac{1}{12\pi} \left(\frac{1}{r_s^2} \frac{\partial^2 E}{\partial r_s^2} - \frac{2}{r_s^2} \frac{\partial E}{\partial r_s} \right) \quad (2.14)$$

While compressibility for an undeformed metals is obtained as

$$\tau = \frac{1}{\frac{1}{12\pi} \left(\frac{1}{r_s^2} \frac{\partial^2 E}{\partial r_s^2} - \frac{2}{r_s^2} \frac{\partial E}{\partial r_s} \right)} \quad (2.15)$$

Considering only the kinetic energy of the free-electron for which $P \propto V^{-5/3}$, the zero kelvin electronic contribution to the bulk modulus is (Elliott, 1997)

$$B_{KE} = \frac{0.586}{r_s^5} (a.u) \quad (2.16)$$

The compressibility of the uniform electron gas is determined by the second derivative of the total energy with respect to volume

$$K = \left(V \frac{\partial^2 E}{\partial V^2} \right)^{-1} \quad (2.17)$$

The compressibility of the ratio of the non-interacting electron gas K_{free} to that of the interacting electron gas K is given as (Bowen Etal, 1974)

$$\frac{K_{free}}{K} = \left[1 - \frac{4}{\pi} \alpha r_s \frac{1}{4} \frac{\pi \alpha}{24} r_s^5 \frac{d}{dr_s} \left(\frac{1}{r_s^2} \frac{dE_c}{dr_s} \right) \right] \quad (2.18)$$

Where r_s is the electron density parameter, $\alpha = (4/9\pi)^{1/3}$ and E_c is the correlation energy per electron which is given by equation (2.5)

For a deformed metal, the average electron density parameter of metal is

$$r_{su} = r_s [1 + (1 - 2\nu)U_{xx}]^{1/3} \quad (2.19)$$

where ν is the Poisson ratio relating the transversal compression to elongation in the direction of applied deformation and U_{xx} is the uniaxial strain.

The bulk modulus and the compressibility of the deformed solid at absolute zero is given by

$$\beta = \frac{1}{12\pi} \left(\frac{1}{r_{su}^2} \frac{\partial^2 E}{\partial r_{su}^2} - \frac{2}{r_{su}^2} \frac{\partial E}{\partial r_{su}} \right) \quad (2.20)$$

and

$$\gamma = \frac{1}{\beta} = \frac{1}{12\pi} \left(\frac{1}{r_{su}^2} \frac{\partial^2 E}{\partial r_{su}^2} - \frac{2}{r_{su}^2} \frac{\partial E}{\partial r_{su}} \right) \quad (2.21)$$

This study generalized the work of Kiejna and Pogoss (2000) due to the shortcomings of the electron density parameter of deformed metals. They failed to account for metal dilation by assuming a constant value for the Poisson ratio of metals which leads to neglect of the uniaxial strain in their computation. In this work, the Bulk modulus and electron compressibility of deformed metals were computed using equation (2.20) and (2.21) and how deformation affects these properties of metals is studied. Also the variations of Bulk modulus and electron compressibility with electron density parameters were computed and studied using equation (2.14) and (2.15).

Results And Discussion:-

Figure 1 shows the variation of bulk modulus with electron density parameter for some monovalent, divalent, trivalent and polyvalent metals. Figure 1 revealed that there is high concentration of electron in the high density region than the low density region for both computed and experimental values of the bulk modulus. These seem to suggest that the higher the density of valence electron and the electronic concentration of metal the higher the bulk modulus and the lower the density of valence electron and electronic concentration of metal the lower the bulk modulus. In the high density region we have the alkaline metals and in the lower density region we have the noble and inner transition metals. Although, the bulk modulus of metals in figure 1 seems not to exhibit a particular trend and these could be due to the fact that there are some basic properties that contribute to the bulk modulus of metals that the model does not take into consideration such as the nature of chemical bonding, impurity atom and atomic size of the metals. The trend exhibited by metals in figure 1 revealed that there is an agreement between the computed and experimental values of the bulk modulus. This is more pronounce in some of the metals the high density region. The experimental value of the bulk modulus used in these work is obtained from solid state Physics by Kittel (1976). Figure 2 shows the variation of compressibility with electron density parameter for group one, group two, group three and transition metals. The trend exhibited by metals in figure 2 seems to be linear and exhibit a neither increase nor decrease in compressibility for metals the high density region but towards where $r_s \geq 2.75$ a.u there is a clear increase in the compressibility of the metals as we go from one metals to another. The trend exhibited by metals in figure 2 shows that compressibility is truly the inverse of bulk modulus. Figure 2 revealed that metals in the high density region have low compressibility while metals in the low density region have high compressibility. These could be due to the variable valence electron contribution in the solid and the nature of their electronic state structure.

Figure 3 shows the variation of bulk modulus with deformation for some monovalent, divalent, trivalent and polyvalent metals. Figure 3 revealed that the bulk modulus of all the metals investigated decreases with an increase in deformation. These could be due to reduction in electron compressible rate, fracture density and the pressure increase during deformation. The trend exhibited by metals in figure 3 shows that potassium has the lowest bulk modulus while molybdenum were found to be having the highest bulk modulus among all the metals subjected to different deformation. These could be due to the high electronic concentration, high equilibrium distance between the electron and high electronic energy level of the free atom in the alkaline metal that cause the bulk modulus of potassium to be the least affected among all the metals subjected to different deformation. Figure 3 also revealed that metals in the high density region has high bulk modulus while metals in the low density region were found to be having low bulk modulus among all the metals subjected to different deformation. These seems to suggest that as deformation increase the pressure and the atomic volume of the metals increases and thereby reduces the strength of interaction between the electron which forces the bulk modulus of the metals to decrease as deformation increases. Furthermore, the trend exhibited by metals in figure 3 revealed that as deformation increases, there is an increase in the electron temperature due to delocalisation as these may cause a decrease in some of the elastic properties that the bulk modulus of metals depend upon.

Figure 4 shows the variation of compressibility with deformation for some metals containing group one, group two, group three and transition metals. Figure 4 revealed that the electron compressibility increase with an increase in deformation for all the metals investigated. These seems to suggest that as deformation increases the collision and temperature between the interacting electron in metals increases and their by forces the compressibility of the electron in metals to increase as deformation increases. Figure 4 also revealed that metals in the high density region has low compressibility while metals in the low density region has high compressibility, these could be due to the

change in the electronic energy level of the free atoms in the solid that causes the number of valence electrons per unit cell to occupy more sites during deformation which result to an increase in the compressibility as deformation increases. Among all the metals subjected to different deformation, potassium has the highest compressibility; this could be due to the high density of the free valence electron, compression factor and electronic concentration in the metal. The trend exhibit by metals in figure 3 and 4 revealed that the bulk modulus and compressibility of metals is strongly affected by deformation.

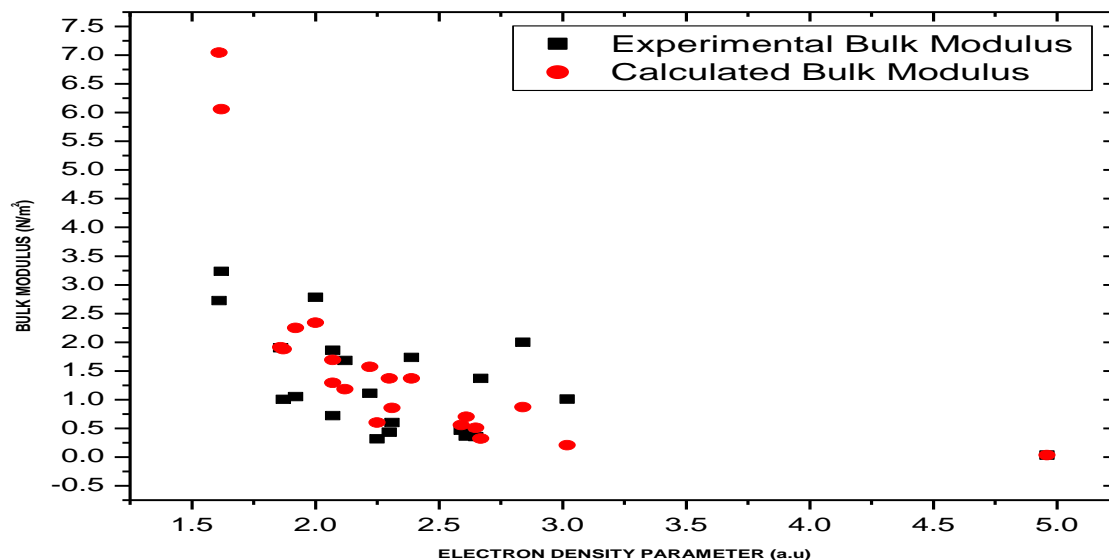


Figure 1:- Variation of Bulk Modulus with Electron Density Parameter for Undeformed Metals.

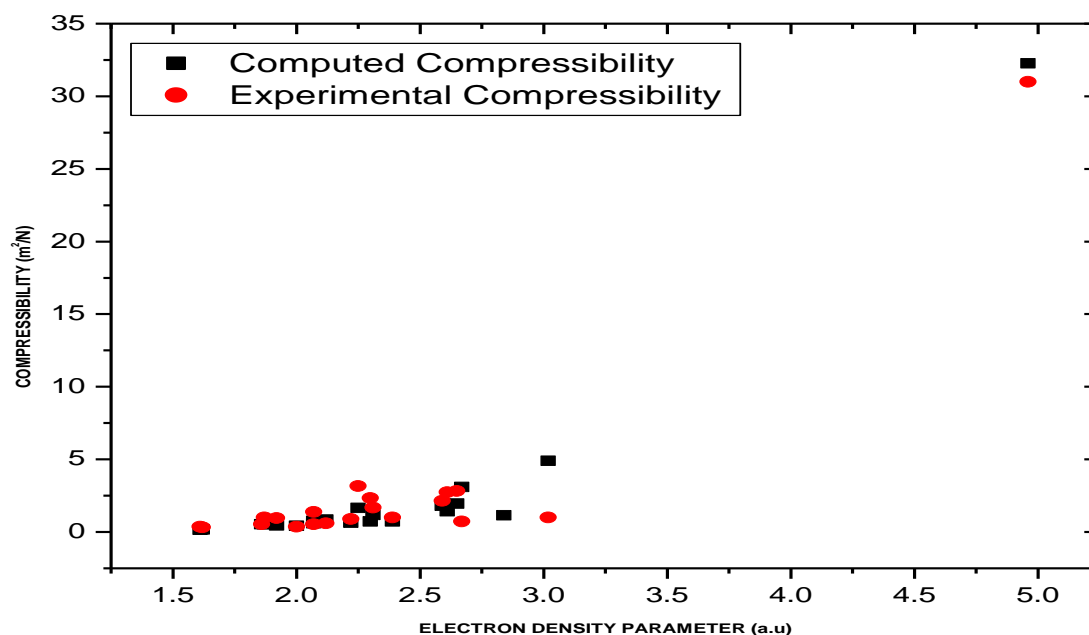


Figure 2:- Variation of Compressibility with Electron Density Parameter for Undeformed Metals

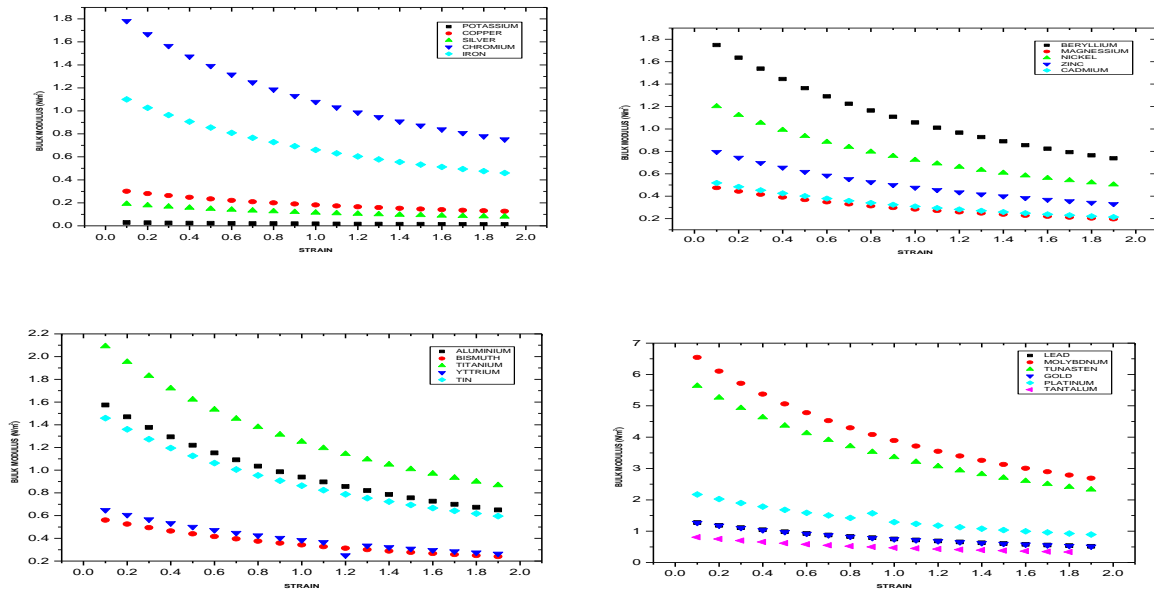


Figure 3:- Variation of Bulk Modulus with Deformation for some Metals

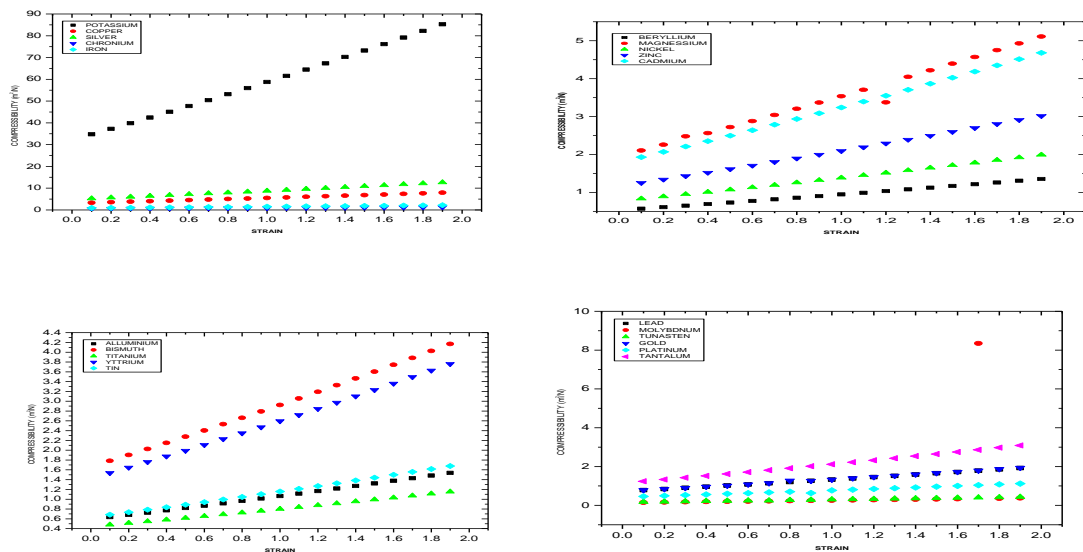


Figure 4:- Variation of Compressibility with Deformation for Some Metals.

Table 1: Bulk modulus of Deformed Metals (Nm^{-2})

Metals	r_s (a.u)	Strain								
		0.2	0.4	0.6	0.8	1.0	1.2	1.4	1.6	1.8
K	4.96	0.0268	0.0236	0.0210	0.0188	0.0170	0.0155	0.0142	0.0131	0.0122
Cu	2.67	0.281	0.248	0.221	0.200	0.181	0.166	0.152	0.141	0.131
Ag	3.02	0.178	0.157	0.140	0.126	0.114	0.104	0.0958	0.0884	0.0821
Be	1.87	1.636	1.445	1.291	1.164	1.058	0.967	0.890	0.823	0.765
Mg	2.65	0.443	0.390	0.347	0.312	0.283	0.258	0.237	0.219	0.203
Cr	1.86	1.688	1.474	1.317	1.187	1.079	0.987	0.908	0.840	0.780
Fe	2.12	1.027	0.906	0.808	0.728	0.661	0.604	0.555	0.513	0.476
Ni	2.07	1.123	0.991	0.884	0.796	0.723	0.661	0.607	0.561	0.521
Zn	2.31	0.745	0.656	0.585	0.526	0.477	0.436	0.401	0.370	0.343
Cd	2.59	0.483	0.425	0.379	0.341	0.309	0.282	0.259	0.239	0.222
Al	2.07	1.469	1.294	1.152	1.036	0.939	0.857	0.787	0.727	0.674
Bi	2.25	0.525	0.465	0.416	0.376	0.342	0.313	0.289	0.267	0.248
Ti	1.92	1.952	1.720	1.533	1.379	1.251	1.142	1.049	0.969	0.899
Y	2.61	0.607	0.533	0.474	0.426	0.385	0.351	0.322	0.297	0.276
Sn	2.22	1.360	1.195	1.061	0.949	0.864	0.788	0.723	0.667	0.618
Pb	2.30	1.187	1.043	0.927	0.832	0.754	0.687	0.630	0.581	0.539
Mo	1.61	6.107	5.372	4.781	4.296	3.892	3.551	3.259	3.008	2.789
W	1.62	5.260	4.630	4.123	3.707	3.360	3.067	2.817	2.600	2.412
Au	2.39	1.183	1.039	0.923	0.828	0.749	0.683	0.626	0.577	0.535
Pt	2.00	2.028	1.784	1.587	1.426	1.292	1.179	1.082	0.998	0.926
Ta	2.84	0.750	0.658	0.584	0.523	0.473	0.431	0.394	0.363	0.336

Table 2: Compressibility of Deformed Metals (m^2N)

Metals	r_s (a.u)	Strain								
		0.2	0.4	0.6	0.8	1.0	1.2	1.4	1.6	1.8
K	4.96	37.259	42.419	47.725	53.169	58.742	64.439	70.253	76.179	82.212
Cu	2.67	3.561	4.033	4.517	5.012	5.518	6.034	6.559	7.094	7.638
Ag	3.02	5.632	6.388	7.163	7.957	8.769	9.597	10.442	11.301	12.175
Be	1.87	0.611	0.692	0.775	0.859	0.946	1.033	1.123	1.215	1.308
Mg	2.65	2.257	2.565	2.881	3.205	3.536	3.875	4.220	4.571	4.929
Cr	1.86	0.599	0.678	0.759	0.842	0.927	1.013	1.101	1.191	1.282
Fe	2.12	0.973	1.104	1.237	1.374	1.514	1.657	1.802	1.950	2.101
Ni	2.07	0.891	1.009	1.131	1.256	1.384	1.514	1.647	1.782	1.919
Zn	2.31	1.343	1.524	1.710	1.900	2.095	2.294	2.496	2.703	2.913
Cd	2.59	2.069	2.351	2.640	2.936	3.239	3.549	3.865	4.187	4.514
Al	2.07	0.681	0.773	0.868	0.965	1.065	1.167	1.270	1.376	1.483
Bi	2.25	1.904	2.151	2.403	2.661	2.924	3.193	3.466	3.744	4.026
Ti	1.92	0.512	0.581	0.652	0.725	0.800	0.876	0.953	1.032	1.112
Y	2.61	1.649	1.876	2.110	2.350	2.595	2.846	3.102	3.362	3.628
Sn	2.22	0.735	0.837	0.941	1.048	1.157	1.269	1.383	1.499	1.618
Pb	2.30	0.843	0.959	1.079	1.201	1.327	1.455	1.586	1.720	1.856
Mo	1.61	0.164	0.186	0.209	0.233	0.257	0.282	0.307	0.332	0.359
W	1.62	0.190	0.216	0.243	0.270	0.298	0.326	0.355	0.385	0.415
Au	2.39	0.845	0.963	1.084	1.208	1.335	1.465	1.597	1.733	1.870
Pt	2.00	0.493	0.561	0.630	0.701	0.774	0.848	0.924	1.002	1.0804
Ta	2.84	1.333	1.520	1.713	1.912	2.115	2.323	2.535	2.752	2.972

Table 3: Calculated Bulk Modulus and Compressibility of Undeformed Metals and their Experimental values obtained from Kittel (1976)

Metals	$r_s(\text{a.u.})$	Cal. Bulk Modulus $\text{Nm}^{-2} \times 10^{11}$	Exp. Bulk Modulus $\text{Nm}^{-2} \times 10^{11}$	Cal. Compressibility $\times 10^{-11} \text{m}^2/\text{N}$	Exp. Compressibility $\times 10^{-11} \text{m}^2/\text{N}$
K	4.96	0.031	0.032	32.25	31.00
Cu	2.67	0.322	1.37	3.10	0.73
Ag	3.02	0.204	1.007	4.90	0.99
Be	1.87	1.876	1.003	0.53	1.00
Mg	2.65	0.511	0.354	1.96	2.82
Cr	1.86	1.914	1.901	0.52	0.53
Fe	2.12	1.181	1.683	0.85	0.59
Ni	2.07	1.290	1.86	0.78	0.54
Zn	2.31	0.857	0.598	1.17	1.67
Cd	2.59	0.557	0.467	1.80	2.14
Al	2.07	1.693	0.722	0.59	1.39
Bi	2.25	0.601	0.315	1.66	3.17
Ti	1.92	2.247	1.051	0.45	0.95
Y	2.61	0.700	0.366	1.43	2.73
Sn	2.22	1.570	1.11	0.64	0.90
Pb	2.30	1.370	0.430	0.73	2.33
Mo	1.61	7.042	2.725	0.14	0.37
W	1.62	6.060	3.232	0.17	0.31
Au	2.39	1.367	1.732	0.73	0.99
Pt	2.00	2.339	2.783	0.43	0.36
Ta	2.84	0.869	2.00	1.15	0.50

Conclusion:-

In this work, a generalized approach for computing and studying the effect of linear deformation on bulk modulus and compressibility of metals based on the structureless pseudopotential formalism is presented. The results obtained for bulk modulus and compressibility of undeformed metals were in agreement with the experimental values which shows the validity of the model used in the computation. There is high concentration of electron in the high density region than the low density region for both computed and experimental values of the bulk modulus. Metals in the high density region have low compressibility while metals in the low density region have high compressibility. These could be due to the variable electron contribution in the solid and the nature of their electronic state structure. The bulk modulus and compressibility of metals is strongly affected by deformation due to the trend exhibited by different metals during deformation (strain).

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RESEARCH ARTICLE

CARBON NANOTUBES: SYNTHESIS, PROPERTIES AND TECHNOLOGICAL APPLICATIONS.

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Abstract

This project reviews synthesis, properties and technological applications of carbon nanotubes. The different synthesis methods of carbon nanotubes are illustrated briefly. The energy dependence of density of states was used to determine the optical properties of CNT. Finally, brief description of optical and electronic properties was presented. Using MATLAB Code the theoretical simulation of electronic band structure as a function of wave vector, the band gap as a function of tube diameter and the density of state as a function of energy band are generated.

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Introduction:-

The history of nanotechnology is generally understood to have begun in December 1959 when physicist Richard Feynman gave a speech, "There are Plenty of Rooms at the Bottom" at an American Physical Society meeting at the California Institute of Technology in which he identified the potential of nanotechnology. Feynman said it should be possible to build machines small enough to manufacture objects with atomic precision, and that if information could be written on an atomic scale, "all of the information that man has carefully accumulated in all the books in the world can be written in a cube of material 12700 nm wide about the size of the smallest piece of dust visible to the human eye."

Nanotechnology and Nanoscience got a boost in the early 1980s with two major developments: the birth of cluster science and the invention of the scanning tunneling microscope (STM). This development led to the discovery of fullerenes in 1985 (Prasada *et al.*, 2008) and carbon nanotube a few years later. In another development, the synthesis and properties of semiconductors was studied; this led to a fast increasing number of metal and metal oxide nanoparticles and quantum dots.

Nanotechnology as one of the most modern technologies has attracted many researchers all over the world to study the different aspects of nanosystems and nanostructures. Among various nanostructures, carbon nanotube (CNT) has an influential role in this technology due to its remarkable mechanical, electrical, thermal and chemical properties. This nanostructure has been employed in different fields including nanomaterial, nanoelectronics and nanomechanics during recent decades. Due to these extensive applications, its modeling and analysis in various conditions are more demanded (Fakhrabadi *et al.*, 2012).

Carbon nanotubes (CNTs) are tubular structures that are typically of nanometer diameter and many micrometers in length. This fascinating new class of materials was first observed by Endo in 1975 and later by Iijima in 1991 in the

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soot produced in the arc-discharge synthesis of fullerenes (Dresselhaus *et al.*, 2004). The structure of a CNT can be described in terms of a single graphite layer (graphene) rolled up to form a single cylinder or concentrically arranged cylinders. The former is referred to as a single-wall nanotube (SWNT) and the latter are called multi-wall nanotubes (MWNTs). MWNTs can form starting from double-walled tubes (DWNTs) to tubes with 50 or more walls or layers.

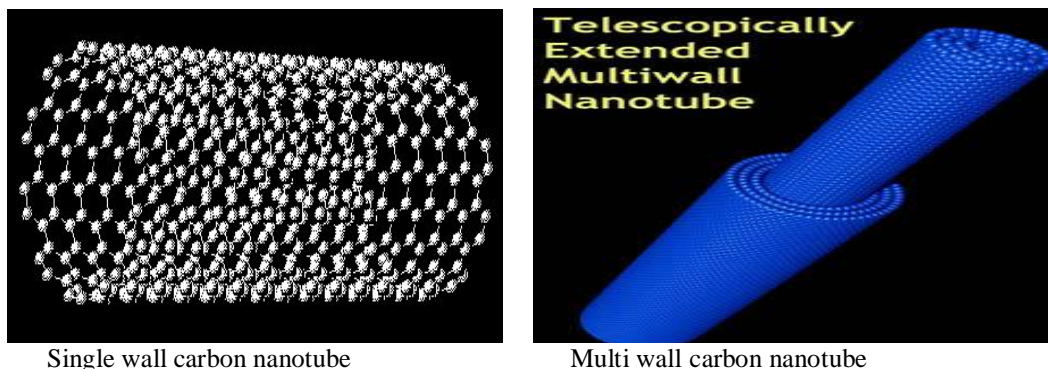


Figure 1:- Structure of single-wall and multi-wall carbon nanotubes (Zhang *et al.*, 2003).

Single-wall nanotubes has only one single layer and with a diameter of 1 to 5 nm. The properties of SWNT are more stable than MWNT so it is more favorable. This is due to the degree of relative stability for carbon CNT bundle interconnects versus the length bundle and the diameter of each individual CNT has been obtained. The obtained results show that with increasing the length of the CNT bundle and the diameter of each individual CNT to be interconnected, then the relative stability increase, and the system become more stable (Fathi, 2009). MWNT is a little bigger than SWNT because MWNT has about 50 layers. MWNT's inner diameter is from 1.5 to 15 nm and the outer diameter is from 2.5 nm to 30 nm. SWNT have better defined shapes of cylinder than MWNT, thus MWNT has more possibilities of structure defects and its nanostructure is less stable (Zhang *et al.*, 2003).

It is a well-known fact that much of the Carbon Nanotubes produced are usually formed along with a mixture of amorphous carbon, fullerenes and other impurities. Cost effective and efficient ways of separation and filtration of Carbon Nanotubes are yet to be discovered. Amongst the available methodologies the three most commonly used techniques includes Arc-Discharge method, Laser Ablation method and Chemical Vapor Deposition (CVD). The CVD has proved to be a comparatively reliable way of synthesizing CNTs (Tripathi *et al.*, 2010).

Within material science, the optical properties of carbon nano tubes refer specifically to the absorption, photoluminescence and Raman spectroscopy. Spectroscopic methods offer the possibility of quick and non-destructive characterization of relatively large amounts of carbon nanotubes. There is a strong demand for such characterization from the industrial point of view: since numerous parameters of the nanotube synthesis can be changed, intentionally or unintentionally, to alter the nanotube quality. Optical absorption, photoluminescence and Raman spectroscopes allow quick and reliable characterization of this nanotube quality in terms of non-tubular carbon content, structure (chirality) of the produced nanotubes, and structural defects. Those features determine nearly any other properties such as optical, mechanical, and electrical properties.

General Objective of this work being reported here was:-

- To review the synthesis, properties and application of carbon nanotubes

Specific Objectives were:-

- ❖ To explain optical and electronic properties of carbon nanotubes
- ❖ To review the applications of carbon nanotubes

Literature Review:-

Fabrication of Carbon Nanotubes:-

Following the rapid advancement of carbon nanotubes and applications, different techniques to produce carbon nanotubes have been developed. Carbon nanotubes can be fabricated using techniques such as arc discharge, laser ablation, and chemical vapor deposition (CVD) methods.

Arc discharge method:-

This was the first available method to fabricate carbon nanotubes. An electric arc discharge is an electrical breakdown of a gas producing a plasma discharge, very similar to a spark, which is the flow of current through a nonconductive medium such as air or an insulator. In a conventional carbon arc discharge fabrication, a plasma discharge is generated in a small gap between two graphite electrodes. In this method, carbon nanotubes are produced at the core in the cathode deposit (Javey *et al.*, 2002).

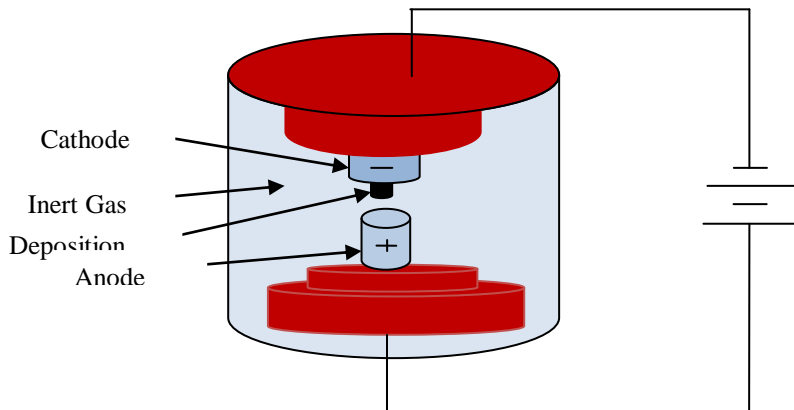


Figure 2:- Schematic diagram of an arc-discharge system

Source: <http://students.chem.tue.nl/ifp03/synthesis.html>.

As shown figure 2 during the arc discharge, two graphite rods are placed in an enclosure that is filled with some inert gas (like helium or argon) at low pressure (between 50 and 700 *mbar*). The carbon rods act as electrodes which are kept at different potentials. The anode is moved close to the cathode until an arc appears and the electrodes are kept at the distance of 1 *mm* for the whole duration of the process that takes about one minute. After the de-pressurization and cooling of the chamber, the nanotubes together with the by-products can be collected. Most nanotubes deposit on the cathode. The synthesis product yield which represents the amount of carbon nanotubes expected in the converted carbon is 60% (Isaacs *et al.*, 2010).

Laser ablation:-

Laser ablation is a process of removing materials from a solid surface by irradiating the surface with a laser beam. It was developed in 1995 to produce CNTs. Figure 3 illustrates the growth setup of laser ablation. In a typical laser ablation process for CNT growth, the graphite block is placed inside an oven and hit by a pulsed laser, and Argon gas is pumped along the direction of the laser point. The oven temperature is set approximately to 1200°C. As the laser ablates the target, carbon is vaporized and carried by the flowing gas onto a cool copper collector. CNTs grow on the cooler surfaces of the reactor as the vaporized carbon condenses. Sometimes, a water cooled surface may be included in the system to collect the CNTs. SWNTs are formed from a composite block of graphite and metal catalyst particles, whereas MWNTs form from pure graphite as the starting material (Guo *et al.*, 1995).

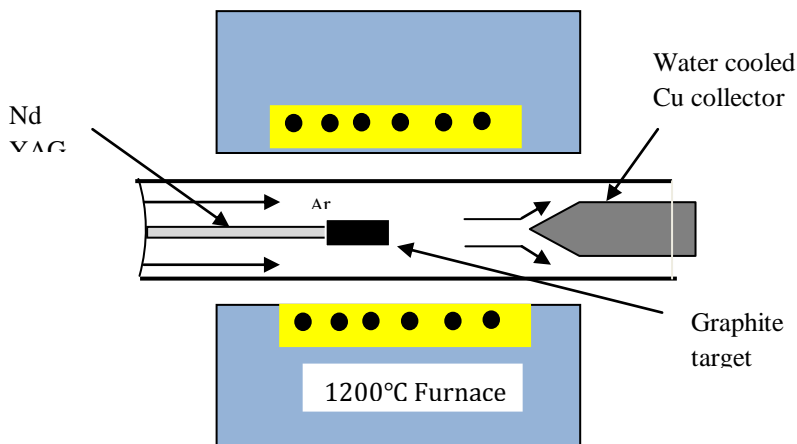


Figure 3:- Laser ablation equipment to produce CNTs.

Source: <http://students.chem.tue.nl/ifp03/synthesis.html>

Disadvantages of Arc-Discharge and Laser ablation:-

Both arc-discharge and laser ablation produce some of the most high quality nanotubes but suffer from the following disadvantages, which limit their use as large scale industrial processes.

- ✓ Both need extensive amount of energy to produce nanotubes. Such a huge amount of energy is not only impossible but also uneconomical for large scale production.
- ✓ Both methods require solid carbon/graphite as a target which has to be evaporated to get nanotubes. It is difficult to get such large graphite to be used as target in industrial process which limits its exploitation as large scale process.
- ✓ Both processes grow nanotubes in highly tangled form, mixed with unwanted form of carbon or catalysts. Thus CNTs produced by these processes require purification to get purified and assembled forms. The designing of such refining processes is difficult and expensive.

All the above mentioned factors severely limit the use of both arc-discharge and laser ablation as large scale processes for production of carbon nanotubes. In order to address these issues other methods were developed.

Chemical vapor deposition (CVD) grown carbon nanotubes:-

During CVD, a substrate covered with metal catalysts, like nickel, cobalt, iron, or a combination is heated to approximately 700°C. As shown figure 4, the growth starts after two gases are passed through the chamber, a carrier gas like nitrogen, hydrogen or argon, and some hydrocarbon gas like acetylene (C_2H_2) or methane (CH_4). The synthesis production yield, which indicates the amount of carbon nanotubes in the converted carbon, reaches 90% (Isaacs *et al.*, 2010). CVD is commonly used for industrial purposes because the method is already well investigated and offers acceptable results on the industrial-scale.

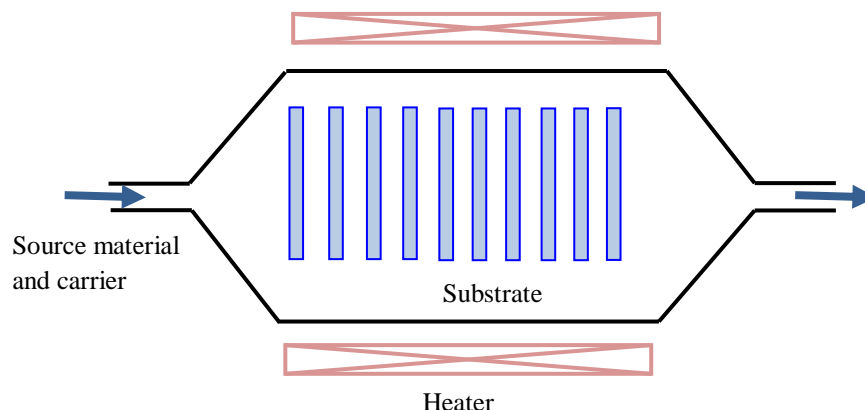


Figure 4:- Schematic diagram of chemical vapor deposition method.

Source: http://en.wikipedia.org/wiki/Chemical_vapor_deposition

Advantages of Chemical vapor deposition:-

This method has many advantages as compared to the arc discharge and the laser ablation method. To mention some:

- Reaction process and reactor design is simple, reaction is easy to control and manipulate.
- Raw materials are abundant and available readily in the form of gases.
- The process does not need huge amount of energy to produce high quality and large quantity of carbon nanotubes.
- Process is capable of producing CNTs directly onto substrates which ease out the process of further collection and separation and eliminates post refining processes to a large extent. Some refining is required in some cases for further purification.
- Process is unique for the production of vertically aligned nanotubes. No other process can produce aligned nanotubes.

More and more research is underway in the world today for the production of large quantities of high purity carbon nanotubes by chemical vapor deposition process. Researchers are developing method and designing reactors, which could be utilized as units for large scale production of carbon nanotubes.

The major pitfall for CVD grown MWNTs is the high defect density in their structures. The defective nature of CVD-grown MWNTs remains to be thoroughly understood, but is most likely due to the relatively low growth temperature, which is not sufficient to provide adequate thermal energy to anneal nanotubes into perfectly crystalline structures.

Properties of Carbon Nanotubes:-

The wide range of electronic, optical, thermal, magnetic mechanical, chemical, physical and structural properties of carbon nanotubes vary according to the different diameter, chirality, length, and direction of 'twist' of the nanotube. Many applications arise from the surprising and desirable properties they exhibit, some of which are already being used in new and improved products. Among these properties of carbon nanotubes, optical and electronic properties are discussed in detail.

Optical properties of carbon nanotubes:-

The optical properties of CNTs are linked closely to their structural and electronic properties and are fully governed by one parameter, its chiral vector, which describes how the carbon-atom honeycomb is organized with respect to the CNT axis. Depending only on this parameter, CNTs may behave as a direct band gap semiconductor or as a metal.

Optical properties of carbon nanotubes derive from electronic transitions within one dimensional density of states (DOS). A typical feature of one-dimensional crystals is that their DOS is not a continuous function of energy, but it descends gradually and then increases in a discontinuous spike. The sharp peaks found in one-dimensional materials are called van Hove singularities. These van Hove singularities would be discussed later in the discussion part.

For an individual CNT, the optical properties of single walled (SWNTs and MWNTs should be considered separately. The optical properties of SWCNTs, especially those with diameters less than 1 nm, exhibit very strong dependence on the detailed atomic structure (chirality) (Bachilo *et al.*, 2002). Chirality-controlled CNT growth or chirality separation is still difficult in experiments. Thus SWCNT arrays usually contain much different chirality with a random distribution, which complicates the engineering of optical properties and functionalities. In contrast, MWCNTs, due to their larger size, have more regular and uniform optical properties. It is thus reasonable to treat each MWCNT as a homogeneous medium and design the structure for different applications (Lidorikis and Ferrari, 2009).

The optical properties of a CNT array further depend on the manner in which the CNTs are arranged, including pattern and intertube distance. Vertical arrays of nanotubes and nanowires have attracted much attention recently in solar, photonic crystal, mode-locked fiber lasers, and nanoantenna applications. For example, absorption enhancement in silicon nanowire arrays has been predicted using electromagnetic calculations and observed in experiments. Also, anisotropic optical scattering is predicted and observed for an individual semiconductor nanowire. In comparison, MWNT arrays possess a similar cylinder array structure. Although enhanced absorption and anisotropic scattering are experimentally observed, they have been less investigated theoretically, and quantitative analysis is still lacking (Ruan *et al.*, 2010).

Raman spectroscopy:-

Raman spectroscopy is one of the most powerful tools for characterization of carbon nanotubes. Without sample preparation, a fast and non-destructive analysis is possible. All allotropic forms of carbon are active in Raman spectroscopy (Arepalli *et al.*, 2004): fullerenes, carbon nanotubes, amorphous carbon and polycrystalline carbon.

Raman spectroscopy provides information about molecular vibrations that can be used for sample identification and quantification. The technique involves shining a monochromatic light source that is laser on a sample and detecting the scattered light. The majority of the scattered light is of the same frequency as the excitation source; this is known as Rayleigh or elastic scattering. A very small amount of the scattered light is shifted in energy from the laser frequency due to interactions between the incident electromagnetic waves and the vibration energy levels of the molecules in the sample.

Raman scattering offers a unique tool to characterize carbon materials such as graphite, diamond and carbon nanotubes, since the amount of ordering and degree of sp^2 and sp^3 bonding leaves a unique Raman fingerprint. Therefore, Raman spectroscopy is used here to identify carbon nanotubes and to characterize the obtained carbon structures by their degree of crystallinity.

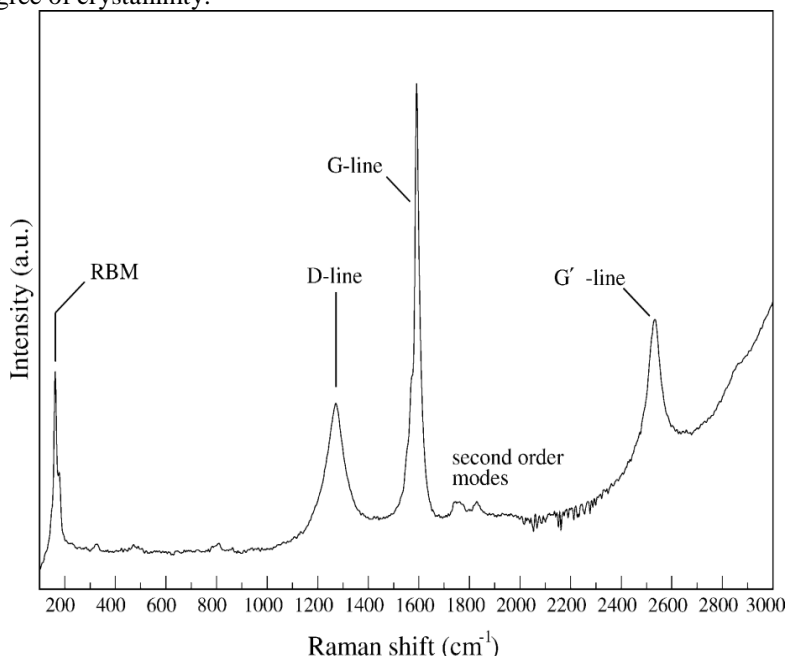


Figure 5:- Raman spectrum showing the most characteristic features of CNTs: Radial Breathing Mode (RBM), the D band, G' band and G band (Belin and Epron, 2005)

From figure 5 Raman spectrums showing the most characteristic features of CNTs are summarized as follows:

- A low-frequency peak $< 200 \text{ cm}^{-1}$ (or bunch of peaks for poly disperse samples) when resonating conditions were met characteristic of the SWNT assigned to an A_{1g} breathing mode of the tubes, whose frequency depends essentially on the diameter of the tube (RBM: radial breathing mode).
- A large structure (1340 cm^{-1}) assigned to residual ill-organized graphite, the so-called D-line (D: disorder).
- The D- line or D-band is observed in the Raman spectra of semiconducting and metallic SWNT at the single nanotube level. The D-band in graphite involves scattering from a defect which breaks the basic symmetry of the graphene sheet, it is observed in sp^2 carbons containing porous, impurities or other symmetry-breaking defects.
- A high-frequency bunch (between 1500 and 1600 cm^{-1}) called G band also characteristic of nanotubes, corresponding to a splitting of the E_{2g} stretching mode of graphite.
- A second order observed mode between 2450 and 2650 cm^{-1} assigned to the first overtone of the D mode and often called G mode; a combination mode of the D and G modes between 2775 and 2950 cm^{-1} .

Radial breathing mode (RBM) corresponds to radial expansion-contraction of the nanotube. Therefore, its frequency ν_{RBM} (in cm^{-1}) depends on the nanotube diameter d (in nm) and can be estimated (Fantini *et al.*, 2004).

$\nu_{\text{RBM}} = \frac{234}{d} + 10$ for SWNT or $\nu_{\text{RBM}} = \frac{234}{d}$ for DWNT, which is very useful in deducing the CNT diameter from the RBM position.

Raman spectroscopy for the RBM is therefore often used to determine the diameter or diameter distribution in SWNT samples and is further used to assign the chiral index (n, m) of individual SWNTs by considering their resonant transitions.

Nonlinear optical properties:-

Non-linear optics (NLO) is the branch of optics that describes the behavior of light in non-linear media, that is media in which the dielectric polarization \mathbf{P} responds nonlinearly to the electric field \mathbf{E} of the light. This nonlinearity is typically only observed at very high light intensities (values of the electric field comparable to interatomic electric fields, typically 10^8 V/m) such as those provided by pulsed lasers.

The rapid development of nanoscience and nanotechnology provides lots of new opportunities for nonlinear optics. A growing number of nanomaterials have been shown to possess remarkable nonlinear optical (NLO) properties, which promotes the design and fabrication of nano and nanoscale optoelectronic and photonic devices (Hasan *et al.*, 2009; Loh *et al.*, 2010). The wonderful carbon allotropes discovered in recent decades are the most representative products of nanotechnology: from 3D carbon nanoparticles (graphite), to 0D fullerenes, to 1D carbon nanotubes (CNTs), and then to 2D graphenes discovered most recently. Interestingly, all of these nano-carbons exhibit diverse NLO properties. For instance, carbon black suspensions show strong thermally-induced nonlinear scattering (NLS) effect and hence optical limiting (OL). For intense ns laser pulses fullerenes show large third-order optical nonlinearity and reverse saturable absorption (RSA) at certain wavelength band. CNTs show ultrafast second- and third order nonlinearities and saturable absorption (SA) in the near infrared (NIR) region and graphenes show ultrafast carrier relaxation time and ultra-broad-band resonate NLO response.

Materials with large second or third order optical nonlinearities are of great interest for data processing in photonic computing. Since the invention of laser in 1960 by Maiman and the first discovery of nonlinear optical (NLO) effect of quartz crystals, there has been a great deal of research directed at finding new materials with higher order NLO properties. Only noncentrosymmetric or polar materials could possess second order NLO susceptibility. Most third order NLO materials are generally too low for practical applications. Since the discovery of carbon nanotubes, which possess optimal properties (like highest thermal conductivity, Young modulus 100 times higher than steel, zero bandgap metal, ballistic conductance, simple structure, nanometer-size and ultra-light weight) by a NEC scientist Sumio Iijima in 1991, a heat of studying and engineering the properties of carbon nanotubes has been raised worldwide.

Electronic properties of carbon nanotubes:-

Carbon nanotubes (CNTs) provide a number of unique and special properties that suggest great promise for nanoelectronics applications. In particular, the high electrical conductivity of quantum wires provides a potential solution for on chip interconnect metals and transistors of future integrated circuits.

One crucial obstacle to overcome in fabrication is controlling whether the CNT is metallic or semiconducting. The critical parameter determining the electronic properties of CNTs is the chiral vector, $c_h = na_1 + ma_2 \equiv (n, m)$, where n and m are integers and a_1 and a_2 are the real space unit vectors of the graphene sheet. When $n - m$ is a multiple of 3, the simple theory leads to a crossing of bands at the Fermi energy, implying that CNT is metallic; otherwise, it is expected to be a semiconductor. Thus, armchair (n, n) , CNTs are expected to always be metallic, whereas zigzag $(n, 0)$, CNTs are semiconductor except when n is a multiple of 3.

Since a carbon nanotube (CNT) is a rolled graphene sheet, the band structure of CNT is also predetermined by electronic structure of graphene. However in contrast to graphene, CNT is a quasi-one-dimensional structure, in which electronic properties depends on how the graphene sheet was rolled up. To classify the variety of ways to wrap the nanotube from a graphene sheet two indexes, n and m are commonly used (Saito *et al.*, 2000). Figure 6 illustrates an example of graphene sheet about to be wrapped into a nanotube with the primitive translation vectors a_1 and a_2 . once wrapped, the length of the chiral vector becomes the tube circumference and the tube diameter d can be calculated from this circumference as:

$$d = \frac{a}{\pi} \sqrt{n^2 + nm + m^2} \quad (2.2.1)$$

The chiral angle is the angle between the translation vector c_h and the $(n, 0)$ line as seen in figure 7:

$$\alpha = \arctan\left(\frac{\sqrt{3}n}{2m+n}\right) \quad (2.2.2)$$

Defined by the integer index pair, (n, m) , the chiral angle ranges from 0 to 30°. Zigzag nanotubes have chiral angles of 0 degrees, armchair nanotubes have chiral angles of 30° and chiral nanotubes have chiral angles between 0 < α < 30°.

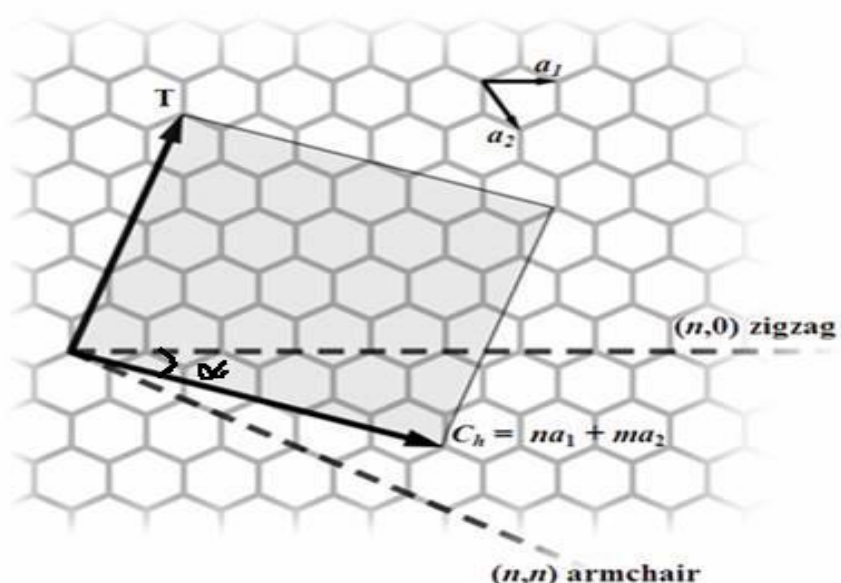


Figure 6:- Graphene model of a CNT showing the index pair notation used to characterize the CNT geometry.
Source: [http://en.wikipedia.org/wiki/ Carbonnanotube](http://en.wikipedia.org/wiki/Carbonnanotube)

$a = 2.46\text{\AA}$ from the graphene structure and by convention $n \geq m$. If $m = 0$, the nanotubes are called zigzag, while if $n = m$, the nanotubes are called armchair. Otherwise, they are called chiral. Therefore varying the way of the rolling up one can arrive at CNT of different chirality and hence to different periodicity of the electronic wavefunction. In the electronic spectrum, such a periodicity manifests itself as delta-shaped peaks known as van Hove singularities which positions depends on a diameter and chirality of the SWNT. In particular the diameter is responsible for the band gap or type (metal or semiconducting) of the nanotube (Kataura *et al.*, 1999). The electronic bands with the corresponding density of one-electron states (DOS) calculated for metallic and semiconducting single walled carbon nanotubes.

The electronic and optical properties of the tubes are considerably influenced by the environment (Charlier *et al.*, 2001). Under externally applied pressure, the small interaction between the tube walls results in the internal tubes experiencing reduced pressure (Puech *et al.*, 2004). The electronic transition energies are in the infrared and visible spectral range.

Generally, one can view carbon nanotubes as giant conjugated molecular wires with conjugation length corresponding to the whole length of the tube. In order to understand their electronic structure, we have to start with graphene, a single sheet of graphite. Carbon has four valence electrons of which three are strongly bound to neighbor atoms giving graphene its very high in-plane rigidity. The fourth electron is delocalized and shared by all the atoms, thus allowing for electronic current transport. However, because of its particular structure, graphene is electronically between semiconductor and metal. It is a semimetal or a zero-gap semiconductor.

Application of Carbon Nanotubes:-

A carbon nanotube is inert, has a high aspect ratio and a high tensile strength, low mass density, high heat conductivity, a large surface area, and a versatile electronic behavior, including high electron conductivity. However, while these are the main characteristics of individual nanotubes, many of them can form secondary structures such as ropes, fibers, papers and thin films with aligned tubes, all with their own specific properties. These properties make them ideal candidates for a large number of applications provided their cost is sufficiently low. The cost of carbon nanotubes depends strongly on both the quality and the production process. High-quality single-wall carbon nanotubes can cost 50–100 times more than gold. However, carbon nanotube synthesis is constantly improving, and sale prices are falling rapidly. The application of carbon nanotubes is therefore a very fast moving field, with new potential applications found every year, even several times per year.

Photovoltaic solar cells:-

Carbon nanotubes (CNTs) have been widely used in solar cells research, for example, CNTs have been integrated in organic photovoltaic devices both as an electron acceptor material and as a transparent electrode. This is because the high conductivity along the tube axis of CNTs helps carrier's separation and collection. In 2005, GE Global Research observed a photovoltaic effect in a pristine nanotube diode device consisting of two CNTs with different electrical properties (Lee, 2005).

Photovoltaic device is a device that converts the energy of light directly into electricity by the photovoltaic effect. It is a crucial part of solar cells. Currently, wafer-based silicon (single crystal, poly crystalline and multicrystalline) solar cells and thin film solar cells based on amorphous silicon, CdTe, CuInGaSe₂, and III-V semiconductors dominate photovoltaic manufacturing (Singh *et al.*, 2008; Gupta *et al.*, 2009). However, they are of low efficiency and expensive, which have limitations for replacement of current energy sources. There is a clear need to look for low-cost and high-efficiency solar cells. Many new kinds of solar cells have been proposed, such as p-n junction solar cells, dye-sensitized solar cells and organic solar cells.

Nanomaterials have been widely used in the above proposed solar cells. The advantages of using nanostructure-based solar cells are, on one hand, their high manufacturing costs as a result of using a low temperature process similar to printing instead of the high temperature vacuum deposition process typically used to produce conventional cells made with crystalline semiconductor material, and on the other hand, their low quantum efficiency which can be improved by using multiple electron-hole pair generation in nanostructures, like quantum dots and carbon nanotube (Hanna *et al.*, 2004; Jin Kim *et al.*, 2008; Stubbs *et al.*, 2010).

Recently, carbon nanotube (CNT) and graphene are playing an important role in such nanostructure-based solar cells. The dispersion of CNTs in a solution of an electron donating conjugated polymer is perhaps the most common strategy to implement CNT materials into organic photovoltaic devices to obtain higher efficiency. Enhancements of more than two orders of magnitude have been observed in the photocurrent from adding the single walled carbon nanotube to the poly matrix (Kymakis *et al.*, 2003). The use of CNTs in DSSC has doubled the efficiency of this kind of photoelectrochemical solar cells (Kongkanand *et al.*, 2007).

Graphene thin films also have been used as anode in dye-sensitized solar cells and organic solar cells layered graphene for photovoltaic devices have been reported to have an even better photovoltaic performance than the single walled carbon nanotube/quantum dots devices (Guo *et al.*, 2010).

As one of major variables of DSSC, the dye absorbed light energy transits from ground state to excited state and gets electron injection. Electrons are injected in a very fast speed of femtosecond or picoseconds unit and oxidized dye is renewed within several nanoseconds. On the other hand, since rejoining speed that the electron becomes dissolved into electrolyte via surface state is slow such as micro seconds or milliseconds and most of photons are injected semiconductor conduction band, those electrons that are not injected meet holes again to be restored and decrease efficiency of solar cell. If we utilize CNT that is advantageous for electrical and thermal conduction as a compound material, we can increase electron injection speed than rejoining speed and increase efficiency of dye sensitized solar cells through movement of more electrons.

Since multi walled carbon nanotube (MWNT) is a material to transfer electricity and heat well, it can function as a basic electrode. Nanotubes function as electrodes to penetrate into broad TiO₂ surface mutually and assist extracting charge carriers efficiently from dye layer. Since these electrodes are very clear under longer wavelength, they are advantageous for solar light spectrums.

Chemical sensors:-

The electrical conductance of semiconductor SWNTs was recently demonstrated to be highly sensitive to changes in the chemical composition of the surrounding atmosphere at room temperature, due to charge transfer between the nanotubes and the molecules from the gases adsorbed onto SWNT surfaces. It has also been shown that there is a linear dependence between the concentration of the adsorbed gas and the change in electrical properties, and that the adsorption is reversible. SWNT-based chemical NO₂ and NH₃ sensors are characterized by extremely short response times unlike conventional sensors (Kong *et al.*, 2000; Chang *et al.*, 2001).

The electrical response has been measured by exposing MWNT films to sub-ppm NO₂ concentrations (10–100 ppb in dry air) at different operating temperatures ranging between 25 and 215°C (Cantalini *et al.*, 2003). For SWNTs, the sensor responses are linear for similar concentrations, with detection limits of 44 ppb for NO₂ and 262 ppb for nitro toluene (Li *et al.*, 2003). High sensitivity to water or ammonia vapor has been demonstrated on a SWNT-SiO₂ composite (Varghese *et al.*, 2001). This study indicated the presence of p-type SWNTs dispersed among the predominantly metallic SWNTs, and that the chemisorptions of gases on the surface of the semiconductor SWNTs is responsible for the sensing action.

Generally speaking, the sensitivities of these new nanotube-based sensors are three orders of magnitude higher than those of standard solid state devices. Another reason for using nanotubes instead of current sensors is their simplicity, the facts that they can be placed in very small systems and that they can operate at room temperature, as well as their selectivity. These advantages allow a limited number of sensor device architectures to be built for a variety of industrial purposes, while the current technology requires a large variety of devices based on mixed metal oxides, optomechanics, catalytic beads, electrochemistry, and so on.

Multifunctional Materials:-

One of the major benefits expected from incorporating carbon nanotubes into other solid or liquid materials is that they endow the material with some electrical conductivity while leaving other properties or behaviors unaffected. As already mentioned in the previous section, the percolation threshold is reached at very low nanotube loadings. Tailoring the electrical conductivity of a bulk material is then achieved by adjusting the nanotube volume fraction in the formerly insulating material while making sure that this fraction is not too large. As demonstrated by (Maruyama and Alam, 2002), there are three areas of interest regarding the electrical conductivity:

- ❖ Electrostatic discharge (for example, preventing fire or explosion hazards in combustible environments or perturbations in electronics, which requires an electrical resistivity of less than 10¹² Ωcm).
- ❖ Electrostatic painting (which requires the material to be painted to have enough electrical conductivity – an electrical resistivity below 10⁶ Ωcm to prevent the charged paint droplets from being repelled).
- ❖ Electromagnetic interference shielding (which is achieved for an electrical resistivity of less than 10 Ωcm).

Materials are often required to be multifunctional; for example, to have both high electrical conductivity and high toughness, or high thermal conductivity and high thermal stability. An association of several materials, each of them bringing one of the desired features, generally meets this need. The exceptional features and properties of carbon nanotubes make them likely to be a perfect multifunctional material in many cases. For instance, materials used in satellites are often required to be electrical conductive, mechanically self-supporting, able to transport away excess heat, and often to be robust against electromagnetic interference, while being of minimal weight and volume. All of these properties should be possible with a single nanotube containing composite material instead of complex multilaterals combining layers of polymers, aluminum, copper, and others.

The special nature of carbon combines with the molecular perfection of single-wall CNTs to endow them with exceptional material properties, such as very high electrical and thermal conductivity, strength, stiffness, and toughness. No other element in the periodic table bonds to itself in an extended network with the strength of the carbon-carbon bond. The delocalized pi-electron donated by each atom is free to move about the entire structure, rather than remain with its donor atom, giving rise to the first known molecule with metallic-type electrical conductivity. Furthermore, the high-frequency carbon-carbon bond vibrations provide an intrinsic thermal conductivity higher than even diamond.

In most materials, however, the actual observed material properties - strength, electrical conductivity, etc. - are degraded very substantially by the occurrence of defects in their structure. For example, high-strength steel typically fails at only about 1% of its theoretical breaking strength. CNTs, however, achieve values very close to their theoretical limits because of their molecular perfection of structure. This aspect is part of the unique story of CNTs. CNTs are an example of true nanotechnology: they are only about a nanometer in diameter, but are molecules that can be manipulated chemically and physically in very useful ways. They open an incredible range of applications in materials science, electronics, chemical processing, energy management, and many other fields.

CNTs Ceramic Applications, CNTs Air and Water Filtration, CNTs Biomedical Applications, CNT Catalyst Supports, CNTs Fibers and Fabrics, CNTs Structural Composites, CNTs Thermal Materials, CNTs Molecular Electronics, CNTs Conductive Adhesives and Connectors, CNTs Energy Storage, CNTs Conductive Plastics, CNTs

Field Emission Applications, and CNTs Thermal Conductivity are the application of this nanotube which is responsible for the current manufacturing industry, agricultural industry and also many other industry.

Other CNT Applications:-

There is a wealth of other potential applications for CNTs, such as solar collection; nanoporous filters; catalyst supports; and coatings of all sorts. There are almost certainly many unanticipated applications for this remarkable material that will come to light in the years ahead, and which may prove to be the most important and valuable ones of all. Many researchers are looking into conductive and or water proof paper made with CNTs. CNTs have also been shown to absorb Infrared light and may have applications in the I/R Optics Industry.

Electronic Band Structure of Carbon Nanotubes:-

In order to examine the electronic structure of carbon nanotubes, it is necessary to define the structural configuration. Carbon nanotube structures are defined by the indices (n, m) inscribed in the chiral vector. The chiral vector is a vector along the perimeter of the carbon nanotube.

Crystal lattice:-

The primitive cell of a carbon nanotube can be described from the unit vectors:

$$\vec{a}_1 = \frac{\sqrt{3}}{2}a\hat{x} + \frac{1}{2}a\hat{y} \quad (2.4.1)$$

$$\vec{a}_2 = \frac{\sqrt{3}}{2}a\hat{x} - \frac{1}{2}a\hat{y} \quad (2.4.2)$$

Where \vec{a}_1 and \vec{a}_2 are the unit cell vectors, a is the lattice constant of graphene with the value $a = \sqrt{3}a_{c-c} = 2.46 \text{ \AA}$, Where a_{c-c} is the nearest distance between two carbon atoms.

Reciprocal lattice:-

The reciprocal lattice vectors \vec{b}_1 and \vec{b}_2 are of the form:

$$\vec{b}_1 = \frac{2\pi}{\sqrt{3}a}\hat{x} + \frac{2\pi}{a}\hat{y} \quad (2.4.3)$$

$$\vec{b}_2 = \frac{2\pi}{\sqrt{3}a}\hat{x} - \frac{2\pi}{a}\hat{y} \quad (2.4.4)$$

Energy dispersion relation:-

The energy dispersion relation for carbon nanotubes can be calculated from the electronic structure of graphene. The energy dispersion of graphene is given by (Heo, 2008):

$$E(k) = \pm\gamma\sqrt{1 + 4\cos\left(\frac{\sqrt{3}ak_y}{2}\right)\cos\left(\frac{ak_x}{2}\right) + 4\cos^2\left(\frac{ak_x}{2}\right)} \quad (2.4.5)$$

Where γ is the transfer integral and k is the wave vector, k_x and k_y are denote the allowed reciprocal wave vectors along the tube and the circumference axis are found using the relation:

$$k_x = \frac{(2n+m)b_1 + (2m+n)b_2}{Nd_R} \quad (2.4.6)$$

$$k_y = \frac{mb_1 - nb_2}{N} \quad (2.4.7)$$

Where N is the number of hexagons within a unit cell and it is given by:

$$N = \frac{2(n^2 + m^2 + nm)}{d_R} \quad (2.4.8)$$

where d_R the greatest common divisor of $(2n + m)$ and $(2m + n)$.

One-dimensional (1D) energy band structure of carbon nanotubes can be adopted from Eq. (2.4.5). For armchair and zigzag carbon nanotubes the energy band structures as follows:

$$E_{1D,arm}(k) = \pm\gamma\sqrt{1 + 4\cos\left(\frac{q\pi}{n}\right)\cos\left(\frac{ka}{2}\right) + 4\cos^2\left(\frac{ka}{2}\right)} \quad (2.4.9)$$

where ka is $-\pi \leq ka \leq \pi$ and q is $0 \leq q \leq 2n - 1$,

$$E_{1D,zig}(k) = \pm\gamma\sqrt{1 + 4\cos\left(\frac{\sqrt{3}ka}{2}\right)\cos\left(\frac{q\pi}{n}\right) + 4\cos^2\left(\frac{q\pi}{n}\right)} \quad (2.4.10)$$

where ka is $-\frac{\pi}{\sqrt{3}} \leq ka \leq \frac{\pi}{\sqrt{3}}$ and q is $0 \leq q \leq 2n - 1$

where γ is the transfer integral and \pm corresponds to the valance and the conduction band of the nanotubes.

Energy gap of carbon nanotubes:-

Furthermore, the (n, m) integers also determine whether a CNT is metallic or semiconducting. When $n - m$ is a multiple of 3, the nanotube is metallic, otherwise the nanotube is semiconducting with an energy gap depending on its diameter as follows:

$$E_{g,v} = \sqrt{3}a\gamma \frac{2\pi}{na} \left(v - \frac{2n}{3} \right) = \sqrt{3}a\gamma \frac{2}{d} \left(v - \frac{2n}{3} \right) \quad (2.4.11)$$

where $d = \frac{na}{\pi}$ is tube diameter and $v = 1, \dots, 2n$

$$E_g = \frac{2\gamma a_{c-c}}{d} = \frac{0.72 \text{ eV}}{d(nm)} \quad (2.4.12)$$

where a_{c-c} is the nearest distance between two carbon atoms which is equal to 0.144 nm and d is the diameter carbon nanotubes. The density of states of carbon nanotubes are given by the following formula:

$$g(E) = \sum_v^{all \text{ band}} \frac{8}{3\pi a_{c-c}\gamma} \frac{|E|}{\sqrt{E^2 - E_{g,v}^2}} \quad (2.4.13)$$

This equation is valid for all semiconductor and metallic carbon nanotubes.

Discussion:-

From section 2.4. we have seen how the energy band structure depends on the wave vector of a given nanotubes. In order to identify whether the given nanotube is metallic or semiconducting, one can simulate the wave vector dependence of energy band structure of carbon nanotubes. With the help of equation (2.4.9) for armchair tubes and equation (2.4.10) for zigzag tubes, we simulate the variation of energy band structure of the tubes as a function of wave vector as showed in figure 7 respectively using MATLAB codes.

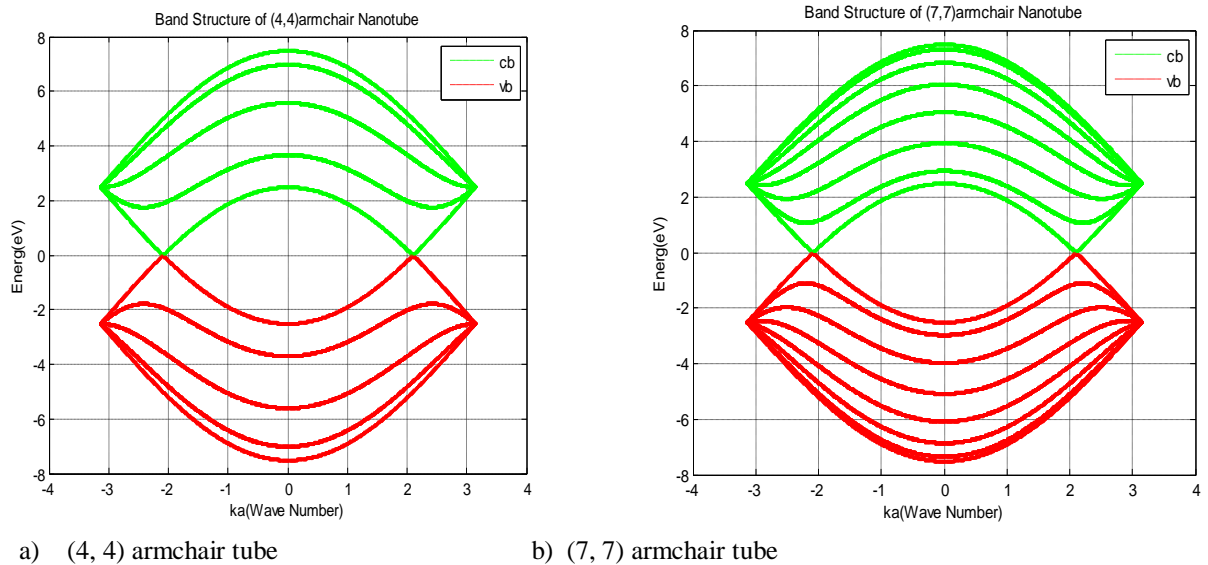


Figure 7:- Band structure of armchair tubes as function of wave vector using Eq. 2.4.9

From figure 7a we observe that for (4, 4) armchair tube has 10 different subbands; half of the total belongs to conduction and half of the total belongs to valence bands. From the half of conduction and/or valence bands two of them are non-degenerate and the remaining three subbands are degenerate. We also observe that the first conduction and the first valence bands meet at a Fermi level ($ka = \pm 2.032$). Because of the degeneracy point between the valence and conduction bands at the band crossing, the (4, 4) nanotube is thus a zero-gap semiconductor. This implies that the tube is metallic. This discussion is also true for (7, 7) armchair tubes as shown in figure 7b except the number of subbands and the degeneracy. In general, all (n, n) armchair nanotubes yield $2n$ conduction subbands and $2n$ valence bands, and of these bands, two are non-degenerate and $n - 1$ are doubly degenerate. They all have a band degeneracy between the highest valence band and the lowest conduction band at ($ka = \pm 2.032$) where the bands cross the Fermi level. The degeneracy comes from the two subbands with the same energy dispersion, but different q values. All armchair nanotubes are expected to exhibit metallic conduction.

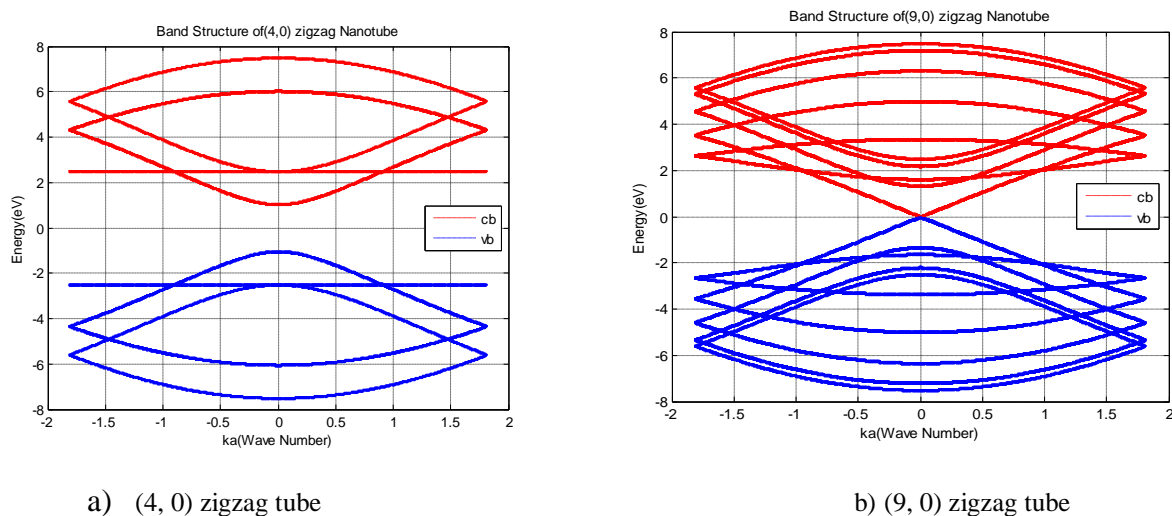


Figure 8:- Band structure of zigzag tubes as function of wave vector using Eq. 2.4.10

From figure 8a we observe that there are 10 sub bands in (4,0) tubes. Half of the total belongs to the conduction band (red color) and half of the total belongs to the valence band (blue color). We observe that the valence and the conduction bands are symmetric with respect to Fermi energy ($E = 0$). Again from figure 8a the valence bands and the conduction bands do not meet along the wave vector ka . Therefore, this tube is a semiconducting one since there is a band gap. In general most of the zigzag nanotubes are $(n, 0)$ a semiconducting. But when n is a multiple of 3, there is no energy gap, then the zigzag nanotubes are metallic, otherwise semiconducting. An example of metallic zigzag nanotube is (9,0). Because as we observed from figure 8b above, the first valence and conduction band meet at $ka = 0$. Simply there is no energy gap between the conduction and valence band in (9,0) zigzag tubes.

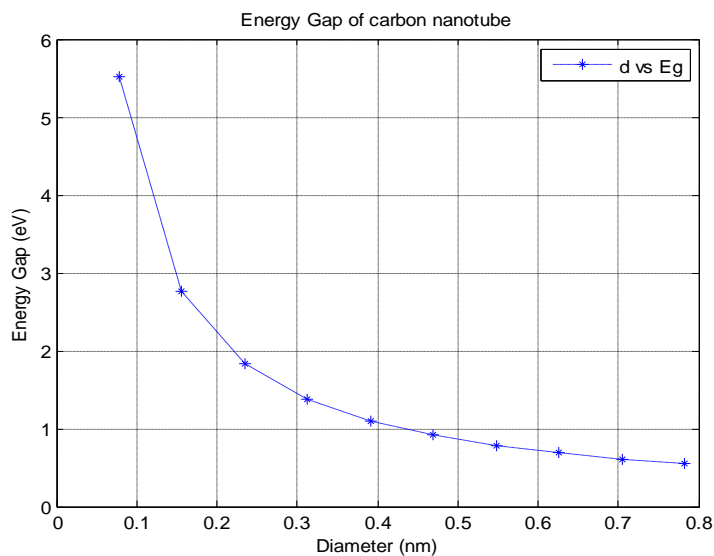
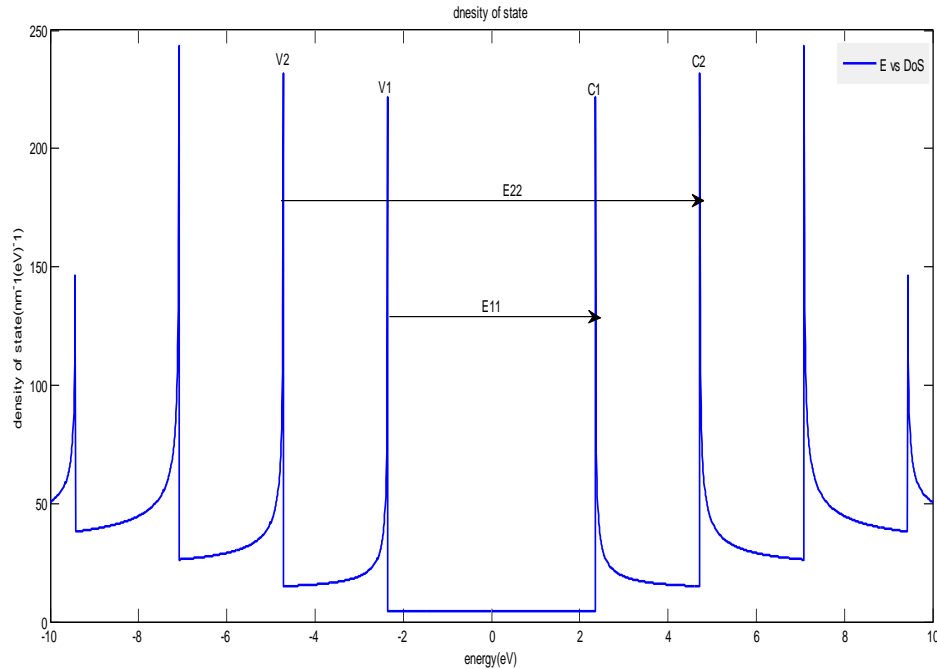
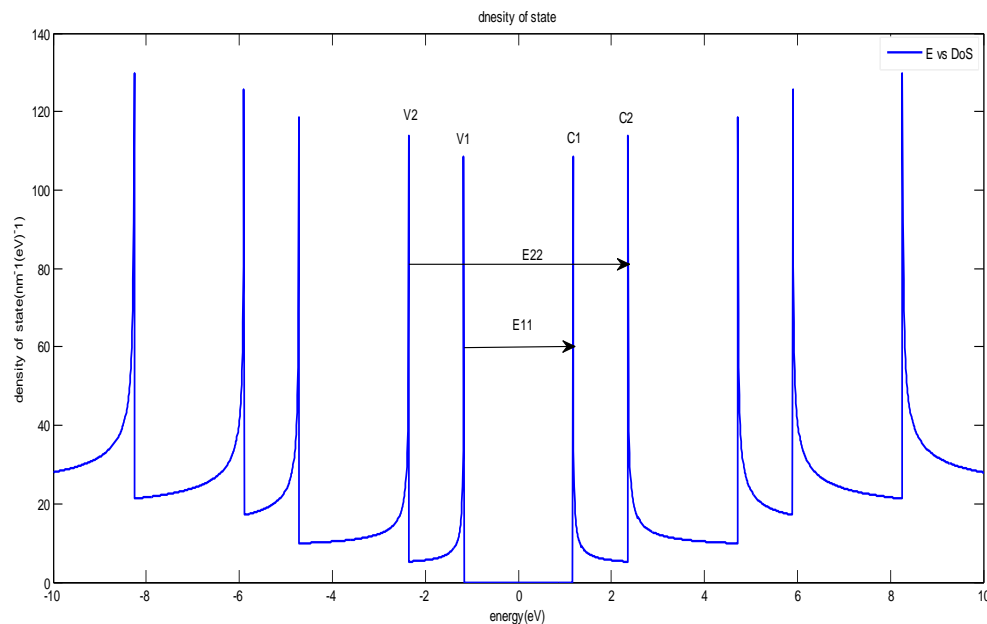


Figure 9:- Energy gap as a function of tube diameter using Eq. 2.4.11

From figure 9 we observe that the band gap for semiconducting tubes is found to be inversely proportional to the tube diameter. The energy gap decreases for larger tube diameters and increases for smaller diameters. Most of the time SWCTs have high energy gap when we compared to MWCTs. The diameter of MWCTs is greater than that of SWCTs. It has been shown that electronic conduction mostly occurs through the external tube for MWNTs.



a) Density of state for metallic tube



b) Density of state of semiconductor carbon nanotube

Figure 10:- Density of state as function of energy band structure of the carbon nanotubes using Eq. 2.4.13

From figure 10a we observe that at the Fermi energy (the highest occupied energy level) the density of states is finite for a metallic tube (though very small) and from figure 10b we observe that at the Fermi energy the density of state is zero for semiconducting tubes. As the energy increased, the sharp peaks in the density of states called the van Hove singularities, appear and specific energy levels. The density of state is highest at the energies where the band structure has a smallest slope and is lowest at the energies where the band structure is steepest. As we observe from the figures the density of state is not a continuous function of energy but it descends gradually and then increases in a discontinuous spike. It is clear that the density of state is inversely proportional to the square root of energy in both

tubes, (i.e., semiconducting and metallic tubes). Van Hove singularities results a remarkable optical properties of carbon nanotubes:

The energies between the van Hove singularities depend on the nanotube structure. Thus by varying this structure, one can tune the optoelectronic properties of carbon nanotube. Such fine tuning has been experimentally demonstrated using UV illumination of polymer-dispersed CNTs (Iakoubovskii *et al.*, 2006).

Optical transitions occur between the $V_1 \rightarrow C_1$, $V_2 \rightarrow C_2$ and so on states of semiconducting or metallic nanotubes and are traditionally labeled as S_{22} , S_{22} , M_{11} , and so on or, if the conductivity of the tube is unknown or unimportant, as E_{11} , E_{22} and so on. Crossover transitions $V_2 \rightarrow C_1$, $V_1 \rightarrow C_2$ and so on are dipole-forbidden and thus are extremely weak, but they were possibly observed using cross-polarized optical geometry (Miyauchi *et al.*, 2006).

Conclusion:-

In this project work synthesis, properties and technological applications of carbon nanotubes are reviewed. From the reviewed the synthesis methods chemical vapor deposition method is the most effective method which produces large quantities and high quality of carbon nanotubes. The technique is commonly used for the industrial purposes because the method is already well investigated and offers acceptable results on the industrial-scale.

The electronic band structure of a semiconductor consists of a valence band, conduction band, and Fermi level that are at energy levels particular to a given semiconductor. The Fermi level is the energy of the highest occupied state of electrons at absolute zero, and the valence band contains the valence electrons that when excited can move to the higher energy levels of the conduction band.

The electronic band structure calculations predict that the (n, m) indices determine the metallic or semiconducting behavior of CNT. It is clear that the result shows zigzag tubes $(n, 0)$ should have two distinct types of behavior; metallic when $n/3$ is an integer, and otherwise semiconductors. The gaps of the semiconducting $(n, 0)$ zigzag tubes should depend inversely on diameter and (n, n) armchair tubes band gap is remain independent of diameter.

The energy dependence of the nanotube density of states as where, the density of states for metallic and semiconductor zigzag nanotubes was compared. It was observed and demonstrated that the density of states near the Fermi level located at $E = 0$ are zero in pure semiconducting nanotubes. But density of states has a value of non-zero for metallic nanotubes even at the Fermi level located at $E = 0$.

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RESEARCH ARTICLE

EFFECT OF IMPELLER ROTATIONAL SPEED ON FLOW BEHAVIOR IN FULLY BAFFLED MIXING TANK.

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Abstract

This paper reports the results of numerical study undertaken to investigate the effect of impeller rotational speed on velocity field in mixing tank. The hydrodynamic of the flow in standard mixing tank generated by retreat curve 3-bladed impellers, (radial flow) is studied. Using ANSYS FLUENT v15.4 to solve the continuity and momentum equations incorporating the RNG K- ϵ turbulence model with the standard wall function available in Fluent. The multiple frames of reference (MFR) model is used for impeller modeling. The results show that the velocity fluctuated increased with the rotational speed.

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Introduction:-

Mixing tanks is one of the techniques that is widely used and play a significant role in industrial processes to homogenize the mixture of two or more fluids/solids by using rotating impeller. The better understanding of the fluids behavior in mixing vessel may improve the performance of impeller and the mixing process. Means e.g. the main aims of using missing is to improve the mass and heat transfer and to generate a homogenized mixture to minimize the settling of the particles at the bottom of the tank. A summary of some recent published works in the literature is given below.

(Wei et al 1997) [1] Studied the influence of the number and width of baffles in mechanically mixing vessel with and without aeration on the fluid hydrodynamics and mixing tank. (Ducoste and Clark 1999) [2] Employed the Computational Fluid Dynamics (FIDAP) code called Fluid Dynamics International, Evanston, IL in the study. The model was a simple geometry consists of the submerged impellers in cubical mixing vessel. The k- ϵ model was used to simulate the turbulence and the flow pattern induced in the vessel. (Masoud Rahimi 2005) [3] Used CFD simulation to study the influence of impellers number and layout on mixing time. A large storage tank of 19000m³ contain three types of crude oil with difference density. (Aoyi et al 2008) [4] Investigated the hydrodynamics of fluid in mixing stirred vessel agitated by Rushton turbine with low clearance condition. The CFD technique and LDV measurements were performed in order to understand the flow pattern and estimated the mixing time. (Angelique Delafosse et. al. 2008) [5] Investigated the hydrodynamics in mixing tank by CFD simulation. Two models were used to determine the dissipation rate and its distribution in mixing vessel. Three dimensional simulations using the commercial CFD code FLENT 6.2.16 based on the Unsteady Reynolds Averaged Navier-Stokes equations (URANS) model and Large Eddy simulation (LES) model was used (Zadghaffari et. al. 2008)[6] Studied the flow field, power and mixing time in mixing tank agitated by a double Rushton turbine. It was observed that a considerable reduction in mixing time can be achieved by increasing impeller rotation speed and reasonable agreement between the simulation and experimental results were obtained. The comparisons pointed the advantages

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of this CFD approach as a computational tool for designing mixing reactors. (Ameur and Bouzit 2012) [7] Studied the effect of blades number on mixing performance in shear thinning fluid. The simulations and calculations of impeller were performed using computer code (CFX11.0). It has been shown that the axial velocity is directly proportional to Reynolds number which results an increasing in cavern size. The maximum values of tangential velocity are located at the tip of blade for any number of blades. The tangential velocity degradation is greater with impeller have two blades when compared with the other cases, while the difference becomes insignificant between an impeller with six and eight blade. (Meriem Ammar et. al. 2012) [8] Studied the effect of tank design on the flow pattern. Three baffles less stirred vessel types have been prepared, cylindrical, curved, and spherical vessel. It has been mentioned that the curved bottom tank reduced the low velocity flow zones, and established more uniformity for entirely tank volume. By the comparison between LDV data and simulation results, it can be consider that the CFD code is valid for predicting the fluid hydrodynamics in mixing vessel. (Sharma and Genitha 2014) [9] Numerically investigated the effect of different impellers on aerobic baffles stirred tank fermenter. The performance of aerobic stirred tank fermenter evolution was performed by using various software packages, Autodesk simulation CFD, and ANSYS FLUENT. The $k-\epsilon$ model, $k-\omega$ model, Shear Stress Transport (SST) model and Scale Adaptive Simulation (SAS) model were used to represent the turbulence. It has been observed that the axial flow component for all impellers was restrained on the bottom of the tank; consequently the overall flow was mainly radial. This will weak the axial velocity distribution at the bottom of mixing vessel, therefore the solid particles stays around the bottom of the tank. The 6-blade Rushton turbine equipped with walled baffles induced the best state for mass transfer. (Arturo J. et. al. 2015)[10] Examined the optimal location of one or two axial impellers on central shaft in tall stirred tank through establishing a connection of Evolutionary Program EP and CFD technique. It has been noted the optimum mixing time and power consumption can be achieved through the utilization of EP method. For mixing process established with one impeller the optimum mixing performed at clearance $C = 0.13418$ m and for two impellers system it's achieved with clearance $C = 0.26441$ and impeller space $h = 0.1601$ m. The evolution process selected the best entities according to operation condition for realizing optimum conditions.

In this paper, the flow field in stirred tank is studied using ANSYS Fluent v15.4 to identify the dead zones in the tank where the fluid is not mixing by presenting the velocity vectors at different planes in the tank. This will give better information which improves tank design in accordance with operation conditions.

Theoretical Analysis:-

CFD Model and Simulation:-

A schematic diagram of the tank and the impeller is shown in Figure 1. The system consists of a flat bottomed cylindrical vessel, the vessel diameter ($D_t = 0.3$ m) of which equals the height of the liquid ($H = D_t$). Four baffles having width, $W = D_t/10$ are spaced equally around the vessel. The shaft of the impeller is concentric with the axis of the vessel. The impeller diameter, D_i , equivalent to $D_t/3$. And the bleed radius 0.3 m and bleed height is $.03$ m. The distance between the tank bottom and the impeller position C is set to $C = D_t/3$. The rotational speed of the impeller, N , is ranging from 60 rpm to 135 rpm increasing step 15 rpm, leading to a tip speed, V_{tip} , ranging 0.314 m/s to 1.05 m/s. The working fluid is water with density, ρ , of 1000 kg/m³ and viscosity, μ , of 1×10^{-3} Pa.s. The mixing tank was design depending on the standard configuration [11], (Georgy 1991)[12]. The retreat impeller showed in Figure 2.

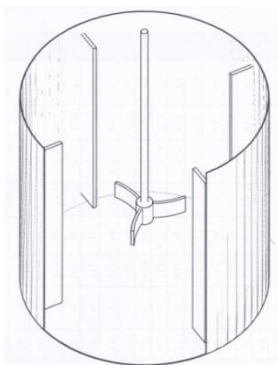


Figure1:- mixing Tank in Present Study

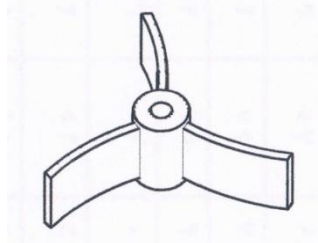


Figure 2:- Retreat impeller Geometry.

Governing equations:-

Continuity equation:-

The net flow of mass across the boundary of a control volume is zero in steady state flow:

$$\nabla \cdot \vec{G} = 0 \quad (1)$$

Where:

$\vec{G} = \rho \vec{V}$, is mass velocity.

Equation (1) can be written relative to cylindrical coordinates as follows:

$$\frac{\partial G_z}{\partial z} + \frac{1}{r} \frac{\partial}{\partial r} (r G_r) + \frac{1}{r} \frac{\partial G_\theta}{\partial \theta} = 0 \quad (2)$$

The subscripts z, r and θ are representing to the axial, radial and tangential components respectively. The \bar{u} , \bar{v} and \bar{w} are the components for the time mean velocity in z, r and θ directions respectively, and u' , v' and w' be the corresponding velocities of fluctuation. The continuity equation can be written as conservation of mass equation with the following form : (Joseph 1997), (Ronald 1984)

$$\frac{\partial}{\partial z} (\bar{u}) + \frac{1}{r} \frac{\partial}{\partial r} (r \bar{v}) + \frac{1}{r} \frac{\partial}{\partial \theta} (\bar{w}) = 0 \quad (3)$$

Momentum Equation:-

The general momentum equations in terms of shear stress τ governing the fluid motion for three dimensions for cylindrical coordinate are [9]:

In Z-direction

$$\rho \left(\frac{\partial U}{\partial t} + U \frac{\partial U}{\partial z} + V \frac{\partial U}{\partial r} + \frac{W}{r} \frac{\partial U}{\partial \theta} \right) = - \frac{\partial p}{\partial z} + \left[\frac{1}{r} \frac{\partial}{\partial r} (r \tau_{rz}) + \frac{1}{r} \frac{\partial}{\partial \theta} \tau_{\theta z} + \frac{\partial}{\partial z} \tau_{zz} \right] + \rho F_z \quad (4)$$

In R-direction

$$\rho \left(\frac{\partial V}{\partial t} + U \frac{\partial V}{\partial z} + V \frac{\partial V}{\partial r} + \frac{W}{r} \frac{\partial V}{\partial \theta} - \frac{W^2}{r} \right) = - \frac{\partial p}{\partial r} + \left[\frac{1}{r} \frac{\partial}{\partial r} (r \tau_{rr}) + \frac{1}{r} \frac{\partial}{\partial \theta} \tau_{\theta r} + \frac{\partial}{\partial z} \tau_{zr} - \frac{\tau_{\theta \theta}}{r} \right] + \rho F_r \quad (5)$$

In- θ -direction

$$\rho \left(\frac{\partial W}{\partial t} + V \frac{\partial W}{\partial r} + \frac{W}{r} \frac{\partial W}{\partial \theta} + \frac{WV}{r} + U \frac{\partial W}{\partial z} \right) = - \frac{1}{r} \frac{\partial p}{\partial \theta} + \left[\frac{1}{r^2} \frac{\partial}{\partial r} (r^2 \tau_{r\theta}) + \frac{1}{r} \frac{\partial}{\partial \theta} \tau_{\theta \theta} + \frac{\partial}{\partial z} \tau_{z\theta} + \frac{\tau_{\theta r} - \tau_{r\theta}}{r} \right] + \rho F_\theta \quad (6)$$

The previous equations of mass conservation and momentum can be combined to form of one general form (Versteeg 1995)

$$\frac{\partial \psi_z}{\partial z} + \frac{1}{r} \frac{\partial (r \psi_r)}{\partial r} + \frac{1}{r} \frac{\partial (r \psi_\theta)}{\partial \theta} = S_\phi \quad (7)$$

For the continuity and momentum the ψ_z , ψ_r , and ψ_θ are the total diffusion fluxes defined by:

$$\psi_z = \rho U \phi - \Gamma_\phi \frac{\partial \phi}{\partial z} \quad (8)$$

$$\psi_r = \rho V \phi - \Gamma_\phi \frac{\partial \phi}{\partial r} \quad (9)$$

$$\psi_\theta = \rho W \phi - \Gamma_\phi \frac{\partial \phi}{r \partial \theta} \quad (10)$$

Where Φ stands for any of the dependent variables and the corresponding values of Γ_ϕ and S_ϕ is indicated in table

(1). For axisymmetric swirling flow $\left(\frac{\partial \phi}{\partial \theta} = 0\right)$, equation (7) becomes:

$$\frac{\partial \psi_z}{\partial z} + \frac{1}{r} \frac{\partial (r \psi_r)}{\partial r} = S$$

$$\rho \frac{\partial}{\partial z} \left(U \phi - \Gamma_\phi \frac{\partial \phi}{\partial z} \right) + \frac{1}{r} \frac{\partial}{\partial r} \left(r V \phi - r \Gamma_\phi \frac{\partial \phi}{\partial r} \right) = S_\phi \quad (11)$$

Table (1):- variables of equations

Equation	Φ	Γ_ϕ	S_ϕ
Conservation of mass Eq(2)	1	0	0
Conservation of momentum in z – direction eq. (4)	U	μ_{eff}	$-\frac{\partial \bar{p}}{\partial z} + \frac{\partial}{\partial z} \left(\mu_{eff} \frac{\partial \bar{u}}{\partial z} \right) + \frac{1}{r} \frac{\partial}{\partial r} \left[r \mu_{eff} \left(\frac{\partial \bar{v}}{\partial z} \right) \right] + \frac{1}{r} \frac{\partial}{\partial \theta} \left[\mu_{eff} \left(\frac{\partial \bar{w}}{\partial z} \right) \right]$
Conservation of momentum in r – direction eq.(5)	V	μ_{eff}	$-\frac{\partial \bar{p}}{\partial r} + \frac{\partial}{\partial z} \left[\mu_{eff} \frac{\partial \bar{u}}{\partial r} \right] + \frac{1}{r} \frac{\partial}{\partial r} \left[r \mu_{eff} \frac{\partial \bar{v}}{\partial r} \right] + \frac{1}{r} \frac{\partial}{\partial \theta} \left[\mu_{eff} \left(r \frac{\partial \left(\frac{\bar{w}}{r} \right)}{\partial r} \right) \right] + \rho \frac{\bar{w}^2}{r} - \frac{2\mu_{eff}}{r} \left(\frac{1}{r} \frac{\partial \bar{w}}{\partial \theta} + \frac{\bar{v}}{r} \right)$
Conservation of momentum in θ -direction eq.(6)	W	μ_{eff}	$-\frac{1}{r} \frac{\partial \bar{p}}{\partial \theta} + \frac{\partial}{\partial z} \left[\mu_{eff} \left(\frac{1}{r} \frac{\partial \bar{u}}{\partial \theta} \right) \right] + \frac{1}{r} \frac{\partial}{\partial r} \left[\mu_{eff} \left(\frac{\partial \bar{v}}{\partial \theta} - \bar{w} \right) \right] + \left[\frac{\mu_{eff}}{r} \left(r \frac{\partial \left(\frac{\bar{w}}{r} \right)}{\partial r} + \frac{1}{r} \frac{\partial \bar{v}}{\partial \theta} \right) \right] - \frac{\rho \bar{w} \bar{v}}{r} + \frac{1}{r} \frac{\partial}{\partial \theta} \left[\mu_{eff} \left(\frac{1}{r} \frac{\partial \bar{w}}{\partial \theta} + \frac{2\bar{v}}{r} \right) \right]$

THE K- ϵ TURBULENCE MODEL:-

Jones and Launder had proposed the following equations for both the turbulence kinetic energy (k) and for energy dissipation (ϵ) [10]

For turbulence kinetic energy (k)

$$\rho \bar{u} \frac{\partial k}{\partial z} + \rho \bar{v} \frac{\partial k}{\partial r} + \rho \bar{w} \frac{\partial k}{\partial \theta} = \frac{\partial}{\partial z} \left(\frac{\mu_t}{\sigma_{k,t}} \frac{\partial k}{\partial z} \right) + \frac{1}{r} \frac{\partial}{\partial r} \left(\frac{\mu_t}{\sigma_{k,t}} r \frac{\partial k}{\partial r} \right) + \frac{1}{r} \frac{\partial}{\partial \theta} \left(\frac{\mu_t}{\sigma_{k,t}} \frac{\partial k}{\partial \theta} \right) - \rho \epsilon + \mu_t G \quad (12)$$

For energy dissipation rate (ϵ)

$$\rho \bar{u} \frac{\partial \epsilon}{\partial z} + \rho \bar{v} \frac{\partial \epsilon}{\partial r} + \rho \bar{w} \frac{\partial \epsilon}{\partial \theta} = \frac{\partial}{\partial z} \left(\frac{\mu_t}{\sigma_{\epsilon,t}} \frac{\partial \epsilon}{\partial z} \right) + \frac{1}{r} \frac{\partial}{\partial r} \left(\frac{\mu_t}{\sigma_{\epsilon,t}} r \frac{\partial \epsilon}{\partial r} \right) + \frac{1}{r} \frac{\partial}{\partial \theta} \left(\frac{\mu_t}{\sigma_{\epsilon,t}} \frac{\partial \epsilon}{\partial \theta} \right) + C_1 \frac{\epsilon}{k} \mu_t G - C_2 \frac{\epsilon^2}{k} \quad (13)$$

Where G refers to the generation term and is given by [37]:

$$G = 2 \left[\left(\frac{\partial \bar{u}}{\partial z} \right)^2 + \left(\frac{\partial \bar{v}}{\partial r} \right)^2 + \left(\frac{\bar{v}}{r} \right)^2 \right] + \left[\left(\frac{\partial \bar{u}}{\partial r} \right) + \left(\frac{\partial \bar{v}}{\partial z} \right) \right]^2 + \left(\frac{\partial \bar{w}}{\partial z} \right)^2 + \left(\frac{\partial \bar{w}}{\partial r} - \frac{\bar{w}}{r} \right)^2 \quad (14)$$

The turbulent kinetic energy (k) and the dissipation rate of the turbulent energy (ϵ) are chosen as the two properties in order to determine the turbulent viscosity μ_t

$$\mu_t = \frac{C_\mu \rho k^{\frac{1}{2}}}{l^{-1}} \quad (15)$$

Where: C_μ is a constant. It is assumed that at a high Reynolds number, (ε) value to be Proportional to $k^{\frac{3}{2}}/l$, the above equation becomes:-

$$\mu_t = \frac{C_\mu \rho k^2}{\varepsilon} \quad (16)$$

The quantities $\sigma_{k,t}$, C_1 , C_2 , $\sigma_{\varepsilon,t}$ and C_μ that appear in the k- ε model and μ_t equations, are the universal k- ε model constants, whose values are reproduced in table (2).

Table (2):-shows the variable of k- ε model.

Equation	Φ	Γ_Φ	S_Φ	C_1	1.44
Turbulent K.E.eq. (12)	K	$\mu_t/\sigma_{k,t}$	$-\rho\varepsilon + \mu_t G$	C_2	1.92
Dissipation rate eq.(13)	ε	$\mu_t/\sigma_{\varepsilon,t}$	$\mu_t G C_1 \frac{\varepsilon}{k} - C_2 \frac{\varepsilon^2}{k}$	C_μ	0.09
				$\sigma_{k,t}$	1
				$\sigma_{\varepsilon,t}$	1.3

The CFD modeling of mixing problem, consist of three steps which are pre-processing, equation solving and post-processing. In first part the problem geometry should be built and meshed. In the second step the partial differential equations describing the flow (Continuity and Navier-Stokes) are discretized on the mesh and solved simultaneously. The boundary and initial conditions should be introduced to the CFD. The turbulence model selected which is describing the effect of turbulence on the bulk flow properties of the fluid. Finally, the obtained results should be analyzed.

The Numerical Solution Setup:-

In the present study, mixing in 21.2 liter of water agitated by Retreat impellers. The mixing tank model mesh type is tetrahedron and it divided in to 58244 nodes as shown in figure 3. The MFR method was applied for modeling the impeller rotation. Also the continuity and Navier-Stokes equations together with the RNG version of the K- ε were used to describe the equation of motion.

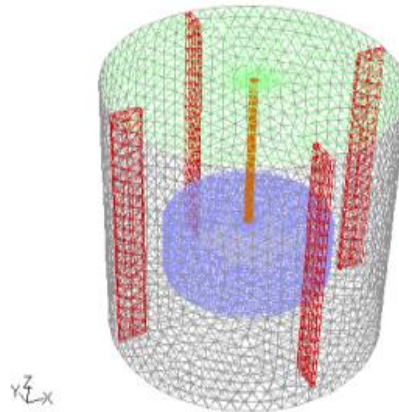


Figure 3:- Mixing Tank Modeling and Mesh Geometry.

Results and Discussions:-

The impeller rotation speed has a great effect on fluid motion in mixing processes. Consequently, the efficiency of settling process for the solid material is affected by the changing of impeller rotation speed. Also, the concentration of chemical additives would become more homogeneous when the optimum impeller rotation speed has been correctly selected.

Figure 4 illustrates the velocity vectors and contours for Z-0 planes for Retreat impeller rotates at 90 rpm. For plane at $\theta = 0^\circ$, the impeller blades pumped the fluid radially toward tank wall that cause fluid to split into two jets. The small jet is circulated downward to the bottom of tank and then returns to the impeller zone so produces an eddy in

the zone below the impeller. The center of eddy is located at radius of 0.113 m from the tank center and 0.061m from the bottom of tank. A Poor or weak mixing region is exists at the center of tank below the impeller.

The second jet of the fluid will be circulated in upward direction, so generates an eddy which is located at same radial direction with lower one but with height of 0.138m from the bottom of the tank. The flow pattern will be same at 30° and 60° plan except the reduction in the fluid flow velocity at the upper zone especially at 30° plan and velocity of fluid near the wall of tank increases in the upward direction. The fluid flow behavior in 90° plan is similar to that observed at 0° plan.

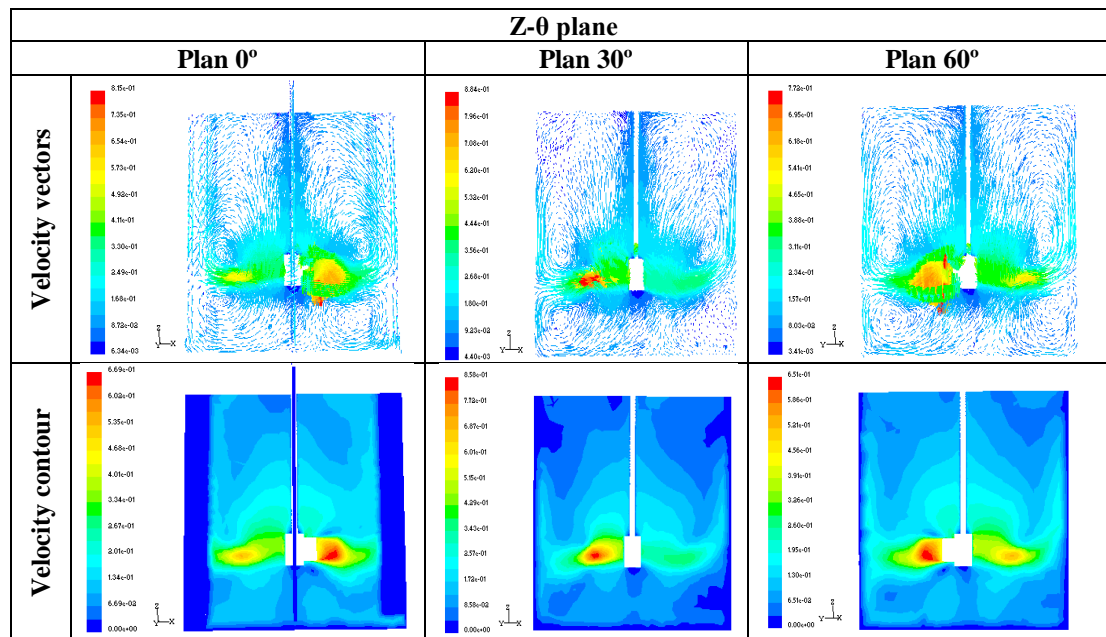


Figure 4:- Velocity Vectors and Contour for retreat Impeller Rotate at 90 Rpm.

Figuer.5 showed the velocity vectors and contours In Z-R direction at height 0.01 m velocity vectors and contour shows the poor mixing zone in the center of tank. The velocity increases toward the wall of tank, and then the velocity decreases near the tank wall, this fluctuation of velocity lead to form zones with high mixing. At height of 0.1m (impeller zone) the velocity linearly increases along the blade of impeller until it reaches to its maximum value at the tip of the blade. Then the velocity is sharply decreases as a result of the pumping direction. At the free surface of fluid the velocity decreases because most of the fluid circulates in the lower zone due to the influences of impeller pumping capacity. The high mixing zone reduces especially at the center of tank but swirling motion continues near the baffles wall.

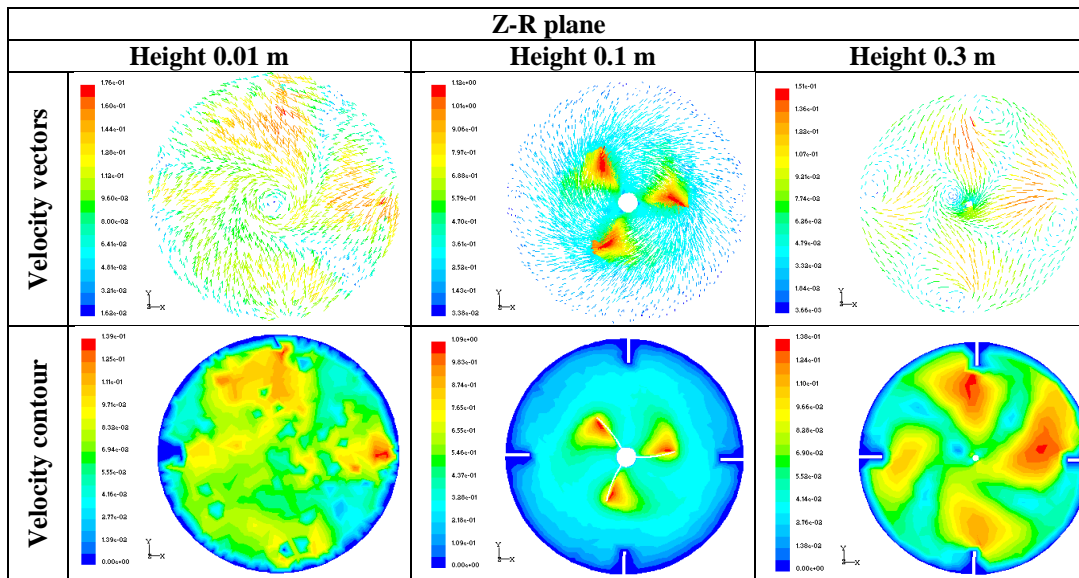


Figure 5:- Velocity Vectors and Contour for Retreat Impeller Rotate at 90 Rpm

Figure 6 showed the velocity vectors and contours in the Z- θ plan and Z-R plan at impeller rotational speed 135 rpm. It can observe that there are no significant changes on the fluid behavior or on the flow pattern. But the velocity magnitude increases as the impeller rotation speed increases.

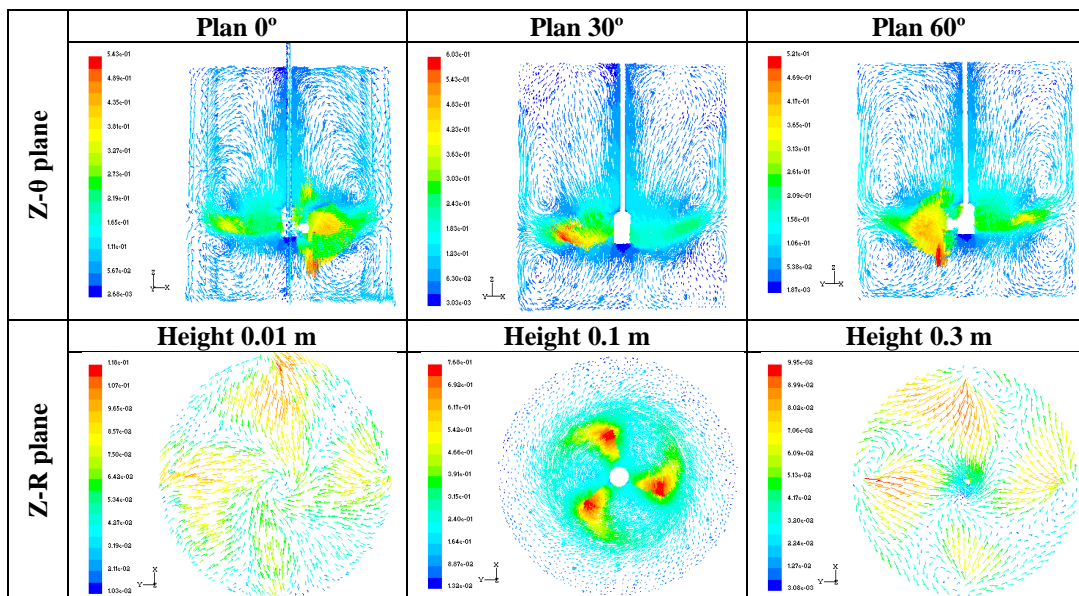


Figure 6:-Velocity Vectors for Retreat Impeller Rotate at 135 rpm

Figures (7-9) illustrates the velocity magnitude distribution with respect to tank radius in Z-R plan with various impeller rotational speeds. It can observe that the velocity fluctuations are not sufficient at low rotational speed and it becomes more sufficient at 90 rpm and the velocity fluctuated increased as rotational speed of impeller increased. This occurs at three difference levels (0.1, 0.1, and 0.3) m in mixing tank. The same thing occurs with velocity distribution at R- θ plan (0°, 30° and 60°) as shown in figures (10-12)

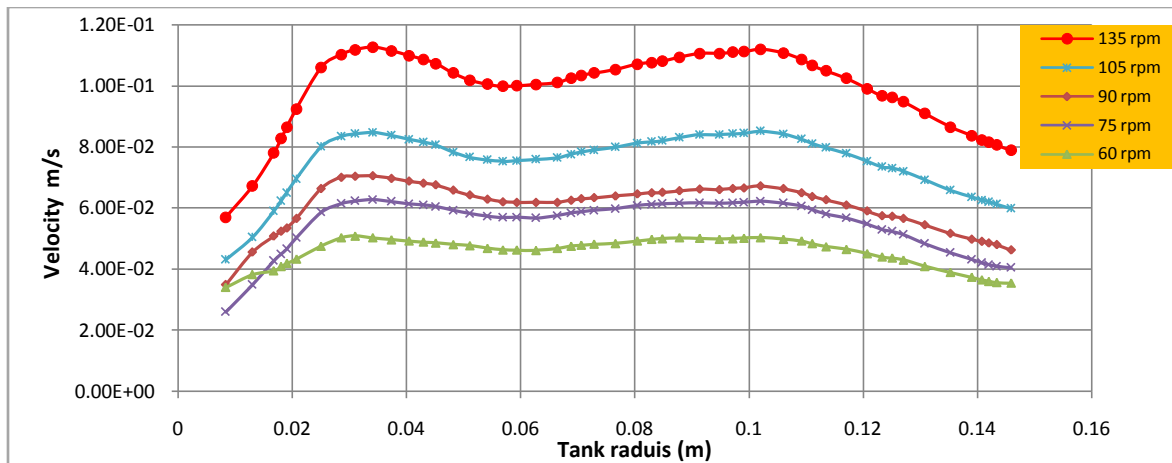


Figure 7:- Velocity Distribution for Retreat Impeller Z-R plan and Height 0.01 m

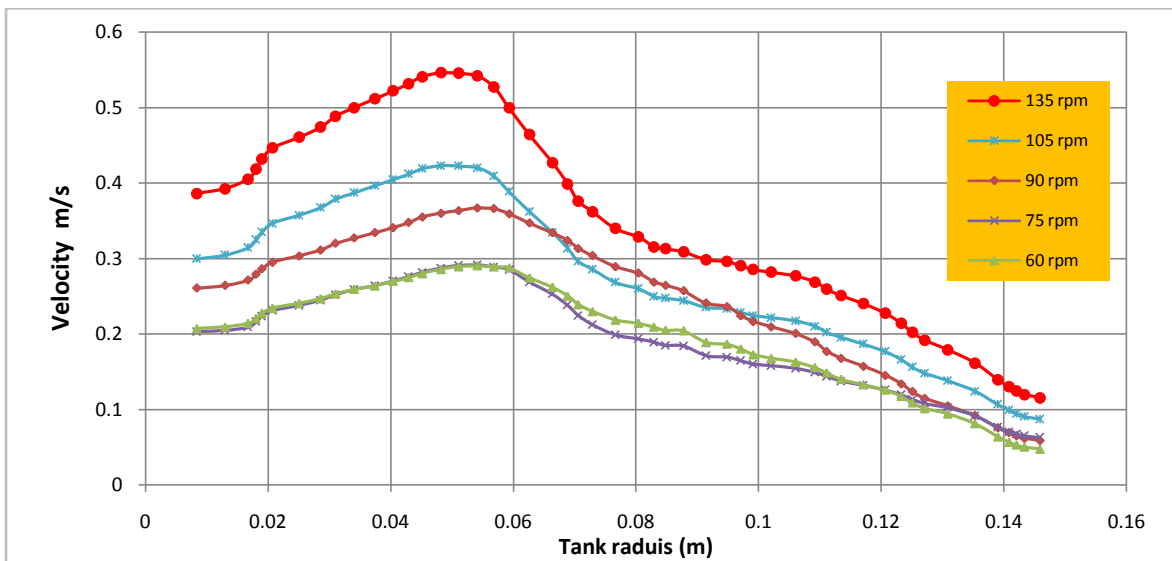


Figure 8:- Velocity Distribution for Retreat Impeller Z-R plan and Height 0.1 m

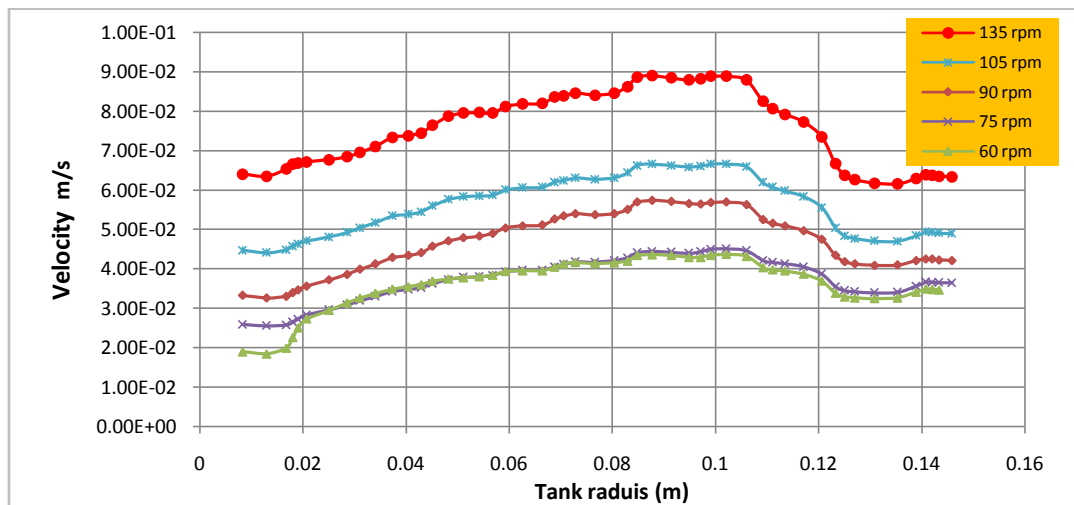


Figure 9:- Velocity Distribution for Retreat Impeller Z-R plan and Height 0.3 m

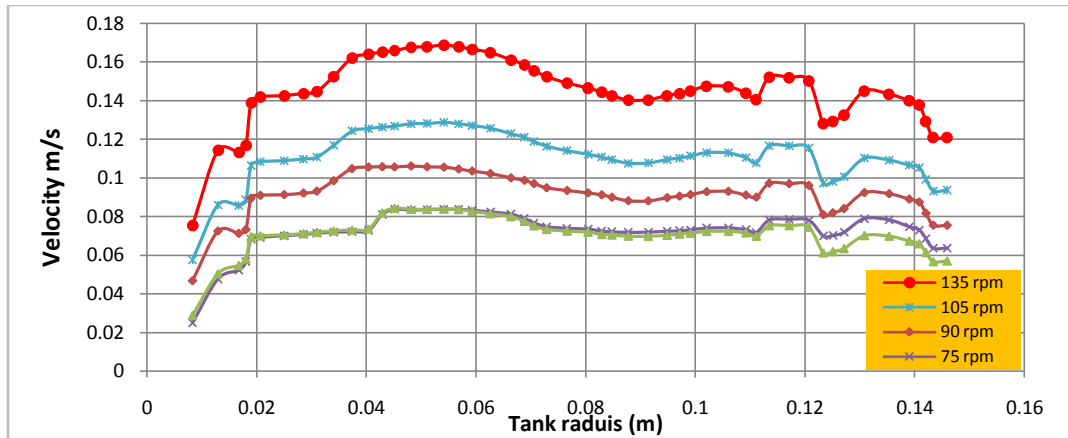


Figure 10: Velocity Distribution for Retreat Impeller at Z-θ plan 0°

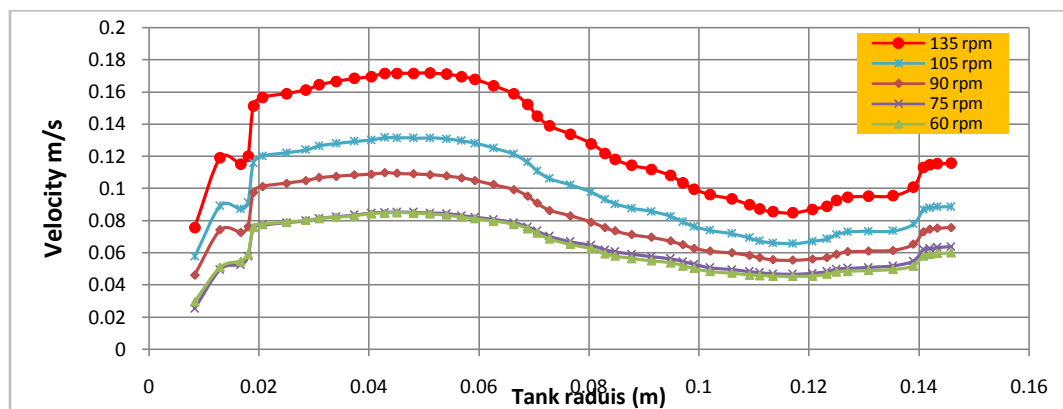


Figure 11:- Velocity Distribution for Retreat Impeller at Z-θ plan 30°

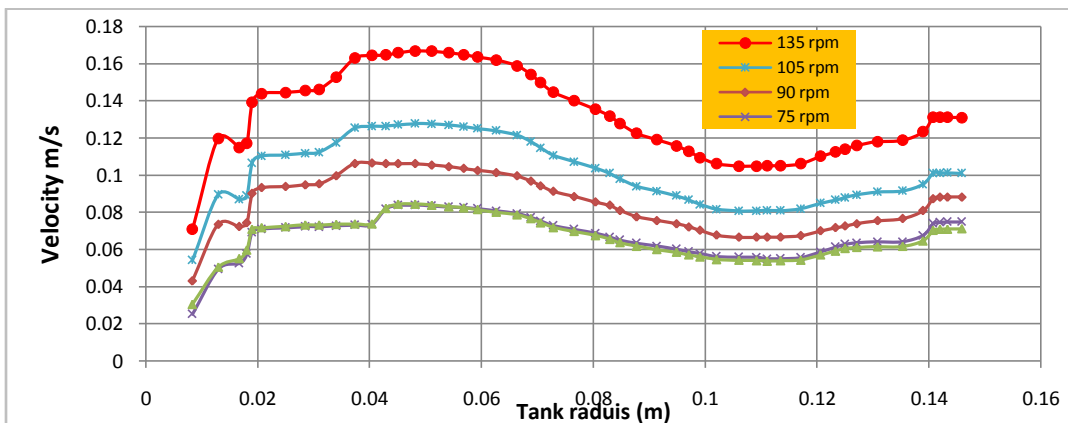


Figure 12:- Velocity Distribution for Retreat Impeller at Z-θ plan 60°

Conclusion:-

The FLUENT v.15.4 is used to analysis the Retreat impeller used for mixing purpose in cylindrical mixing tank. The following conclusions and the results of the study:

1. The numerical analysis gives good results of velocity distribution and mixing rate with the tank. Such results can be used for describing the flow behaviour.
2. Velocity vectors show that the poor mixing zones are generated in the mixing tank.
3. The velocity fluctuation of the fluid increases as rotational speed increased.

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Nomenclature

Symbols	Description	SI Unit
a	Acceleration	m/s ²
C ₀	Constant	
C ₁	Constant	
C ₂	Constant	
C _u	Constant	
D _i	Impeller diameter	m
D _t	Diameter of mixing tank	m
E _k	Turbulent kinetic energy in equation (3-52)	m ² /s ²
C	Impeller clearance	m
H	Liquid height in mixing tank	m
N	Impeller rotation speed	rev/min
p	Pressure	N / m ²
r	Radius of mixing tank	m
S _r	Source term in r-direction of momentum equation	
S _θ	Source term in θ-direction of momentum equation	
S _k	Source term in k-ε model	
S _ε	Source term in k-ε model	

t	Mixing time	s
U	Velocity vector in z - direction	m / s
u	Velocity in z-direction	m / s
V	Velocity vector in θ - direction	m / s
v	Velocity in r-direction	m / s
W	Velocity vector	m / s
w	Velocity in θ – direction	m / s
ρ	Density	kg / m ³
ε	Dissipation rate of kinetic turbulence energy	m ² / s ³
ε_w	Dissipation rate of kinetic turbulence energy at wall	m ² / s ³
εe	Eddy viscosity	Pa.s
Γ	Arbitrary parameter	

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RESEARCH ARTICLE

DESIGNING THE TRAFFIC PHASES USING GRAPH COLOURING AND BOOK THICKNESS TO REDUCE THE TRAFFIC FOR A SELECTED JUNCTION IN COLOMBO CITY.

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Key words:-

Cycle Time, Traffic Phases, Entering point and exit point, Traffic movement, Planarity, Vertex colouring

Abstract

Traffic signaling systems can be optimized by reducing the cycle time byway of minimizing the traffic phases. In this study the concept of graph colouring and the book thickness were used to develop an algorithm in order to minimize the number of traffic phases at Borella junction in the Colombo city and optimize the traffic signaling system. The proposed algorithm facilitates a mechanism to reduce the Cycle Time of that junction by reducing the number of traffic phases in a traffic signaling system. As a result, the number of traffic phases of Borella junction was reduced from five to four.

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Introduction:-

Road traffic is a critical problem in Sri Lanka especially in the Colombo city and suburbs. According to the ComTrans report [5], a huge speed drop due to the heavy traffic is clearly visible during the peak hours. During the peak hours, lengthy vehicle queues are there in many junctions even though they are controlled through the traffic light signaling system. Most of the junctions with traffic light systems are operated manually by the traffic police during the peak hours due to heavy traffic.

Moreover, a direct connection between the Cycle Time of a traffic signaling system and the traffic queue formation can be witnessed, since the other traffic movements should be stopped when one traffic movement is allowed. Then the queues will form in the traffic movements which were stopped within the time period of inter green. Similarly when the unnecessary excess amount of green time was allocated for certain traffic movements, all the other vehicles in other traffic movements should be stopped and it will be result in the queue formation in those traffic movements. In that situation if the green time given for a certain traffic movement is not adequate to flow the length of the newly formed queue, the traffic congestion in that area will increase further.

Since the Cycle Time contains with the green time of each phase with inter green time, the reduction of the Cycle Time needs to be considered when forming a system to reduce the traffic. The Cycle Time of a certain traffic signaling system depends on the number of traffic phases in the relevant junction. Therefore obtaining the minimum number of traffic phases will lead to optimize the traffic signaling system.

Graph theoretical concepts such as planarity, book thickness together with graph colouring can be used to avoid intersecting edges. Also there can be seen a method of managing the signaling process using graph theory with the concept of grouping the traffic movement (Baruah and Baruah 2012). Moreover the concept of circular arcs in Graph theory has been used when installing the traffic light system (Darshankumar and Jhala 2014), (Hosseini and Orooji 2009).

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The graph colouring is a method which has used to avoid edge intersection. Therefore the formulation of the traffic phases is possible when the concepts in graph colouring are used. The concept of graph colouring has used to find the different types of accident zones in traffic flows (Myna 2015). However they have used the edge colouring to obtain the accident zones. Similarly the vertex colouring concept was used in many applications including air traffic flow management (Barnier and Brisse 2002), scheduling radio transmission networks (Ramanathan and Lloyd 1993), time table scheduling and Biprocess tasks (Marx 2004).

Borella is a junction where currently traffic signaling system is operated but considered as a junction where huge traffic congestion occurs during the peak hours. Thus graph theoretical concepts such as planarity, book thickness and graph colouring were used in this study to come up with more suitable model and optimize the performance of the model.

Methodology:-

There is a need for the identification of a graph for the selected junction. Planar graphs from the identified graph should be obtained while minimize the number of planar graphs. The junction with traffic signaling system should be converted to a graph by properly identifying the relevant edges and vertices. Since the objective of this study is to optimize traffic signaling through the reduction of number of traffic phases, a proper attention must be paid to the traffic movements. The graph will reflect a representation of the traffic movements. A junction can be converted to a graph by taking the entering or exit point as vertices and the road segments connected by them as edges. Moreover the corresponding traffic movements can be represented through directed edges, since those road segments will represent the traffic movements from entering point to the junction and from junction to exit point.

After converting the traffic movements of the selected junction to a graph, the existence of planar graphs must be examined. Since each planar graph includes the traffic movements which doesn't intersect with each other, it can be regarded as traffic phases to the relevant junction. The process of obtaining the traffic phases can be considered through obtaining planar graphs using graph colouring.

For this purpose (to apply graph colouring), an edge of the graph should be formed between two vertices if those traffic movements are intersecting. Then according to the concept of graph colouring, the different colours will be assigned to the vertices which connected through edges. Then two vertices which have same colour will represent two traffic movements which doesn't intersect while the two vertices with different colour will provide the traffic movements which intersect with each other. As a result, the chromatic number of the graph is obtained. Hence minimization of planar graphs can be obtained through this process which will provide the set of traffic movements (edges in the former graph) which doesn't intersect with each other. And the chromatic number will provide the minimum number of colours that can be used when generating the planar graphs which is referred as the book thickness.

The program which constructed to implement the above algorithm will be facilitated to input the traffic movements which are intersecting with each other as a set of edges to obtain the set of traffic phases as the output.

Algorithm:-

- Step 1: Construct a graph (G) for the selected junction in terms of the edges (E) and vertices (V).
- Step 2: Select any vertex $v_i \in V$ and create two empty sets (S_1 & S_2).
- Step 3: Assign a colour to v_i and add it to the set S_1 .
- Step 4: For $v_j \in V$ when $v_i \neq v_j$ and if v_i, v_j is not connected then assign the same colour to v_j and add to S_2 , otherwise assign a different colour and save in a different set S .
- Step 5: Take another v_k from S and repeat the step 4.
- Step 6: Repeat the process till all the vertices has assigned colours such that no two vertices which are connected has same colours.
- Step 7: Return the chromatic number.
- Step 8: Build a book (book embedding) using all planar graphs which were obtained from the chromatic number.

The traffic phases are designed by converting the planar graphs to the traffic movements in a manner that each vertex represents an edge in the graph. There are the vertices that will be used to represent the entering and exit points while the edges represent traffic movements. Hence each planar graph in above process can be identified as a representation of a traffic phase.

A suitable method of calculating the Cycle Time was used to model the signaling system after the traffic phases were designed. Here the following equation was used to obtain the green time for each traffic phase after obtaining the traffic phases. It was developed by considering the time which a car (not a vehicle since the calculations were done using passenger car equivalence) with average speed will be needed to pass the entering and exit points in a certain traffic movement along with the number of vehicles in a certain traffic length. The traffic length represents the optimum number of vehicles which needed to be flowed to reduce the possibility of occurring the traffic congestion in the nearest junction which connected to the entering point.

$$T_i = t + (N-1) \cdot v \cdot 1000 / (3600 \cdot c),$$

where T_i – Green time for the relevant traffic movement,

t – Time needed for first vehicle to pass the junction,

N – Number of vehicles in the traffic length in passenger car equivalence,

v – Average vehicle speed and

c – Average length of a car.

Then the maximum T_i for a relevant traffic phase can be considered as the corresponding green time for that phase and using all the green times for the traffic phases in the junction and the inter green time, the cycle time can be calculated. Thereafter the results from the new algorithm and manual results can be compared to validate the new method.

Results:-

Formation of the graph was done by considering the traffic movements in the junction where entering/exit point of each road connected to the junction was labelled as A, B, C, D and E (See the Fig. 1)

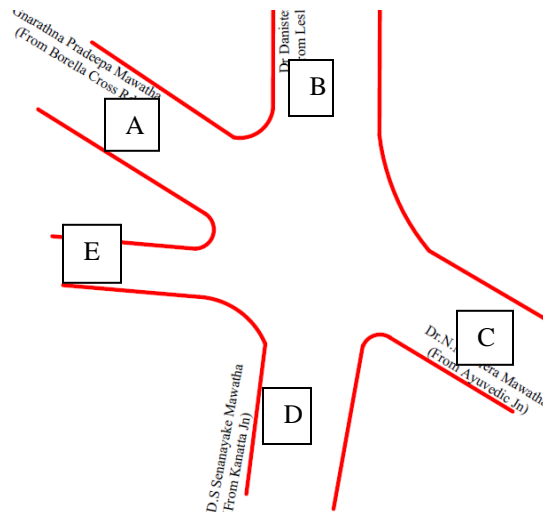


Fig. 1:- The map of Borella Junction.

The complete graph model $G=(V, E)$ is given by Fig. 2 where V represents the set of all intersecting traffic movements and E represents set of all edges to represent the relation of intersection between two traffic movements. For example the traffic movements BD and AC intersect with each other. Then both traffic movements were included in V and $[BD, AC]$ was taken as an edge.

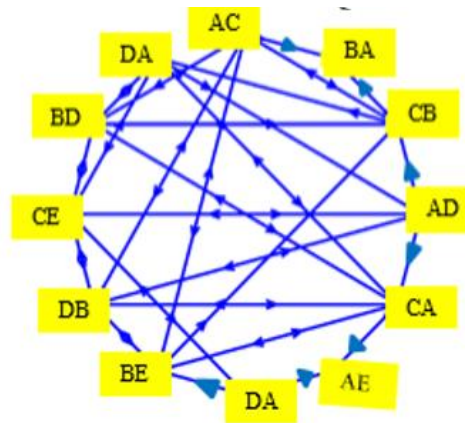


Fig. 2:- Graph of the traffic movements which intersect with each other in Borella junction.

Above algorithm was used to construct a procedure in Maple 12. Then the minimum colours that needed in the vertex colouring was provided by the program along with the relevant planar graphs as output. Since here the number of planar graphs which was used as the traffic phases was minimized, the minimum number of colours will be the book thickness.

The program for the above mentioned algorithm had provided the output of the above graph (in Fig. 2) as follows; $\{AC, AD, AE\}$, $\{BA, BD, BE\}$, $\{CA, CB, CE\}$ and $\{DA, DB, DC\}$ in different sets. The traffic movements that not included here do not intersect with any other movement and hence the flow of those movement can be allowed in any time.

The above four sets represent four planar graphs which can be obtained through a simple transformation. Then the above sets of vertices were transformed to a more comprehensive form where the book embedding was easily obtained. There each vertex which represent a traffic movement was transformed to an edge by taking the entering and exit points as vertices. For example if the vertex AC in set one was considered, it will transformed to the edge $[A, C']$ in the planar graph G_1 . Here C' represents an exit point while A represent entering point. Similarly from converting each vertex in the set one, the planar graph G_1 was formed. Then $G_1 = (V_1, E_1)$ where $V_1 = \{A, C', D', E'\}$ and $E_1 = \{[A, C'], [A, D'], [A, E']\}$. In a similar process all the planar graphs were obtained. Afterward the book embedding was obtained considering whether the number of pages were minimized. There it was compared that the chromatic number obtained through the above procedure was equivalent to the book thickness. The book embedding which was obtained using above planar graphs are given in Fig. 3.

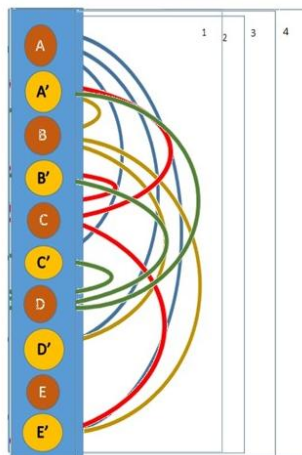


Fig. 3:- The book embedding obtained using above planar graphs.

Since the traffic movements which doesn't intersect with each other will not affect the planarity in each graph which obtained earlier, the edges was added to each page without violating the planarity. Each page of the book embedding

was clearly given as the pages in Fig. 4 along with the edges for the traffic movements which doesn't intersect each other (Those traffic movements were given in light green colour). Here the number of pages were optimized and the book thickness is four.

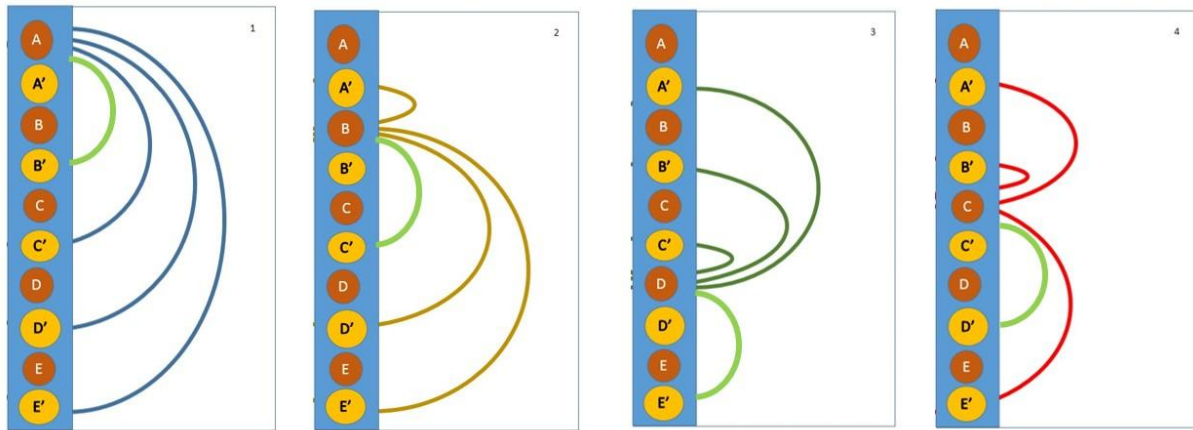


Fig 4:- Pages obtained through book embedding.

Moreover each edge of the above book embedding represent a traffic movement and therefore the pages were taken as the traffic phases. For example, phase 1 was taken as $[[A, C], [A, D], [A, E]]$.

The Cycle Time for the junction was calculated using above mentioned process. Here the Cycle Time was obtained for the current process and for the traffic phases obtained by the book embedding. Then the green time for each phase has been around 9.1 seconds and inter green time was taken as 4.8 seconds.

There is five traffic phases in the current traffic signaling system in this junction, which has the Cycle Time of 69.5 seconds. For the traffic phases obtained from book embedding, the Cycle Time was 55.6 seconds. The number of traffic phases were reduced to four through this approach and as a result of that approximately 14 seconds was reduced from the Cycle Time.

Discussion:-

The output of the program of above mentioned algorithm only reflects the traffic movement which at least intersecting once. Then the traffic movements which doesn't intersect with any other traffic movement can be allowed without any restriction and it will not be reflected in the Cycle Time. Because those traffic movements always will have free flow since it will not be encountered any restriction.

However there should be a proper knowledge of the road capacity of the road segment which the flow projects in, because allowing the flow of that road segment might cause the traffic congestion when other flows are projected in to the same road segment concurrently.

Conclusion:-

When the concept of graph colouring was used to design the traffic phases, the number of traffic phases were reduced ultimately reducing the Cycle Time.

Furthermore the Cycle Time calculation which was developed was considered the vehicle density in a certain traffic length when calculating the green time and therefore it was considered as a proper indication of the traffic flow. Here both current and the proposed processes were compared and the proposed process is better than the current process when considering the number of phases and Cycle Time.

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RESEARCH ARTICLE

EFFECTIVENESS OF PALIVIZUMAB IN PREVENTING RSV HOSPITALIZATION IN HIGH RISK INFANTS WITH CHD & PREMATURE INFANTS.

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Abstract

Respiratory syncytial virus (RSV) is the major cause of bronchiolitis and viral pneumonia in children younger than 1 yr of age.

Infection with respiratory syncytial virus (RSV) is one of the major causes of childhood respiratory morbidity and hospitalization. Palivizumab, a humanized monoclonal antibody, has been recommended for high risk infants to prevent severe RSV-associated respiratory illness.

Key ward: palvizumab + bronchiolitis + RSV

A randomized clinical trial was done in Karballa Teaching Hospital For Pediatrics from October 2015-march 2017 to studies the protective effect of palivizumab prophylaxis for reducing RSV-associated hospitalizations in infants with congenital heart disease and premature infants.

We studied 84 patients divided to 2 groups (group A & B): 30 patients with congenital heart disease their age less than 1 year considered group A and 54 premature neonates who are delivered at the season of RSV (October – March) considered group B.

Group A (30 patients): subdivided to 2 subgroups: A1 & A2, both had hemodynamically significant CHD (large VSD & PDA, complete AV canal with heart failure and on medical treatment, TOF and D-TGA). 18 patients who received 5 doses of palvizumab monthly (from October- March) considered as group A1 and 12 patients who received 3 doses monthly considered as group A2, with monthly follow up for signs of any respiratory illness.

Group B (54 patients): subdivided to 2 subgroups: B1 & B2 both were premature neonates their gestational age less than 35 weeks of gestation and their body weight less than 1500 gm. 25 patients they were received 5 doses of palvizumab monthly for 5 months from October-March considered as group B1 and 25 patients they were received 3 doses of palvizumab monthly considered as group B2. 2 patients received 2 doses only and developed irritability & cough therefore we stopped palvizumab injection, 2 patients received one dose and escape from follow up (therefore those 4 patients were excluded from the study). premature neonates with severe sepsis, severe jaundice, severe congenital malformation, history of birth asphyxia and complex congenital heart disease also were excluded from the study.

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All 4 groups were received palvizumab 15mg/kg intramuscular monthly with regular follow up each months for any sign of respiratory distress.

For each group we select control group C1(30 patients with CHD)&group C2(50 premature infants) :they didn't received palvizumab ,both groups were taken from cardiology clinic,outpatients clinic &premature department &compared with each group.

The results for group A1, 3 patients developed mild bronchiolitis without hospitalization,for group A2 only one patient(8.33%) need hospitalization .

The results for group B1, 7 of them they developed mild bronchiolitis and they didn't need hospitalization,for groupB2, 8 of them they developed mild bronchiolitis and one of them(4%) need hospitalization for 2 weeks.no death was reported.

Regarding control group: 30 patients with CHD(group C1) who didn't received palvizumab,21(70%) patients they were need hospitalization &1 death was reported . and from 50 patients premature infants(group C2) ,39 patients(78%) developed bronchiolitis and they need admission &4 death was reported.

In this study we found breast feeding had significant factor that contribute to the reduction of severity of bronchiolitis and rate of hospitalization.

From this study we recommend :the usage of palvizumab as prophylaxis against RSV for premature infants & hemodynamically significant CHD because its effectiveness in reduction of morbidity &mortality. And encourage breast feeding for its effectiveness against severe disease in infants below 1 year.

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..... **Introduction:-**

Respiratory syncytial virus (RSV) is the major cause of bronchiolitis and viral pneumonia in children younger than 1 yr of age.

RSV is an enveloped RNA virus with a single-stranded negative-sense genome that replicates entirely in the cytoplasm of infected cells and matures by budding from the apical surface of the cell membrane.Because this virus has a nonsegmented genome, it cannot undergo antigenic shift by reassortment like the influenza viruses do.(1)

Infection with RSV does not grant permanent or long term immunity. Reinfections are common and may be experienced throughout life. Other viruses identified as causing bronchiolitis are human metapneumovirus, influenza, adenovirus, and parainfluenza.(2).

The virus can live for a half an hour or more on hands. The virus can also live for up to 5 hours on countertops and for several hours on used tissues.(3).

Epidemiology:-

RSV is distributed worldwide and appears in yearly epidemics. In temperate climates, these epidemics occur each winter over 4-5 mo.During the remainder of the year, infections are sporadic and much less common. In the Northern hemisphere, epidemics usually peak in January, February, or March. RSV outbreaks often overlap with outbreaks of influenza and human metapneumovirus but are generally more consistent from year to year and result in more disease overall, especially among infants younger than 6 mo of age.

RSV is one of the most contagious viruses that affect humans. Infection is nearly universal among children by their 2nd birthday. Reinfection occurs at a rate of at least 10-20% per epidemic throughout childhood, with a lower frequency among adults. In situations of high exposure, such as daycare centers, attack rates are nearly 100% among previously uninfected infants and 60-80% for second and subsequent infections.

Asymptomatic RSV infection is unusual in young children. Most infants experience coryza and pharyngitis, often with fever and frequently with otitis media caused by a virus in the middle ear or bacterial super infection.

Transplacentally acquired anti-RSV maternal immunoglobulin G serum antibodies, if present in high concentration, appear to provide partial but incomplete protection. These immunoglobulin Gs may account for the lower severity of RSV infections during the 1st 4-6 wk of life, except among infants born prematurely, who receive less maternal immunoglobulin. Breastfeeding provides substantial protection against severe disease, an effect that may pertain only to female infants and not male infants.(1)

Clinical manifestation:-

Typically, the first sign of infection in infants with RSV is rhinorrhea.

Cough may appear simultaneously but more often does so after an interval of 1-3 days, at which time there may also be sneezing and a low-grade fever. Soon after the cough develops, the child who experiences bronchiolitis begins to wheeze audibly. If the disease is mild, the symptoms may not progress beyond this stage. Auscultation often reveals diffuse fine inspiratory crackles and expiratory wheezes. Rhinorrhea usually persists throughout the illness, with intermittent fever. Chest radiograph findings at this stage are frequently normal.

Signs of severe, life-threatening illness are central cyanosis, tachypnea of >70 breaths/min, listlessness, and apneic spells. At this stage, the chest may be significantly hyperexpanded and almost silent to auscultation because of poor air movement.

Chest radiographs of infants hospitalized with RSV bronchiolitis have normal findings in approximately 30% of cases, with the other 70% showing hyperexpansion of the chest, peribronchial thickening, and interstitial infiltrates. Segmental or lobar consolidation is unusual and pleural effusion is rare.

Fever is an inconstant sign in RSV infection. In young infants, particularly those who were born prematurely, periodic breathing and apneic spells have been distressingly frequent signs, even with relatively mild bronchiolitis. RSV infections in profoundly immunocompromised hosts may be severe at any age of life.(1)

Bronchiolitis and pneumonia resulting from RSV are more common in boys than in girls by a ratio of approximately 1.5 : 1. Other risk factors with similar impact include 1 or more siblings in the home, white race, rural residence, maternal smoking, and maternal education <12 yr. The medical factors in infants associated with highest risk are chronic lung disease of prematurity, congenital heart disease, immunodeficiency, and prematurity.(1).

The incubation period from exposure to first symptoms is approximately 3-5 days. The virus is excreted for variable periods, probably depending on severity of illness and immunologic status. Most infants with lower respiratory tract illness shed infectious virus for 1-2 wk after hospital admission. Excretion for 3 wk, and even longer, has been documented. Spread of infection occurs when large, infected droplets, either airborne or conveyed on hands or other fomites, are inoculated in the nasopharynx of a susceptible subject.(1).

Nosocomial infection during RSV epidemics is an important concern. Virus is usually spread from child to child on the hands of caregivers or other fomites. Adults undergoing reinfection also have been implicated in spread of the virus. Contact precautions are sufficient to prevent spread when compliance is meticulous, as the virus is not usually spread by small particle aerosol.(1).

Pathogenesis:-

Bronchiolitis is caused by obstruction and collapse of the small airways during expiration. Airway narrowing likely is caused by virus-induced necrosis of the bronchiolar epithelium, hypersecretion of mucus, and round-cell infiltration and edema of the surrounding submucosa. These changes result in formation of mucus plugs obstructing bronchioles, with consequent hyperinflation or collapse of the distal lung tissue.(1).

Bronchiolitis is a disorder most commonly caused in infants by viral lower respiratory tract infection. It is the most common lower respiratory infection in this age group. It is characterized by acute inflammation, edema, and necrosis of epithelial cells lining small airways, increased mucus production, and bronchospasm.(2)

A large number of soluble factors, such as cytokines, chemokines, and leukotrienes, are released and may predispose some individuals to more severe disease. There is also evidence that genetic factors may predispose to more severe bronchiolitis.(1)

Diagnosis:-

Bronchiolitis is a clinical diagnosis. RSV can be suspected with varying degrees of certainty on the basis of the season of the year and the presence of the virus in the community.(1)

RSV plays a causative role in an estimated 40-75% of cases of hospitalized bronchiolitis.(1)

History & physical examination: Clinicians should diagnose bronchiolitis and assess disease severity on the basis of history and physical examination. Clinicians should not routinely order laboratory and radiologic studies for diagnosis.(2)

Clinical signs and symptoms of bronchiolitis consist of rhinorrhea, cough, wheezing, tachypnea, and increased respiratory effort manifested as grunting, nasal flaring, and Clinical signs and symptoms of bronchiolitis consist of rhinorrhea, cough, wheezing, tachypnea, and increased respiratory effort manifested as grunting, nasal flaring, and intercostal and/or subcostal retractions preceded by viral upper respiratory tract prodrome.(2) Older children most often have only mild, cold-like symptoms, such as croupy cough (often described as a "seal bark" cough), stuffy nose, or low-grade fever. (3). Rarely, RSV infection can cause death in infants. However, this is unlikely if the child is seen by a health care provider in the early stages of the disease. Children who have had RSV bronchiolitis may be more likely to develop asthma(3)

The course of bronchiolitis is variable and dynamic, ranging from transient events such as apnea or mucus plugging to progressive respiratory distress from lower airway obstruction. Young infants with bronchiolitis may develop apnea, which has been associated with an increased risk for prolonged hospitalization, admission to intensive care, and mechanical ventilation.(2) Apnea can be the first sign of bronchiolitis in an infant. This occurs more commonly in infants born prematurely and infants who are younger than 2 months. Difficulty feeding related to nasal congestion and rapid breathing, which can result in dehydration.(4)

risk factors for severe disease include prematurity, hemodynamically significant congenital heart disease, chronic lung disease (bronchopulmonary dysplasia, cystic fibrosis, congenital anomaly), and the presence of an immunocompromised state.(1)(2)(3)(4).

Radiography: may be useful when the hospitalized child does not improve at the expected rate, if the severity of disease requires further evaluation, or if another diagnosis is suspected. (2)

Routine laboratory tests are of minimal diagnostic use in most cases of bronchiolitis or pneumonia caused by RSV.

The white blood cell count is normal or elevated, and the differential cell count may be normal with either a neutrophilic or mononuclear predominance. Hypoxemia as measured by pulse oximetry or arterial blood gas analysis is frequent and tends to be more marked than anticipated from the clinical findings. A normal or elevated blood CO₂ value in a patient with a markedly elevated respiratory rate is a sign of respiratory failure.

Definitive diagnosis of RSV infection is based on the detection in respiratory secretions of live virus by cell culture. The presence of viral RNA (detected by a molecular diagnostic test using reverse transcription PCR) or viral antigens (detected by a rapid diagnostic test, usually a membrane blotting test incorporating antibody detection of viral proteins) is strongly supportive in the right clinical setting. Endotracheal tube lavage fluid from patients intubated for mechanical ventilation can be tested. The specimen should be placed on ice, taken directly to the laboratory, and processed immediately for culture, antigen detection, or PCR analysis (1)

Treatment:-

Treatment at home usually includes antipyretic, increased oral fluid, Parents should encourage their child to drink an adequate amount of fluids. and saline nose drops (with bulb suctioning for infants). Monitoring at home periodically for signs or symptoms of worsening. Specifically, this includes monitoring for an increased rate of breathing, worsening chest retractions, nasal flaring, cyanosis, a decreased ability to feed or decreased urine output. Parents

should contact their child's healthcare provider to determine if and when an office visit is needed, or if there are any other questions or concerns. Smoking in the home should be avoided.(4)

Recovery — Most children with bronchiolitis who are otherwise healthy begin to improve within two to five days. However, wheezing persists in some infants for a week or longer, and it may take as long as four weeks for the child to return to his or her "normal" self.(4).

The treatment of uncomplicated cases of bronchiolitis is symptomatic. Humidified oxygen and suctioning are usually indicated for hospitalized infants who are hypoxic. Intravenous fluid can be given for moderately dehydrated infants. NG tube feeding is helpful when sucking is difficult because of tachypnea.(1)

Ribavirin is an antiviral agent delivered through an oxygen hood, face mask, or endotracheal tube with use of a small-particle aerosol, indicated for:

- ❖ Infants younger than 1 yr of age with hemodynamically significant congenital heart disease
- ❖ Children 24 mo of age or younger with profound immunocompromising conditions during RSV season
- ❖ Infants in the 1st yr of life who have either congenital abnormalities of the airway or neuromuscular disease that compromises handling of respiratory secretions
- ❖ Administration in the 2nd yr of life is recommended for children who required 28 or more days of oxygen after birth and who have on going treatment for chronic pulmonary disease (oxygen, steroids and diuretics)(1).

In 2006, the AAP, in conjunction with the American Academy of Family Physicians (AAFP), the American College of Chest Physicians (ACCP), and the American Thoracic Society (ATS), published guidelines for the diagnosis and management of bronchiolitis in children 1 through 23 months of age. These guidelines were updated in 2014 and include the following recommendations (5) :

- Diagnosis and severity should be based on history and physical findings and not on laboratory and radiologic findings
- Bronchodilators should not be routinely used
- Corticosteroids should not routinely be used
- Ribavirin should not be used
- Risk of serious bacterial infection, especially in infants 30-90 days old with bronchiolitis is low. Antibacterials should be used only upon proven coexistence of bacterial infection
- Nutrition and hydration should be assessed.
- Supplemental oxygen should not be routinely used for patients with saturations above 90% on pulse oximetry; continuous pulse oximetry monitoring may not be necessary
- Chest physiotherapy has not shown to benefit infants with bronchiolitis
- Deep suctioning may provide temporary relief
- Nebulized hypertonic (3%) saline may improve symptoms of bronchiolitis when length of stay is expected to exceed 3 days
- Palivizumab prophylaxis should only be administered to selected children
- Hand decontamination is indicated to prevent nosocomial spread
- Infants should not be exposed to passive smoking
- Breastfeeding is recommended.

Prevention : There are several ways to prevent severe bronchiolitis:

- ❖ Avoid smoking in the child's home because this increases the risk of respiratory illness.
- ❖ Wash hands frequently with soap and water, especially before touching an infant. Hands should ideally be wet with water and plain or antimicrobial soap, and rubbed together for 15 to 30 seconds. Hands should be rinsed thoroughly and dried with a single-use towel.
- ❖ Use alcohol-based hand rubs.
- ❖ Avoid other adults and children with upper respiratory infection.
- ❖ A yearly vaccination for influenza virus is recommended for everyone older than 6 months, especially for household contacts of children younger than five years.
- ❖ Infants who are younger than 24 months with specific types of chronic lung disease and infants who are younger than 12 months who were born before 29 weeks, have specific types of heart disease, or have other risk

factors for severe respiratory syncytial virus (RSV) infection may be given a special medication (palivizumab) to prevent severe RSV infection requiring hospitalization.(4).

Breastfeeding and avoidance of cigarette smoke are also presumed to decrease the incidence and/or severity of viral respiratory tract infections (5)(6).

Immunoprophylaxis: Palivizumab(synagis):-

Palivizumab, a humanized murine monoclonal immunoglobulin G-1 directed against an epitope on the F glycoprotein of RSV, is produced by recombinant DNA technology, and has 95% human and 5% murine amino acid sequences. Rare cases of anaphylaxis are the only recognized serious adverse event. Standard dosing is 15 mg/kg administered intra muscularly every 30 days during RSV season for a maximum of five dose. Palivizumab is available in 50 mg or 100 mg vials.(6).

Palivizumab was licensed in June 1998 by the Food and Drug Administration for the reduction of serious lower respiratory tract infection caused by respiratory syncytial virus (RSV) in children at increased risk of severe disease.(7).

The American Academy of Pediatrics has updated its guidance for the use of palivizumab in infants and young children at greatest risk of hospitalization attributable to RSV infection:

Preterm Infants Without Chronic Lung Disease of Prematurity or Congenital Heart Disease:

Palivizumab prophylaxis may be administered to infants born before 29 weeks, 0 days' gestation who are younger than 12 months at the start of the RSV season. For infants born during the RSV season, fewer than 5 monthly doses will be needed. Infants 29 weeks, 0 days' gestation or later may qualify to receive prophylaxis on the basis of congenital heart disease (CHD), chronic lung disease (CLD), or another condition. Palivizumab prophylaxis is not recommended in the second year of life on the basis of a history of prematurity alone.(7).

Infants With Hemodynamically Significant CHD:

Certain children who are 12 months or younger with hemodynamically significant CHD may benefit from palivizumab prophylaxis. Children with hemodynamically significant CHD who are most likely to benefit from immunoprophylaxis include infants with acyanotic heart disease who are receiving medication to control congestive heart failure and will require cardiac surgical procedures and infants with moderate to severe pulmonary hypertension.(7)

Decisions regarding palivizumab prophylaxis for infants with cyanotic heart defects in the first year of life may be made in consultation with a pediatric cardiologist.(7)

Children younger than 2 years who undergo cardiac transplantation during the RSV season may be considered for palivizumab prophylaxis.(7)

Because 5 monthly doses of palivizumab at 15 mg/kg per dose will provide more than 6 months (>24 weeks) of serum palivizumab concentrations above the desired level for most children, administration of more than 5 monthly doses is not recommended within the continental United States. For qualifying infants who require 5 doses, a dose beginning in November and continuation for a total of 5 monthly doses will provide protection for most infants through April and is recommended for most areas of the United States.(7).

Discontinuation of Palivizumab Prophylaxis Among Children Who Experience Breakthrough RSV Hospitalization: If any infant or young child receiving monthly palivizumab prophylaxis experiences a breakthrough RSV hospitalization, monthly prophylaxis should be discontinued because of the extremely low likelihood of a second RSV hospitalization in the same season (<0.5%).(7).

Use of Palivizumab in the Second Year of Life: Hospitalization rates attributable to RSV decrease during the second RSV season for all children. A second season of palivizumab prophylaxis is recommended only for preterm infants born at <32 weeks, 0 days' gestation who required at least 28 days of oxygen after birth and who continue to require supplemental oxygen, chronic systemic corticosteroid therapy, or bronchodilator therapy within 6 months of the start of the second RSV season.(7).

Lack of Therapeutic Efficacy of Palivizumab: Passive antibody administration is not effective in treatment of RSV disease and is not approved or recommended for this indication.(7)

Effect of Palivizumab Prophylaxis on Subsequent Wheezing:

Prophylaxis is not recommended for primary asthma prevention or to reduce subsequent episodes of wheezing.(7) Clinicians may administer palivizumab prophylaxis in the first year of life to certain infants with hemodynamically significant heart disease.

Clinicians may administer up to a maximum of 5 monthly doses of palivizumab (15 mg/kg per dose) during the RSV season to infants who qualify for prophylaxis in the first year of life. Qualifying infants born during the RSV season may require fewer doses. For example, infants born in January would receive their last dose in March.(7) palivizumab reduced the risk of hospitalization due to RSV infection by 55% and 45%. Via monthly intramuscular (IM) injection, to be administered throughout the duration of the RSV season.

Palivizumab targets the fusion protein of RSV, inhibiting its entry into the cell and thereby preventing infection.(8)

Other potential target groups for palivizumab prophylaxis include:(9)

1. Children younger than one year of age with neuromuscular disorders impairing the ability to clear secretions from the upper airways or pulmonary abnormalities.
2. Children younger than two years of age who are immunocompromised (e.g. those with severe combined immunodeficiency; those younger than two years of age who have undergone lung transplantation or hematopoietic stem cell transplantation) during the RSV season.
3. Children with Down syndrome who have additional risk factors for lower respiratory tract infections.

Side effects of palvizumab: (10)

- Sore throat
- Runny nose
- Redness or irritation at injection site
- Vomiting
- Diarrhea

Some more serious side effects include:

- Severe skin rash
- Itching
- Hives (urticaria)
- Difficulty breathing

Adverse events such as injection site reactions (2.3%), fever (1.5%), diarrhea (<1%), and nervousness/irritability (<1%).(11)

palivizumab, a humanized monoclonal antibody that binds RSV F protein, have been shown to reduce RSV-associated hospitalization. Palivizumab the preferred choice of RSV immunoprophylaxis because of its lack of concern with fluid overload, particularly in children with pre-existing cardiac or pulmonary disease, ease of administration, lack of interference with immunization schedules.(11)

palivizumab was able to neutralize both subtype A and B strains of RSV

and it was safe and well-tolerated, there were no significant changes in urinalysis, hematological values, blood urea nitrogen, creatinine, and transaminase levels.(11)

Monthly prophylaxis resulted in a 55% relative reduction in RSV hospitalization. Significant decreases in hospitalizations were seen in children greater than 5 kg (51%), less than or equal to 5 kg (57%), and in infants born

before 32 weeks gestation (47%). Palivizumab recipients also had significantly reduced hospital days, days with supplemental oxygen requirement, moderate/severe lower respiratory tract infections, and intensive care unit (ICU) admissions.(11)

Risk factors for RSV hospitalization:

Reported host risk factors include the following: congenital malformations, congenital airway anomaly, neuromuscular impairment, birth weight, gender, lack of breastfeeding, duration of breastfeeding, cord serum anti-RSV antibody concentration, small for gestational age, Down syndrome, epilepsy, cord blood vitamin D concentration, family history of atopy, viral load, malnutrition, multiple births, and singletons versus multiple birth subjects.

Environmental risk factors include the following: environmental pollution, crowded living conditions, living at increased altitude, meteorological conditions, low parental education, low socioeconomic status, child care attendance, size of child care facility, month of birth, smoke exposure, maternal smoking during pregnancy, and proximity to hospital care. One publication suggested that malformations of the urinary tract increase the risk of RSV hospitalization.(7)

A study was done in Spain for premature infant found that, age ≤ 10 weeks at start of RSV season, breast-feeding ≤ 2 months, at least 1 school age sibling, at least 4 people living in the household (excluding the infant and school-age siblings), and a family history of wheezing were significantly associated with an increased risk of RSV hospitalization .(11)

Storage and administration of palivizumab:-

The storage must be in the refrigerator, not allowed to be freeze and must be used within 6 hours once you mix it. Palivizumab can be used after dilution with 1 mL of sterile water, should be added slowly to the palivizumab vial and then gently swirl the vial (do not shake it) for 30 seconds. then let the vial sit at room temperature for 20 minutes until the solution becomes clear. The solution should be clear and free of floating material. (12).

Palivizumab is administered intramuscularly at a dosage of 15 mg/kg once a month. The drug is packaged in single-dose liquid solution vials at 50 mg/0.5 mL and 100 mg/1.0 mL and does not contain preservative. A vial cannot be stored once it is opened. Anaphylaxis has occurred after palivizumab administration after initial exposure or reexposure, with some cases of severe hypersensitivity reactions reported.(13).

Palivizumab does not interfere with the immune response to live or inactivated vaccines. The childhood immunization schedule should be followed for all children, regardless of palivizumab use.(13)

Material and Methods:-

A randomized clinical trial was done in Karballa Teaching Hospital For Pediatrics from October 2015-march 2017 to studies the protective effect of palivizumab prophylaxis for reducing RSV-associated hospitalizations in infants with congenital heart disease and premature infants.

The study included 84 patients with CHD and premature infants, 2 premature infants were received 2 doses only and developed irritability & cough therefore we stopped palvizumab injection and 2 premature infants received one dose and escape from follow up(therefore those 4 patients were excluded from the study).

The remaining 80 patients were divided to 2 groups(group A&B) : 30 patients with congenital heart disease their age less than 1 year considered group A and 54 premature neonates who are delivered at the season of RSV(October –March) considered group B. Group A(30 patients) :subdivided to 2 subgroups: A1&A2, both had hemodynamically significant CHD(large VSD &PDA,complete AV canal with heart failure and on medical treatment,TOF and D-TGA) . 18 patients who received 5 doses of palvizumab monthly(from October- March) considered as group A1 and 12 patients who received 3 doses monthly considered as group A2,with monthly follow up for signs of any respiratory illness.

Group B (50patients): subdivided to 2 subgroups:B1&B2 both were premature neonates their gestational age less than 35 weeks of gestation and their body weight less than 1500gm .25 patients they were received 5 doses of

palvizumab monthly for 5 months from October-March considered as group B1 and 25 patients they were received 3 doses of palvizumab monthly for 3 months considered as group B2.

Exclusion criteria:-

1. Premature infants with severe sepsis ,severe jaundice,severe congenital malformation and history of birth asphyxia.
2. complex congenital heart disease

All 4 groups were received palvizumab 15mg/kg intramuscular(antrolateral aspect of the thigh) monthly with regular follow up each months for any sign of respiratory distress and side effects.the vial of palvizumab was diluted with 0.5ml of steril water for 50 mg vial and 1ml for 100mg , the sterile water was added slowly to the palvizumab vial and then gently swirl the vial for 30 seconds. then the vial was left at room temperature for 20 minutes until the solution becomes clear and then injected by well trained nursing staff.the patients was observed for any sign of allergy or anaphylaxis for half an hour.

For each group we select control group(group C)80 patients :30 patients with CHD(group C1) &50 premature neonats(group C2) was taken from cardiology clinic,outpatients clinic &premature department &compared with each group.

For each patient and control there was a list of information that include: the name of patient,age,body weight,date of palvizumab injection(first,second,3rd,4th and fifth dose),amount of palvizumab,side effects,type of feeding(breast,mixed and artificial),diagnosis(premature or CHD),if the patient developed bronchiolitis,if the patient need hospital admission,phone or mobile number.

The patients were followed monthly and examined for any signs of respiratory illness and asked the parents for any noticeable side effects.

For all patients we asked them to continue their routine vaccination according to the vaccination program in Iraq,because the palvizumab not interfere with routine vaccination.

THE RESULTS: we in ruled in this study 80 patents with the following results:

For group A1:

The hospital admission was zero percent

The number of patients that developed mild bronchiolitis was 3 patients and no death was reported.

For group A2:

The hospital admission was 8.3%

The number of patients that developed mild bronchiolitis were 4 patients and no death was reported.

For group B1:

The hospital admission was zero percent.

The number of patients that developed mild bronchiolitis were 7 patients and no death was reported.

For group B2:

The hospital admission was 4%

The number of patients that developed mild bronchiolitis were 8 patients and no death was reported.

For group C1:hospital admission was 21(70%) and one death was reported(3.3%),as shown in table (1).

For group C2: Hospital admission was 39(78%) and 4 death was reported(8%).as shown in table (2).

No significant side effects was reported.

Palvizumab didn't interfere with routine vaccination program

Breast feeding had significant factor that contribute to the reduction of severity of bronchiolitis and rate of hospitalization as shown in table(3).

Table (1):- the effectiveness of palvizumab in reduction hospital admission between congenital heart disease group and control group:

Patients group	Total No.	No.of patients developed mild bronchiolitis	No.of hospital admission	No. of death
Group A1	18	3 ¥ #	zero ¥ *	Zero ¥ #
Group A2	12	4 #	1(8.3%) *	Zero #
Control group(C1)	30	3	21(70%)	1(3.3%)

¥ No significant difference ($p>0.05$) as compare with group A2 .

#No significant difference ($p>0.05$) as compare with control group

* Significant difference ($p<0.05$) as compare with control group

Table (2):- the effectiveness of palvizumab in reduction hospital admission between premature group and control group:

Patients group	Total No.	No.of patients developed mild bronchiolitis	No.of hospital admission	No. of death
Group B1	25	7 ¥ *	Zero ¥ *	Zero ¥ #
Group B2	25	8 *	1(4%) *	Zero #
Premature infants(C2)	50	1	39(78%)	4(8%)

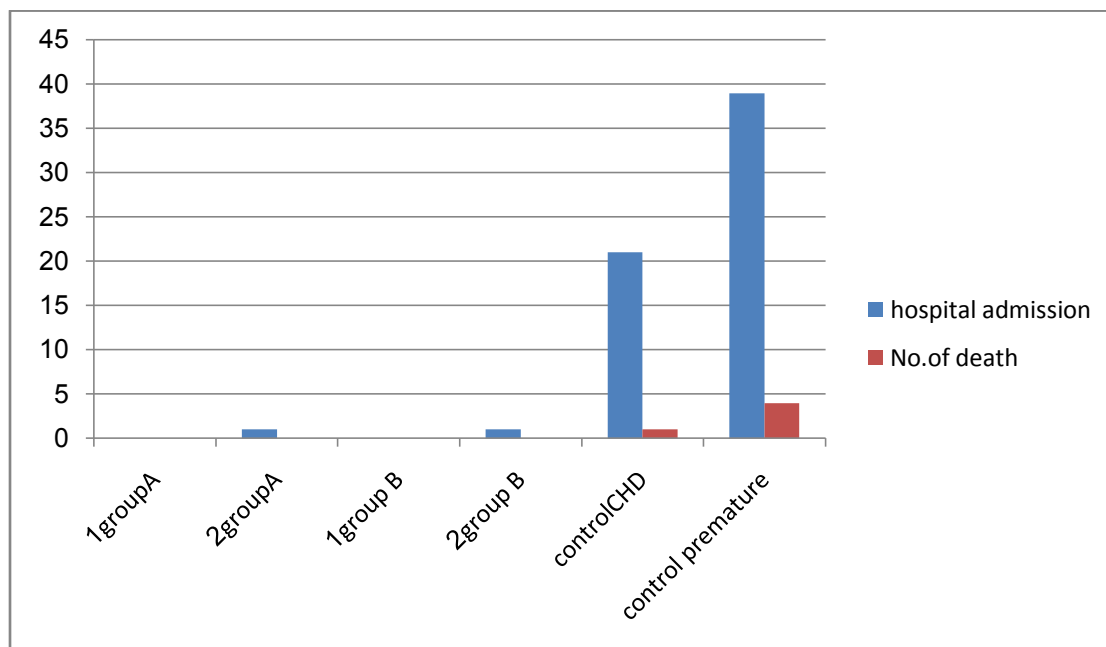
¥ No significant difference ($p>0.05$) as compare with group B2 .

#No significant difference ($p>0.05$) as compare with control group

* Significant difference ($p<0.05$) as compare with control group

Table (3):- the effectiveness of breast feeding between patients and control groups:

Group of patients	Total No.	Breast feeding	mixed	artificial
Group A	30	24(80%)	4	2(6.6%)
Group B	50	25(50%)	19	6(12%)
Patients with CHD(C1)	30	3(10%)	4	23(76.6%)
Premature patients(C2)	50	4(8%)	10	36(72%)

**Figure (1):-** hospital admission and death between patients whom received palvizumab and control groups.

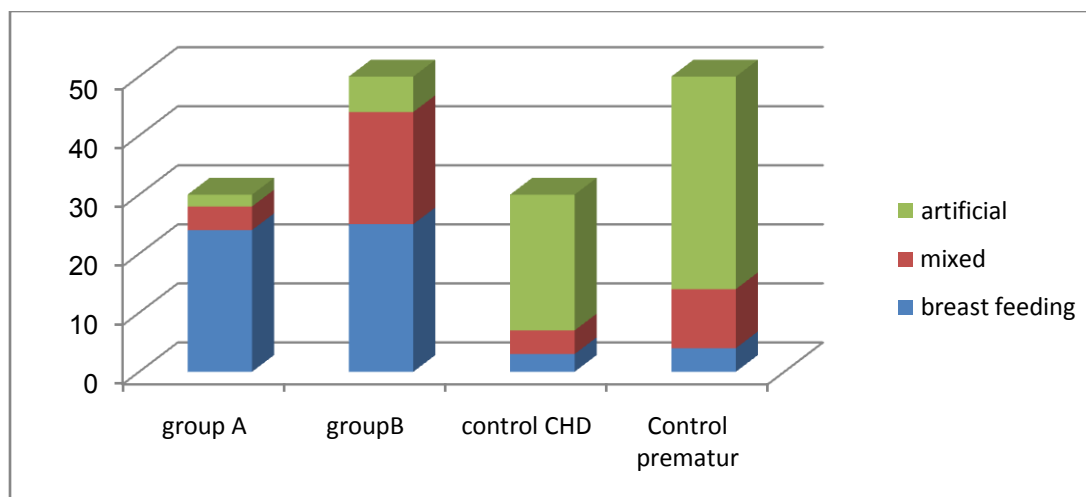


Figure (2):- effectiveness of breast feeding between group A&B and control(group C):show significant number with breast feeding didn't developed severe bronchiolitis

Discussion:-

Guidance from the American Academy of Pediatrics (AAP) for the use of palivizumab prophylaxis against respiratory syncytial virus (RSV) was first published in a policy statement in 1998. Guidance initially was based on the result from a single randomized, placebo-controlled clinical trial conducted in 1996–1997 describing an overall reduction in RSV hospitalization rate from 10.6% among placebo recipients to 4.8% among children who received prophylaxis. The results of a second randomized, placebo-controlled trial of children with hemodynamically significant heart disease were published in 2003 and revealed a reduction in RSV hospitalization rate from 9.7% in control subjects to 5.3% among prophylaxis recipients.(13).

From our study we found there is significant reduction in RSV hospitalization rate from 70% in control group to 8.33% among infants with CHD were prophylaxed with palvizumab for 3 doses and to zero percent for those infant who received 5 doses of palvizumab and no death was reported(P.value less than 0.05).regarding premature infants, the reduction rate of hospitalization was from 78% in control group to 4% among premature infants were prophylaxed with 3 doses of palvizumab and to zero percent for premature infants who received 5 doses of palvizumab .

The mortality rate was zero in both prophylaxed groups and 3.3% for patients with CHD & 8 % for premature infants whom didn't receive palvizumab.

Therefore in this study we found there is significant reduction in morbidity and mortality.

The effectiveness of palvizumab in this study indicat that RSV is the main cause of bronchiolitis in our country. From our study we found that breast feeding had significant preventive measure that contribute to decrease the incidence and severity of bronchiolitis in both groups,this result go with the guidelines were updated in 2014 and recommend the use of breast feeding is a must. (5)(6)(1).

The results of Farber didn't go with our study, Farber found that the effect of palivizumab administration on hospitalization for bronchiolitis with or without an RSV diagnosis among healthy preterm infants born at 29 to 36 weeks of gestation during their first RSV season occurring in 2012, 2013, or 2014, limited utilization and benefits of palivizumab were observed and no benefits were observed for those born between 33 and 36 weeks. , new diagnostic testing for a wide variety of respiratory pathogens has demonstrated that multiple viruses, such as coronaviruses, human metapneumovirus, influenza virus, parainfluenza virus, and rhinovirus, circulate at the same time as RSV and cause bronchiolitis syndromes that are indistinguishable clinically from RSV.(14)

Palivizumab, a humanized monoclonal anti-RSV antibody, was shown to reduce hospitalizations and the clinical severity of RSV infection in high risk infants and children by 55% in a randomised controlled trial. (15)

The result of our study go with (IMPact-RSV study group 1998) was a multi-center, randomized, double-blinded, placebo-controlled trial that enrolled 1502 children (500 placebo and 1002 palivizumab recipients). Eligible participants were ≤ 35 week's gestational age, they found that the monthly prophylaxis resulted in a 55% relative reduction in RSV hospitalization (10.6% placebo, 4.8% palivizumab) with significant relative reductions in premature infants (78%). (11).

My study like two studies (16, 17) which included any premature infant born at GA < 36 weeks reported a statistically significant reduction in RSV hospitalization among prophylaxed compared to nonprophylaxed infants (19–29% rate reduction), but one study (18) which included late preterm infants (GA 33 to 35 weeks) reported a non significant reduction of 0.6%.

our study go with another studies of a total of 1287 children less than or equal to 2 years of age with hemodynamically significant CHD that was uncorrected or palliated were randomized in a double-blind, placebo-controlled trial conducted in Canada, France, Germany, Sweden, United Kingdom, and the United States found that Palivizumab prophylaxis was significantly associated with a 45% relative reduction in hospitalization rates. RSV hospitalization rates were 9.7% in the placebo group and 5.3% in the palivizumab group. (11)

A similar rate of hospitalization (4.6%) was found in 108 Japanese infants with CHD who received palivizumab with no children requiring mechanical ventilation and no mortality (Saji et al 2005). Other investigators have found similar or lower incidences of RSV hospitalizations in patients with hemodynamically significant CHD than the 9.7% rate reported in the placebo arm of Feltes et al (2003) who were not given palivizumab prophylaxis (Duppenthaler et al 2004; Meberg and Bruu 2006). RSV hospitalization rates in patients with severe CHD in Norway were 9.2% (Meberg and Bruu 2006) and only 2.4% in Switzerland (Duppenthaler et al 2004). (11)

Like our study, in 3 studies (19, 20, 21) which included infants born at GA < 32 weeks, the rate of RSV hospitalization was lower in infants who received palivizumab, the reported reduction ranged between 1.2% and 12.4% and this reduction was statistically significant and this is go with my study

Like our study, a study (22) based on hospital record review investigated the effect of palivizumab among 266 children aged < 2 years with CHD, they found that the rates of RSV hospitalization in prophylaxed children were 19% lower than for nonprophylaxed infants from a different RSV season.

Another cohort study (22) among children with CHD reported a reduction in RSV hospitalizations from 7–9 cases/year when children received palivizumab ad hoc to 2–3 cases/per year when prophylaxis was administered systematically (5 doses) and in accordance with recommendations.

Like our study, a cohort study (23) that included premature infants of GA 29 to 32 weeks also reported a lower RSV hospitalization rate in children prophylaxed according to the standard recommendation of 5 doses, compared with the rate in children who received inadequate prophylaxis, that is, children who did not receive the recommended five full doses (3.3% versus 8.1%,).

Our study didn't go with the only study (24) that stratified by gestational age did not find a significant reduction in hospitalization rate in prophylaxed infants born at GA 33 to 35 weeks without CLD. The review of the studies suggests that groups of children who are likely to benefit most from palivizumab prophylaxis are premature infants born at GA ≤ 32 weeks and children with CLD.

Recommendations:-

From this study we recommend:

1. the use of palvizumab in premature infants less than 35 weeks of gestation and their body weight less than 1500 gm as immune-oprophylaxis against severe bronchiolitis
2. the use of palvizumab in hemodynamically significant congenital heart disease: large VSD, large PDA, complete AV canal with heart failure, TOF and D-TGA.
3. 5 monthly doses of pavizumab is very effective in reducing the RSV associated hospitalization.
4. 3 monthly doses of palvizumab is also effective in preventing severe bronchiolitis specially if it was given during the RSV season

5. Breast feeding is recommended in all infants less than one year because of its protective effect against severe bronchiolitis.
6. We need further studies to see the effectiveness of palvizumab in patients with chronic lung disease, cystic fibrosis, neuromuscular disease and immunocompromised patients in order to reduce the rate of hospital admission
7. No significant side effects or anaphylaxis from intramuscular injection of palvizumab.
8. Palvizumab should be supplied for each pediatric hospital to be used in high risk infants.
9. Palvizumab not interfere with routine vaccination.

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List of abbreviation:-

CHD: Congenital Heart Disease
 RSV :Respiratory Syncytial Virus
 VSD: Ventricular Septal Defect
 PDA :Patent Ductus Arteriosus
 AV Canal: AterioVentricular Canal
 TOF :Tetralogy of Fallot
 D-TGA: D-transposition of Great arteries
 CO₂ :Carbon dioxide
 NG tube: Nasogastric tube
 CLD: Chronic Lung Disease

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RESEARCH ARTICLE

EFFECT OF FROZEN STORAGE AND CRYOPROTECTANTS ON FUNCTIONAL PROPERTIES OF TILAPIA (*Oreochromis mossambicus*) FISH

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Key words:-

Tilapia, frozen storage, cryoprotectants, apparent reduced viscosity, calcium ATPase Activity, gel filtration profile

Abstract

In the present investigation, changes in functional properties of proteins from tilapia (*Oreochromis mossambicus*) in the form of whole tilapia (WHT), water washed minced meat of tilapia without addition of cryoprotectants (WMT) and with cryoprotectants (WMCT) during frozen storage have been evaluated. Gradual reduction in moisture content and marginal increase in protein content was observed in all the samples with progressive frozen storage. The reduction in non protein nitrogen (NPN) content was lower in WMCT as compared to WHT and WMT. All the samples exhibited decreased calcium-ATPase activity as a function of frozen storage. WMCT had a higher activity index during frozen storage as compared to WHT and WMT. The enzymatic activities decreased with progressive frozen storage period with relatively higher activity in WHT. Proteins from washed meat with cryoprotectants had minimum change in reduced viscosity profile at single protein concentration during different periods of frozen storage. Changes in structure of proteins were evident from apparent reduced viscosity and gel filtration profile as a function of frozen storage.

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Introduction:-

Being a rich source of proteinaceous material, fish is consumed worldwide as a source of nutrition. Tilapia is becoming an increasing substitute for traditional white fish species. The consumption of tilapia in the year 2014 was around 5.4 MMT worldwide (Fernandes et al., 2015). Around 4% of the catch spoils as a result of lacuna in transportation and storage conditions (Jackson et al., 1997). In order to increase the consumer preference for tilapia, value addition is one of the options. Due to the presence of bones, preparation of value added products is problematic. To overcome the problem, production of mince from fish by mechanical means is a good alternative. Fish mince needs freezing and frozen storage and can be a good source of raw material for production of various value added products (Venugopal and Shahidi, 1998).

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Water washed mince meat blended with additives, frozen and stored under frozen storage is termed as surimi (Park and Lin, 2005). Few white flesh varieties are commercially used for production of large quantities of surimi. In India, surimi production accounted for 58000 million metric tonnes in the year 2011 (Park and Lin, 2013). Gelation behavior of fish on heat processing is an important characteristic in surimi production. Use of tilapia for production of surimi has been attempted (Pan et al., 2010). However, the information on changes in properties of tilapia proteins added with cryoprotectants during frozen storage is limited.

Due to the perishable nature of tilapia, frozen storage is required in order to maintain its quality. However, after prolonged storage even at temperatures below -20°C , marked undesirable sensory changes in the product take place. There are many reports on frozen storage of tilapia in relation to its biochemical, sensory and microbiological quality (Ng and Bahurmiz, 2009). Protein denaturation and/or aggregation may take place during frozen storage of tilapia meat (Shenouda, 1980). Protein denaturation leads to alteration of functional and gelation properties (Badii and Howell, 2001). Cryoprotectants are generally used to prevent these undesirable changes in the myofibrillar protein and to preserve maximum functionality of surimi (Yoon and Lee, 1990). It is well documented that these sugars work by stabilizing actomyosin, increasing surface tension (Arakawa and Timasheff, 1982) and the amount of bound water, preventing protein water loss (Buttkus, 1970) and maintaining protein solubility (Herrera and Sampedro, 2002). A mixture of sucrose and sorbitol is the preferred as cryoprotectant for fish surimi as both have excellent cryoprotective effects on fish myofibrillar protein (Yoon and Lee, 1980).

Utilization of tilapia has been attempted to produce surimi and have been found to be suitable for surimi production (Park et al., 1990). Properties of the protein of fish intended for surimi production play a pivotal role in the end product quality (Murthy and Rajanna, 2011). In order to produce mince based products from tilapia, it is essential to have understanding of properties of proteins during frozen storage so as to improve the textural properties. The aim of this study was to evaluate the effect of freezing and frozen storage on the quality of tilapia with and without cryoprotectants. We investigated freeze-induced denaturation of samples by evaluation of Ca^{++} -ATPase activity, MIPase activity of tilapia fish mince with and without addition of cryoprotectants.

Methodology:-

Raw material:-

Fresh tilapia (*Oreochromis mossambicus*) caught from natural freshwater body near Mysore, Karnataka, India was used for the study. The length of fish used was 21 to 28.5 cm, weighing 150 to 340 g. The fish samples were washed and iced in the ratio of 1:1 (fish: ice) and transported to the laboratory immediately after harvest. Head and entrails were removed manually, washed with chilled water (3°C) and subjected to further analysis.

Chemicals and reagents:-

Sodium chloride, sodium di-hydrogen phosphate, di-sodium hydrogen phosphate, tri-chloro acetic acid (TCA) were procured from E-Merck India Limited. Ellman reagent was procured from Sigma Chemical co., St. Louis, USA. The synthetic Krehalan casings (copolymer of vinylidene chloride and vinyl chloride) of size 50mm x 280mm (diameter x length) were obtained from Kruhea Chemical industry Co. Ltd., Japan. All chemicals and reagents used in the present study were of AR or GR grade.

Freezing and frozen storage:-

From minced meat, surimi was prepared from minced meat (Nishiya et al., 1961) and mixed with cryoprotectants (w/w) such as sodium tri-poly phosphate (0.2%), sorbitol (4%) and sucrose (4%). The fish meat packed in polyethylene bags was frozen in air blast freezer at -35°C . The total time for freezing was 4 to 6 hours. Whole fish were frozen in similar fashion. All three, washed meat (WMT), washed meat with cryoprotectants (WMCT) and whole fish (WHT) frozen samples were stored in deep freezer (Vest frost, Holland) at $-20\pm 2^{\circ}\text{C}$ during the period of study. Samples were drawn at regular intervals up to 360 days frozen storage period.

Changes in properties of proteins from whole fish, washed meat and washed meat with cryoprotectants of tilapia fish meat during frozen storage:

Changes in moisture, total protein content and non-protein nitrogen (NPN) content:-

The samples were macerated well using a pestle and mortar and the macerated meat was used. Moisture and protein in the meat were determined as per AOAC, 2000. NPN content was determined by TCA method (Velankar and Govindan, 1958). About 5 g of meat was ground with 10 ml of 15% TCA for 5 min using pestle and mortar. The

slurry was filtered and made up to 50 ml with distilled water and 5 ml of aliquot was taken for nitrogen estimation by Kjeldahl method.

Changes in apparent reduced viscosity:-

Total proteins from tilapia meat were extracted using phosphate buffer (50 mM, pH 7.5) containing 1 M NaCl referred to as extraction buffer (EB) and the solution was used for viscosity measurements. Ostwald viscometer with capillary diameter of less than 0.5 mm was used. The temperature of the viscosity measurements was done at $25 \pm 1^\circ\text{C}$. The ratio of fish meat to buffer for extraction of proteins was 1:10. The clear supernatant obtained was taken for viscosity measurements after determining protein concentration by Lowry's method (Lowry et al., 1951). Appropriate dilutions of protein solution were made using EB as solvent so as to get different concentration of protein solution. A known quantity of protein solution (10 ml) was taken into viscometer and equilibrated in a water bath at $25 \pm 1^\circ\text{C}$ for 15 min. Time taken by the protein solution to pass through the two markings on the viscometer was monitored with the help of a stop watch (Rocar, Switzerland) with 0.2 sec sensitivity. The solution was taken up to mark by suction and the flow time in seconds required to flow between the markings in viscometer was recorded. For each protein concentration 3 readings were taken and the average flow time was used for calculation.

Apparent reduced viscosity was calculated using the following formula (Yang, 1961) and expressed as ml/mg of protein in the solution. Apparent reduced viscosity was plotted against different protein concentration (mg/ml).

$$\eta_{red} = \frac{t_1 - t_0}{t_0 \times C}$$

Where;

η_{red} = reduced viscosity (ml/mg)

t_1 = flow time for protein solution (sec)

t_0 = flow time for solvent, EB (sec)

C = concentration of protein in the solution (mg/ml)

Changes in calcium ATPase activity:-

Calcium ATPase activity was measured by using spectro-photometric method (Noguchi and Matsumoto, 1970). About 1 g of meat was homogenized in 10 ml of 0.05M Tris-HCL buffer, pH 8.0. The homogenate was centrifuged at $8000 \times g$ using IEC B22 high speed refrigerated centrifuge for 15 minutes at 4°C . The supernatant thus obtained was used as a source of enzyme. The reaction mixture consisting of 0.06 ml of 0.05 M adenosine tri-phosphate (ATP) solution, 0.4 ml calcium chloride (0.1 M), 2 ml (0.05 ml) Tris-HCl buffer (pH 8.0). About 0.4 ml meat extract was added to reaction mixture and incubated for 5 minutes at $28 \pm 1^\circ\text{C}$. The reaction was stopped by adding 2 ml of 15% TCA. All the assays were done with both samples blank as well as reagent blank in triplicate. The mixture was filtered, the inorganic phosphorous in the filtrate was determined (Taussky and Shorr, 1952). To the 2 ml filtrate, 3 ml of ammonium molybdate ferrous sulphate solution (freshly prepared) was added. The intensity of the colour developed was read at 660 nm. The concentration of liberated inorganic phosphate was measured using standard curve obtained by using potassium di-hydrogen phosphate as a standard. The activity was expressed as $\mu\text{g P}_i/\text{mg protein/ minute}$. The initial calcium ATPase activity was taken as 100 and designated as activity index.

Changes in modori inducing proteases (MIPase) activity:-

Modori inducing protease in tilapia meat was determined spectrophotometrically (An et al., 1994). The crude enzyme extract was prepared by homogenizing 5 g of fresh tilapia meat with 15 ml phosphate buffer (50 mM; pH 7.5) at 3000 rpm for 2 minutes in the homogenizer (Nihon Seiki Kogyo Co., Tokyo, Japan). The homogenate was centrifuged at $8000 \times g$ for 15 minutes. The supernatant was diluted with phosphate buffer in the ratio of 1:1 and used as crude enzyme extract. The substrate solution was prepared by dissolving 4 mg casein (according to Hammerstein) in 1.25 ml phosphate buffer (50 mM, pH 7.5) and making up to 2 ml with distilled water. To determine the modori inducing protease activity, 2 ml of the substrate solution was pre-incubated at $55 \pm 1^\circ\text{C}$ for 5 minutes in water bath. To this 0.5 ml of crude enzyme was added and again incubated at $55 \pm 1^\circ\text{C}$ for 1 hour for the enzymatic reaction to occur. The reaction was terminated by adding 1 ml cold 20% TCA. The slurry was centrifuged at $8000 \times g$ for 15 min at 4°C . The supernatant containing the hydrolyzed oligo peptide was collected without disturbing the pellet. The oligo peptide content in the supernatant was determined by measuring the absorbance at 280 nm in a UV- VIS Spectrophotometer (Spectronic 21 Bausch and Lomb, USA). The blank was prepared in the same manner except that the crude enzyme extract was added immediately after TCA precipitation. The enzyme

activity was expressed as the difference of absorbance at 280 nm between the sample and the blank. The concentration of protein in the crude enzyme was estimated by measuring absorbance at 280 nm.

Changes in gel filtration profile:-

Gel filtration profile of WHT, WMT and WMCT samples as a function of frozen storage was carried out on a sepharose 6B gel packed in a column of 1.5 x 80 cm (dia x height) using EB as an eluent. Total bed volume of the column was 160 ml. The void volume (V_o) of column was determined using blue dextran and was found to be 50 ml. Two grams of meat sample was homogenized with 18 ml EB using a homogenizer (Nihon Seiki Kogyo, Co., Japan) and the homogenate was centrifuged at 9000 X g using IEC refrigerated centrifuge for 15 min at 4°C. A clear supernatant (1.5 ml) was loaded on to the top of gel column after determining the protein concentration. The protein concentration loaded to the column varied between 10-12 mg/ml. Flow rate was adjusted to 30 ml/hour and fractions of 3 ml were collected manually. The protein concentration was measured by taking absorbance at 280 nm using a UV/VIS spectrophotometer (Spectronic – 21, Bausch and Lomb, USA). A plot of absorbance at 280 nm against elution volume was obtained to get the gel filtration profile.

Statistical Analysis:-

In order to assess the effect of frozen storage period and cryoprotectants from different sources on various properties of tilapia meat, two-way analysis of variance (ANOVA) without replication was carried out using MS-Excel.

Results and Discussion:-

Physical characteristics of fresh tilapia meat:-

The fishes used in the present study were relatively big in both size and weight. The dressing yield (after removal of head, entrails) was 64.3% and the meat yield from the whole fish was 44%. Normally bigger size fish will give higher yield compared to small size fish. The yield of separated meat of the fresh water fishes like common carp and silver carp varies from 40-47 % (Arekere, 1993; Siddaiah et al., 2001).

Changes in moisture, total protein and non-protein nitrogen (NPN) content as a function of frozen storage period:-

The changes in moisture content of whole fish (WHT), washed meat (WMT) and washed meat with cryoprotectants (WMCT) during frozen storage are given in Fig. 1a. The initial moisture content of washed meat was higher than whole fish meat. The increased moisture content of washed meat may be attributed to hydration of myofibrillar proteins (Suvanich et al., 2000; Karthikeyan et al., 2004). A reduction in moisture content was recorded as a function of frozen storage period in all the samples. It is usual to find reduction in moisture content of fish and fishery products during frozen storage because of dehydration (Shenouda, 1980). The moisture content of WMCT sample registered minimum changes during frozen storage. The variation in the moisture content of WHT, WMT and WMCT as a function of frozen storage was significant ($p < 0.05$). Significant difference was also observed between WHT, WMT and WMCT for similar storage days ($p < 0.05$). Similar type of reduction in moisture content was observed in croaker fish washed meat samples (Basavanagouda, 2001) and in pink perch surimi (Nopianti et al., 2012). Cryoprotectants added in the surimi reduced the protein aggregation by increasing the hydration and prevented drastic fall of moisture content.

The total protein content of samples as a function of frozen storage period showed a marginal increase in all the samples and is given in Fig. 1b. The initial protein content of whole fish and washed meat was found to be 17.47 and 12.80% respectively. Reduced protein content on washing has also been reported (Fogaca et al., 2015). Addition of cryoprotectants to washed meat reduced the total protein content. There was a gradual increase in protein content in all the three samples during frozen storage. Protein content of WHT sample was significantly higher than WMT and WMCT throughout the storage period ($p < 0.05$). WMCT sample registered a higher increase in protein content than WMT possibly due to higher reduction in moisture content.

The changes in non-protein nitrogen (NPN) content of tilapia meat samples during different periods of frozen storage are given in Fig. 1c. There was a significant decrease in NPN content in all the samples throughout the storage period ($p < 0.05$). The reduction in NPN content was higher WHT than WMT followed by WMCT. The results of analysis of variance indicated that the changes in NPN values were altered both due to storage and sample condition. The loss of NPN constituents in frozen fish may be observed due to the leaching effect during thawing. The initial NPN content of whole fish meat was 365 mg%, which was more than that of washed meat indicating that washing has reduced the NPN content. Similarly washed meat of pink perch showed reduction in NPN content

(Karthikeyan et al., 2006). The loss of NPN constituents in frozen fish meat is mainly due to leaching effect during thawing.

Changes in apparent reduced viscosity as a function of frozen storage period:-

Measurement of viscosity of protein samples has been used to determine the protein denaturation. A plot of frozen storage period vs apparent reduced viscosity at a single protein concentration (3 mg/ml) was obtained (Fig. 2a). All the three frozen stored samples initially showed an increasing trend and later the value decreased. The decrease was more rapid in washed meat samples without cryoprotectants. Denaturation of myofibrillar proteins lead to changes in viscosity due to the change in particle axis ratio (Suzuki, 1981). It could be seen that proteins from washed meat with cryoprotectants had minimum change in reduced viscosity profile at single protein concentration during different periods of frozen storage. This is due to the preferential hydration of protein molecules induced by added cryoprotectants. Similar reduction in apparent reduced viscosity has been reported for horse mackerel, blue whiting and croaker (Colmenero and Borderias, 1983; Basavanagouda, 2001). The apparent reduced viscosity of protein from tilapia clearly indicated that washed meat with cryoprotectant has undergone lesser degree of conformational changes than that of proteins from whole fish and washed meat.

Changes in calcium ATPase activity as a function of frozen storage period:-

The calcium activated ATPase activity which arises from globular head of myosin, S1 fraction is generally taken as an index of denaturation of fish proteins during ice storage and frozen storage (Lehninger, 1993; Lin and Park, 1998). The calcium ATPase activity of WHT, WMT and WMCT as a function of frozen storage period is given in Fig 2b. The reduction in enzyme activity index was higher in whole fish meat than washed meat and washed meat with cryoprotectants. The decrease in ATPase activity of all the three frozen tilapia samples was evident during different periods of frozen storage. Washed tilapia meat with cryoprotectants had a higher activity index during different periods of frozen storage in comparison to whole and washed tilapia meat. The effect of cryoprotectants in minimizing the aggregation/ denaturation is the possible reason for the retention of higher ATPase activity. Ca^{++} ATPase activity of actomyosin from tilapia fillets stored over 2000 atm showed apparently falling trend and the protein denatured evidently (Ko and Hsu, 2002). The present investigation indicates sucrose and sorbitol could minimize the reduction in Ca^{++} ATPase activity during frozen storage by retarding the deteriorative reaction. Similar observations have been recorded for frozen stored tilapia (*Sarotherodon nilotica*) meat added with cryoprotectants (Zhou et al., 2006).

Changes in modori inducing protease (MIPase) activity as a function of frozen storage period:-

The MIPase are group of cathepsins present in muscle cell, hydrolyzing myofibrillar proteins or any other suitable substrates. The modori inducing protease activity of tilapia samples during different periods of frozen storage is given in Fig. 2c. The activity of enzyme decreased with increase in frozen storage period and relatively higher activity was observed in whole fish meat. It is evident that the initial MIPase activity of WHT was significantly higher than that of WMT and WMCT. It clearly suggests that MIPase activity is associated with sarcoplasmic fraction of the muscle and hence reduction in activity was observed in washed meat because of washing. Degradation of protein network during gelation process by cathepsin has been studied extensively. Tilapia surimi is known to have high proteolytic activity at 60°C (Yongsawatdigul et al., 2000). Sarcoplasmic fraction and MIPase activity are associated hence, washing reduces the MIPase activity.

Changes in gel filtration profile as a function of frozen storage period:-

Changes in gel filtration profile of WHT, WMT and WMCT samples during different from storage periods are given in Fig. 3. The gel filtration profile of total proteins from WHT showed more of low molecular weight components at the initial stage with further marginal increase in the high molecular weight components with progressive storage up to 90 days after which a gradual decrease in concentration was observed. The elution volume of high molecular weight fraction was shifted from 67.89 ml in the fresh sample to 90.65 ml at the end of 240 days of storage. At the end of 360 days of storage the elution volume was shifted to 74.4 ml. This clearly demonstrates that there is initial dissociation after which the association phenomenon has taken place. Altered elution profile indicated association-dissociation phenomenon (Binsi et al., 2007). In WMT sample, decrease in high molecular weight components at the end of 240 days storage period was recorded whereas WMCT sample revealed increase in high molecular weight components up to 90 days followed by low molecular weight components during storage period. WMT sample showed the dissociation phenomenon immediately after freezing whereas, WMCT sample exhibited relatively higher concentration of high molecular weight components throughout the storage period. The indication of high molecular weight components at relatively higher concentration indicated lesser extent of degradation. The data

obtained on apparent reduced viscosity and gel filtration profile supported that there is a minimum conformational change in proteins in WMCT than WHT and WMT. Structural change of proteins during frozen storage was evident from reduced viscosity and gel filtration profile. Changes in protein structure as a function of frozen storage of common carp meat, tilapia were also reported (Ganesh et al., 2006; Murthy et al., 2011). Peak reduction may be attributed to removal of free amino acids, peptides and other low molecular weight compounds (Suvanich et al., 2000).

With the increasing production of tilapia catch, it is necessary to think towards better utilization of tilapia in order to get higher economic returns. Better utilization relies on understanding of fish in relation to the protein properties. Properties of proteins exhibited changes in structure as evident from gel filtration profile and apparent reduced viscosity values in all the samples namely whole fish, meat without and with cryoprotectants as a function of frozen storage. In order to utilize the catch throughout the year, use of cryoprotectants and frozen storage play an important role. Water washed frozen stored tilapia meat with cryoprotectants could be used in product development.

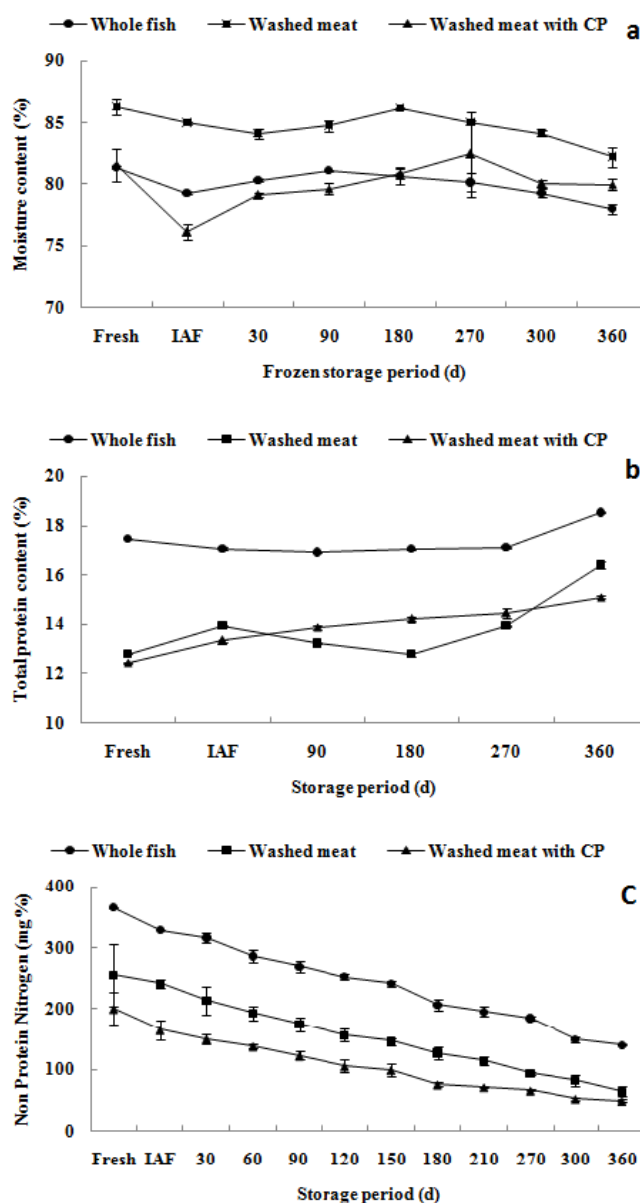


FIGURE 1:- Changes in composition of whole tilapia (WHT), washed tilapia meat without cryoprotectants (WMT) and washed tilapia meat with cryoprotectants (WMCT) as a function of frozen storage; a. Moisture content (%); b. Protein Content (%); and c. Non-protein nitrogen (NPN) content (mg%)

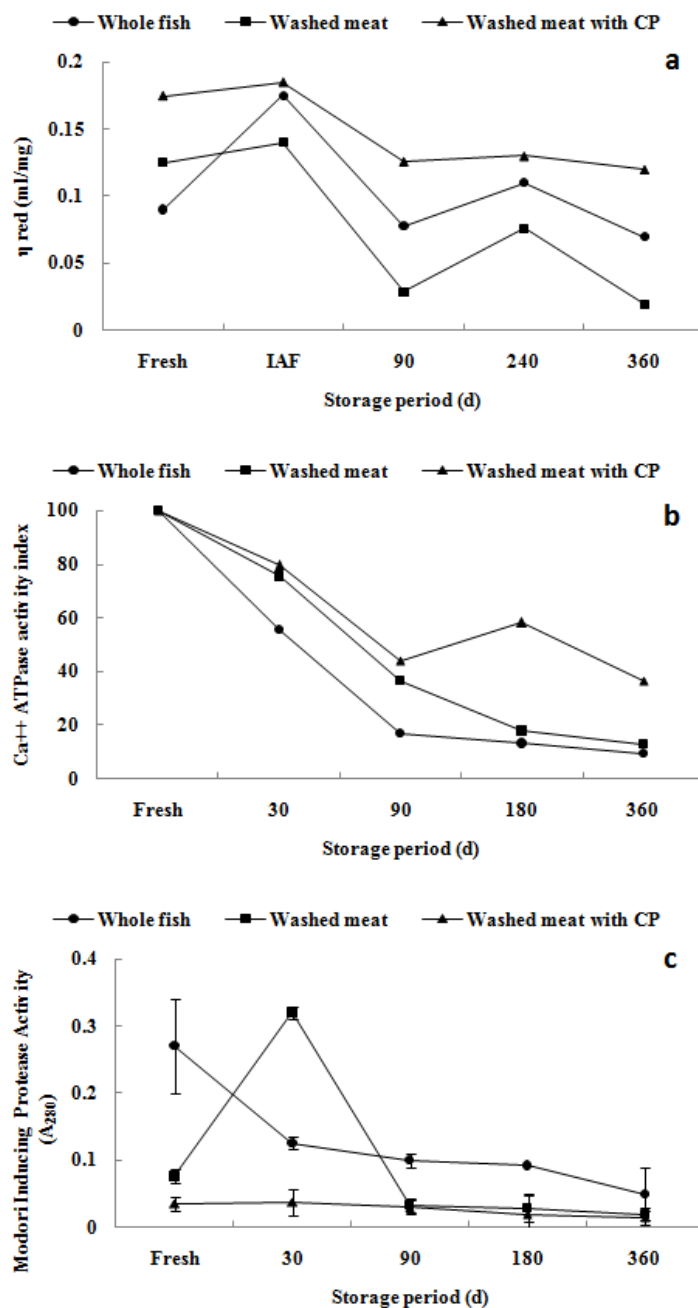


FIGURE 2:- Changes in properties of proteins of whole tilapia (WHT), washed tilapia meat without cryoprotectants (WMT) and washed tilapia meat with cryoprotectants (WMCT) as a function of frozen storage; a. apparent reduced viscosity (ml/mg); b. Calcium ATPase activity index; and c. Modori inducing proteases activity in terms of absorbance

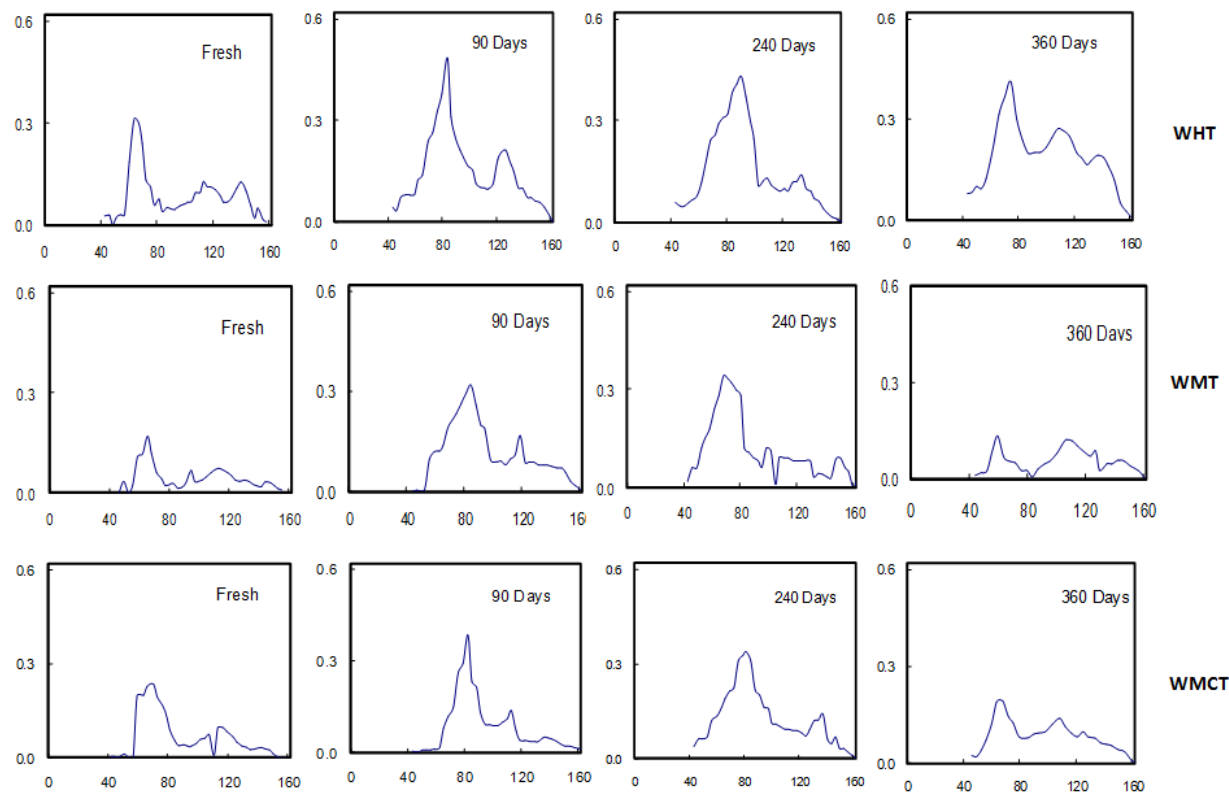


FIGURE 3:- Changes in gel filtration profile of proteins of whole tilapia (WHT), washed tilapia meat without cryoprotectants (WMT) and washed tilapia meat with cryoprotectants (WMCT) as a function of frozen storage

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RESEARCH ARTICLE

ETHNOBOTANICAL STUDY AND CONSERVATION STATUS OF MEDICINAL PLANTS IN AND AROUND YAYO FOREST, OROMIA REGION, SOUTHWEST ETHIOPIA

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Abstract

The study was conducted to investigate the Ethnobotanical Study and Conservation Status of Medicinal Plants in and around Yayo forest. The purpose of the study was to document and analyze information on the use, conservation and sustainable use of medicinal plants. Data collection was accomplished through active participation of healers and familiar elders who practice traditional medicine locally. A variety of Ethnobotanical techniques were used to collect the data: semi-structured interview, field observation and group discussion. Data were analyzed quantitatively and by Microsoft office excels. A total of sixty nine (69) plant specimens belonging to thirty four (34) families and sixty one (61) genera were documented and identified both in the field and the National Herbarium, Addis Ababa University. From the total documented medicinal plant species, about 42.63% are used to treat human ailments while 36% are used to treat livestock ailments and 22.27% are used to treat both livestock and humans. Among the documented families, family Fabaceae had the highest species richness followed by Asteraceae, Euphorbiaceae, Lamiaceae, Moraceae, and Poaceae. To protect and conserve this forest, appropriate management strategy and awareness creation is very crucial. As it was recommended, some cultural believes and traditional practices associated with traditional medicines were found to be contributed much for the conservation of medicinal plants in the study area.

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Introduction:-

Ethnobotany is a multidisciplinary science defined as the interaction between plants and people or the study of the relationship between plants and people: From "ethno" - study of people and "botany" - study of plants (Debela Hunde, 2001). It focus on how plants have been or are used, managed and perceived in human societies and includes plants used for food, shelter, medicine, divination, cosmetics, dyeing, textiles, for building, tools, ornamentation, currency, clothing, rituals, social life and music (Choudhary *et al.*, 2008).

Basic quantitative and experimental ethnobotany includes basic documentation, quantitative evaluation of use and management and experimental assessment. Today, ethnobotanical surveys include applied projects that have the potential to improve poverty levels of these people, allowing them to make more educated decisions about their

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future directions (Graves, 1996). These new approaches enhance the quality of the science, provide compensation for the cultural groups and take into account environmental concerns.

This modern approach is based on an interdisciplinary team usually composed of an Ethnobotanist, an anthropologist, an ecologist and a physician (Etana Tolasa, 2007). The preliminary results of a study on behalf of WHO has shown that the number of individuals using medicinal plants is large and on the increase, even among young people. It is not just in developing countries that medicinal plants are important (Endalew Amenu, 2007).

Medicinal plants and traditional medicine play an important role in the health care system of most developing countries. In Ethiopia about 80% of human population and 90% of livestock rely on traditional medicine (Choudhary *et al.*, 2008). In light of the importance of these resources that provide the health care needs for the majority of human and livestock population in Ethiopia, and also the fact that medicinal plants could be used as sources of revenue for farmers (STCP, 2005). Efforts made to conserve forests in general and medicinal plants in particular. However, it did not have significant impact as they did not take into account the interests of the local community by failing to include farmers in decision making processes (Teklu Tesfaye and Thomas, 2004). Lack of continuity of the already initiated conservation efforts, tenure insecurity, lack of awareness and participation by local communities in different project phases, impacts of population growth and resettlement are the major factors contributing to forest degradation (STCP, 2005).

Today many medicinal plants face extinction or severe genetic loss, but detailed information is lacking. For most of the endangered medicinal plant species no/less conservation action has been taken (Erlangung and Fakultat, 2009). Therefore, this study was designed to give scientific information and methodology on how to use medicinal plants in sustainable manner and how to conserve these medicinal plants.

Materials and Methods:-

Study Area:-

The research was conducted at Yayo district particularly in and around Yayo forest, which is found in Ilu Abba Bor zone, Oromia Regional State that is found at 564 km Southwest of Addis Ababa, the capital City of Ethiopia and 36Km from the zonal capital of Mettu Town. The study area is located between 8°2'42" North and 8°31'18" North latitude and 35°37'48" East longitude. The district has a total area of 162,901 hectares (1,629.01km²) that constitutes 9.97 percent of the total land of the zone.

Yayo is one of the few areas in the country that is endowed with a variety of vegetation types including few species found only in Ethiopia. This forest covers about 90,890.7 hectares constituting 58.8 percent of the total area of the district. Yayo forest represents the largest (10,000 ha) undisturbed forest fragment kept for the conservation of wild *Coffea arabica* population earlier identified as a potential Coffee gene reserve (Teklu Tesfaye and Thomas, 2004). The mean annual temperature is about 23°C ranging between 18.59°C mean minimum annual temperature and 27.88°C mean maximum temperature. The physical conditions and variations in altitudes have resulted in a great diversity of Climate, Soil and Vegetation which in turn, has brought about the evolution of many plant species with large diversity. The rainfall pattern of the districts varies annually from 1,191.6 to 1,960.7mm showing variations from year to year. It is a unimodal type of rainfall that increases from May to October and declines in November. The district has three agro-climatic zones which includes highland (*Badda*); temperate (*Badda-daree*) and lowland (*Gammoojji*). Such diverse climatic conditions and habitats partly contributed to the occurrence of high species diversity in plants and animals, making Ethiopia.

Study Design:-

Materials used for the study was Plant cutter, Plant frame, Magazine, Blotter and Belt. The interview of key informants, focus group discussion, direct observation, and household survey were the main techniques used in the primary data collection. In-depth interview was conducted with experts and professionals at different levels of administrative staff, community representatives, elders, traditional healers and farmers. The plant specimens were identified in the field and by consulting experts. In addition to this it was compared with plants specimens which was already collected and identified from different parts of the country and found in the National Herbarium of Addis Ababa University, Ethiopia; and by referring the flora book of Ethiopia and Eretria. The name of family, genus and species as well as growth habit were identified and recorded.

Methods of Data Collection:-

Quadrants Methods: Quadrants (20m×20m or 400m²) in size were laid down purposefully along transects at 100 meters distance in three sites which was randomly selected in the site of study area for collecting of plant specimens. Frequency of medicinal plant individuals were counted and recorded. GPS was used to record the data of altitude, latitude and longitude of each quadrant of the study area. The collected data were recorded, summarized and entered into Microsoft offices excel for analysis. In the case of data analysis different methods for analyzing of quantitative data were used.

Results:-

Medicinal Plant Resources of the Study Area:-

A total of 69 medicinal plants were recorded in the sites of study area. Out of these collected and identified plant species, the greater numbers of Medicinal plants were from the wild vegetation. These species belong to thirty four (34) different families and sixty one (61) genera. Among these thirty four (34) families, family Fabaceae was represented by ten (10) species followed by Asteraceae, Euphorbiaceae, Lamiaceae, Moraceae, and Poaceae were represented by four (4) species for each. Family Meliaceae was represented by three (3) species while Acanthaceae, Boraginaceae, Malvaceae, Musaceae, Myrtaceae, Rutaceae, Verbenaceae and Zingiberaceae were represented by two (2) species. The remaining sixteen families had one (1) species each (Figure 1).

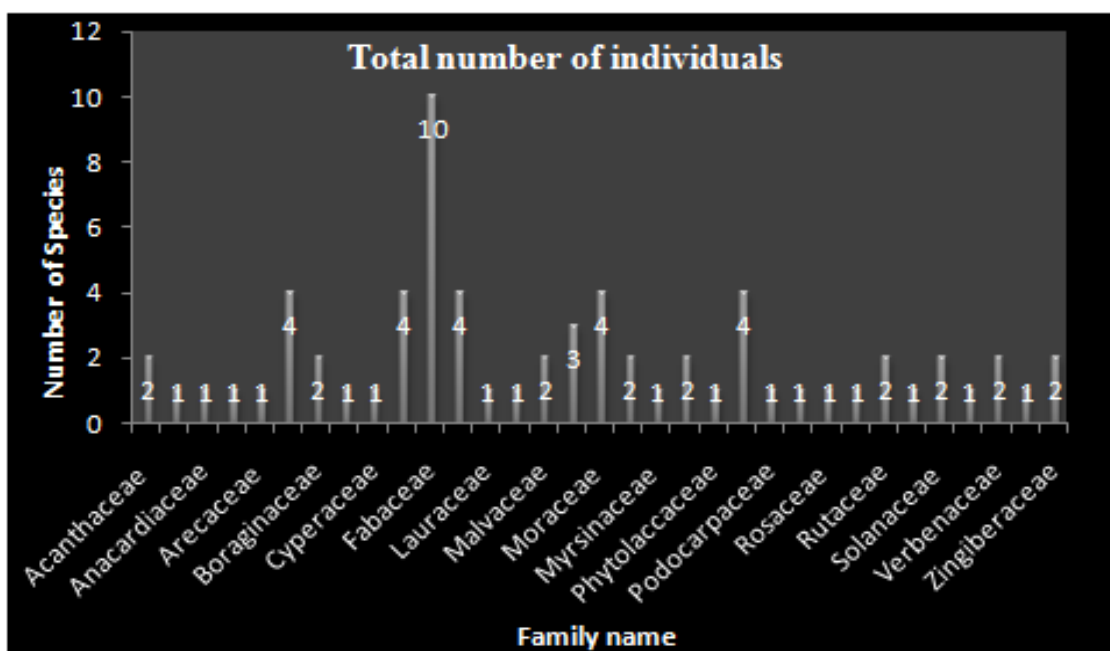


Figure 1:- The families' name with total number of individuals documented in the study area

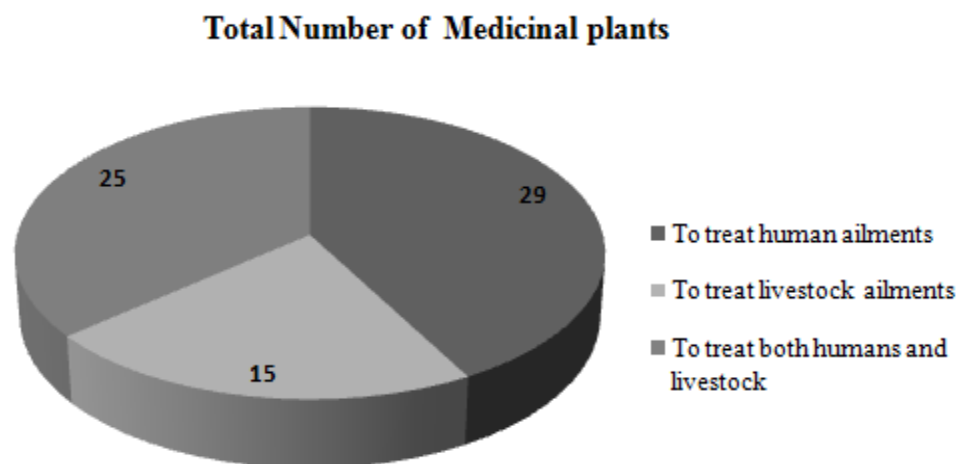
The result point out that relatively more family Fabaceae were found at the sites of study area and they are used as medicinal values for treatment of disease for both human beings and livestock compared to the remain families. Concerning habit diversity; herbs were the most common and stood first with (23 species), followed by trees (22 species) and shrubs (19 species, and climbers (2 species) and the remaining were like epiphytes.

From the total documented medicinal plant species from the study area, about 42.63% were used to treat only human ailments while about 36% were used to treat livestock ailments and about 22.27% were used to treat both livestock and humans. The sustained existence and utilization of such a large number of medicinal plants by people in the study area indicates that the majority of the people used and continued to use indigenous medicinal practices for different ailments (Table 1).

Table 1:- The percentage use of medicinal plants to treat various ailments found in the study area

Useful Medicinal Plants to Treat Various Ailments	Total percentage
To treat human ailments	42.63%
To treat livestock ailments	36%
To treat both humans and livestock	22.27%

From 69 plant species, 29 plant species were used to treat human ailments while 25 plant species were used to treat livestock ailments and 15 plants species were used to treat both humans and livestock respectively (Figure 2).

**Figure 2:-** The number of useful medicinal plants used for treatment of various ailments

Threats and Conservation of Medicinal Plants:-

Threats to Medicinal Plants:-

The cause of threats to medicinal plants can be generally grouped in to natural and human induced factors. However, as reported in this study most of the causes for the threats to medicinal plants and associated knowledge were the anthropogenic factors such as deforestation due to over exploitation of plants for different uses/charcoal, fire woods, construction woods, overgrazing, cutting and burning of plants to create new agricultural lands ,medicinal plants trade for different uses and others.

The results of the present study showed that agricultural expansion, fuel woods (charcoal and fire woods), harvesting for construction and urbanization and over grazing and medicinal plants for the threats to medicinal plants and associated knowledge of the plants in the study area. The other reason for the threats of the knowledge of medicinal plant was modernization and refusal to practice/ inherit the knowledge by new generation. As reported by the informants, the expansion of modern health institutions, schools some environmental and cultural modifications reason for the losses (threats) of the knowledge of medicinal plant.



Figure3:- Collecting of plants for firewood and other purposes by local community.

Discussion:-

Medicinal principles are present in different parts of the plant like root, stem, bark, heartwood, leaf, flower, fruit or plant exudates. These medicinal principles are separated by different processes; the most common is being extraction. Extraction is the separation of the required constituents from plant materials using a solvent (Paroda, 1993).

Descriptions of the most frequently reported medicinal plants used to treat different ailments

***Vernonia amygdalina* Del. (Astraceae) Eebicha (Afaan Oromoo):-**

Is shrub or small tree usually branched from near the base and it is 2-10m high. The leaves are used to wash the materials during the preparation of locale beer (Tella) and katikala (Areke) with that of *Rhamnus prinoides*. In Ethiopia it is found in many parts including Bale, Gamo-gofa, Gonder, Gojam, Tigray, Shewa, and Ilu Abba Bor (Edwards *et al.*, 2000). Elsewhere in Ethiopia, the leaves and bark are bitter and used in local medicine for instance, in the study area it is used for the treatment of skin infections and sudden sickness (Dhukkuba tasaa).

***Solanum incanum* L (Solanaceae) Hiddii (Afaan Oromoo):-**

Solanum incanum is a shrub up to 1.5m high, but often less, seasonally deciduous, found in heavily grazed areas, Waste places, in altitude of above 2100m, Solanaceae. In Ethiopia it is found in Gonder, Sidamo, Bale, Shewa, Tigray, Gamo-gofa and Ilu Abba Bor. This form is common in Somalia and the Sudan and widespread in tropical Africa, the Middle East and India (Edwards *et al.*, 2000).

***Ocimum lamifolium* Hochst .ex Benth. (Lamaceae) Damakesee (Afaan Oromoo):-**

Is sub shrub or shrub, 0.7-3m tall a very common species of primary and secondary mountain forest and Bush lands, tall grasslands, rarely also cultivated as an ornamental. In Ethiopia it is found in Gojam, Gonder, Kefa, Shewa, Ilu Abba Bor, and Wollega. In Africa it found in East Africa, Malawi, Democratic republic of Congo and Cameroon (Edwards *et al.*, 2000). In the study area this plant is used for treatment of head ache, eye infection and other related diseases.

Descriptions of the most frequently reported medicinal plant species used to treat livestock:-

***Nicotiana tabacum* L. (Solanaceae) Tamboo (Afaan Oromoo):-**

An erect and it is annual or biennial herb growing to 2.5m high. It is a cultivated plant in home gardens in some parts of the country and sometimes escaped into waste ground and along streams at altitudes range of between 1700 and 2400m, Solanaceae. In the study area, it is found in farmers' home garden or under the shade of life fence. It has a medicinal use for internal parasites, Trypanosomiasis, eye infection and headache. The local people use this plant

to treat for human disease). Leaf of *Nicotiana tabacum* is pounded with root of *Carissa spinarum*, mixed with water and cup of tella given to patient. In addition, dried and powdered leaf of *Nicotiana tabacum* sniffed by human for relive to headache, and its leaves with the leaves of *Capparis cartilaginea* used to treat cough.

Descriptions of the most frequently reported medicinal plants used to treat both human and livestock disease ***Croton macrostachyus* Del. (Euphorbiaceae) Bakkannisa (Afaan Oromoo):**

It is shrub or tree, 2-25m long. Found in forest margins and secondary woodlands, extending in to disturbed areas and along edges of roads, mostly in soils of volcanic origin, an altitude range so far recorded for the plant is 500-2350m, Euphorbiaceae. In Ethiopia, this plant is found in Tigray, Gonder, Gojam, Wello, Shewa, Arsi, Wollega, Ilu Abba Bor, Kefa, Sidamo, Bale and Harerge. In Africa it is distributed west to Guinea, south to Angola, Zambia, Malawi and Mozambique (Edwards *et al.*, 2000). The stem bark of this species is also known for its medicinal value in treating leprosy. In the study area this plant is more effective for the treatment of ringworms, gonorrhea and rabies.

***Phytolacca dodecandra* L.Herit. (Phytolaccaceae) Handodee (Afaan Oromoo)**

It is semi- succulent, scrambling shrub to 10m tall or more. It grows in ever green bush lands, forest edges and distributed places in altitudinal range of 1500-3000m, Phytolaccaceae. In Ethiopia, this plant is found distributed in Tigray, Bale, Gamo-Gofa, Gonder, Wollo, Gojam, Wellega, Shewa, Ilu Abba Bor, Kefa, Arsi, Sidamo and Hararge regions. It is also found in other African countries in Eritrea, Madagascar and in tropical and South Africa. It is widely used as soap specially for cleaning cotton cloths, also as Molluscicides for the control of the snails that are vectors of Bilharzias. The importance of this plant due to molluscidal properties has led to setting up of the Endod Foundation, with its head quarter in Addis Ababa. Endod varieties with high molluscidal content have been identified and are now being cultivated by farmers. In the study area this plant is used for treatment of malaria and rabies (Edwards *et al.*, 2000).

Conservation of Medicinal Plants:-

Although various threats have profound effect on medicinal plants, local people attempt to grow medicinal plants in home garden though the effort is minimal. For instance, some people and/ or office have started conserving the plants by in-situ method (in original/natural habitat), fenced/protected pasture land at different worship areas (churches, mosques, etc) in their (farms' field/farm margins and so on. And also conserving by ex-situ method/ outside the original/ natural habitat/ like in and around their farmers' home gardens, live fences of the gardens, plantation fields, and so on were started in the study area. As reported by Moa Megersa (2010), home gardens are central target for in-situ and ex-situ conservation of traditional medicinal plants.

Conclusions:-

The study reported that medicinal plants have revealed that there was an insight of the local indigenous knowledge on the utilizing of medicinal plants from the study area. The study was also inferred that sixty nine (69) different plant species belonging thirty four (34) families and sixty one (61) genera were identified and recorded. Of these different families documented; family Fabaceae was found to be represented by ten (10) species followed by Asteraceae, Euphorbiaceae, Lamiaceae, Moraceae, and Poaceae were represented by four (4) species for each. Family Meliaceae was represented by three (3) species while Acanthaceae, Boraginaceae, Malvaceae, Musaceae, Myrtaceae, Rutaceae, Verbenaceae and Zingiberaceae were represented by two (2) species. The left over families had one (1) species for each.

Out of sixty nine documented medicinal plant species from the study area about 29 (42.63%) of them were found to be used to treat human ailments while about 15 (36%) were used to treat livestock ailments and about 25 (22.27%) were used to treat both livestock and humans. The main use of medicinal plants are to treat human diseases than livestock ailments by local people as they pointed that; the local communities of the study area seek to find traditional medicine for their ailments first and then after for livestock ailments. Consequently, the local people possibly gained smaller understanding of medicinal plants to treat livestock ailments than knowledge of medicinal treating human ailments. Some local communities are knowledgeable about the use and conservation of medicinal plants. This is due to a given Indigenous practices somewhat contributed to the sustained use, management and conservation of medicinal plants and multiple use of indigenous trees. Traditional medicinal plants harvested mostly from wild followed by home gardens. Furthermore, the use of medicinal plants for different purposes in addition to their medicinal value such as charcoal and firewood, construction, useful materials and other anthropogenic activities. Thus, directly or indirectly leads to threat medicinal plants. Regardless of this fact, traditional healers still

depend to a greater extent on naturally growing species, as they believe those species in the wild vegetation are more powerful in the treatment of different ailments and health problems.

The results of the study also revealed that many wild species of medicinal plants are under pressures from various human induced factors. In addition, disinterest of young generation on traditional medicine; put the continuity of traditional medicinal knowledge under question. Because of young generation shows lack of attention to make use of and to be aware of medicinal plants from elder, the knowledge of traditional medicine possibly get rid of in the near future except that appropriate documentation is made. Furthermore, creation of awareness for young generation and other concerning body about medicinal plant uses and how elders used traditional plants to be continued to treat their livestock ailments, human ailments in proper manner and to use it in sustainable manner for future generation as well.

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RESEARCH ARTICLE

EVALUATION OF THE EMERGENCE AGITATION INCIDENCE IN CHILDREN WHO UNDERWENT DEEP SEDATION FOR TOOTH EXTRACTION.

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Abstract

Purpose: Emergence agitation (EA) is a condition where factors associated with patient, surgery and anesthesia play role in occurrence, and that may occur in any age groups—particularly well-defined in children and delay discharge. Emergence agitation has been investigated for medical interventions under general anesthesia in children, but not for dental treatment under deep sedation. The objective of this study was to investigate the incidence of EA in pediatric cases scheduled for tooth extraction under deep sedation and its relationship with drugs for sedation.

Materials and Methods: 120 cases, less than 12 years old and in ASA I-II group according to “American Society of Anaesthesiologists (ASA)” risk classification were scheduled for elective tooth extraction under deep sedation without premedication. The patients were admitted in operating room just according to our anesthesia protocol. With the aim of achieving Richmond Agitation Sedation Scale (RASS)=(-4), In Group K who allowed IV (Intravenous) cannula insertion: 2-3 mg.kg⁻¹ IV Ketamine was administered. In Group KM who also allowed IV cannula insertion: 0.5 mg of Midazolam was added to 2-3 mg.kg⁻¹ IV Ketamine. In Group S: 8% Sevoflurane in 50% O₂/N₂O through face mask (connected to a Mapleson C circuit) was administered to the Group S who did not allow inserting IV cannula. Detecting the RASS as -4 local infiltration anaesthesia was applied for the extraction of associated teeth. Following the procedure recovery was evaluated at T₀, than 15 min. later (T₁₅), 30 min. later (T₃₀), 45 min later (T₄₅), 60 min later (T₆₀) with RASS.

Results: The RASS values at T₀ were statistically significantly higher in Group K than those of other groups. No significant differences were found between groups for RASS mean at T₁₅ RASS values at T₃₀, T₄₅, T₆₀ minutes were statistically significantly lower in Group S than Group K and Group KM.

Conclusions: We concluded that use of ketamine alone or in combination with low dose of midazolam for short-time surgical procedures did not cause agitation compared to sevoflurane, increased

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quick recovery and child-parent satisfaction and efficiency of operating room.

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Introduction:-

Dental fear and anxiety complicates and sometimes even makes treatment impossible in children (1). It is required to use sedation or general anesthesia in patients for whom dental treatment cannot be performed under local anesthesia and behavioral and psycho-pedagogical techniques remain inadequate (2, 3). The sedation is a method that is often used for dental treatment of children for performing dental treatment in safe and comfortable conditions; reduces the time apart for the child-parent and the occupancy time of patient beds, and increases the efficiency of operating room. The sevoflurane, midazolam and ketamine are therefore commonly used anesthetics. The sevoflurane is the preferred inhalation agent because it has low solubility and low risk for hepatotoxicity, is less irritant, maintains hemodynamic stability, and results in quick recovery (4, 5). However, the sevoflurane is also reported to cause agitation more often in pediatric patients than other anesthetics during recovery from general anesthesia (5, 6). The ketamine is a non-competitive N-methyl-D-aspartate receptor antagonist that is commonly used anesthetics for outside anesthesia. Ketamine allows for sedation, analgesia and amnesia without suppressing the functions of respiratory tract and circulatory system (7). But, hallucinogenic properties of the ketamine is its unwanted effect. Midazolam is one of the benzodiazepine class sedative, and dose-dependent, anxiolytic, sedative-hypnotic and safely used with presence of specific antidote, working by increasing GABA activity in the central nervous system (8). However, low doses of midazolam is reported to cause agitation not sedation in some cases (9).

The EA was first defined in 1960 and is a problem that occurs within the first 30 minutes after surgery, often spontaneously improves within 15 minutes, includes disorientation, agitation, delusion, hallucination, discomfort, unstoppable crying, cognitive changes and impaired memory, and delays the discharge (9-13). It has been reported that selected anesthetics may play role in the incidence of EA in general anesthesia practice for medical reasons (9). Although there is not only one cause for EA, it is important to know the risk factors causing this condition in order to reduce the contributing factors and treat it (7). The factors playing role in occurrence of EA may be grouped in association with patient, surgery and anesthesia (9). The EA has been investigated in detail in the literature for the pediatric population and medical procedures performed under general anesthesia (e.g., strabismus surgery, adenotonsillectomy), but it has not been sufficiently studied for the dental treatment performed under deep sedation (9, 13, 14). Therefore, the objective of this study was to examine the characteristics of postoperative recovery for evaluating the incidence of EA and its relationship with drugs used for anesthesia in pediatric cases scheduled for tooth extraction under deep sedation (15).

Materials and Methods:-

With the approval of local ethics committee of Faculty of Dentistry, Ankara University, this study was performed with data of 120 cases, less than 12 years old, ASA I-II group according to 'American Society of Anesthesiologists (ASA)' risk classification, who were scheduled for tooth extraction under deep sedation because it was not possible to perform tooth extraction under local anesthesia. The informed consent form was read and signed by the children's parents.

The cases whose parents did not provide informed consent, who had drug allergy, mental-motor retardation, uncontrolled systemic disease and psychiatric complaint or diagnosis, and were on antiepileptic or sedative medication and above 12 years of age were excluded from the study.

Patient's age, body weight (kg), height (cm), ASA risk group and number of teeth extracted were recorded in the study protocol. The anesthesia protocol used for tooth extraction in our clinic was followed. After 6-hour fastening time, a topical anesthetic (EMLA®) was applied on the hand dorsal surface of all cases 30 minutes before transferring them to the operating room. All the cases were transferred to the operating room without premedication. Group K allowed to insert intravenous (IV) cannula and intravenously received 2-3 mg.kg⁻¹ of Ketamine (Ketalar®, Pfizer, 50 mg ml⁻¹, Istanbul, Turkey). Group KM (Ketamine-Midazolam, n=40) intravenously received 0.5 mg Midazolam (Dormicum®, Roche, 5 mg.ml⁻¹, Fontenay-sous-Bois, France) in addition to 2-3 mg.kg⁻¹ of Ketamine. Group S disallowed to insert intravenous cannula and received 8% Sevoflurane in the 50% O₂/N₂O (Sevoflurane®, Abbott, Italy) through a face mask (connected to a Mapleson C circuit). The aim was to achieve Richmond Agitation SedationScale(RASS, Table 1): -4 (deep sedation) in all of the groups. Research method was planned to obtain

(RASS: -4) sedation level. For that reason if we could not obtain RASS = -4 level, another doses of ketamine (0.05 mg kg⁻¹ iv ketamine in Group K, KS) or additional Sevoflurane (in Group S) inhalation was planned. The EA was assessed another anesthesia personnel who was blinded to which anesthetic agent was administrated to the patients. The cases were oxygenized through nasal cannula for 4 L/min during the procedure, and noninvasive Systolic Arterial Pressure (SAP), Diastolic Arterial Pressure (DAP), Heart Beat Rate (HR), and peripheral oxygen saturation (SPO₂) were monitored. When desired sedation level (RASS=-4) was obtained, dental surgeon performed infiltration anesthesia (Ultrakain DS[®], 1 mL/40 mg Articaine hydrochloride+0.006 mg epinephrine hydrochloride, Sanofi Aventis, Istanbul) on the teeth planned for extraction, and then dental procedure was performed. The anesthetic gasses were terminated in Group S when RASS was -4; spontaneous breathing was maintained and oxygenation through nasal cannula was performed during the procedure. Anesthetic agents were administered performed by Anesthesiologist to obtain RASS=-4 level. Doses of anesthetics, procedure time and unwanted side effects were recorded by same Anesthesiologist. At the end of the procedure, another anesthesia personnel (allied health staff) who were blinded groups were recorded RASS at minute 0 (T₀) in the operating room and the RASS at minutes 15., 30., 45. and 60. [respectively (T₁₅), (T₃₀), (T₄₅), (T₆₀)] were recorded in the wards with parents. The cases numbered for 2 months were grouped by the anesthetist according to anesthetics administered, and the data of first 40 cases in each group was included in the study.

Statistical Analysis:-

The data was analyzed using SPSS version 20.00. The descriptive analysis was performed to determine mean and standard deviation of data. ANOVA was used to analyze intergroup differences. Tukey test was performed to analyze differences between groups that had equal variances, and post hoc Dunett's test was performed when the variances were not equal between the groups. Normal distribution of data was assessed by Kolmogorov Smirnov test and the data had normal distribution ($p < 0.005$). The homogeneity test was performed to determine whether or not intergroup variances were homogeneous.

Results:-

The data of all cases (n=120) was included in the study. The groups were created according to anesthetics administrated in accordance with our clinical protocols (Group K: Ketamine, n=40, Group KM: Ketamine + Midazolam, n=40, Group S: Sevoflurane, n=40).

No statistically significantly differences were found in comparison of groups for height, body weight, procedure time, and number of teeth extracted. In comparison of groups for the age of cases, the age of cases in Group KM was statistically significantly higher than that of cases in Group K ($p < 0.05$). No significant differences were found between the Group KM and the Group S (Table 2).

In comparison of study groups for RASS, statistically significantly differences were found between the groups ($p < 0.001$). The RASS values at minute 0 were statistically significantly higher in Group KM than that of other groups. While no statistically significantly differences were found between the groups for mean RASS at 15, the RASS values at minute 30., 45. and 60. were statistically significantly lower in Group S (Table 3).

Comparison of groups by number of cases in RASS score groups (Table 4, 5, 6, 7, 8 and Graphic 1)

Discussion:-

In the study groups, the targeted sedation level [RASS=(-4) score (deep sedation)] was achieved with anesthetics administered and the tooth extraction was successful in all of the cases.

RASS was used by *Kerson et al.* (15) to assess consciousness and awareness in the pediatric population, to titrate sedative agents, and to evaluate childhood delirium, and is one of the validity and safety proven scales that is intuitive, easy to use, includes agitation and sedation scales together, and has the highest adaptation among practitioners (16). The score "0" represents ideal level on the scale, and the values up to "+4" represent increasing agitation, and values up to "-5" represent increasing sedation level (17).

According to the memorandum published by ASA in 1999 and revised in 2004, the *deepsedation* is a sedation manifestation during which consciousness is depressed by or totally disappears by drugs, defense reflexes are partially maintained, patients cannot be easily aroused and occasionally respond to physical and verbal stimulus,

interventions may be required to maintain the airway patency and spontaneous ventilation but no cardiovascular functions are affected (18).

Our clinic practice aims at RASS:-4 (deep sedation) to achieve a proper mouth opening during dental treatment and successfully perform the procedure. We obtained RASS:-4 level in all of the cases, started the procedure and extracted the teeth without any undesirable effects. Upon completion of the procedure, we assessed the emergence agitation with RASS.

The factors establishing the incidence and severity of EA are multifactorial (7). However, one of the factors contributing to the development of EA is *anesthesia time*. No statistically significant differences were found between the groups for the procedure time and number of teeth extracted (7,9, 12, 14).

Children with EA are unaware of the things around them and cannot be calmed by parents and other caretakers (19). Literature reports that EA mostly occurs between the ages of 2 and 5, but preschoolers are at risk (7, 9, 20, 21). In comparison of study groups by mean age, the age of cases in Group KM was statistically significantly higher than that of cases in Group K ($p < 0.05$), but not different from the Group S (Table 2). After the procedure was completed, 12.5% of cases in Group S were agitated, 22.5% of cases were restless at minute 0 (Table 4). None of the cases in two other groups were agitated from the completion of procedure and to the discharge. In our opinion, this may be due to anesthetics selected as well as the age characteristics of children.

One factor responsible for EA is personality traits of child and stressed induction of anesthesia (11). The children in all groups were the cases for who tooth extraction was attempted under local anesthesia but could not be performed due to dental anxiety and fear. Therefore, the fear and anxiety level of all cases in the study was higher than those who were treated under local anesthesia. Kain *et al.* (22) concluded that there was a relationship between the preoperative anxiety and the postoperative behavior disorder. The cases in Group KM and Group K allowed intravenous cannulation but not the cases in Group S, and the number of agitated and restless cases in Group S was greater. We therefore judge that preoperative anxiety level may have an effect on the development of EA.

Another responsible factor among other factors causing EA is the presence of pain (9, 21). However, the surgeon performing the procedure administered infiltration anesthesia to all the cases during tooth extraction, and none of the cases complained about the pain for 60 minutes after the procedure.

Another factor that is associated with EA is a noisy environment distant from family (9, 12, 20, 21). During the study, deep sedation was administered in a quiet and peaceful environment and all the cases were exposed to the same conditions.

The other factors that increase the incidence of EA include quick recovery of patients and possibility of airway obstruction. All of the cases included in this study were in the ASA I risk group and none of them had systemic diseases that might cause airway obstruction. We oxygenated our cases in 4L/min through nasal cannula during the procedure and did not have airway obstruction that might disturb the oxygenation or similar unwanted side effects. When we examine the groups for quick recovery of consciousness in an environment without family, we consider that anxiety caused by preoperative dental anxiety and fear was quickly remembered because midazolam had amnesic effects that was used in standard dose in addition to Ketamine in Group KM, Ketamine used in Group K had amnesic effects, and had no such effect in Group S; therefore the number of agitated and restless cases in Group S was statistically significantly higher than that of other groups.

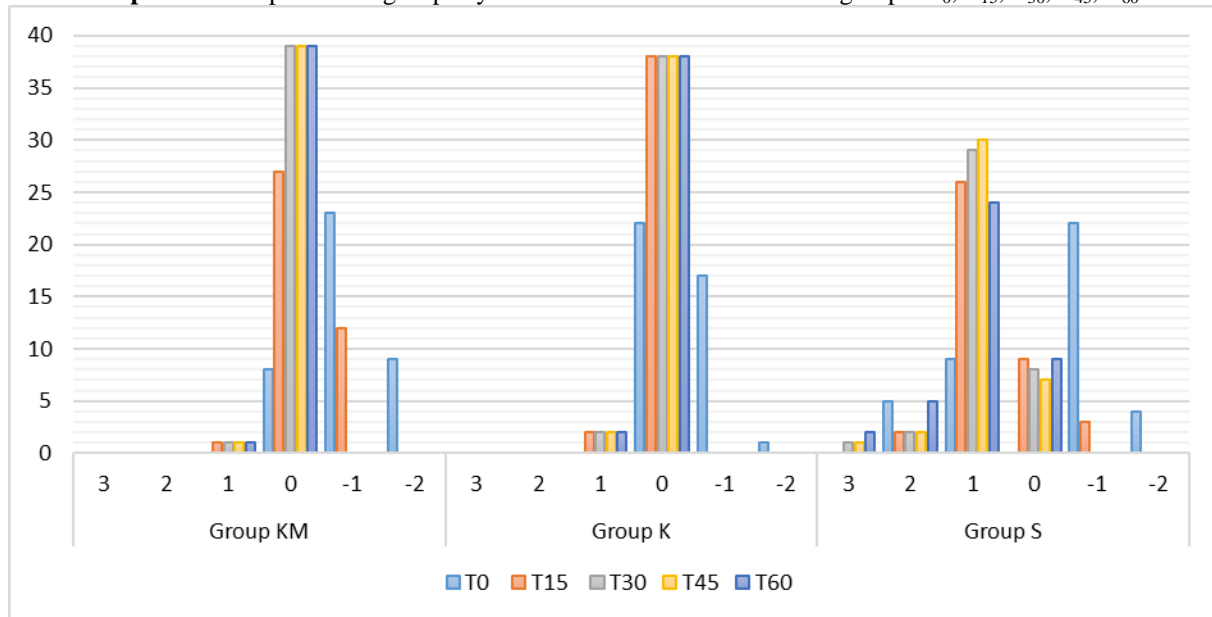
The factor that is mostly responsible for EA is anesthetics. The pediatric anesthesiologists underline that they observe higher agitation with Sevoflurane and associate this with low blood/tissue solubility and its EEG (electroencephalography) changes (6, 23). Işık *et al.* suggested that EA was also observed for administration of sevoflurane for MRI where no surgical intervention was performed (24). Cravero *et al.* (25) compared the sevoflurane and halothane anesthesia in children who underwent magnetic resonance imaging, and reported that no agitation was observed in cases received halothane and that 33% of cases received sevoflurane had agitation. The authors concluded that quicker recovery with sevoflurane and sudden awakening of child in a foreign environment resulted in agitation. The studies comparing propofol and sevoflurane reported that recovery was quicker with propofol and the agitation was higher in the sevoflurane group (26, 27). In the present study, 55% of cases in Group K, 20% of cases in Group KM and 0 case in Group S were alert and calm following completion of procedure. The

recovery was quicker in Group KM and Group K than that of Group S, and the number of agitated and aggressive cases was higher in this group as compared to two other groups. Therefore, we consider that recovery time may have an effect on the EA.

There are contradictory data for premedication with midazolam: a number of studies suggest that it is effective in reducing preoperative anxiety and EA in children received anesthesia with sevoflurane (28). Despite this, *Cole et al.* pointed out that EA was higher after isoflurane and halothane anesthesia in children received midazolam for premedication (30%-40%)(19). They explained that this might be due to a gap caused by midazolam in the memory of children, sudden awakening in an environment that was totally strange to children, and an intense sense of fear accompanied by irritating stimulus. *Shrestha S et al.* (29) compared placebo and midazolam by orally administering (0.5 mg/kg) paracetamol, and reported that 96.7% of children in midazolam group and 53.3% of children in placebo group were calm when leaving their parents. The authors also reported that induction of anesthesia was calm in 73.3% of children in midazolam group and 33.3% of children in placebo group, and no differences were found between two groups for recovery. *Lapin et al.* (28) and *Ko et al.* (30) reported that premedication with midazolam reduced the EA; *McGraw et al.* (31) reported that it had no effects on the EA, moreover children developed postoperative unfavorable behavioral changes. We administered 0.5 mg/kg standard dose of midazolam to all of the cases in Group KM in addition to ketamine. We found that less cases were alert and calm and more cases were drowsy and sedated at minute 0 according to RASS at the end of procedure compared to the group received Ketamine alone (Group K). We judge that standard dose of midazolam added to ketamine deepened the anesthesia, slightly prolonged the recovery time, however, we did not have any agitated cases in these two groups for 60 minutes following completion of procedure.

Literature has no studies that evaluate the effect of ketamine on the postoperative agitation in children who underwent dental treatment. On the other hand, there are studies reporting that premedication with ketamine reduced postoperative agitation compared to placebo (7, 32). *Hadi et al.* (7) intraoperatively administered bolus low dose of ketamine to cases undergoing adenotonsillectomy with sevoflurane, and reported reduced incidence and severity of agitation developed with sevoflurane. *Chen et al.* (33) reported that dexmedetomidine and ketamine reduced EA and pain in strabismus surgery. We did not have agitated cases in the groups (Groups KM and K) received ketamine neither.

In comparison of study groups for postoperative RASS values, the cases in Group KM were statistically significantly drowsy at minute 0 compared to the other groups, and we associated this deepened anesthesia with midazolam. In regard to RASS values at minute 30, 45 and 60, we observed that cases in Group S were statistically significantly restless and agitated than other cases. We consider that sevoflurane delays recovery in short-time surgical procedures, which is in consistent with literature, and results in agitation because it has no amnesic effects. Finally, we used anesthetics (Ketamine, Ketamine-Midazolam and Sevoflurane) in cases of tooth extraction to achieve deep sedation, which are not compared alone in the literature for incidence of recovery and agitation. We achieved quicker recovery with ketamine alone (Group K) and did not have any agitated cases. We concluded that use of ketamine alone or in combination with low dose of midazolam for short-time surgical procedures did not lead to agitation compared to sevoflurane, increased quick recovery and child-parent satisfaction and efficiency of operating room, and that preoperative anxiety or agitation level may be an indicator for development of postoperative behavior disorder. However, the limitations of this study include selection of anesthetics according to characteristics of cases and lack of randomization. Secondly, age distribution was not comparable in the groups. Therefore, we consider that postoperative, randomized and controlled studies on similar age groups are needed.

Graphic 1:-Comparison of groups by number of cases in RASS score groups T₀, T₁₅, T₃₀, T₄₅, T₆₀**Table 1 :-**Richmond Agitation SedationScale (RASS)

Numbers correspondings RASS scoring	Scoring	Clinical status
1	+4	Combative
2	+3	Very agitated, aggressive
3	+2	Agitated (frequent non-purposeful movement)
4	+1	Restless
5	0	Alert and calm
6	-1	Drowsy
7	-2	Light sedation
8	-3	Moderate sedation
9	-4	Deep sedation
10	-5	Unarousable

Table 2:-Demographic characteristics by groups (mean± SD)

	Group KM (n=40)	Group K (n=40)	Group S (n=40)	F	P
Age (year)	6.15 ±1.95*	5.10 ±1.33	5.37 ±1.62	4.30	0.016*
Height (cm)	107.30±18.87	100.00±13.36	107.17±15.48	2.70	0.071
Body weight (kg)	21.83±5.16	20.00±4.89	22.78±6.70	2.49	0.087
Procedure time (minute)	7.87 ±2.77	7.05±2.56	6.75±2.25	2.10	0.126
Number of teeth extracted	2.85 ± 1.99	2.40±1.62	2.67±1.83	0.61	0.541

Table 3:-Groups as in terms numbers of the correspondings RASS scoring (mean± SD)

RASS/Time	Group KM	Group K	Group S	F	P
T ₀	6.02±0.65	5.47±0.55	5.27±1.28	7.596	0.001
T ₁₅	5.27±0.50	4.95±0.22	4.32±0.69	35.596	0.000
T ₃₀	4.97±0.15	4.95±0.22	4.10±0.59	70.484	0.000
T ₄₅	4.97±0.15	4.95±0.20	4.07±0.57	78.561	0.000
T ₆₀	4.97±0.15	4.95±0.22	3.95±0.93	43.507	0.000

Table 4:-Number of cases according to RASS values at T₀ n (%)

Groups	Agitated (+2)	Restless (+1)	Alert and calm (0)	Drowsy (-1)	Light sedation (-2)	Total
Group KM	0	0	8 (20)	23 (57.5)	9 (22.5)	40
Group K	0	0	22 (55)	17 (42.5)	1 (2.5)	40
Group S	5 (12.5)	9 (22.5)	0	22 (55)	4 (10)	40
Total	5	9	30	62	14	120

Table 5:-Number of cases according to RASS values at T₁₅ n(%)

Groups	Agitated (+2)	Restless (+1)	Alert and calm (0)	Drowsy	Total
Group KM	0	1 (2.5)	27 (67.5)	12 (30)	40
Group K	0	2 (5)	38 (95)	0	40
Group S	2 (5)	26 (65)	9 (22.5)	3 (7.5)	40
Total	2	29	74	15	120

Table 6:-Number of cases according to RASS values at T₃₀ n (%)

Groups	Very agitated, aggressive (+3)	Agitated (+2)	Restless (+1)	Alert and Calm (0)	Total
Group KM	0	0	1 (2.5)	39 (97.5)	40
Group K	0	0	2 (5)	38 (95)	40
Group S	1 (2.5)	2(5)	29 (72.5)	8 (20)	40
Total	1	2	32	85	120

Table 7:-Number of cases according to RASS values at T₄₅ n(%)

Anesthetic agent	Very agitated, aggressive(+3)	Agitated (+2)	Restless (+1)	Alert and Calm (0)	Total
Group KM	0	0	1 (2.5)	39 (97.5)	40
Group K	0	0	2 (5)	38 (95)	40
Group S	1 (2.5)	2 (5)	30 (75)	7 (17.5)	40
Total	1	2	33	84	120

Table 8:-Number of cases according to RASS values at T₆₀ n(%)

Groups	Very agitated, aggressive (+3)	Agitated (+2)	Restless (+1)	Alert and Calm (0)	Total
Group KM	0	0	1 (2.5)	39 (97.5)	40
Group K	0	0	2 (5)	38 (95)	40
Group S	2 (5)	5 (5)	24 (60)	9 (22.5)	40
Total	2	5	27	86	120

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RESEARCH ARTICLE

KINETIC AND MECHANISTIC STUDY OF PALLADIUM (II) CATALYZED OXIDATION, DEAMINATION AND DECARBOXYLATION OF GLYCINE BY ALKALINE PERMANGANATE.

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Abstract

Kinetics of oxidation of glycine by permanganate in presence of Palladium(II) catalyst was studied spectrophotometrically. It was found that the reaction exhibits first order dependence on [catalyst] & [oxidant] and less than unit order in [glycine] and [alkali]. The data suggests that oxidation proceeds via formation of a complex between the substrate and the catalyst, which then reacts with one mole of permanganate in a slow step to form a glycine free radical followed by a fast step to yield the products. The activation parameters of the rate determining step were computed and are discussed. The reaction constants involved in the mechanism were calculated and there is good agreement between observed and calculated rate constants under different experimental conditions.

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Introduction:-

The kinetic investigations on the oxidation of amino acids are important because of their biological significance and a precise understanding of the mechanism of such biological redox reactions helps in the synthesis of reaction products. Amino acids act as building blocks in protein synthesis and also play a significant role in metabolism. Presently large numbers of oxidation studies on α -amino acids are being carried out using various oxidants. Glycine, is a non-essential, simplest amino acid and mild reductant, requires drastic conditions or powerful oxidants for studying its kinetic behavior. The oxidation of glycine has been studied from mechanistic point of view with potassium permanganate in aqueous phosphate buffers by Joaquin et al [1], where the reaction is auto catalyzed by one of the products, colloidal manganese dioxide. The uncatalyzed and the surfactant catalyzed oxidation of glycine by acidic permanganate [2] have been studied in which the reaction followed first order dependence in both glycine and permanganate. Amino acids have been oxidized by number of reagents [3]. The oxidations of amino acids are of interest as they produce different oxidation products for different oxidants [4, 5]

Permanganate is one such powerful multi-electron oxidant which can exist in various oxidation states, among which +7 is its highest oxidation state, which occurs in the oxocompounds like MnO_4^- , Mn_2O_7 , MnO_3F . Out of which MnO_4^- is the most commonly used well known oxidant species to carry out kinetic studies in acidic, neutral and alkaline media. Oxidation by permanganate ion is used in organic synthesis especially in phase transfer catalysis [6-12].

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Palladium (II) is known to catalyze various reactions [10]. Most studies using Palladium (II) as catalyst have been used in the form of Palladium (II) chloride [11] and the nature of its active form in such reaction remains obscure. Hence the effect of chloride on the reactions was studied in order to study the active species of palladium in chloride medium. The uncatalysed reaction of glycine and permanganate has been reported earlier [13]. Here we have described the results of the title reaction in order to know the active species of oxidant, reductant and catalyst in such media and to interpret the possible mechanism.

Results and Discussion:-

Reaction Order:-

As the oxidation of glycine by permanganate in alkaline medium proceeded with a measurable rate in the absence of Pd(II) catalyst, the catalysed reaction is understood to occur in parallel paths. The reaction orders were determined from the slopes of $\log k_c$ versus (concentration) plots by varying the concentrations of oxidant, reductant, alkali and catalyst, while keeping other parameters constant.

Effect of oxidant, Substrate and Catalyst:-

The oxidant permanganate was varied in the range of $0.5-5.0 \times 10^{-4} \text{ mol dm}^{-3}$ and the linearity of the plot of $\log [\text{MnO}_4^-]$ versus Time indicates the order in $[\text{MnO}_4^-]$ is unity. This was also confirmed by varying $[\text{MnO}_4^-]$, which did not show any change in pseudo-first order rate constants k_c (Table 1). The dependence of rate on substrate [glycine] was studied by varying the concentration of glycine in the range of $1.0 - 6.0 \times 10^{-3} \text{ mol dm}^{-3}$ keeping the concentrations of all the other reactants constant at three different temperatures 298, 303 and 308K. The rate increased with increase in [glycine] (Table-1) and the order with respect to [glycine] was found to be fractional. Further, the plots of $[\text{Pd(II)}] / k'$ vs $1/[\text{glycine}]$ (Fig.1) are linear with positive intercepts on rate axis indicating that the reaction obeys Michelis-Menton behavior. The concentration of Palladium(II) catalyst effect was studied by varying the concentration in the range of $(0.2 \text{ to } 2.0) \times 10^{-6} \text{ mol dm}^{-3}$ and it was found that rate constants increased with increase $[\text{Pd(II)}]$. The order in $[\text{Pd(II)}]$ was found to be unity.

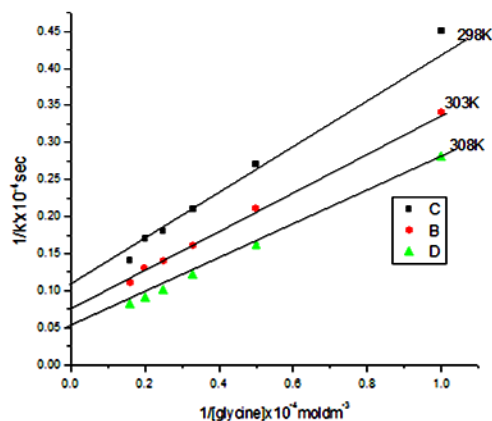


Fig 1:- Effect of concentration of glycine on Palladium (II) catalyzed oxidation of glycine by permanganate in alkaline medium

Effect of alkali:-

The effect of alkali on the rate of the reaction was studied at constant concentrations of glycine, permanganate, Pd(II) and ionic strength at 30°C . The rate constants obtained were found to increase with increase in [alkali] (Table 1) and the order with respect to alkali is found to be fractional order.

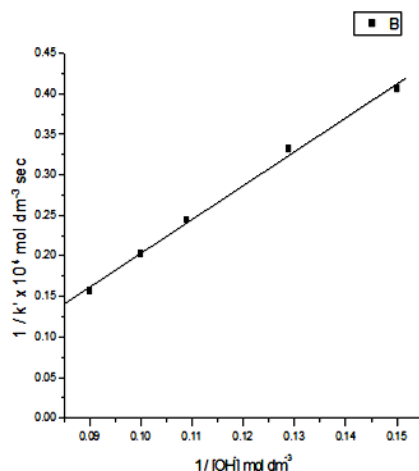


Fig 2:- Effect of $[OH^-]$ on the pseudo first order rate constant, k' on Palladium (II) catalyzed oxidation of glycine by permanganate in alkaline medium.

Effect of ionic strength:-

The effect of ionic strength on the reaction rate was studied by varying the concentration of sodium perchlorate in the reaction mixture in the range of 0.2- 0.7 mol dm^{-3} at constant [glycine], [permanganate], [Pd(II)] and [alkali]. The rate constants obtained were found to be constant (Table 1) which shows that the ionic strength has negligible effect on the rate of the reaction.

Effect of the product:-

The effect of the product, manganese(VI) on the reaction rate, was studied by carrying out kinetic runs keeping the concentrations of all other reactants constant and varying the concentration of manganese(VI) from $(0.25-2.5) \times 10^{-4}$. The pseudo-first order rate constants thus obtained are found to remain practically constant indicating that manganese (VI), one of the products does not have any significant effect on the rate of the reaction (Table 1).

Table 1:- Effect of [Glycine], $[MnO_4^-]$, $[OH^-]$ and Pd(II) catalyzed oxidation of Glycine by permanganate in aqueous alkaline medium at 25°C ($I = 0.5 \text{ mol dm}^{-3}$).

$[Gly] \times 10^3$ mol dm^{-3}	$[MnO_4^-] \times 10^4$ mol dm^{-3}	$[OH^-]$ mol dm^{-3}	$[Pd(II) \times 10^5]$ mol dm^{-3}	k_{obs} $10^4 (\text{s}^{-1})$	$k_{cat} 10^4 (\text{s}^{-1})$
0.5	2.0	3.0	1.0	2.22	2.27
1.0	2.0	3.0	1.0	2.85	2.83
2.0	2.0	3.0	1.0	3.57	3.62
3.0	2.0	3.0	1.0	4.76	4.80
5.0	2.0	3.0	1.0	5.55	5.6
2.0	0.5	3.0	1.0	6.24	6.22
2.0	1.0	3.0	1.0	6.34	6.21
2.0	2.0	3.0	1.0	6.22	6.20
2.0	3.0	3.0	1.0	6.15	6.23
2.0	5.0	3.0	1.0	6.28	6.25
2.0	2.0	1.0	1.0	5.23	5.15
2.0	2.0	2.0	1.0	6.45	6.38
2.0	2.0	3.0	1.0	7.64	7.52
2.0	2.0	4.0	1.0	8.67	8.51
2.0	2.0	5.0	1.0	9.89	9.58
2.0	2.0	3.0	0.2	3.26	3.21
2.0	2.0	3.0	0.6	6.6	6.18
2.0	2.0	3.0	1.0	6.87	6.79
2.0	2.0	3.0	1.5	7.57	7.58
2.0	2.0	3.0	2.0	8.32	8.21

Test for free radical:-

The free radical test has been carried out by taking glycine, sodium hydroxide in a thumborg tube and acrylonitrile and permanganate in the bent tube. After evacuating the system the solutions were mixed by tilting the tube. The reaction mixture was kept aside and after 24 hours a soft precipitate was observed which indicates the formation of free radical.

Effect of Temperature:-

The influence of temperature on the rate of reaction was studied for the reaction at 25, 30, 35 and 40°C. With varying $[\text{OH}^-]$ by keeping other conditions constant. The rate was found to be increased with increase in temperature. The rate constants, k , for the slow step of Scheme 1 were obtained from the slopes and the intercept of the plots $[\text{Pd(II)}]/k$ versus $1/[\text{OH}^-]$ and were used to calculate activation parameters. The values of k ($\text{dm}^3 \text{mol}^{-1} \text{s}^{-1}$) were 5.2×10^4 , 6.2×10^4 , 7.5×10^4 and 8.32×10^4 at 25, 30, 35 and 40°C respectively. The activation parameters for the rate determining step were obtained by the least square method of plot of $\log k$ versus $1/T$ are given in Table 3 and are good agreement with uncatalysed reaction.

Table 2:- Thermodynamic activation parameters for the Pd(II) catalysed oxidation of glycine by alkaline permanganate at 25°C ($I = 0.20 \text{ mol dm}^{-3}$).

Parameters	Values	
	Uncatalyzed	Catalyzed
E_a (kJmol^{-1})	28.49 ± 0.02	26.62 ± 0.5
ΔH (kJmol^{-1})	25.93 ± 0.05	24.14 ± 0.04
ΔG (kJmol^{-1})	117.18 ± 0.4	109.22 ± 0.23
ΔS (kJmol^{-1})	-306.23 ± 1.62	-285.45 ± 1.45

Discussion:-

Glycine, an optically inactive non-essential amino acid exists in the form of zwitter ion(gly) $\text{H}_3\text{N}-\text{CH}_2-\text{COO}^-$ in neutral medium, in the form of the cationic species $\text{H}_3\text{N}-\text{CH}_2-\text{COOH}$ in acid medium and in the form of anionic species (gly-) $\text{H}_2\text{N}-\text{CH}_2-\text{COO}^-$ in alkaline medium. pH metric method using computer augmented programs SCPHD and MINQUAD75[18] has been used to determine the pKa values of glycine for carboxylic acid and amino groups. Glycine exists predominantly in the anionic form, $\text{H}_2\text{N}-\text{CH}_2-\text{COO}^-$ (gly-), under the experimental conditions employed, in presence of alkali.

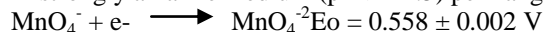
It is interesting to note that the probable species of Pd(II) chloride in alkaline medium. It is noted that the reaction is not influenced by chloride ions, which is usually expected in Pd(II) catalysed reactions[19]. In alkaline solutions Pd(II) is mostly presented as hydroxilated species, $[\text{Pd(OH)}(\text{Cl})]$, represented by $\text{Pd(II)}^\#$ in **scheme 1**, which is in accordance with earlier work[20] and not as its chloride complex as in the other works[19].

Permanganate acts as powerful oxidant in acid as well as alkaline media. Its oxidation reactions are very interesting because of the several oxidation states to which it can be reduced. The stable reduction product of manganese is largely determined by the pH of the medium.

In solutions of low alkali and in neutral media, manganese (VII) is reduced to manganese (IV) [21, 22].



In strongly alkaline medium ($\text{pH} > 12.5$) permanganate is usually reduced to manganese (VI).

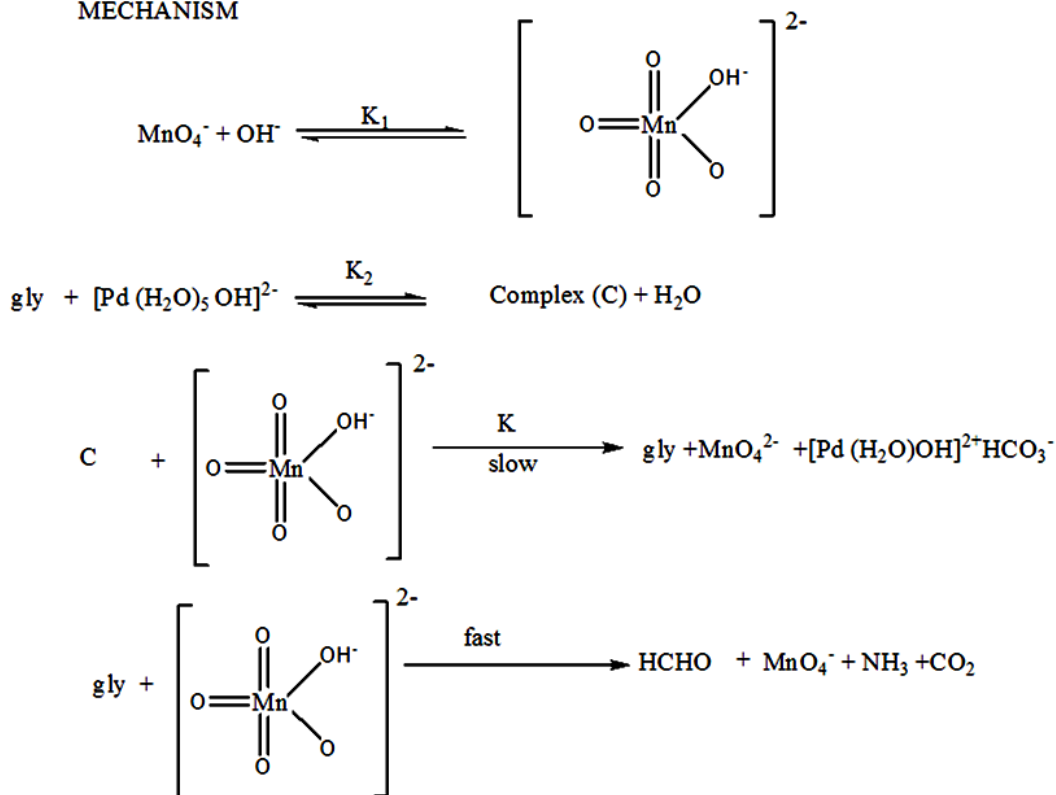


Most of the oxidations by permanganate in neutral and alkaline media proceed through intermediate complex formation between oxidant and substrate [23, 24]. In alkaline medium, MnO_4^- is the only active species of permanganate known to exist. Though oxidations by permanganate involve multi electron changes in neutral and acidic media but in strong alkaline media ($\text{pH} > 12.5$) they involve only one electron change, forming manganese (VI). In the present investigation in the presence of pd(II) catalyst the stoichiometry of the reaction indicates that permanganate is reduced to manganese(VI) state only and no evidence is obtained for further reduction of manganese(VI) by glycine to manganese(IV).

The results suggest that first alkali combines with permanganate to form an alkali permanganate species in an equilibrium step which is also supported by the fractional order of $[\text{OH}^-]$ and reciprocal of Michalis-Menton plot,

which is linear with positive slope (Figure.2). Glycine reacts with Pd(II) species to form complex. The complex further reacts with alkali permanganate in a slow step to form glycine free radical, which further reacts with another permanganate species in a fast step to form the products. Based on these observations the following mechanism has been proposed.

MECHANISM



SCHEME-1

The spectral evidence for such complex is obtained from the UV-Visible spectra of pd(II) and mixture of Pd(II) and glycine. A bathochromic shift λ_{max} 4 nm from 220-224 nm and hyperchromicity at λ_{max} 224-380 was observed. Thus the above scheme leads to rate law given as

$$\text{Rate} = -d[\text{MnO}_4^-] / dt = K K_1 K_2$$

$$\begin{aligned}
 \text{Rate} = -d[\text{MnO}_4^-] / dt &= K K_1 K_2 [\text{gly}] [\text{MnO}_4^-] [\text{Pd(II)}] [\text{OH}^-] \\
 &= \frac{K K_1 K_2 [\text{gly}] [\text{MnO}_4^-] [\text{Pd(II)}] [\text{OH}^-]}{(1 + K_1 [\text{OH}^-]) (1 + K_2 [\text{gly}]) (1 + K_1 [\text{MnO}_4^-]) (1 + K_2 [\text{Pd(II)}])} \quad \text{----- (2)}
 \end{aligned}$$

$$\text{Rate} = \frac{d[\text{MnO}_4^-]}{dt} = \frac{KK_1K_2 [\text{gly}] [\text{MnO}_4^-][\text{OH}^-] [\text{Pd(II)}]}{1 + K_1[\text{OH}^-] + K_2 [\text{gly}]}$$

$$\frac{\text{Rate}}{[\text{Pd(II)}]} = K_{\text{obs}} = \frac{KK_1K_2[\text{gly}][\text{OH}^-]}{1 + K_1 [\text{OH}^-] + K_2 [\text{gly}] + K_1K_2[\text{gly}][\text{OH}^-]} \quad \text{-----} \quad (3)$$

$$\frac{[\text{Pd(II)}]}{K_{\text{obs}}} = \frac{1}{KK_1K_2[\text{gly}][\text{OH}^-]} + \frac{1}{KK_2[\text{gly}]} + \frac{1}{KK_1[\text{OH}^-]} + \frac{1}{K} \quad \text{-----} \quad (4)$$

Rate Law

Further equation (4) predicts the plots of $[\text{Pd(II)}]/k$ versus $1/[\text{glycine}]$ and $[\text{Pd(II)}]/k$ versus $1/[\text{OH}^-]$ (Fig.1,2) is a linear with a positive intercept supports the Pd(II) - Glycine complex, which is reported earlier [25]. Experimentally observed plots are the one similar predicted from the rate equation thus supporting the proposed mechanism. Also the rate constants of the rate determining step, k , are determined from the intercepts of the plots of $[\text{Pd(II)}]/k$ versus $1/[\text{glycine}]$ at three different temperatures (25, 30 & 35°C) and the activation parameters E_a and ΔS^\ddagger are computed using linear least square method and were found to $24.36 \pm 0.03 \text{ KJ}^{-1} \text{ mol}^{-1}$ and $-266.13 \pm 2.62 \text{ JK}^{-1} \text{ mol}^{-1}$ respectively. A high negative value of entropy indicates that the transition state is rigid in nature. This type of radical intermediate has been observed in earlier work [26] on alkaline –permanganate oxidation of amino acids.

Conclusion:-

The observations in the present study reveal that anionic form of glycine, $\text{H}_2\text{N}-\text{CH}_2-\text{COO}^-$ (gly-) and MnO_4^- are the reactive species in alkaline medium and formaldehyde is the product of oxidation. It is interesting to note that the oxidant species $[\text{MnO}_4^-]$ requires a $\text{pH} > 12$ otherwise the system becomes disturbed and the reaction proceeds further to give reduced product of the oxidant as manganese (IV). Pd (II) catalyst is known to catalyze various reactions, especially in alkaline medium. Which catalysis the reaction in a measurable velocity at a concentration of $1.0 \times 10^{-6} \text{ Mol dm}^{-3}$. The active species of the catalyst was found to be PdCl.

Experimental:-

Materials and Methods:-

A stock solution of glycine (E.Merck) was prepared afresh by dissolving in required volume of water and its strength is determined by the acetuous-perchloric acid method [14].

A stock solution of permanganate is prepared by dissolving potassium permanganate in double distilled water. The solution is boiled gently for 15-30 minutes, cooled and filtered through a sintered glass crucible. It is standardized by titrating with sodium oxalate [15].

A stock solution of manganese(VI) is prepared by heating potassium permanganate in 8.0 mol dm^{-3} sodium hydroxide until a clear green color is produced and is standardized spectrophotometrically [16] at 610 nm using a molar absorbance value of $1530 \text{ dm}^3 \text{ mol}^{-1} \text{ cm}^{-1}$. All the chemicals used in this investigation were of analytical grade. Double distilled water is used throughout the investigation. NaOH and NaClO_4 were used to provide required alkalinity and ionic strength respectively.

A palladium(II) stock solution was prepared by dissolving a known weight of palladium(II) chloride (S.D. Fine Chem.) in $0.20 \text{ mol dm}^{-3} \text{HCl}$ and standardized against EDTA [17]. For some kinetic runs in the absence of chloride, the chloride ion in the palladium(II) stock solution was precipitated with AgNO_3 and removed by repeated centrifugation. The resulting clear palladium(II) solution contained less than $5.0 \times 10^{-5} \text{ mol dm}^{-3}$ of Cl^- and Ag^+ . Such extremely low concentrations of Cl^- and Ag^+ were found to have no significant effect on the reaction. The chloride concentration was maintained between 2.0×10^{-5} and $4.0 \times 10^{-5} \text{ mol dm}^{-3}$ with KCl.

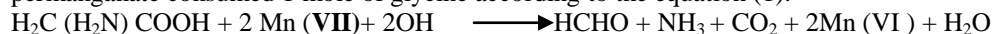
Kinetic measurements:-

Kinetic measurements were carried out at $30 \pm 0.1^\circ\text{C}$ in alkaline medium under the conditions [glycine] is tenfold excess over permanganate ion at constant ionic strength. The reaction was initiated by mixing the thermostated solutions of MnO_4^- , palladium(II) and glycine, which also contained the required amount of NaOH and NaClO_4 to maintained requisite alkalinity and ionic strength respectively. The progress of the reaction was followed by measuring the absorbance of permanganate at 526 nm using Milton Roy 1201 UV-VIS Spectrophotometer with 1cm path length glass cells. As no other species except manganese(VII) has any significant absorbance at this wave length under the conditions employed, the absorbance of the solution is taken as the residual concentration of manganese(VII) at time 't'. The plots of log (absorbance) versus time are found to be linear up to 85% completion of the reaction and the rate constants were reproducible within $\pm 5\%$.

The effect of carbonate on the rate of reaction was studied and it is found that added carbonate has no effect on the rate of reaction. The effect of dissolved oxygen on the rate was also studied in the presence of N_2 . No significant result was observed in the presence of N_2 and air. However fresh solutions were used while conducting the experiments.

Stoichiometry and product Analysis:-

In order to identify the products formed, different sets of reaction mixtures containing different reactant concentrations at constant ionic strength and alkali were kept to react at 30°C . After 24 hours the residual permanganate in each case was determined spectrophotometrically at 526nm. The results showed that 2 mole of permanganate consumed 1 mole of glycine according to the equation (1).



The main product formaldehyde is identified by chromotropic acid test [6] in which, a drop of the reaction mixture is mixed with 2ml of sulphuric acid (72% solution) in a test tube and a little of solid chromotropic acid is added. The test tube is heated for 10 minutes in a water bath at 30°C . A bright violet colour appeared in the test tube confirming the presence of formaldehyde as the main product along with ammonia and carbon dioxide, which are identified by Nessler's reagent and lime water respectively and manganate by its visible spectrum. The main product aldehydic was also confirmed by its IR spectrum, which shows a band at $2845\text{--}2019\text{ cm}^{-1}$ due to aldehydic stretching and was found that it does not undergo further oxidation. The concentration of catalyst, Pd(II), remained unchanged at the end of reaction, as found by estimating the palladium as azide complex [17] at 315 nm ($\epsilon = 12000\text{ dm}^3\text{ mol}^{-1}\text{ cm}^{-1} \pm 10\%$).

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RESEARCH ARTICLE

STEM CELL REGENERATIVE THERAPY IN ORAL AND MAXILLOFACIAL REGION: A SYSTEMATIC REVIEW.

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stem cells; regeneration ; embryonic stem cells; dental pulp; stem cell therapy; regenerative dentistry; oral mucosal lesions; cancer management.

Abstract

In the modern era, where biology and biotechnology have replaced the chemistry, we are exploring “biological solutions to biological problems.” The breakthrough of stem cells has produced new possibilities in the regeneration of different organs and tissues. The scope of application of stem cells in dentistry is vast and includes continued root formation, in pulp/dentin tissue regeneration, in replantation and transplantation, and reconstruction of the periodontium, in cancer management and research. In few years plenty of studies have been conceded which showed that stem cells and tissue engineering are giving rise to a separate branch termed “Regenerative Dentistry” that will have its own position in future dental practice. This article provides an insight into the various aspects of stem cells therapies in oral mucosal lesions and its future perspectives.

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Abbreviations:-

BMSC'S –Bone marrow stem cells; MSC'S- Mesenchymal stem cells; ADSC's- Adipose derived stem cells ; DPSC's- Dental pulp stem cells ; SHED- Stem Cells from Exfoliated Deciduous teeth; PDLSC's- Periodontal ligament stem cells; SCAP- Stem cells from Apical papilla; DFPC's- Dental Follicle Precursor Cells; ESC's- Embryonic stem cells; IPSC's- Induced Pluripotent Stem Cells; CSC's- Cancer Stem Cells; PLGA- Poly Lactic co glycolic acid; PGA- Poly Glycolic Acid

Introduction:-

Stem cells are the precursors of the body tissue which are undifferentiated biological cells that can differentiate into specialized cells and can divide (through mitosis) to produce extra stem cells.^(1,2) They are also defined as immature or undifferentiated cells that are capable of generating daughter cells identical to themselves or of differentiating into diverse cellular phenotypes. These cells are originated in multicellular organisms^(3,4). Human dental tissues have a limited potential to regenerate but the discovery of dental stem cells have developed new and surprising scenario in regenerative dentistry.

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History Of Stem Cells:-

Stem cells also known as “progenitor or precursor” cells and are clonogenic cells capable of both self-renewal and multi-lineage differentiation. In 1868 the term “stem cell” for the first time appeared in the works of German biologist Haeckel^(4,5). Wilson coined the term stem cell⁽⁵⁾. In 1908 Russian Histologist Alexander Maksimov postulated existence of hematopoietic stem cells at congress of hematologic society in Berlin⁽⁶⁾. Then the term “stem cell” was proposed for scientific use.

Stem Cell Properties:-

A classic stem cell should possess two properties namely *self renewal* and *potency*^{7,8,9}.

- **Self-renewal** is the capacity of the cell to undergo numerous cycles of cell division maintaining the undifferentiated state. An ideal stem cell should have the capacity of self renewal (the ability of the cell to proliferate to about 40-60 population doublings).
- **Potency** means the differentiation capacity of the stem cells.

Classification Of Stem Cells^{(10,11):-}

There are different types of stem cells that are usually considered for their potential use in research. They can be classified on the basis of the following:

1. The extent to which they can differentiate into different cell types.
2. Source of stem cells.

Extent to which they can Differentiate:-

- a. **Totipotent cells:** These stem cells have the potential to differentiate into any kind of cell in the body.
- b. **Pluripotent cells:** These are true stem cells, with the potential to differentiate into any differentiated cells in the body, except the cells of placenta or other supporting tissues of the uterus.
- c. **Multipotent cells:** These are true stem cells, but only differentiate into a limited number of cell types, **e.g.** the bone marrow contains multipotent stem cells that give rise to all the cells of the blood but not to other types of cells. Multipotent stem cells are of three major types - *ectodermal* (skin and nerves), *mesodermal or mesenchymal* (bone, cartilage, muscle and adipose tissue) and *endodermal* (intestines and other organs).

On the Basis of Source^{(12,13):-}

- a. Embryonic stem cells
- b. Embryonic germ cells
- c. Adult stem cells
- d. Induced pluripotent stem cells

a. Embryonic stem cells^{(14):-}

Embryonic stem cells (ESCs) are derived from 4 to 5day old embryo, (early-stage embryo, 4-5 days old, consist of 50-150 cells) or earlier morula-stage embryo. At this stage, the embryos are spherical and are known as blastocysts. Each blastocyst consists of 50 to 150 cells and includes three structures: an outer layer of cells, a fluid filled cavity and a group of about 30 pluripotent cells at one end of the cavity called the inner cell mass(Figure 1) .

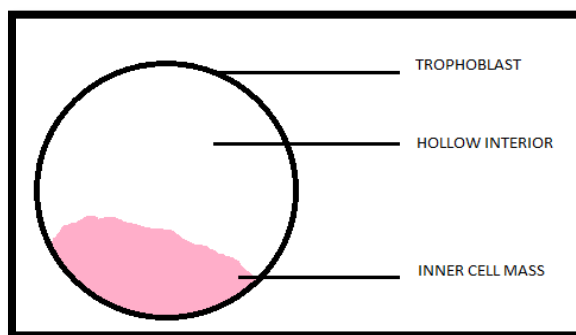


Figure 1:- Embryonic stem cells from blastocyst . COURTESY: K. Saraswathi Gopal et al : Stem cell and the regenerative dentist: International Journal of Contemporary Dental and Medical Reviews (2015)

b. Embryonic germ cells (or foetal germ cells):- Embryonic germ cells are pluripotent stem cells derived from the so-called primordial germ cells, which are the cells that give rise to the gametes (sperm and eggs) in adults. These can be isolated from the precursor to the gonads in aborted tissues.

c. Adult Stem Cells:- Adult stem cells are characterised as *haematopoietic* or mesenchymal. Hematopoietic stem cells are capable of forming all types of white blood cells and red cells. Sources of Haematopoietic stem cells are Umbilical cord blood, Umbilical cord, bone marrow and peripheral blood. Haematopoietic stem cells have been used to treat blood cancers such as leukaemia and other blood disorders. *Mesenchymal* are capable of forming a wide variety of connective tissues such as bone, muscle, cartilage, fat, tendons-but not internal organs or skin. Sources of Non-haematopoietic Stem Cells or Mesenchymal Stem Cells are adipose tissue, bone marrow and almost all body tissues including the pulp tissue of the teeth⁽¹³⁾.

Bone marrow^(14,15):- Bone marrow stem cells (BMSCs) can be harvested from **sternum** or **iliac crest**. It is composed of both hematopoietic stem cells and mesenchymal stem cells (MSCs). The advantage of bone marrow is that it has a larger volume of stem cells and can be differentiated in to wide variety of cells. Isolation of BMSCs can be carried out only under general anesthesia. Bone marrow contains both types of stem cells and has been most extensively researched.

Adipose tissue^(8, 16):- They can be harvested from the lipectomy or liposuction aspirate. Adipose derived stem cells (ADSCs) contain a group of pluripotent mesenchymal stem cells. Advantage of adipose tissue is that it is easily accessible and abundant in many individuals.

d. Induced pluripotent stem cells:-⁽¹⁷⁾

Induced pluripotent stem cells (iPSCs) are adult cells that have been genetically reprogrammed to an embryonic stem cell-like state by being forced to express genes and factors important for maintaining the defining properties of embryonic stem cells. They express stem cell markers also and are capable of generating cells characteristic of all three germ layers. iPSCs may prove to be useful tools for drug development and modeling of diseases and in transplantation medicine. In this evolving concept 3-4 genes found in the stem cells are transferred into the donor cells using appropriate vectors. The stem cells thus derived by culturing will have properties almost like embryonic stem cells. This path breaking discovery may have a major role in future stem cell therapy.

Source Of Stem Cells From Dental Tissues^(18,19):-

Stem cells from oral and maxillofacial region predominantly contain mesenchymal stem cells. Deciduous teeth, wisdom teeth and tooth extracted for orthodontic treatment are the major sources of dental tissue. In oral and maxillofacial area different types of dental stem cells were isolated and characterized (Figure : 2). They include

- **Dental pulp stem cells (DPSCs)**
- **Stem cells from exfoliated deciduous teeth (SHED)**
- **Periodontal ligament stem cells (PDLSCs)**
- **Stem cells from apical papilla (SCAP)**
- **Dental follicle progenitor cells (DFPCs)**

Dental pulp stem cells (DPSCs) were successfully isolated by Gronthos et al⁽¹⁸⁾., in 2000. They were able to demonstrate odontoblast like cells from DPSCs producing ectopic dentin in the immunocompromized mice.

Stem cells from exfoliated deciduous teeth (SHED)⁽¹⁹⁾ were identified to be cells of higher proliferation rate, with increased population doublings, immature multipotent clonogenic cells isolated from deciduous teeth that can differentiate into several cell types.

Periodontal ligament stem cells (PDLSCs) were isolated from periodontal ligament of 25 human third molars by Seo BM et al in 2004⁽²⁰⁾. They demonstrated cementoid cells, adipocytes when transplanted into immunocompromized rodents.

Stem cells from apical papilla (SCAP) Sonoyama et al in 2008⁽²¹⁾ isolated mesenchymal stem cells from apical papilla which are capable of forming odontoblast like cells in vivo.

Dental follicle progenitor cells (DFPCs) Morszeck C et al in 2005⁽²²⁾ obtained stem cells from dental follicle which can form cementoblasts, periodontal ligament cells and osteoblasts.



Figure 2:- Stem cells of dental origin

Dental Stem Cells Advantages ^(6,24) :-

The advantages of stem cells from oral and maxillofacial region is that

1. Have high plasticity.
2. It can be cryopreserved for longer period.
3. It showed good interaction with scaffold and growth factors.
4. The dental pulp stem cells will be better fitting tool due to easy surgical access, the very low morbidity of the anatomical site.

Stem Cells Storage And Transport ^(24,25) :-

Tissue samples containing stem cells are placed in a screw top vial containing Hypothermosol, mesencult basal medium or phosphate-buffered saline media, which nourishes it during transport. The sample should reach the processing storage facility before 40 hours. In the laboratory the samples were trypsinized and cryopreserved at -150°C to yield colonies of stem cells. The required cell type can be manipulated by utilizing right inductive signals and appropriate growth factors to the stem cells.

Stem Cell Application In Oral Diseases:-

Clinical applications of dental stem cells will continue to emerge in the near term and longer term. Currently dental stem cell research focuses on **regeneration of dentine, pulp and teeth, alveolar bone, regeneration of periodontal ligament, and various other applications of mesenchymal and hematopoietic stem cells in repair of craniofacial defects, oral mucosal lesions and salivary gland regeneration after radiation therapy.**

Regeneration of teeth ^(25,26) :-

Tooth regeneration represents a new era in dentistry as the concepts of repair is being shifted to regeneration. Regenerative dentistry restores a tissue defect to its original form and function by biological substitutes. Dental tissue stem cells/ progenitor cells can differentiate into dental cell lineages, and are used to regenerate some dental tissues in the treatment of tooth defects and tooth loss. Research focuses on whole tooth regeneration using a strategy of transplanting artificial tooth germ and allowing it to develop in the adult oral environment. Two means of regenerating teeth include **conventional tissue engineering**, in which the application of cells in a carrier material *in vitro* under the influence of a stimulus leads to tissue regeneration. The second process of tooth regeneration is using **dental epithelium and mesenchymal cells** ^(27,28) *in vivo* after direct implantation, which is based on knowledge of embryogenesis and physiological tooth development during childhood.

Handa K (2002) ⁽²⁹⁾ isolated progenitor cells from bovine dental follicles. In invitro conditions these cells formed clusters of spheroid like cells and in invivo conditions, cementum matrix formation took place by these cultured dental follicle cells. Gronthos et al 2002⁽²⁵⁾ demonstrated both in vivo and in vitro in animals that dental pulp stem cells (DPSCs) were able of forming ectopic dentin and related pulp tissue⁽²⁵⁾. Batouli et al. 2003⁽³⁰⁾ demonstrated the capacity of dental pulp stem cells to form a dentin pulp-like complex by transplanting them in immunocompromised mice. After 16 weeks, the pulp-like tissue contained a fibrous connective tissue, blood vessels, and odontoblasts associated with newly formed dentin indicating the possibility of using those stem cells for the repair of injured dental structures. Duailibi *et al.* 2004⁽³¹⁾, in their studies were able to form tooth from single cell suspensions of cultured rat tooth bud cells. They confirmed bioengineered rat teeth grown in 12 weeks with Poly Lactic co glycolic

acid (PLGA) and Poly glycolic acid (PGA) scaffold. Honda *et al* 2005⁽³²⁾ developed tissue engineered teeth, when implanted into omentum of rat utilizing porcine tooth bud cells and PGA fiber engage scaffold that reminds of the model of odontogenesis. Young *et al* 2005⁽³³⁾ using porcine tooth bud cells, PLGA and PGA scaffolds produced a crossbred tooth bone for the cure of tooth loss beside with alveolar bone resorption. Huang *et al* 2008⁽³⁴⁾ in his review article summarized new protocol for endodontically involved immature permanent teeth in which minimal instrumentation was done in it followed by disinfection with triple antibiotic paste. Treated tooth is coated with mineral trioxide aggregate (MTA) and filled with glass ionomer cement. Periodical observation was done to ascertain root maturation.

Periodontal regeneration:-

Recently new techniques of cell transplantation have been developed to regenerate periodontium using Periodontal ligament cells (PDLs) isolated from extracted human teeth and Dental follicle stem cells (DFSCs) which could become an alternative cell source for periodontal regeneration therapy. When transplanted into immunocompromised mice, PDLs have shown to regenerate collagen fibers and cementum/periodontal ligament-like structures. Periodontium can also be regenerated by culturing of periosteum, non-dental stem cells such as bone marrow mesenchymal stem cells, and adipose-derived stem cells. Studies have shown that grafted autologous cultured cell membrane derived from periosteum into a surgically created class III furcation defect in dogs, lead to the formation of cementum, periodontal ligament-like tissues, and alveolar bone in 3 months after the grafting procedure. Another research reported a treatment using cells derived from periosteum in human periodontal disease by Kawaguchi *et al* 2004⁽³⁵⁾. It is also confirmed by Hasegawa *et al* 2005, that periodontal defects can be managed by reimplantation of these cells⁽³⁶⁾. Autologous mesenchymal stem cells from iliac crest in combination with platelet rich plasma from peripheral blood was used for periodontal regeneration. Significant closure of bone defect and improvement of attachment level was observed after one year follow up. It also showed good healing and regeneration of interdental papilla⁽³⁷⁾. Nagatomo *et al* 2006⁽³⁸⁾ in their experimental studies found that PDL cells having stem cell properties can regenerate periodontium. Transplantation of PDL derived cells into animal models were shown to regenerate periodontal tissue. Iwata *et al* 2009⁽³⁹⁾ harvested and expanded primary canine PDL cells *in vitro* and also made into transplantable constructs containing Poly glycolic acid (PGA) Scaffold and PDL cell sheets. The transplantable constructs in combination with porous tricalcium phosphate (b-TCP) induced regeneration of periodontal structures, including alveolar bone, cementum, and periodontal fibers. Liu *et al* 2008 regenerated periodontal tissue in miniature swine using scaffolds seeded with periodontal ligament derived stem cells⁽⁴⁰⁾. PDLSCs can differentiate into cells that can colonize on biocompatible scaffold, suggesting an easy and efficient autologous source of stem cells for regeneration of dental tissues⁽⁴¹⁾. Marie MK *et al* 2009 in their experimental on goat was able to regenerate periodontal tissues around titanium implant using autologous bone marrow stem cells with scaffold⁽⁴²⁾.

Repair of craniofacial defects:-

Craniofacial defects results from post-cancer ablative surgery, craniofacial osseous deficiencies can also arise from infection, trauma, congenital malformations and progressively deforming skeletal diseases. Although autologous bone graft is considered the best option, it has the limitation of donor sites. Use of skeletal or dental stem cells can be used in future to repair craniofacial region and may provide a promising alternative approach for reconstruction of craniofacial defects. Lagenbach *et al* 2010⁽⁴³⁾ in their *in vitro* studies used microspheres (scaffold free tissue construct) to close the bone defects. They found osteogenically differentiated microspheres with outgrowing cells can be used to fill up bone defects. This new procedure has added advantage of permitting the transplantation of more cells and better integrity compared with cell suspensions or gels⁽⁴⁴⁾. Degenerative bone diseases including TMJ defects can be bioengineered with stem cells. Cells from various sources like articular cartilage cells, fibroblasts, mesenchymal stem cells have been used to reconstruct TMJ⁽⁴⁵⁾. Bone tissue engineering endeavours to repair large bone losses using three dimensional scaffolds to deliver vital cells to the defective site.

Stem Cell Therapy in Oral Mucosal Lesions:-

Among oral mucosal lesions, stem cell research is presently focused on the treatment of certain lesions only. These oral mucosal lesions are:

Ulcerative lesions: oral ulcers and wounds, oral mucositis, pemphigus vulgaris

Potentially malignant disorders: Oral submucous fibrosis (OSMF), Oral lichen planus (OLP).

Malignant lesions: oral carcinoma.

The main mechanism behind the use of stem cells in these lesions is discussed in detail.

Oral ulcers and wound healing ⁽⁴⁶⁾:-

Optimum healing of a cutaneous wound requires a good integration of the complex biological and molecular events of cell migration and proliferation and extracellular matrix deposition, angiogenesis and remodeling. Large wounds take a long time to heal, and defective healing leaves behind unacceptable scars and strictures.

Bone marrow-derived mesenchymal stem cells (BM-MSCs) are self-renewing, expandable stem cells and are able to differentiate into adipocytes, osteoblasts, and chondrocytes. These cells possess the ability to engraft at the site of injury and promote tissue regeneration and wound healing through synergistic downregulation of proinflammatory cytokines and increased production of soluble factors with antioxidant, antiapoptotic, and proangiogenic properties.

In oral wounds, they exhibit increased re-epithelialization, cellularity, intracellular matrix formation and neoangiogenesis, thereby accelerate wound healing. Hence, MSC therapy can be a promising therapeutic modality for oral ulcers and wounds. El-Menoufy *et al* 2010⁽⁴⁷⁾. Submucosally injected autologous Bone Marrow-Mesenchymal Stem Cells suspended in phosphate buffered saline around formocresol induced oral ulcers in dogs. There was increased expression of both collagen and vascular endothelial growth factor (VEGF) genes in Mesenchymal Stem Cells-treated ulcers compared with controls. Similar results were seen by Aziz Aly *et al* 2012.⁽⁴⁸⁾ in a study on formocresol induced oral ulcers in dogs using Bone Marrow and adipose-derived stem cells. Hence He concluded Mesenchymal Stem Cells transplantation may help to accelerate the healing of oral ulcers [Figure 3].

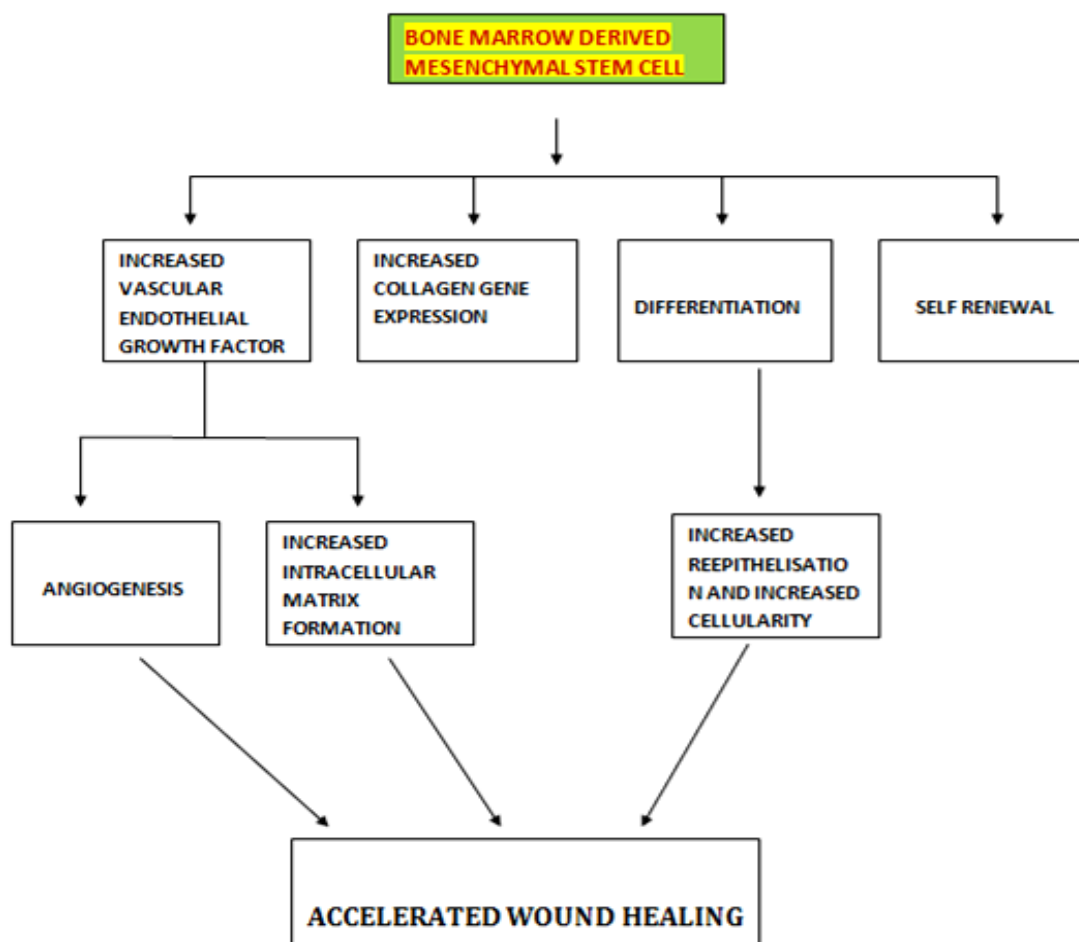


Figure 3:- Role of mesenchymal stem cells in wound healing Courtesy : Journal of Pharmacy & Bioallied Sciences : Jan-Mar 2015; 7 (1): 2-8

Oral mucositis:-

Oral mucositis is one of the most debilitating side effects which occur with chemo or radiotherapy. Management of mucositis is fairly symptomatic. Presently mesenchymal stem cells have been explored in its management by virtue of their immunomodulatory, anti inflammatory functions as well as regenerative properties. Their therapeutic efficacy can further be increased by transgenic approach or preconditioning them with certain factors like pro-inflammatory cytokines.

Zhang *et al* 2012.⁽⁴⁹⁾ injected spheroid Gingiva-Derived Mesenchymal Stem Cells (GMSCs) in mice with chemotherapy-induced oral mucositis and found that treatment with GMSCs decreased the severity and incidence of ulceration and restored the papillae structure, the lining, and thickness of the epithelial layer as compared with those of untreated disease group. The improved therapeutic benefits of spheroid-derived GMSCs may be attributed to their increased capabilities for engraftment and survival at the injury sites, trans-differentiation into epithelial cells, and preconditioning to hypoxic and oxidative challenges. Hence GMSCs could prove a sure shot therapy in oral mucositis following the cancer therapies.

Pemphigus vulgaris:-

Pemphigus vulgaris is a potentially life-threatening disease, primarily affects the mucous membranes of patients over the age of 50 years characterised by formation of autoantibodies, directed against desmosomal glycoproteins (dsg1, dsg3) present on the cell surface of the keratinocyte resulting in the formation of intraepithelial bullae and mucosal ulceration. The effective treatment of pemphigus vulgaris is long-term use of corticosteroids that itself has detrimental systemic complications. The immunomodulatory and antiinflammatory properties of stem cells can be utilized in the treatment of the condition.

In a research Vanikar *et al.*(2007) ⁽⁵⁰⁾ performed allogenic hematopoietic stem cell transplant (HSCT) with nonmyeloablative low-intensity conditioning in nine patients of Pemphigus vulgaris and found that the existing skin lesions started to regress within 24 h of stem cell treatment . In a similar study conducted by Vanikar *et al* in 2012, consisted of clinical trial on patients with pemphigus vulgaris to evaluate the effects of allogenic Hematopoietic Stem cell Transplantation into thymus, bone marrow and peripheral circulation on central and peripheral arms; it was found that the recovery began within 24 h of HSCT and new lesions stopped after 6 months⁽⁵¹⁾. Stem cell therapy in pemphigus not only has shown promises in treatment but also brings about a shift towards non steroidal approach in autoimmune diseases. However the use of stem cell therapy for oral pemphigus is yet to be ascertained and needs new clinical trials.

Stem Cell Therapy In Potentially Malignant Disorders:-**Oral submucous fibrosis:-**

Oral submucous fibrosis is a chronic, insidious disease associated with both significant morbidity (including pain, reduced mouth opening) and an increased risk of malignancy. Various agents like *Areca nut*, gutkha, spices etc., are known to cause insult to oral mucosa by increasing cytokine production and release of reactive oxygen species; which in turn results in increased synthesis of collagen, decreased collagen breakdown, compromised vascularity and increased tissue oxidative stress, ultimately resulting in clinical OSMF⁽⁵²⁾.

Stem cell therapy is primarily aimed at neoangiogenesis by releasing cytokines and growth factors (paracrine effect)⁽⁵³⁾. This may result in increased free radical scavenging by antioxidants (either naturally occurring or exogenous). Neoangiogenesis may also facilitate the removal of senescent cells from the lesions by supplying more number of scavenging defense cells and reversal of hypoxia in the diseased tissue. Stem cell therapy may help to stimulate resident tissue stem cells to transform into new fibroblasts, which may help in the removal of disintegrated biochemically and morphologically altered collagen fibers(Figure 4)

Sankaranarayanan *et al* 2013.⁽⁵⁴⁾ have demonstrated the effectiveness of stem cell treatment in OSMF patients by injecting 0.5–1 ml of marrow-derived stem cell concentrate into labial and buccal mucosa and tongue under local anesthesia. They found reduction in blanching, decrease in the burning sensation while consuming spicy food, significant increase in the mouth opening and the results were found to be sustained in the follow-up period from 6 months to 5 years

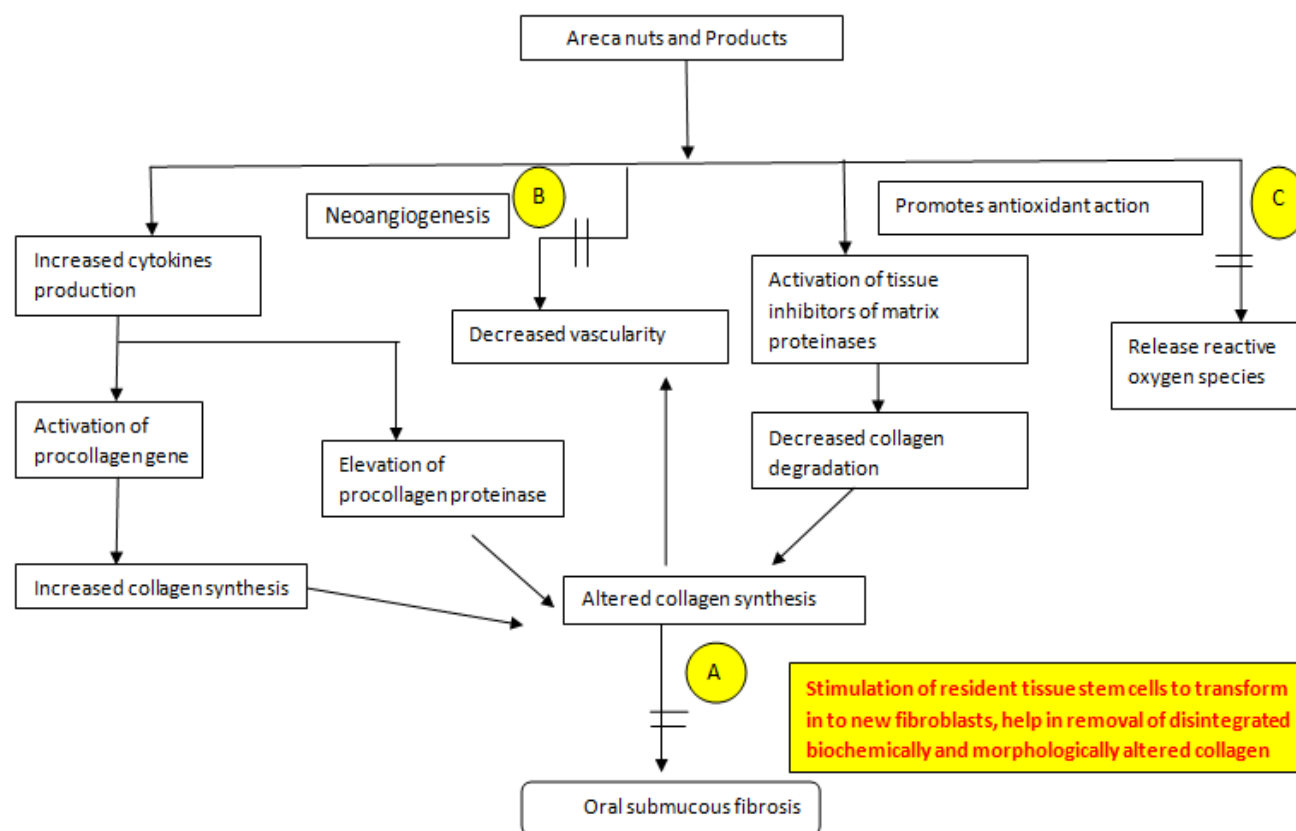


Figure 4:- Role of stem cell therapy in oral submucous fibrosis at various levels. (a) Removal of pathologically altered collagen and stimulation of healthy collagen. (b) Promoting neoangiogenesis. (c) Promoting antioxidant action. **Courtesy:** Journal of Pharmacy & Bioallied Sciences : Jan-Mar 2015; 7 (1): 2-8

Oral lichen planus:-

Oral lichen planus (OLP) is a T cell-mediated, chronic mucocutaneous disease with uncertain etiopathogenesis. Various factors like stress, genetics, systemic diseases, drugs, dental restorative materials and viruses are known to cause the disease either by an antigen-specific mechanisms like activating cytotoxic T-cells and nonspecific mechanisms like mast cell degranulation and matrix metalloproteinase activation. Both of these cause the disruption of the basement membrane, which in turn triggers apoptosis of basal epithelial cells⁽⁵⁵⁾. Recent advance in the treatment of lichen planus employ the use of immunomodulating agents such as tacrolimus which may treat the disease. In the past several years, studies have focussed on immunosuppressive properties of mesenchymal stem cells on various immune cell type as shown in **figure 5**. Based on these studies it is proposed that mesenchymal stem cells can be utilized to treat oral lichen planus via systemic infusion or local application^(56,57)

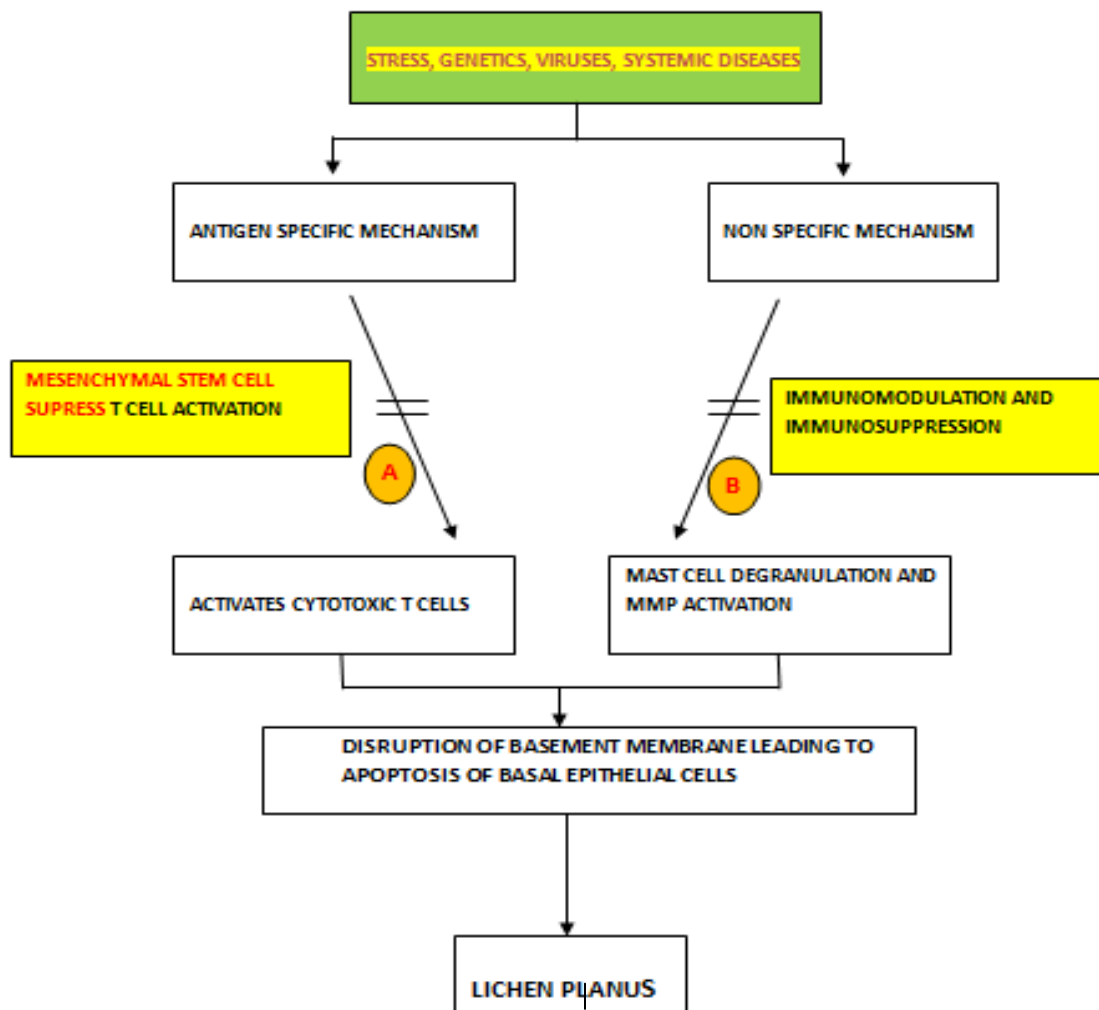


Figure 5:- Action of stem cells in modulation of immune response in oral lichen planus (shown at points A and B).

Courtesy: Journal of Pharmacy & Bioallied Sciences : Jan-Mar 2015; 7 (1): 2-8

Malignant lesions:-

Oral carcinomas:-

Surgery and external beam radiation has been the mainstay of treatment for head and neck squamous cell carcinomas. Studies in a wide variety of malignancies have demonstrated that a distinct subpopulation of tumor cells, termed Cancer stem cells (CSCs) ⁽⁵⁸⁾, contain the ability to undergo self-renewal and differentiation and hence have the ability to initiate tumorigenesis and support ongoing tumor growth. Stem cells here play a dual role-in carcinogenesis and in the development of possible new cancer treatment options in future. For past so many years stem cells have been used in the replenishment of blood and immune system damage during treatment of cancer by chemotherapy or radiotherapy.

Apart from their use in the immuno-reconstitution, the stem cells have been reported to contribute in the tissue regeneration as they have extraordinary capacity to regenerate and differentiate. Recently, a new model of carcinogenesis was suggested, which lead to the cancer stem cell hypothesis. According to this hypothesis, not all epithelial cells are capable of generating a tumour, and only long- surviving cells such as stem cells, can undergo the summative oncogenic alterations required for carcinogenesis ^(59,60).

The Mesenchymal stem cells (MSCs) have been used in the cell-based bone reconstruction following chemotherapy and surgery in malignancies like osteosarcoma and Ewing sarcoma⁽⁶¹⁾

Another important aspect of their use in cancer therapy is the use as delivery vehicles. Systematic delivery of drug or gene therapy has promising future but is currently limited by various factors such as immune detection, nonspecific accumulation in normal tissues and poor permeation. Stem cells can act as cell-based carriers that may target the desired site⁽⁶²⁾. The recent concept of use of stem cells as delivery vehicles came from the fact that the tumors send out chemo-attractants such as Vascular endothelial growth factor (VEGF) to recruit mesenchymal stem cells (MSC) to form the supporting stroma of the tumor^(63,64). However, further work is required to understand the role of stem cells in cancer therapies, with the eventual goal of eliminating the residual disease and recurrence.

Salivary gland regeneration after radiation therapy:-

Radiation induced hyposalivation is a major problem in the treatment of head and neck cancer. Studies have shown that stem cell therapy may provide a means to reduce radiation-induced hyposalivation and improve the quality of life of patients. The ability of salivary gland tissue to regenerate after atrophy has provided knowledge about location and isolation of cell populations that contain salivary gland stem cells. It has been determined that within the ducts of salivary glands reside cells capable of proliferation and differentiation known as stem/ progenitor cell population. Application of specific growth factors to these cells induces differentiation into functional units⁽⁶¹⁾. Stem cells isolated from mouse salivary glands have shown to rescue saliva production in irradiated salivary glands.

Pringle et al in 2016⁽⁶⁵⁾, showed engraftment and functional capabilities of human adult epithelial stem cells in salivary gland demonstrated in vitro self renewal and differentiation into multilineage organoids. He also showed in vivo functionality, long term engraftment and functional restoration in Xenotransplantation model.

Applications of dental stem cells in medical field:- ⁽⁶⁶⁾

Dental pulp stem cells were proven to differentiate into functionally active neurons, and when implanted dental pulp stem cells induce endogenous axon guidance, suggesting their potential as cellular therapy for neuronal disorder. Thus based on these findings, stem cell research has hope in the field of medicine directed toward: brain damage, spinal cord injury, Parkinson's disease, Alzheimer's disease, multiple sclerosis and other neurological and neurodegenerative disorders. The Tissue regeneration concept hopes to repair heart damage, baldness, deafness, blindness, impaired vision, muscle damage, diabetes mellitus, etc. Stem cell research is a boon to drug research as the drug application can be directly given on to human cells, and this will provide more relevant data than testing on animal models.

Conclusion:-

Stem cell research in dentistry has given opportunity for dentists to assume a leading role not only in the treatment of dental pathosis and also in medical diseases. With the advances in the stem cell biology, dental stem cells will hopefully be able to correct cleft palate, periodontal defects, save injured teeth, jaw bones and most strikingly regenerate the entire teeth structures. Though certain studies have confirmed the effectiveness of stem cell therapy treatment in pemphigus vulgaris, potentially malignant disorder like Oral submucous fibrosis, the research is mainly confined to animal models in oral mucositis, cancer management and Lichenplanus, thus more human research trials are needed to ascertain the role of stem cells in their management. The evolution of more methods of stem cell therapy in the future will give simple, definitive and effective treatment of most of the oral mucosal diseases.

Conflicts of interest: none

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RESEARCH ARTICLE

DATA STREAM CLUSTERING ISSUES AND CHALLENGES-A SURVEY

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Abstract

In recent years, advances in both hardware and software technology has allowed us to automatically record transactions and other information everyday at a rapid rate. Huge volumes of web, sensory and transactional data are continuously generated everyday as data streams, which need to be analyzed online as they arrive. Analysis of data streams have been researched extensively because of its emerging, imminent, and broad applications. One of the important method is clustering have been widely studied in the data mining community. Many existing data mining methods cannot be applied directly on streaming data because of the fact that the data needs to be mined in single pass. Furthermore, in data stream processing temporal locality is also quite important, because the essential patterns in the data may change and therefore, the clusters in the past history may no longer remain relevant to the future. In this paper we explore various issues and challenges on clustering data streams.

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Introduction:-

Nowadays, many applications with huge amount of data which are caused limitation in data storage capacity and processing time. Traditional data mining algorithms are not suitable for this kind of applications so they should be changed or designed with new algorithms.

The main challenge is the *data-intensive* mining using a bounded number of resources to perform an analysis to an unlimited dataset. Furthermore, traditional Data Mining Algorithms work with a static dataset and the algorithm can afford to read the data several times on the other hand Stream Mining can afford the data reading once so the algorithms for this subfield of data mining are based on a single scan.

The data generation rates in some data sources become faster than ever before. This fast generation of continuous streams of information has challenged our storage space, computation and communication capabilities in computing systems. Models and techniques have been proposed and developed over the past few years to address these challenges [3].

A data stream is a massive, infinite, temporally ordered, continuous and rapid sequence of data elements [1]. Research on data stream is motivated by emerging applications involving massive data sets such as customer click streams, supermarket, telephone records, stock market, meteorological research, multimedia data, scientific experiments data and engineering experiments data and sensor type of data. A new generation of mining

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algorithms are needed for real-time analysis & query response for these applications since the majority of conventional data mining algorithms can only be applied to static type of data sets that may be updated periodically in big chunks, but not in continuous streams of data. While data mining has become a reasonably well recognized field now, the data stream problem poses a no. of unique challenges which are not easily solved by traditional data mining methods.

Some of issues of data stream [2] like dynamic nature, Infinite size and high speed, unbounded memory requirements, Lack of global view, handling the continuous flow of data impose a great dispute for the researchers dealing with streaming data sets. Unlike traditional data sets, it is impossible to store an entire data stream or to scan through it multiple times due to its incredible volume. New methods may keep on growing in data streams at different times, to deal with this any data stream processing algorithm must continuously update their models to adapt to the changes.

Data streams pose special challenges to several data mining algorithms, not only because of the huge volume of the online data streams, but also because of the fact that the streaming data may show temporal association. Such temporal association may help disclose important data evolution characteristics, and they can also be used to develop efficient and effective mining algorithms [3]. Moreover, data streams require online mining, in which we wish to mine the data in a continuous fashion. Furthermore, the system needs to have the capability to perform an offline analysis as well based on the user interests. This is similar to an online analytical processing (OLAP) framework which uses the paradigm of pre-processing once, querying many times [5].

Rest of the paper is organized as follows. Section 2 presents types of data streams, its characteristics, applications. Overviews of data stream clustering techniques are presented in section 3. Research issues and challenges are discussed in section 4. Finally, section 5 summarizes this paper.

Data Streams: -

A data stream is a real-time, continuous, ordered sequence of items. It is not possible to control the order in which items appear nor is it feasible to locally store a stream in its entirety [7]. There are two kinds of data streams, transactional data streams and measurement data streams [8].

Transactional data streams: These are the data streams which recorded the communication between data attributes, for example; purchasing item details in credit card, phone calls details of callers to dialed parties.

Measurement data streams: Consists of data from sensors on things of interest. These types of data streams which monitored the changes of entity states, for example; traffic information at router interfaces, weather forecasting data at weather stations and traffic data in sensor networks.

Characteristics [4] of data streams as follows:-

- Huge volumes of continuous data, possibly infinite
- Quick changing and requires high-speed, real-time response
- Data stream captures properly our needs of data processing today
- Random access is expensive—single scan algorithm
- Store only the summary of the data
- Most streaming data are at pretty low-level or multi-dimensional in nature, needs multi-level and multi-dimensional processing

Some of the applications evolving data streams:-

- Telecommunication calling records
- Transaction flows of credit card
- Network traffic flow and monitoring
- Stock exchange details in financial market
- Power supply and manufacturing details
- Sensor, monitoring and surveillance flows
- Video streams, RFIDs
- Security monitoring details
- Web logs and Web page click streams

- Massive amount of data sets

There are some of the main differences between Traditional data & Data Stream [9] shows in Table-1:

Table 1:-Traditional Vs Stream

	Traditional	Stream
No. of passes	Multiple	Single
Processing time	Unlimited	Restricted
Memory Usage	Unlimited	Restricted
Types of Results	Accurate	Approximate
Distributed	No	Yes

There are some of the differences between Traditional Data base Management System (DBMS) & Data Stream Management (DSMS) [10] shows in table-2:

Table 2:-Traditional DBMS Vs DSMS.

DBMS	DSMS
Random access	Sequential access
No real time services	Real time requirements
Unbounded Disk store	Bounded main memory
Access plan determined by query processor, physical data base design	Unpredictable because of variable data
Data at any granularity	Data at fine granularity
One time queries	Continuous queries

Most data mining and knowledge discovery techniques assume that there is a finite amount of data generated by an unknown, stationary probability distribution, which can be actually stored and evaluated in multiple steps by a batch mode algorithm. For data stream mining, however, the successful development of algorithms has to take into account the following restrictions [5]:

- Data objects arrive continuously;
- There is no control over the order in which the data objects should be processed;
- The size of a stream is (potentially) unbounded;
- After processing, data objects are discarded. In practice, one can store piece of the data for a given period of time, using a forgetting mechanism called to discard them later;
- The unknown data generation process is possibly non-stationary, i.e., its probability distribution may change over time.

Data Stream Clustering:-

Imagine an enormous amount of dynamic stream data, many applications involve the automated clustering of such data into groups based on their similarities. Although there are many powerful clustering algorithms for data sets in static form, clustering data streams involves other constraints on such algorithms, as any data stream model requires algorithm to make a single scan over the data, with restricted memory and limited processing time. Several algorithms have been developed for clustering data streams described as below:

STREAM –A k-median based Stream Clustering Algorithm is presented by Guha, Mishra, Motwani and O'Callaghan [11]. It consists of two phases and follows divide and conquer approach. In first phase, it divides the data streams in relevant buckets and then finds 'k' clusters in each bucket by applying k-median clustering algorithm. It stores cluster centers only and cluster centers are weighted based on the number of data points belongs to corresponding cluster and then ignore the data points. In second phase weighted cluster centers are clustered in small number of clusters. Though its space and time complexity is low but it cannot use to concept evolution in data.

CluStream [12] clustering evolving data streams is proposed by Aggarwal et al. It divides the clustering process in following two online component and offline components. The summary of data in the form of micro clusters is stored in online component. Micro-cluster is the temporal extension of clustering feature of BIRCH [11]. Summary statistics details of data are stored in snapshots form which gives the user flexibility to specify the

time interval for clustering of micro-clusters. Offline component apply the k-means clustering algorithm to cluster micro-clusters into larger clusters.

ClusTree [13] any time Stream Clustering is proposed by Kranen et al. It divides the clustering process in following two online and offline components. Online components are used to learn micro clusters. The micro clusters are arranged in hierarchical tree type of structure. Any variety of different offline components can be developed. It is a self adaptive algorithm and delivers a model at any time.

DenStream [14], a new approach for discovering clusters in an evolving data stream. The dense micro-cluster called as core-micro-cluster is introduced to summarize the clusters with different arbitrary shape, while the potential core-micro-cluster and outlier micro-cluster structures are projected to maintain and differentiate the potential clusters and outliers. A novel pruning strategy was designed based on these concepts, which guarantees the precision of the weights of the micro clusters with limited memory.

HPStream (clustering of high dimensional data streams) [15] is proposed by Aggarwal et al. This clustering technique uses a Fading Cluster Structure (FCS) to stores the summary of data streams and it gives more significance to recent data by fading the old data with moment in time. To handle high dimensional data selects the subset of dimensions by projecting on original high dimensional streaming data. Number of dimensions and dimensions are not same for each cluster. This is due to the fact that significance of each dimension in each cluster may differ. It is incrementally updatable and highly scalable on number of dimensions. Moreover it cannot find out the cluster of different arbitrary shapes and it requires domain knowledge for identifying the number of clusters and average number of projected dimension parameters.

E-Stream [16] is a data stream clustering technique which supports following five type of advancement in streaming data: Appearance of new cluster, Disappearance of an old cluster, Splitting of a large cluster, combining of two similar type of clusters and change in the behavior of cluster data itself. It uses a fading cluster structure with histogram to approximate the streaming data. Though its performance is better than HPStream clustering algorithm but it needs many parameters to be specified by user.

HUE-Stream [17] extends E-Stream which is described earlier, to support uncertainty in mixed data (heterogeneous data). A distance function with probability distribution is introduced in between two objects to support uncertainty in categorical attributes. To detect change in clustering structure, the proposed distance function is used to combine(merge) similar clusters and find the closest cluster of a given incoming data and proposed histogram management is used to split cluster in categorical data.

POD-Clus (Probability and Distribution-based Clustering) [18] is a model based clustering technique for streaming data. It is applicable to both clustering by example and clustering by variable scenario. For summarizing and updating the cluster information incrementally, it uses a cluster synopsis which comprises the mean, standard deviation, and number of points for each cluster. It maintains concept evolution by allowing new cluster appearance, splitting of a cluster, combining of two similar clusters and disappearance of a cluster.

Issues and Challenges: -

Data stream mining is an inspiring field of study that has raised many challenges and research issues to be addressed by the database and data mining communities.

The following are the some of the research issues and challenges [18, 19]:

Handling the continuous flow of data streams: this is a data management issue. Traditional database management systems are not capable of dealing with such continuous high data rate. Novel indexing approach, storage and querying methods are required to handle this non-stopping fluctuated flow of information streams.

Unbounded memory requirements due to nonstop continuous flow of data streams: most of the machine learning methods represent the main source of data mining algorithms. Most of machine learning methods require data to be resident in memory while executing the algorithm for analysis. Due to the huge amounts of the generated streams, it is absolutely a very important concern to design space efficient techniques that can have only one scan or less over the incoming streaming data.

Required result accuracy: design a space and time efficient techniques should be accompanied with acceptable result accuracy. Approximation algorithms as described earlier can assure error limits.

Modeling changes of mining results over time: in some cases, the user is not interested in mining data stream results, but how these results are changed over time. If the number of clusters generated for example is changed, it might represent some changes in the dynamics of the arriving stream. Due to dynamic nature of data streams using changes in the knowledge structures generated would advantage many temporal-based application analysis.

Visualization of data mining results on small screens of mobile devices: conventional data mining visualization results on a desktop is still a research issue. Visualization in small screens of a PDA for example is a real challenge. Imagine a businessman is being streamed and analyzed data on his PDA. Such results should be efficiently visualized in a way that enables.

The specific challenges [20] in the context of data stream clustering scenario are as follows:

- Streams usually have massive volume, and it is often not possible to store the data explicitly on disk. Therefore, the data needs to be processed in a single look, in which all the summary data required for the process of clustering needs to be stored and maintained. The time needed to process each record must be small and constant. Otherwise, the model creation process would never be able to take up with the data stream.
- The patterns in the data stream may continuously evolve over time. From a stream mining perspective, this implies that the underlying clustering models need to be updated continuously. A usable model must be available at any time, because the end of data stream computation may never be reached, and an analyst may need results at any point in time.
- Different domains of data may pose different challenges to data stream clustering. For example, in a massive domain of discrete attributes, it is impossible to store summary representations of the data clusters efficiently without increasing the computational complexity of the problem significantly. Therefore, space-efficient methods need to be designed for massive domain clustering of data streams.

Another challenge that should be handled by data stream clustering algorithms is the ability of properly dealing with outliers, and also of detecting changes in the data distribution. The dynamic nature of evolving data streams, where new clusters often emerge while old clusters fade out, imposes difficulties for outlier detection. In general, new algorithms should provide mechanisms to distinguish between seeds of new clusters and outliers. Regarding the challenge of dealing with non-stationary distributions, the current and naive strategy employed by most available algorithms is to implicitly deal with them through window models.

Even though more robust change detection mechanisms have been implemented in generic frameworks, we believe future data stream clustering algorithms should explicitly provide mechanisms for performing change detection. Dealing with different data types imposes another challenge in data stream clustering. Different data types such as categorical and ordinal values are present within several application domains. In addition, complex data structures like DNA data and XML patterns are largely available, thus a more careful attention should be given to algorithms capable of dealing with different data types.

Conclusion: -

Now a days, continuous massive generation of stream data has lead to new trend in the field of data mining named as Data Stream Mining. So in this paper, we discussed various issues elevated by data streams and to be had an overview of various technology used for generating synopsis data structures from continuous generation of stream data. From our study we can conclude that streaming data evolves immense volumes of temporally data changing so, traditional techniques of data mining cannot be applied straightforwardly. Research in data stream mining is in early stage. If the problems caused by data streams are solved and if more efficient and interactive mining methods which are user friendly are developed, it is likely that in the near future stream mining will play vital role in business world, as it deals with many applications which involves mining from continuous data streams.

Bearing in mind that clustering data streams is a relevant and challenging task, we believe that much effort should be addressed to developing more sophisticated evaluation criteria, high-quality benchmark data, and a sound methodology for reliable experimental comparison of new data stream clustering algorithms.

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RESEARCH ARTICLE

LEGAL AID PRINCIPLE: (DIGNIFIED JUSTICE THEORY PERSPECTIVE)

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Abstract

It is a postulate in the Dignified Justice Theory perspective (Dignified Justice), that the law might only be found in the spirit of the people (*Volksgeist*, or the legal system, and in this case the Pancasila Legal System), in which the research of finding of the law has to be undertaken. The spirit of the people as such is manifested in the existing laws and regulations in the legal system in question. The same case apply in the effort to understand and describe the principle of legal aid in Indonesia, research has to be conducted in order to find it in the existing laws and regulations in the Pancasila Legal System. It has been found that an important principle of legal aid in the Pancasila Legal System is that, legal aid is the instrument of the state to protect stability in terms of close the gap between the haves and the have nots parties living in the country. Such efforts as to try as best as possible to close the gaps; may also be termed as the effort of the law in upholding human dignity (*nguwongke uwong*); in particular, serving those who has been struggling to have themselves freed from entangled legal problems encountered in the process of the court or out of the court settlements.

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Introduction:-

Referring to the direction or perspective in the what we called the Dignified Justice Theory or Dignified Justice that legal norms and principles can only be found in a particular legal system or in the spirit of the people (*Volksgeist*) which has been set as the focus of an observation and a study. Therefore, the principle or rule which are related to the legal aid as a legal concept or legal institution in Indonesia can only be found in the existing laws and regulations. There are several existing laws and regulations in Indonesia (the Pancasila Legal System), in particular the Indonesian Criminal Procedures Act, (Law Number 8 of 1981) which governing or containing the principle on legal aid as such.

Legal Aid in Laws and Regulations in the Pancasila Legal System:-

Norm and principle on legal aid in Indonesia could be found in the Definitional Section, Part 1 number (3) letter (e), of the Law Number 8 of 1981, it has been stipulated that: "every person which is connected with a criminal case is necessarily be given access to legal aid which is mostly given in order that the litigant is enabled to take every efforts in defending his or herself". This is the most important principle of legal aid in the Pancasila Legal System

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governing the due process of law stated in the Law Number 8 of 1981. This principle has a particular function, i.e. to give protection to the dignity of human being, or in other words to protect the human rights.

Concerning the issue of legal aid, an interesting issue must be discussed in this paper. The issue in question is the principle of protection to the dignity and self of human being. Accordingly, this principle is a derivative principle stated in the Indonesian Constitution, and it could also be termed as one of the manifestations of the spirit of the Indonesian people (the Indonesian *Volksgeist*) which is found its ultimate source in the Pancasila as the name of the First Agreement of the Indonesian people. Furthermore, with regard to the issue of principle of legal aid, there are also several legislations (Acts of Parliaments and several instruments) has to mentioned below.

In its history, legal aid has been regulated with clear stipulation in the Indonesian Law Number 14 of 1970 on The Basic Principles of Judicative Power. More than that, even when the Law Number 14 of 1970 was replaced by the Republic of Indonesia Law number 4 of 2004 on the same matter, in order to have it adjust to the Law Number 8 of 1981 on Criminal Procedures, the main principle of legal aid as mentioned above has still been uphold until the coming into force of the very recent Law Number 48 of 2009 governing all the Basic Principles of Judicial Power in the Pancasila Legal System.

The principle of legal aid which has justified the existing Indonesian legal aid system as mentioned above has become part of the Indonesian Criminal Justice System. To be viewed from the Dignified Justice Perspective, the principle of legal aid as such has become the principle of law, since it is derived from Pancasila. In this view, Pancasila is the supra-principle (*grundnorm*). Since the Dignified Justice recognizes that a scientific theory has various levels of application. The level is hierarchical, with lower levels subordinated to the higher ones. The highest is the core of the principle of law, and all other level are subordinated to Pancasila.

Although different with Kelsen (the Germany Jurist), in the Dignified Justice perspective once even the lowest level of principle has been derived from the highest principle (i.e. Pancasila), the (lowest) rule as such can also be termed as Pancasila, or *grundnorm*, since it contained the spirit of the people (*Volksgeist*) i.e., Pancasila. Some philosophers in Indonesia has considered Pancasila as an ideology, although in legal field it has to be called the First Agreement of the Indonesian People. Pancasila is the ultimate of all the sources of laws.¹

Pancasila is the highest norm the place where all norms under it is depended. In this case, the principle that every person which has got involved in any legal cases is obligated to be granted rights to have access to legal aid which solely to be given to him in order that he or she can take every efforts to defend his or her interest.² This could be considered as the philosophy of legal aid in general, and also the principle of legal aid which apply to the have not litigants, those who are economically weak to pay the litigation or criminal justice process.

Juridically the existence of the Indonesian Criminal Procedures Code, in which the principle of legal aid is stipulated, would not only depend its being from the Pancasila as the spirit of law in Indonesia. It is also the derivative principle of rule of law, as clearly stated in the Indonesian Constitution (the Basic Act of the State of the Republic of Indonesia of 1945). Therefore the principle of legal aid has to be understood as a logical consequence that Indonesia is also recognizing the principle of Negara Hukum (the rule of law).

As it has been generally known that the principle of the rule of law in the Pancasila Legal System as stated in the General Definition Section of the Basic Act of the State of the Republic of Indonesia of 1945 concerning The System of Government. It has been stated there that: "The State of the Republic of Indonesia is based on (*rechtsstaat*), not to be based on mere power (*machtstaat*)". This principle has been underlined and restated in the Article 1 section (3) of the Basic Act of the Amendments to the Constitution of the State of the Republic of Indonesia of 1945 with several Decrees promulgated by the People Consultative Assembly of the Republic of Indonesia. It is stated that: "the State of the Republic of Indonesia is the state of law".

¹ *Pancasila the Ultimate of All the Sources of Laws (A Dignified Justice Perspective)*, Journal of Law, Policy and Globalization, International Institute for Science, Technology and Education (IISTE), Vol. 54, October 2016.

² Teguh Prasetyo, *Hukum dan Sistem Hukum Berdasarkan Pancasila*, Cetakan Kesatu, Media Perkasa, Yogyakarta, 2013, hal., 69.

It has also been given a meaning to such a principle, that the State of Indonesia also strongly uphold the human rights and guaranteeing that all its citizens is equal to the law and government, and being obligated to upholding that law and the government without exceptions what so ever. The principle of legal aid has become fundamentally essential, and intrinsic, what a nation state has made a declaration that it is a state of law or based its principle on the rule of law. The principle of legal aid might also be found implicitly if further research has to be done to find it in another historical constitutional documents.

In the constitutional history of the Republic of Indonesia, stipulations on the concept of legal aid has been there for a very long time on the Document, called Five Year Term of National Development Plan. In that Document, legal aid was considered as part of national strategy stated in that Document. (the Third National Development Plan). The stipulation of principle of legal aid continue to be stated and implementing in the next national Plan, until the last National Plan, i.e. the fifth. On the lowest level of instrumental regulation, the principle of legal aid was stated in the Ministerial of Justice Decree Number: M.02 UM. 90.08 of 1980 on Guidelines for the Implementation of Legal Aid.

Latter on, the Ministerial Decree was replaced by Ministry of Justice Instruction of The Republic of Indonesia Number M. 08-UM.06.02 of 1992 on the Amendment to the Ministry of Justice Instruction Number M.24-UM.06.02 of 1985 on the Guidelines to the Implementation of Legal Aid for the Section of the Society which Has Been Considered as the Have Not.

While in the Ministerial Instruction Number M.24-UM.06.02 of 1985, legal aid was regarded as general, in the Minister of Justice Instruction Number M. 08-UM.06.02 of 1992 on the Amendment to the Justice Minister Instruction Number M.24-UM.06.02 of 1985, the principle of legal aid has put much weight as the legal aid program for the underprivileged of societies or the have not.

The meaning to the concept of legal aid has been enriched and also has been adjusted to the spirit of the Indonesian people to direct it to its mission as the tool for national stability. By national stability it meant, to maintain the balance in order that the increasing gap between the have and the have not in the society could be closed. This problem of economic segregation in the Indonesian society at that time was very large, and therefore was considered as dangerous to the national stability. Legal aid was given more emphasis in order to combat the economic gaps between the rich and the poor in the society.

Later on, the principle of legal aid has been given more emphasis on the help that also been given to the rich, and therefore the principle has been changed from structural principle to neutral principle. Legal aid has been given to all the people, without considering their social and economic background. Although still, legal aid is understood as legal instrument that containing the structural dimension. Legal aid has been conducted to serve the section of the society that was considered as marginalized and outcast.

The court has been used to channel the free of charge legal aid services to the section of the society, the marginalized society as mentioned above. In this case, the district court all over Indonesian has been appointed as the institutions that channels the legal aid service as such. This is understood, as when the legal aid was implemented, the judicial power was under the control of the executive, as made possible by the Law Number 14 of 1970 on the Judicial Power.

Its meaning has also been expanded, to cover the involvement of universities and law schools in the Archipelago. The doctrine of the higher education which has to be understood as universities has to get involve in teaching, research and serving the society (tri dharma) was utilised. Legal aid was placed under the higher education doctrine of serving the society. Many legal aid department at the universities all over the country was set up to serve the need of legal aid. Legal aid has been implemented via accompanying the parties in the disputes or any criminal cases, whether it is in or out-side the court. Private cases, criminal cases, including cases in which the individual has to face the Government in the Administrative Court has been the targets of the legal aid programme.

It is interesting to note, that legal aid at this stage has also been given to the Government who has to face the trial at the Administrative Court with the individual who has brought it to the court. This is the case which has been named as cases of abuse of governmental power. (*onrechtmatige daad zaken/onrechtmatigover heidsdad*).

The Aim or Purposes of Legal Aid:-

It must also be mentioned here regarding the aim and purposes of legal aid in the Pancasila Legal System. Firstly, for humanity. This particular purpose, is in line with the purposes postulated by the Dignified Justice perspective which has been constructed with the aim to create a dignified justice.³ One, from many of the orientations of the Dignified Justice perspective is to make human as human or using the Javanese phraseology *nguwongke uwong*. The legal aid programme is given in order that it could lightening the burden of life of the section of the society which is considered as the have not. This will increase the opportunity for the most part of the society to have justice and also legal protections.

Secondly, the aim of legal aid is to risen the legal awareness. It is expected that legal aid would educate, or in terms used by the Dignified Justice theory, is *nguwongke uwong* for those part of the society which is considered as the have nots, and that it could make them manage to live as members of the society with the required level of understanding, with which they are aware of their rights and obligations as member of society and as citizens. Some has saidt, "through the increasing legal awarnes of the society, the people can understand their rights and obligations, and also the utilities of the legal aid itself"⁴.

In the Era of President Joko Widodo, through the Ministry of Law and Human Rights, it has been alocated resources to manage and implementing the principle of legal aid. The emphasis has been back to the idea that legal aid has to be directed to help the marginalized and underprivileges part in the society. The task to undertake and implementing the legal aid program has been considered as a noble task, since it has been directly making contact with the neede part of the society. It has also interesting to submit here that the noble task of legal aid is associated with the term using in Part I Introduction, number (2) the Attachment of the Instruction of the Ministry of Justice of the Republic of Indonesia Number M.08-UM. 06.02 of 1992, on the Guideline of the Implementation of the Legal Aid Program for the Poor. In this regulation, the poor has been termed as *buta hukum* (lay person). In this particular aspect, legal aid has been considered as noble task since it has opened the "door" for justice to all of the people, particularly for the poor⁵.

The concept of poor in the legal aid program has been connoted as the people who has no money, and live under the poverty line, as used by the economist. This understanding has been reiterated in the Law Number 13 of 2011 on the Poor. In this law, the concept poor have to be restricted to the people who has no money, and live under the poverty line. The laws have stated that the poor means the people who have no money to pay for legal services given by the legal practitioners. No indication that the term poor have to be related with the "legal blind" or person who does not have a clear understanding of the law.

In the past, however, as mentioned on the regulations stated above, the poor that has to be given legal aid was also related with the "legal blind". The person who was ask for his or her expertice to give legal aid, in the law at that time has been considered as those who do the noble task and this in the old Latin frase was *officium nobile*. It might be also interesting to mention it here that the structure in the relation between the giver of the legal aid and its receiver has been regarded as sub-ordinative, one is higher then the other.

In the beginning, the lower status as such was understood as a section in the society that does not have the ability to see the law ("legal blind"). Therefore, the concept poor at that time has been understood as similar to the meaning people who does not know the law. Meanwhile, the party that was regarded as having the higher status was called *officium nobile* since they understood the law, and that they were able to give their legal service. In the position as *officium nobile* as such, it has been overwhelmed with virtues such as *charitable*. Therefore, the State, as dictated by the Constitution has invited their readiness as *ofcium nobise* to dedicate their live to the modern institution of the law with the task of legal aid.

³Teguh Prasetyo, *Keadilan Bermartabat Perspektif Teori Hukum*, Cetakan Pertama, Nusa Media, Bandung, 2015.

⁴ Muhadar, *Beberapa Catatan tentang Bantuan Hukum dalam Perkara Pidana Prodeo dalam Kaitannya dengan Sosial Ekonomi*, Fakultas Hukum Universitas Hasanuddin, Ujung Pandang, Ujung Pandang, 1985, hal., 47.

⁵ Ministry of Justice Instruction Republic of Indonesia Number: M. 08-UM.06.02 of 1992. According to Ismael Saleh, the Ministry of Justice at that time: "all person have always hope that poverty will not be the obstacle to have access to justice and law protection". *Ibid.*, The Attachment of the Ministry of Justice Instruction Republic of Indonesia Number: M. 08-UM.06.02 Tahun 1992.

Apart from those meaning of legal aid as mentioned above, legal aid has also been considered as a part of the criminal policy to prevent the what so called criminogenic factor. It means that legal aid has become an institution to prevent people become the criminal perpetrator or victims. Again, in this aspect, legal aid has been linked to the function of protecting the *national security*. This has been reflected in the phrase used in the Constitution that: "The state protecting all the Indonesian nation and all one's birthplace Indonesia...".

Such a formulation is used as a principle statement in the Preamble of the Indonesian Constitution, that legal aid is not monopoly the business of the state alone. The task has been linked with the concept outsourcing the sovereignty with the tight control still at the hands of the Government as the holder of the sovereign power. The task has been transferred to the practising lawyers (*Advokat*), as they have the skills, knowledges and expertise to be given power in order to accompany their client before the law (the court).

Practising Lawyers in Structural Legal Aid:-

Advokat or practising lawyers are the profession who give legal services, whether in or out-side the court of law. This principle has been formulated in the Article 1 Number (1) the Law Number 18 of 2003 on Practising Lawyer (*Advokat*).

According to Article 56 section (1) of the Criminal Procedures Act:

In case if the defendant or the accused was charged with the crime that threatened with the capital punishment or imprisonment up to fifteen years or more or for those who are poor which was threatened with five years or more which does not have their own legal council, the officer in charged on all level of investigations and court proceeding is obligated to appoint legal counsel for them.

It has to be added that legal services is also to be understood as legal aid, and it could be given by a legal counsel or practising lawyer (*Advokat*) to give legal advice, legal consultation, legal opinion, legal audit, defend either in or out of the court and also accompanying in the criminal cases, including handling of trade matters, arbitration and labour.⁶ For all of that functions, the practising lawyer has now been regarded in Indonesia as law enforcement officers. This was not the case before the Act. Therefore the Practising Lawyers as part of the "Law enforcement as a system has several components", that they must mastered: a) legal substance; b) legal structure; and c) legal culture⁷. The Law Number 18 of 2003 on Practising Lawyer (*Advokat*) is the Indonesian *Volkgeist* that dictating all of the aspects in the law enforcement system as mentioned above. This has strengthening their character as *officium nobile*.

It has to be noticed that now in Indonesia, as law enforcement officers, the practising lawyers are now having the same status as the police, public prosecutors, and also judges. At the same time, when they give legal aid to their client, they have to face the other law enforcement officers as such; which is tend to sided with those in society who have influence and economic power. Therefore it is important for the practising lawyers to understand the concept of structural legal aid. The meaning of structural legal aid is that the legal aid has to side with the individual or part of the society which is marginalized, as this is the postulate in the Dignified Justice as well. If they have to defend workers (employees) who are in conflict with the employers the lawyer has to side with the employees according to the law⁸.

In the structural legal aid, the mind set (the system) of the lawyers has to be changed. The change of legal culture for the benefit of the client has to be seriously considered. They have to think about the new legal culture, which is purely based on law, not in the political power as happened in the past Indonesia. They have to be consistent, for example with the dictate of the law to conduct the legal process with speedy, low cost, and also simple and opened or transparent procedures. They have to change the bureaucratic culture of the court and criminal justice system to side with their poor client.

In the structural legal aid, the practising lawyers have to be concerned with the broad approach in their tasks. They have to considered aspects such as conflicts dimensions between centre vs. peripherals and also aspects such as

⁶ Ropaun Rambe, *Teknik Praktek Advokat*, Grasindo, Jakarta, 2001, hal., 10.

⁷ Teguh Prasetyo dan Abdul Halim Barkatullah, *Filsafat, Teori dan Ilmu Hukum Pemikiran Menuju Masyarakat yang Berkeadilan dan Bermartabat*, Cetakan Kesatu, RajaGrafindo Persada, Jakarta, 2012, hal., 312.

⁸T. Mulya Lubis, *In Search of Human Rights*, Gramedia Pustakan Utama, Jakarta, 1993, hal., 55.

social, politics, economy, and also cultural. Legal formal approach has to the main priority, but material aspects in laws such as social factors must not be forgotten. Legal aid has to be constructed as a social movement.

They have to create a short of *power resources* to face the sections in the society which is tended to oppress the poor.⁹ The vision of structural legal aid as seen above is important. Particularly, since the legal culture, and also legal structure recognized by the Indonesian Criminal Procedures Law is based on the principle of *aquisatoir*. With this system in mind, the suspects, accused or convicted criminal, for example has all the rights to have legal aid. Since the process of investigation in the Police office, the suspects have to be accompanied by legal counsels.

This in order that they may give proper treatment in that legal process. It has been stipulated in Article 69 of the Indonesian Criminal Procedures Act that: “legal council has the right to have contact with the suspects since he or she is got caught by the police or detained in every stage of the proceedings according to ways that has been stated in the Act”. This principle has been provided in order that the legal counsel has every opportunities to implement his duties and noble task accordingly.

Conclusion:-

Legal aid, in particular the service to providing support for or assist parties which has been involved in disputes or criminal charges in the court in the Pancasila Legal System is by its nature a legal institutioun which has been provided in the legal system to protect society as a whole and in particular protecting the weaker section in the society. In the Pancasila Legal System, legal aid has to side with the poor, wheter those who are weak economically and also those who lack knowledge about the law and all its complexcities. This task of the lawyer (Advocates) who give the legal aid as such must be regarded as a noble task. This task is in line with the ideal of justice in the Dignified Justice, i.e. to make human as human within the Law.

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⁹T. Mulya Lubis, *Loc. Cit.*

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RESEARCH ARTICLE

THREE PHASE FOUR WIRE UNIFIED POWER QUALITY CONDITIONER FOR POWER QUALITY IMPROVEMENT

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B.E- Final Year Students.

Manuscript Info

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Abstract

This paper presents a novel structure for a three phase four-wire (3P4W) distribution system utilizing unified power quality conditioner (UPQC). The 3P4W system is realized from a three-phase three-wire system where the neutral of series transformer used in series part UPQC is considered as the fourth wire for the 3P4W system. A new control strategy to balance the unbalanced load currents is also presented in this paper. The neutral current that may flow toward transformer neutral point is compensated by using a four-leg voltage source inverter topology for shunt part. Thus, the series transformer neutral will be at virtual zero potential during all operating conditions. The simulation results based on MATLAB/Simulink are presented to show the effectiveness of the proposed UPQC-based 3P4W distribution system. Index Terms- Active power filter (APF), four-leg voltage-source inverter (VSI) structure, three-phase four-wire (3P4W) system, unified power quality conditioner (UPQC).

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Introduction:-

The use of sophisticated equipment/loads at transmission and distribution level has increased considerably in recent years due to the development in the semiconductor device technology. The equipment needs clean power in order to function properly. At the same time, the switching operation of these devices generates current harmonics resulting in a polluted distribution system. The power-electronics-based devices have been used to overcome the major power quality problems. To provide a balance, distortion-free, and constant magnitude power to sensitive load and, at the same time, to restrict the harmonic, unbalance, and reactive power demanded by the load and hence to make the overall power distribution system more healthy, the unified power quality conditioner (UPQC) is one of the best solutions. A three-phase four-wire (3P4W) distribution system can be realized by providing the neutral conductor along with the three power lines from generation station or by utilizing a delta-star (Δ -Y) transformer at distribution level. The UPQC installed for 3P4W application generally considers 3P4W supply. This paper proposes a new topology/structure that can be realized in UPQC-based applications, in which the series transformer neutral used for series inverter can be used to realize a 3P4W system even if the power supplied by utility is three phase three-wire (3P3W). This new functionality using UPQC could be useful in future UPQC-based distribution systems. The unbalanced load currents are very common and yet an important problem in 3P4W distribution system. This paper deals with the unbalanced load current problem with a new control approach, in which the fundamental active powers demanded by each phase are computed first, and these active powers are then redistributed equally on each of the phases. Thus, the proposed control strategy makes the unbalanced load currents as perfectly balanced source currents using UPQC. The proposed 3P4W distribution system realized from existing 3P3W UPQC-based system

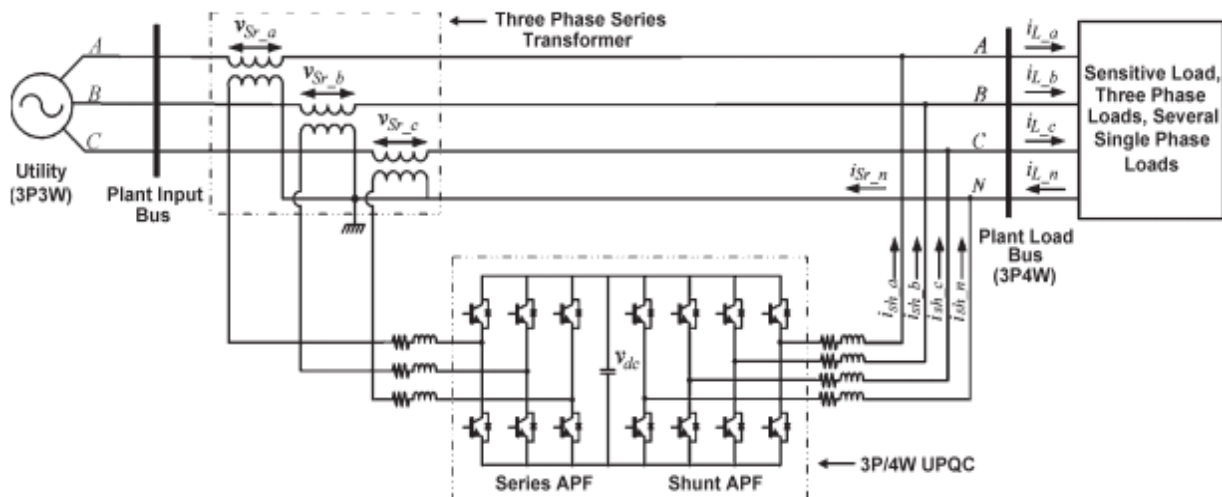


Fig:-Proposed 3P4W system realized from a 3P3W system utilizing UPQC.

Proposed 3p4w Distribution System Utilizing Upqc:-

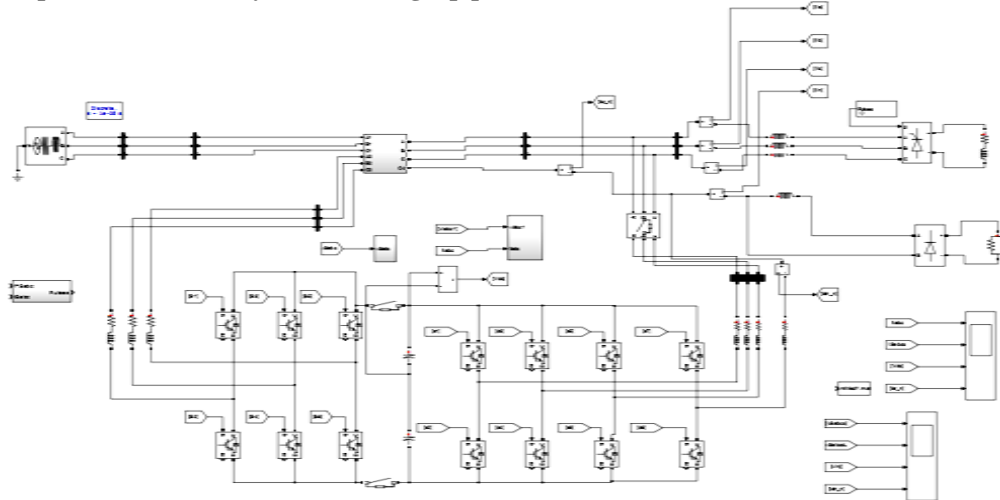


Figure: - Simulation Block Diagram of 3P4W system realized from a 3P3W system utilizing UPQC.

Generally, a 3P4W distribution system is realized by providing a neutral conductor along with three power conductors from generation station or by utilizing a three-phase Δ -Y transformer at distribution level shows a 3P4W network in which the neutral Conductor is provided from the generating station it shows a 3P4W distribution network considering a Δ -Y transformer. Assume a plant site where three-phase three-wire UPQC is already installed to protect a sensitive load and to restrict any entry of distortion from load side toward utility

If we want to upgrade the system now from 3P3W to 3P4W due to installation of some single-phase loads and if the distribution transformer is close to the plant under consideration, utility would provide the neutral conductor from this transformer without major cost involvement. In certain cases, this may be a costly solution because the distribution transformer may not be situated in close vicinity. Recently, the utility service providers are putting more and more restrictions on current total harmonic distortion (THD) limits, drawn by nonlinear loads, to control the power distribution system harmonic pollution. At the same time, the use of sophisticated equipment/load has increased significantly, and it needs clean power for its proper operation. Therefore, in future distribution systems and the plant/load centers, application of UPQC would be common. It shows the proposed novel 3P4W topology that can be realized from a 3P3W system. This proposed system has all the advantages of general UPQC, in addition to easy expansion of 3P3W system to 3P4W system. Thus, the proposed topology may play an important role in the

future 3P4W distribution system for more advanced UPQC based plant/load center installation, where utilities would be having an additional option to realize a 3P4W system just by providing a 3P3W supply. The UPQC should necessarily consist of three-phase series transformer in order to connect one of the inverters in the series with the line to function as a controlled voltage source.

If we could use the neutral of three-phase series transformer to connect a neutral wire to realize the 3P4W system, then 3P4W system can easily be achieved from a 3P3W system. The neutral current, present if any, would flow through this fourth wire toward transformer neutral point. This neutral current can be compensated by using a split capacitor topology [2], [9], [10] or a four-leg voltage-source inverter (VSI) topology for a shunt inverter [2], [11]. The four-leg VSI topology requires one additional leg as compared to the split capacitor topology. The neutral current compensation in the four-leg VSI structure is much easier than that of the split capacitor because the split capacitor topology essentially needs two capacitors and an extra control loop to maintain a zero voltage error difference between both the capacitor voltages, resulting in a more complex control loop to maintain the dc bus voltage at constant level. In this paper, the four-leg VSI topology is considered to compensate the neutral current flowing towards the transformer neutral point. A fourth leg is added to the existing 3P3W UPQC, such that the transformer neutral point will be at virtual zero potential. Thus the proposed structure would help to realize a 3P4W system from a 3P3W system at distribution load end.

Simulation Block Diagrams and Results:-

The simulation results for the proposed 3P4W system realized from a 3P3W system utilizing UPQC.

Utility voltage are assumed to be distorted with voltage THD of 35.41%. The resulting load current profile has THD of 12.1%. The UPQC should maintain the voltage at the load bus at a desired value and free from distortion. The plant load is assumed to be the combination of a balanced three phase diode bridge rectifier followed by an RL load, which act as a harmonic generating load and three different single. This would eventually result in easy expansion from 3P3W to 3P4W systems. A control strategy to generate balanced reference source current under unbalanced load condition is also proposed in this paper.

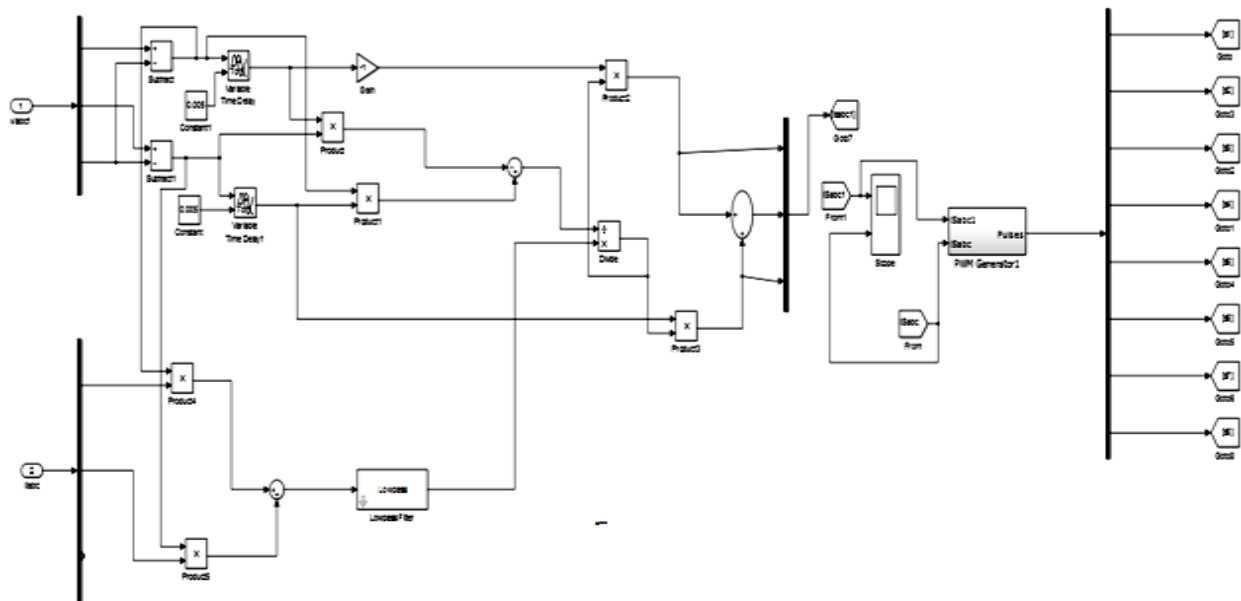


Fig: - Shunt Active Power Filter.

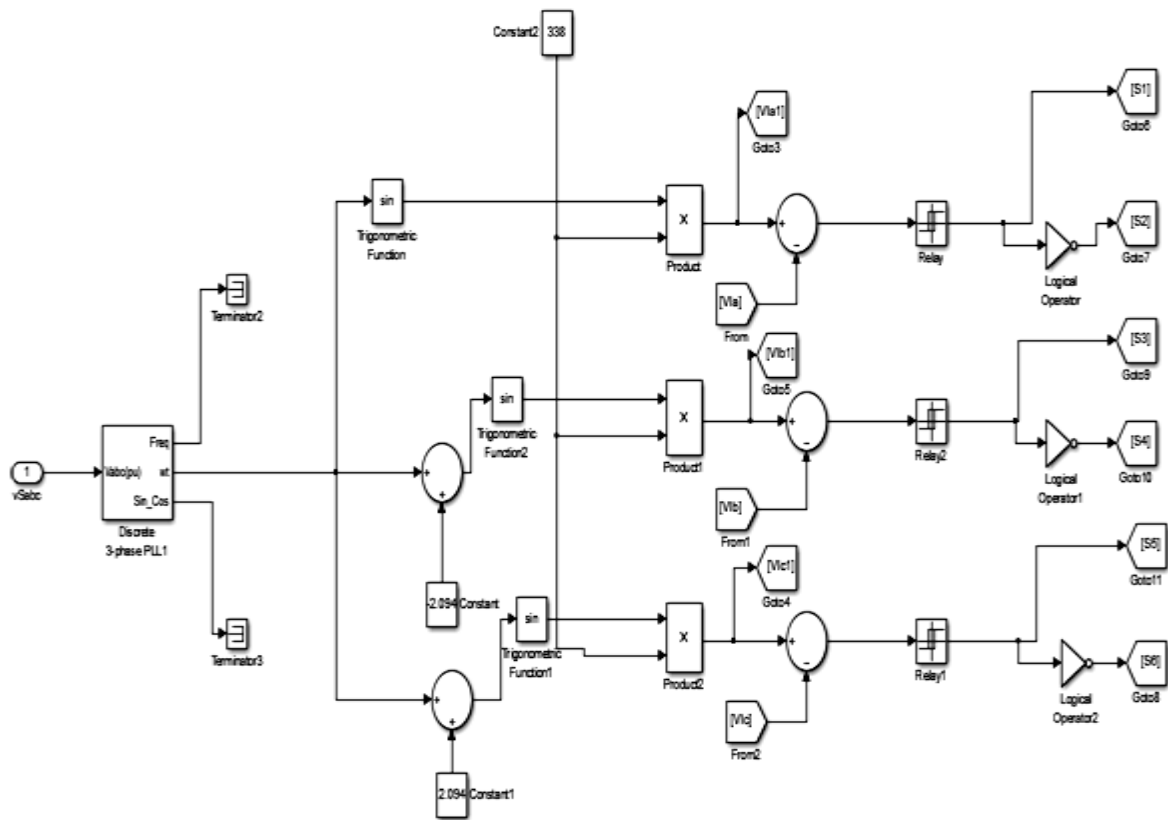
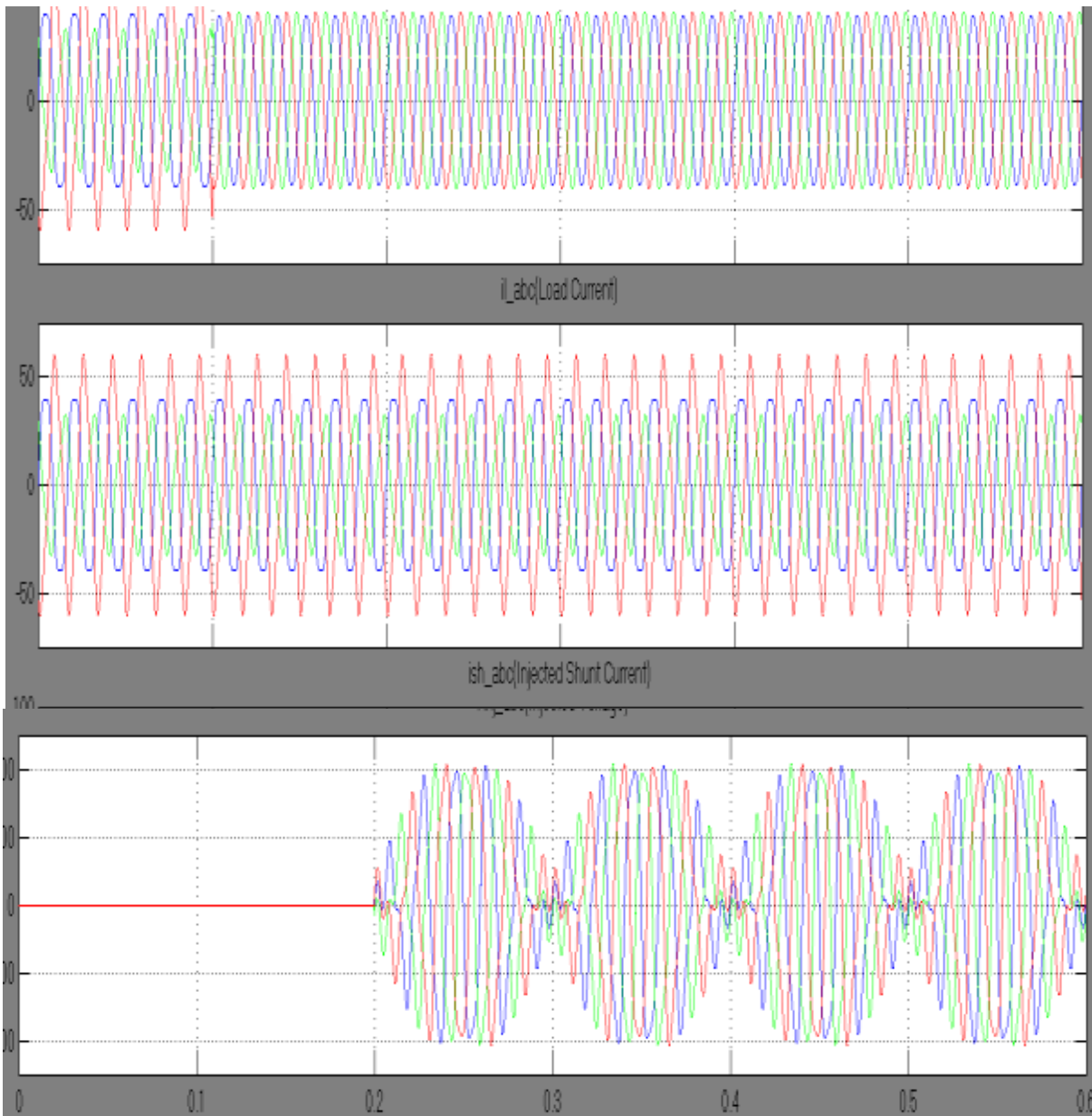


Figure: - Series Active Power Filter

The series active power filter injects the required compensating voltages through series transformer, making the load voltage free from distortion (THD = 1.51%) and at a desired level in load voltage. The series active power filter injected voltage profile

The compensated source currents are perfectly balanced with the THD of 2.26% .The compensating current injected through the fourth leg of the shunt APF. The load neutral current profile, the shunt APF effectively compensates the current flowing toward the transformer neutral point. Thus, the series transformer neutral point is maintained at virtual zero potential Simulation Block Diagram of 3P4W system realized from a 3P3W system utilizing UPQC.



Conclusion:-

The design of a unified power quality conditioner (UPQC) connected to 3P4W distribution system has been presented in this project. Where UPQC is installed to compensate the different power quality problems, which may play an important role in future UPQC- based distribution system. The simulation results shows that the distorted and unbalanced load currents seen from the utility side act as perfectly balanced source currents and are free from distortion. Here we can absorb the power quality problems like voltage and current unbalanced and reduced the total harmonic distortion (THD) of 3P4W system utilizing 3P3W system to connect the UPQC. The neutral current that may flow toward the transformer neutral point is effectively compensated such that the transformer neutral point is always at virtual zero potential.

Scope for Future Work:-

Proposed model for the UPQC is to compensate input voltage harmonics and current harmonics caused by non-linear load. The work can be extended to compensate the supply voltage and load current imperfections such as sags, swells, interruptions, voltage imbalance, flicker, and current unbalance. Proposed UPQC can be implemented using Multi converter-unified power quality conditioning system (MC-UPQC).

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RESEARCH ARTICLE

GC-MS ANALYSIS AND ANTIMICROBIAL ACTIVITY OF FIXED OIL FROM SAUDI *LEPIDIUM SATIVUM* (CRUSIFEREAE) SEEDS.

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Key words:-

Lepidium sativum, Fixed oil, GC-MS, Antimicrobial Activity.

Abstract

In this study we planned to identify and quantify the lipid composition of Saudi *Lepidium sativum* seed and to evaluate the potential antimicrobial activity of the extracted fixed oil. GC-MS analysis of the fixed oil revealed the presence of 16 components. Major constituents are: β -amyrin(31.33%), 9,12,15-octadecatrienoic acid methyl ester(15.97%), 9-octadecenoic acid methyl ester(11.93%), α -amyrin(9.32%), 11-eicosenoic acid methyl ester (6.64%), 9,12-octadecadienoic acid (6.03%), hexadecanoic acid methyl ester(5.24%). In cup plate agar diffusion bioassay, the oil which was extracted by two different methods (soxhlet and maceration) was evaluated for antimicrobial activity and the effect of the method of extraction on the antimicrobial potency was investigated.

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Introduction:-

Lepidium sativum (Crusifereae) is an annual plant about 50cm in height. The plant can grow easily tolerating difficult environmental conditions(Sharma and Agrawal, 2011; Wadhwal et.al, 2012). It has many branches on the upper parts and white-pinkish flowers(Derek, 1997; Boswell and Sowerby, 1863). The plant is genetically related to mustard and watercress and is known in some regions as garden cress, garden pepper cress, pepperwort, pepper grass(Cassidy and Hall, 2002; Staub and Bduchert, 2008).

Lepidium sativum L. contains significant amount of iron, calcium, folic acid beside vitamins A and C. It also contains protein(25%); leucine (8.21%); glutamic acid(19.3%) and methionin(0.97%). Seeds mainly contain alkaloids, calcium, iron, carotene, riboflavan, uric acid, phosphorus, thiamine and niacin. Seed oil contains palmitic, linoleic, stearic, behenic, oleic, arachidic and ligneric acids(Hiba and Wasfeih, 2014).

Lepidium sativum is a key species in African system of medicine where it is mainly used against bronchitis. In some Asian communities the plant is used against a wide array of human disorders(Baquar, 1989; Duke et.al, 2002). It was reported that garden cress possesses anti-inflammatory, analgesic, anticoagulant(Al-Yahya, 1994), diuretic(Patel et.al, 2009), antihypertensive (Maghrani et. al., 2005), antirheumatic (Ahsan et.al, 1989), antidiarrheal, antispasmodic, laxative(Rehman et.al., 2011) and hypoglycemic(Patole, 1998) properties.

Aqueous extracts of seeds exhibited significant water excretion in spontaneously hypotensive models without any significant change in heart rate.. The petroleum ether extract of seeds showed antimicrobial activity against some

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standard human pathogens, while oral administration of seeds proved a hypoglycemic effect(Hiba and Wasfeih ,2014).

It was reported that *Lepidium sativum* juice possess chemoprotective effect(Hiba and Wasfeih ,2014).The aqueous methanolic extract of seeds showed important improvement in various parameters of pulmonary function in a clinical experiment(Najeeb-Ur-Rehman et.al.,2012). In another clinical study , the seeds were evaluated for the management of osteoarthritis. Seeds showed considerable relief of joints pain , swelling, stiffness and other symptoms associated with osteoarthritis(Nita and pandye,2009).

It seems that *Lepidium sativum* seeds are well tolerated . In clinical experiments of the effect of seed administration on bronchial asthma, none of the test subjects showed the presence of adverse effects or any other problems physically or at hematological profile(www.bioline.org.). Feeding model animals with *Lepidium sativum* seeds (2% w/w) for six weeks was non-toxic , while a dose of 50% (w/w) was lethal(www.worldscientific.com).

Materials and Methods:-

Materials:-

Plant material:-

Lepidium sativum seeds were purchased from the local market-Ryad-Saudi Arabia and authenticated by direct comparison with a herbarium sample.

Instruments:-

A Shimadzo GC-MS-QP2010 Ultra instrument with a RTX-5MS column (30m,length ; 0.25mm diameter ; 0.25 μ m, thickness)was used.

Methods:-

Extraction of oil:-

Powdered *Lepidium sativum* seeds (400g) were exhaustively extracted with n-hexane (soxhlet).The solvent was removed under reduced pressure and the oil was kept in the fridge at 4°C for further manipulation.Another seed sample (400g) was exhaustively macerated with n-hexane.

Esterification of oil:-

A Methanolic solution of sodium hydroxide was prepared by dissolving (2g) of sodium hydroxide in (100ml) methanol.A stock solution of methanolic sulphuric acid was prepared by mixing (1ml) of concentrated sulphuric acid with(99ml) methanol.

The oil(2ml) was placed in a test tube and (7ml) of alcoholic sodium hydroxide were added followed by(7ml) of alcoholic sulphuric acid. The tube was stoppered and shaken vigorously for five minutes and then left overnight.(2ml) of supersaturated sodium chloride were added,then (2ml) of normal hexane were added and the tube was vigorously shaken for five minutes. The hexane layer was then separated.(5 μ l) of the hexane extract were mixed with (5ml) diethyl ether . The solution was filtered and the filtrate(1 μ l) was injected in the GC-MS vial.

GC-MS analysis:-

Lepidium sativum oil was analyzed by gas chromatography – mass spectrometry. A Shimadzo GC-MS-QP2010 Ultra instrument with a RTX-5MS column (30m,length ; 0.25mm diameter ; 0.25 μ m, thickness)was used. Helium (purity; 99.99 %) was used as carrier gas. Oven temperature program is given in Table 1, while other chromatographic conditions are depicted in Table 2.

Table 1:- Oven temperature program.

Rate	Temperature (°C)	Hold time (min ⁻¹)
-	150.0	1.00
4.00	300.0	0.00

Table 2:- Chromatographic conditions.

Column oven temperature	150.0°C
Injection temperature	300.0°C
Injection mode	Split
Flow control mode	Linear velocity
Pressure	139.3KPa
Total flow	50.0ml/ min
Column flow	1.54ml/sec.
Linear velocity	47.2cm/sec.
Purge flow	3.0ml/min.
Spilt ratio	- 1.0

Antimicrobial Assay:-**Preparation of bacterial suspensions:-**

One ml aliquots of 24 hours broth culture of the test organisms were aseptically distributed onto nutrient agar slopes and incubated at 37°C for 24 hours.

The bacterial growth was harvested and washed off with sterile normal saline, and finally suspended in 100 ml of normal saline to produce a suspension containing about 10⁸-10⁹ colony forming units per ml. The suspension was stored in the refrigerator at 4°C until used. The average number of viable organism per ml of the stock suspension was determined by means of the surface viable counting technique.

Serial dilutions of the stock suspension were made in sterile normal saline in tubes and one drop volumes (0.02 ml) of the appropriate dilutions were transferred by adjustable volume micropipette onto the surface of dry nutrient agar plates. The plates were allowed to stand for two hours at room temperature for the drop to dry, and then incubated at 37°C for 24 hours.

Preparation of fungal suspensions:-

Fungal cultures were maintained on dextrose agar incubated at 25°C for four days. The fungal growth was harvested and washed with sterile normal saline, and the suspension was stored in the refrigerator until used.

Testing for antibacterial activity:-

The cup-plate agar diffusion method was adopted, with some minor modifications, to assess the antibacterial activity. (2ml) of the standardized bacterial stock suspension were mixed with 200 ml of sterile molten nutrient agar which was maintained at 45°C in a water bath. (20 ml) Aliquots of the incubated nutrient agar were distributed into sterile Petri dishes. The agar was left to settle and in each of these plates which were divided into two halves, two cups in each half (10 mm in diameter) were cut using sterile cork borer (No 4), each one of the halves was designed for one of the test solutions. Separate Petri dishes were designed for standard antibacterial chemotherapeutics.

The agar discs were removed, alternate cups were filled with 0.1 ml samples of each test solution using adjustable volume microtiter pipette and allowed to diffuse at room temperature for two hours. The plates were then incubated in the upright position at 37°C for 24 hours. The above procedure was repeated for different concentrations of the test solutions and the standard chemotherapeutics. After incubation, the diameters of the resultant growth inhibition zones were measured in triplicates and averaged.

Results and Discussion:-**GC-MS analysis of *Lepidium sativum* seed oil:-**

The GC-MS analysis of *Lepidium sativum* seed oil was conducted and the identification of the constituents was accomplished by comparison with the MS library (NIST) and further confirmed by the observed fragmentation pattern. 80-95% match was observed.

Identification of oil constituents:-

The GC-MS spectrum of the studied oil revealed the presence of 16 components (Table 3). The typical total ion chromatogram (TIC) of the hexane extract is shown in Fig.1.

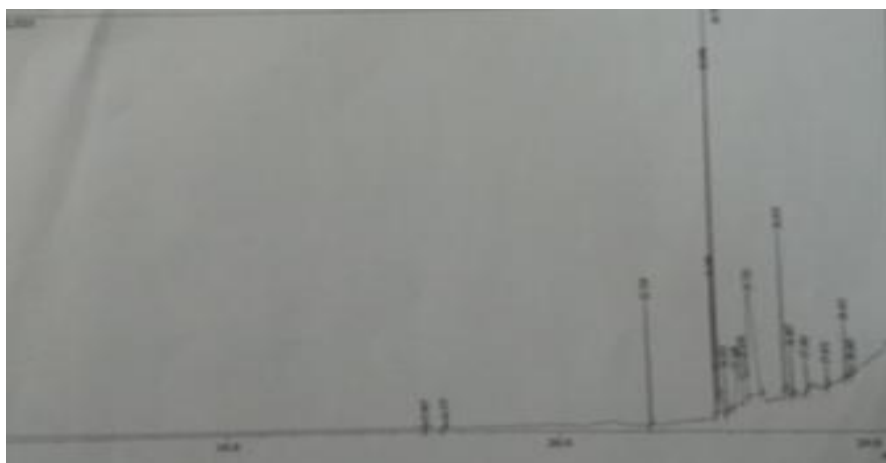


Fig.1:- Total ion chromatograms of hexane extract.

Table 3:- Constituents of *Lepidium sativum* seed oil.

Peak#	R.Time	Area	Area%	Name
1	15.967	103007	0.31	1s,4R,7R,11R-1,3,4,7-Tetramethyltricyclo[
2	16.577	139023	0.42	Butylated Hydroxytoluene
3	22.720	1720641	5.24	Hexadecanoic acid, methyl ester
4	24.640	1978338	6.03	9,12-Octadecadienoic acid (Z,Z)-, methyl e
5	24.696	3914945	11.93	9-Octadecenoic acid (Z)-, methyl ester
6	24.720	5239919	15.97	9,12,15-Octadecatrienoic acid, methyl este
7	24.935	573522	1.75	Methyl stearate
8	25.268	3059739	9.32	.alpha.-Amyrin
9	25.479	826694	2.52	Urs-12-en-24-oic acid, 3-oxo-, methyl ester
10	25.723	10281524	31.33	.beta.-Amyrin
11	26.655	2179660	6.64	11-Eicosenoic acid, methyl ester
12	26.867	633944	1.93	Eicosanoic acid, methyl ester
13	27.282	761783	2.32	9-Octadecenamide, (Z)-
14	27.872	315933	0.96	Phenol, 2,2'-methylenebis[6-(1,1-dimethyl
15	28.423	865133	2.64	13-Docosenoic acid, methyl ester
16	28.605	225495	0.69	Docosanoic acid, methyl ester
		32819300	100.00	

The following major constituents were detected:

β -Amyrine(31.33%):-

The EI mass spectrum of β -amyrine is shown in Fig. 2. The peak at m/z 426, which appeared at R.T. 25.723 in total ion chromatogram, corresponds $M^+[C_{30}H_{50}O]^+$. The peak at m/z 411 corresponds to loss of a methyl function.

9,12,15-Octadecatrienoic acid (15.97%):-

The EI mass spectrum of 9,12,15-octadecatrienoic acid (as methyl ester) is shown in Fig. 3. The peak at m/z 292, which appeared at R.T. 24.720 in total ion chromatogram, corresponds $M^+[C_{19}H_{32}O_2]^+$. The peak at m/z 277 corresponds to loss of a methyl function.

9-Octadecenoic acid methyl ester(11.93%):-

The EI mass spectrum of 9-octadecanoic acid methyl ester is shown in Fig. 4. The peak at m/z 296, which appeared at R.T. 24.696 in total ion chromatogram, corresponds $M^+[C_{19}H_{36}O_2]^+$. The peak at m/z 264 corresponds to loss of a methoxyl function.

α -Amyrine(9.32%):-

The mass spectrum of α -amyrine is displayed in Fig. 5. The peak at m/z 426, which appeared at R.T. 25.268 in total ion chromatogram, corresponds $M^+[C_{30}H_{50}O]^+$. The peak at m/z 411 corresponds to loss of a methyl function.

11-Eicosenoic acid(6.64%):-

The mass spectrum of 11-eicosenoic acid is shown in Fig. 6. The peak at m/z 324 (R.T.26.655) corresponds $M^+[C_{21}H_{40}O_2]^+$. The signal at m/z 292 is due to loss of a methoxyl function.

9,12-Octadecadienoic acid methyl ester(6.03%):-

The EI mass spectrum of 9,12-octadecadienoic acid methyl ester is depicted in Fig. 7. The peak at m/z 294, which appeared at R.T. 24.640, corresponds $M^+[C_{19}H_{34}O_2]^+$. The signal at m/z 263 accounts for loss of a methoxyl group.

Hexadecanoic acid(5.24%):-

Mass spectrum of hexadecanoic acid methyl ester is shown in Fig.8. The peak at m/z 270 (R.T. 22.720) corresponds $M^+[C_{17}H_{34}O_2]^+$. The signal at m/z 239 corresponds to loss of a methoxyl function.

13-Docosenoic acid(2.64%):-

The EI mass spectrum of 13-docosenoic acid is displayed in Fig. 9. The peak at m/z 352, which appeared at R.T.28.423 in total ion chromatogram, corresponds $M^+[C_{23}H_{44}O_2]^+$. The signal at m/z 320 corresponds to loss of a methoxyl group.

9-Octadecenamide(2.32%):-

The mass spectrum of 9-octadecenamide is shown in Fig.10. The peak at m/z 281, which appeared at R.T.27.282, corresponds $M^+[C_{18}H_{35}NO]^+$. The peak at m/z 238 corresponds to loss of acetyl function.

Eicosenoic acid(1.93%):-

The EI mass spectrum of eicosenoic acid is depicted in Fig. 11. The peak at m/z 326, which appeared at R.T.26.867 in total ion chromatogram, corresponds $M^+[C_{21}H_{42}O_2]^+$. The peak at m/z 295 is due to loss of a methoxyl group.

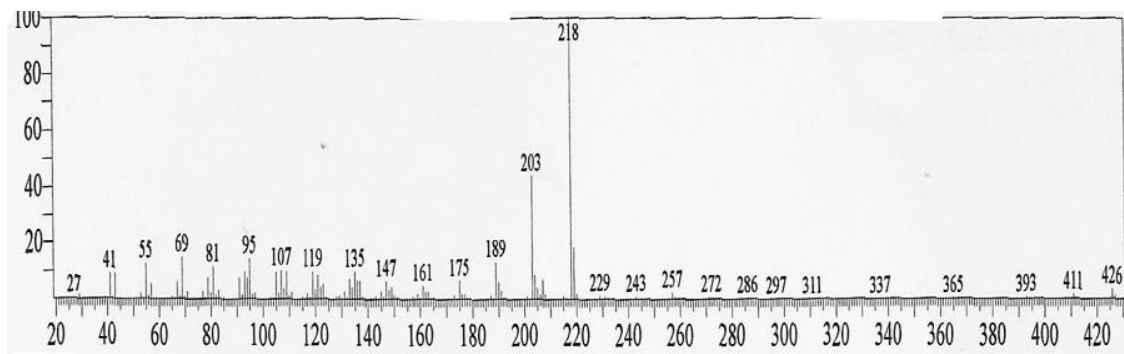


Fig 2:- Mass spectrum of β -amyrine.

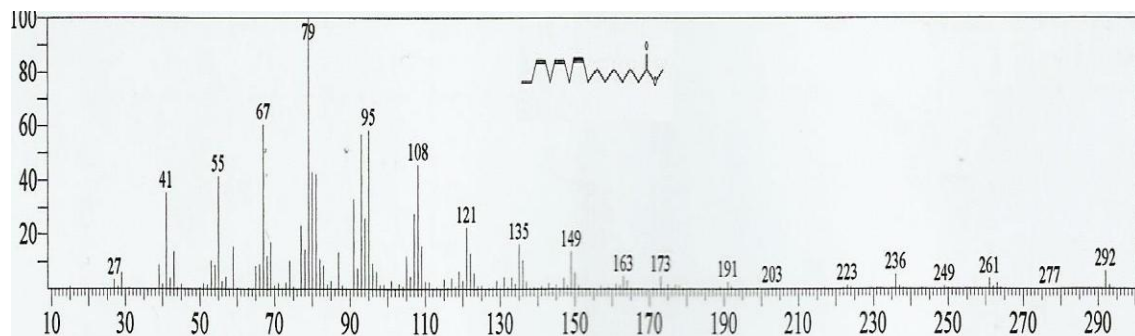


Fig 3:- Mass spectrum of 9,12,15-octadecatrienoic acid methyl ester.

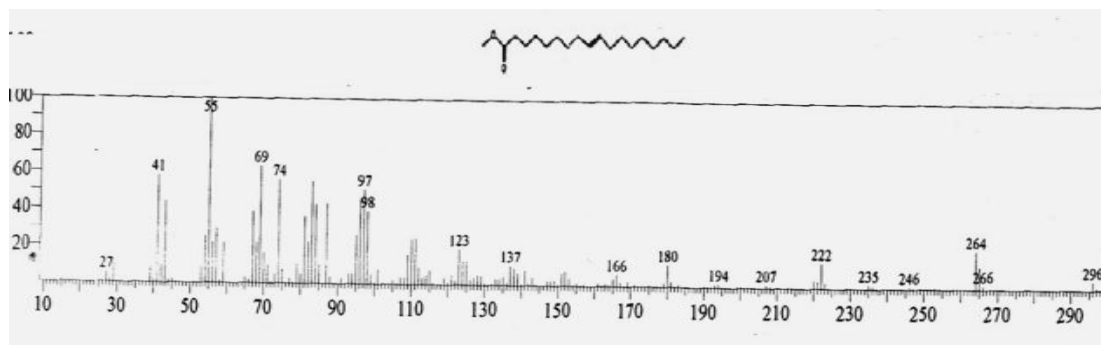


Fig 4:- Mass spectrum of 9-octadecanoic acid methyl ester.

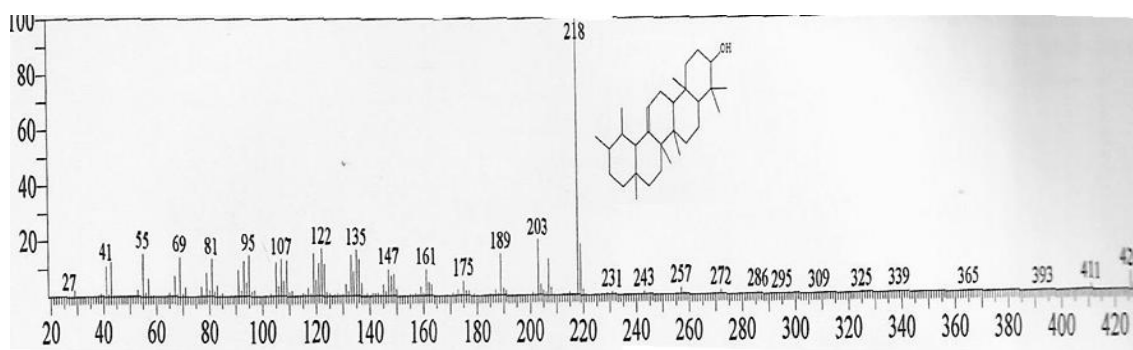


Fig 5:- Mass spectrum of α -amyrine.

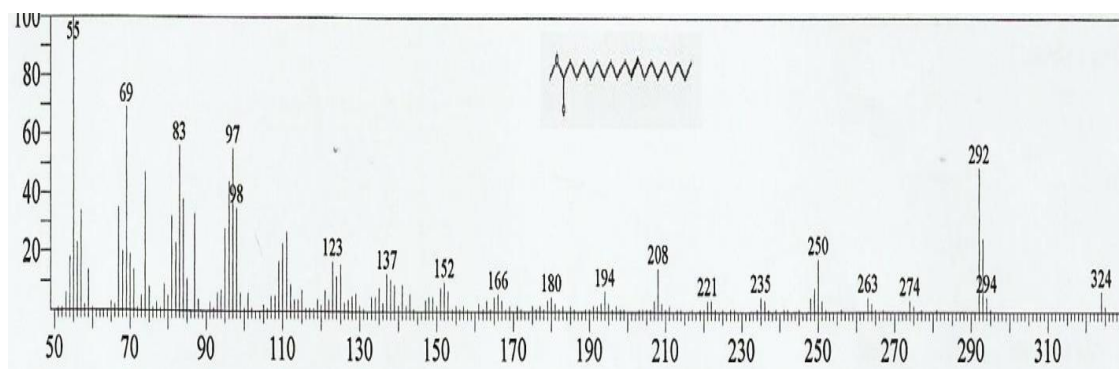


Fig 6:- Mass spectrum of 11-eicosenoic acid.

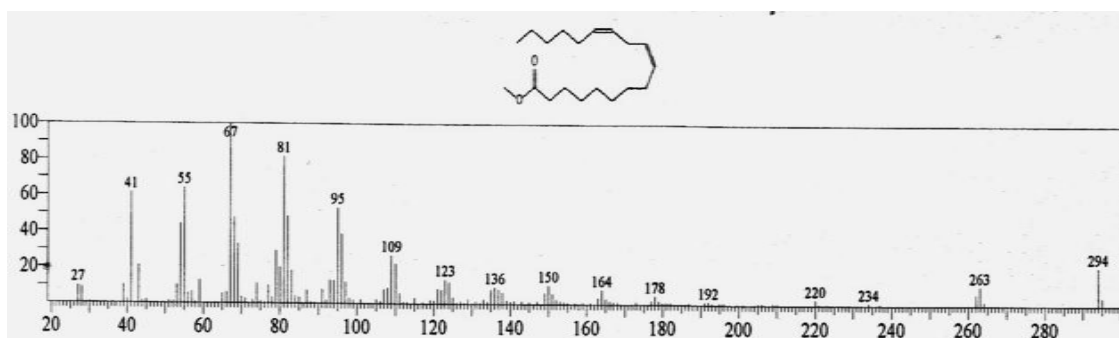


Fig 7:- Mass spectrum of 9,12-octadecadienoic acid methyl ester.

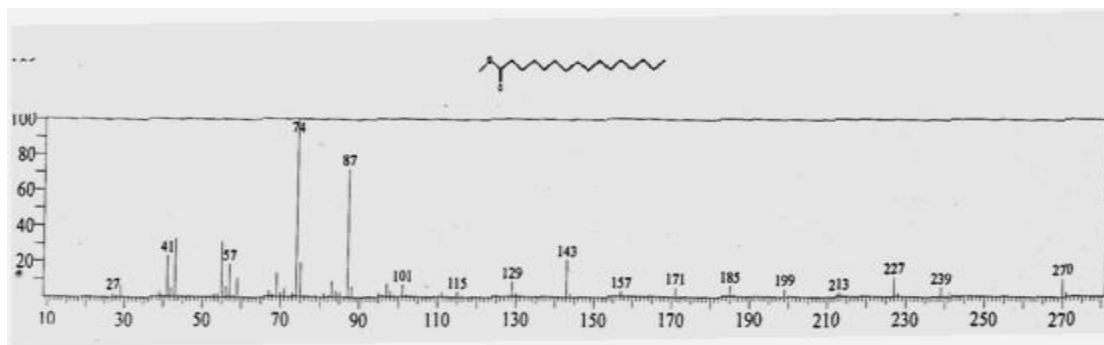


Fig 8:- Mass spectrum of hexadecanoic acid methyl ester.

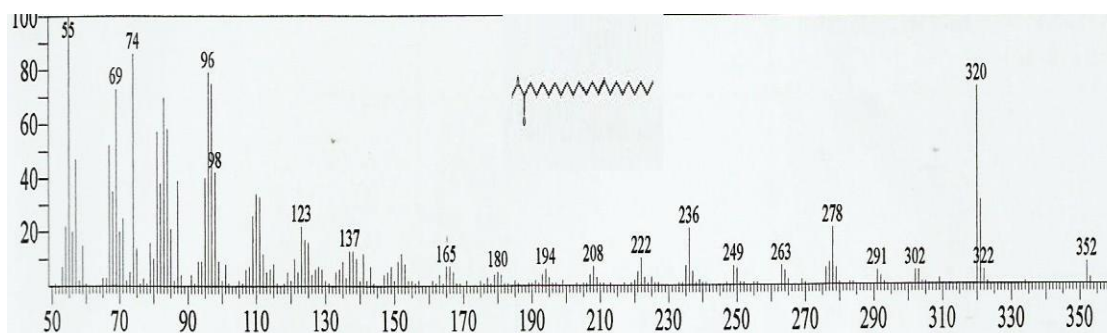


Fig 9:- Mass spectrum of 13-docosenoic acid.

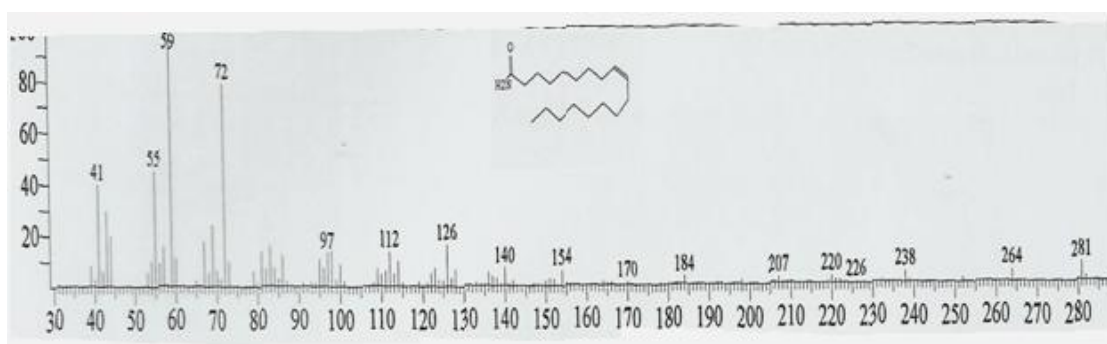


Fig 10:- Mass spectrum of 9-octadecenamide.

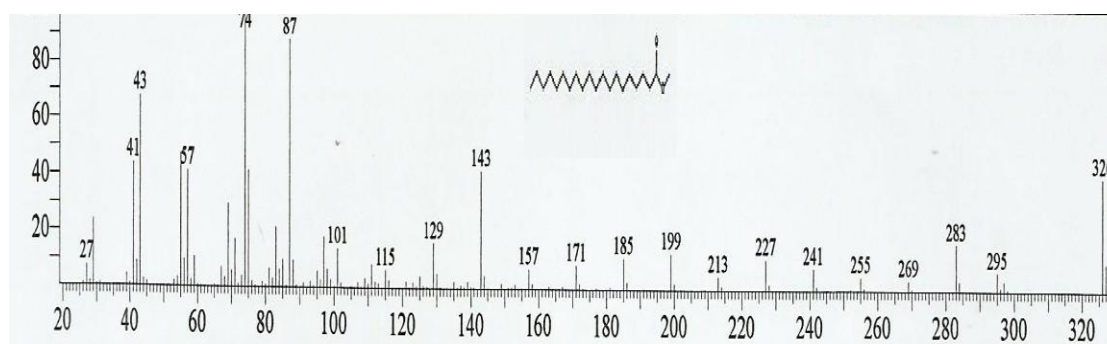


Fig 11:- Mass spectrum of eicosenoic acid methyl ester.

Antimicrobial assay:-

Lepidium sativum seed oil was extracted by two different methods (A; soxhlet and B; maceration) to evaluate the effect of heating(soxhlet) on the antimicrobial potency of the extracted oil. Table (4)shows that the oil extracted by maceration is partially active against *Escherichia coli* at concentrations of 10 and 50 µg/ml , while the soxhlet

sample was devoid of activity at these concentrations. At 50 µg/ml both samples were partially active against *Klebsiella pneumoniae*. Furthermore, the soxhlet sample exhibited partial activity against *Bacillus subtilis* at a concentrations of 10 and 50 µg/ml while the macerated sample was inactive at these concentrations. However, both samples were inactive against *Staphylococcus aureus* at test concentrations. It seems that the potency of such extracts depends largely on the type of test organism and the concentration of the sample. Similar trends were observed for the minimum inhibition concentration (Table 5).

Table 4:- Minimum Inhibition Zone (mm)

Gram	Strain	A			B			Amp	Kan	Strip		Nys
		5	10	50	5	10	50	5	10	10	10	10
-ve	<i>Escherichia coli</i>	-	-	-	-	10±0.4	12±0.3	10±0.4	15±0.4	19±0.3	25±0.4	0
-ve	<i>Klebsiella pneumoniae</i>	-	9±0.4	10±0.4	7±0.0	8±0.3	10±0.4	9±0.5	10±0.4	19±0.0	26±0.4	0
-ve	<i>Acinetobacter baumannii</i>	-	10±0.5	11±0.4	8±0.0	10±0.4	10±0.7	-	12±0.3	19±0.3	12±0.3	0
-ve	<i>Pseudomonas aureginosa</i>	-	-	-	-	7±0.4	8±0.4	-	12±0.3	14±0.3	20±0.5	0
+ve	<i>Staphylococcus aureus</i>	-	-	-	-	-	-	-	15±0.5	17±0.5	15±0.5	0
+ve	<i>Bacillus subtilis</i>	-	11±0.4	11±0.3	-	-	-	9±0.0	13±0.4	19±0.4	30±0.4	0
C	<i>Candida albicans</i>	7±0.0	7±0.4	7±0.5	7±0.4	9±0.3	11±0.6	8±0.4	N	16±0.5	N	12±0.5
F	<i>Aspergillus flavus</i>	7±0.0	9±0.4	11±0.4	-	8±0.0	8±0.3	-	N	15±0.5	N	15±0.5

-ve: gram negative, +ve: gram positive, C: colony forming, F: filamentous, -: no activity.

Table 5:- Minimum inhibition concentration (µg/ml).

Gram	Strain	A			B			Amp.	Kan.	Strip.	Nys.
		100	250	500	100	250	500	10	10	10	10
-ve	<i>Escherichia coli</i>	R	R	R	R	R	I	S	S	S	N
-ve	<i>Klebsiella pneumoniae</i>	R	R	R	R	R	R	S	S	S	N
-ve	<i>Acinetobacter baumannii</i>	R	R	R	R	R	I	S	S	S	N
-ve	<i>Pseudomonas aureginosa</i>	R	R	R	R	R	R	S	S	S	N
+ve	<i>Staphylococcus aureus</i>	R	R	R	R	R	R	S	S	S	N
+ve	<i>Bacillus subtilis</i>	R	R	I	R	R	R	S	S	S	N
C	<i>Candida albicans</i>	R	R	R	R	R	I	N	S	N	S
F	<i>Aspergillus flavus</i>	R	R	I	R	R	R	N	S	N	S

-ve: gram negative, +ve: gram positive, C: colony forming, F: filamentous,

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RESEARCH ARTICLE

MONTE CARLO MODELING OF A ^{252}Cf -BASED DETECTION SYSTEM FOR LANDMINES

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Monte Carlo modeling

Abstract

Simulations using Monte Carlo N-particle transport code MCNP5 were carried out to model a ^{252}Cf -based system for detection of buried landmines. The study explores the performance of the neutron backscattering methods in providing elemental characterization for the buried landmine. The net elastically back-scattered (EBS) neutron energy spectra at both fast and thermal neutron ranges were calculated. The net (EBS) neutrons from the major constituent elements of a landmine simulant (TNT) have shown definite structures that can be used for identification of buried landmines.

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Introduction:-

The most common explosives used in landmines are TNT ($\text{C}_7\text{H}_5\text{N}_3\text{O}_6$) and RDX ($\text{C}_3\text{H}_6\text{N}_6\text{O}_6$). As their composition indicates, they are composed of four basic elements: hydrogen, carbon, nitrogen and oxygen. Although many organic materials buried in soil are also composed of the same elements, use can be made of the fact that explosives have concentrations that are different than in soil and in most common organic materials. One way for the detection and identification of explosives is the use of neutrons. Several investigations were carried out on the advantages and limitations of neutron-based techniques used for the detection and identification of anti personnel landmines (e.g. Csikai, 1999, ElAgib and Csikai, 1999, Datema et al., 2000, Hussein and Waller, 2000, Kiraly et al., 2001, Brooks et al., 2004). The incident neutrons will interact with the nuclei of the major chemical elements in the mine (H, C, N and O), emitting elastically backscattered fast and thermal neutrons spectra which can act as fingerprints of the these chemical elements.

The elastically backscattered (EBS) neutrons can be detected by a suitable detector capable of differentiating the EBS neutrons according to their energy and their flux. The concentration of hydrogen, carbon, nitrogen or oxygen can be evaluated by calculating the elastically backscattered net relative yield of the neutrons and observing the different patterns of their energy spectra (Hussein, et al., 2005). One other detection approach based on neutron backscattering is to simply detect the hydrogen content in soil by measurement of thermal backscattered neutrons from hidden explosives. The change in hydrogen concentration in soil can be made by calculating the intensity of low energy neutrons reflected back from soil (Hussein and Waller, 2000). The chemical elements of interest for the detection of explosives require neutron sources of different energies in order to be observed.

Hydrogen is best observed through nuclear reactions initiated from very low energy neutrons. Other elements such as C, N, and O require neutron energies of several MeV to be observed at all. To satisfy this, the required neutron source should produce high energy neutrons for the detection of elements such as C, N, and O, and low energy neutrons for elements such as H (Vourvopoulos and Sullivan, 2006). Such a task can be accomplished with the use of a spontaneous fission source such as ^{252}Cf . The present work was done by considering a simple sample-source-

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detector geometry, simulating the elastically backscattered (fast and thermal) neutrons from the constituent elements of the explosive material and studying the variation in the net flux with the source energy spectrum. Such geometry was chosen in order to tally the EBS fast and thermal neutrons spectra.

MCNP Modeling & calculation procedure:-

Monte Carlo simulation of a land-mine localization device using the neutron backscattering method was reported by Datema et al. (2002). The general-purpose Monte Carlo N-Particle (MCNP) code, as described by the X-5 Monte Carlo Team (2003), was used in the present study. The code accounts for all neutron reactions given in a particular cross-section evaluation (such as ENDF/B-VI). The evaluated data are processed into a format appropriate for MCNP with the help of codes such as NJOY (MacFarlane et al., 1982). Continuous nuclear cross section data based on the ENDF/B-VI were used in the present computations. The net elastically backscattered neutron spectra were computed for the major elements of a landmine when buried in soil. Calculations were performed using a fixed point source with enough histories to have the statistical error less than 2% in all energy bins.

The sample-source-detector geometry used in the present study is shown in Fig. 1.

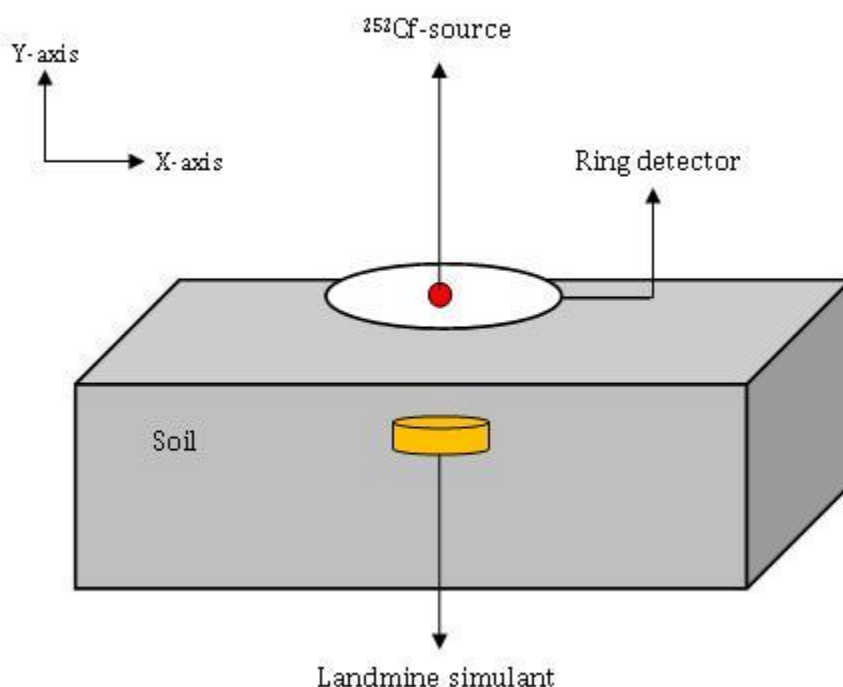


Fig 1:- Geometry used in the MCNP simulations.

The model consists of a soil of dimensions 200cm×150cm×100cm, with the sample as an explosive material in the form of a cylindrical cane TNT of 5cm radius×5cm height, buried 5cm deep in soil. A point neutron isotopic ^{252}Cf source was used in the study. The source was located at vertically above the soil at $y = +10\text{cm}$. Measured and normalized neutron spectra of ^{252}Cf employed in the calculations were taken from Griffith, et al. (1990). A ring detector with 10cm radius was centered on the ^{252}Cf source. This is tally the elastically backscattered (EBS) neutrons from the major constituent elements of the buried landmine. The net elastically backscattered neutrons spectra were calculated by subtracting the background spectrum (soil) from the signal (soil and sample). The calculations were carried out in duration of 100s.

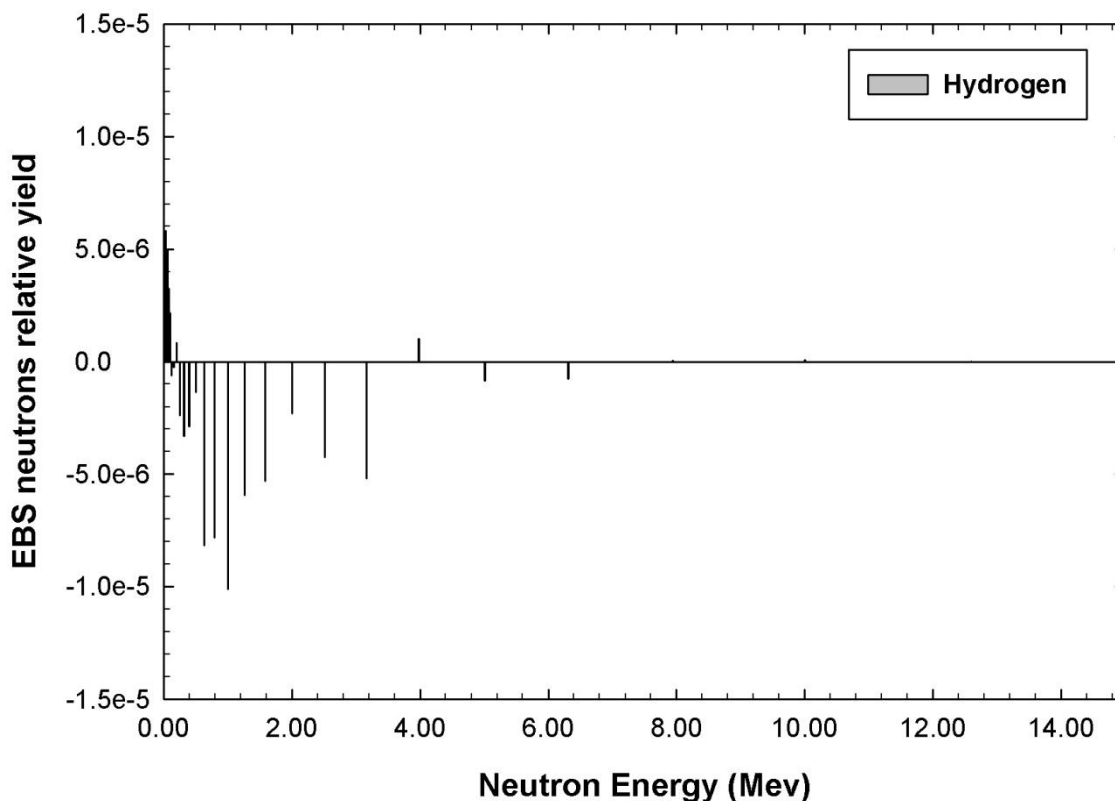
The composition of soil and TNT explosive as modeled in the MCNP simulations is shown in Table 1, with the data taken from Maucec and Rigollet (2004) and Hussein et al. (2005). Mass densities (in g.cm⁻³) of H (0.02), C (2.23), N (0.81), O (1.14) and Si (2.33), that were used in the MCNP simulations, were taken from the same sources.

Table 1:- Composition of soil and TNT explosive as modeled in MCNP simulation [8,11]

Material / Mass Density (g.cm ⁻³)	Elemental Mass Fraction / Mass Density (g.cm ⁻³)					
	H	C	N	O	Si	Al
Soil	0.0146			0.5520	0.3607	0.0731
1.12	0.016			0.618	0.404	0.082
TNT (C ₇ H ₅ N ₃ O ₆)	0.0217	0.370	0.185	0.4229		
1.65	0.0358	0.610	0.306	0.689		

Results and Discussion:-

The net EBS neutron energy spectra from H, C, N, O and landmine stimulant are shown in Figs. 2-6, respectively. Fig. 2 represents the net EBS neutron energy spectra from H. A few peaks of low energy elastically backscattered (LEBS) neutrons are observed in the low neutron energy range (0.022MeV -0.052 MeV). Negative indications are observed, because fast elastically backscattered (FEBS) neutrons are higher from the constituent elements of soil.

**Fig. 2:-** The EBS neutron energy spectra from H.

The net EBS neutron energy spectra from C are shown in Fig. 3. More relatively high peaks of LEBS neutrons are observed at the low neutron energy range (0.39MeV-0.64MeV). Compared with H, it is reasonable, since C is less effective as a moderator compared to hydrogen. Relatively high peaks of FEBS neutrons are also observed at the energies 1.58MeV, 2 MeV and 3.51MeV.

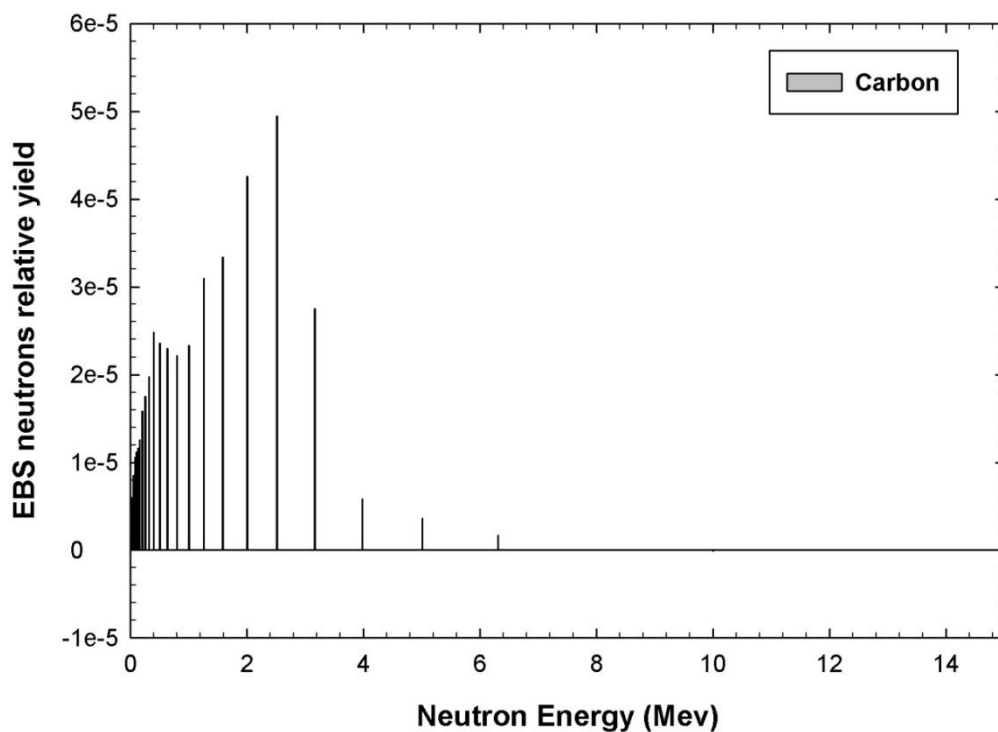


Fig. 3:- The EBS neutron energy spectra from C.

The net EBS neutron energy spectra from N are shown in Fig. 4. Intensive peaks of LEBS neutrons are observed at the low neutron energy range (0.022 MeV-0.64 MeV). Relatively high peaks of FEBS neutrons are also observed at the energies 1.21 MeV, 2.32 MeV, 3.35 MeV and 3.25 MeV.

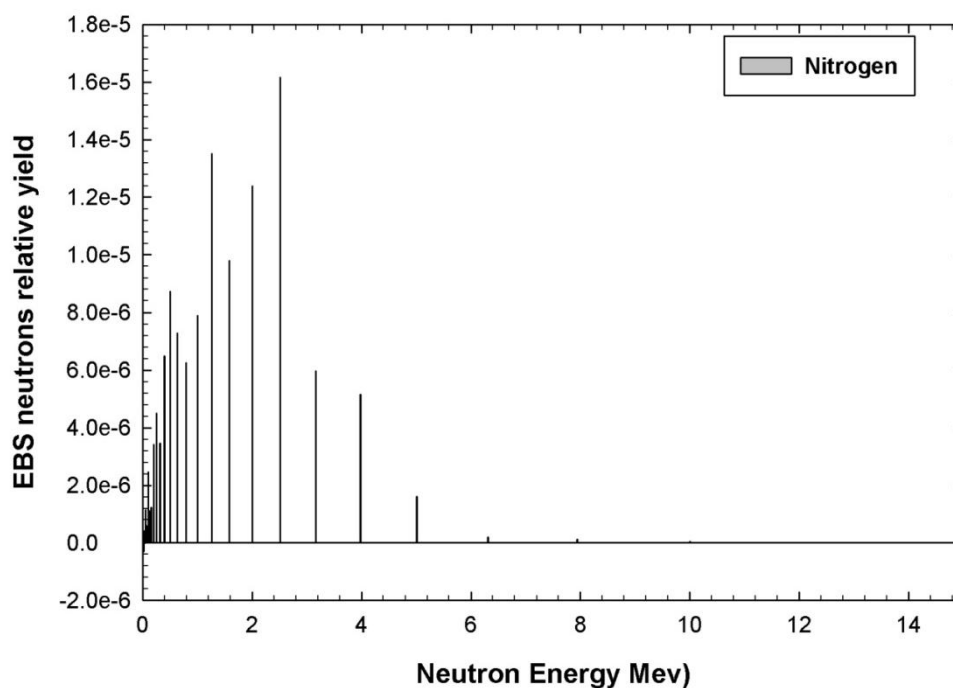


Fig. 4:- The EBS neutron energy spectra from N.

Fig. 5 shows the net EBS neutron energy spectra from O. A high peak of LEBS neutrons is observed at 0.64 MeV. Relatively high peaks of FEBS neutrons are observed at energies 2.33 MeV and 3.27 MeV

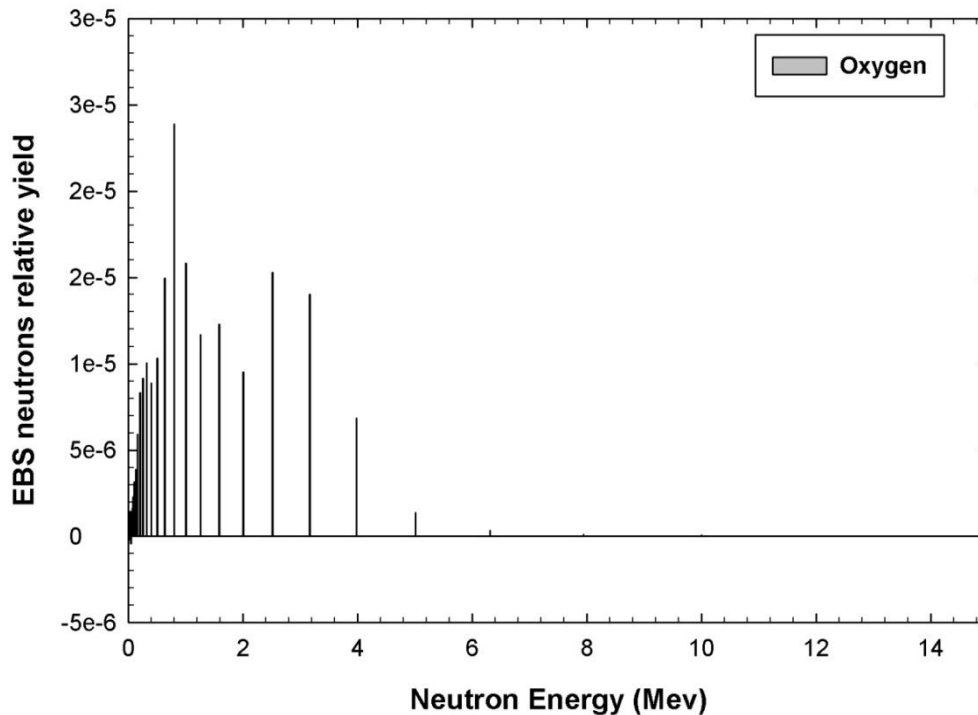


Fig. 5:- The EBS neutron energy spectra from O.

Fig. 6 represents the net EBS neutron energy spectra from landmine. Relatively high peaks of LEBS neutrons are observed at the energies 0.324 MeV, 0.531 MeV and 0.74 MeV. In addition, relatively higher peaks of FEBS neutrons are observed at 3.35 MeV. It is clear from the above results that the EBS neutrons spectra are spread throughout the slow and fast neutron energy regions. Effective detection of such neutron yields can be achieved by employing a detector capable of detecting both slow and fast neutrons.

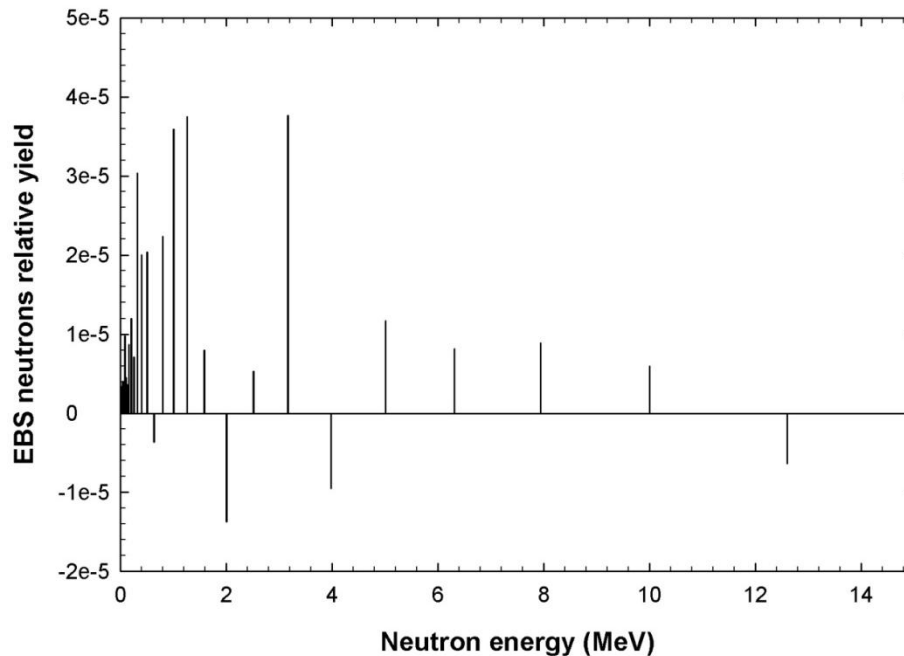


Fig. 6:- The EBS neutron energy spectra from landmine.

Conclusion:-

In the present work, Monte Carlo code was used to model a point ^{252}Cf neutron source centering a ring detector of 10cm radius. The energy spectra of the net EBS neutrons from the major constituent elements of landmine; H, C, N and O were explored. The EBS neutron energy spectra of fast and thermal neutrons of the major constituent elements of landmine stimulant have shown definite and detectable structures that can be used for the identification of a buried landmine.

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RESEARCH ARTICLE

CORRELATION OF ALT/AST RATIO WITH INSULIN RESISTANCE IN METABOLIC SYNDROME.

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Key words:-

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Abstract

Background: In recent years, non-alcoholic fatty liver disease is considered as a novel component of insulin resistance and metabolic syndrome, which is found to be associated with mild elevation of liver enzymes.

Aim of the Study: The aim of this study was to correlate the ALT/AST ratio with insulin resistance calculated by HOMA-IR (Homeostatic model assessment-Insulin resistance) method among patients found to have metabolic syndrome.

Design and Methodology: This was an observational cross sectional study which included 60 subjects of metabolic syndrome defined by International Diabetes Federation criteria. The relevant clinical examination and basic investigations were done. Fasting insulin levels were analysed by chemiluminescence method. Liver function tests were done by fully automated Analyzer Synchron CX-9 using Roche kits, calibrators and Randox control Sera. Data was processed and analyzed by SPSS version 17.0 software. The Correlation between ALT/AST ratio and HOMA-IR was assessed using The Pearson correlation test.

Results The ALT/AST Ratio was found to have positive correlation with HOMA-IR (Pearson's correlation coefficient is +0.742 and the significance was <0.001). This study shows that ALT/AST ratio can be used as screening tool of insulin resistance in the community.

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Introduction:-

The aminotransferases (transaminases) are sensitive indicators of liver cell injury. They include the aspartate aminotransferase (AST) and the alanine aminotransferase (ALT). AST is found in the liver, cardiac muscle, skeletal muscle, kidneys, brain, pancreas, lungs, leukocytes, and erythrocytes in decreasing order of concentration. ALT is found primarily in the liver and is therefore a more specific indicator of liver injury [1].

The aminotransferases are normally present in the serum in low concentrations. These enzymes are released into the blood in greater amounts when there is damage to the liver cell membrane resulting in increased permeability. The normal range for aminotransferases varies widely among laboratories, but generally ranges from 10-40 U/L [1].

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The liver plays an important role in maintaining normal glucose concentrations during fasting as well as postprandially. A number of recent studies have suggested that abnormal hepatocellular function is associated with obesity, insulin resistance, and type 2 diabetes [2], [3].

The metabolic syndrome is a condition characterized by a cluster of features including glucose intolerance, insulin resistance, abdominal obesity, atherogenic dyslipidemia, elevated blood pressure, a proinflammatory and a prothrombotic state [4]. It increases the mortality and morbidity due to cardiovascular diseases significantly. Insulin resistance is the key feature of metabolic syndrome. It means greater amount of insulin are required to elicit normal biologic response at a cellular organ or whole body level [4].

In recent years, nonalcoholic fatty liver disease (NAFLD), as a novel component of insulin resistance and metabolic syndrome (MS), has drawn the attention of many researchers. NAFLD encompasses a wide spectrum of liver diseases ranging from simple benign steatosis to steatohepatitis, fibrosis and cirrhosis. This condition which is associated with long-standing elevations in liver enzymes is related to higher risk of adverse cardiovascular events, oxidative stress, endothelial dysfunction, and MS [5]. Although not all patients with NAFLD are obese, obesity is considered the most important risk factor [5], [6]. Typically, liver biopsy is the ultimate test for diagnosis and delineation of the extent. It is usually not done except for situations of conflicting diagnosis. Liver ultrasonography although not sensitive enough to differentiate simple steatosis from more advanced hepatic involvement, is widely used [7]. Recently ALT/AST ratio has attracted great interest as potential novel marker of insulin resistance.

Aim of the Study:-

The aim of this study was to correlate the ALT/AST ratio with insulin resistance calculated by HOMA-IR (Homeostatic model assessment-Insulin resistance) method in metabolic syndrome patients.

Materials and Methods:-

This was an observational cross sectional study conducted among 60 patients of age ≥ 18 years with metabolic syndrome. This study was approved by the Ethical committee of the institution.

Inclusion Criteria:-

The study population included the adult patients of age ≥ 18 years who were diagnosed to have Metabolic syndrome based on following International Diabetes Federation criteria.

Waist circumference ≥ 90 cm in men and ≥ 80 cm in women with two or more of the following:-

Fasting triglyceride ≥ 150 mg/dl or specific medications.

HDL cholesterol < 40 mg/dl and < 50 mg/dl for men and women respectively or specific medications.

Blood pressure ≥ 130 mm systolic or ≥ 85 mm diastolic or previously diagnosed hypertensive or specific medications.

Fasting blood glucose ≥ 100 mg/dl or previously diagnosed type 2 Diabetes mellitus.

Exclusion Criteria:-

The following cases were excluded from the study:-

History of alcohol intake.

History of liver disease due to autoimmune, viral, genetic, drug induced causes.

Severe cardiac/renal/hepatic dysfunction.

Patient on insulin therapy for treatment of diabetes mellitus.

Methodology:-

A detailed history and examination of the study population were noted and anthropometric measurements, including height, weight, BMI, waist circumference and hip circumference were noted and the waist hip ratio was calculated (WHR). 10 ml of fasting venous blood sample was taken with aseptic precautions and distributed in EDTA, plain and fluoride containers. EDTA sample was used for complete blood count, fluoride vial sample for blood glucose and serum for estimation of Liver function tests, Kidney function tests and Lipid profile. Fasting insulin level in a plain test tube was analysed by chemiluminescence method. HBsAg and Hepatitis C were tested by Elisa method. Liver function tests, kidney function test and lipid profile were checked using fully automated Analyzer Synchron CX-9. Normal ALT and AST levels were considered to be 10-40 IU/L.

Insulin resistance is calculated by following formula using homeostasis model assessment of insulin resistance,

$$\text{HOMA-IR} = \frac{\text{fasting insulin } (\mu\text{U/ml}) \times \text{fasting glucose}(\text{mmol/l})}{22.5}$$

Statistical analysis of the study:-

Data was processed and analysed by SPSS version 17.0 software. Sample statistics i.e, mean, median and standard deviation were calculated for quantitative data. Chi-square test and difference of proportion were applied for qualitative analysis. Mean and standard deviation were compared by student t test. Mann Whitney's test and Kruskal Wallis test were used for non-normally distributed data. The Correlation between ALT/AST ratio and HOMA-IR was assessed using the Pearson correlation. Statistical significance was set at $p \leq 0.05$.

Results:-

68% of patients were from the group of age 50-69 years and 8% of patients were from the group of age ≥ 70 years and 23% from the group of age 30-49 years (table 1). The mean age is found to be 57.05 ± 8.83 years. The study population ranged from 36 years to 74 years. There were 30 males and 30 females. There was no statistically significant difference between the age distribution of males and females ($p=0.806$).

Patients who were both Diabetic and hypertensive patients (43.33%) were more in number compared to isolated diabetes (26.67%) and isolated hypertension (28.33%). The Blood pressure did not show any statistically significant difference among sex groups and different age groups but diastolic BP was significantly higher with people having higher BMI. The mean BMI was 29.44 ± 2.83 kg /m². Majority (93.33%) of study group were with BMI > 25 kg/m² and females were found to have higher mean BMI. 68.3 % patients among the study group had WHR ≥ 0.95 . The mean fasting Blood sugar among study group was 170.12 ± 36.37 mg/dl.

The mean ALT level among the study group is 51.45 ± 9.12 IU/L. The mean AST level among the study group is 44.18 ± 6.19 IU/L. The Study group is divided based on normal cut off as 40 IU/L for both the enzymes and it was found that 56 patients (93.33%) were found to have ALT levels ≥ 40 IU/L and 36 patients (60%) were found to have AST ≥ 40 IU/L (table 2). The mean ALT/AST Ratio among the study group is 1.173 ± 0.186 . The ALT/AST Ratio among the study group ranged from 0.69 to 1.60. 83.3% of patients were in the group ALT/AST RATIO ≥ 1 (figure 1).

The mean HOMA-IR level among the study group is 4.14 ± 2.27 . The HOMA-IR among the study group ranged from 1.23 to 11.28. The definite cut off of HOMA-IR to identify insulin resistance is not defined yet as the people in different areas of world also differs in their ethnicity and clinical background. On considering patients whose HOMA IR ≥ 2 as insulin resistant and HOMA < 2 as non insulin resistant, it was found that about 81.7% patients among the study group were insulin resistant.

The ALT/AST Ratio was correlated to HOMA-IR among the study group using Pearson Correlation test and it was found that Pearson's Correlation coefficient is +0.742 and the significance (2 tailed) was < 0.001 (Figure 2). Hence the correlation was found to be statistically significant. The correlation between ALT enzyme levels and HOMA-IR was also found to be statistically significant ($r = +0.58$ and $p < 0.001$).

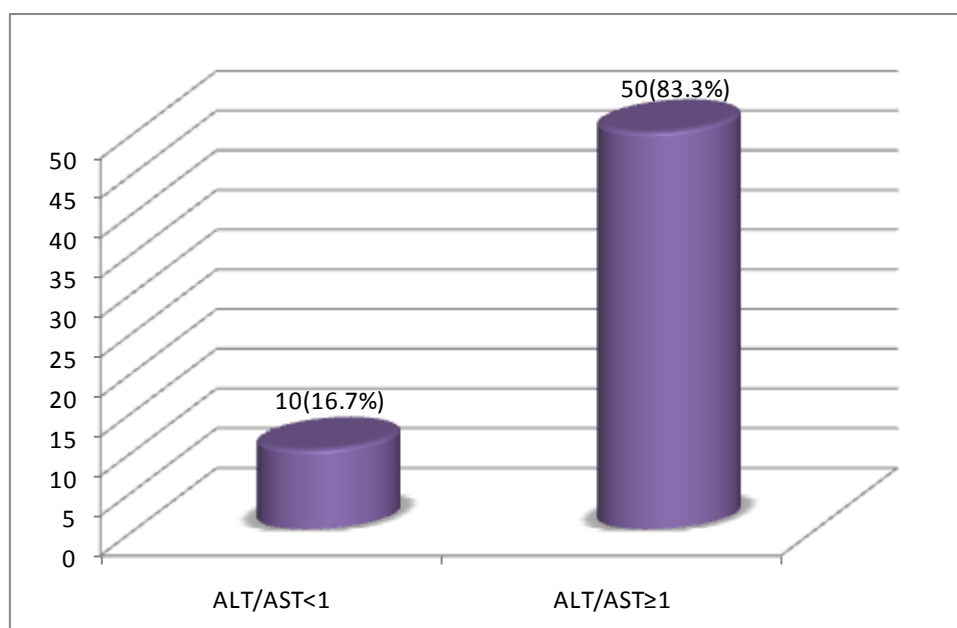
There was positive correlation between ALT/AST Ratio with fasting Blood Sugar and fasting insulin levels with significance (2 tailed) value < 0.001 . The ROC curve of ALT/AST Ratio is calculated in relation to HOMA-IR value as gold standard test (figure 3). Among the study group it was found that about 49 patients had HOMA-IR > 2 where as 11 patients had HOMA-IR < 2 . The area under the curve is 0.862 and it was found that the cut off of HOMA-IR value-1.06 has the sensitivity of 93.9% and the specificity of 81.8 % but the cut off of 1.07 has the sensitivity of 91.8 % and specificity of 81.8%. Since ALT/AST Ratio is easy to use as a screening tool of insulin resistance the cut off of 1.06 is considered better to achieve better sensitivity.

Table 1:- Age and Sex distribution of study group.

Sex	Age 30-49 years	Age 50-69 years	Age ≥ 70 years
Male	6(20%)	21(70%)	3(10%)
Female	8(26.66%)	20(66.66%)	2(6.67%)
Total	14(23%)	41(68%)	5(8%)

Table 2:- Frequency distribution of ALT, AST and HOMA-IR.

Parameter	Frequency	Percentage
ALT<40 IU/L	4	6.7%
ALT ≥ 40 IU/L	56	93.3%
AST<40	24	40%
AST ≥ 40	36	60%
ALT/AST<1	10	16.7%
ALT/AST ≥ 1	50	83.3%
HOMA-IR ≥ 2	49	81.7%
HOMA-IR <2	11	18.3%

**Figure 1:-** Distribution of ALT/AST Ratio among study group.

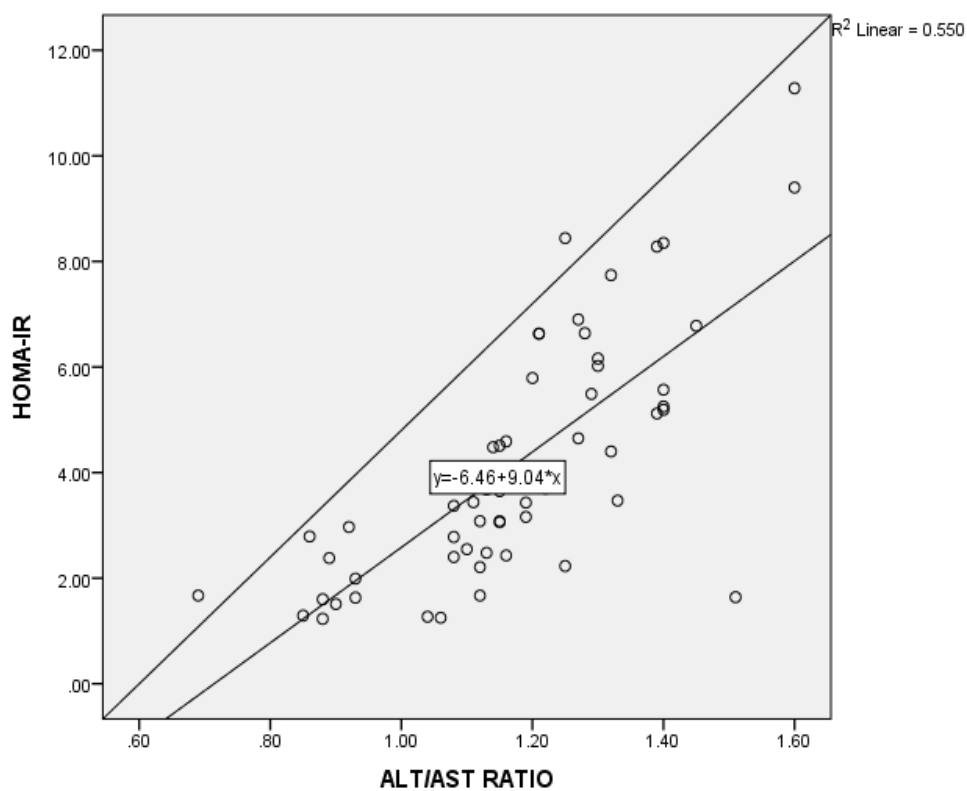


Figure 2:- Scatter Plot showing correlation of ALT/AST ratio to HOMA-IR.

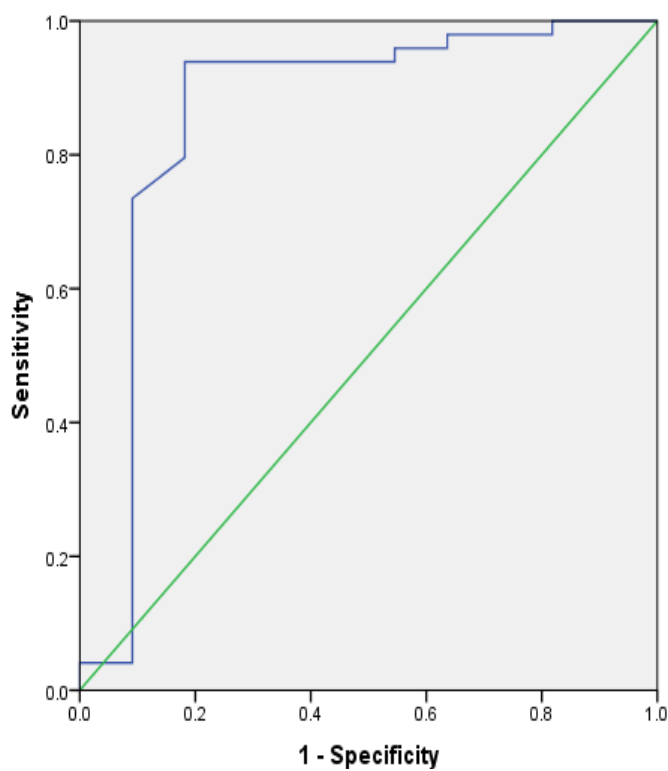


Figure 3:- ROC curve of ALT/AST ratio in relation to HOMA-IR.

Discussion:-

The mean age of this study population was found to be 57.05 years which is similar to study in Eastern India by Prasad et al [8] which showed that prevalence of metabolic syndrome increases with age and maximum number of patients were from age 50-59 years. Another study by Kumar et al [9] in Warangal, Andhra Pradesh also showed maximum prevalence was found in age group 51 -60 years with a mean of 52.8 ± 11.6 years. Deepa et al [10] conducted a study conducted in Chennai among metabolic syndrome patients defined by IDF criteria showed mean age to be 44 ± 11 years. Tan et al [11] studied metabolic syndrome among Asians which also showed increasing prevalence of metabolic syndrome and maximum prevalence was among age group 60-69 years (50.8%). Kanjilal et al [12] conducted a study among metabolic syndrome patients from Bangalore and Mumbai also showed maximum prevalence among the age group 50-59 years.

26 patients among our study group (43.33%) were suffering from both diabetes and hypertension and 16 patients were only diabetic and 17 patients were only hypertensive. There was one patient with impaired glucose tolerance. This shows that patients with both Diabetic and hypertensive patients were more in number compared to isolated diabetes and isolated hypertension. But Prasad et al [8] it was shown that hypertension is the most common abnormality observed among metabolic syndrome patients with about 89.2% prevalence.

The Mean BMI among our study group was 29.44 kg/m^2 and 93.33% of study group had BMI $>25 \text{ kg/m}^2$. BMI was significantly higher among females. Prasad et al [8] showed that 67% of patients of metabolic syndrome were of BMI $>25 \text{ kg/m}^2$. Garg et al [13] in their study observed that mean BMI was $28.1 \pm 2.1 \text{ kg/m}^2$ similar to our study. Kumar et al [9] observed mean BMI of $25 \pm 4 \text{ kg/m}^2$ in their study. Bhat et al [14] observed a mean BMI of $25.01 \pm 3.24 \text{ kg/m}^2$ in their study. Indulekha et al [15] observed a mean BMI of $25.6 \pm 4.2 \text{ kg/m}^2$ in their study. Deepa et al [10] observed a mean BMI of $25.7 \pm 3.1 \text{ kg/m}^2$ in their study.

The mean Waist circumference /Hip circumference ratio was 0.95 and the standard deviation was 0.046. It was found that 68.3 % patients among the study group had WHR ≥ 0.95 . In a study from Qatar among metabolic syndrome patients it was found mean WHR was 0.89 ± 0.09 . Deepa et al [10] observed a mean WHR of 0.94 ± 0.08 . Garg et al [13] in their study observed a mean WHR of 1.18 ± 0.1 in males and 1.21 ± 0.1 in females.

In a study from Texas by Hanley et al [16] it was shown that ALT, ALP and CRP were found to have positive association with various components of metabolic syndrome .It was also found that there was negative association between AST/ALT ratio to components of metabolic syndrome. In another study among Mexican Children with Metabolic syndrome it was showed that higher prevalence of elevated ALT compared with those without the syndrome (30.9% vs. 9.4%), as well as higher mean serum ALT levels (42.1 IU/L , 95%CI 33.4–50.7 vs. 23.9 IU/L , 95%CI 21.0–26.8), and higher prevalence of an AST/ALT ratio <1 (47.3% vs. 18.2%) [17]. Among Korean adolescents, a study by Park et al showed positive association of ALT with components of metabolic syndrome [18]. AST/ALT ratio showed positive correlations with HDL-C and negative correlations with BMI, WC, TG, and HOMA-IR in a study from Taiwan [19]. ALT levels were significantly higher in subjects with MS compared to those without MS in another study from China [20].

In our study, ALT/AST Ratio was found to correlate with HOMA-IR value which is calculated using fasting blood sugar and fasting insulin levels among metabolic syndrome patients (pearson's correlation coefficient is +0.742 and the significance was <0.001). The HOMA-IR cut off of about 1.06 had the sensitivity of 93.9% and the specificity of 81.8 %. At this cut off, Positive Predictive Value was 95% and Negative Predictive Value was 75%. Positive Likelihood Ratio was 5.16 and Negative Likelihood Ratio was 0.07. Hanley et al [16] in their study "The Insulin Resistance Atherosclerosis Study" showed that elevated ALT was independently associated with insulin resistance by HOMA-IR method. In another study from China it was found that obesity and elevated ALT are significantly associated with insulin resistance by HOMA-IR method [21]. In a study among Japanese population [22] ALT/AST ratios were independently and significantly associated with HOMA-IR and the optimal cut-off point to identifying insulin resistance for these markers yielded ALT/AST ratio of ≥ 0.82 in non-obese subjects and ≥ 1.02 in overweight subjects to identify insulin resistance which is very similar to our study. In another study from China, both ALT and GGT concentrations were linearly correlated with HOMA-IR and independently associated with HOMA-IR [23]. To the contrary, in a study by Esteghamati et al, both Serum ALT and AST were significantly higher in patients with MS [24].

Conclusion:-

ALT/AST Ratio was found to have positive correlation with HOMA-IR. Fasting blood sugar and fasting insulin, ALT enzyme levels were also found to have positive correlation with HOMA-IR. The best cut off of ALT/AST Ratio to identify insulin resistance was 1.06 with sensitivity of 93.9% and the specificity of 81.8 % among study group. 93.3% of patients with metabolic syndrome had raised ALT and 83.3% patients with metabolic syndrome had ALT/AST ratio >1.

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RESEARCH ARTICLE

LEXICAL-SEMANTIC FEATURES OF TRANSLATING IDIOMS FROM ENGLISH INTO UZBEK

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Abstract

The manuscript deals with investigation of lexical-semantic specifics of translation of English idioms into the Uzbek language with maximum possible preservation of primarily semantic, lexical and morphological content. The author presents all possible ways of an adequate translation of lexical-semantic meanings and images, mechanisms and principles of translation of English idioms into the Uzbek language. Theoretical part of the manuscript is proved by numerous examples of English idioms and their Uzbek equivalents or analogues.

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Methods and Materials:-

Methodological ground of the present investigation covers English idioms and their possible Uzbek equivalents or analogues; theoretical issues in the aspect of theory of translation. The main issue among them is Uzbek linguist in the aspect of translation Ganisher Rakhimov's book printed in the Uzbek language "Tarjima nazariyasi va amaliyoti" ("Theory and practice of translation"). – Tashkent, 2016. – 176 p. In this book the author proposes modern methodology and principles of adequate and correct translation of English idioms and proverbs into the Uzbek language. The present investigation is based on his concept of translation. The following methods were used in investigation: descriptive method, componential analysis, comparative method, method of lexical translation and other methods.

Results and Discussion:-

There is an important role of language, its lexical-semantic functions and artistic issues in esthetic translation. By using both effective and efficient proportion means of the language, translator can form adequate and equivalent ways of artistic translation. Hence, translator should be able to have deep life experience, emotional and logical observation knowing and understanding emotions clearly as writers. The most important part is, by knowing the secrets of cultural and mentality, keeping nationalism of the translation work, translator can create other cultural environment. Otherwise translator can describe cultural and national importance of the translation which readers can't get other foreign culture. Thereby, there is an important role of understanding adequately lexical-semantic significance in artistic translation. In this case there is a great interest determine the principles of translation of English idioms into Uzbek.

Idioms are simulation word phrase unit, it applies semantic unit than basis of interior construction and it demonstrates the functionality of the language¹. That is the language is not based on certain models, it uses

¹ Amosova N.N. The basis of the English phraseology. – Leningrad: Leningrad State University, 1963. – P. 24.

repeatedly every time and applies as an integral part of the language. In spite the changes of the idioms in interior construction; it can't affect its main features. So, idiom is semantic structure. Idiom includes its care and periphery. The care is invariable semantic part of idiom (mare's nest-nonsense, spiel the beans-reveal a secret). This part is usually changes its real meaning. In some cases the real meaning of idioms are stored (husband's tea-very weak tea, dressed up to the nines-dressed to perfection). Idioms are linguistic reality of the language, its semantic richness and colorfulness, the diversity of its potential units. Usually idioms can be translated in such cases as:

- Idioms aren't compatible by meaning and content of the expressions in translated language
- Idioms are partly appropriate by the meaning in expressions in translated language, even though they haven't adequate meaning and equivalent.
- Idioms are full logically and content suitable in expressions.

In both translated language lexical opportunities are refused. The aim of refusing is, to make a clear total solution. The aim of the solution is, find a translation of the word which can be reflect the both culture of the language. For example, there are specific sides of the word "time" in English. Beside linguistic, semantic and grammatical features, it has psycho-linguistic features of the word "time". For instance, there is an "Indian time" and "European time" concept in India. And they have different meaning. If they organize some event according Indian time, participants can come at any suitable time for them, it isn't violation of ethical norms if they are late, but in European time they must be on time. So, time concept has special features in translation. Time concept is logical and semantic differs from Uzbek language. Searching semantic unity of the concept will require efforts. So, in artistic translation each word of original text is rich with its meaning. Opinion idiom is a structure of a word unit. Concentrated in this part of the idioms is necessary to understand fully the idea. Each idiom has exact, clear and concise meaning. By the metaphor way of ideas many idioms are clear and concise. Poetic vivid expressions are used to express the idea is called figurative expression.

In order to translate idioms we need to understand and reform figurative expressions. Figurative expressions in idioms reform by the help of epithet, metaphor, allusion, assimilation, repeated, antithesis and contra-position, the most important part of idioms and its translation is to understand clearly figurative expression and to create its duplication in other language. For instance, *to give smb the finger*² – *birovning qo'ynini puch yong'oqqa to'ldirmoq*. But in idiom it is clear as to give present smb the finger. In fact, we can't give finger itself to smb. By this, imaginative content of artwork is found. Giving nothing but promise to give smth situation based on the metaphor of the idiom will be suitable and adequate in translation. That's why this idiom translated as "*birovning qo'ynini puch yong'oqqa to'ldirmoq*" into Uzbek.

Adequacy of meaning takes a great role in translating idioms. Adequacy of translation is to clarify the real meaning of the text and to save the rule of the language. The translation of idioms refers to the adequacy of the content, repeat and rebuilds it. Because, by translating English idioms into Uzbek, it is difficult to save language rules. Because they aren't refer to two similar types of languages.

According to G. Rakhimov, to find analogue to an idiom means to find a similar expression in the translating language which has full or partial semantic meaning: *Business before pleasure*. – *Mehnat, mehnatning tagi rohat*³.

The ability of the unit of speech expresses the function of another speech unit is called the equivalent of the language. In this case the unit of speech can be suitable for both languages. Equivalent idioms mean essentially the same unit in the same language and live in the conditions of the same conditions. Using and expressing the translation of idioms in the same way is called the equivalence of idioms. The main unit of speech in translation isn't a word, we call it syntagma. Syntagma is a group of word meaning that is used in relation to understanding the basis of semantic-syntactic combination. That's why, in idioms translation we use a lot of variation than in-variation.

In translating the idioms we need to pay attention to the following features:

- Translating the text should carry out on the basis of original text, work with it and know it as a best way in translation.

² Longman dictionary of English Idioms – Oxford, 1979. – 1st edition. – P. 312.

³ Ganisher Rakhimov. Tarjima nazariyasi va amaliyoti. (Theory and practice of translation). – Tashkent: "Uzbek national encyclopedia" State publishing house, 2016. – P. 127.

- Using actively metaphor, assimilation, part of artistic language such as laconism, allusion, associative, adequacy and equivalence in translation.
- To occupy the properties of the original authors technique and try to give this technique by translating the text.
- Saving social status and mental characteristics of English describe the characteristics which are expressed through the language option and translate it into Uzbek. (For example: in English there isn't a word "sen" so while translating we translate it as a "siz" "you").
- Taking into consideration of the idea and literary method of the author, when giving the meaning of the translated work.
- To observe the full text of the original work reproduce of the author's style, rhythmic repetition of the original text circumstance is the effective use of the art possibilities of language.
- The peculiarities of translation language of the work are to keep heroes language as a mean of creating artistic image and use.
- The wealth of Uzbek language, to know Uzbek fables and proverbs deeply, and to use opportunities of the Uzbek language effectively⁴.

Conclusion:-

To follow the rules of proportionality and compatibility, equivalence and adequacy, association and observance of phraseology in translation the idioms into Uzbek we need to pay attention to the following features:

1. Clarity. To pay attention to the author's idea. Learning and thinking deeply different shades and variations is important than meaning itself. Specific manifestation of meaning and colorfulness plays an important role in translation of idioms. Relationships between author's artistic and aesthetic search for the author's style of instruction. Not to violate linguistic and the meaning of the original text.
2. Laconic. English language avoids different kind of reality, pathos and luxurious. In translation especially translation of idioms the main idea and linguistic unit gives by laconism. To avoid a word which expresses different meanings.
3. Clarify and distinct. Each of the text is hidden special meaning. Meaning wears a dress of words. This dress may be shine and luxurious or it may be clear, right and unique. English language is such language which can describe these clear ideas. Clarify and distinction of idioms helps to identify full idea of meaning and to give it readers. To understand the meaning clearly in translation is very important. In order to come one solution words must be analyzed, systematized and collected. Therefore, the translation language should be not far from author's work, on the other hand idea expresses simply, smooth and clearly.
4. Full possibilities of the literature language in translation. Works will be created on the basis of the norms of the literary language and grammar. English psychology, on the other hand Uzbek psychology describes clearly in translation. Translation reflects the two linguistic norms as a result of their strict observance of the two languages. These rules are shown in translation of idioms. Because in translating idioms stylistic, semantic-linguistic and meaning is more important than linguistic request.
5. Analogue. Between English and Uzbek language linguistic social and cultural relations are carried out. At the same time English and Uzbek language is in one language group, but by grammatical meaning they are differ from each other. Except this, learning all possibilities of both languages and finding Uzbek analogues to English idioms. For example: *daryodan tomchi – drop in the bucket*.
6. Find out number of stable and adequate idioms. Translating idioms we should pay attention meaning adequacy. The adequacy of each idiom relies on imaginative of the content. Each idiom represents the most efficient use of language, its wealth and requires the use of language effectively.
7. To use a variety of methods. Complex of variety method helps to be proportionate of idioms. To find out analogues, adequacy and equivalence of meaning, to provide identification of the meaning, anatomic translation and using stylistic complex is important in translation.

Consequently, artistic translation is targeted to clear aim it has many unique and unplanned features. To understand these specific features involved specific people in translation.

⁴ Rasulov R. Umumiy tilshunoslik. (General linguistics). – Tashkent: Fan, 2007. – P. 63.



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RESEARCH ARTICLE

REVIEW PAPER ON YAWNING DETECTION TO PREVENTS ROAD ACCIDENTS

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Abstract

For the safety purpose of drivers yawning detection is very important. There are many important applications of yawning in a drivers fatigue detection, humans well behavior, driving behavior and understanding the language of tongue disability person. It is observed that heavy vehicle drivers keep going continuously driving without giving a frequent rest period. Considering above applications, an automatic detection of yawning is one important system component. Uptil now yawning detection system does not satisfy the real time requirement which is having a high computational complexity and does not satisfies the challenges like facial obstruction, ease of implementation, accuracy and safety. Uptil now there is no any idea suggested or presented on vehicle side hardware. In this paper we will be reviewing hardware of engine start or stop control by using microcontroller.

In previous, for yawning detection two different algorithms are used such as for face viola-Jones and mouth detection contour activation algorithm. Previously presented systems are in position to recognize other persons faces while finding the biggest face in all of the frame that we are interested and other faces also considered; hence face search time is not optimized. By studying these disadvantages of traditional yawning detection method possible improvement algorithm is proposed.

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Introduction:-

In earlier years the amounts of road accidents have been suddenly increased. These accidents because of continuous driving ability, not getting proper sleep. Especially in the transportation industry the heavy vehicle drivers keep continuously driving without giving a rest period. Due to this most of the accidents are occurs. Because of this it has become an area of great socioeconomic concern. Driver Fatigue is one of major reasons causing most fatal road accidents around the world.

The global status report on road safety 2013 shows that survey or information from 182 countries, calculated about 99% of the world's population. The report shows that around the world the number of deaths is due to the road accidents are high at 1.24 million per year. About 3,400 people die every day in world due to road accidents. Tens of millions of people are injured or disabled per year. Cyclist, pedestrians, children and the elderly are the most susceptible of road users. So it is very necessary to design such a prototype system which prevents road accidents by detecting driver's inattention, driver's drowsiness and giving a audio sound when yawning is detected. In this paper

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the aim is to develop a prototype system which detects yawning to prevent road accidents. This system includes a video camera either externally attached or the inbuilt camera in laptop which extracts video to detect face and mouth features. This system is non intrusive real time monitoring system. The programming for this is done in MATLAB 2013a using the image processing toolbox.

The purpose for so much attention towards drivers yawning detection is that suddenly increases road accidents and for the people's safety. Once fatigue has been detected, a number of activities we can performed for the safety purpose of person who drives the vehicle, mainly producing the persons alert voice, fluctuations in steering wheel or persons chair, showing text, providing large amount of oxygen for person himself.

There are many other applications for yawning detection is helpful in to be concerned with home health care technique. That proves yawning is one important method to detect the people's health information and modify their lifestyle. There are also more applications for yawning detection such as operator attentiveness to determining whether the operators such as heavy machines, nuclear reactor controls and monitors, and air traffic controllers in critical system is paying attention or not. Another application is that the system used in to determine the communication purpose of human being with language defect, particularly to detect false estimation.

For all of the above systems that require a natural investigation of involuntary intake, the value of technique is more important to create it cost effective. For that purpose VBM is very co-operative. VBM is a technique where a camera or optical sensor captures the photo from various scenes. Later this photo is used in process of yawning detection through manipulation component. This camera and manipulation component is called as intelligent camera. Because of this, VBM technique is most popular and useful now a day. Today the VBM, studied a experimental answer for the operations like investigation of persons natural or well being characteristics, for example facial expression or self propelled functional technique. Yawning detection system has number of important applications such as drivers fatigue detection, drivers behavior detection and also other automotive applications to prevents road accidents. Driving is a most common activity in people's everyday life. So it is very necessary to make driving safe for the peoples. The main reason to discover yawning detection method is significant increases the probability of car accidents. Most of the yawning detection system does not satisfy the real time requirement have a high computational complexity. So now a day's very few yawning detection system uses in a luxury cars to prevent car accidents. They do not have a sufficient accuracy. There are three main reasons why existing system is not used in our car system is

1. Computational complexity
2. Facial obstruction and
3. Lighting conditions

The main goal of this yawning detection method is lower the computational complexity and make this system safer to the person by studying real boundaries and can designing in actual intelligent cameras.

Literature Survey:-

In 2008 author described drivers fatigue detection based on trailing the jaws and to survey on controlling and recognising the involuntary intake. Author explained a technique to trace and trail the mouth cascade of classifiers is applied also the yawning images. After locating the mouth from face images using classifiers the SVM technique is applied to distinguish jaws also for to detect yawning then alert fatigue.

This is the non intrusive real time monitoring system. Many researches deal with yawning detection uses the geometric feature of the mouth techniques, but there are some disadvantages of that technique that is the left and right mouth corners are important feature point .But in this lip position are difficult to detect at that time the lips move, then lip detection makes more difficult. Also, geometric features liable to pose and have more difference for individual. This paper author used cascade of classifiers to train and trail with the help of viola-Jones. Then SVM is used for data classification.

After studying this paper we have conclude that, because of using SVM (support vector machine) computational cost is reduced and provide better generalization. There is disadvantage, two algorithms is used, hence increases computational time and this viola-Jones technique is not efficient to run in a real time on smart cameras if implemented as above that using cascade of classifiers [1].

In 2009 author describes to locate and tracking the drivers mouth using two CCD cameras. This is the real time monitoring system. Camera A is fixed; it is used to locate drivers head position. Camera B is movable or controllable, which is used to locate the mouth and extract the mouth information. In this first collecting all the videos from camera A and using image processing algorithm locate the drivers face. Then second with the help of face position in camera A shoot driver head position all the time by sending command to camera B through serial port. Then to detect and track driver's mouth in camera B, use haar- like features. Then at last by comparing the ratio of mouth height and width, yawning is detected.

After studying this paper we have concludes that, through this method resolution ratio of driver's mouth is higher than single camera. Also the feature information is higher accuracy. There is some disadvantage of this method, once the position between the driver or two cameras is changed, and then the relation between angle control volume and pixel control volume is also changed. So this technique is not intelligent, it needs some future work [2].

Authors Dr. Sheifali Gupta, Er. Garima describes a prototype system for detection of driver's drowsiness to prevent road accident. This system is non intrusive real time monitoring system. This system detects facial features and extracts video with the help of a video camera either externally attached or inbuilt camera in laptop. Eye closure and yawning is detected followed by alert. In this programming is done with the help of MATLAB 2013a with the help of computer vision toolbox.

After studying this paper we have concluded that, the aim of this paper is to prevent the road accidents by detecting eye closure and involuntary intake of drivers. A 1.3 Mp video camera used to capture the real time videos and for processing. In this the vision based system is used because of its accuracy and non intrusiveness. Only the disadvantage is that, a night vision camera is not used to capture the images in dark or dim light to increase the usability [3].

Narender Kumar, Dr. N.C. Barwar Presented eye blinking and yawning detection system in real time for monitoring drivers drowsiness. This system is the non intrusive real time monitoring system. In this system if the drivers eyes are remains closed for certain period of time and his mouth remains open to yawning then driver is said to be drowsy and then alarm is sounded. In this the detection of facial features viola Jones algorithm is applied and for mouth detection contour activation algorithm is used to find the contour of the lips.

After studying this paper we have concluded that, this system was tested in different light conditions for different subjects that are without eye glasses, with eye glasses, without moustache and with moustache. In case of females there is also same set up, programming, webcams for night and late experiments. Without eye glasses eye blink rate detected more accurate. Only the disadvantage is that different algorithm is used for face and mouth detection, so not easy to implement and uses OpenCV method in that considering the biggest face we are interested, also other faces are considered, hence face search time is not optimized [4].

In 2016 article author proposes yawning detection is important application in driving behaviour monitoring. In this author used to either faces or jaws detection very efficient algorithm that is viola-Jones algorithm. Later used back projection theory to determine yawning. The author used in yawning detection method for implementing and testing smart embedded camera is used. This is the real time monitoring system.

After studying this paper we have concluded that, by comparing existing method this system gives correct yawning detection rate. In this the camera is installed on the dash board of the car or in front mirror. The result of face and mouth detection also the involuntary intake or tiredness alarm indication is display on monitor. Only the disadvantage is that the author is not address the challenge of lighting condition, safety, facial obstruction. They only implementing up to detection of yawning, there is no further implementation on hardware side for safety purpose. They only improve computational complexity [5].

Algorithm used:-**Face detection algorithm**

Comparing to previous system this yawning detection method is explaining the structure of a quick or recapture, well organized face investigation algorithm known as viola-Jones algorithm. Firstly in face investigation method, each and every information given by prepared facial characteristics of every lump can be removed or reserved mainly five different folders in intelligent camera for maintains the calculating period. After some time for the investigation algorithm this folders which are answer of prepared facial characteristics are applied. As related to that same direction, this controlling method uses store costs rather than preparing or making division or using whole photo for searching facial characteristic at starting.

Every folder has above sets of information.

- a. Facial characteristic point
- b. Facial characteristic costs
- c. Facial characteristic load
- d. Perform division

In the face investigation, for viola-Jones algorithm it is possible nothing but since the different persons are multiple face dimensions, rather since the people have capability to near or taking out against the camera in related period, according to bigger or shorter face dimensions. For that purpose facial characteristic is necessary to measured.

One of important advantage of this yawning detection system, by comparing the Open CV method of viola-Jones, this method ends detecting the different face later on searching the primary face of every structure that is needed. Specially face with original topic, so remaining faces not studied. Hence face investigate period is saved. The face which is any time nearest to the camera this face is investigate primary in this method. The other persons faces is not considered and they are not large than the original persons face.

The diagram 1 shows instances for the vehicle riding situation, in that persons face and mouth investigate correctly. Also, for storing prepared facial characteristic in folders is help straightly through camera and ignoring prepared face investigation, the effect of this method is gains the rate of face investigation step and the capability of controlling method.

Mouth detection Method:-

After detecting the face, the down partial face is selected for the goal detection area. This is the identical method like a face investigation. Later on searching the largest mouth in structure all the information nearest to mouth area and dimension send towards yawning detection.

Fig 1:-Persons face and mouth searched between different peoples.

Fig 2:- Yawning sequence.



Yawning detection Method:-

The last part is yawning detection. The image is color image, so it is necessary to convert it into the gray scale image. The back-projection theory is used for that purpose.

After conversion of that grayscale photo the design take care not any involuntary intake in the structure. This is searched by comparing black and white elements. For that two rules are necessary to fulfilled.

- Relation between black element in present folder and in relating folder should be larger than starting point.
- Relation between black element in mouth area and white element in area surrounding to the mouth should be larger than second starting point.

Suppose these situations are fulfilled, according the design investigate specific folder shows involuntary intake and that procedure is copied for all of the later folders of motion picture.

Proposed Work:-

After studying literature survey and understanding need globally these papers are modified and a novel technique is build called yawning detection.

Below fig 3 displays the projected figure of yawning detection method. In this system the camera is installed in front of mirror or dash board in the car. MATLAB is used to build mathematical model of this system. In this viola-Jones algorithm is used for detecting face and mouth of driver. Viola-Jones algorithm is already installed in MATLAB. In hardware side microcontroller PIC16F877A is used along with other peripherals. PIC is family of Harvard architecture microcontroller made by micro chip technology.

Hardware side foot acceleration in that rack and pinion mechanism is used, which converts the rotary motion into the linear motion. Then amount of force is coming is passing on to the potentiometer, then that force is read on to the ADC.

The microcontroller understands only digital logic that is either 0 or 1 also incoming voltage is AC; hence it needs to convert that AC voltage into DC volt. So ADC is used. The engine ON and OFF is depend on servo motor and in servo motor PWM that is pulse width modulation wave is inbuilt.

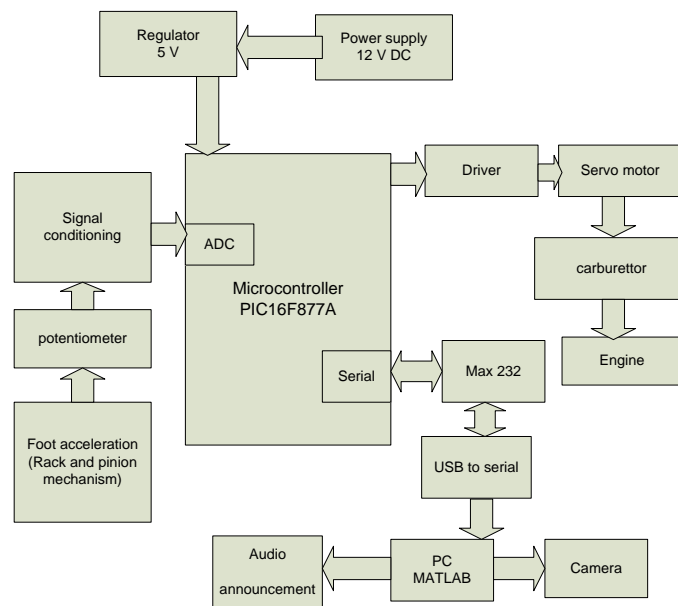


Fig 3:- projected yawning detection Method.

Conclusion:-

As we discussed until now yawning detection system, this system gives better accuracy, safety, easy implementation and reliability, reduce complexity, and increase efficiency. The previous papers have some disadvantages like no any implementation on vehicle side hardware and biggest candidate is the final detected mouth. In this paper we have reviewing hardware on PIC microcontroller and image processing part on MATLAB. For face and mouth detection this system monitors the condition based on delay and then takes decision. We will be reviewing hardware of engine start or stop using microcontroller. Also we will be taking care of acceleration of vehicle.

In car intex smart camera is placed in instrument panel, also in the foreground of the mirror. All the actions performed by the camera are running on embedded based system with help of this yawning detection method. To create yawning detection method act on a computationally limited platform & address the problem of lighting condition, facial obstruction much effort was made in allocating or improving algorithm and codes to act in actual period. Also do not need any complex hardware.

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RESEARCH ARTICLE

IMPACTS OF FLOOD AND ITS MANAGEMENT – A CASE STUDY OF BIHAR.

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Abstract

Floods are most recurring and frequent natural hazard in India causing enormous loss to life and livelihood, and damage to infrastructure. In India, Bihar is one of the worst affected states due to floods. As per one estimate, in Bihar flood affects about 6.880 million hectares of land out of about 9.416 million hectare, which is about 73.06%. It not only affects the infrastructure but also the socio-economic life in the state. So, there is a need to minimize negative consequences and ill effects of flooding by means of flood management. In Bihar, most of the times, the decision makers go for structural measures like construction of embankments, flood retention walls, flood levees and channel improvements, etc. However, it is felt that structural measures itself is not sufficient to reduce the adverse impacts of floods in the state. Hence non-structural measures like flood plain management policy, building bye-laws, flow and silt management policy are also required. It contemplates the use of flood plains judiciously, simultaneously permitting vacating of the same for use by the river whenever the situation demands. The purpose of this paper is to highlight the severity of flood impacts on development of state and to discuss the possible flood management measures in context of Bihar.

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Introduction:-

Floods have been a recurrent phenomenon in India causing huge losses to lives, properties, livelihood systems, infrastructure and public utilities affecting the development of the region. Bihar is one of the States of the India bounded by Nepal in the north. Main drainage system of the state, the River Ganga flow eastward direction in a stretch of 432 km across Bihar and divides the state in two unequal halves. The plains in north of the River Ganga are drained by two major rivers, the River Kosi and the Gandak (Fig.1). In addition to this there are several smaller systems such as the Adhwara group of rivers, the Bagmati, the Bhutahi Balan, the Burhi Gandak, the Ghaghara, the Kamala, and the Mahananda. All these rivers mainly originate in Nepal from the Himalayas whereas the River Kosi also includes its catchment in Tibet. Thus, the rivers of North Bihar mostly share basins outside the country in Nepal and Tibet. The water carried from the part of catchment lying in Tibet also passes through Nepal. To fulfil the requirement of fuel for domestic purposes and reclamation of area for occupational needs, there is an increasing deforestation in Nepal which further causes degradation in vegetative cover in the catchment areas. Due to this practice, soil has been increasingly getting eroded from these areas. So the rivers carries significant sediment load from the upper part of catchments. These sediments in addition to inadequate carrying capability of the rivers cause congestion in natural drainage leading to inundation and floods. So, flood is a state of hydrological extreme of high

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water level in a stream channel or on bank that results in inundation of land that isn't ordinarily submerged. Generally it occurs in event of heavy rainfall and becomes a hazard once it causes loss to the lives, livestock and livelihood. "The trend shows that the intensity and frequency of floods in Bihar Plains have increased over the years causing human suffering and destruction of agriculture, habitation and infrastructure" (NIDM, 2007).

The National Flood Commission, set by Government in 1976 put the country's flood prone area at about 40 MHa out of total area of 329 MHa. This report, thus, revealed a rapid increase in flood proneness in just over a decade. Most of these area falls under Ganga and Brahmaputra basins. The most flood prone basins are those of the Ganga and Brahmaputra in Uttar Pradesh, Bihar, West Bengal and Assam. These five states are the most flood prone. But the commission analyzed the share of damage went up from 25 to 50 percent of the total and chronically flood prone area in case of Bihar.

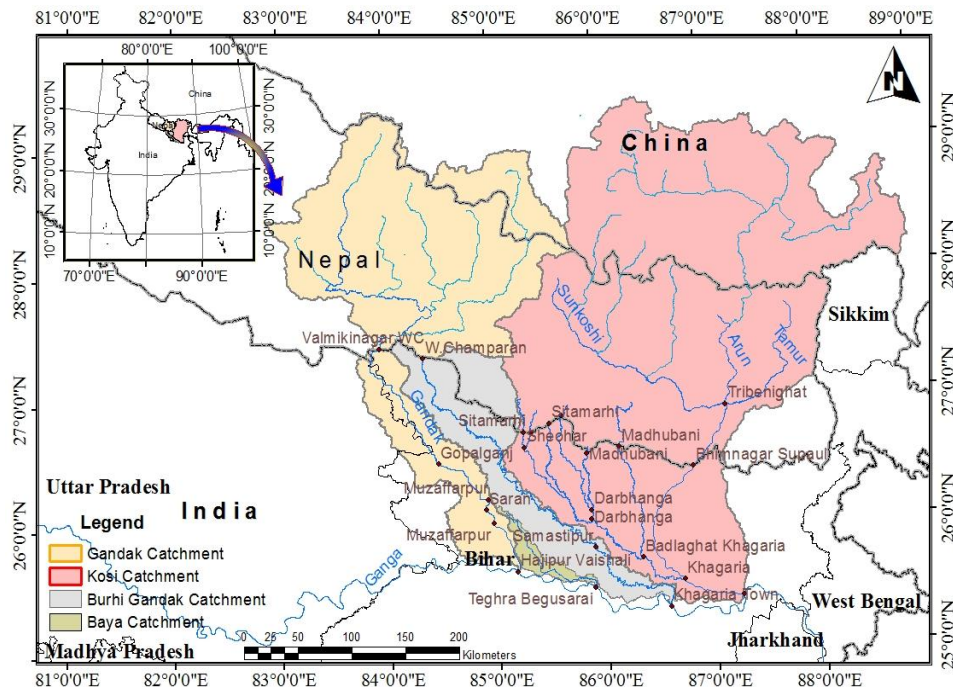


Figure 1:- Map Showing Catchment of Major Rivers in North Bihar

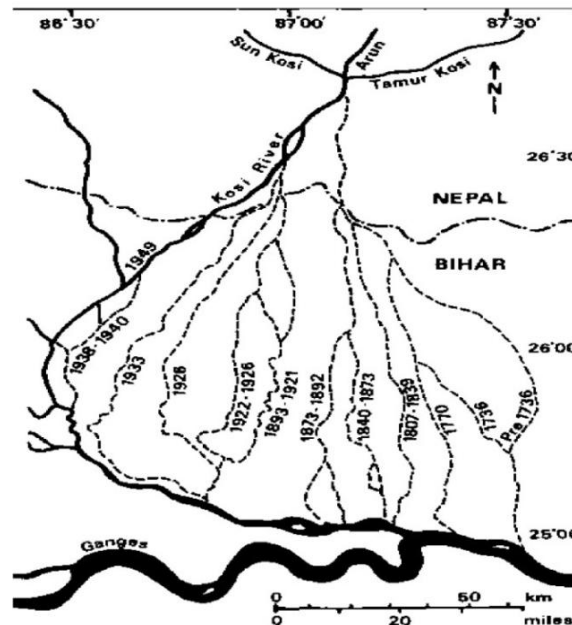
Three fourths area and population of Bihar lie in the natural flood plains of the Ganga and its Himalayan tributaries. About 6.880 out of 9.416 million hectare comprising 73.06 percent of the state is flood affected which accounts for approximately one-sixth of the flood prone area in India. Geographically, northern Bihar is prone to flood as major rivers flowing through North Bihar originates from Nepal and the Tibetan region of China. About 65% of the catchment area of Bihar's rivers fall in Nepal and Tibet and only 35% of the catchment areas lie in Bihar (Sinha et. al.).

The upper part of catchment is liable for the morphological activities within the downstream reaches. It carries huge sediment load with its flow every year which is responsible for change in morphological behaviour of the stream. The matter of the flooding is implicitly guided by sedimentation in river. This sedimentation has gradually reduced the capability of the river to drain extreme flows leading to flooding within the plains.

The variation of minimum and the maximum flows in the Himalayan Rivers is significantly high. This causes huge seasonal variation in river discharge. In addition to this terrain of North Bihar is almost flat having elevation at Indo-Nepal border ranging from 90-95 m whereas elevation at confluence of these rivers in Ganga is of the order of 40-45m.

So, Vulnerability to flood of Bihar lies in its flat topography, wide variation in flow in Himalayan rivers, excessive rainfall levels (more than 1200 mm annually), annual rainfall concentration during only three monsoon months and high sediment loads. The plains of Bihar and adjoining Nepal are drained by rivers that carry high discharge and very high sediment loads, which then fall out onto the plains of Bihar. "Gradients vary from 22 cm per km near the

Indo-Nepal boundary to 7.5 cm per km near the confluence of the rivers with the Ganga main stem” (Sinha et. al., 2012). The flat terrain and the major seasonal variations in water volume in the rivers cause extensive flooding in the northern plains. In addition, as the rivers reach the plains and lose momentum, they begin to meander. Rivers like the Kosi are notorious for changing course. “As per the records it is evident that during 1736 to 1953 the river Kosi has shifted its course through a distance of approximately 112 Km. towards westward” (Kumar D., 2015) (Fig.2). Further Kosi embankment was constructed with a series of spurs on both banks confining the course of river Kosi.



(Source: Kumar D., 2015)

Figure 2:- Kosi River Course.

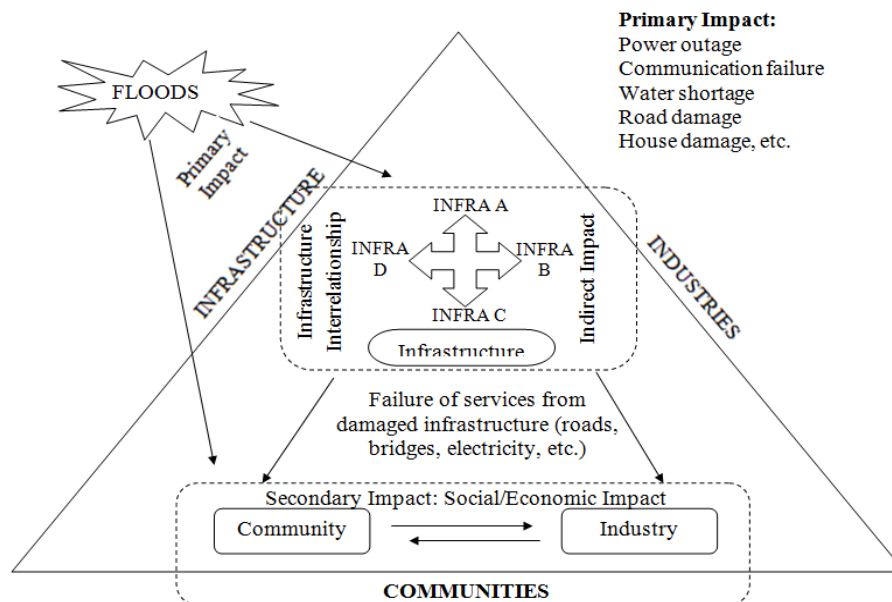
The embankments on both sides spaced five to sixteen km apart have confined the lateral shift of the river within the embankments; however flooding is still a challenge in the area. Human intervention in the flood plain is also making this problem more acute.

Impacts of Flood:-

Flood impacts can be broadly divided into primary impact that occur due to contact with water like loss of life, livestock and property, damage caused to infrastructures like road, bridges, railways, disruption in services like electricity, telephone lines, water supply lines, etc. and secondary impact that occur because of the flooding, such as disruption of services, prolonged health and malnutrition impacts. Secondary impacts cause indirect losses and its ill effect continues on-long time scales and sometimes may be more important. These include prolonged health impacts due to failure of water supply and sewage disposal system, shortage of food and other essentials due to failure in transportation network. A frame work of flood impacts is shown in fig.3.

Infrastructure networks sustain various activities and services of the communities and industries. Due to failure of services from damaged infrastructure economic activities in the region are also poorly affected. It may lead to partially or completely shut down of industries and business establishments causing reduced production followed by adverse impact on economic growth. There may be some long term impacts like sediment deposited by flooding may destroy farm land for coming few years leading to cultivator and labours economically vulnerable. Based on the assessment of losses in monetary values, they are termed as tangible if can be assessed in terms of monetary values and if not then termed as intangible losses. Intangible losses are generally higher in less developed region because of the more consequences of flooding especially in case of frequent and devastating floods.

“Impacts of flood depend on maximum depth and duration of flooding, the extent of inundation in the floodplains, velocity of flow and rate of rise of flood levels” (Gautam et al.). Flash flood causes immediate and huge impacts, whereas gradually occurring floods allows much time for evacuation and protection of properties.



(Source: Deshmukh, A. (2010))

Figure 3:- Framework of Flood Impacts.

Flood Losses in Bihar:-

Almost every year floods severely damages property, both movable and immovable, destroys standing crops and food grains and badly cripples the infrastructure in Bihar. The loss of life and limb caused due to flood events cannot be compensated. However in monetary values, it costs several crores every year. The flood brings untold miseries to the people as they have to leave their damaged houses and spend a long time in relief camps, raised platforms or in temporary shelters. Government has to allocate huge amount for rehabilitation, and relocation of people which requires diversion of capital required for maintaining production. Table 1 shows the extent of flood loss in Bihar for last 15 years.

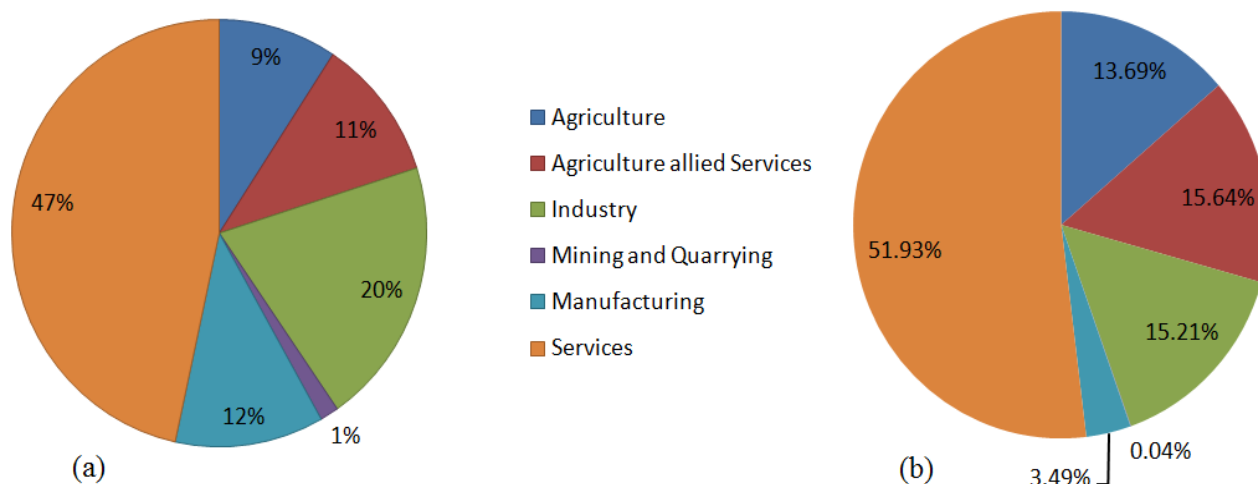
Table.1:- Flood losses in Bihar from 2000 to 2015.

Year	River which caused flood	Number of Affected					Affected Area		House Damaged		Loss	
		Dist.	Block	Vill.	Human	Animal	Agric	Non-Agric	Fully	Partly	Live	Live-stock
		(Nos.)			(in Lac)		(Million ha.)		(Nos.)		(Nos.)	
2015	Gandak, Kosi	8	32	237	4.850	0.140	0.0090	0.0030	157	12	27	1
2014	Gandak	20	116	2018	30.000	1.000	3.6000	0.3000	3235	2977	158	34
2013	Gandak, Kosi	20	126	4810	72.340	5.600	1.6800	0.6900	9344	117572	253	6480
2012	Gandak, Ganga	11	28	277	2.400	0.080	0.0968	0.0100	322	151	15	0
2011	Ganga	25	166	3577	71.430	6.627	3.5420	0.2877	25840	59342	249	183
2010	Kosi	9	44	679	7.180	3.570	0.1602	0.0390	6187	8983	32	0
2009	Bagmati, Gandak	16	91	1546	22.030	1.346	0.1710	0.9339	3867	3807	97	2
2008	Kosi	18	116	2585	49.952	12.166	0.6405	0.2119	104965	151461	258	878
2007	Gandak, Kosi	22	269	18832	244.420	27.130	1.3323	0.5510	369360	414968	1287	2423
2006	Bagmati, Ganga	14	63	959	10.890	0.100	0.1520	0.0297	9218	9419	36	31
2005	Bagmati, Gandak	12	81	1464	21.040	5.350	0.3343	0.1261	1031	4507	58	4
2004	Gandak, Bhutahi	20	211	9346	212.990	86.860	2.0990	0.6010	361017	568756	885	3272
2003	Gandak, Ganga	24	172	5077	76.020	11.960	0.9943	0.5140	43163	41261	251	108
2002	Bagmati, Kamala	25	6	8318	160.180	52.510	1.4446	0.5244	89021	329993	489	1450
2001	Gandak, Kosi,	22	194	6405	90.910	11.700	0.9042	0.2910	128419	93589	231	565
2000	Kosi	33	213	12351	90.180	8.090	0.6570	0.1476	183670	159421	336	2568

(Source: Disaster Management Department, GoB)

Flood Impacts on Growth of Bihar:-

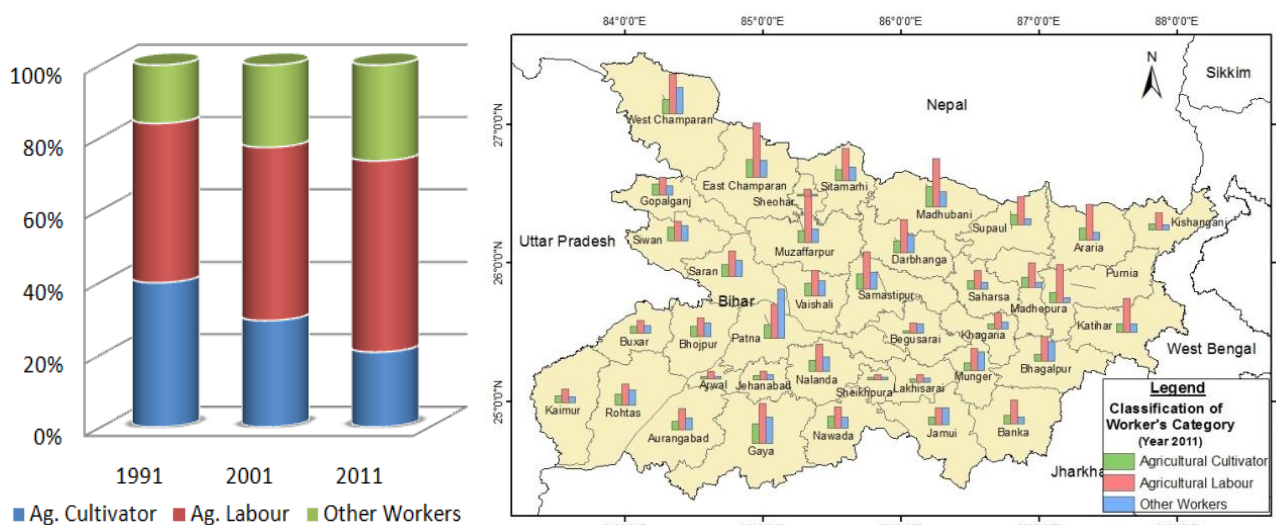
Bihar is one of the states in India whose economy largely depends either on agriculture or in agricultural activities. It's share in Net State Domestic Product (NSDP) of Bihar is 29.33% which is considerably higher in comparison to contribution in national GDP which is 20.06% (Central Statistical Organisation, Govt. of India, 2014) (Fig.4(a) and 4(b)).



(Source: Directorate of Statistics and Economics, Bihar)

Figure 4:- (a) Share of various sectors in India's GDP (b) Share of various sectors in NSDP of Bihar.

Beside this fact, about 78% of population are involved in agriculture and its allied activities either as cultivator or agricultural labour (Fig.5(a)). According to government of Bihar 73% of the farmers are small and marginal farmers, who not only cultivate their land but serves as agricultural labour. The situation is more vulnerable in the region of North Bihar with large flood prone area (Fig.5(b)). So, agriculture is the backbone of Bihar's economy and socio-economic status. Hence sustainable development of Bihar principally depends upon agriculture sector.

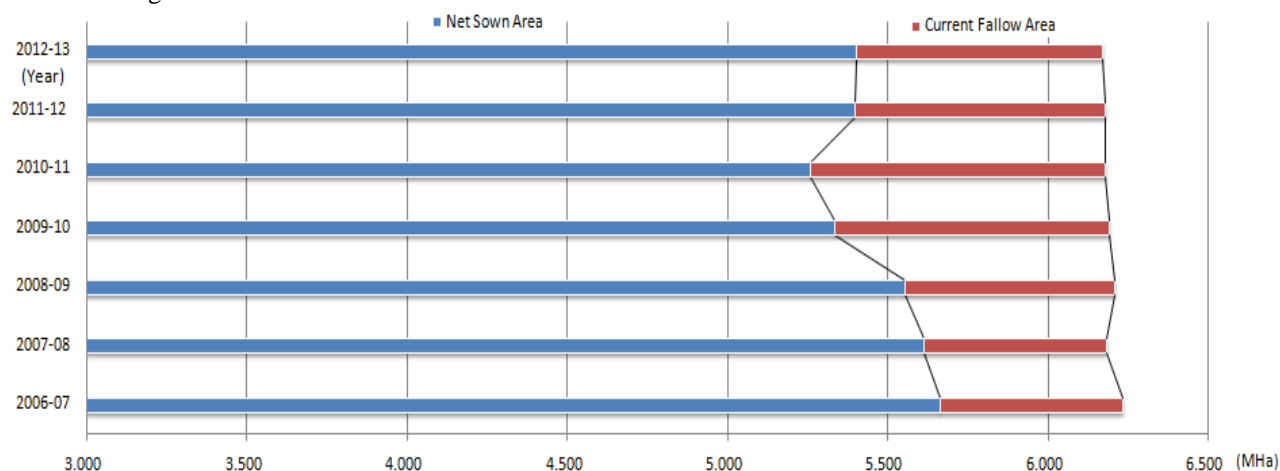


(Source: Directorate of Statistics and Economics, Bihar)

Figure 5:- (a) Classification of workers category in Bihar (b) District wise proportion of various workers category

In occurrence of flood, hectares of agricultural land get submerged and silted, which makes it uncultivable for few consequent years and remains as fallow field (Fig.6). A significant increase in fallow land can be visualised after 2008-09 which is the consequence of Kosi flood in year 2008. The area became uncultivable due to high silt deposition in the farm land. Several hectares of lands gets eroded and grains preserved by households gets destroyed which results into many small and marginal farmers becomes landless. Women from vulnerable families work as

agricultural labour but due to flood this activity is completely stopped. Fig. 7 summarises the direct physical impacts of floods on agriculture sector.



(Source: Directorate of Statistics and Economics, Bihar)

Figure 6:- Yearly variation in fallow area in Bihar

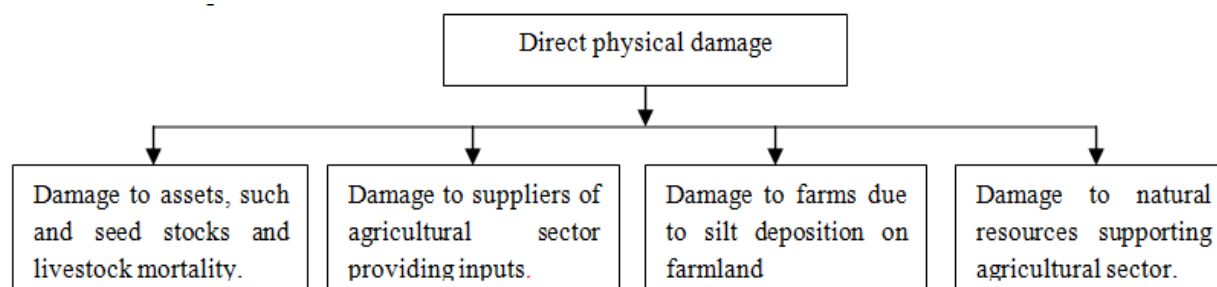


Figure 7:- Direct physical impact of flood on agricultural sector.

Generally the small cultivators as being on marginal level, takes agriculture inputs on credits from the local input dealers but due crop failure they are not in a position to re-pay these amounts. Further they do not remain in the position to manage for the next year and migrates in search of livelihood. This ultimately affects the agricultural production in consequents years also. Such adverse impacts of flood on agriculture badly affect the economy of state and its growth. These ill effects are shown in fig. 8.

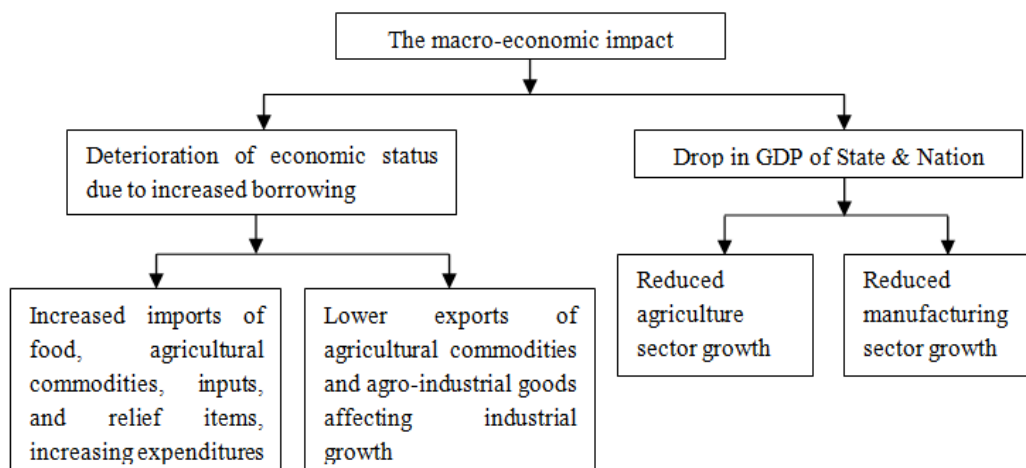
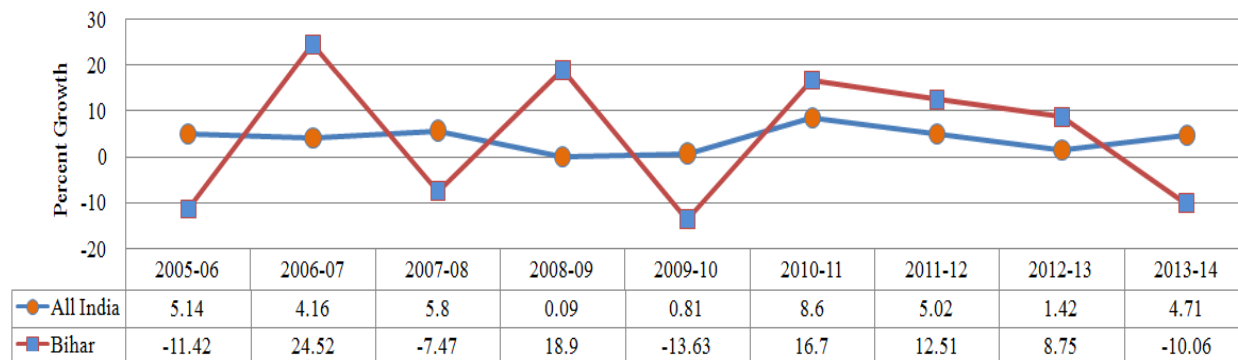


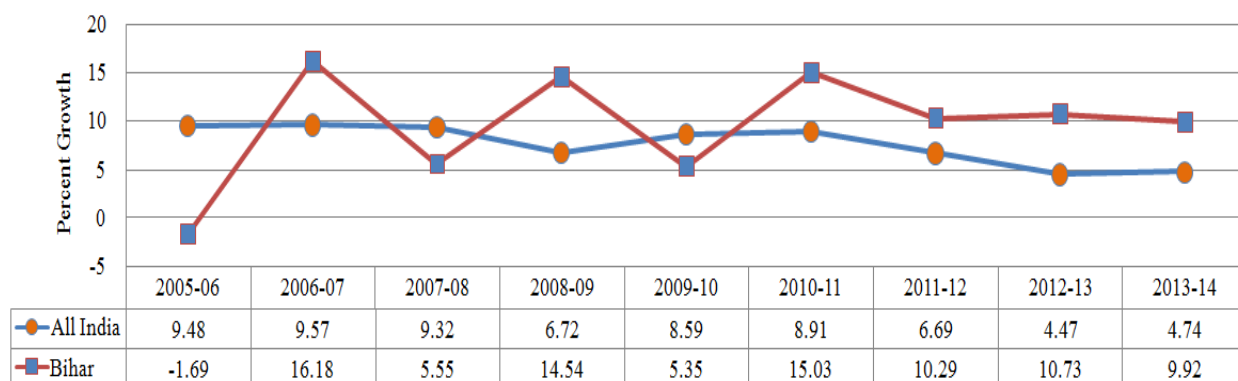
Figure 8:- Economic impact of poor agricultural growth.

Due to recurrent flood events the miserable growth rate in agriculture and its allied services in Bihar for last one decade can be seen in fig. 9(a). As agriculture is the mainstay of Bihar's economy so poor growth in agriculture sector directly affects the GSDP of the state. Comparisons of Growth Rates (GNP of India & GSDP of Bihar) are shown in fig.9(b). The overall GSDP of Bihar shows almost similar trend as that of GSDP in agricultural and allied sectors.



(Source: CSO, 2015)

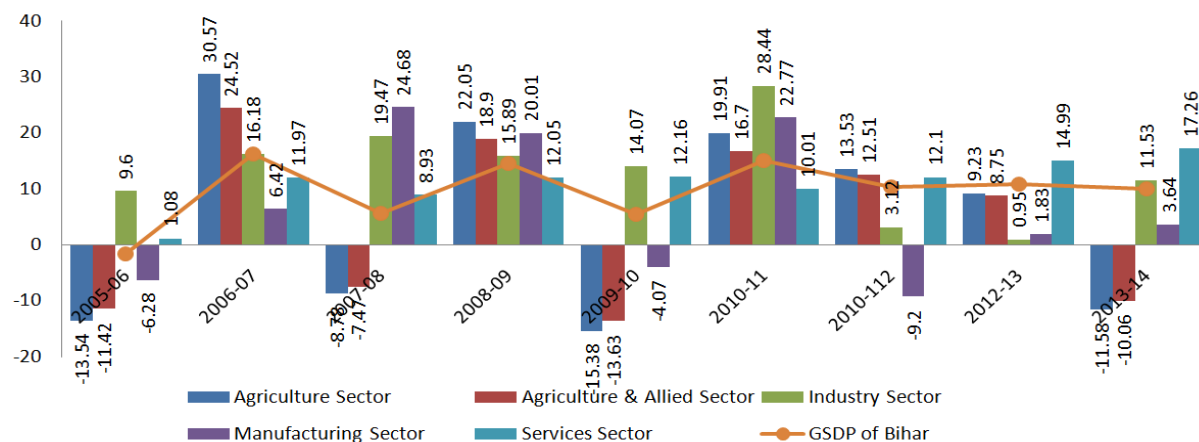
Figure 9 (a):- Yearly Growth Rate (National and Bihar) in Agriculture & Allied Sector



(Source: CSO, 2015)

Figure 9 (b):- Yearly Growth Rate (GNP of India & GSDP of Bihar)

Next to agriculture sector which is contributing to NSDP of Bihar and equally affected by negative impacts of floods are industrial and manufacturing sector. These sectors are closely linked with infrastructure like power supply, water supply, communication, etc. In case of any disruption in these services, the production will be affected immediately. In event of flood, these services are affected primarily causing its consequence as temporary stoppage of production due to lack of electricity or water, disruption of raw material availability and inflow, temporary unavailability or shortages of labour, etc. During the flood, the industrial sector may confront damage that may include buildings, equipment and machinery, stocks of raw materials and of manufactured goods. In addition, after flood the sector may face changes in its production flows, including production decline and possible higher costs of production arising from the destruction of assets or other causes such as lack of raw materials, strategic inputs of water and electricity, temporary absence of labour, lack of working capital, etc. Furthermore, some branches of industry – such as the food producers – that rely on the production obtained from agricultural sectors for raw materials may suffer production declines even if no damage has happened to their facilities. In case of damage to building, equipment and machinery there is a need of funding to ensure returning the sector to its normal level of functioning or production. Fig. 10 shows the percentage growth or decline in various sectors in Bihar. Positive peak refers to potential of state whereas the negative fall shows the impact of flood during the year and its consequence. So flood is playing a vital role in miserable growth rate of state. Observation on growth rate in various sectors in last decade shows the highly unsustainable pattern.



(Source: Directorate of Statistics and Economics, Bihar)

Figure 10:- Yearly Percentage growth rate in various sectors.

Flood Management in Bihar:-

In the above context, to minimize negative consequences and ill effects of flooding to maintain the sustainable growth of the state, there is a need of planning for flood management. As floods cannot be controlled completely so planning for flood management implies management of flood in such a manner to get maximum benefit in the given circumstances. Flood management measures can be broadly categorised in two types, (i) structural measures involving like construction of embankments, flood retention walls, flood levees, channel improvements, detention basins, and other such structural components whereas (ii) non-structural measures includes land use planning, redevelopment of flood prone areas, flood insurances, watershed management, flood forecasting, disaster mitigation and preparedness, etc. Both the measures are not mutually exclusive rather complementary to each other should be taken as integrated approach for best and effective result.

Structural Measures:-

In Bihar, mostly flood management works consists of constructing and maintaining the embankments (Fig.11). It comes under the category of short-term structural measures.

Construction of flood embankments:-

These are very popular structural measure adopted in the state not because of their efficacy but due to being cheap and quicker to construct. By March, 2013, a total of about 3732 km of embankments has been constructed and 1555 km is under construction which is scheduled to be completed by 2017 (WRD, Govt of Bihar, 2014).

Channel improvements:-

The existing embankments are under increasing pressure due to rise in the bed level of rivers due to heavy silts brought by the rivers from its upstream catchment area which lies in Nepal. It has been observed in Gandak that during the past three decades, the rise of bed level had been of the order of 1.8 meters approx. This type of aggradations necessitates perpetual raising and strengthening of the embankments. But this raising river bed levels, decreases their carrying capacity and causing drainage congestion in the countryside. To overcome such issues, method of improving the natural drainage or rivers by dredging and de-silting is a better option. It re-develops the carrying capacity of channels and enables it to flow at safe levels and within its banks. But this measure is not generally adopted due to its high cost of execution and others associated problems.

Embankment protection works:-

Embankment protection works are broadly classified in two categories, (i) Anti-erosion works and (ii) Flood-fighting works. Anti-erosion works are carried out during the period of January to May every year for the restoration of embankments damaged during the flood period (monsoon months) due to excessive discharge in rivers and construction of any additional embankment protection structures like spurs, aprons, flood levees. Flood fighting works are carried out during flood period on need basis to protect the embankments during flood events. These are of emergent nature.

In addition to above, it has also been observed so many times that creation of appropriate flood storage in reservoirs and detention basins will provide the long-term solution for the flood problems in the area. No such dam has been constructed either in Bihar or in Nepal on any of the tributaries of the Ganga which may provide flood moderation in the region. The major issue in this regard is there is no suitable site available in Bihar for dam building on any of the north Bihar Rivers as they originate from Nepal.



Fig 11:- Embankment and embankment protection works as structural flood control measure.

Hence, it is felt that structural measures itself is not sufficient to reduce the adverse impacts of floods in the state. There is need to adopt non-structural measures too as a judicious mix for effective flood management practices.

Non-structural Measures:-

But, losses in recent years contemplate that structural measure alone can't solve the problem. So it is felt necessary to adopt non-structural measure along with structural measures as an integrated approach to efficiently mitigate flood impacts. These non-structural measures includes,

Land-use planning:-

Use of land in flood prone area should be given consideration to master plan or governing laws. The land on a stream should be used for parks and recreational purposes, moreover as for ecological reserves. Infrastructures like washroom, picnic facilities etc. should be made flood proof. Private developments like golf courses may be brought up. The necessary purpose here is to integrate the land-use coming up with for flood-prone lands into the broader plans for the urban and surrounding space.

Zoning of flood-prone lands:-

To reduce flood damage in future the effective way is to prevent development in flood prone lands. So the best approach is zoning of such land, however the land use planning as discussed should be adopted so the land contains an outlined use. Zoning can be used to scale back damages from flooding and must be so flexible to acknowledge that different types of land use are compatible (Fig.12).



Figure 12:- Zoning of flood prone lands.

As in flood prone areas having water velocities are low and not causing serious erosion, agricultural use of lands is acceptable. Particularly in case of Bihar, where agricultural land is limited so in view of self sufficiency in food supply, flood prone lands can judiciously be used as agricultural lands.

Redevelopment of flood-prone areas:-

A major flood disaster can be taken as opportunity to correct past planning errors. Removal of flood-prone development as well as conversion of the land to a defined and restricted use is an option to be considered. It should be more cost-effective in long run to physically relocate flood-prone development as a part of a disaster reduction programme.

Compensation and incentives:-

Compensation is provided to victims after the event of disasters. It helps them to restore their living up to some extent. But as part of disaster help, compensation must have a goal to reduce the future flood damages. So, instead to just paying for damage, the funds should be centred on flood proofing, buyouts and relocation to avoid recurrences of such events. Similarly, incentives should be developed to encourage flood proofing and relocations from flood prone areas. It should be supported through cost shared program having shares of government and owners as well.

Flood Insurance:-

Flood insurance works on the policy of distributing the losses among the wider segment of society. Peoples living in flood plains should be made aware regarding the flood insurance. The authority is needed to advice the peoples of the requirement of flood insurance and its benefits. Then in case of flood disasters, the losses of the affected peoples will be paid from the accumulated premiums of policy holders. So, flood insurance can be adopted as an important measure to reduce the flood impacts and its losses. “*The Pradhan Mantri Fasal Bima Yojana*” is an initiative in this regard but needs to be implemented more effectively and for all assets like livestock and livelihood in affected areas.

Silt management:-

The Lower reach of river Kosi and its contributing tributaries are severely affected floods every year. As the Kosi with its tributaries like Adhawara group, Bagmati and Kamla Balan originates from the hills and reaches to plains of Nepal Terai and Bihar, their flow velocity drops significantly. It results in increase of depth of flow and hence silt carrying capacity reduces. Due to reduction in water carrying capacity, silts get deposited (fig.13) at river bed resulting rise of river bed and consequently erosion of banks. The mouth of the channels oftenly gets obstructed and hence shifting of river course takes place taking new area under threat of flood. This development is additional attributed causing increase in the water level that ultimately results into overtopping and breaches of embankments and flooding within the basin. Therefore it needs watershed management with an effective silt management policy to resolve such issue.



Figure 13:- Silt deposition in Kosi River at Saharsa.

Flood forecasting and warning:-

Flood forecasting and early warning system is a predominant non-structural flood protection measure. On the basis of mathematical modeling, experts can convert the past information like discharge, stage and inundation area from data base to present on the basis of rainfall, moisture and snow cover. For small catchments time lag between rainfall and peak flood is very small leading to flash floods. So quantitative precipitation forecasting and installing of radars is advisable in such areas. A framework for flood forecasting and warning system is shown in fig.14. In

case of large rivers, it allows sufficient time for respond. Accurate forecasting is necessary as well as its formulation into reliable warning and effectively disseminated to the communities at risk to take necessary steps and reduce losses.

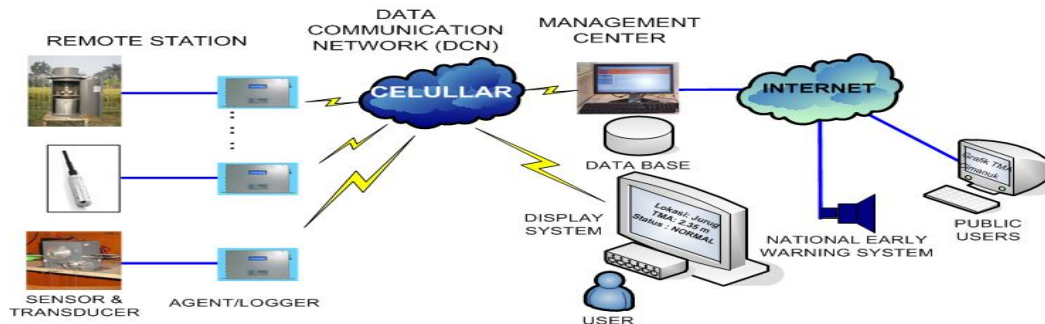


Figure 14:- Framework for flood forecasting and warning system.

Conclusion:-

Flooding is a perennial problem in the state of Bihar with devastating impact on the livelihood of people. The effects of damages to life and property caused by flooding were severe for several years. The damage to infrastructure also badly affects the industry sector. Agriculture is the mainstay to our large growing population and the back bone of state's economy playing primary role in its development. However, due to recurrent floods in the state, agriculture and allied sectors have accounted miserable growth rate but still play a vital role in the development of Bihar. It has been observed that flood events badly affect the agriculture sector and the GSDP of state in almost similar fashion. Majority of rural population belong to the lower socio-economic strata of the society. Small and marginal farmers, landless labour, and petty traders dominate the rural economy of the state, which suffers from extreme poverty conditions due to recurrent losses due to floods. In spite of the government's measures of flood management, households continue to live with suffering on an account of severe damage to their life and assets. It is not possible to avoid floods, but flood preparedness can be developed and its impact can be minimized. With appropriate planning and management, it is possible to reverse the uneven trend of growth in state and check the migration of the people in search of their livelihood. To minimize the negative consequences of flood impacts, flood management works so far implemented comprise of structural measures like, construction and maintenance of embankments, aprons in selected portions of river banks, land spurs and other necessary flood protection works. But structural measures alone cannot solve the problem. So there is need to adopt non structural measures as well. Non-structural flood management measures such as land use regulations, flood forecasting and warning, flood proofing and disaster prevention, preparedness and response mechanisms should be adopted. The awareness of the community at risk of flooding should be raised and maintained with a clear understanding of their role in responding to emergency situations appropriately. They need to be developed so as to adaptation to the flood risk and living with them in flood plains and simultaneously permitting vacating the same for use by the river whenever the situation demands.

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RESEARCH ARTICLE

A STUDY OF ERRORS AND MISCONCEPTIONS IN SCIENCE IN RELATION TO SCIENTIFIC ATTITUDE AMONG SECONDARY SCHOOL STUDENTS.

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Key words:-

Errors, Misconceptions, Scientific Attitude, Concept Achievement Test.

Abstract

To promote effective and meaningful learning there is need to identify misconceptions as prevalence of misconceptions among students not only presents an obstacle to learning but also interferes in further learning. A descriptive study was conducted on a sample of 912 students studying in ninth class in various schools of Punjab. For the study Concept Achievement Test in Science was constructed and standardized and was administered to identify errors and misconceptions among students. Scientific Attitude Scale (SAS) developed and standardized by Gakhar and Kaur (1995) was used for measuring the attitude relevant in science education. The results related to patterns of errors indicate that secondary school students with unfavourable scientific attitude committed more errors as compared to their counterparts with favourable scientific attitude. The suggestions for educational implications are also discussed.

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Introduction:-

Attitude has been identified as an important correlated towards achievement of students. Chen, Hui-Ling (2001); and Mehra (2004) found mathematics attitude to be related to achievement while findings of studies by Bandopadhyaya (1984); Paul (1986); Kar(1990); and Nelliappan (1992); showed significant relationship between scores on scientific attitude and achievement in science. On the other hand Many researches were done in various subjects to identify the naïve ideas (misconceptions) of students that can create learning difficulties for students and ultimately causing low achievement (Arnaudin and Mintzes, 1985; Boyes and Stainstreet, 1991; Driver, 1993; Marques and Thompson, 1997; Palmer, 1998). Researches were conducted to identify misconceptions of various concepts of science by Eryilmaz and Surmeli(2002), Cetin(2004), Haki(2005) and Kutuluay(2005) who revealed that errors among secondary school students were due to mistakes, lack of knowledge and misconceptions. While identifying errors and misconceptions among students they referred mistakes as the incorrect answers given by the students who have correct scientific conceptions while lack of knowledge as the incorrect answers given by the students who have incorrect scientific conceptions and have no confidence for their wrong conceptions, in case the students have confidence for their wrong conceptions, these were referred as misconceptions. There are a lot of researches in the area of science achievement but there is paucity of researches in the area “errors and misconceptions in learning of scientific concepts” in Indian context. So the present study is an endeavor in this direction to provide empirical evidence with regard to problems in learning of scientific concepts with objectivity and scientifically.

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Objectives:-

- To find out the level of performance of secondary school students on Concept Achievement Test (CAT) in science in relation to scientific attitude.
- To find out the percentage of secondary school students who committed errors and misconceptions on Concept Achievement Test (CAT) in science in relation to scientific attitude.
- To study the patterns of errors and misconceptions on Concept Achievement Test (CAT) in science in relation to scientific attitude.

Hypothesis:-

The students with favourable scientific attitude will differ from students with unfavourable scientific attitude in their pattern of errors and misconceptions on Concept Achievement Test (CAT) in science.

Method:-

A descriptive method of research was used in the conduct of the study. The study was completed in two phases:

Phase I: Construction and standardization of Concept Achievement Test (CAT) in science by the researcher for the identification of errors and misconceptions to include the concepts of adaptations, habitat, biosphere, ecosystem, food chain and food web, functions of ecosystem, biomass and biodiversity from the PSEB prescribed science text book of ninth class.

Phase II: Field Work

The preliminary draft of the test contained fifty three test items to be responded on three tiers. This draft was administered to 220 students of ninth class from various schools in Patiala. On the basis of the indices of the item difficulty and in the view of the experts 23 items were discarded and 30 items were selected. Then that test was administered to 125 secondary school students of Patiala to find out the reliability and validity of the test. First tier included multiple choice items having one right answer and three distracters. The second tier required students to write reason for the response. Third tier asked students about their confidence for the answer in first two tiers. Total achievement score of each student was calculated according to the students' multiple choice item scores, reasoning part scores and the confidence level scores together.

Sample:-

A sample of 912 students of 9th class studying in high and senior secondary government schools was selected randomly selecting 33 schools from seven districts of Punjab namely, Patiala, Ropar, Ludhiana, Bathinda, Mansa, Fatehgarh Sahib and Sangrur.

Tools:-

Following tools of research were used for the collection of data:-

1. Concept Achievement Test constructed and standardized by researcher herself.
2. Scientific Attitude Scale (SAS) developed and standardized by Gakhar and Kaur (1995) was used for measuring the attitude relevant in science education.

Findings:-

The major findings of the study are summarized below: The achievement of secondary school students of Punjab on Concept Achievement Test (CAT) in science was moderate.

- The secondary school students with favourable scientific attitude performed significantly higher than their counterparts with unfavourable scientific attitude on Concept Achievement Test (CAT) in science. The level of performance on Concept Achievement Test (CAT) in science was directly/positively affected by achievement and attitude. The secondary school students with unfavourable scientific attitude committed more errors in the concept areas of 'adaptations', 'habitat', 'food chain and food web', 'functions of ecosystem' and 'biomass and biodiversity'.
- The secondary school students with unfavourable scientific attitude significantly have more objectively false conceptions as compared to their counterparts with favourable scientific attitude on Concept Achievement Test (CAT) in science.
- There is no significant difference in the mean misconceptions of secondary school students with favourable and unfavourable scientific attitude on Concept Achievement Test (CAT) in science.

- It is found that students with unfavourable scientific attitude have significantly more mean mistakes, lack of knowledge and misconceptions as the sources of errors as compared to their counterparts on Concept Achievement Test (CAT) in science.

Testing of Hypothesis:-

The hypothesis formulated was accepted. The results related to patterns of errors indicate that secondary school students with unfavourable scientific attitude committed more errors as compared to their counterparts with favourable scientific attitude.

Educational Implications:-

In the light of the findings of the present study regarding errors and misconceptions in science learning among secondary school students, following implications emerged that can be used by teachers to improve the delivery of science education.

1. The findings of this study could facilitate teachers in their planning and implementation of relevant measures to reduce the incidence of students' misunderstandings about the concept of environment. A teacher should focus on students cognitive level to eliminate misconceptions, because most of the students in high schools and all students in elementary and junior high schools are in concrete levels; therefore, the major focus of instruction for those students should link between concept and concrete experiences and expect difficulties on linking concrete experiences to abstract concepts because any science concept has a relationship with other concepts, so students have to link the ideas and other concepts.
2. The teacher plays a pivotal role in identifying, reconstructing and internalizing the scientific knowledge among the students. The teacher should encourage students to test their conceptual frameworks in discussion with other students and by thinking about the evidence and possible tests. He should revisit common misconceptions as often as he/she can. He should allow students to have sufficient experience of the first concept before introducing the next one.
3. Teachers should involve use of animations, videos, activities and alternative methods of teaching viz. concept mapping (representing knowledge in graphs), clustering (finding groups for data), mental modeling (asking open ended questions), problem solving and laboratory skills.
4. The results of the study showed that students made errors, due to mistakes, lack of knowledge or due to misconceptions. These misconceptions resist changing and obstructing the learning process. The teachers are required to take students misconceptions into account. The more the teachers know about their students' misconceptions the more guidance they will be able to provide them to learn. This could contribute to the professional development of science teachers. Smith and Anderson (1993) and Lawrenz (1986) advocated that during (preservice and in-service) teacher education programmes the teachers should be given chances to identify misconceptions held by the pupils in their classrooms.
5. Identification of the students' misconceptions in science for the unit of environment by Concept Achievement Test (CAT) in science can give some feed back to the text book editors.
6. Students' misconceptions and alternative ideas can be taken into consideration by curriculum developers and remediation techniques of them should be designed. If these techniques can be combined with small group work with demonstration and appropriate hands-on activities, they should be very powerful methodology for science classrooms.
7. To remove misconceptions among students the teachers should provide more structured opportunities for students to talk about ideas at length, both in small groups and whole class discussions.
8. This work has identified new material which can be utilized by science educators both in the classroom and in teachers' pre-and in-service courses. This new material includes a number of misconceptions. These new pieces of information add to the overall picture of students' conceptual development and with this knowledge, educational practitioners will be in a better position to make necessary changes and improvements in their classroom practices to enhance the student learning.

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RESEARCH ARTICLE

ISOLATION OF *ENTEROBACTERIACEAE* FROM RAW SEAFOODS SOLD IN FISH MARKETS IN EASTERN PROVINCE OF SAUDI ARABIA.

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Abstract

Aim of this investigation is to provide the general information about prevalence of members of *Enterobacteriaceae* in raw seafoods sold in fish markets in Eastern province of Saudi Arabia. Studies were carried out to isolate and identify different bacterial species belongs to the family of *Enterobacteriaceae* in 448 seafood samples comprising Fishes (353), Prawns (49) and Cuttlefishes (46), collected during the period from January 2015 until May 2016. Bacterial isolates were characterized on specific culture media and further confirmed using API 20E Strips and identified by "api web". Out of 448 samples analysed, 304 (67.85%) samples showed positive isolation of *Enterobacteriaceae*. *Morganellamorganii* showed highest prevalence (18.52%), followed by *Proteussp* (12.27%), *Citrobactersp* (8.03%), *Enterobactersp* (6.69%), *Providenciasp* (5.58%), *Hafniaalvei* (4.46%), *Escherichia coli* (4.24%), *Klebsiellasp* (2.90%), *Rahnellaaquatilis* (2%), *Serratialiquefaciens* (1.78%) and *Pantoeaagglomerans* (1.33%). The presence of these organisms in the fresh seafood samples showed that the source of contamination may be from environment, catching, unhygienic handling and transportation. Thus it is strongly recommended that the improvements in handling and processing are needed to minimize the prevalence of pathogenic bacteria by implementing preventive measures and application of appropriate procedures for hygiene practices to monitor the quality of raw seafoods.

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Introduction:-

Fish is a valuable and nutritious food, and an essential source of high quality and cheap animal protein crucial to balance of diets in marginally food secure communities (United Nation, 2004). Production of Fish and Shellfish by aquaculture is the fastest growing sector of world food economy. World aquaculture output has been growing at 11% per annum during the past decade and has reached 33.3 million tonnes in the year 1999 (Biradaret *et al*, 2001).

Unlike meat and poultry, fish are more liable to contamination with pathogenic bacteria from human reservoirs which may contaminate the water depending on the fishing and also may be further contaminated during handling, processing and packaging. While the muscle flesh of fish, which is the main edible part is normally sterile but microorganisms can penetrate from the skin and the gut to the flesh (FAO, 1983).

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Fishery products have been recognized as a major carrier of food-borne pathogens. The contamination can occur prior to harvest, during harvest and processing operations, distribution, storage and preparation of the product. In a simplified overview, poor quality products, spoiled or decomposed are rarely responsible for food poisoning because they are usually discarded before consumption. Food poisoning in seafood products such as scombroid or histamine poisoning, normally is the result of mishandling during or after preparation. The microbial flora of seafood is related to the environment from which the fish are harvested (Wekellet *al*, 1994; Falana, 2005).

Fresh fish is a very perishable product and spoil due to microbiological activity, chemical oxidation of lipids and autolysis. However, microbial spoilage is the main mechanism affecting fresh fish quality. As the bacteria grow they utilize nutrients and produce by-products. It is well established that the accumulation of metabolic products is the primary cause of the organoleptic rejection of fresh fish. It is also known that spoilage is caused only by a fraction of the initial microbial population, known as specific spoilage organisms (SSO), which produce metabolites responsible for off-flavors and cause the organoleptic rejection of the product. The fraction of the initial micro-biota that dominate and the metabolic products that are produced are determined mostly by the temperature and atmospheric conditions during storage (Gram and Huss, 1996).

The *Enterobacteriaceae* count is considered as an index to determine the seafood quality. Some human pathogens such as *Escherichia coli*, *Klebsiellasp*, *Salmonellasp*, *Citrobactersp* and *Proteus sp*, when isolated from fish and fish products gives an identification about environmental fecal pollution of fish. These organisms have been found to survive and multiply in the gut, mucus and tissues of fish and that render fish acting as potential vector of human disease over long periods (Onyangoet *al*, 2009; Wogu and Maduakol, 2010).

Contamination of seafood with *Salmonella* is a major public health concern. Salmonellosis is a worldwide problem responsible for food poisoning outbreaks but it is mainly difficult to determine whether the contamination of fishes occurred in their aqueous habitat or during their handling and marketing (Etzelet *al*, 1998). Coliforms and Fecal coliforms in fish demonstrate the level of pollution of their environment because these organisms are not the normal bacterial flora in fish. (Cohen and Shuval, 1973).

Therefore, the intention of this study is to determine the contamination level of seafood samples with *Enterobacteriaceae* collected from the Eastern province of Saudi Arabia. And to take recommendations to reduce risk associated with the consumption of seafood.

Materials and Methods:-

A total of 448 samples comprising of Fishes (353), Prawns (49) and Cuttlefishes (46) were randomly collected from retail and wholesale fish markets in Damman during the period from January 2015 until May 2016. The collected samples were individually kept in sterile polyethylene bags embedded with ice and transported rapidly to the laboratory in an insulated box.

Sample processing for Bacteriological Examination:-

Bacteriological analyses of collected seafoods were performed within 2 hours of sampling. All media used in the present study were from Oxoid, U.K. and prepared according to the standard procedures. Media were sterilized by autoclaving at 121°C at 15 psi pressure for 15 minutes, unless otherwise specified. Sterility of the media was checked by incubating at 37°C for 24 hours. The microbiological analysis was performed as per the standard methods adopted from the Online Bacteriological Analytical Manual, USFDA (BAM 8th edition Online).

***Salmonella* species:-**

25 g of the sample was homogenized in 225 ml of Lactose Broth (LB) and incubated overnight at 37°C. After the initial pre enrichment step, samples were selective enriched in Tetrathionate Broth (TTB) at 37°C and Rappaport Vassiliadis (RV) medium at 42°C and incubated overnight. A loopful of growth from each of these media was streaked on Bismuth Sulphite Agar (BSA), Xylose Lysine Deoxycholate Agar (XLDA) and Hektoen Enteric Agar (HEA). After 24 (HEA, XLDA) – 48 (BSA) hours of incubation at 37°C, both atypical and typical colonies were picked up, purified and identified further by subsequent biochemical tests which included Gram staining, Motility, Oxidase production and then identified at the genus and species level using API 20E (Biomerieux, France) test kits. The identification was obtained by using the identification software “api web”.

Other Enterobacteriaceae:-

25 g of the sample was homogenized in 225 ml of sterile physiological saline in a filter stomacher bag and made in to slurry using the BagMixer® (Interscience, France) and the diluted samples were plated on MacConkey Agar. After 24 hours of incubation at 37°C, both pink and colourless colonies were picked up, purified and identified further by subsequent biochemical tests which included Gram staining, Motility, Oxidase production and then identified at the genus and species level using API 20E (Biomérieux, France) test kits. The identification was obtained by using the identification software “api web”.

Results and Discussion:-

The *Enterobacteriaceae* are all occurring on fish products as a result of contamination from the animal/human reservoir. This contamination has normally been associated with fecal contamination or pollution of natural water or water environments, where these organisms may survive for a long time (months) or through direct contamination of products during processing (FAO).

This study was conducted to determine the incidence of *Enterobacteriaceae* in Seafoods (353 Fishes, 49 Prawn and 46 Cuttlefish) for sale in wholesale and retail outlets of fish markets in Eastern province of Saudi Arabia. The results related to the incidence and identification at the species level of *Enterobacteriaceae* from Fishes, Prawn and Cuttlefish are summarized in Table 1. The overall percentages of prevalent bacterial species are shown in Table 2.

Out of 448 samples analysed, 304 (67.85%) samples showed positive isolation of *Enterobacteriaceae*. The ranges of incidence levels of *Enterobacteriaceae* were from Fishes 233 out of 353 (66%), Prawn 44 out of 49 (89.79%) and Cuttlefish 27 out of 46 (58.69%).

The obtained results were revealed that the dominant bacterial species occurred most frequently in the three types of seafood samples includes *Morganellamorganii* with 83 isolates (18.52%), followed by *Proteus* spp with 55 isolates (12.27%), *Citrobacter* spp with 36 isolates (8.03%), *Enterobacter* spp with 30 isolates (6.69%), *Providencia* spp with 25 isolates (5.58%), *Hafnia alvei* with 20 isolates (4.46%), *Escherichia coli* with 19 isolates (4.24%), *Klebsiella* spp with 13 isolates (2.90%), *Rahnella aquatilis* with 9 isolates (2%), *Serratia liquefaciens* with 8 isolates (1.78%) and the lower isolate was *Pantoea agglomerans* with 6 isolates (1.33%).

Salmonella was not detected in any of the samples analysed in this study, which was in agreement with previous studies (Popovic *et al*, 2010; Rodriguez *et al*, 2011; Figenet *et al*, 2014) in seafood products. Conversely, different workers (Shabarinath *et al*, 2007; Kumar *et al*, 2009; Bakret *et al*, 2011) reported varying incidence rates of *Salmonella* in seafood.

Morganellamorganii is widely distributed in nature, commonly found in the environment and the intestinal tracts of humans, mammals and reptiles as a part of the normal flora. Generally *Morganellamorganii* can act as an opportunistic pathogen that mainly causes wound and urinary tract infections. The drug resistance of *Morganellamorganii* is increasing in recent years, which often results in Clinical treatment failure (Hui Liu *et al*, 2016). In the present study *Morganellamorganii* showed highest prevalent (18.52%) in all the analysed seafoods. In a study conducted by Sureeratphuvasate (2008), he clearly demonstrated the ability of *Morganellamorganii* to produce Histamine in fish samples. Histamine is a biogenic amine formed through the decarboxylation of the amino acid histidine by the enzyme histidine decarboxylase. Consumption of fish containing histamine may result in illness known as Scombroid poisoning with a variety of symptoms, including rash, facial flushing, sweating, vomiting, diarrhea, head ache, burning sensation and metallic taste in the mouth.

Proteus sp. were involved in the spoilage of seafoods which can result in potential health hazard to the consumers. It can cause enteric infection in humans, summer diarrhea in infants, sinusitis, otitis as well as urinary tract infections (Bryan 1980; Varnam and Evas, 1991). The isolation of *Proteus* sp. from fish samples has been reported in previous studies by Tosun *et al* (2016). Sanaa O. Yagoub (2009) isolated *Proteus* spp in fish with an incidence of 10.2%. In the present study, the percentages of *Proteus mirabilis* and *Proteus vulgaris* were 7.58% and 4.68% respectively. Some investigations involve *Proteus* genus as an indicator of fecal contamination in marine fish, although their use as an indicator is not very common (ILSI, 2011).

In this study, members of *Citrobacter* were identified, represented by species in the ranges of *Citrobacter freundii* (4.24%), *Citrobacter braakii* (2.67%) and *Citrobacter koserii* (1.11%). Olugboje *et al* (2015), detected the presence of

Citrobactersp in fish samples of commercial importance collected from Nigeria. In a study conducted by Tosunet *al* (2016), *Citrobactersp* showed highest prevalence (45.56%) among all other *Enterobacteriaceae* from Horse Mackerel fish sold in public markets of Istanbul, Turkey. *Citrobactersp* are widely distributed in soil, water, food and the intestinal tract of humans and animals. They make up the normal flora of the human gut and were previously considered as a purely environmental contaminants or non-pathogenic colonizers, but known to be opportunistic pathogens, as they can cause serious infections, sepsis, food borne outbreaks of gastroenteritis, respiratory infections and urinary tract infections, especially in high-risk groups such as infants and immune compromised adults (Rusulet *al*, 1991; Dos santoset *al*, 2015).

The *Enterobactersp* can be found in soil, water, sewage, intestinal tract of man and animals (Banwart, 1989). In the present study, several species of *Enterobacter* has been isolated which includes *Enterobacter cloacae* (2.67%), *Enterobacter aerogenes* (2.23%), *Enterobacteramnis* (1.11%) and *Enterobactersakazakii* (0.66%). In a study conducted by Mai F. Elsheriefet *al* (2014), he isolated *Enterobactersp* along with other members of *Enterobacteriaceae* from the farmed and retailed fishes of Tilapia and Mugilcephalus. *Enterobactersp* can act as opportunistic pathogens that rarely cause primary diseases in humans. They are described as agents associated with respiratory infections, skin, urinary tract, bones, joints, central nervous system and gastro intestinal tract infections (Dos Santos *et al*, 2015).

In this study, members of *Providencia* were isolated, identified and represented by species in the percentages of *Providenciastuartii* (2.67%), *Providenciaertgeri* (1.56%) and *Providenciaalcalifaciens* (1.33%). Sanaa O. Yagoub (2009), isolated *Providencia* spp along with other members of *Enterobacteriaceae* including, *Escherichia coli*, *Enterobactersp*, *Klebsiellasp*, *Citrobactersp*, *Salmonellasp*, *Shigella* sp, *Serratiasp* and *Proteussp* from raw fish sold in fish market in Khartoum state of Sudan. *Providenciasp* are generally considered to be commensals in the gastrointestinal tract of humans, warm blooded animals, often also in soil and sewage water. *Providenciaertgeri*, *Providenciastuartii* and *Providenciaalcalifaciens* has been isolated from samples of human feces, either as part of the human intestinal microbiota as well as a cause of gastric disorder, such as traveler's diarrhea. In 2001, *Providenciaalcalifaciens* was reported in a major outbreak of food borne illness among children in Japan. They can act as opportunistic pathogen involved in urinary tract infections, keratitis, dacryocystitis, conjunctivitis and endophthalmitis. The bacteria *Providenciastuartii* is reported as more resistant species of the family *Enterobacteriaceae*. The emerging of resistance in *Providenciasp* is a clinical concern as it may lead to increased treatment cost and delays in treatment, which can be fatal to the patient (Dost Santos *et al*, 2015).

Hafniaalvei, a member of the family *Enterobacteriaceae* and the only species of the genus *Hafnia* described, is part of the human gastrointestinal flora, but can be found in environmental habitats such as surface water and food. *Hafniaalvei* is normally considered as a colonizing organism and rarely appears to be a pathogen. Recent findings suggest that it is a rare but may contribute to opportunistic infections in humans apart from diarrhea; it can cause extra intestinal infections such as septicaemia, liver abscess, peritonitis and pneumonia (Podschunet *al*, 2001; Dos Santos *et al.*, 2015). In the present investigation, the prevalence range of *Hafniaalvei* is around 4.46% in seafoods, followed by *Escherichia coli* with the percentage of 4.24% and members of *Klebsiellasp*, represented by its species in the ranges of *Klebsiellaterrigena* (1.56%) and *Klebsiellapneumoniae* (1.33%). Isolation of *Escherichia coli* and *Klebsiellasp* from the seafood samples analysed in this study was in agreement with the previous studies done by different workers (Yagoub, 2009; Mai F. Elsheriefet *al*, 2014; Romero Jarero Jorge, 2016; Tosunet *al*, 2016) who reported varying incidence rates of such organisms in seafoods.

Infections associated with *Escherichia coli* through the ingestion of seafoods are directly related to fecal contamination. *Escherichia coli* is normally present in the intestinal flora of humans and warm-blooded animals which may transfer to the foods due to the poor hygienic conditions, cross contamination or contaminated water (Huss *et al*, 2004; Renata Albuquerque costa, 2013). *Klebsiellasp* can also act as an opportunistic pathogen that cause wide range of infections including foodborne outbreaks of gastroenteritis, urinary tract infections, pneumonia, bacteremia, neonatal sepsis and wound infections (Rahkiaet *al*, 1998; Podschunet *al*, 1998).

In the present study members of *Enterobacteriaceae* such as *Rahnellaaquatilis*, *Serratialiquefaciens* and *Pantoeaagglomerans* were isolated in the ranges of 2%, 1.78% and 1.33% respectively from the analysed seafoods. All the above organisms can act as opportunistic pathogen in humans. *Rahnellaaquatilis* is typically associated with fresh water, since been isolated from blood, surgical wounds, urine, sputum, bronchial washings and stool, usually in immune compromised patients or in young children (KaleyTash, 2004). *Serratialiquefaciens* also an opportunistic

pathogen which is capable of colonizing a wide variety of surfaces in water, soil, the digestive tracts of rodents, plants, insects, fish and humans (Grimont and Grimont, 1978). *Pantoea agglomerans* inhabits plants, soil, and water, reported as both commensal and pathogen of animals and humans. They rarely cause disease in healthy individuals, such as septic arthritis, outbreak secondary to contaminated parental nutrition, occupational respiratory infections and skin allergy (Maria Carla Liberto *et al*, 2009).

Most of the *Enterobacteriaceae* members isolated and identified in this study were comes under the group of Coliforms and Fecal Coliforms. Austin and Austin (1985) have demonstrated the presence of *Escherichia coli*, *Enterobacter* sp, *Citrobacter* sp, *Salmonella* sp, *Proteus* sp, in fish that lives in contaminated marine water by domestic waste water. Priyanka Rani Majumdar *et al* (2014), stated that the untreated and the improper way of sewage disposal system in the water body is one of the main sources for microbial water contamination which results in the accumulation of these bacterial pathogenic species in the commercial edible fishes. Moreover, unhygienic handling by infected people or healthy carriers, during the capture, transport and processing may also act as a source of contamination by pathogenic organisms in seafoods.

Table 1:-List of Bacterial species isolated from Fishes, Prawn and Cuttlefish.

FISHES	PRAWN	CUTTLEFISH
<i>Enterobacter cloacae</i>	<i>Enterobacter aerogenes</i>	<i>Proteus mirabilis</i>
<i>Enterobacter aerogenes</i>	<i>Enterobacter amnigenus</i>	<i>Proteus vulgaris</i>
<i>Enterobacter amnigenus</i>	<i>Enterobacter cloacae</i>	<i>Providencia stuartii</i>
<i>Enterobacter sakazakii</i>	<i>Enterobacter sakazakii</i>	<i>Providencia rettgeri</i>
<i>Citrobacter freundii</i>	<i>Citrobacter freundii</i>	<i>Morganella morganii</i>
<i>Citrobacter braakii</i>	<i>Citrobacter braakii</i>	<i>Hafnia alvei</i>
<i>Citrobacter koseri</i>	<i>Citrobacter koseri</i>	<i>Citrobacter braakii</i>
<i>Providencia stuartii</i>	<i>Proteus vulgaris</i>	<i>Escherichia coli</i>
<i>Providencia alcalifaciens</i>	<i>Proteus mirabilis</i>	<i>Klebsiella pneumoniae</i>
<i>Providencia rettgeri</i>	<i>Morganella morganii</i>	<i>Rahnella aquatilis</i>
<i>Proteus mirabilis</i>	<i>Escherichia coli</i>	
<i>Proteus vulgaris</i>	<i>Hafnia alvei</i>	
<i>Klebsiella terrigena</i>	<i>Rahnella aquatilis</i>	
<i>Klebsiella pneumoniae</i>	<i>Serratia liquefaciens</i>	
<i>Morganella morganii</i>	<i>Pantoea agglomerans</i>	
<i>Escherichia coli</i>		
<i>Hafnia alvei</i>		
<i>Rahnella aquatilis</i>		
<i>Serratia liquefaciens</i>		
<i>Pantoea agglomerans</i>		

Table2:- Prevalence of Bacterial species among Fishes, Prawn and Cuttlefish.

S. No	Bacterial Isolates	Bacteria l Genus	Overall No. (%) of Prevalence in Samples			No. (%) of Prevalence for Total 448 Samples	No. (%) of Prevalence of Bacterial Genus
			Fishes (353)	Prawn (49)	Cuttlefish (46)		
01.	<i>Morganellamorganii</i>	<i>Morganellasp</i>	63 (17.84 %)	14 (28.57%)	06 (13.04%)	83 (18.52%)	83 (18.52%)
02.	<i>Proteus mirabilis</i>	<i>Proteus spp</i>	28 (07.93%)	02 (04.08%)	04 (08.69%)	34 (07.58%)	55 (12.27%)
03.	<i>Proteus vulgaris</i>		14 (03.96%)	03 (06.12%)	04 (08.69%)	21 (04.68%)	
04.	<i>Citrobacterfreundii</i>	<i>Citrobacter spp</i>	16 (04.53%)	03 (06.12%)	---	19 (04.24%)	36 (08.03%)
05.	<i>Citrobacterbraakii</i>		07 (01.98 %)	03 (06.12%)	02 (04.34%)	12 (02.67%)	
06.	<i>Citrobacterkoserii</i>		04 (01.13%)	01 (02.04%)	---	05 (01.11%)	
07.	<i>Enterobacter cloacae</i>	<i>Enterobacter spp</i>	11 (03.11%)	01 (02.04%)	---	12 (02.67%)	30 (06.69%)
10.	<i>Enterobacter aerogenes</i>		06 (01.69%)	04 (08.16%)	---	10 (02.23%)	
08.	<i>Enterobacter amnigenus</i>		03 (00.84%)	02 (04.08%)	---	05 (01.11%)	
09.	<i>Enterobacter sakazakii</i>		02 (00.56%)	01 (02.04%)	---	03 (00.66%)	
11.	<i>Providencia stuartii</i>	<i>Providencia spp</i>	09 (02.54%)	---	03 (06.52%)	12 (02.67%)	25 (05.58%)
13.	<i>Providencia rettgeri</i>		06 (01.69%)	---	01 (02.17%)	07 (01.56%)	
12.	<i>Providencia alcalifaciens</i>		06 (01.69%)	---	---	06 (01.33%)	
14.	<i>Hafnia alvei</i>	<i>Hafnia spp</i>	13 (03.68%)	03 (06.12%)	04 (08.69%)	20 (04.46%)	20 (04.46%)
15.	<i>Escherichia coli</i>	<i>Escherichia spp</i>	14 (03.96%)	04 (08.16%)	01 (02.17%)	19 (04.24%)	19 (04.24%)
16.	<i>Klebsiella terrigena</i>	<i>Klebsiella spp</i>	07 (01.98%)	---	---	07 (01.56%)	13 (02.90%)
17.	<i>Klebsiella pneumoniae</i>		05 (01.41%)	---	01 (02.17%)	06 (01.33%)	
18.	<i>Rahnella aquatilis</i>	<i>Rahnella spp</i>	07 (01.98%)	01 (02.04%)	01 (02.17%)	09 (02.00%)	09 (02.00%)
19.	<i>Serratia liquefaciens</i>	<i>Serratia spp</i>	07 (01.98%)	01 (02.04%)	---	08 (01.78%)	08 (01.78%)
20.	<i>Pantoea agglomerans</i>	<i>Pantoea spp</i>	05 (01.41%)	01 (02.04%)	---	06 (01.33%)	06 (01.33%)
TOTAL NUMBER OF ISOLATES			233 (66.00%)	44 (89.79%)	27 (58.69%)	304 (67.85%)	304 (67.85%)

Conclusion:-

The monitoring and control of seafood quality is one of the main goals in the seafood industries. The findings of this study revealed that the raw seafoods sold in fish markets in the eastern province of Saudi Arabia could be a source of food borne bacterial pathogens. The presence of such contaminating bacteria could be attributed to cross contamination from the environment, source and unhygienic handling by the sellers. Hence, the findings of this study may be considered as an additional knowledge to enhance proper controlling of the storage and shelf life of seafoods by implementing preventive measures such as HACCP and application of Sanitation Standard Operating

Procedures (SSOP) in fish markets to monitor the quality of seafoods by minimizing the prevalence of the pathogens.

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RESEARCH ARTICLE

STEREOMICROSCOPIC EVALUATION OF DENTINAL DAMAGE CAUSED BY K-FILES, PROTAPER UNIVERSAL, NEONITI A1 AND iRaCe FILES-AN *IN VITRO* STUDY

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Abstract

Aim: To compare the dentinal damage induced by stainless steel hand and rotary nickel-titanium instruments using K files, ProTaper Universal, Neoniti A1 and iRaCe files. **Materials and Methods:** Sixty single rooted premolars were collected and randomly divided into four groups of 15 teeth each. Group 1- Stainless steel K Files, group 2- ProTaper Universal, group 3- Neoniti A1 file and group 4 - iRaCe rotary instruments. Cleaning and shaping was done with the respective files. The roots were then cut horizontally at 3mm, 6mm and 9 mm from apex and were examined under stereomicroscope. The sections were checked for dentinal defects. **Statistical analysis:** Groups were analysed with the Fishers exact test. **Results:** It was found that all instruments examined caused dentinal defects with maximum defects in the apical third. Neoniti A1 file showed least number of dentinal defects compared to other file systems. **Conclusion:** The present study revealed that all the instruments could result in an increased chance for dentinal defects.

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Introduction:-

The main objective of endodontics is a three dimensional seal of the root canal system. There are various steps in root canal treatment and each step has its importance in the success of the treatment. The biomechanical preparation is one of the steps in the root canal treatment for removal of bacteria and debris from the root canal to receive an obturating material.^{1,2,3}

Traditionally, the shaping of the root canal was achieved by the use of stainless steel hand files.⁴ Stainless steel root canal instruments clean the canal superficially and can create canal aberrations such as ledges, zips, and elbows.⁵ To eliminate these shortcomings of stainless steel instruments, nickel titanium (NiTi) instruments have been introduced.

It offers many advantages over conventional files. They are flexible (Walia et al. 1988), have increased cutting efficiency (Kazemi et al. 1996) and have improved time efficiency (Ferraz et al. 2001). Furthermore, Ni-Ti instruments maintain the original canal shape during preparation.⁴

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ProTaper Universal (DENTSPLY Tulsa Dental Specialties) utilized multiple tapers of an increasing or decreasing percentage on a single file. This revolutionary, progressively tapered design limits each file's cutting action to a specific region of the canal.⁶

Recently Neoniti A1, a single file nickel titanium (NiTi) has been introduced which completes the canal preparation with only one instrument, this file system is made by the EDM (Electric Discharge Machining) process which is responsible for the unique behavior of the file⁷ which is claimed to relieve stress on the instrument because of its peculiar cutting action requiring even lesser time than rotary full-sequence systems.⁸ It is assumed that this movement reduces the risk of cyclic fatigue caused by tension and compression.

iRaCe (FKG) provides a quick, safe and effective protocol for preparation of curved root canals. These recently introduced NiTi rotary files have the same design features as RaCe files. The working part has alternating cutting edges. This design avoids "screwing-in" effect and allows better control of the instrument's progression.⁹

Rotary files have increasing taper design that increases the cutting efficiency and relatively removes more dentin compared to other systems which leads to formation of more complete and incomplete dentinal cracks.¹⁰ Vertical root fractures, which is one common complication associated with mechanical canal preparation¹¹, initiated from this dentinal crack (Wilcox et al. 1997), can lead to extraction.

A vertical root fracture is a longitudinally oriented fracture of the root, extending throughout the entire thickness of dentin from the root canal to the periodontium. It may be initiated in the crown or at the root apex or in some cases, along the root between these two points.¹²

Dentinal microcracks can occur during tooth extraction and tooth-sectioning procedures, and pre-existing defects may not be detected using root sectioning and microscopic observation techniques.¹³ A crucial goal in endodontic research is to overcome the potential problem of dentinal microcrack formation during instrumentation with rotary or reciprocating instruments.¹³ The purpose of this study is to evaluate dentinal defects caused by different NiTi file systems and K files.

Material and Methods:-

Sixty single rooted premolars were collected, cleaned with periodontal scaler and stored in purified water. Teeth with curved roots, calcified canals, developmental anomalies or extra canals were discarded. The coronal portions of all teeth were removed with diamond disk with water cooling leaving roots of 15mm in length. All the root surfaces were observed with stereomicroscope under 24x to rule out preexisting cracks or any craze lines.

Apical patency was checked using a 10 no. K file. Working length was determined by leaving 10 no. K file in the canal so that it was just visible at the apical foramen and from that length 0.5mm was subtracted. The specimens were divided into 4 groups each group containing 15 specimens each.

Group 1- K Files stainless steel files (Mani)

Group 2- ProTaper Universal (DENTSPLY Maillefer)

Group 3- Neoniti A1 (NEOLIX, FRANCE)

Group 4- iRaCe (FKG Swiss endo)

In all the groups each canal was irrigated with 3% sodium hypochlorite and saline alternatively. Along with RC-Prep (EDTA 15% and urea peroxide 10%) used between each sequential instrument in canal preparation.

Hand file group was prepared using stainless steel K files up to apical size 25 at the working length and step-back technique was used up to 40 no. K file.

In the ProTaper Universal group, the following sequence was used for canal preparation at 300 rpm using a crown down technique. Sx file was used for coronal enlargement and S1, S2, F1 and F2 corresponding to apical size 25 used at working length.

In the Neoniti A1 (25/0.08) file group, the file was taken to the middle third using 3 or 4 circumferential brushing actions followed by taking it to the working length using a pecking motion at 300 rpm.

In the iRaCe group, the file R1 in rotation was introduced into the canal and reached up to working length with long back and forth strokes and shaping of the canal was finished with R2(25/.04) up to working length. These files were used at 600 rpm.

For efficient cleaning of the canals EndoActivator (Dentsply Tulsa Dental) was used in all the groups, with no.25 tip for 30 seconds by agitating the solution vigorously.¹⁴

Examination of roots:-

After the biomechanical preparation, roots were sectioned from 3mm, 6mm and 9 mm from the apex using a diamond disk under water cooling. Digital images of all the section were captured using a 24x stereomicroscope by using a digital camera (Olympus, Tokyo, Japan). Specimens were checked for dentinal defects by two examiners. Defects were classified using Wilcox *et al.* classification¹⁵ with modification.

Defects were classified as 'no defect', 'fracture' and 'all other defects'.

"No defect" was root dentine without any lines or cracks, neither the external surface nor the internal root canal wall had no defects.

"Fracture" was the teeth with crack that was extending from the root canal space to the outer root surface (Figure 1).

"Other defects" were other cracks that did not extend from the root canal to the outer root surface. This includes the internal and external craze lines and partial cracks.

"Internal craze lines" included cracks extending from the canal lumen wall into the dentin without reaching the root surface; however, "external craze lines" were defined as cracks extending from the root surface into dentin without reaching the canal lumen.

Samples were considered defective if at least one of the three sections showed any defect. Data were collected and statistical analysis was done.

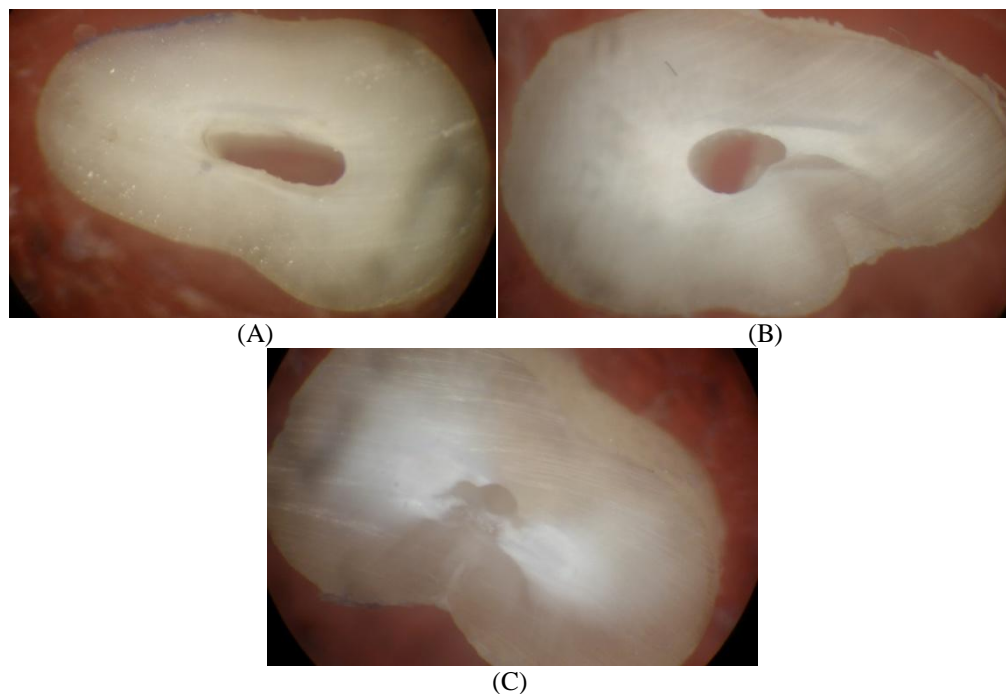


Figure 1:- (A) No defect (B) Partial Crack
(C) External Craze line

Statistical analysis:-

The data was analysed using statistical software SPSS (SPSS Inc., Chicago). Fisher's exact test was used to determine statistically significant difference in the appearance of defected roots between the experimental groups.

Fisher's exact test was also performed to determine the defects at different horizontal sections in each group. The level of significance was set at $P < 0.05$.

Results:-

Table 1 shows the number and percentage of cracks in each sections and the total number of defective samples. Figure 2 is a bar chart representing the percentage of defects in each group. Neoniti A1 group showed lowest defect (1/15) followed by ProTaper Universal and K files which showed same number of defects (3/15). iRaCe group showed maximum number of defects (4/15). Statistical significant difference was seen between Neoniti A1 group and iRaCe group ($P < 0.05$). No significant difference was found between other groups ($P > 0.05$). Only in the middle section, iRaCe and K files produced significantly more defects than ProTaper Universal and Neoniti A1 ($P < 0.05$).

Table 1:- Number and percentage of cracks.

	Group 1		Group 2		Group 3		Group 4	
	Number	%	Number	%	Number	%	Number	%
Apical	0	0%	0	0%	1	6.7%	1	6.7%
Middle	3	20%	1	6.7%	0	0%	4	26.7%
Coronal	0	0%	2	13.3%	1	6.7%	0	0%
Total	3	20%	3	20%	1	6.7%	4	26.7%

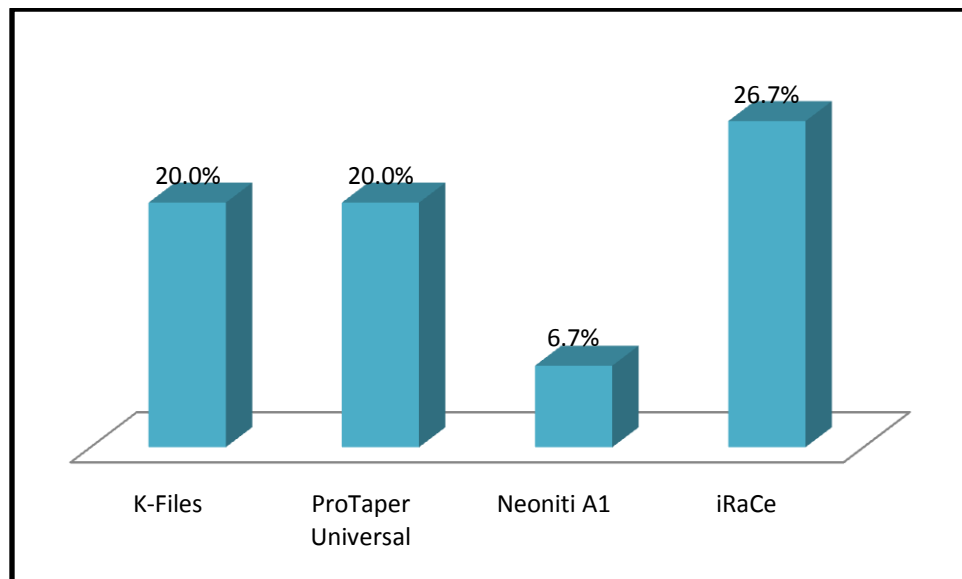


Figure 2:- Percentage of defects.

Discussion:-

NiTi instruments are used in rotational motion and rotational force is applied to root canal walls. This can create dentinal defects in root dentin.¹⁶ The aim of the present study was to detect the dentinal damage in the form of cracks and craze lines caused by K files and NiTi instruments.

In our study, we selected premolars because these teeth are probably more prone for forces of instrumentation. These teeth have got smaller dimensions and thin dentinal walls. If large tapered files cannot induce cracks in premolar, it is unlikely that rotary files induce cracks in other teeth.¹⁷

Only partial cracks were observed in this study, which indicates that fractures do not occur immediately after root canal preparation. In a study by Bier et al.¹⁸ the samples prepared with either hand or rotary instruments did not show any complete fracture. However, overtime these dentinal defects like craze lines or incomplete cracks may turn into fractures. But it has a potential effect on root fracture that makes these defects clinically significant.¹⁹

Rotary files cause more number of defects may be attributed to various reasons: Tip design, cross-section geometry, constant or variable taper, pitch and flute form.^{16,18,20,21,22} Crown down technique used in the canal preparation can also be considered as a contributor in the formation of dentinal cracks.^{21,21,23}

In this study K file group as well as ProTaper Universal group showed dentinal defects (20%). In a study by Dilek et al.¹⁹ all instruments including hand files showed dentinal defects. Though all the teeth selected in this study were having straight canals apical foramen is located eccentrically in 68 to 80% of the teeth.²⁴ This indicates that even in straight roots canals are not straight and can curve apically. In fact all the roots have some degree of curvature.²⁵ Stainless steel files are stiffer than NiTi files and this could be the reason why the K file group showed more defects.²⁶ This is in contradiction to other studies which showed least number or no defects in the hand file group.^{18,27,28} This may be due to the fact that hand files removed less amount of dentin because of the less taper of the hand files.^{18,20}

ProTaper Universal files caused significantly more cracks. Finishing files of ProTaper Universal files has got more taper. This may cause the dentinal cracks by generating increased stress on the dentinal walls as compared to other rotary system.²⁹ These files have got more active rotating movement that may result in that may result in high levels of stress concentrations in root canal walls that may result in crack formation.³⁰

Neoniti A1 file used in this study showed least number of defects. A literature search did not reveal any previous studies regarding the effect of this file system on root dentin. These files are made by EDM (Electric Discharge Machining) process offers unique features such as progressive flexibility, sharp cutting edges and built in abrasive properties.⁷ The relatively high flexibility of Neoniti A1 file may be the reason for the less number of cracks in this study.³¹ Moreover, Neoniti A1 is a single file system. Only one file is required for the complete shaping of the canal and this may also contributes to the less number of defects.³¹ As the EDM technology is a newly introduced method for the manufacture of files, future studies with more focus on dentinal defects in root dentine are therefore suggested.

Among the groups iRaCe showed maximum number of defects (26.7%). The RaCe files have a sharp cutting edge with convex triangular cross-section. They have asymmetrical longitudinal design. A set of cutting edge alternates with the second set pitched at a different angle leading to two different cutting edges on the same file. This could cause stress concentration at specific points rather than distribute it along the entire length of file.³² This concentration of stresses could have led to significantly more cracks seen with this system.²⁶ Moreover these files were used at a higher rpm (600). This can be the other reason for the higher number of cracks with these files.^{18,23,33,34} On the contrary Peters et al. (2014) and Ceyhanli et al (2015) reported that high rotational speeds enhanced cutting efficiency and decreased defect formation.

Limitations:-

1. Use of different speed settings for each rotary system.
2. Simulation of periodontal ligament was not done in the present study. Capar et al ³¹ stated that simulation of the periodontal ligament is necessary for investigating the influence of forces on formation of crack or fracture strength. Moreover, the periodontal ligament has viscoelastic property. It plays a vital role in stress dissipation due to application of load on the teeth.
3. Pre-existing defects due to excessive force during tooth extraction or during sectioning of the tooth that could not be evaluated.

Conclusions:-

Even though this in vitro study did not reflect the clinical settings, within the limitations of this study it can be concluded that all the instruments examined caused dentinal defects. More defects were observed in the apical one third followed by middle one third. Neoniti A1 file showed least number of dentinal defects compared to other file systems. More studies using techniques such as micro CT is suggested for further evaluation.

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RESEARCH ARTICLE

DETERMINATION OF RESIDUAL SOLVENTS IN NEOSTIGMINE METHYLSULFATE BY HEADSPACE GAS CHROMATOGRAPHY.

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Abstract

A static headspace gas chromatographic (GC-HS) method is developed for quantitative determination of residual organic solvents such as acetone, ethyl acetate and acetonitrile used in the synthesis of Neostigmine Methylsulfate. Separation was achieved on capillary column DB-624 of length 30 m having internal diameter 0.32 mm and 1.8 μ m film thickness using a Flame ionization detector with gradient column oven temperature program. The headspace condition was optimized with the vial equilibration temperature of 100°C and time at 30 min. The injection was carried out in split mode, with a split ratio of 10:1. N-methyl-2-pyrrolidone is selected as a diluent to obtain good sensitivity along with the recovery. Analytical method validation was performed within the requirements of ICH validation guidelines Q2 (R1). System suitability parameter set with tailing factor, theoretical plate and relative standard deviation as required with various pharmacopoeias are checked. Limits of detection and quantitation, precision, linearity and accuracy along with robustness are performed, and acceptable results are obtained. The proposed developed method is demonstrated to be simple, sensitive, linear, accurate and robust, hence can be used to determine the residual organic solvents in Neostigmine methylsulfate drug substance.

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Introduction:-

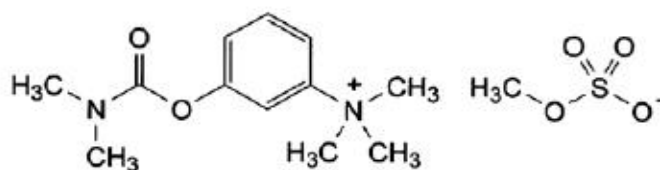
Neostigmine methylsulfate is a acetylcholinesterase inhibitor used in the treatment of myasthenia gravis and to reverse the effects of muscle relaxants such as gallamine and tubocurarine and is currently recommended by the WHO for the treatment of neurotoxic snakebite [1-4].

In the synthesis of drug substances various organic solvents are used at various stages. These solvent may be used as a medium for the reaction or purification of intermediates or drug substances. Intermediates and drug substances are dried at a specific temperature to remove the used solvents, but traces of them may carry forward to drug substance as impurities which are called as residual solvents.

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Residual solvents are unwanted chemical which does not have any no therapeutic benefit and in some cases they are toxic. Based on their toxicity, they are classified into three categories Class 1 solvents are known or suspected to be carcinogens to human and environmental hazards, hence these should be avoided in the production of drug substances, unless their use is strongly justified in a risk-benefit assessment. Class 2 solvents are non-mutagenic and known or suspected to be carcinogens in animals, therefore these solvents should be limited in order to ensure patient safety. Class 3 solvents with low toxic potential to humans; no health based exposure limit is needed. Residual solvents are generally determined using Headspace gas chromatography (GC-HS) techniques. Analytical methods used for determination of residual solvent should be validated as per ICH guidelines. The aim of this study is to develop and validate the GC-HS method of analysis for residual solvents namely acetone, ethyl acetate and acetonitrile in Neostigmine methylsulfate. Some of the methods for identification of residual solvent in drugs are also discussed [5-9].



Neostigmine Methylsulfate

Fig. 1:- Structure of Neostigmine methylsulfate

Table 1:- Residual solvent with there class and limits.

Solvent	Class	Limit (ppm)
Acetone	3	5000
Ethyl acetate	3	5000
Acetonitrile	2	410

Materials:-

Reagent and Chemicals:-

Neostigmine Methylsulfate sample was received from Analytical research and development department of Indoco research centre (Navi Mumbai). GC grade Acetone, Ethyl acetate and Acetonitrile were purchased from Merck (India). N-methyl-2-pyrrolidone used as diluent for preparation of sample and standard was purchased from Spectrochem.

Instrumentation:-

Perkin Elmer, Clarus 500 gas chromatographic system with a Turbo matrix headspace gas sampler and Flame ionization detector (FID) was used for separation and detection of solvents. Sartorius (Germany) analytical balance was used for weighing the materials.

Methodology:-

Method optimization:-

Various GC columns such as DB-Wax, DB-1 and DB-5 were used of various dimensions, but the best separation was achieved on DB-625 (Make-Agilent) with dimension 30 m length and 0.32 mm of internal diameter and coated with stationary phase of 1.8 μ m film thickness. Details of other optimized gas chromatographic and headspace parameters are given in Table 2 and Table 3 respectively. For suitability of a system, tailing factor kept was not more than 2.0, theoretical plate not less than 5000 and % RSD not more than 5.0% was kept for peak area of each solvent for six standard solution replicate injections.

Table 2:- Optimized gas chromatographic conditions.

Detector	Flame Ionization Detector
Column oven temperature program	Initial 40°C, hold for 8.0 minutes
	Increase @ 20°C per minute to 200°C
	Hold at 200°C for 4.0 minutes
Detector Temperature	220°C
Injector Temperature	200°C
Attenuation	-6
Split Ratio	10:01
Carrier Gas	Nitrogen
Carrier Gas Flow	1.50 mL/min
Run time	20 min
Range	01

Table 3:- Optimized headspace conditions.

Oven equilibration temperature	100°C
Needle temperature	110°C
Transfer line temperature	120°C
Thermostat time/Vial incubation time	30.0 minutes
Pressurization time	3.0 minutes
Injection time	0.05 minutes
Withdrawal time	0.5 minutes
GC cycle time	34.0 minutes

Preparation of solutions:-**Blank solution:-**

Transferred 1.0 mL of diluent into headspace vials and sealed the vials immediately using PTFE septa with aluminium crimp cap.

Standard stock solution:-

Transferred 5.0 g of Acetone, 0.410 g of Acetonitrile and 5.0 g of Ethyl acetate into a 100 mL volumetric flask containing about 20 mL diluent and made up to mark with diluent.

Standard solution:-

Transferred 1.0 mL standard stock solution into a 100 mL volumetric flask and made upto mark with diluent. Transferred 1.0 mL of diluted solution into headspace vials and sealed the vial immediately using PTFE septa with aluminium crimp cap.

Test solution:-

Transferred 0.10 g sample into a headspace vial, added 1.0 mL of diluent and sealed the vial immediately using PTFE septa with aluminium crimp cap.

Procedure:-

Conditioned the column at 200°C and then equilibrated at 40°C. Injected blank solutions, six standard solution and test solution. Recorded the peak area of each solvent and calculated solvent content in Neostigmine Methylsulfate by formula below

$$\text{Ethyl acetate/Acetone/Acetonitrile (ppm)} = (\text{AT/AS}) \times (\text{WS/WT}) \times 100$$

Where, AT is area of solvent in the test sample, AS is average area of corresponding solvent in the standard solution, WS is weight of corresponding solvent taken for standard solution preparation (g) and WT is weight of Neostigmine Methylsulfate test sample (g).

Analytical Method Validation:-

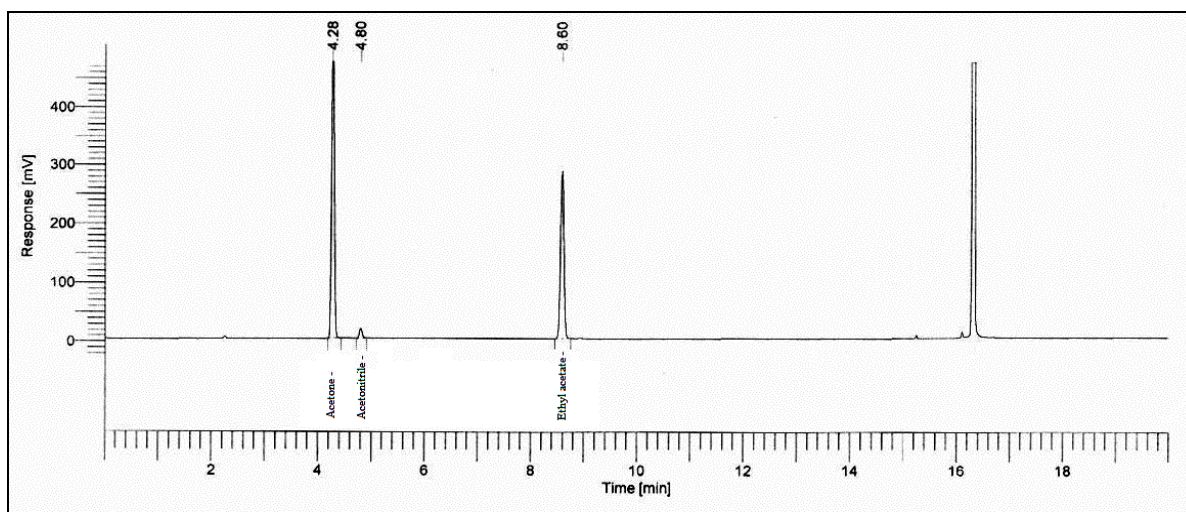
The analytical method validation work was conducted according to the ICH (International Conference on Harmonization) guidelines and with reference to other literature [10-16]. The parameter with which analytical method is validated is Specificity, Limit of detection, Limit of quantitation, Linearity, Accuracy, Precision and Robustness.

Specificity:-

Capability of the method to measure the analyte peak (solvent) response in the presence of other components is termed as specificity. For this, blank, standard, test sample and spiked test sample solutions were injected and observed the chromatogram for any interference from blank and test sample peaks at a retention time of solvent peaks. It was observed that there was not interference at a retention time of solvent peaks (Fig. 3). Also the retention time of the solvent peaks for standard and the spiked test sample were matching (Table 4).

Table 4:- Retention time of solvent peaks

Solvent	Retention time (min)	
	Standard solution	Spiked test sample
Acetone	4.280	4.276
Acetonitrile	4.804	4.801
Ethyl acetate	8.592	8.592

**Fig 3:-** Neostigmine Methylsulfate sample spiked with solvents.**Limit of detection (LOD) and Limit of quantitation (LOQ):-**

For the establishment of LOD and LOQ, six standard solutions of known concentration were injected and average signal-to-noise ratio for solvent peak was determined. Concentration for each solvent was calculated for which signal to noise is 3 and 10 and reported as the limit of detection and limit of quantitation respectively (Table 5).

Limit of detection (ppm) = $3 \times C/S$

Limit of quantitation (ppm) = $10 \times C/S$

Where C is the concentration of solvent in standard solution (ppm) and S is the signal to noise ratio of that solvent peak.

Table 5:- Detection and quantitation limits.

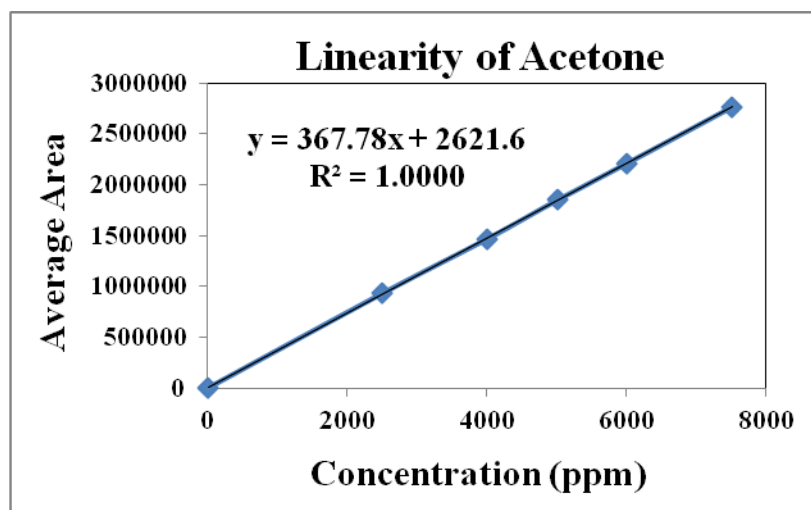
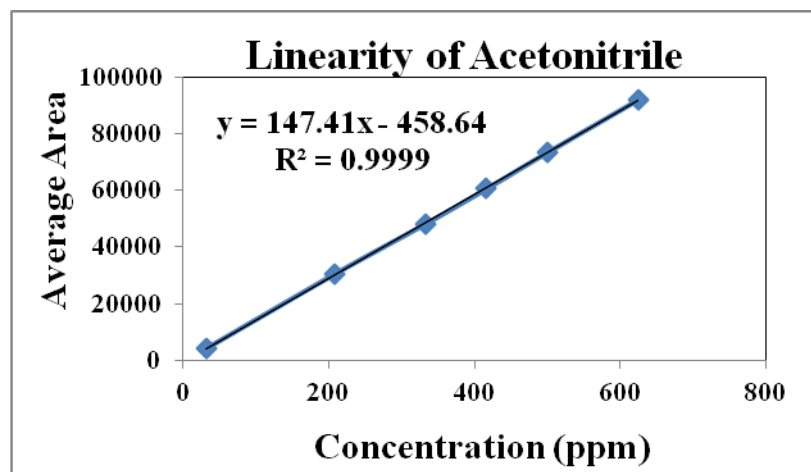
Solvent	Limit of detection (ppm)	Limit of quantitation (ppm)
Acetone	3.97	13.23
Acetonitrile	9.49	31.65
Ethyl acetate	7.87	26.23

Linearity:-

Six linearity solutions were prepared ranging from LOQ to 150% of limit level concentration of each solvent. Injected each solution in triplicate and calculated average peak area for each solvent. Linearity curves were drawn by plotting the graph of the average peak area of solvent against its concentration in ppm in linearity solutions (Fig. 4-6). Reported slope, intercept, % y intercept, regression coefficient (r^2) and range for which linearity is established (Table 6).

Table 6:- Linearity and range of solvents.

Solvent	Slope	Intercept	% y intercept	r^2	Range (ppm)
Acetone	367.78	2621.6	0.14	1.0000	13.23 to 7514.58
Acetonitrile	147.41	458.64	-0.76	0.9999	31.65 to 624.65
Ethyl acetate	232.83	2226.1	0.19	1.0000	26.57 to 7547.85

**Fig. 4:-** Linearity plot for Acetone.**Fig. 5:-** Linearity plot for Acetonitrile.

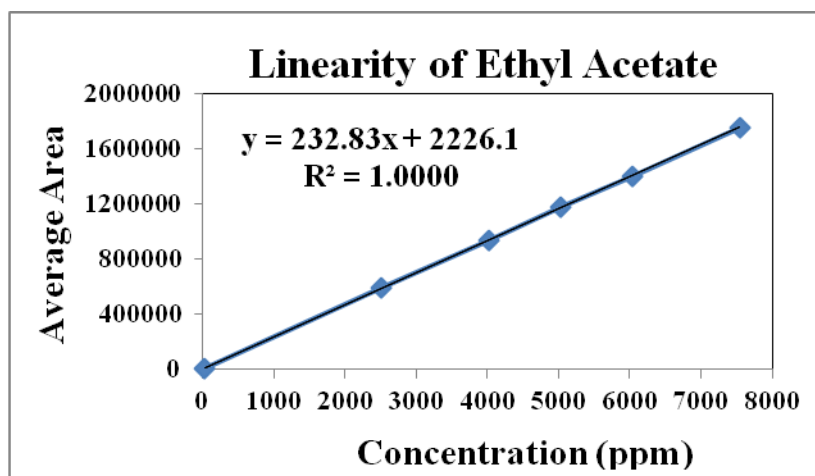


Fig. 6:- Linearity plot for Ethyl acetate

Accuracy:-

Accuracy was established by performing the recovery studies where each solvent was spiked in the test sample at four different concentration levels, i.e. LOQ, 80, 100 and 120 % of target concentration. Each level was prepared in triplicate. The solvent content for each accuracy sample was calculated as per methodology and reported as observed concentration. To calculate the recovery for solvent, the ratio is taken of the observed solvent content and the theoretical solvent content spiked. Calculated percent accuracy for each sample and reported the average for each level. Recovery for each solvent was between 80% and 120% (Table 7).

Table 7:- Recovery of Acetone, Acetonitrile and Ethyl acetate.

% Recovery	Acetone	Acetonitrile	Ethyl acetate
LOQ	94.79	109.51	103.68
80%	95.32	92.54	100.77
100%	98.14	93.19	104.03
120%	94.97	88.43	100.75

Precision:-

System precision was carried out by analysing six standard solutions prepared as per methodology. % RSD for the peak area for each solvent was found to be less than 5.0 %. Six samples were prepared by spiking each solvent with LOQ concentration and RSD for peak area was calculated which was less than 5.0 %. For repeatability (Inter-day) and intermediate precision (Intra-day and Inter-day), six test solutions were prepared by spiking the solvents at limit level concentration. RSD observed for each solvent content for repeatability and intermediate precision solution was less than 5.0 % also cumulative RSD for repeatability and intermediate precision was less than 5.0 %.

Table 8:- Precision for acetone, acetonitrile and ethyl acetate.

Parameter	% RSD for peak area		
	Acetone	Acetonitrile	Ethyl acetate
System precision (Standard solution)	1.56	1.53	1.55
Precision at LOQ	1.30	1.56	1.36
Repeatability (Intra-day)	0.64	0.96	0.58
Intermediate precision (Inter-day)	0.61	1.01	0.68
Cumulative (Intra-day and Inter-day)	0.64	1.54	0.67

Robustness:-

For robustness, two deliberate changes were done with respect to carrier gas flow rate and vial incubation temperature. Each change consists of one upper set and one lower set (Table 9). For each set, three preparations were done by spiking the solvents in the test sample at limit level and analysed. % RSD for each solvent content individually for three determinations and cumulative with six determinations of repeatability was observed for robustness solutions. RSD was found to be less than 5.0 % (Table 10).

Table 9:- Robustness parameter changes.

Changes	Lower set	Upper set
Change in carrier gas flow rate	1.4 mL/min	1.6 mL/min
Change in vial incubation temperature	95°C	105°C

Table 10:- Robustness parameter changes.

Sample ID	Individual % RSD			Cumulative % RSD		
	Acetone	Acetonitrile	Ethyl acetate	Acetone	Acetonitrile	Ethyl acetate
Robustness-1	0.53	1.26	0.43	0.82	0.99	0.84
Robustness-2	0.59	1.13	0.44	0.61	1.19	0.54
Robustness-3	0.27	0.13	0.17	0.90	0.79	0.84
Robustness-4	1.28	0.48	1.26	0.90	0.96	1.13

Result and Discussion:-

The retention time of the solvent peak of standard solution matches with that of the spiked test sample solution. No interference was observed at a retention time of the solvent peak from blank and test sample. The percentage recovery obtained for each solvent was in the range of 80% to 120%, which is within ICH acceptance. Precision parameter shows the RSD was less than 5.0 % for all the solvents in system precision, repeatability and intermediate precision at 100% concentration. Linearity was observed in the concentration range of LOQ to 150% with r^2 values greater than 0.999 and y-intercept less than 5.0% showing a good correlation between the response and solvent concentration. The calculated limit of detection and limit of quantitation for Acetone was 3.97 ppm and 13.23 ppm, Acetonitrile was 9.49 ppm and 31.65 ppm and for Ethyl acetate 7.87 ppm and 26.23 ppm respectively. The method is robust as in robustness parameter deliberate changes were made for which individual and cumulative RSD values for each set were less than 5.0 %.

Conclusions:-

Headspace gas chromatographic method developed can be successfully applied for quantitative determination of Acetone, Acetonitrile and Ethyl acetate in Neostigmine Methylsulfate bulk drug sample on a manufacturing level in the pharmaceutical industry. The method is validated and found out to be specific, linear, accurate, precise and robust, Acceptable data for all method validation parameters tested and found out to be satisfactory.

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RESEARCH ARTICLE

EVOLUTION OF “WILFUL DEFAULTS” MANAGEMENT IN INDIA: COMPENDIUM OF INSTRUCTIONS

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Abstract

India is a country which discerns between “wilful” and other defaulters. The issue of wilful default by a large number of borrowers had been receiving the attention of Government of India, Reserve Bank of India, and credit institutions since 1986. This study is an attempt to critically examine the regulatory framework on “wilful defaults”.

This paper has been divided into three phases. Phase I introduces the concept of wilful defaults and the scheme framed for disclosure of such information. Phase II was during 2001 to 2005, when the monitoring tools were strengthened and penal measures were framed. During Phase III greater transparency and accountability was brought in the process of identification of a “wilful” defaulter. A centralized mechanism of credit information and dissemination was put in place to alert the lenders against the borrowers who abuse the public funds and jeopardize the health of the institutions.

The relevance of this study at this juncture, is very crucial for credit risk management of the financial institutions in India. A thorough understanding of the policy prescriptions would enable these institutions to employ an ex-ante approach to detect early stages of strategic defaults and thus avert high costs associated with it.

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Introduction:-

“If you owe your bank manager a thousand pounds, you are at his mercy. If you owe him a million pounds, he is at your mercy” -- J.M. Keynes

Genesis:-

The issue of “wilful default” and withholding of credit facilities to defaulting borrowers, to enforce credit discipline had been receiving the attention of the regulatory authorities in India for more than three decades now. Reserve Bank of India (RBI), the central bank of the country, in 1986 had noticed that the credit institutions are sanctioning term loans to borrowers for setting up of new units or expansion of their existing businesses even when such borrowers had not respected the repayment commitments of their existing debts. Reserve Bank had, therefore, issued guidelines to all scheduled commercial banks to ascertain the cause of such defaults and delay in repayment of the principal or the interest. The guidelines further stated that the same may be condoned only if these have been due to genuine factors which were beyond the control of the defaulters. Otherwise, not meeting the financial obligations for a prolonged period indicates some serious lacunae in the financial management of the borrowing company. In such cases, it is expected that the borrower resolves the issues of the existing unit rather than

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considering expansion or enlarging his business by setting up new ones. Banks were directed not to consider fresh loans in respect of such borrowers either exclusively or jointly with other banks / financial institutions. To overcome this, banks were directed to ascertain the overdue position from other credit institutions before sanctioning fresh term finance.

With respect to working capital finance, as stoppage or withdrawal of such facility may lead to closure of the business and adversely affect the workers, the banks were directed to withheld such credits and not sanction fresh loans only when it could be established that the management of the defaulting borrowers are indulging in siphoning off the company's funds or such other malpractices. The guideline states that the banks may even freeze the operations of the account, if warranted, for bringing round the management of the company to comply with the required financial discipline. Credit institutions were also required to assess whether there is need to consider change in management of such recalcitrant borrowers.

Introduction of a reporting mechanism to Reserve Bank:-

To assess the overall position in respect of these persistent defaulters in the banking system, the Reserve Bank introduced a reporting mechanism in the year 1986. The details of the same are as under:

Table 1:-Details of annual report to be submitted by the banks.

S. No.	Periodicity	Amount and nature of the loan	Description of the report to be submitted
1.	Annually by 31 January of the following year. (First such report for the calendar year 1986 to be submitted not later than January 31, 1987).	Borrowers enjoying aggregate working capital limits of Rs.50 lakhs and above from the banking system.	From banks to Reserve Bank Action taken on: (i) the persistent defaulters in repayment of principal / interest on terms loans and deferred payment guarantees. (ii) Management of the defaulting companies who have siphoned off funds from the companies. (iii) Impact of such action on the units. (If it is a consortium arrangement, leader to furnish the report) Report to indicate: (i) nature of offence / default (ii) name of the promoter group (iii) the period of default (iv) steps taken by the bank.
2.	Annually	Borrowers enjoying aggregate working capital limits of Rs.10 lakhs and over, and up to Rs.50 lakhs	From banks to its Board of Directors In order to strengthen their internal management, the report as detailed at point number 1 above for Rs.10 lakhs and above, to be submitted annually by the banks to its Board members.
3.	Quarterly - to be submitted within fifteen days after the close of the quarter. (beginning from the quarter ending March 1987)	Borrowers enjoying aggregate working capital limits of Rs.50 lakhs and above from the banking system.	From banks to Reserve Bank At shorter interval (quarterly), report to contain snapshot of the defaults: (i) names of the defaulting borrowers (ii) nature of action taken in each case.

The lending institutions were directed to inculcate the necessary financial discipline with regard to the borrowers who abhorrently exploit the system with impunity. The impacts of such action were analyzed by these reports.

Evolution of wilful default management:-**Phase I – 1990 to 2000**

The journey of the wilful defaults management can be divided into three phases. In the first phase i.e., the period from 1990 to 2000, the Reserve Bank introduced the definition of “wilful defaults” and also framed a scheme of disclosure of information on defaulting borrowers.

Definition of a “Wilful Default”:-

RBI (1990) defined “wilful” defaults as “*defaults other than those caused by genuine factors beyond the control of the borrowers*”. During that period there was a rapid increase in number of wilful defaulters and also it was brought to the notice of the central bank that such borrowers do not co-operate with the lending institutions when appropriate corrective measures are initiated by these institutions. Reserve Bank attached considerable importance to the feedback received on the subject and reviewed the matter.

With a view to strengthen the existing monitoring mechanism, it was decided to increase the frequency of reporting from annually to half-yearly and also enlarge the scope by including the other borrowers belonging to the same group which had wilfully defaulted. With the introduction of the revised reporting arrangement, RBI advised the banks to discontinue the then existing yearly reporting. The details of the revised reporting mechanism are as under:

Table 2:-Details of the half-yearly report to be submitted by the banks.

S. No.	Periodicity	Amount and nature of the loan	Description of the report to be submitted
1.	Half-yearly reports as on March 31 and September 30 of every year. To reach RBI within a month from the end date. (First such report for March 1990 to be submitted by end of April 1990)	Borrowers enjoying fund based aggregate credit facilities (term loans and working capital) of Rs.2 crores and above from the banking system.	From banks to Reserve Bank The details of each borrower have to be submitted separately along with a summary sheet on the number of cases and names of the wilful defaulters. The details of each of the borrower had to be reported in a prescribed format with complete particulars such as name, health code, nature of the business, constitution of the company, credit facilities enjoyed by the same group, with brief remarks indicating the circumstances for becoming irregular and the measures proposed to be taken to regularise it.

RBI framed a Scheme of Disclosure of information on defaulting borrowers:-

In 1994, the issue of defaulting borrowers again came to the fore with the rapid increase of non-performing assets of the banks and financial institutions. The Honourable Union Finance Minister in his Budget Speech on February 28, 1994, announced that RBI will put in place an arrangement for circulating the names of the defaulting borrowers amongst lending institutions. This mechanism would alert and guard these institutions against borrowers who have defaulted in their dues to other lenders. Government further mentioned that RBI would also publish a list of defaulting borrowers in cases where suits have been filed by these institutions.

Accordingly, RBI framed a Scheme of Disclosure of information on defaulting borrowers of banks and financial institutions for collection / dissemination of information from / to the banking companies. RBI issued a circular under the powers vested to it by the provisions of Chapter IIIA of the Reserve Bank of India Act, 1934. Under the scheme all the commercial banks and all India financial institutions were advised on April 23, 1994 to furnish to RBI the prescribed details of defaulting borrowers with outstanding amount (both funded and non-funded) of Rs.1 crore and above which were classified as “doubtful” and / or “loss” and suit filed accounts on half-yearly basis (March 31 and September 30).

This scheme had the following objectives:-

- to alert banks and financial institutions and to put them on guard against borrowers who have defaulted in their dues to other lending institutions.
- to make public the names of the defaulting borrowers against whom suits have been filed by the banks and financial institutions.

As stated in the budget, RBI had started publishing such information in a booklet form for use of these institutions since 1994.

Establishment of Central Vigilance Commission by Government of India:-

The matter of “wilful default” received further impetus with the establishment of Central Vigilance Commission (CVC) by the Government of India in 1998. The Central Vigilance Commission Ordinance 1998 under Section 8(1)(h) directs that the power and function of the CVC will be the following:

- (a) “Exercise superintendence over the vigilance administration of the various Ministries of the Central Government or Corporations established by or under any Central Act, Government companies, societies and local authorities owned or controlled by that Government”.
- (b) “Improving vigilance administration is possible only if system improvements are made to prevent the possibilities of corruption and also encourage a culture of honesty.”

Directions issued by the Central Vigilance Commission on “wilful defaults”:-

In exercise of the above powers conferred on the CVC, it issued instructions on November 27, 1998 on “Improving vigilance administration in Banks” which *inter alia*, included directions on “Lack of communication between banks” and directed banks, financial institutions and RBI on the following for compliance:

- (a) “all cases of wilful default of Rs.25 lakh and above will be reported by banks and RBI as and when they occur or are detected.
- (b) whether a matter is a case of “wilful default” will be decided in each bank by a Committee of Officers.
- (c) RBI will circulate the information received from the banks of wilful default every three months. The data with the RBI will also be accessible directly by the banks concerned.
- (d) there should be greater intra-bank communication about wilful default, frauds, cheating cases, etc., so that the same bank does not get exploited in different branches by the same defaulting parties.”

Scheme framed by RBI for reporting the names of the “wilful defaulters”:-

Pursuant to the instructions of the Central Vigilance Commission, a scheme was framed by RBI under which all the scheduled commercial banks and all notified All India financial institutions were required to submit the details of the wilful defaulters to RBI. The scheme came into effect from April 1, 1999.

Objective of the Scheme:-

Dissemination of information in respect of wilful defaulters, amongst the credit institutions would make them more vigilant while considering requests for new or additional loans from these defaulters. The risk of plundering of public money can thus be mitigated.

Definition of “wilful default” revised by RBI:-

Hitherto the definition of “wilful default” given by RBI only indicated that if the defaults are caused other than by genuine factors, it would deem to have been “wilful”. However, in 1999, RBI expanded the scope of the definition of a “wilful default”.

The revised definition broadly covers the following:-

1. “Deliberate non-payment of the dues despite adequate cash flow and good net worth.
2. Siphoning off of funds to the detriment of the defaulting unit.
3. Assets financed have either not been purchased or have been sold and proceeds have been misutilised.
4. Misrepresentation / falsification of records.
5. Disposal / removal of securities without bank’s knowledge.
6. Fraudulent transactions by the borrower.”

Mechanism prescribed for identification of “wilful default”:-

The Reserve Bank had advised the credit institutions that *prima facie* for a default to be categorized as “wilful” it must be intentional, deliberate and calculated. Further, for the purpose of identifying the cases of wilful default, the banks and financial institutions were advised to form a Committee consisting of three General Managers / Deputy General Managers or equivalent to them. However, it was made amply clear that the wilful defaulters should be identified based on their track record and the decision of the credit institutions should not be based on isolated transactions or incidents. Further, these credit institutions need to report the cases of wilful defaults at their overseas branches also, if such disclosure is permitted under the laws of the host country. RBI also instructed that names of the “Professional Directors” and “Nominee Directors” of the financial institutions, Central or State Governments need not be reported. It was further clarified that names of the directors who are stakeholders only should be reported in the list. In case of consortium / multiple banking arrangements, the credit institutions will report wilful

defaults to other participating / financing institutions also. Further, any fresh limit / renewal or enhancement of the loan in respect of wilful defaulters can be considered only by the Board of Directors of the credit institutions.

Reporting mechanism on wilful defaulters strengthened:-

The reporting mechanism in respect of “wilful defaulters” was further strengthened by the Reserve Bank in the year 1999. The periodicity of the reporting was reduced from half-yearly to quarterly and also the threshold amount was brought to Rs.25 lakhs from the earlier figure of Rs.2 crores.

Table 3:-Details of the quarterly report to be submitted by the banks.

S. No.	Periodicity	Amount and nature of the loan	Description of the report to be submitted
1.	Quarterly – detected after March 31, 1999	All non-performing borrowal accounts with outstanding (funded facilities and such non-funded facilities which are converted into funded facilities) aggregating to Rs.25 lakhs and above.	The data has to be reported by the credit institutions to RBI in a prescribed format. The format contains all details of the borrower viz., name, address, amount outstanding, names of the directors and details of the reporting bank and branch etc.

Inclusion of “Consent Clause” in the loan agreements:-

As banks were under an obligation to maintain secrecy of their customers’ accounts, Reserve Bank (October 1999) under section 21 and 35A of the Banking Regulation Act, 1949 issued directions to all commercial banks and the notified all India financial institutions to obtain consent from the borrowers for publicizing their names if they commit default in repayment of their dues. Therefore, the circular had directed the credit institutions to include a condition to that effect in the loan agreement. However, after enactment of CICRA, this clause was withdrawn in July 2013.

Phase II – 2001 to 2005

Review of wilful default management:-

Report of the Parliament’s Standing Committee:-

The Parliament’s Standing Committee on Financial Institutions - Objectives, Performance and Future Prospects, in its Eighth Report submitted to the Parliament on December 20, 2000 had underlined the need for effective action by the credit institutions’ against the entrepreneurs who exploit the public funds and thereby jeopardize the health of the institutions. This Committee had *inter-alia* also made certain recommendations relating to wilful defaulters, diversion and siphoning of funds, change of management of borrowing companies and other related issues. The following are some of the major conclusions / recommendations of the Committee in respect of wilful defaults:

Comprehensive system of credit information:-

The Committee was of the view that all the credit institutions should have access to an efficient system of credit information. This would prevent the headstrong defaulters from availing fresh credit from other lenders despite their deplorable past. This database of credit information should be a comprehensive system and should include details of the defaulters, wilful defaulters including the information on the promoters and the group companies who have abused the public funds with impunity. Further, the Committee recommended that the ambit of credit institutions sharing the said information should also be enlarged and more institutions should be made eligible for this purpose. Further, these institutions should also adhere to the directions issued by the RBI on wilful defaulters so that uniformity in interpretation, identification, classification and reporting is ensured in the system. This would also facilitate in having a standardized format for compilation of the data in respect of these defaulters.

Strengthen the process of recovery:-

The Committee had noticed that the procedure for recovering the dues from the “wilful” defaulters is similar to that of the “genuine” defaulters. The data revealed that recovery of dues from the wilful defaulters was not very encouraging and it showed that only a small percentage had been brought back. It had to be noted that lenient view taken on such defaulters who are robbing public money would jeopardize the health of the credit institutions and would also adversely affect the economy. Hence, the committee concluded that the process of recovery and the action taken against such persistent defaulters should be strengthened.

Recommendation of penal measures:-

The Committee recommended stringent penal measures including filing of criminal cases against the companies which have siphoned off bank funds, have resorted to falsification of accounts, misrepresented and fraudulently processed their transactions. Further, it stated that promoters of such companies should be debarred from availing the institutional finance for floating new ventures for at least a period of 15 years. The report further states that wilful defaulters should not be allowed to become directors on the Board of any Government controlled or owned companies / corporations. The Committee recommends that if such defaulter is already on the Boards of these companies or corporations, immediate steps should be taken for removal of such director. Also, the credit institutions should take proactive steps to change the management of the companies which have wilfully defaulted.

Deny access to capital markets:-

The Committee has recommended that if a company is accessing the primary market for raising funds through equity or debt issue and has in its Board a wilful defaulter/s then it would be mandatory on the part of the company to mention this fact in their prospectus and offer documents. This would enable the investors to factor this point while considering investment on such issues. These steps were felt necessary to have a deterrent effect on the wilful defaulters who enter the capital market.

Recommendation on Guarantees:-

With regard to guarantees the Committee recommended that the entrepreneurs, who had the capacity to honor the guarantees but were delinquent in meeting their commitments when invoked, should also be debarred from becoming directors on the Board of Government owned and controlled companies / corporations. Further, such entrepreneurs should also be debarred from financial assistance from credit institutions for a period of 15 years.

Constitution of a working group by RBI in 2001:-

Government of India, Ministry of Finance, had desired RBI to examine the recommendations of the above report and furnish comments thereon to the Government. Hence, in May 2001, RBI in consultation with Government of India, constituted a "Working Group on Wilful Defaulters". The said group was chaired by Shri S.S. Kohli, the then Chairman of the Indian Banks' Association. After receiving the recommendations of this working group in November 2001, RBI constituted another in-house Working Group. This group examined the recommendations of Shri Kohli's Committee report and thereafter, various directions and guidelines on "wilful defaulters" were issued by the Reserve Bank to the credit institutions, by way of Circulars.

Filing of suits to recover dues from Wilful Defaulters:-

RBI in the year 2001 reviewed the information received in respect of wilful defaulters. They had observed that though there were large amounts of outstanding against these defaulters, yet the credit institutions had not initiated any legal action against them. Hence, RBI issued the following directions in May 2001:

Table 4:-Details of legal action to be taken up by the banks.

S. No.	Details and Amount outstanding	Action to be taken
1.	Cases of wilful default having outstanding amount of Rs. One crore and above	Examine each case and file suits if not already done by the institution.
2.	Cases of cheating / fraud by the defaulting borrowers having outstanding of Rs. One crore and above.	Examine each case and file criminal cases against the borrowers.
3.	Cases of defaulting borrowers having outstanding of less than Rs. One crore	Examine each case and take appropriate action including legal action.

Definition of "Wilful Default" redefined:-

RBI (2002) issued the landmark circular on wilful defaulters and action there-against on May 30, 2002. The definition of "wilful default" was redefined and it superseded the earlier definition given in 1999. RBI stated that "A wilful default would be deemed to have occurred if any of the following events is noted":

- "the unit has defaulted in meeting its payment / repayment obligations to the lender even when it has the capacity to honor the said obligations.
- the unit has defaulted in meeting its payment / repayment obligations to the lender and has not utilized the finance from the lender for the specific purposes for which finance was availed of but has diverted the funds for other purposes.

- (c) the unit has defaulted in meeting its payment / repayment obligations to the lender and has siphoned off the funds so that the funds have not been utilized for the specific purpose for which finance was availed of, nor are the funds available with the unit in the form of other assets.”

“Diversion of funds” and “siphoning of funds” – meaning of the terms:-

In the year 2002, the meaning to be construed for the terms “diversion of funds” and “siphoning of funds” was also framed by RBI as hereunder:

“Diversion of funds” would be construed to include any one of the undernoted occurrences:

- utilization of short-term working capital funds for long-term purposes not in conformity with the terms of sanction;
- deploying borrowed funds for purposes / activities or creation of assets other than those for which the loan was sanctioned;
- transferring borrowed funds to the subsidiaries / Group companies or other corporates by whatever modalities;
- routing of funds through any bank other than the lender bank or members of consortium without prior permission of the lender;
- investment in other companies by way of acquiring equities / debt instruments without approval of lenders;
- Shortfall in deployment of funds vis-à-vis the amounts disbursed / drawn and the difference not being accounted for.

“Siphoning of funds” would be construed to have occurred if any funds borrowed from banks / FIs are utilized for purposes un-related to the operations of the borrower, to the detriment of the financial health of the entity or of the lender. But RBI in its circular has stated that the decision as to whether a particular instance amounts to “siphoning of funds” or not, would be the judgement of the credit institutions, based on the objective facts and circumstances of the case.

Directions on monitoring the end use of funds:-

In 2002, RBI further directed the credit institutions to strengthen their monitoring mechanism and credit risk management. RBI had stated in its circular that these lending institutions should enhance the quality of their loan portfolio by strengthening their internal controls. The institutions in addition to obtaining certificates from the chartered accountants with regard to utilization of the loan funds, should put in place an independent monitoring mechanism with due diligence, to verify the end use of funds. The said mechanism should form part of the loan policy document and appropriate systems and procedures should be laid down by the lending institutions. Some of the illustrative measures that could be followed by the credit institutions were also indicated in this circular, for example, undertaking periodical in depth scrutiny of borrowers’ books of accounts, balance sheets and other reports. Regular inspection of his assets, periodical visits to the site, stock audit and comprehensive management audit should be taken up to detect systemic flaw, if any, on the credit administration.

Introduction of penal measures and access denied to capital market:-

To deter the growth of wilful defaults in the financial sector, RBI directed certain penal measures to be applied on the defaulters identified as “wilful”. However, the credit institutions were directed to ensure that there is no misuse and also that an isolated case is not made as a basis for imposing these provisions. RBI emphasized on putting in place a transparent mechanism for the whole process and also stated that the discretionary powers should be kept at, the bare minimum.

RBI also decided to share the list of wilful defaulters with Securities and Exchange Board of India which is the regulatory authority of the capital market to enable them to deny access of the same to such defaulters. This measure was a major step as it closed the other channel of raising funds by the wilful defaulters. These steps were applicable for all the borrowers who were identified and declared as “wilful” with an outstanding balance of Rs.25 lakhs and above.

The other penal measures which need to be initiated by the institutions were:

- Wilful defaulters to be debarred from any kind of institutional finance for a period of five years from the date of publication in the list of RBI.
- Institutions may initiate legal process or criminal proceedings wherever necessary.
- Adopt proactive approach for change of management.

- Companies not to induct wilful defaulters on its board and if such person is already a member on the Board, effective steps to be initiated immediately for removal of such member from the Board.
- Guarantees not honored when invoked by a company of the same group, such group company will also be reckoned as wilful defaulter by the lender.

The Role of Auditors:-

In order to strengthen the role of audit, RBI had directed the credit institutions to lodge a complaint with the Institute of Chartered Accountants of India, where it is observed that the auditor was negligent or deficient in conducting the audit of the wilfully defaulted company.

Introduction of Grievance Redressal Mechanism:-

Till 2003 the credit institutions did not have a grievance redressal mechanism in respect of wilful defaulters. Hence, RBI in July 2003 reviewed the position and directed the institutions as under:

- Higher functionaries of the credit institutions to constitute the identification committee to impart more objectivity to the process. Hitherto the identification committee consisted of three General Managers / Deputy General Managers or equivalent to them. In 2003, RBI revised the constitution of the said Committee and directed that the same should be headed by an Executive Director and two General Managers / Deputy General Managers should form part of the Committee, as members.
- Identification committee should specify the reasons for classifying a borrower as a “wilful” defaulter and advise the borrower accordingly. The borrower should also be provided reasonable time of say around 15 days, to represent his case, if he desires to do so.
- A system of “hearing” to the borrowers who represented that they have been wrongly classified as “wilful” was also put in place by creating a grievance redressal mechanism. The redressal team was headed by the Chairman and Managing Director of the institution and included two other senior officials as its members. Final view to be taken by this Committee. RBI (2004)
- RBI reiterated that the credit institutions should identify and declare a borrower as “wilful” defaulter with requisite evidence and support the same with proper documentation.

Observations and Recommendations of the Joint Parliamentary Committee:-

The Joint Parliamentary Committee (JPC) on Stock Market Scam and matters relating thereto was constituted on April 27, 2001, which presented its report to both the houses of Parliament on December 19, 2002. This Committee in the course of their examination *inter alia* observed that in a number of cases the funds taken from banks / financial institutions was not used for the purposes for which the funds were lent but had been diverted by the borrowing entities. In this context, the Committee examined the extant legal provisions and following were the observations:

- Banking Regulation Act, 1949 does not include any provision as to the rights and responsibilities of borrowers or depositors.
- Loan funds are governed by the agreements, contracts and other documents entered into between the banks and their customers. Thus, it is the responsibility of the lending institutions to ensure the proper end-use of funds by periodical monitoring of the borrowal accounts.
- Diversion of funds has to be considered as per the provisions of the Contract Act for civil remedy and as per the Indian Penal Code 1860 for penal recourse, if any. However, criminal law rests on the principle of *mens-rea* which requires the proof of certain degree of malafide intention at the time of commission of the alleged crime (dishonest intention, fraudulent intention).
- Sections 405 and 415 of the Indian Penal Code (IPC) relating to criminal breach of trust and cheating needs to be considered whether relevant in cases of diversion of funds.

Interpretation of Sections 405 and 415 of IPC:-

As per the judicial interpretation of implications of diversion of funds vis-a-vis provisions of Section 405, the Courts have observed that, “*If a sum of money is advanced by way of loan no criminal breach of trust is committed even if the borrower uses it for a purpose other than that for which the advance was made*”.

As regards Section 415 of IPC, in order to make an offence of cheating, the presence of dishonest intention from the beginning is necessary. In case of borrowing of funds from the bank, the proposal submitted before the bank depicts a genuine purpose for borrowing. It is only at the subsequent stage that the borrowed funds are utilized for some other purpose. Thus it would be difficult to prove the presence of dishonest intention on the part of the borrower

from the beginning and in the absence of such proof the offence of cheating as provided under section 415 of IPC cannot be made out.

Therefore, unless the provisions of IPC are suitably amended as to make such acts on the part of borrowers as an offence punishable under IPC, there is no recourse available for penalizing the borrowers.

Recommendations of the JPC Report:-

The JPC suggested exploring the possibility of (i) criminal action against wilful defaulters and (ii) obtaining a certificate from the borrower certifying that the funds have been used for the purpose for which it was borrowed.

The JPC concluded that as the activity of diversion of funds is not culpable either under the Banking Regulation Act or under the Indian Penal Code, *“it is essential that such offences are clearly defined under the existing statutes governing the banks, providing for criminal action in all such cases where the borrowers divert the funds with malafide intention. However, the Committee agreed that such penal provisions should be used sparingly and after due diligence and caution, at the same time it is also essential that banks closely monitor the end use of the funds and obtain certificates from the borrowers certifying that the funds have been used for the purpose for which these were obtained. Wrong certification, should attract criminal action against the borrower.”*

Recommendations of the Standing Technical Advisory Committee on Financial Regulation:-

RBI (2004) in consultation with the Standing Technical Advisory Committee on Financial Regulation examined the recommendations of the Joint Parliamentary Committee report of 2002. Guidelines were issued to the credit institutions on “Checking of wilful defaults and measures against wilful defaulters” and on the need for initiating criminal action against the concerned borrowers. They are:

- it is essential that offences of breach of trust or cheating construed to have been committed in the case of loans should be clearly defined under the existing statutes governing the banks, providing for criminal action in all cases where the borrowers divert the funds with malafide intentions.
- it is essential that banks closely monitor the end-use of funds and obtain certificates from the borrowers certifying that the funds have been used for the purpose for which these were obtained.
- wrong certification should attract criminal action against the borrower.

RBI further advised the credit institutions to:-

- put in place a transparent mechanism for initiating criminal proceedings.
- based on the facts and circumstances of the case, consider initiating criminal action against wilful defaulters, under the provisions of Sections 403 and 415 of the Indian Penal Code (IPC) 1860.
- With due approval of their Board, and after careful consideration and due caution, use these penal provisions effectively and determinedly.

Enactment of the Credit Information Companies (Regulation) Act, 2005:-

RBI in 1999 constituted a Working Group under the chairmanship of Shri N.H. Siddiqui to examine the possibilities of setting up of a Credit Information Bureau in India. It was recognized that in order to arrest the growth of the non-performing advances of the credit institutions, it is necessary to put in place an institutional mechanism for collecting and furnishing information on the borrowers of these institutions.

Formation of Credit Information Bureau (India) Ltd. (CIBIL):-

Based on the recommendations of Shri Siddiqui's Committee, Credit Information Bureau (India) Ltd. (CIBIL), being a company, was formed and registered under the Companies Act, 1956. CIBIL was set up by the State Bank of India in association with Housing Development Financial Corporation in January 2001. The Working Group, however, observed that it would not be possible to set up a credit information company within the existing legal framework. Therefore, with a view to provide the necessary legislative support to the business of credit information, it was proposed to enact legislation for regulation of credit information companies and to facilitate efficient distribution of credit.

The 15th report of the Standing Committee on Finance on the Credit Information Companies (Regulation) Bill 2004 was presented to the Parliament on February 25, 2005. *This Act would provide for regulation of credit information companies and facilitate efficient distribution of credit and for matters connected therewith or incidental thereto.* On

June 23, 2005 the Government of India had notified the Credit Information Companies (Regulation) Act, 2005 and the Act came into force with effect from December 14, 2006.

Reporting to Credit Information Bureau (India) Ltd. (CIBIL):-

Credit institutions had to submit the list of suit-filed accounts on wilful defaulters of Rs.25 lakhs and above as at end March, June, September and December every year only to Credit Information Bureau (India) Ltd. (CIBIL) from the quarter ended on March 31, 2003. However, they had to submit the quarterly lists of wilful defaulters where suits have not been filed only to RBI in the prescribed format.

Phase III – 2006 to 2015

Introduction of Master Circular on “Wilful Defaulters”:-

Till July 1, 2006 RBI had been issuing circulars to credit institutions from time to time, containing instructions on matters relating to wilful defaulters. Master Circular on Wilful Defaulters incorporating all the instructions and guidelines on the subject is being issued annually in the month of July since 2006.

The scope of the definition of wilful defaulters expanded by RBI in 2008:-

Pursuant to the Hon'ble Supreme Court's Order relating to writ petition Civil No.291 of 1998 titled Common Cause (A registered Society) Vs. Union of India &Anr., RBI had received a suggestion to expand the scope of definition of "wilful default". The suggestion was examined by RBI and it was decided to expand the scope of definition of "Wilful Defaulter". The additional criterion is as indicated below:

“The unit has defaulted in meeting its payment / repayment obligation to the lender and has also disposed of or removed the movable fixed assets or immovable property given by it for the purpose of securing a term loan without the knowledge of the bank / lender.”

Issuance of Certificate of Registration to four Credit Information Companies:-

Four companies were granted Certificate of Registration by RBI to carry on the business of credit information in terms of Para 5 of the Credit Information Companies (Regulations) Act, 2005 (CICRA).

1. Experian Credit Information Company of India Pvt. Ltd. (Date of Registration: February 17, 2010)
2. Equifax Credit Information Services Pvt. Ltd. (Date of Registration : March 26, 2010)
3. CRIF High Mark Credit Information Services Pvt. Ltd. (Date of Registration: November 25, 2010)
4. Credit Information Bureau (India) Ltd.(Date of Registration: March 5, 2012)

Credit Information Bureau of India Ltd. (CIBIL) has been functioning as a Credit Information Company (CIC) from January 2001 onwards and credit institutions have been reporting data to CIBIL as per the instructions of RBI.

Reporting of credit information to Credit Information Companies:-

With issuance of Certificate of Registration to four Credit Information Companies by RBI, credit institutions were instructed in 2010 to submit the list of suit-filed accounts of wilful defaulters of Rs.25 lakh and above as at the end of March, June, September and December every year to CIBIL and / or any other credit information company which has obtained certificate of registration from RBI in terms of Section 5 of the Credit Information Companies (Regulation) Act, 2005 and of which it is a member. It was further clarified that if outstanding amount falls below Rs. 25 lakhs the banks / financial institutions need not report and also in respect of cases where banks have agreed for a compromise settlement and the borrower has fully paid the compromised amount, the same need not be reported.

Dissemination of Suit filed accounts by CICs on their websites:-

In order to have a system of wider dissemination, RBI in 2011 advised the Credit Information Companies to disseminate credit information covering data supplied by credit institutions on suit-filed accounts on their respective websites.

Withdrawal of “Consent Clause”

Section 17 of the CICRA, 2005 provides for collection and furnishing of the credit information by Credit Information Companies. Hence, RBI (2013) states that CIC Act now provides statutory backing for sharing of the credit information by credit institutions to the credit information companies. Therefore, with CIC Act coming into force, the consent clause becomes redundant and hence the same need not be insisted upon by the institutions.

Reporting of data to Credit Information Companies and not to RBI:-

Till September 2014, RBI was collecting the data on non-suit filed wilful defaulters of Rs.25 lakh and above and the same was being disseminated by RBI to the credit institutions to enable them to take credit decisions after factoring in the details of this list in their credit appraisal. However, this system predates the enactment of Credit Information Companies Act, 2005 (CICRA), when there was no system of centralized credit information on borrowers. With the establishment of these four Credit Information Companies in India, such a system was now put in place. Therefore, the credit institutions were advised to furnish the requisite data to these Companies, and not to RBI. Further, the periodicity was also made monthly or more frequent basis instead of quarterly hitherto, as this would enable such information to be available to the institutions on a real time basis.

Modifications / clarifications issued in the guidelines – 2015:-

As the reporting and dissemination of the names of the wilful defaulters started strengthening and consequences of the same was felt in the economy, it also gave rise to many references / queries from the banking sector and other agencies. RBI examined all such references and the issues emanating from them, made suitable modifications in the guidelines and issued a detailed circular on the same on January 7, 2015.

Objective:-

To put in place a systematic mechanism of all the credit information available in respect of wilful defaulters and disseminate the same periodically amongst the credit institutions in order to alert and caution them from further lending to these borrowers.

Clarification on term “lender” and “Unit” in the definition of “Wilful Default”**Table 5:-**Description of the term “lender” and “Unit”

Lender	<ul style="list-style-type: none"> Covers all banks and financial institutions To which amount is due on account of banking transactions Includes off balance sheet transactions.
Unit	<ul style="list-style-type: none"> Individuals Juristic persons Any type of business enterprise Persons responsible or In-charge for the management of the affairs in case of business enterprise.

“Grievances Redressal Mechanism” now titled as “Mechanism for identification of Wilful Defaulters”

With a view to bring in accountability and greater transparency the process put in place by the credit institutions for identifying, declaring and reporting a borrower as “wilful” defaulter, has been revised. The title of the mechanism has also been reformed from “Grievances Redressal Mechanism” to “Mechanism for identification of wilful defaulters”.

The changes brought are as under:**Table 6:-** Procedure for identification of “wilful defaulters” – old and new.

Step-wise procedure of “Grievances Redressal Mechanism”	Step-wise procedure of “Mechanism for identification of wilful defaulters”
With a view to imparting more objectivity in identifying cases of wilful default, decisions to classify the borrower as wilful defaulter should be entrusted to a Committee of higher functionaries headed by the Executive Director and consisting of two GMs/DGMs as decided by the Board of the concerned bank / financial institution.	The evidence of wilful default on the part of the borrowing company and its promoter/whole-time director at the relevant time should be examined by a Committee headed by an Executive Director and consisting of two other senior officers of the rank of GM/DGM.
The decision taken on classification of wilful defaulters should be well documented and supported by requisite evidence. The decision should clearly spell out the reasons for which	If the Committee concludes that an event of wilful default has occurred, it shall issue a Show Cause Notice to the concerned borrower and the promoter/whole-time director and call for their submissions and after considering their submissions issue an

the borrower has been declared as wilful defaulter vis-à-vis RBI guidelines.	order recording the fact of wilful default and the reasons for the same. An opportunity should be given to the borrower and the promoter/whole-time director for a personal hearing if the Committee feels such an opportunity is necessary.
The borrower should thereafter be suitably advised about the proposal to classify him as wilful defaulter along with the reasons therefor. The concerned borrower should be provided reasonable time (say 15 days) for making representation against such decision, if he so desires, to a Grievance Redressal Committee headed by the Chairman and Managing Director and consisting of two other senior officials.	The Order of the Committee should be reviewed by another Committee headed by the Chairman / CEO and MD and consisting, in addition, of two independent directors of the Bank and the Order shall become final only after it is confirmed by the said Review Committee.
Further, the above Grievance Redressal Committee should also give a hearing to the borrower if he represents that he has been wrongly classified as wilful defaulter.	As regard a non-promoter/non-whole time director, it should be kept in mind that Section 2(60) of the Companies Act, 2013 defines an officer who is in default to mean only the following categories of directors: (i) Whole-time director (ii) where there is no key managerial personnel, such director or directors as specified by the Board in this behalf and who has or have given his or their consent in writing to the Board to such specification, or all the directors, if no director is so specified; (iii) every director, in respect of a contravention of any of the provisions of this Act, who is aware of such contravention by virtue of the receipt by him of any proceedings of the Board or participation in such proceedings and who has not objected to the same, or where such contravention had taken place with his consent or connivance. Therefore, except in very rare cases, a non-whole time director should not be considered as a wilful defaulter unless it is conclusively established that i) he was aware of the fact of wilful default by the borrower by virtue of any proceedings recorded in the Minutes of the Board or a Committee of the Board and has not recorded his objection to the same in the Minutes, or, ii) the wilful default had taken place with his consent or connivance. A similar process should be followed when identifying a non-promoter/non-whole time director as a wilful defaulter.
A final declaration as 'wilful defaulter' should be made after a view is taken by the Committee and the borrower should be suitably advised.	

The major changes which have been brought about in the mechanism are:

1. In addition to the borrowing company, the evidence of wilful default is being examined in respect of its promoter / whole-time director as they represent the core management of the company who are responsible for such default.
2. The reviewing committee which takes the final decision on the borrower being wilful has in addition to the Chairman / CEO and MD of the bank two independent directors of the bank now. Inclusion of independent directors in the committee makes the process more fair and unbiased.

3. In view of the limited role of non-promoter / non-whole time directors (Nominee and Independent directors) in the management of a company's debt contracts, their names shall now be excluded from the list of wilful defaulters, except in the rarest circumstances.

Clarifications issued on guarantees furnished:-

As there were references received from the credit institutions with regard to guarantees issued by individuals, group companies and non-group companies in respect of the wilfully defaulted company, hence clarifications on the same were issued by Reserve Bank in its circular of 2015.

RBI had advised that in terms of Section 128 of the Indian Contract Act, 1872, "*The liability of the surety is coextensive with that of the principal debtor unless it is otherwise provided by the contract.*" It is therefore, clarified that the credit institutions can go ahead with invoking the guarantee against the guarantor / surety even without exhausting the available remedies against the wilful defaulter, who is the principal debtor. Further, it states that when the credit institution makes a claim on the guarantor, the liability of the guarantor is immediate.

The major instructions on this aspect are as under:

Table 7:-Action to be taken by banks on guarantees

Type of Guarantee	Action to be taken by credit institution
Wilful default of a single borrowing company in a Group.	Institution to consider track record of the individual company with reference to its repayment performance to its lenders.
Guarantees furnished by the companies within the Group on behalf of the wilfully defaulting units.	If the guarantee is not honoured when invoked, such group companies should also be reckoned as "wilful defaulters"
Claim made on the guarantor on account of the default made by the wilful defaulter.	As the liability of the guarantor is immediate, in case he refuses to pay in spite of having means to make payment, such guarantor would also be treated as a "wilful defaulter".

Strengthened the role of auditors:-

Hitherto, whenever the credit institutions observed that the auditors of the borrowers were negligent or deficient in conducting the audit of the wilfully defaulted company they were instructed to lodge a formal complaint against such auditors with the Institute of Chartered Accountants (ICAI).

In 2015, RBI had further strengthened the mechanism of audits. The credit institutions have been directed that in addition to the complaint being filed with the ICAI, they should also forward the same to the Department of Banking Supervision, Central Office of RBI and the Indian Banks' Association (IBA) for records. IBA in turn, would circulate the names of these Chartered Accountant firms amongst the credit institutions, who should consider this aspect before assigning further task to these firms. RBI would also share such information with other financial sector regulators / Ministry of Corporate Affairs (MCA) / Comptroller and Auditor General (CAG).

Specific Certificate from auditors on end-use of funds:-

The credit institutions should award a separate mandate to the auditors if they desire to have a specific certificate from them regarding diversion / siphoning of funds. However, if such a certificate is called for from the auditors, the credit institution should ensure that an appropriate covenant exists in the loan agreement which enables them to award such a mandate from the borrowing company or its auditors. Credit institutions may also engage their own auditors for obtaining specific certificate on the end use of funds, in addition to their own "due diligence" in monitoring the end use of funds lent.

Role of Internal Audit / Inspection spelt out:-

The credit institutions were also directed that while conducting their internal inspection or audit of their branches periodically, they should look into the defaulters' accounts exhaustively and scrutinize carefully to check whether there are diversion and / or siphoning of funds. The cases where "wilful" defaults are found, it should be reported to the Audit Committee of the bank.

Directors' to be identified by "Director Identification Number" (DIN):-

Ministry of Corporate Affairs (MCA), Government of India has introduced the concept of Director Identification Number (DIN) with insertion of Section 266A and 266G of Companies (Amendment) Act, 2006. Hence, as per the

extant notification all the existing and intending Directors have to obtain DIN within the prescribed time-frame from the MCA.

RBI (2015) had directed all the credit institutions to include the DIN against the names of the directors so that directors are correctly identified and names of the directors who do not appear in the list of wilful defaulters are not denied credit due to the similarity in the names with that of the person who is in the said list. The institutions should carefully examine this issue while scrutinizing the credit proposals and in case of doubt should independently verify the names from other sources.

Way Forward:-

The twenty-seventh Standing Committee Report on “Non-performing assets of Financial Institutions” presented by the Ministry of Finance, Government of India to both the houses of Parliament on February 24, 2016 *inter alia* states that the “wilful defaulters owe public sector banks a total of Rs.64,335 crore, which constitutes 21% of the total non-performing assets.” As a measure of public accountability the said Committee has recommended that as a deterrent measure, credit institutions must pay attention to their top 30 wilful defaulters and make their names public. Such measure will help in recovering the existing debt from such defaulters and also prohibit these borrowers from availing further credit from the system. The report also states that to facilitate publishing of names of the wilful defaulters, if need be, the provisions of RBI Act or any other extant law governing prohibiting the same should be amended. There is need to examine the recommendations / conclusions of this report and mitigate the huge credit risk and stressed assets piled up in respect of “wilful defaults”.

A thorough understanding of the above policies and regulatory prescriptions would enable credit institutions to employ an ex-ante approach to detect early stages of wilful defaults and thus avert high costs associated with identification, classification, reporting and legal recourse undertaken by filing a suit.

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RESEARCH ARTICLE

PSYCHOSOCIAL BURDEN ON PRIMARY CAREGIVERS OF CHILDREN WITH DOWN SYNDROME

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Abstract

Down syndrome is the commonest chromosomal disorder causing mild to moderate intellectual disability. A child with disability is a serious stress factor for parents. In India persons with intellectual disability have traditionally been cared for by their families. Multifaceted factors have made parenting vulnerable to stress. Indian studies are lacking on care arrangements of children with Down syndrome and their caregiver's psychological issues. A cross sectional study was conducted to assess care arrangements of children with Down syndrome and caregiver burden and depression. 70 caregivers of children less than 18 years attending the Down syndrome clinic at Government Medical College Hospital, Thrissur from January 2013 to July 2014 were enrolled in the study. Subjects were interviewed by questionnaire method using internationally accepted measuring tools like client service receipt inventory, burden interview, caregiver activity survey and general health questionnaire. Mothers were the primary givers in 69(99%) of cases. 29(40%) caregivers experienced moderate to severe burden. Depression was detected in 44(63%). There was a significant positive correlation between caregiver burden and level of depression (correlation coefficient 0.62, p value 0.001). Severe burden was reported in caregivers having children with hypothyroidism (p value-0.026) and hearing impairment (p value-0.001). Depression was more prevalent in older mothers (p value-0.05) but less in mothers who were better educated (p value-0.005) and those who were employed (p value-0.03).

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Introduction:-

Down syndrome is the commonest chromosomal disorder causing mild to moderate intellectual disability. The worldwide incidence is 1 in 733 live births (1). A mentally retarded child in a family is usually a serious stress factor for parents. It often requires reorientation and re-evaluation of family goals, responsibilities and relationships (2). Multifaceted factors like social isolation, financial constraints, poor marital relationships, behavioral, health and developmental problems of the child and above all limited family and social support have made parents vulnerable to stress. In India the majority of persons with intellectual disability have traditionally been cared for by their families (3).

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Parents of children with Down syndrome perceived more care giving difficulties and child related stress like distractibility, demanding or unacceptable behavior. Parent related stress like incompetence, depression and health problems were also more in these parents (4).

Children with Down syndrome when intervened early by speech therapy, physiotherapy and occupational therapy and given proper medical attention for different health issues, can have a better long term outcome as compared to other genetic causes of intellectual disability.(5). Hence the need for their long term care .But this also poses challenges to the caregivers and adds to their emotional and social stress. Indian studies done were mainly on caregivers of children with intellectual disability as a broad category which also included Down syndrome. Hence this study was planned on care arrangements of children with Down syndrome and the caregiver's psychological issues and is probably the first of its kind from South India done solely on caregivers of children with Down syndrome.

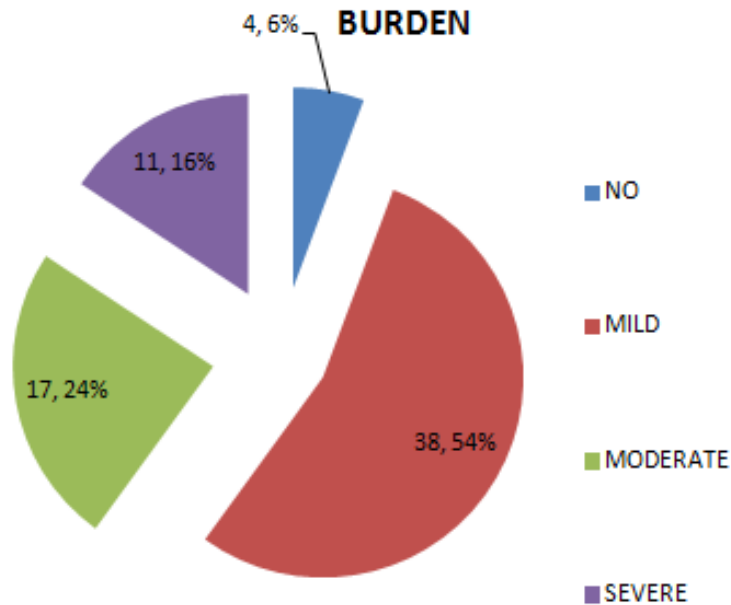
Materials and Methods:-

A cross sectional study was done on 70 caregivers of children below 18 years attending the Down syndrome clinic at Government Medical College Hospital(GMCH),Thrissur from January 2013 to July 2014 ,a period of one and a half years . Subjects were interviewed by questionnaire method using internationally accepted research methods and measuring tools, which included *client service receipt inventory*, *burden interview*, *caregiver activity survey* and *general health questionnaire*. Data was obtained by personal interview using standard proforma and consent, from caregivers of children attending Down syndrome clinic at GMCH, Thrissur. *Client service receipt inventory (CSRI)* is a research method and instrument for the collection of data on service utilization and related characteristics of people with mental disorders as the basis for calculating the cost of care. . It is a questionnaire for collecting retrospective information about study participant's use of health and social care services, usual living situations, employment and income, service receipts and medication profile(6) *The Zarit Burden Interview* has been specially designed to address the stresses experienced by caregivers of patients with dementia and chronic illnesses . It consists of a set of 22 questions to the caregivers for assessing the impact of the patient's disabilities on their life. Higher scores indicate greater caregiver distress(7). *The Caregiver Activity Survey (CAS)* is a tool developed to measure the time caregivers spend aiding Alzheimer's patients with their day-to-day activities and this would help to determine the economic impact of the illness(8). *The General Health Questionnaire* is a method to quantify the risk of developing psychiatric disorders. The instrument targets on the inability of the caregiver to carry out normal functions and the appearance of distress in order to assess well being in a person. The format is 60 item test (9). The test exists in several alternate forms: GHQ-30(30 items), GHQ-28 and GHQ-12. In this study the GHQ 12 version was employed to assess depression. The advantage of GHQ 12 is that it can be used in busy clinical settings and in situations where the patients are illiterate ,the questionnaire can be read out to them(10).These tools were modified suitably to use in caregivers of disabled children. Data was analysed using SPSS version 18.

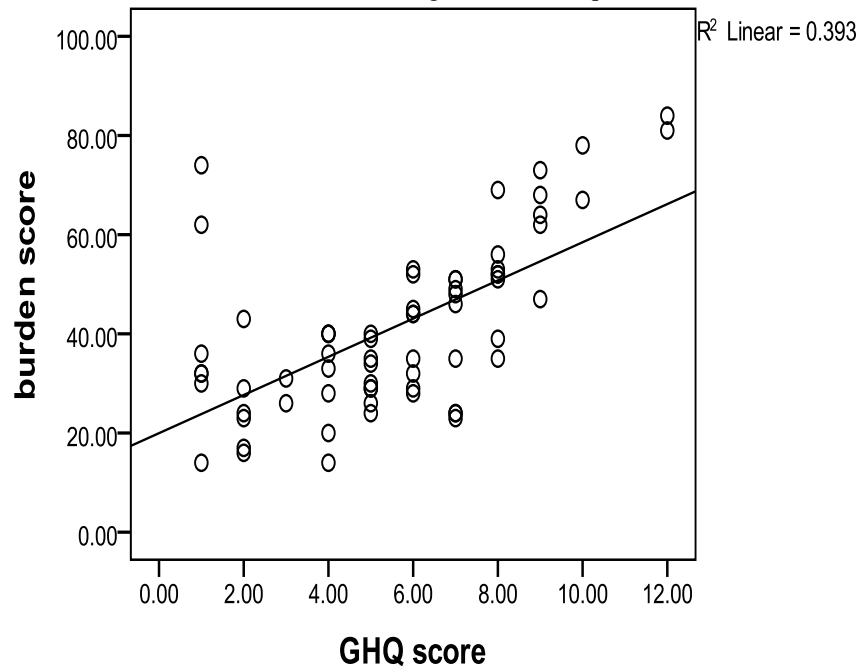
Results:-

A total of 70 caregivers were interviewed. The median age of the children cared for was **4.5** years and the male to female ratio was almost equal. Mothers were the primary caregivers in all families.. Grandparents were informal carers for 32 (46%) and mothers sole caregivers for the rest 38(64%). 40 mothers (57.1%) were more than 30 years at the time of delivery .Even though **51(73%)** mothers had up to secondary education and above, only **20(29%)** were working. Out of 22 working mothers, 14(63%) had to quit or cut down their working hours. The majority of children (68 , 96%) stayed with their parents .Out of the 38 children attending school 24(73%) went to normal schools and 14(27%)to special schools.Only 33 (47%) received any financial assistance from the government.Living conditions of many were not satisfactory. Overcrowding was found in many homes. Among the daily caregiver activities, feeding was reported to be the most time consuming part.

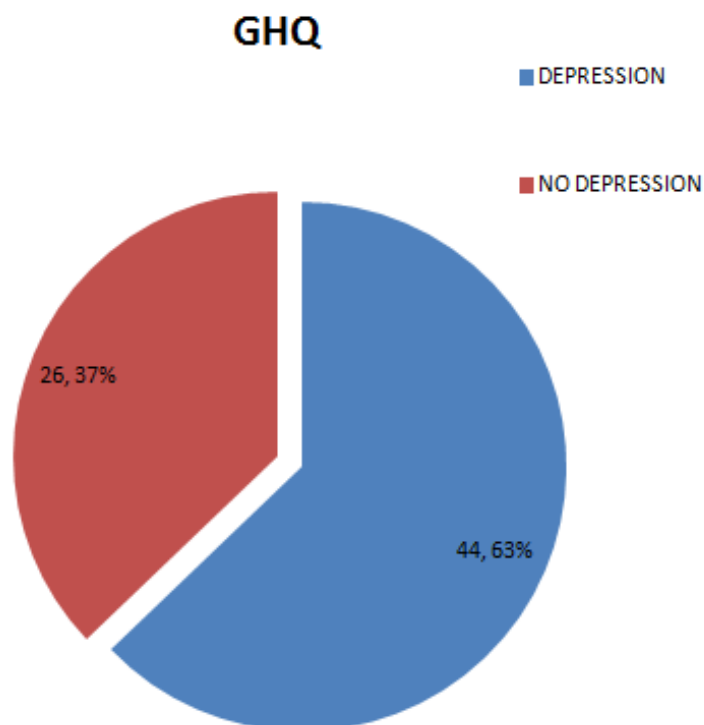
Severe burden was perceived in 11(16%), moderate burden in 17(24%), and mild burden in 38(54%). 4 (6%) caregivers perceived no burden which was assessed using Zarit Burden Interview Score.

Figure 1:- Severity of burden in caregivers as per Zarit Burden Interview.

A positive correlation was seen between increasing burden (Zarit burden interview score) and level of depression (General health questionnaire score). (correlation coeff-0.62, p value-0.001).

Figure 2:- Correlation between burden interview score and general health questionnaire score (GHQ).

Caregiver depression was detected in 44 (63%) cases according to general health questionnaire 12 score.

Figure 3:- Caregiver Depression As Per General Health Questionnaire (Ghq).

Hypothyroidism(p value-0.026) and hearing impairment(p value-0.001) in children imparted severe burden to their caregivers. Severe burden was not reported by caregivers with friend's or relative's help.(p value-0.005). Child needing more assistance in their daily activities increased burden of the caregiver.(p value-0.035).

Depression was also found to be more associated in caregivers of children with hypothyroidism (p value-0.005) & hearing impairment (p value-0.005).It was more prevalent in older mothers than younger ones. (p value-0.05) and less in better educated mothers (p value-0.005) and caregivers who continued with their jobs (p value-0.030).

Discussion:-

Most children in the study group were cared for at their homes and mothers were the primary caregivers in almost all cases. Among the mothers about two third were the sole caregivers and hence they alone had to bear the entire burden of care giving . For this , many had to quit or cut down their jobs. Among the children attending school, three fourth went to normal school which shows a change in outlook among people towards disability with their community and educational inclusion rather than segregation. Less than half received financial assistance from the government. Medical professionals need to be sensitized about the burden on the caregiver so that they adopt a more favourable approach while issuing disability certificates and pensions About 40% of caregivers experienced moderate to severe burden .In a study from India by Bhatia et al (2015)severe burden was detected in 39% of caregivers of mentally disabled children(11). Depression was found in 63%. Prevalence of depression was 70% in a study from Pakistan by Azeem et al (2013)on parents of children with intellectual disabilities (2). But in a European study by Van Riper et al the rate of depression was 30% which was comparatively lesser(12). This could be due to better economic and educational status of caregivers and their constant contact with the health care system. There they have support groups that allow sharing, contact with each other, provide information about the child's condition and assistance in choosing healthy coping strategies(12).

A positive correlation was found between caregiver burden and depression. It could be that the increasing burden contributing to depression or the depression among caregivers that made them feel burdened. In an Australian study, Jenna Serr, BS et al (2007) also found positive correlation between depression and caregiver burden. They also found that burden was more in mothers compared to fathers(13). Children with more disabilities caused more burden

and depression among caregivers (14,15,16).Hearing impairment and hypothyroidism in children increased the burden of caregivers, may be the hearing impairment causing difficulty in communication and hypothyroidism further contributing to worsening of their cognitive impairment.

Age of the mother was found to be significantly associated with depression. Older mothers perceived more depression than younger ones. But Anna J Esbensen et al(2011)in her study on North American parents found older maternal age at the time of birth of the child ,experienced less depression and better well being compared to younger mothers. It could be attributed to greater maturity and financial stability and larger social support networks among older mothers there(17).

Better educated and working mothers were less burdened and depressed. In a study by Cuskelly M et al (2014) similar finding was obtained where older and better educated parents and constant contact with health care services were associated with less depression(14).Sujatha sethu et al (2013)in her study from India found that parents' education and economic status of the family did not make any difference to the perceived stress and burden(18,19). Working mothers reported less stress and more life satisfaction despite the extra demands of work, child care and family. Working outside provided mothers with social contacts and friends, a different role, a change of daily demands and an increase in income.(4). Presence of relative's (informal carers) help was associated with lesser degree of caregiver burden. Feldman M et al (2011)in his study also had similar experiences(20,21).

Conclusions:-

Mothers are the sole caregivers in our country and they have to be involved in activities apart from care giving. Strategies focussing on the physical, mental and psychological wellbeing and upliftment of mothers have to be adopted. Fathers and other family members have to be motivated to play better roles in the care of their child with disability. The community also should be involved and support systems and groups created to reduce the burden on the caregiver. Preventive health care guidelines have been made available to both parents and doctors . Health facilities for the differently abled in government sector have to be improved ; so also the awareness and utilization of existing services Registries have to be created for different types of disabilities The caregiver should also to be included as a recipient of care.

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RESEARCH ARTICLE

SEASONAL VARIABILITY OF THE OCEAN MIXED LAYER DEPTH IN MOROCCAN UPWELLING AREAS DERIVED FROM IN SITU PROFILES.

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Mixed Layer Depth (MLD), Upwelling variability, CTD Profile, optimal threshold, gradient criterion.

Abstract

The determination of the ocean mixed layer depth (MLD), in relationship with the upwelling phenomenon, is important to a wide variety of oceanic investigations including ocean productivity, air-sea exchange processes, and long-term climate change. The MLD can be estimated using different methods including the threshold value criteria or the gradient method. The aim of this study was to create scripts combining both approaches using an optimal threshold value 0.5 °C of the temperature relative to the surface and a maximum gradient of each parameter temperature and density determined depending on its profile in water column. Moreover, we used surface distributions of Temperature, salinity, oxygen and Fluorescence in sea surface to detect the upwelling activity. The CTD data was collected along the Atlantic coast of Morocco during the winter season of 2011/2012 and the summer season of 2012. Our results revealed high seasonal variability of the MLD. The MLD is generally much deeper in winter as compared to summer all over the Atlantic coast of Morocco following the spatial and temporal variability of the upwelling phenomenon.

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Introduction:-

Understanding the seasonal variability of the mixed layer depth in the Moroccan Atlantic coast, in relationship with the upwelling phenomena in this region, which is the key to improve our knowledge about the Atlantic flow towards the Moroccan coast. The atmosphere and the ocean are connected through their limit layers where the turbulence is dominant and the MLD is strongly conditioned by air-sea exchanges (Wang et McPhaden 2000).

The mixed layer (ML) is defined as a surface layer with nearly no variation in density with depth (Clayson, et al 2000). The MLD is highly variable in time and space, ranging from 10 m in the equatorial zones and up to 400 m in the high latitude regions in winter season (Brainerd et Gregg 1995).

In the other hand, the upwelling is an oceanographic phenomenon identified by cool sea surface temperature and high concentrations of fluorescence. The Atlantic coast of Morocco is amid regions characterize by the coastal upwelling in the world (Lazier et Mann 1989).

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The upwelling observed along the Atlantic coast of Morocco is characterized by a cold sea surface temperature (SST) and high ocean productivity (Lazier et Mann 1989). This phenomenon was studied along the Moroccan Atlantic coast since the 50's. An upwelling index is calculated as the difference of temperature in the surface waters and the deep-sea water. The upwelling phenomenon is studied by the National Fisheries Research Institute, Casablanca (INRH) since 90's. They defined four major areas of upwelling activity in the Atlantic coast of Morocco (Makaoui, et al. 2005) (Fig.1).

The objective of this study is to understand the variability of the MLD and its relationship with the activity of the upwelling phenomenon in the Moroccan Atlantic coast.

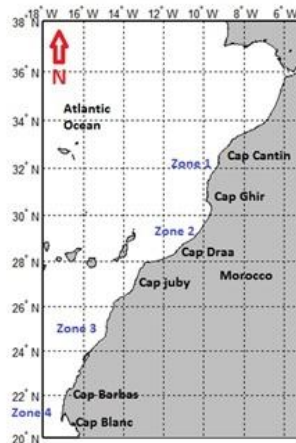


Figure 1:- The zone of Study and The four zones of upwelling along the Morocco Atlantic coast according to (Makaoui et al., 2000).

Material and Methods:-

The study area is part of the African North-West coast, between latitudes 36°N and 21°N (Fig. 1). The results of this study were based on CDT data (Conductivity, Depth, and Temperature). The data used in this study were the results of the framework of the project CCLME "Protection of the marine ecosystem of the current of the Canary Islands" (GCP/INT/023/GFF). The project was financed by the global environment facility, with the collaboration of the project EAF-Nansen «Strengthening of the base of the knowledge for the implementation of an Eco-systematic approach of sea fishing in developing countries» (GCP/INT/003/NOR). The project was realized by the FAO, two Eco-systematic campaigns were realized aboard the Norwegian N/R 'Fridjof Nansen' covering the sub-region of Guinea, in the South, to Morocco in the North. The first campaign was realized from October 20th until December 21st, 2011 and the second from May 08th until July 22nd, 2012.

The SST, salinity, windspeed, and the chlorophyll-a data are from the EU-Copernicus-Marine database. The satellite data for the SST and salinity are combined products from satellite observations (Sea Level Anomalies, Mean Dynamic Topography and Sea Surface Temperature) and in-situ (Temperature and Salinity profiles) on a 1/4 degree regular grid. The wind is estimated from ASCAT retrievals. The analyses are estimated as monthly averaged data over global ocean with spatial resolution of 0.25x0.25 degrees in latitude and longitude. For the distributions of the chlorophyll-a is estimated from the OC5ci algorithm, a combination of OCI and OC5 (Gohin, F., et al., 2008.), developed at PML. Separate chlorophyll products are produced from MODIS and VIIRS data.

Usually, because of the difficulty measuring the turbulence, the MLD is often deduced from ocean data CTD profiles. Different methods for determination of the MLD were found in literature. We can detect the MLD from the gradient of density (Keith, et al. 1995). Otherwise, the MLD is defined by the gradient of temperature (Montegut, et al. 2004) (Mignot, et al. 2009). Moreover we can define the MLD with a threshold value between 1 °C and 0.1 °C, that means that the MLD is defined as the depth in which the temperature is equal to the temperature of surface minus the threshold value.

The 0.2°C is the best value adapted to the global detection of the MLD in the seasonal and internal timescales (Montegut, et al. 2004). In the other hand, the 0.8 °C is the optimal threshold value deduced from statistical

comparisons between observations and climatology (Levitus 1982). We noticed that the threshold value is different depending on the literatures. For the detection of the upwelling areas, distributions of temperature, salinity and fluorescence are necessary, because the upwelling is detected by the appearance of cold, less salt water and a maximum of fluorescence, which are often, visualized on the surface near the coast.

Several studies have noted methods to define the MLD using either density or temperature profiles (Thomson et al. 2003)(Talley, et al. 2009)(Lorbacher, et al. 2006) and various threshold criteria (Kara, et al. 2000). In our study we defined the MLD with a gradient of Temperature and a gradient of density. In addition, we used the most popular method of the threshold value ΔT because it is simple and can be applied to profiles with various vertical resolutions (Sato et al. 2009). In our case we used the $\Delta T = 0.5^\circ\text{C}$ as the threshold value for the North Atlantic (Montegut, et al. 2004).

The threshold method is usually defined as the depth where the property has changed by a certain value from the surface, for the case of temperature

$$T(z) - T(0) = \Delta T$$

Where $T(0)$ is the value at the surface and ΔT is the specified difference criterion.

The gradient methods are defined as the depth where the gradient surpasses a specified value, for the case of density.

$$\frac{\Delta\sigma}{\Delta z} = \left(\frac{\partial\sigma}{\partial z}\right)$$

Where $\Delta\sigma$ are respectively the difference of the temperature and density over a vertical distance Δz .

The figure 2 shows the variation in temperature, and density with depth at station N°1016 (Latitude = 09 11.13 N, Longitude = 015 22.51 W, and the estimation of the MLD for the two parameters. The MLD was estimated at 28 m by the gradient method of temperature and density versus depth.

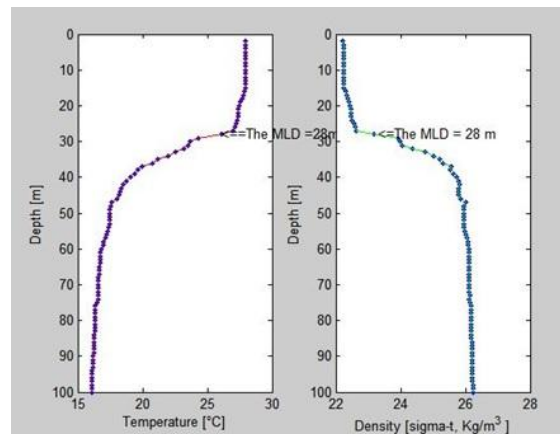


Figure 2:- Vertical distribution at 1016 station (Latitude = 09 11.13 N, Longitude = 015 22.51 W) of the temperature, and the density, plus the estimation of the MLD.

Results:-

In situ Results:-

In this work, we estimated the MLDs with the threshold value of temperature, the gradient of temperature and the gradient of density in winter and summer season. The Figures 4 and 6 presents the seasonal climatology of the Moroccan Atlantic cost MLD obtained from the analysis of individual in situ profiles. The comparison between the three methods showed similar results. In addition, we made satellite distributions of the average SST, salinity, Chl-a and wind in February 2011 and July 2012 to liken the in situ results and the satellite results.

The results revealed that in winter season, the upwelling activity detected by sea surface temperature and sea surface salinity is marked south of Boujdor cape especially between Dakhla (24°N) and cape of Blanc, where the water salinity is under 36.1psu (Fig.3). In this season, the MLD as calculated by the threshold value methods is over 50 m offshore and the depth of ML is near the surface in the South of Boujdor Cape (Fig. 4). This situation is reflected on

the sea surface chlorophyll which show a high fluorescence in large South area (Fig. 3). Contrarily to the winter season, in summer season, the activity of upwelling is both important in south and north of the Atlantic coast. Fresh and low saline water was detected along the coast (Fig. 5), following by the MLD shallowing to the surface from Cape of Cantin to Cape of Barbas as compared with winter (Fig. 6).

Satellite Results:-

This study has been conducted using E.U. Copernicus Marine Service Information. The satellite distributions of the SST (Fig. 7), show that in winter season we have temperature between 15 and 18°C we have a case homogenize temperature, but in summer season we have a large scale of temperature from 15 to 25°C. the fresh water in the cost it can be explained by the activity of the upwelling in summer season. The Figure 8 confirm this hypothesis because we have less salinity 36 PSU in the regions where the upwelling is active. Moreover, the distributions of Chl_a indicate that the peak is about 3 mg/m³ in summer and less than 1.2 mg/m³ in winter (Fig. 9). All this parameters are related to the upwelling phenomena (Makaoui, et al. 2005). In addition, we made the average wind direction and speed for February 2011 and July 2012 (Fig. 10). The distributions of the wind illustrate that we have an intense winds in summer about 6 to 10m/s from the North and the North-Est. Otherwise, in winter the intensity of wind is low, compared to summer about 2 to 8 m/s from the North and the North-Est.

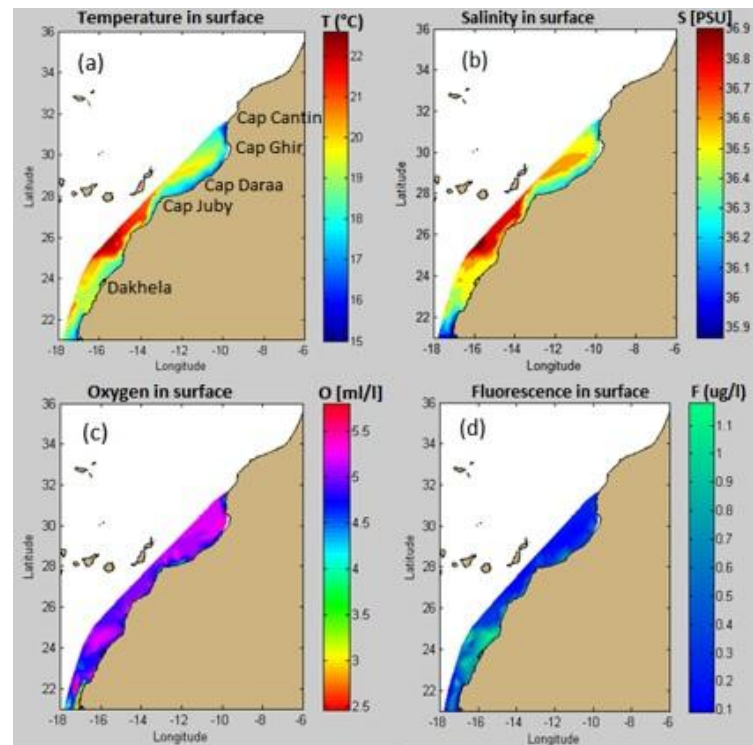


Figure 3:- Winter surface distribution of (A) temperature, (B) salinity, (C) oxygen and (D) fluorescence.

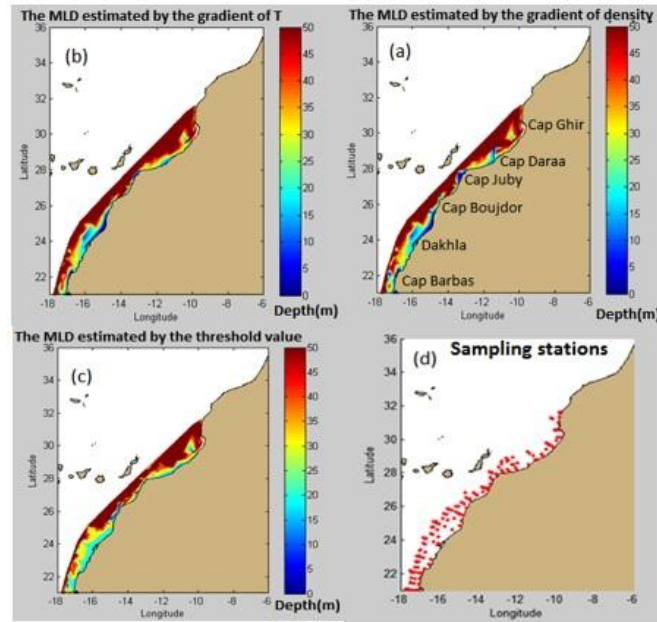


Figure 4:- Localization Of The MLD In Meter Using 3 Methods, (A) Estimation By The Gradient Of Density, (B) Estimation By The Gradient Of The Temperature And (C) Estimation Of The MLD By The Threshold Value $Dt=0.5$ °C. (D) Is The Sampling Stations In Winter.

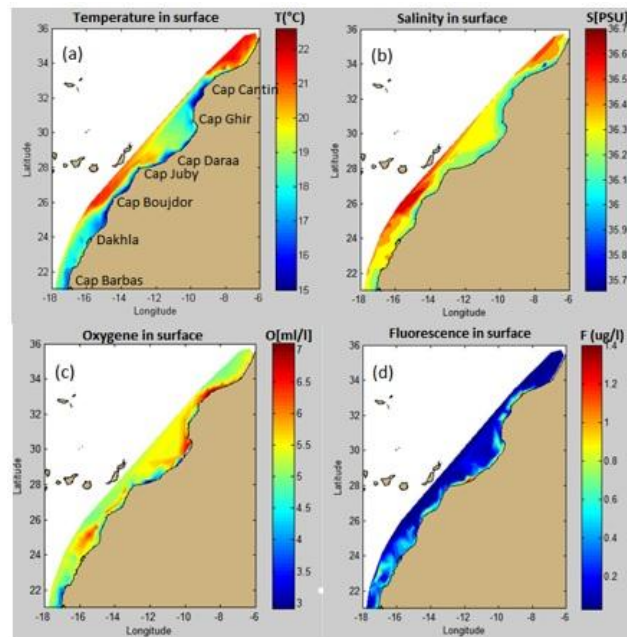


Figure 5 :- SUMMER SURFACE DISTRIBUTION OF THE OCEANIC PHYSICAL PARAMETERS, (A) TEMPERATURE, (B) SALINITY IN SURFACE, (C) OXYGEN IN SURFACE AND (D) FLUORESCENCE IN SURFACE

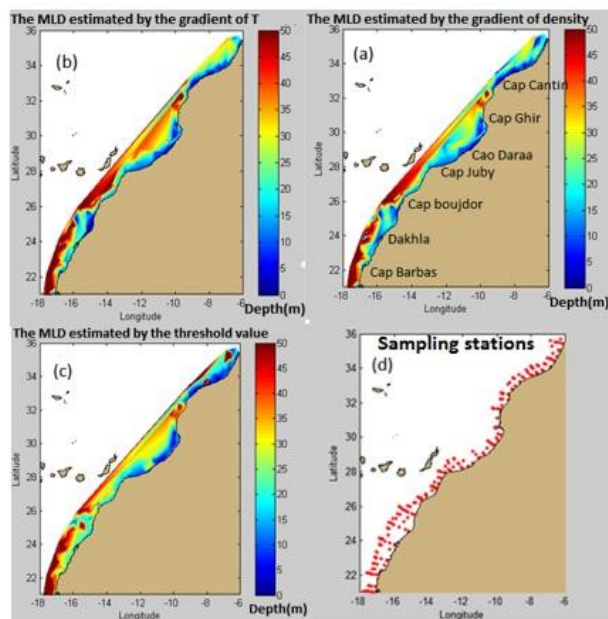


Figure 6:- Localization Of The MLD In Meter By 3 Methods Different With, (A) Estimation By The Gradient Of Density, (B) Estimation By The Gradient Of The Temperature And (C) Estimation Of The MLD By The Threshold Value $\Delta t=0.5\text{ }^{\circ}\text{C}$. (D) Is The Sampling Stations For Summer.

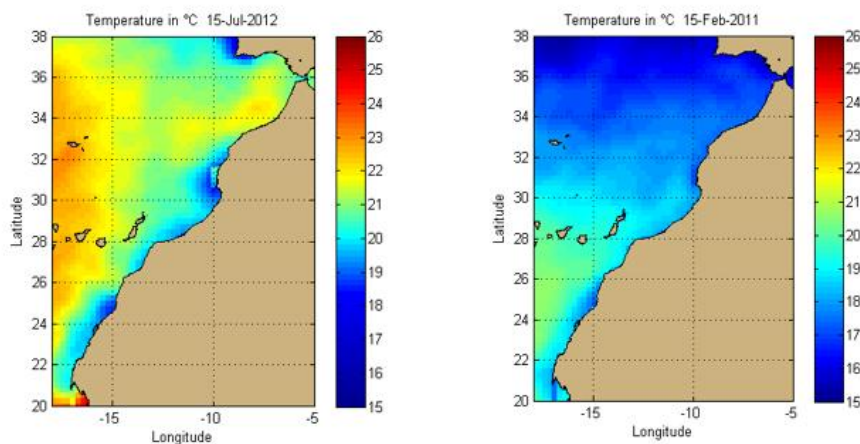


Figure 7:- Sea surface temperature ($^{\circ}\text{C}$) in July 2012 left and February 2011 right.

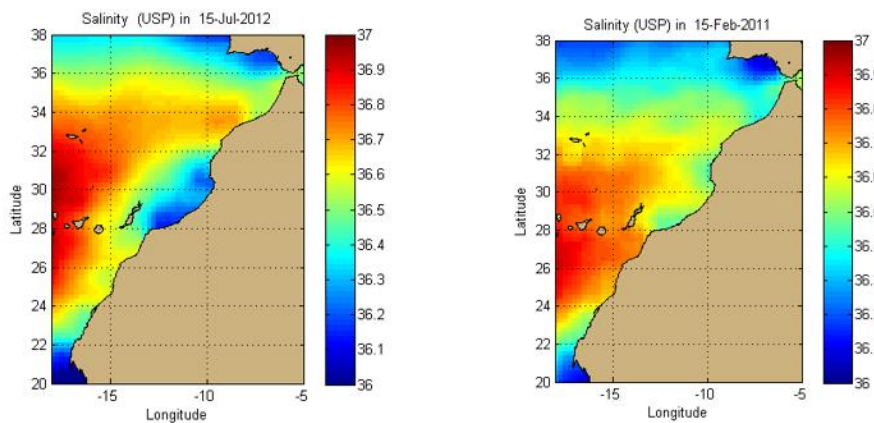


Figure 8:- sea surface salinity (PSU) in July 2012 left and February 2011 right.

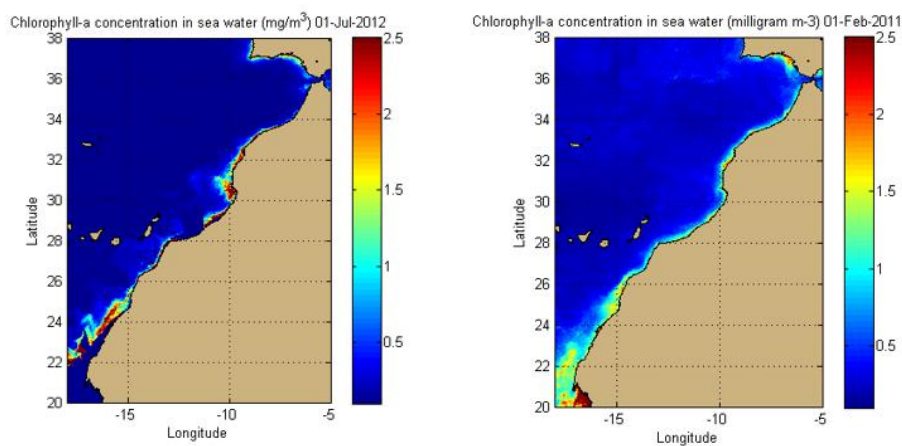


Figure 9:- Chlorophyll-a concentration in seawater (mg/m³) in July 2012 left and February 2011 right.

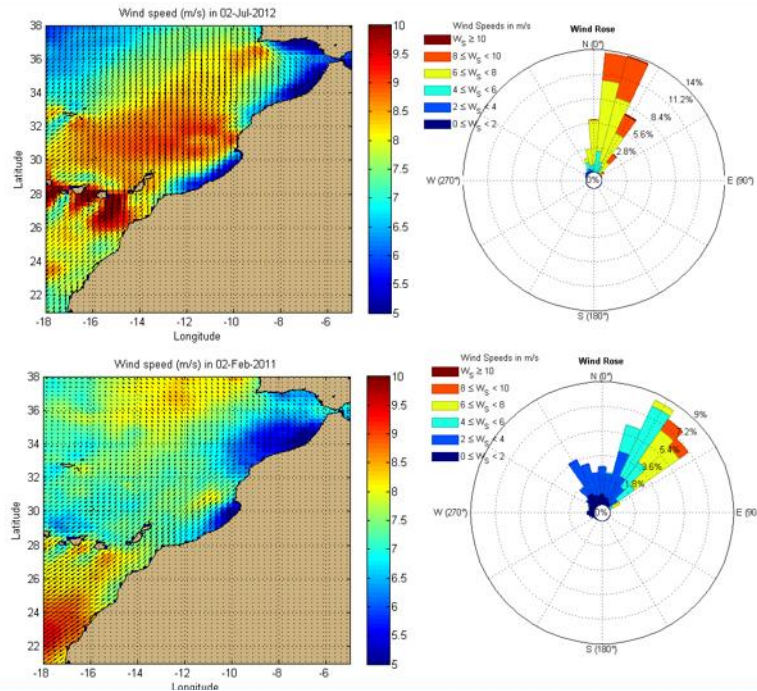


Figure 10:- wind speed (m/s) top left and top right the wind rose in July 2012, bottom left wind speed (m/s) and wind rose in February 2011.

Discussion:-

In the Moroccan Atlantic coast, we found that temperature-based MLDs are similar to density-based MLDs. This implies that salinity is a less important factor than temperature in determining the MLD distribution. On the other hand, the low salinity in the upper ocean near the Moroccan coast associated with the low depth of the ML exists. However, none of the earlier studies has addressed the relationship between the MLD and the upwelling.

Our results of the MLD in the winter exceeds 50 m. However, in the areas (-18° – -15° E and 21° – 25° N) in winter were less than 50 m, due to highly activity of the upwelling, but detailed analysis is required to confirm the relation and is beyond of our scope. In summer season, the activity of upwelling is both important in South and North of the Atlantic coast. Fresh and low saline water was detected along the coast. The MLD reached the 10m with maximum of the oxygen concentration and cold sea surface temperature. It can be explain by the cool water raised to the surface and reach of nutriment caused by the upwelling activity.

We find that overall good agreement in surface distribution between mixed layer and the fluorescence. The mixed layer shallow corresponding with the elevated fluorescence, while relatively low fluorescence concentration corresponds to deep mixed layer. It can be explained that, in summer, the activity of the upwelling contributes to the elevated fluorescence concentration and to pushing the MLD to the surface.

Conclusion:-

In this work, we studied the relationship between the MLD and the upwelling activity in the Moroccan Atlantic coast for two different seasons using two different methods to estimate the MLD in the region. Our results show that the MLD is highly variable between seasons and regions. The problem of the limited data is magnified since much of the MLD variability is linked to the seasonal cycle.

The MLD reached the surface along the coast less than 50m, according of the upwelling activity during the summer season. We found that the MLD is in a good agreement with the activity of the upwelling during this season along the Morocco Atlantic coast. While in winter season, when the upwelling is active in the South of Morocco the MLD shallow in the South areas.

The good correspondence in distribution between MLD and Upwelling activity during the season suggests that MLD climatology is important in understanding the spatial pattern of the upwelling dynamics in the Moroccan Atlantic Coast.

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RESEARCH ARTICLE

STUDY ON ECONOMIC SUSTAINABILITY OF SEWAGE SLUDGE TREATMENT PLANTS IN CHINA AND FINLAND.

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Sludge treatment, Anaerobic digestion, Energy, Transportation, Land scarcity

Abstract

Arising metropolitans, industrialization and urbanization caused urgency to solve the sludge problem appropriately in the world, especially in developing countries such as China; sewage sludge treatment technology and site selection is crucial for cost reduction; the objective of the research is to investigate the economic sustainability of Shanghai Bailonggang sewage sludge Treatment Plant and the newly constructed sewage sludge treatment plant in Changsha. The economic sustainability of a wastewater treatment plant in Helsinki, Finland is discussed as well in this paper as a contrast subject. The results have showed that increased economic burden and growth of plants globally increased amount of sludge that affected sludge treatment location scarcity to energy intensive transportation methods issuing anaerobic digestion to be vital in wastewater treatment plant to utilize energy recovery. Viikinmäki has 25 341MWh energy recovery which has officially reported to covers 60% of plant's needs and heat recovery by engines with AD are altogether 8 922MWh (100%) which is total wastewater treatment plants heat using capacity.

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Introduction:-

High rising of demand for clean water is becoming a more serious issue all over the world. Currently large scale urbanization is carrying on in China. Increasingly, more wastewater is produced; besides, water quality has deteriorated significantly over the last few decades. All these urge the municipal governments to solve wastewater and sewage sludge problem. In metropolitan cities like Shanghai some old wastewater treatment plants located at city center may have to be torn down, wastewater and sewage sludge are transported to suburb areas for treatment and disposal. The city growth and urbanization have placed a critical burden to Shanghai with sanitation and wastewater treatment. Water and power supply requirements (**Fig 1.**) have increased within 50 years massively by 32 million cubic meters of water and 1300 million kWh. Discharge of wastewater has increased annual rate of 67.7 million tons (**Cui & Shi, 2012**). All happens due to land constraint in downtown areas. As a result, super-long pipelines and powerful pump stations have to be constructed for transportation of megatons of sewage sludge from downtown to suburb areas. Within Chinese studies economics is the most important criterion in decision making and it's the most common in developing countries when considering wastewater or sludge treatment plants (**Massoud, Tarhini, & Nasr, 2009**). Commonly used indicators were globally cost of operation, technology, maintenance or

replacement (Annelies J. Balkema a, 2002). The strongest factor to technology selection has been cost, which reflected to sludge treatment in China by less interests about AD. Currently only 50 Chinese WWTP's, that is less than 5% of Chinese sewage treatment plants, used anaerobic digestion in the wastewater and sludge treatment process. Anaerobic digestion has been reported to be the lowest environmental and economic burden by significant sludge reduction in volume, thus it decreased further sludge, recovered energy, however required expensive treatment preparations (Xu, Chen, & Hong, 2014).

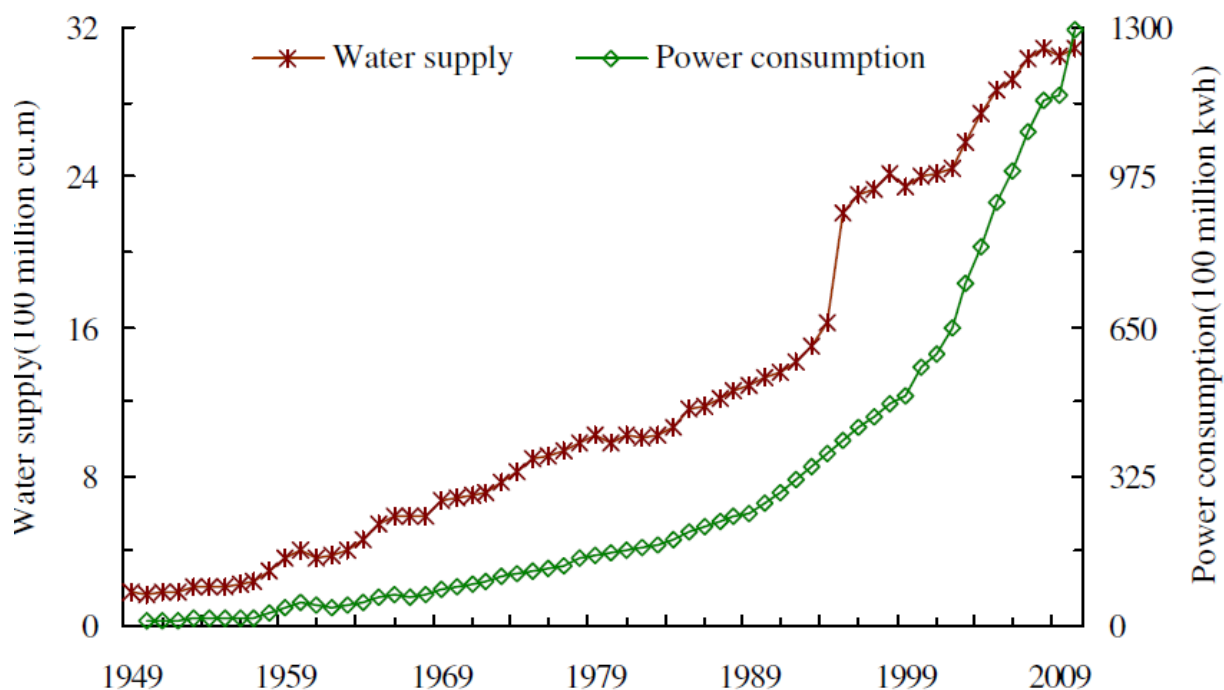


Figure 1:- Water supply increase and power consumption in shanghai from 1949 until 2009 (Cui & Shi, 2012)

Bailonggang WWTP, which is the largest WWTPs in Asia, and one of the largest WWTPs in the world is located in Shanghai and has a capacity of 2,000,000 m³/d. Initially this WWTP used chemically enhanced primary treatment (CEPT) as its technology, but it has been upgraded to a secondary treatment plant. The major technology used in this WWTP is the multi-mode A²/O (anaerobic/anoxic/oxic) process, which is characterized by its flexibility to switch and optimize flow rates of mixed liquid and returned sludge. A two-stage centrifugation process is used for sludge thickening and the sludge is dewatered to water content below 80%. In the advanced sludge dewatering process, the dewatered sludge of Bailonggang WWTP and the three adjacent WWTPs are combined and mixed with gravity-thickened sludge to reach water content of 93%, then conditioned by CaO (20%) and FeCl₃ (8%), and then pressed to reach water content below 60% by filter press. Daily sludge amount being treated: 204 tDS/d (design parameter), sludge quantity: 4080 m³/d (with MC of 80%, design parameter), 156.3 ± 23.8 tDS/d (operating parameter), daily produced methane gas: 44512 m³/d (design parameter), 16503 m³/d (operating parameter), average yearly gas production: 10.73 m³/m³ sludge with MC of 80%, methane gas yield: 0.82 m³/kg VSS, average daily gas consumption: 21106 m³/d. (Ren et al., 2015).

In contrast, the newly built sewage sludge treatment plant in Changsha suburb area receives 500 tons a day of dewatered sewage sludge with MC (moisture content) of 80% from all WWTPs located in Changsha City. The dewatered sludge is transported to this newly built plant by trucks. Then it is diluted to 86% MC with clean water. Afterwards, it enters the reactor for thermal hydrolysis. Thermal hydrolysis is a two-stage process combining high-pressure boiling of waste or sludge followed by a rapid decompression. This combined action sterilizes the sludge and makes it more biodegradable, which improves digestion performance. Thermal hydrolysis can quickly realize the reduction of sludge. The thermal hydrolysis of dewatered sludge is realized by steam injection at a temperature of 170°C for 30 minutes. The HAD of hydrolyzed sludge is done within a HRT (hydraulic retention time) of 15 days and reaches volatile reduction of more than 40% on dewatered biological sludge. The digested sludge is further

dehydrated to MC of 60% and further heated to MC of 40%, which can be applied as landfill cover soil material. (Liu Xiao Jun, 2015).

Current situation in Finland of sludge treatment has harnessed AD reaching over 50% in WWTP's that is used mostly in the largest treatment plants. The main and largest WWTP's in Helsinki region, Viikinmäki has treatment plant size of 278 011 m³/day including Suomenoja 100 191 m³/day, those treat wastewater in Helsinki, Espoo and Vantaa area. Viikinmäki sludge pre-treatment's primary sedimentation tank pumps around 3.4% DS (dry solids) sludge to 4x10,000m³ anaerobic chambers with temperature of 36-37°C for 14-20 days total by daily 2,400m³ flow rate. AD's gas is collected to 4x2,500m³ chambers to stay for 3-4 days (HSY, 2013), meanwhile sludge processed to temporary storage until dried by 4x160kW powerful, 1,700l centrifuges. These centrifuges have rated input flow of 67m³/h, 2.0tTS/h, those spend polyelectrolyte 100t annually performing 28% DS, which equals 60 000t of dry sludge (Mari Heinonen, 2016). Treatment methods of average Finnish sludge treatment are presented in table 1. eleven questionnaire treatment plants are displayed in table 2. revealing distances of sludge transportation treatment. An estimated optimal sludge content in Finnish wastewater sludge has an average of 20% DS, which equals 80% MC (moisture content) represented in Chinese data.

Methods and Techniques:-

Sludge treatment in Finland is regularly based on following the favorable treatment as following Table 1 and 2; composting (529 RMB/t), digestion with composting (483 RMB/t), thermal drying (791 RMB/t), thermophilic digestion (385 RMB/t), incineration (676 RMB/t), kemicond (368RMB/t) and lime stabilization (245 RMB/t). Kemicond process is thickening or digestion, sludge conditioning with H₂O₂ and H₂SO₄, processed with NaOH and polymers to drying by helix vise, centrifuge, belt press or chamber press. Lime stabilization utilizes burnt CaO to react with sludge reaching pH level of 12, halting biological activity and sanitizing sludge in 60°C. Kemicond and lime stabilization won't release any useful energy, composing them inefficient practices in Finland. Incineration is generally avoided, nonetheless used in conditions without proper treatment preparations. Thermophilic digestion is less likely used and commonly it has 0, 20, 40% replaced by food industry slurry. Commonly digestion in Finland had been used in half of the plants according to a table 2. however it has a secondary sludge composting possibility or thermal drying, that can utilize heat produced from biogas. Alternative heat sources respectively as follows external steam (489 RMB/t), external biogas (431 RMB/t) for thermal drying. Composting seen in table 2. is used almost always as an end product that doesn't depend on size or distance of the treatment plants (Pöyry Environment Oy, 2007). It is considered to be environmentally sustainable technology, however economic sustainability is to reproduce useful material from sludge, which is not considered in this treatment cost of composting.

Table 1:- Finnish methods for sludge treatment 80% DS. (Pöyry Environment Oy, 2007), (Elina Lohiniva, 2001)

Sewage sludge treatment technology	Operation investment (€/t)	Operating investment (RMB/t)
Gravity concentration	40	280
Anaerobic digestion	44-94	217 - 553
Dehydration	60	420
Heat drying	63-163	791
Heat hydrolysis	70-123	675
Kemicond	39-66	368
Lime stabilization	30-40	245
Sewage sludge disposal technology		Operating investment (RMB/t)
Landfilling	0-132	0-924
Land use	92-110	644-770
Composting	71-80	529

Table 2:- Finnish WWTP sludge questionnaire in 2009 (Tukiainen, 2009)

Plant number	Distance to next treatment	Unit	Sludge amount tons per year	DS %	Sludge pre treatment	Sludge after treatment
1	7	km	967	20,0 %	Centrifuge	Compost, Dumb waste
2	60	km	200	24,0 %	Thickening, Centrifuge	Compost
3	10	km	2559	24,0 %	Centrifuge	Compost
4	6	km	4800	15,0 %	Thickening, Shaftless Helix Vise	Compost
5	0	km	4900	26,0 %	Thickening, Digestion, Centrifuge	Compost
6	12,5	km	13237	17,0 %	Thickening, Centrifuge	Digest
7	16	km	9970	22,4 %	Thickening, Centrifuge	Compost
8	0,2	km	6559	30,0 %	Thickening, Digestion, Centrifuge	Compost
9	0	km	30000	22,0 %	Centrifuge	Compost, Chemical Kemicond
10	40	km	19279	31,0 %	Thickening, Digestion, Centrifuge	Compost, Digest, Dumb waste
11	40	km	60500	30,0 %	Digestion, Centrifuge	Compost

Chinese WWTP have estimated from five options: 1.Anaerobic digestion with land application, 2.Composting with land application, 3 Lime stabilization with land application, 4.Lime stabilization and landfill, 5.Incineration with landfill to apply. From those choices the most environmental friendly method was chosen to be 1.Anaerobic digestion with land application (Xu et al., 2014). Sludge treatment in China is approximately based on 71.5%, 40.6%, 38%, 5% and 3.5% by using thickening, dewatering, drying, anaerobic digestion and composting. End of life treatment process agricultural use 45%, landfill 31% and incineration 3,5% (Xu et al., 2014). Sludge in agricultural use contains a wide diversity of pathogens, therefore its use could be banned or restricted (Gianico, Braguglia, Cesarini, & Mininni, 2013). Two of the most powerful economic outcomes are disposal of sludge and income acquired from electricity sold (Tomei et al., 2016) ,noticing cost cannot exceed the benefits (Annelies J. Balkema a, 2002). Noticed 50 WWTPs (5%) in China have AD struggle with stable operation and investment costs compared with non-AD WWTPs. Those payoffs of biogas production from Chinese sludge in AD doesn't have specific conditions to economic benefits which influences to site selection and technical schemes. (Zhou, Barjenbruch, Kabbe, Inial, & Remy, 2016). In the following table 3. gravity concentration, anaerobic digestion, landfilling and land use operation investments approximately follow 20RMB/t, 38 RMB/t, 75RMB/t and 42RMB/t. The cost of capital investment gives a direction of using sludge treatment by favoring cheaper options and economic stability.

Table 3:- Chinese sewage sludge operation and capital investment of Changsha sludge treatment when moisture content reaches 95%. (Liu Xiao Jun, 2015)

Sewage sludge treatment technology	Capital investment (RMB/t·d) ($\times 10^4$)	Operating investment (RMB/t)
Gravity concentration	4.5	20
Anaerobic digestion	27	38
Dehydration	14	30
Heat drying	35	175
Heat hydrolysis	2	50
Sewage sludge disposal technology	Capital investment (RMB/t·d) ($\times 10^3$)	Operating investment (RMB/t)
Landfilling	125	75
Land use	0	42

Table 4:- Chinese sewage sludge operation and capital investment of Bailonggangsludge treatment when moisture content reaches 80%. (Liu Xiao Jun, 2015)

Sewage sludge treatment technology	Capital investment (RMB/t·d) ($\times 10^4$)	Operating investment (RMB/t)
Gravity concentration	18	80
Anaerobic digestion	72	152
Dehydration	56	120
Heat drying	140	700
Heat hydrolysis	8	200
Sewage sludge disposal technology	Capital investment (RMB/t·d) ($\times 10^3$)	Operating investment (RMB/t)
Landfilling	500	300
Land use	0	168

Techniques:-

Continuous process of sludge treatment calculates operation cost by following table 4. and equation of continuous process TR, where belt press has 500 tons/d sludge treated with moisture content of 0.99 by operation cost of 20 RMB/t that represents daily operation cost of $3,333 \text{ RMB} \times 10^3/\text{t}$. Following **Fig: 2, 3, 4, 5, 6** are using by same replicated equation. In this replicated model sludge cost depends on moisture content of previous treatment followed to the next treatment, which considers before treatment amount of sludge with process cost from table 3 in China and table 1 in Finland by noticing the MC and starting amount of sludge.

Continuous process of sludge treatment $TR = S_0 * TrC * MC_{-1}$

Where,

TR represents calculated sludge treatment operation cost or calculated capital investment S_0 represents treatment of sludge per day

TrC represents treatment unit cost for operation or treatment unit cost of capital investment

MC_{-1} represents moisture before current treatment

Table 5:- Data provided for TR calculation to Changsha, Bailonggang and Viikinmäki. (Liu Xiao Jun, 2015), (Tukiainen, 2009), (Pöyry Environment Oy, 2007)

Changsha process	MC (%)		Bailonggang process	MC (%)		Viikinmäki process	MC (%)
Start	0,99		Start	0,99		Start	0,99
Belt press	0,8		Anaerobic-anoxic-aerobic	0,99		Sedimentation	0,964
transportation	0,8		Centrifuge two stage 1	0,8		Anaerobic Chamber 1	0,85
Dilution	0,86		Centrifuge two stage 2	0,8		Anaerobic Chamber 2	0,85
Thermal hydrolysis	0,91		Thickening	0,8		Anaerobic Chamber 3	0,85
AD	0,4		Gravity thickening	0,93		Anaerobic Chamber 4	0,85
Dehydration	0,6		Conditioning	0,93		Centrifuge 1	0,72
Heat drying	0,4		Filter press	0,6		Centrifuge 2	0,72
landfilling	0,4		Landfill	0,6		Centrifuge 3	0,72
						Centrifuge 4	0,72
						Composting	0,6

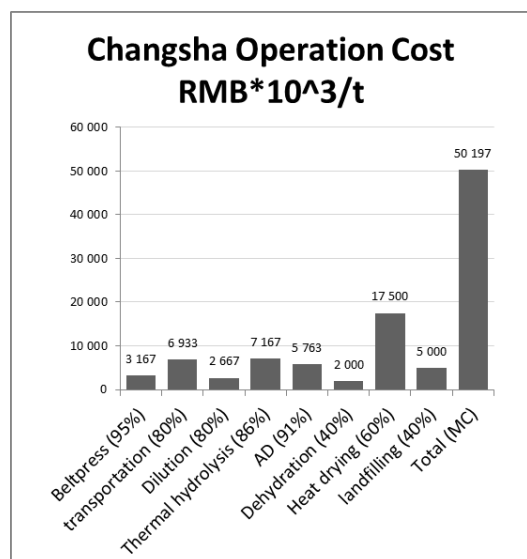


Figure 2:-Calculated operation cost of Changsha

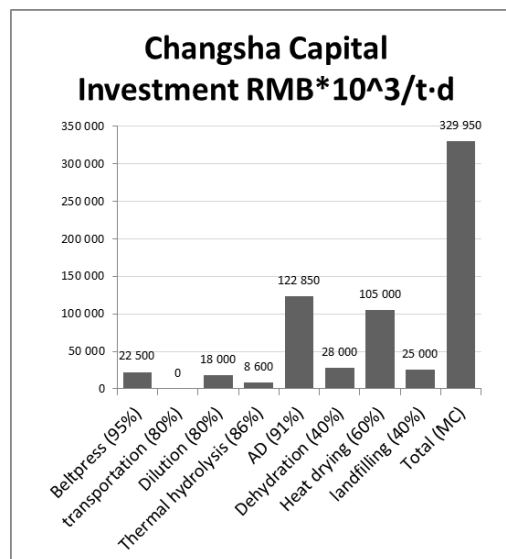


Figure 3:-Calculated capital investment of Changsha

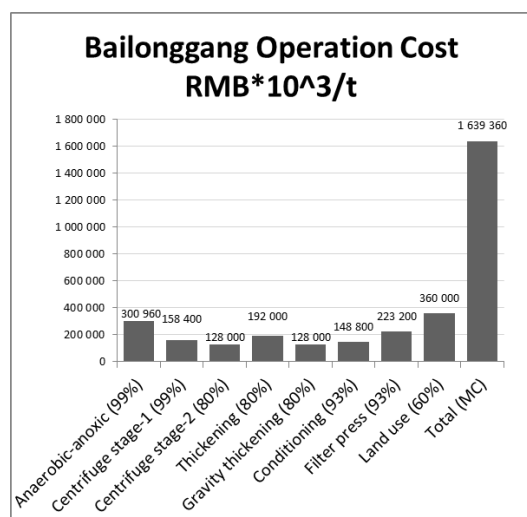


Figure 4:- Calculated operation cost of Bailonggang.

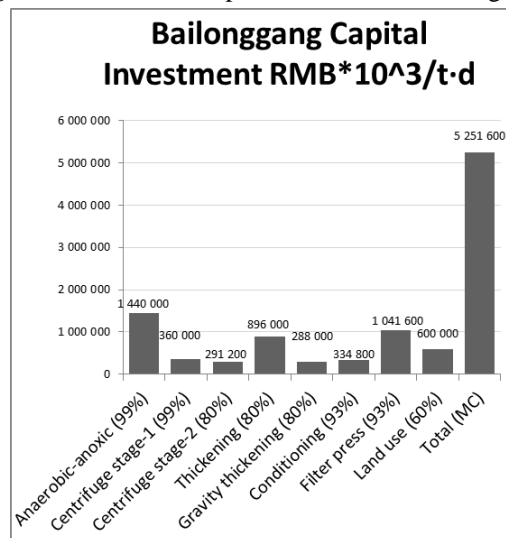


Figure 5:- Calculated capital investment of Bailonggang.



Figure 6:- Calculated operation cost of Viikinmäki.

Result and Discussion:-

The average Chinese WWTP final disposal includes thickening, conditioning, dewatering, stabilization and drying (Yang, Zhang, & Wang, 2015). As seen in Fig 2. Changsha operation cost is relatively high in heat drying and transportation. AD in Fig 2-6 have operation costs of Viikinmäki ($38,044 \text{ RMB} \cdot 10^3/\text{t}$) and Changsha ($5,763 \text{ RMB} \cdot 10^3$) those represented a significant cost difference and treatment efficiency of AD in Viikinmäki by 4 digesters. Anaerobic digestion ($270 \times 10^3 \text{ RMB}/\text{t} \cdot \text{d}$) has higher capital investment than heat drying ($350 \times 10^3 \text{ RMB}/\text{t} \cdot \text{d}$) in Fig 3, however heat drying can be energy demanding which can be seen in Fig 2. Changsha's operation costs ($17,500 \times 10^3 \text{ RMB}/\text{t}$) results. Viikinmäki's AD ($38,044 \times 10^3 \text{ RMB}/\text{t}$) is comparatively higher to Changsha AD ($5,763 \times 10^3 \text{ RMB}/\text{t}$), but Changsha capital investment in Fig 3. has fluctuation by high cost peak in AD ($122,850 \text{ RMB} \cdot 10^3$) and heat drying ($105,000 \text{ RMB} \cdot 10^3$), when compared to Fig 5. Bailonggang's, that has more stable capital investment. Changsha Fig 2. and 3. lead to a result of inefficient technical schemes, where transportation, AD and heat drying have the fluctuation higher in operation cost and capital investment. Fig 4. Bailonggang, Fig 6. Viikinmäki had more stable treatment costs and Fig 5. Bailonggang capital investment had more stabilized investments in treatment of sludge in comparison of Changsha. Those differences between Changsha, Bailonggang and Viikinmäki implicated financial shortage affected technical schemes of Changsha in constraint of the space, thus price of land raised higher and transportation became an issue. Transportation of wastewater or sludge is necessary in larger cities due to price of land constraint, when populations in cities grows bigger (Cui & Shi, 2012). "In the future centralized approaches are non-sustainable with high investment and require maintenance and update, which is burden for location selecting (Ma, Xue, González-Mejía, Garland, & Cashdollar, 2015)". Location selection and Lifetime had common prediction to whole collection systems or of parts of WWTP's had to be renewed every 50-60 years, besides the long lifetime WWTP's required many periodic of maintenances, that can be seen in electricity and heating low values in Table 6. and 7. during June, July and August. (Libralato, Volpi Ghirardini, & Avezzu, 2012)

Table 6:-Viikinmäki biogas electricity production from sludge in 2015.(Johanna Castrén, 2016).

Month	Bought, MWh	Produced MWh	Used in Process MWh	Total consumption MWh
January	1 278	2 169	3 030	3 447
February	1 083	2 086	2 810	3 169
March	1 195	2 295	3 116	3 490
April	952	2 283	2 872	3 235
May	798	2 484	2 890	3 282
June	793	2 378	2 786	3 171
July	794	2 354	2 748	3 148
August	2 572	667	2 888	3 239
September	1 093	2 140	2 864	3 233
October	1 040	2 286	2 933	3 326
November	1 291	2 033	2 941	3 324
December	1 356	2 166	3 146	3 522
Total	14 245	25 341	35 024	39 586

Table 7:-Viikinmäki Heat production by engines, boilers, HRV "Heat recovery ventilation", AD and sold to customers 2 601MWh annually.(Johanna Castrén, 2016).

Month	Produced by Engines MWh	Produced by Boilers MWh	Produced in HRV or MVHR MWh	Sold to Customers MWh
January	1 790	1 194	1 059	312
February	1 817	750	965	204
March	1 976	726	996	188
April	1 968	588	827	297
May	2 109	376	758	206
June	1 998	74	633	149
July	1 799	103	321	116
August	519	1 328	460	100
September	1 716	413	418	140
October	1 973	344	764	233
November	1 752	653	736	289
December	1 925	739	985	367
Total	21 342	7 288	8 922	2 601

Sludge treatment coordination and location selection is critical for future demands, Changsha contractors can momentarily bring a lot of sludge in which case all the sludge cannot be immediately dealt within, the storage space can end and feeding material can also become a shortage. Coordinating of sludge will also take into account the introduction of different stages and schemes of sludge to treatment plants. For example in Finland composting plant is imported from different sources, treated at the same time very wet sludge, requiring supporting material and promoting very wet and dried sludge at the same time for treatment (Tukiainen, 2009). Table 2 illustrates Finnish wastewater treatment plants sludge transportation distances, those lead to a fact treatment plants are in urban or semi distances to next treatment, whereas composting is chosen to be the most common treatment method within nearby location(Hong, Hong, Otaki, & Jolliet, 2009). Sludge treatment centralized approaches are non-sustainable with high investment, difficult maintenance and upkeep, including WWTP location selection by land scarcity(Ma et al., 2015).Decentralized approaches are more flexible, however transportation is typically in China 50-80km to be equivalent of 0,65RMB/km, respectively 52rmb of maximum transportation cost for 80km (Liu Xiao Jun, 2015). Affordable transportation distances in Finland for AD sludge and agricultural sludge are 25km's, industrial and municipal sludge distances can be 150-250km's at maximum. Table 2. sludge treatment questionnaire of schemes follow commonly at site of WWTP or nearby location between 0-15km's, estimated 20% transports sludge outside of WWTP over 15km's. Treated sludge end product as composting 0-15km's, over 15km's and external party represent 50%, 20%, 30%. In any huge scale industrial plant's sludge is worth pre-dry thermally, whether alternative option is to suburb area incineration, transportation of sludge becomes non-profitable between 50-100km distances or more.

The main transportation method from centralized plant to urban is pumping sludge, which is responsible for about 20 percent of the world's electricity consumption. (Tukiainen, 2009) Helsinki suburb area has over 500 wastewater pumping stations, 30 pumping stations located above ground and rest located underground. Pumping is an electricity consumer, used in many industrial processes and the highest electricity required process. Reduction in the amount of sludge can be clearly seen in handling costs and environmental impact, although reducing the length of the pipeline is a principle solution to minimize pumping energy consumption.

Thermal dryers in plants consume a lot of energy, this can make a fully dried sludge (>85% TS) to incineration process a possible solution as sludge disposal. Under 45% TS causes commonly a dust problem, this is for centralized plants a critical decision of sludge to pump suburbs. Transported sludge with lime stabilization increases amount of sludge. Whether quantity grows larger, preparations of ammonia recovery are necessities. Some sludge disposals in forms of incineration require large scale preparations to establish facility in any residential neighborhood and affects residents by environmental concerns (Eggimann, Truffer, & Maurer, 2015). Focusing of the pumping process is the most efficient and common way to transport sludge from place A to B. Energy intensive pumping has a side-effect, "centralized plants returning recycled water can require tremendous energy due to pumping"(Naik & Stenstrom, 2016). Pumping focuses an importance to reduce the consumption of electricity, which can be costly in certain situations. The two considerations those have the greatest impact on the final economic outcome are sludge disposal cost and income deriving from electric energy sale. In average 75% of wastewater capital needs are for pipe repair (Ma et al., 2015) however electricity use accounts for approximately 80% of drinking water treatment distribution cost and 25%–40% of the operating budgets for wastewater facilities. Urbanization, land scarcity, cities growing bigger, the longer the distances to transport water and the higher the energy demand becomes(Ma et al., 2015).

There is a strong dependency on electrical energy supply, total investments in China have increased by 99.08% and 2.78 times nearly in ten years (Zhang et al., 2015). Scheme of a typical Chinese WWTP uses 60% of energy in water treatment, those 40% left are for sludge treatment. (Liu Xiao Jun, 2015). Table 6 and 7 indicate produced electricity and heat of Viikinmäki, which officially can cover 60% of electricity in whole wastewater treatment process and 100% of heat consumption that is officially announced by Viikinmäki. Provided data in wastewater and sludge process is $25341\text{MWh} / 35\,024\text{MWh} = 72\%$ percentage of electricity in this WWTP process according to data in table 6, which is revealed to be 60% by Viikinmäki WWTP.AD-model from Viikinmäki's 60% recovering would cover the scheme of typical Chinese WWTP's scheme or alternatively sludge treatment with 20% energy leftover to be used for example in pumping process. Energy as resource to trade cost or buyback in operation process represents a huge possibilities whether treatment plants last longer than pipe-chain of life.Nowadays the in-plant pumping and aeration in common primary and secondary treatment often consists of more than 60% of the total plant electricity use. The further sludge treatment and disposal is also energy-intensive because of dewatering. Viikinmäki's biogas electricity production line in table 6. focuses AD into 4 digestion chambers. Chambers pay-off (25 341MWh) of the

energy produced in whole wastewater treatment process, rest of the energy is bought from local district electricity (14 245MWh). Comparing the overall production of biogas to overall consumption of electricity in treatment plant can utilize around 72% of electricity (Johanna Castrén, 2016). Total consumption of WWTP is (39 586MWh), which uses (35 024MWh) in main process. Leftover electricity is counted as in maintenance, backup electricity or used in service tasks. Energy loss in this situation can be estimated as building heating or upkeep of the control service systems.

Conclusion:-

China favors low cost medium efficiency treatment methods, whereas AD is useful in the future for large scale and medium scales WWTP's. Anaerobic digestion investment in China is expensive according table 3 and 4, moreover it's more expensive in Helsinki Viikinmäki with energy recovery by 60% announced officially by Viikinmäki that is 39 586MWh of electricity and heat recovery measured in 2015 by 40 153MWh annually, which covers 100% heating process in total wastewater plants usage. Digestion cost of sludge treatment is generally more expensive in Finland compared to Changsha or Bailonggang and more efficient, when moisture content is considered to be equally same in Viikineva, Bailonggang and Changsha. However large investments and controlled anaerobic digestion process have huge benefits in sludge treatment process for energy recovery and controlling operation cost. Investing incineration or AD to nearby WWTP commits a possibility to utilize energy recovery that can reduce burden of transportation or sludge volume reduction. Transportation of sludge by trucks is causing extra costs in Changsha that can be avoided by structuring nearby sludge treatment plant considering example of Viikinmäki WWTP, which has over 500 wastewater pumping stations. After all growing cities and treatment plants of Bailonggang and Changsha will face land scarcity in the future, which would need preparations to relocate treatment plants to suburb areas.(van Afferden, Cardona, Lee, Subah, & Muller, 2015) Technical schemes in the two Chinese WWTPs might not be reasonable enough from the energy recovery and economic perspective, but for today's China, those combinations of technologies may be the only reasonable way to solve the sewage sludge problem without scars due to financial shortage, constraint in the space of the existing WWTPs, sky-high land price and land scarcity in Chinese metropolitan areas.

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RESEARCH ARTICLE

LEGAL PROTECTION OF CONVENTIONAL CRIME VICTIM (RESTORATIVE JUSTICE APPROACH).

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Abstract

The rights of crime victims, particularly crime victims or conventional crime in relation to the efforts to achieve restorative justice in the criminal justice system in Indonesia. This research was a socio-legal research, which orientation is focused on legal and non-legal aspects and the operation of law in society. The results of the research indicates that the essence of legal protection for crime victims in the handling of conventional crime in Indonesia is not yet optimal to take attention and gives legal protection to conventional crime victim in achieving fairness, expediency and legal certainty as a legal purpose. Conventional crime victims are an integral part of a legal system, which received less attention in obtaining their rights in the criminal justice system in Indonesia. Legal protection of conventional crime victim in Indonesia has been set in legislations, but it is not optimal because the mostly of legislation is more focused of legal protection to the perpetrators rather than the victims. As a result, the implementation of legal protection for conventional crime victim is not optimal, so it is very important the understanding of community, the victim and also law enforcement officers in the implementation of rules and mechanisms of legal protection against conventional crime victims to get their rights.

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Introduction:-

Indonesia is a constitutional state. It is implied in the Preamble of the 1945 Constitution of the Republic of Indonesia which makes specificity that Indonesia intended to protect the entire Indonesian nation and homeland, promote the welfare, educating the nation, and participates in establish the world order.

Indonesia as a constitutional state provides protection for its citizens by establishes an institution that able to provide justice in the form of free judiciary and impartial. It is based on the argument that every human being from birth bears the rights and obligations that are free and rights, and the holding power of a country should not diminish the meaning of freedom and the humanity rights. A form of protection against the society which should be done by the state is to provide legal protection through a process of judicial in a crime act or called as criminal justice system.

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A party who very wanted the protection in a crime is a crime victim. The victim has important role to be given attention and protection in the criminal justice system. This view is based from the idea that the victim as aggrieved party in a crime, so it should receive the attention and care in order to provide protection against their interests.¹

In Indonesia, criminal justice system in general put the interests of victims are represented by the public prosecutor, as well as the wider community. Victim losses may be material that can be measured by money, and immaterial includes fear, pain, sadness, surprise psychic and others. Victim protection in the form of compensation for material loss could be charged directly to the perpetrator, but for the restitution of immaterial losses in some countries (if the perpetrators who cannot afford), then the burden to the state.²

Provisions regarding the protection of victim crime in the Code of Criminal Procedure are more dominant in the provision regarding restitution, by grouping into 4 (four) forms of victims rights, as follows:³

- a. Right to control the actions of investigators and public prosecutors, namely right to object to the termination of investigation and/or prosecution in their capacity as an interested third party as stipulated in Article 109 and 140 paragraph (2) Criminal Code Procedure;
- b. Victim's rights in their position as a witness, as stipulated in Article 168 of Criminal Code Procedure;
- c. Rights for victims' families in case the victim dies, to allow or not the police to conduct a post-mortem or grave digging for autopsy. These rights are stipulated in Article 134 and 136 in the Criminal Code Procedure;
- d. Right to demand restitution for losses suffered as causes of criminal acts in their capacity as the suffered party as stipulated in Article 98 and 101 in the Criminal Code Procedure.

The disadvantages of provisions the Criminal Code Procedure in regulating the victim in view of Barda Nawawi Arief,⁴ as follows:

- a. There is no legal effort to be conducted by the victim if the victim is not satisfied with a court decision. In contrast with the suspect that can make appeal or judicial review. Victims are represented by the prosecutor as a public prosecutor can only accept the decision.
- b. The protection of criminal victims is only stipulated in Chapter XII (Article 98-101) of Criminal Code Procedure, which allows the combination of restitution case to the criminal case.
- c. Article 99 of the Criminal Code Procedure asserts that the restitution which can be decided by the judge is only the costs incurred by the suffered party; while the other losses can only be sued through the civil judicial process takes a long time and begins a new procedural.
- d. The judge can set a specific requirement that the convicted person within a certain time, which is shorter than the probation, having to retribute the whole or part of losses that caused by the criminal act or in other words that the victim's rights as result of criminal acts only applies when the judge sentenced him/her to probation, while in a crimes that cause huge losses or violent, probation difficult to be given.
- e. Cost charged to the perpetrator for restitution is only restricted to the value that is materially.

The disadvantages indicate that the existence and the legal position of crime victims in the criminal justice system is not favorable to crime victims, due to the fundamental problem, namely the victim is only as a witness (complainant or victim). Victims are not included in the elements involved in the criminal justice system; it is not similar to the dependant, the police and the prosecutor.

In principle, the protection of victim has been stipulated in several laws in Indonesia as the embodiment of human rights in the constitution and the rights of victims in the Criminal Code Procedure. The criminal justice system prioritizes the protection of human rights, but if the provisions concerning it be in-depth considered, only the rights of suspects/defendants that many prominent, while the rights of crime victims is very little regulated.

¹Anshari Dimiyati.(2013). *Kebijakan Perlindungan Hukum Terhadap Saksi dan Korban*. Tesis. Universitas Diponegoro : Semarang. p. 3

²Lies Sulistiani. (2010). *Perlindungan Saksi dan Korban di Indonesia*. Press Release No.02/LPSK/PR/II/2010. Lembaga Perlindungan Saksi Republik Indonesia: Jakarta. p. 4

³Muhadar et al. (2010). *Perlindungan Saksi dan Korban dalam Sistem Peradilan Pidana*. Putera Media Nusantara : Surabaya. p. 21

⁴Ibid. Page. 6

Crime victims who are basically a party that suffered in a criminal act, because it has not gained as much protection as provided by the Act to the offender. Crime victims are placed as evidence that provide information, i.e only as a witness, so little possibility for victims to obtain flexibility in fighting their rights. Victims are not given the authority and not actively involved in the process of investigation and trial, so he/she lost the chance to fight their rights and restore their condition as result of a crime. The condition raises a tendency that the right of victim is not protected when the offenders have been sentenced to criminal sanctions by the courts, whereas the issue of justice and respect for human rights does not only apply to offenders but also crime victims. Therefore, the authors are interested in conduct further research and study on the rights of crime victims, particularly crime victims or conventional crime in relation to the efforts to achieve restorative justice in the criminal justice system in Indonesia.

Method of Research:-

This research was a socio-legal research, which orientation is focused on legal and non-legal aspects and the operation of law in society. Law is conceived not only in the normative dimension but also as an empirical phenomenon that can be observed in the context of reality in the community. In other words, this research examines law both in the aspect of *law in books* and *law in action*. The main objective of this research was to examine whether a normative rule (postulate) can or cannot be used to solve a legal problem in reality (in concreto).⁵

This study describes a problem in the community, the procedure applicable in the community and situations, attitude, views, ongoing process, effects of a phenomenon, careful measurement of the phenomenon in the community. The author developed the concept, gather facts, but did not test the hypothesis. Fieldwork was conducted by using in-depth interviews to obtain data in the form of views, thoughts and opinions of the informants. Primary data were intended to obtain confirmation as well as complementary research based on the library study.

Conception of Legal Protection:-

Guilarmo S. Santos⁶ argued that the concept of constitutional State is understood as a condition in a society where the law in a democratic State is determined by the people which are nothing other than the regulation of relationship between the people. Furthermore, Bothink in Ridwan H.R.⁷ argued that the constitutional State is a State where the freedom of power holders is restricted by law. The other view as proposed by Burkens as quoted Attamimi⁸ argued that the constitutional State (*rechtsstaat*) are simply as the State that put law as the basis of State power and the implementation of the power in all its forms is done under the rule of law. H.W.R. Wade⁹ argues that in a constitutional State, everything must be done according to law; it specifies that the government must be subject to the law, not the law should be subject to the government.

Constitutional State in Munir Fuady's view¹⁰ is an administration system that is governed by the law applicable and equitable arranged in a constitution, in which all people in the country, both the governed and the governing, should be subject to the same laws, so everyone is treated same and different people are treated differently on the basis of distinctions rational, irrespective of color, race, gender, religion, region and confidence, and the governments' authority is limited by a principle of distribution of power, so that the government does not act arbitrarily and not in violation rights of the people, hence to give the public an appropriate role and its role in a democratic capabilities.

Budiyanto,¹¹ the theory of constitutional State is generally divided into 2 (two) types, i.e the theory of formal constitutional State and the theory of material constitutional State. *First*, the theory of constitutional State was pioneered by Immanuel Kant. His theory resulted in a country is passive; it means that the duties of a State is only maintaining order and security in the country, or the country simply as "*night watchman*", whereas in social and economic affairs, the State should not interfere. *Second*, the theory of material constitutional State (*welfare state*) was pioneered by Kranenburg. This theory states that countries other than the duty to foster public order, and it

⁵Bambang Sunggono. (1991). *Metode Penelitian Hukum*. PT RajaGrafindo Persada. Jakarta. p. 91.

⁶As cited in Mushammad Asrun. (2001). *Krisis Peradilan Mahkamah Agung di bawah Soeharto*. ELSAM. Lembaga Studi dan Advokasi Masyarakat : Jakarta . Pages. 40-41

⁷Ridwan, H.R. (2007). *Hukum Administrasi Negara*. PT RajaGrafindo : Jakarta. p. 49

⁸Hamid A. Attamimi. (1990). *Peranan Keputusan Presiden RI Dalam Penyelenggaraan Pemerintahan Negara*. Dissertation. Graduate School Universitas Indonesia : Jakarta. p. 8

⁹H.W.R Wade. (1970). *Administrative Law*. Third Edition. Clarendon Press : Oxford University. Page. 6

¹⁰Munir Fuady. (2009). *Negara Hukum dan Demorasi*. Kencana Prenada Group : Jakarta. Page. 3

¹¹Budiyanto. (2000). *Dasar-dasar Ilmu Tata Negara*. Erlangga : Jakarta. Page. 50

also responsible for developing and realizing the welfare of people. This theory is widely practiced in developing countries, such as Indonesia.¹²

As definitions above can be said that the definition of a constitutional State is every act of ruler or its people should be based on the law and as well as put the purpose of the constitutional State, which guarantee the rights of its people. One characteristic of the constitutional State is the realization of an effort to protect the people or legal protection.

Protection comes from the word *protect* that means put themselves under something to hidden. The protection has the sense of an act, to protect, provide assistance.¹³

According to J.C.T. Simorangkir and Woerjono Sastropranoto, law is an application of restoration justice in the framework of legal protection for victims of conventional crime victim among others by carrying out *penal mediation* in order to restitute to the conventional crime victims. However, the restitution does not mean that criminal elements were done then be lost. The restitution will only relieve the perpetrators. Therefore, the concept of restorative justice cannot be implemented purely without a retributive justice, meaning that the restitution to the victims did not causes the offender was not convicted. The development of sentencing is currently in *restorative justice models* with not negate *retributive justice model* called the balance perspective sentenced is a balance between the offender's interests, the victim and the community¹⁴ are regulations that are forcing, which determine human behavior in a community environment created by the agency official. R. Soeroso¹⁵ argued that the law is a set of rules made by the authorities with the aim to arrange the social life that have a characteristic to commanding and forbidding, and force by impose sanctions for those who break them.

Mochtar Kusumaatmadja¹⁶ argued that the definition of law should not only look at the law as a set of rules and principles that govern human life in society, but should also include institutions and processes necessary to realize law in reality.

Legal protection is a protection that given to the subject of law in the form of legal instruments preventive and repressive, written and unwritten. In other words, legal protection as a description of the legal function i.e a concept in which law can provide a justice, order, certainty, usefulness and peace.¹⁷

Facility, Principles and Form of Legal Protection:-

The term of legal protection for the people is a term that does not include "toward the government" or the action of government. The reason as stated by Philip M. Hadjon¹⁸ that:

- a. The term of *people* already implies a sense as opposed to the term of government. The term of *people* is essentially means the governed, (*geregeerde*). Thus, such term is more specific than the other terms in foreign languages such as *folks*, *people*.
- b. In essence, the term of *people* means the governed and not included the term "towards the government" or on the action of government due to the inclusion of the term might create an impression that there is a confrontation between the people who governed by the government as the ruling.

Philip Hadjan¹⁹ in his view that puts the legal protection as anything that enabling a person can do and defend their rights as determined by law and its relation to the action of government as a central point of the Philips M. Hadjon, suggests that there are 2 (two) types of legal protection for the people, namely:

- a) Preventive legal protection

¹²*Ibid*. Page. 51

¹³Poerwadarminta. *Kamus Bahasa Indonesia*. Balai Pustaka : Jakarta. Page. 540

¹⁴J.C.T. Simorangkir and Woerjono Sastropranoto. (1959). *Hukum Indonesia*. Gunung Agung: Jakarta. Page. 32

¹⁵Soeroso. (2006). *Pengantar Ilmu Hukum*. Sinar grafika : Jakarta. Page. 120

¹⁶Mochtar Kusumaatmadja. (2003). *Pengantar Hukum Indonesia*. PT. Alumni : Bandung. Page. 65

¹⁷*ibid*

¹⁸Philipus M. Hadjon. (1987). *Perlindungan Hukum Bagi Rakyat Indonesia Sebuah Study Prinsip-prinsipnya, Penanganannya oleh Pengadilan dalam Lingkungan Peradilan Umum dan Pembentukan Peradilan Administrasi Negara*, PT. Bina Ilmu : Surabaya. Page 1.

¹⁹*Ibid*. 2

Preventive legal protection provides a chance for people to file an objection or opinion before a government decision in definitive. Thus, the preventive protection aimed at preventing disputes.

b) Repressive legal protection

In a repressive legal protection, the people is given a chance to file an objection after the result of a government decision which is definitive in the sense that repressive protection is solve problem. In addition to the preventive and repressive legal protection is also known as the formal and substantive legal protection. Formal legal protection is a legal protection refers to the implementation of the rights and formally adopted in legislation and regulation or agreement. The substantive legal protection is a legal protection refers to the implementation of the rights attached to the substantive law. This means that although it is not applied in the Act but its implementation is attached to the people or their activities.²⁰

Victims of Crime:-

Definition of Victims:-

The word of victim is derived from the Latin word “*victim*”²¹ then the sense of victim in the *Declaration of Basic Principles of Justice for Victims of Crime and Abuse of Power* are person either individually or collectively have suffered losses either physical, mental, emotional and decay (impairment) against their fundamental rights through acts or not, but it is a violation of national criminal law in addition also based on internationally recognized norms relating to the human rights.²²

Victims in the Indonesian General Dictionary areman or person who suffers an accident because the self-act (desire etc) or others. I.S. Susanto argued that the victim was divided into 2 (two), in both narrow and broad senses. Victims in the narrow sense is the victim of a crime, whereas in a broader sense also includes the victims in various fields such as pollution victims, victims of abuse and so forth.

The victim is a person who suffered a loss in physical, mental or financial as a result of a criminal offense (as aneffect) or a factor the incidence of criminal offense (as a cause). The victim is defined as someone who has suffered a loss as a result of a crime and his/her sense of justice is directly disrupted as a result of his/her experience as a target/criminal target.²³

Stanciu²⁴ argued that in the broad sense a victim is a people who suffer as result of injustice. There are 2 (two) basic characteristic to the victims is suffering and injustice. Victim is not only been seen as a result of illegal acts that the real cause can also create injustice, and then causing victim. The definition is also restricted in the narrow sense as stipulated in the positive law. Indonesian positive law which gives decisive definition of victim, among others, Article 1 number 2 of Witnesses and Victims Protection Acts asserting that the victim was a person who suffered in physical, mental, and/or economic loss as result of a criminal offense.

Declaration of Basic Principles of Justice for Victims of Crime and Abuses of Power defines victims as follows:

“*Victims*” means persons who, individually, or collectively, have suffered harm, including physical or mental injury, emotional suffering, economic loss or substantial impairment of their fundamental rights, through acts or omissions that are in violation of criminal laws operative within Member States, including those laws proscribing criminal abuse of power.²⁵

²⁰Muhammad Tahir Azhary. 1995. *Negara Hukum Indonesia Analisis Yuridis Normatif Tentang Unsur-Unsurnya*. UI-Press, Jakarta. Page. 1.

²¹Ira Dwiati.(2007). *Perlindungan Hukum Terhadap Korban Tindak Pidana Perkosaan Dalam Peradilan Pidana*. Thesis. Universitas Diponegoro : Semarang. Page. 76

²²Annex IV Declaration of Basic Principles of Justice for Victims of Crime and Abuse of Power dalam United Nation Office for Drug Control and crime Prevention, Handbook on Justice for Victims, Centre of International crime prevention.

²³Sujoko. (2008). *Implementasi Tuntutan Ganti Kerugian Dalam Pasal 98 KUHP Terhadap Tindak Pidana Perkosaan di Wilayah Hukum Semarang*. Universitas Diponegoro : Semarang. Page. 1

²⁴*Ibid*. Page. 29

²⁵United Nation Declaration of Basic Principles of Justice for Victims of Crime and Abuse of Power 29 November 1985. Available online at: <http://www.unrol.org/files/BASICP~4.PDF>

Classification of Crime Victims:-

Crime victim conception is also formulated in the *Declaration of Basic Principles of Justice for Victims of Crime and Abuse of Power*, namely:

a. Victims of crime include:

- 1) *Direct victims*, i.e victim who directly suffering with criminal offense with the characteristics of victims were either individually or collectively, suffered losses in the form of physical injuries, mental, emotional suffering, loss of income and the suppression of basic human rights, caused by the act or omission that is formulated in the criminal law or caused by the abuse of power.
- 2) *Indirect victims*, the emergence of victim as cause of interference of a person in helping the victims directly (*direct victims*) or participating in the prevention of victim, but he also became a victim of crime, or they depend their life to the direct victims, such as : wife/husband, children and relatives.
- b. Victims of abuse of power, the victim is a person who individually or collectively suffered harm, including physical or mental injury, emotional suffering, economic loss or the suppression of the basic human rights, through acts or omissions that do not constitute a violation of National Criminal Act but internationally recognized norms relating to human rights.

Sellin and Wolf made a classification of victims into 5 (five) categories, namely:²⁶

- 1) *Primary victimization*, i.e individual victim. So, their victim is individual or group.
- 2) *Secondary victimization*, in which the victim is a group as a legal entity.
- 3) *Mutual victimization* occurs because the attitude or behavior of victims who agree to crimes against him/her.
- 4) *Tertiary victimization* victims arising from the implementation of the social order.
- 5) *No victimization*, here it does not mean there is no victim, but the victims were not immediately known.

Steven Schafer, in relation to the role of the victim suggested several types of victim in relation to the accountability, namely:²⁷

- 1) *Unrelated victims*, are those who do not have any relationship with the perpetrator unless the offender who has committed a crime against him/her. In this type of responsibility lies fully in the hands of the perpetrators.
- 2) *Provocative victims* are those who are doing something against the perpetrators and consequently they become victims. Victims in this case were the main perpetrators. In this type of responsibility lies with the two parties that is victim and perpetrator.
- 3) *Precipitative victims*, the behavior of victims who unwittingly encourage perpetrator to do evil. In this type of responsibility lies with the perpetrator.
- 4) *Biologically weak victims* are those who have certain physical and mental shape that encourages people to do evil against him/her, as an example of small children, the elderly, and women, people who are physically or mentally. In this type, those accountable were public and the government, because it is not able to protect helpless victims.
- 5) *Socially weak victims* are those that are not considered by the public as a member, for example, immigrants and minorities. In this type, the accountability lies on criminals and society.
- 6) *Self-victimizing victims* are those who become victims because of his/her own actions, such as drug addiction, homosexuality and gambling. In this type, the accountability lies fully on the perpetrators who are also victims.
- 7) *Political victims* are those who suffer because of their political opponents. In this type that no one can be justified.

The Handling of Legal Protection for Victims of Crime:-

Barda Nawawi Arief said that the positive criminal law in force recently the protection of victims is more as an "abstract protection" or "*indirectly protection*" it means various formulations of criminal offenses in the legislations during this *per set* there is *in abstract* directly protection against rights and legal interests of victims.²⁸

²⁶ Bedi Setiawan Al Fahmi. (2009). Perlindungan Korban Tindak Pidana Perkosaan Dalam Proses Peradilan Pidana Perspektif Pembaharuan Hukum Acara Pidana Indonesia. *Jurnal Hukum dan Pembangunan* No. 1 Vol. 16 Januari 2009. Page. 65

²⁷ Rena Yulia. (2011). Viktimologi: Perlindungan Hukum Terhadap Korban Kejahatan. hlm 164-165. *Jurnal Intelek* Volume 7 No. 3 Desember 2011. Page. 54

²⁸ Barda Nawawi Arief. (2001). *Masalah Penegakan Hukum Dan Kebijakan Hukum Pidana dalam Penanggulangan Kejahatan*. Citra Aditya Bakti : Bandung. Page. 56

Article 1 point 6 of Act No. 13 of 2006 on Witness and Victim Protection as amended by Act No. 31 of 2014 on the Amendment of Act No. 13 of 2006 on Witnesses and Victims Protection, confirmed that the protection is all effort rights fulfillment and the provision of assistance to provide security to witnesses and/victim that must be implemented the Agency of Witness and Victim Protection or other Institutions in accordance with the provisions of this Act. The sense of victims protection can be seen into 2 (two), namely:

1. As legal protection for not being a crime victim (it means the protection of Human Rights or individual legal interests).
2. As protection to obtain guarantee/legal compensation for the suffering/loss of suffered people (it is similar to the sympathetic of victim). This type that can be vindication/rehabilitation, recovery of equanimity among others with forgiveness, compensation, such as restitution, guarantees/social welfare benefits and so on.

The purpose of victim protection were providing security to victims, particularly when providing information on any criminal justice process, providing encouragement and motivation to the victim not to be afraid to undergo criminal justice process, restore the confidence of victims in social life, and a sense of fairness, not just to the victims and their families, but also to the community.²⁹

In general, in theory there are 2 (two) models of the victim protection, namely.³⁰

1. The procedural rights model

In France, this model is called "*partie civile model*" (*civil action system*). In short this model emphasizes the active role of victims in the criminal justice process, such as helping the public prosecutors, involved in every level of examination of the case, shall be heard if the convict is released conditionally, and so forth. In addition, with the participation actively in the criminal justice process, victims can regain self-esteem and confidence. The involvement of the victim has a positive aspect in law enforcement, and also has a negative side because active participation of victims in the criminal justice process execution can lead to personal interests over the public interest.

The theory as background to the formation of the prosecutor institution, as stated by Jan JM van Dijk, the Hague, that "*historically this has been the main justification for the establishment of the office of the public prosecutor*".

Furthermore, another reason put forward a group that opposes the procedural right to the victim is given the role of individual to the victim in trial process or prosecution of perpetrators, making it participate to responsible for the trial, and the results of that process so that the burden of this responsibility will be pressure heavy enough for victims in many respects. Pressure can come from a person with whom the victim made contact and/or caused by police or prosecutors who will take advantage of their rights in the public interest. Offender and his lawyer will seek to influence the behavior of victims during the process and sometimes using intimidation.

2. The services model

This model emphasizes compensation or restitution and making an effort to gain condition of traumatized victims, fear and distress as a result of crime. A policy of sentenced formulation system that oriented in the positive law of victims to come include a policy of victim protection formulation in material criminal law, a policy of victim protection formulation under formal criminal law and a policy of victim protection formulation in criminal law enforcement. The substance of positive law in question is the Draft Law on the Criminal Code Procedure of 2008 and the Draft Law on the Criminal Code Procedure of 2009.

Additional penalty in the draft law on the Criminal Code Procedure in the elucidation of Article 67 paragraph (1) is defined as "compensation payments" that may be imposed only if the judge "clearly stated in the formulation of crime". The provisions of Article 54 in the Criminal Code Procedure assert that the punishment aimed at resolving the conflict posed by crime, restoring balance, and bring a sense of peace in society." The provisions of Article 55 paragraph (1) determines that the Guidelines of Punishment must be considered judges includes the influence of criminal offenses against victims or their families; forgiveness from the victim and/ or her/his family." Explanation of the provisions of paragraph (1) contains Guidelines of Punishment were very helpful in considering the measure or judge the severity of the punishment to be imposed. Taking into account the matters specified in the guidelines is expected sentence or punishment imposed is proportionate and can be understood both by society and the convict.

²⁹Article 4 of Act No. 13 on 2006 on *Witness and Victim Protection*

³⁰Ahmad Kamil. (2012). *Mediasi Penal dalam Penanganan Tindak Pidana*. Mahkamah Agung Republik Indonesia : Jakarta . Page. 4

The details of this provision are not limiting, meaning that the judge may add other considerations other than those listed in this paragraph (1).

The provisions of Article 71 c, d and g specifies that “by keeping into account Article 54 and 55, imprisonment may be imposed so far, if encountered the following circumstances: 1. loss and suffering of victims is not too big; 2. the defendant has paid compensation to victims of crime victims; 3. Victim of crime encourage the occurrence of criminal offense.” Elucidation of Article 71 confirms that the provisions of Article 71 are intended to assist the judge in determining the penal measure to be imposed.

Article 54 and 55 provides guidelines for the judge can convict proportionately and effectively and the provisions of Article 71 of this as a rule of sentencing for the judge in the case will not impose imprisonment in conditions that are described in Article 71. Formulation of the provisions of Article 77 is not operational because there are “provisions of the criminal types that can be imposed judge” after he did not impose imprisonment. Such provisions are also not listed in the explanation. The provisions of Article 65 paragraph (1) under the paragraph “types of criminal” in addition to prison, also listed as principal criminal, namely; closing, supervision, fines and community work. The problem is if the provisions of Article 71 are met, what type of punishment can be imposed judge to determine criminal dose to be imposed proportionate and effective.

The compensation or restitution to victims of crime has been adopted by other countries, especially the Anglo-Saxon legal systems. New Zealand is one of the countries that recognize compensation to victims in a legal setting. In New Zealand, the compensation of victims has been approved as a type of punishment in its legislation. This is motivated by the view that in terms of the nature of suffering or material losses suffered by the victim as a result of criminal acts committed by others, it is only fair perpetrators of criminal acts (the other person) who provide compensation.³¹

Another example is in Japan. Rationale and culture of the Japanese people against the Constitution is not contrary to the ideology that is universal, but rather relied on the ideology of the special (particular), because in this way, Japan can suppress the crime rates into the lowest in the world. Law enforcement practices in Japan prefer an agreement or consensus as compared with the settlement via litigation.³²

Restorative Justice:-

Restorative justice is a concept that responded the development of the criminal justice system by focusing on the needs of victims and community involvement that feels marginalized by mechanisms that work in the criminal justice system that exists today.³³ Muladi argued his view that in restorative justice, the position of case must be changed, no longer in the interest of order, but for the sake of victims and the recovery of material and psychological. The point is how to prevent offenders from imprisonment, but remain responsible.

Van Ness suggested that restorative justice is a theory of justice that promotes the recovery of losses due to malicious behavior, in which the complete recovery through a process of inclusive and cooperative. If viewed from the recovery of various conflicts, the essential elements of the definition of restorative justice by some experts is more priority to reconciliation or improvement efforts between the perpetrator, the victim, and the community rather than retaliation.³⁴

Restorative justice is a fair solution that involved offenders, victims, their families and other relevant parties in a criminal act jointly seek solutions to the offense and its implications by emphasizing restoration to the original condition

Restorative Justice is a concept of punishment, but as a concept of punishment is not confined to criminal law provisions (formal and material). It also must be observed in terms of criminology and the prison system. From the

³¹Roeslan Saleh. (1984). *Segi Lain Hukum Pidana*. Ghalia Indonesia : Jakarta. Page. 21.

³²Romli Atmasasmita. (2001). *Masalah Santunan Terhadap Korban Tindak Pidana*,” *Majalah Hukum Nasional Departemen Kehakiman*. 1992. Pages 44-45

³³Eva Achjani Zulfa. 2010. *Pergeseran Paradigma Pemidanaan*. Lubuk Agung : Bandung. Page. 65

³⁴*Ibid*

facts, the applicable criminal system does not guarantee integrated justice, namely justice for perpetrators, justice for victim, and justice for the people. This has encouraged the concept of "Restorative Justice."³⁵

Bagir Manan argued that restorative justice contains principles, such as build participation together between the offender, victim and community groups to resolve an incident or crime, putting perpetrators, victims, and the community as "stakeholders" working together and immediately tried to find a solution which is perceived to be fair to all parties (win-win solutions).³⁶

Restorative justice approach offers different views and approaches in understanding and dealing with a criminal offense, as depicted on the definition put forward by Dignan, as follows:

Restorative justice is a new framework for responding to wrong doing and conflict that is rapidly gaining acceptance and support by educational, legal, social work, and counseling professionals and community groups. Restorative justice is a valued-based approach to responding to wrongdoing and conflict, with a balanced focus on the person harmed, the person causing the harm, and the affected community.

The definition requires a certain condition that puts restorative justice as a fundamental value that used in response to a criminal offense. In this case, the focus of attention required balance between the interests of offenders and victims and take into account the impact of the settlement of the criminal cases in the community.

The goal of restorative justice is to encourage the creation of a fair judicial and encourage the parties to participate therein. Victims feel that their sufferings are cared and compensation agreed balanced by suffering and loss suffered. The main objective is to empower the victim restorative justice, where offenders are encouraged to pay attention to recovery.

Restorative justice attaches great importance to the fulfillment of material, emotional, and social victim. The success of restorative justice, as measured by the large of loss has been recovered by offender, not measured by how sentence imposed by the judges. Its essence, as far as possible the perpetrator is removed from the criminal proceedings and prison. But, as said by Kent Roach, restorative justice is not only provides an alternative to prosecution and imprisonment, but also ask the responsibility of the perpetrators. Criminal acts in restorative justice are interpreted as a violation of the law and the state, besides facing the perpetrator is the victim and the community, not the government.

The process for resolve disputes, restorative justice is no longer use conventional methods that have been used in the criminal justice system, which only focuses on finding who is right and who is wrong, and to find what is appropriate punishment given to the guilty party. While in resolve disputes through restorative justice are no longer either case, desired by restorative justice is a restoration of the offender so that he will no longer commit crimes, the recovery also helped addressed to the victim as an injured party and the relationship between the victim, the offender and society to the course life can return to normal.

The theory of punishment recognizes a system derived from the conception of individualization of criminal and known as the *double track system*. This system introduces two penalties together that are criminal and action sanctions. This system puts the two in parallel and equal. Both equality are based on the premise that element of suffering through criminal sanctions and the elements of imprisonment through action sanction is two things that are equally important.

These linkage is described by Gerber and Mc Anany argued that criminal sanctions or retributive is not fully removed in sentencing because under certain conditions despite the implementation of sanctions moving towards rehabilitation as goal fully but still there should be criminal prosecution and instead sanctions only relying on criminal sanctions (retributive) proved ineffective to restore social and moral qualities of a criminal in order to

³⁵Joint Decree between the Chairman of the Supreme Court, the Attorney General of the Republic of Indonesia, the Head of the Indonesian National Police, Minister of Justice and Human Rights of the Republic of Indonesia, Minister of Social Affairs of the Republic of Indonesia and the Minister of Women Empowerment and Child Protection of the Republic of Indonesia.

³⁶Lies Sulistiani. (2010). *Perlindungan Saksi dan Korban di Indonesia*. Press Release No.02/LPSK/PR/II/2010. Lembaga Perlindungan Saksi Republik Indonesia : Jakarta. Page. 4

integrate more with the community. This is the reason for the need to integrate between criminal and action sanctions.³⁷

Double track system requires the element of suffering and imprisonment are equally accommodated in the system of criminal law sanctions. This view was expressed also by Albert Camus,³⁸ who stated that the application of criminal sanctions there needs to be equality between *punishment* and *treatment*. *Punishment* is a logical consequence of freedom is misused by offenders but on the other hand a perpetrator remains of a man who has the right to reach the new values and new adjustments in order to be a good human being.

Hart³⁹ argued that a theory of punishment that is morally acceptable is able to demonstrate the complexity of the prosecution and described it as a compromise between different principles and contradictory that any punishment is complex. Complexity is expressed by Hart according to the author showed that the punishment in view of Hart combines the suffering sanctions and for imprisonment. Arief⁴⁰ said that the fundamental difference between criminal sanctions and action sanctions is the criminal question questioned why the imprisonment is held while the action sanctions questioned to what the imprisonment is held. Criminal sanctions are reactive to an act, while action sanctions are anticipatory against the perpetrators of such acts. The focus of criminal sanction is perpetrators act through an affliction to deter while the focus of criminal sanctions is to give assistance for change.

J.E. Jonkers also argued that criminal sanctions emphasized for crimes committed while action sanctions focused on goals that are social.⁴¹ Description and views on criminal sanctions and action sanctions indicate that criminal sanctions oriented to sanctions for action, while the action sanctions oriented to the community protection. The debate of philosophy and theory of punishment always have ups and downs in its development, which leads to the emergence of new strategies in sentencing, as follows:

a) *Indeterminate sentence*

Indeterminate sentence is a strategy of criminal sanction that provides flexibility to judges in sentencing. The development of this strategy gained sharp criticism because they provide greater uncertainty than benefits. The uniformity for any criminal offense generates inequality in the process of criminal prosecution.

b) *Just desert model*

Just desert model is a strategy that is based on fairness with regard punishment as prevention and retribution. *Prevention* aimed at preventing a perpetrator to repeat offense or crime in later while *retribution* aimed to provide vengeance for a criminal offense committed.

c) *Restorative model*

Restorative model is a model of punishment that proposed by the abolitionists. This model is often confronted with retributive models.

Conclusion:-

The essence of legal protection for crime victims in the handling of conventional crime in Indonesia is not yet optimal to take attention and gives legal protection to conventional crime victim in achieving fairness, expediency and legal certainty as a legal purpose. Conventional crime victims are an integral part of a legal system, which received less attention in obtaining their rights in the criminal justice system in Indonesia. Legal protection of conventional crime victim in Indonesia has been set in legislations, but it is not optimal because the mostly of legislation is more focused of legal protection to the perpetrators rather than the victims. As a result, the implementation of legal protection for conventional crime victim is not optimal, so it is very important the understanding of community, the victim and also law enforcement officers in the implementation of rules and mechanisms of legal protection against conventional crime victims to get their rights.

The implementations of restoration justice in the framework of legal protection for conventional crime victim were by carrying out *penal mediation* to compensate the conventional crime victims. However, the compensation does not mean that criminal elements were done then be lost. The compensation will only relieve the perpetrators. Therefore, the concept of restorative justice cannot be implemented purely without a retributive justice, it means that the

³⁷Sholehuddin. (2010). *Sistem Sanksi Dalam Hukum Pidana*. PT RajaGrafindo : Jakarta. Page 54

³⁸*Ibid.*

³⁹*Ibid.* Page. 56

⁴⁰Barda Nawawi Arief. 2002. *Perbandingan Hukum Pidana*. RajaGrafindoPersada : Jakarta Page. 51

⁴¹*Ibid*

compensation to victims did not causes the offender was not convicted. Currently, the direction of sentencing is in restorative justice models with not negate retributive justice model in which called as a balance perspective sentencing that is a balance between the interests of offenders, victims and communities or called as criminal prosecution balance perspective.

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RESEARCH ARTICLE

THE GROWTH PERFORMANCE AND YIELDS OF STEVIA (*Stevia Rebaudiana* BERTONI) RESPONSES TO DIFFERENT TYPES OF FERTILIZER.

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Abstract

A crop that is nourishing using fertilizer usually gives a better growth performance and yield. Different types of fertilizer giving a different impact on the growth performance and yield of a crop. The presence of different types of fertilizer such as organic, inorganic and semi organic fertilizer boost crops in different rate. As we know that the use of fertilizer seems to be an obligation to the farmer, identifying the best types of fertilizer that can bring the best of the crop should be determined. This experiment was conducted to study the growth performance and yield of Stevia (*Stevia rebaudiana* BERTONI) responses to different types of fertilizer. The study was carried out at UiTM Jasin share farm by using a randomized complete block design (RCBD) for every treatment. Data collection or the perimeter for plant height, number of leaf and leaf area were recorded once every seven days while the yield of this plant were recorded at the end of the experiment. Data analysis was carried out by using Minitab and Microsoft Excel. At the end of this experiment, Stevia plants were harvested and the yield is being calculated. The results obtained in this study indicate that there are significant different in the number of leaves, area of leaves and the yields harvested toward the application of different fertilizer. It is concluded that the best fertilizer to be applied to Stevia plant is semi organic fertilizer.

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Introduction:-

Belonging of the Asteraceae (Compositae) family, *Stevia rebaudiana* (Bertoni) also known as herbaceous perennial plant (Brian., 2003). Nowadays, stevia is one of the importance plants as substitute to the sugar that made up by the sugarcane because the stevia's leaves are applied as non nutritive high potency of natural sugar especially in Asia especially Japan, Korea, China and South America. Besides, the increasing cases such as obesity, diabetes and cardiovascular disease are due to the high consumption of sucrose in daily use such as beverages and food. Geuns (2003) studied the diterpene glycosides that produce by *Stevia rebaudiana* Bertoni called as steviol glycosides mainly natural, high potency of low caloric sweeteners where it has been used as sugar substitute.

Stevia rebaudiana leaves are rich in glycosides that contribute to sweet taste with low caloric value and very vital for diabetes as natural substitute for sugar (Kinghorn., 1987). Regarding to the previous studies that has been conducted, the stevia leave's extract has beneficial effect on human health as a natural source of antioxidant (Xi et

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al.,1998), used avoid the formation of cavities and plaque in teeth, non toxic (Elkins.,1997),cardiotonic (Machado *et al.*,1986) and also beneficial against microbes *Streptococcus mutan*, *Pseudomonas aeruginao*, and *Proteus vulgaris* (Yabu *et al.*,1997).

Materials And Method:-

Location of study:-

The experimental site is situated at UiTM Jasin share farm. The essential material for the study that is the three types of fertilizer, organic fertilizer, chemical fertilizer and semi organic fertilizer which are the variables in this study. The other material included stevia cutting, poly bag and net.

Planting materials:-

The stem cutting process will be made in a slant form from the young apical shoot for MS012 variety. Consequently, the end of cut plant will be dipped in IBA concentration. The dipped stem cuttings will be placed in the micro cutting chamber propagation box which is contained a medium of perlite and light expanded clay aggregates (LECA). The stevia's parent was supplied by Agriculture Department District of Tampin, Negeri Sembilan.

Experimental treatment and design:-

After two weeks of propagation, the cuttings were transferred to the pot which contained medium of coco pit with rice husk and soil. Experimental treatments comprised the different types of fertilizers i.e. T₀ -control, T₁- Organic fertilizer (FAA), T₂- Inorganic fertilizer (18+33+18+TE), T₃- Semi organic fertilizer(12+7+25).The dosage recommended for organic fertilizer(FAA) 2ml fertilizer + 1000ml of water, inorganic fertilizer (18+33+18+TE) 2.5g fertilizer + 1000ml of water, and semi organic fertilizer (12+7+25) 20 ml fertilizer + 1000 ml of water. All types of fertilizers being applied once a week in the liquid form. Treatments of experiment were conducted in RCBD with five replications for each treatment.

Data collection and analysis:-

The data of growth parameter was recorded based on the plant height, number of leaves, leaves area and the yields of stevia. The data of growth parameter was taken once a week for five weeks. Recorded plant height was measured from poly bags top soil surface up to the highest leaf tip by straightening all leaves using ruler. Every seven days interval, the total number of leaves was counted. Leaf area was recorded in seven days interval by using Portable Leaf Area tool. Total yield of plants were recorded by plant fresh weight being subtracted to the amount of dry weight of the plant. Dry weight of plant was recorded after reduction of moisture from the plant by using oven. The analysis was carried out by using Minitab 16 software. All the data were analyzed by using Analysis of Variance (ANOVA), and descriptive analysis. The different between the treatments means data were compared using Turkey Family Error Rate Test.

Result and Discussion:-

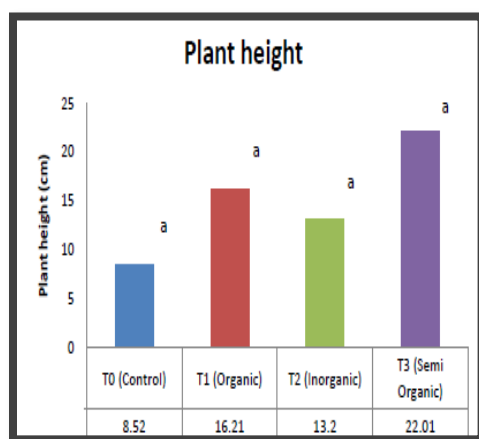


Figure above shows the mean of plants height planted with different application of fertilizer for 5 weeks. From the graph, plant applied with semi organic fertilizer (T₃) show the highest mean of plant height followed by T₁ and T₂. The lowest mean of plant height is the T₀. However, there is no significant different between all the treatments based

on the plant height. By referring to the ANOVA table, it can be concluded that there is no significant different in the plant height of Stevia response to different types of fertilizer. It is because of the P-value in the table (0.237) is lower than the confident interval applied, 0.05.

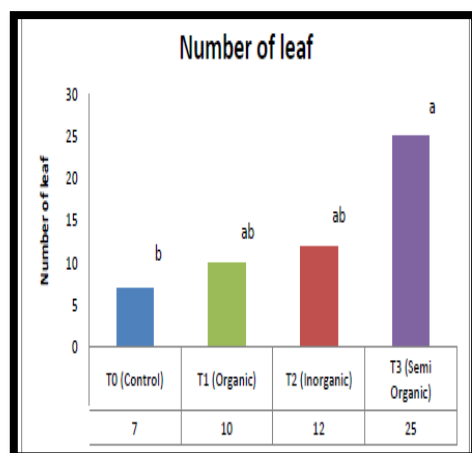


Figure above shows the mean from the number of leaf from different treatment for 5 weeks. Stevia in the T_3 shows the highest average number of leaf. The treatment shows that it is suitable to apply in order to get the highest number of leaf. The plant that is left without applying any fertilizer (T_0) shows the lowest number of leaf to the plant. The higher mean of T_3 in the leaf number was due to the higher presence of humic acid that accelerates plant cell division and promotes growth.

From the analysis using Turkey's family error rates, the means of T_3 and T_0 indicates there is a significant different as shown by the different letters in the graph. By referring to the ANOVA table, it shows that there significant different in the number of leaf of Stevia applied with different types of fertilizer. This is because the P-value in the table (0.029) is lower than the confident interval used for the test, 0.05.

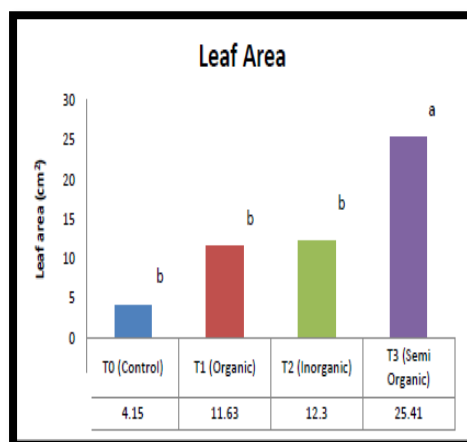
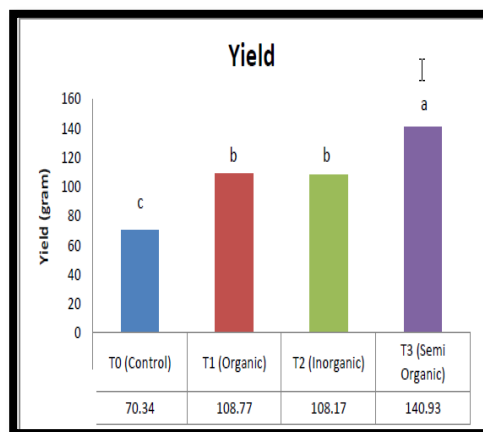


Figure 4.3.2 above shows the mean data of the leaf area from different treatment for 5 weeks. Stevia that is treated with T_3 (semi organic) indicates the highest mean among the treatment applied to the plant. Stevia that is applied with T_0 (control) shows the lowest mean of leaf area. This is due to the nutrient that is absorbed by plant in treatment 3 is faster cause by the presence of humic acid in the fertilizer. From the analysis using Turkey's family error rates, the mean of T_3 shows letter A while the other treatment mean that are T_0 , T_1 and T_2 shows letter B indicates that there is significant different from T_3 toward the other treatment. By referring to the ANOVA table, it can be said that there is significant different in the leaf area of Stevia. It is due to the P-value in the table (0.002) which is lower than the confident interval applied, 0.05.



The above figure shows the mean of the Stevia yield for all the treatment. Stevia that is applied with T₃ (semi organic) fertilizer has the highest mean weight which is more than 140 gram. Stevia that is not applied by any types of fertilizer shows the lowest mean reading that is less than 71 gram. The higher mean of the yield in T₃ indicates that the plant absorb a lots of nutrient from the soil and fertilizer and store it in their plant body. From the Turkey's family error rate, there are several different mean on the treatment applied to the plant. First, the mean of T₃ indicates by letter 'a' show significant different toward the mean of T₀, T₁ and T₂ as the letter indicates them are different from T₃. Then, there is significant different in the mean of T₀ with T₁ and T₂ as the letters shows in the graph is different from each treatment. The ANOVA table above was gained by using Minitab 14. From the table above, it can be concluded that there is significant different between the treatments applied and the yield of the Stevia plant. This is because of the P-value in the ANOVA table (0.000) which is lower than the confident interval use, 0.05.

Conclusion And Recommendation:-

By referring to the result of the study, it can be concluded that Stevia plant (*Stevia Rebaudiana* Bert) shows better growth performance and yields in the application of semi organic fertilizer. This is due to several finding that were obtained during this experiment. Stevia plants that were applied using semi organic fertilizer indicated the best growth in the term of growth performance and yields. This is supported by the significant different of those three parameter tested in the study. Thus, the findings from the study have provided information on the types of the fertilizer that generates the highest yield and plant growth performance toward the farmers. From this research, it is recommended for the farmers to practice the application of semi organic fertilizer to their crops. The used of semi organic significantly increase the yields thus will provide them with more income.

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RESEARCH ARTICLE

LOW SERUM VITAMIN D LEVELS IN EGYPTIAN ADULTS WITH CHRONIC PRIMARY IMMUNE THROMBOCYTOPENIA: A SINGLE CENTER STUDY.

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ITP, 25(OH) D, vitamin D

Abstract

Objectives: Immune thrombocytopenia (ITP) is a disorder characterized by immune-mediated accelerated platelet destruction and suppressed platelet production. Low vitamin D levels have been found in several autoimmune diseases, such as rheumatoid arthritis, SLE. The mechanisms underlying the link between vitamin D with autoimmunity are not completely understood. No currently available studies about vitamin D status in primary ITP patients.

Aim: To evaluate vitamin D levels in patients with primary chronic ITP and compare these levels with normal control subjects and patients with thrombocytopenia due to other non-immune causes **Methods:** The study included 80 adult subjects, 40 ITP patients (They were segregated into 20 responders and 20 non-responders), 20 cases of thrombocytopenia due to non ITP causes and 20 healthy control subjects. Measurement of serum 25monohydroxyvitamin D was done with ELISA.

Results: Vitamin D levels were significantly lower in patients with ITP (range=2-40ng/ml; mean±SD=17.29±10.96 ng/ml) and thrombocytopenia due to non-ITP causes (range=10-40ng/ml; mean±SD=21.05±8.31 ng/ml) in comparison to normal healthy controls (range = 10-65 ng/ml; mean±SD=36.70±16.30 ng/ml) (P=0.000), but there was no statistically significant difference between levels in ITP versus non- ITP thrombocytopenia (P=0.225). There was no statistical significant difference between responders & non-responders

Conclusions: Vitamin D levels are lower among chronic ITP patients in relation to healthy control subjects which might point out to a role in aetiopathogenesis of ITP attributable to vitamin D deficiency.

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Introduction:-

Immune thrombocytopenia (ITP) is a disorder characterized by immune-mediated accelerated platelet destruction and suppressed platelet production^[1]

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In patients with ITP, antiplatelet auto-antibodies frequently appear to be directed against GpIb/IX and GPIIb/IIIa expressed on platelets and megakaryocytes^[2]

Abnormalities of cell-mediated immunity are known to contribute to the pathologic process. Like many other autoimmune diseases, ITP has a T helper cell type 1 bias and a reduced activity of T-regulatory cell^[2]

Vitamin D, a steroid hormone, which is known to play an essential role in calcium homeostasis, but since the receptor for vitamin D (VDR) is found in cells in almost every tissue, and the enzyme 1- α hydroxylase necessary for the final activation of vitamin D also has a widespread distribution, it appears obvious that vitamin D must have a multitude of function^[3]

Lower vitamin D levels have been found in several autoimmune diseases, such as rheumatoid arthritis, systemic lupus erythematosus, systemic sclerosis, type 1 diabetes mellitus, multiple sclerosis, inflammatory bowel diseases, and, autoimmune gastritis^[4]

The mechanisms underlying the link between vitamin D with autoimmunity are not completely understood, but probably are associated with its anti-inflammatory and immunomodulatory functions^[5]

Several studies confirm that vitamin D enhances the innate immune response, whereas it exercises an inhibitory action on the adaptive immune system by inhibiting the proliferation of type 1T helper cells, increasing the quantity of type 2 T helper cells, increasing the quantity of CD4+/CD25+ T-regulator cells, inhibiting B cell proliferation, generation of B memory cells, plasma cell differentiation and immunoglobulin production^[6]

No currently available studies about vitamin D status in primary ITP patients.

SUBJECTS AND METHODS:-

Subjects:-

The current case-control study was conducted on 80 adult subjects (age \geq 18years) including 40 ITP patients, 20 patients with thrombocytopenia secondary to other non-immune diseases, and 20 healthy control subjects. All patients were recruited from the Clinical Hematology Unit, Internal Medicine Department, Faculty of Medicine, AinShams University, Cairo, Egypt.

An informed consent was taken from all subjects participating in the present study according to declaration of Helsinki.

The subjects enrolled in this study were divided into the following groups:-

- **Group-I:** Included 40 adult patients diagnosed as having chronic primary ITP who met the diagnostic criteria of ITP as previously described in the international consensus report published in 2010^[7]. This group was subdivided into:
 - **Group-Ia:** Included 20 patients who were responders to treatment.
 - **Group-Ib:** Included 20 non-responder ITP patients.
- **Group-II:** Included 20 patients with secondary thrombocytopenia. This group included: 8 patients with acute myeloid leukemia (AML), 5 patients with myelodysplastic syndrome (MDS), and 7 patients with aplastic anemia. All the patients with the above diseases were undertreatment.
- **Group-III:** Included 20 sex and age matched healthy controls subjects with normal platelet count and no concurrent illnesses or medications at the time of blood withdrawal.

Inclusion criteria:-

- Adult (age \geq 18 years).
- Primary chronic ITP.
- ITP patients who received the first line treatment by steroid therapy.

Exclusion criteria:-

- Age < 18 years.
- Secondary ITP.
- ITP patients on second line therapy like azathioprine or TPOs
- Chronic renal disease.

- Chronic liver disease.
- Malnutrition and malabsorption.

Plan of treatment in ITP patients:-

Patients were treated by the first line steroid therapy (i.e. prednisolone 1mg/kg) for 3-4 weeks followed by tapering. Patients on second line therapy (e.g. Azathioprine, TPO and danazol, ...etc.) were excluded from this study, and the patients were subdivided into responders and non-responders.

Definition of response:-

Patients were considered responders if they have platelet count between 30 and 100x10⁹/l, doubling of the baseline platelet count and no clinically overt bleeding^[7].

While patients were considered resistant "Non-responders" if they have any platelet count lower than 30x10⁹/l or less than doubling of the baseline count, active bleeding, or steroid dependence^[7].

Methods:-

Assessment of serum level of 25 (OH) D:-

Sample collection:-

For each subject, about 5 ml of venous blood was drawn under complete aseptic conditions and dispensed into a labeled vacutainer containing gel and clot activator, and serum was separated by centrifugation for 5 minutes, then stored at temperature -20°C till assay for measurement of 25(OH) vitamin D was done by ELISA assay using a commercially available ELISA kit (Calbiotech, A Life Science company). The values were expressed in ng/ml.

Definition of vitamin D deficiency:-

Vitamin D deficiency is defined as a 25(OH) D level of 20 ng/ml or less, vitamin D insufficiency as 21 to 29 ng/ml, and vitamin D sufficiency as 30ng/ml or greater.^[8]

Statistical analysis:-

Data were collected, revised, coded and entered to the Statistical Package for Social Science (IBM SPSS) version 20. Qualitative data were presented as number and percentages while quantitative data were presented as mean, standard deviations and ranges when parametric and median with interquartile ranges (IQR) when non parametric.

The p-value was considered significant as the following:

P > 0.05: Non significant

P < 0.05: Significant

P < 0.01: Highly significant.

RESULTS:-

Demographic data and disease characteristics

Subjects enrolled into the study were 80 adult subjects, they were divided into 40 chronic ITP patients (equally subdivided into responders and non-responders), they were 36 females and 4 males with a female: male ratio of 9:1 with mean age± SD of 33.41 ± 9.67 years ranging from 18-50 years, 20 cases of thrombocytopenia due to other reasons with 16 females (80%) and 4 males (20%) with mean age ± SD of 36.70 ± 13.30 years ranging from 19 – 60 years, and 20 adults apparently healthy control subjects , 17 females and 3 males with age range of 18 – 44 years and mean ± SD of 29.90± 8.07 years.

The three groups were matched as regards age (P=0.123) and sex (P=0.560).

Demographic data and disease characteristics amongst the 3 studied groups are illustrated in table number (1).

Hemoglobin levels across the studied groups

Hemoglobin levels were significantly lower in patients with thrombocytopenia due to other (non-ITP) causes (mean±SD =7.80±1.03 g/dl) than ITP patients (mean±SD=11.31±1.48 g/dl) (P=0.000) or healthy normal controls (mean±SD= 12.66±1.37 g/dl) (P=0.000).

Platelet counts across the studied groups

As regards platelet counts, they were significantly reduced in ITP patients and patients with thrombocytopenia due to other causes as compared to healthy controls ($P=0.000$).

The mean platelet volumes were significantly higher in ITP patients ($\text{mean} \pm \text{SD} = 11.16 \pm 2.14$ fl) than patients with thrombocytopenia due to non-ITP patients ($\text{mean} \pm \text{SD} = 9.10 \pm 3.40$ fl) and healthy controls ($\text{mean} \pm \text{SD} = 9.79 \pm 1.01$ fl) ($P=0.004$).

Vitamin D levels across the studied groups:-

25 monohydroxyvitamin D levels were significantly lower in patients with ITP (range=2-40 ng/ml; $\text{mean} \pm \text{SD} = 17.29 \pm 10.96$ ng/ml) and thrombocytopenia due to non-ITP causes (range=10-40 ng/ml; $\text{mean} \pm \text{SD} = 21.05 \pm 8.31$ ng/ml) in comparison to normal healthy controls (range = 10-65 ng/ml; $\text{mean} \pm \text{SD} = 36.70 \pm 16.30$ ng/ml) ($P=0.000$), but there was no statistically significant difference between levels in ITP versus non-ITP thrombocytopenia ($P=0.225$), this is well illustrated in figure (1) attached to the appendix.

Calculating the degree of deficiency of vitamin D in the 3 groups, it was found that 27 (67.5%) of the ITP patients were deficient in vitamin D; with recording values ≤ 20 ng/ml. Of those 27 patients, 15 patients are severely deficient (≤ 10 ng/ml). Other 6 ITP patients (15%) were insufficient and only 7 of them (17.5%) were having sufficient values of vitamin D.

When these data are compared to healthy normal controls, we observed that only 4 (20%) of them were deficient, 1 of them had severe deficiency. Other 4 were insufficient while 12 (60%) had normal levels.

In thrombocytopenia due to non-ITP causes, half of them were deficient (10) while 5 (25%) were insufficient. And 5 only (25%) had normal values).

Vitamin D levels in ITP patients in relationship to response:-

When further segregating ITP patients into responders and non-responders; we found that at enrollment time, the platelet count was significantly lower among non-responders (median $= 64.5 \times 10^9$ /L) than responders (median $= 200.5 \times 10^9$ /L) with highly significant P value ($P=0.000$). At the same time there was no significant difference in hemoglobin level between non responders and responders ($\text{mean} \pm \text{SD} = 11.18 \pm 1.63$ g/dl versus 10.65 ± 1.56 g/dl respectively) ($P=0.296$).

The two subgroups (non-responders and responders) showed no statistical significant difference as regards vitamin D levels being as following ($\text{mean} \pm \text{SD} = 15.73 \pm 10.00$ ng/ml in responders in relation to $\text{mean} \pm \text{SD} = 18.85 \pm 11.89$ ng/ml in non-responders with a P-value of 0.374).

25(OH) D levels when correlated to age, disease duration, dose of steroids given and platelet counts in ITP patients, they showed no significant correlation.

DISCUSSION:-

Immune thrombocytopenia (ITP) is an autoimmune disorder characterized by accelerated platelet destruction and suppression of platelet production, resulting in low platelet count^[9]

Vitamin D is recognized as a true steroid hormone that exerts several biological activities, including regulation of the immune system. So, its deficiency can contribute to the pathophysiological mechanisms for the development of autoimmune diseases^[10]

The aim of this study is to assess vitamin D status in adult patients with primary immune thrombocytopenia (ITP) and to correlate it with response to first line treatment (steroid therapy).

The present study included forty chronic ITP patients (36 females and 4 males), twenty patients with thrombocytopenia secondary to other non-immune diseases (16 females and 4 males), and twenty normal healthy subjects (17 females and 3 males). There were no statistically significant differences between the three groups as regards sex and age (with p values= 0.560 and 0.123 respectively).

In this study, 90% of the ITP patients were females and only 10% were males with female to male ratio 9:1, which reflects a higher incidence of ITP in females. This observation is consistent with the results of *Schoonen et al.*^[11] who found that the incidence of ITP was statistically significantly higher in women compared to men. And also *Saeidi et al.*^[12] who found that the ITP prevalence was higher in females those aged above 16 compared to males of the same age with ratio 7: 3. As an autoimmune disease; ITP is expected to have higher incidence in females.

The mean age of ITP patients in our study was 33.41 ± 9.67 years, this was same as that described by *Cines and Busnel*^[13] who stated that the incidence of ITP is more common in women aged between 18 and 40 years. On the other hand, *Neylon et al.*^[14] estimated a higher incidence of ITP among those aged 60 years and older.

The platelet count in ITP patients (median of $114 \times 10^9/L$) and in the patients with thrombocytopenia due to non-ITP causes (median of $36.5 \times 10^9/L$) were significantly decreased in comparison to healthy controls ($296 \times 10^9/L$) ($p < 0.001$). On the other hand, the haemoglobin levels were significantly lower in patients with thrombocytopenia due to non-ITP causes (mean \pm SD = 7.80 ± 1.03) than in normal healthy controls (mean \pm SD = 12.66 ± 1.37) and ITP patients (mean \pm SD = 11.31 ± 1.48) with p value = 0.000, this finding was due to the nature of the diseases included in this group of patients (AML, aplastic anaemia, and MDS) that are characterized by the manifestations of bone marrow failure. The ITP patients show mean haemoglobin level around the normal because it is the disease of isolated thrombocytopenia.

In this study; we observed that the mean platelet volumes (MPV) were significantly higher in patients with ITP (mean \pm SD = 11.16 ± 2.14 fl) than that in patients with thrombocytopenia due to non-ITP causes (hypoproliferative thrombocytopenia) (mean \pm SD = 9.10 ± 3.40 fl) and in normal controls (mean \pm SD = 9.79 ± 1.01 fl) with p value 0.002 and 0.035 respectively. This result agrees with the finding reported by *Ntaios et al.*^[15]; *Negash et al.*^[16] who found that the MPV is significantly elevated in ITP patients than in patients with hypoproliferative thrombocytopenia. In disagreement with this result, a study conducted by *Borkataky et al.*^[17] who found no significant difference in the MPV between the destructive thrombocytopenia groups and the control group. However, larger platelets have more granules, aggregate more rapidly with collagen, have a higher thromboxane A2 level, and express more glycoprotein Ib and IIb/IIIa receptors than smaller platelets, which may explain the lower bleeding tendency in ITP patients^[18].

Results in the present study revealed non-significant difference in serum 25 (OH) D levels among ITP females and male patients. In concordance to our results, study by *Elsammak et al.*^[19] who evaluated the vitamin D status of a cohort of healthy young Saudi Arabians in the Eastern region of Saudi Arabia. A sample of 139 blood donors studied. They found that serum 25(OH) D levels did not differ significantly between males and females. And this result is consistent with the findings of *Amital et al.*^[20] who conducted their study on SLE patients from several European and Israeli cohorts and found that vitamin D level did not differ by gender. However the results of this present study did not agree with the results of a study by *Wright et al.*^[21] conducted on SLE patients who found that female sex was a predictor for vitamin D deficiency. To our knowledge; no similar studies were conducted on ITP patients before.

In the present study, we did not find a significant correlation between vitamin D levels and age or disease duration in ITP patients. This finding is in line with the results of a study by *Emam et al.*^[22] who studied a comparable number of Egyptian SLE patients (40 SLE patients and 20 age and sex matched controls) and did not find significant correlation between vitamin D level and age or disease duration. And another study by *Rossini et al.*^[23] conducted on rheumatoid arthritis patients did not find an association between vitamin D level and disease duration. Also *Dias de Castro et al.*^[24] in their study on patients with Inflammatory Bowel Disease found no correlation between vitamin D and age or disease duration. However, *Karatay et al.*^[25] studied 32 patients with Behcet's disease and found that age was one of the predictors of 25(OH) D levels.

In the present study, we have found significantly lower mean 25(OH)D levels among ITP cases compared to healthy control group, (17.29 ± 10.96 ng/ml) vs (36.70 ± 16.30 ng/ml) respectively with p value < 0.001 .

When ITP patients and control subjects were classified according to vitamin D status, 33 out of 40 ITP patients (82.5%) had inadequate (insufficient and deficient) 25(OH)D levels (< 30 ng/ml): of which 6 patients (15 %) being insufficient (21-29 ng/ml), 27 (67.50%) being deficient (≤ 20 ng/ml). In contrast, 4 out of 20 healthy control subjects (20%) were found to be insufficient, 4 (20%) being deficient. Out of our knowledge, there was no previous studies evaluating vitamin D levels in patients with ITP in Arabian country, but there were on other autoimmune disorders

such as SLE disease. The frequency of 25(OH) D deficiency among patients with ITP in our study is similar to that reported in a study conducted by *Emam et al.* [22] on 40 SLE Egyptian patients and 20 controls which revealed low vitamin D levels in patients with SLE compared with healthy controls, and (85%) of SLE patients had inadequate (insufficient and deficient) vitamin D levels.

Despite Egypt is a sunny country, many contributing factors lead to low vitamin D level in our population in general as in most Middle East countries, including the dark skin, use of sun screen, the cultural and religious practice of wearing clothes that cover the entire body, and the limited amount of vitamin D obtained from dietary sources due to lack of fortification and even lack of measures to confirm adequacy of fortification of fortified staple foods [26]. Also *Fragoso et al.* [27] explained this to be a result from increased modern indoor life activities.

In the present study, the result is concordant to a study by *Fattizzo et al.* [28] conducted on 103 patients with autoimmune cytopenias (included 44 patients with ITP, 35 with primary autoimmune hemolytic anemia, 5 with Evans' syndrome, and 19 chronic idiopathic neutropenia) and 40 controls, which revealed low 25 (OH) D level in patients with ITP compared with healthy controls (mean \pm SD= 2.3 ± 1.8 vs 6 ± 6 ng/ml, $P < 0.001$), they also found, that 25(OH) D levels were significantly lower in patients with primary autoimmune hemolytic anemia, Evans' syndrome, and chronic idiopathic neutropenia than in controls (mean \pm SD= 2.4 ± 1.6 ng/ml, 1.3 ± 0.5 ng/ml, 2.1 ± 1.3 ng/ml vs 6 ± 6 ng/ml, $P < 0.001$).

Also, in agreement with our results a Chinese study conducted on 45 ITP patients showed that the levels of 25(OH)D (10.6 ± 7.7 ng/ml) and $1\alpha,25(\text{OH})_2\text{D}$ (69.9 ± 29.0 pg/ml) of the ITP patients were lower than those of healthy controls (13.7 ± 9.1 ng/ml, 87.3 ± 19.9 pg/ml) ($P < 0.05$) [29].

In the present study there was no statistical difference between the ITP patients and patients with thrombocytopenia secondary to other causes (non ITP) as regard vitamin D levels. Serum 25(OH) D levels were significantly lower in patients with thrombocytopenia due to non ITP causes than normal healthy controls (mean \pm SD = 21.05 ± 8.31 ng/ml) vs (mean \pm SD = 36.70 ± 16.30 ng/ml) ($p < 0.001$). In fact, the patients included in this group (patients with thrombocytopenia secondary to other causes) unfortunately were patients with acute myeloid leukemias, myelodysplastic syndrome, and aplastic anemia, and vitamin D deficiency has been implicated in the pathogenesis of these hematological and onco-hematological disorders. However, many of the diseases with thrombocytopenia were associated with vitamin D deficiency, and we could not enroll patients with thrombocytopenia secondary to HCV or HBV because patients with liver diseases were excluded from this study as liver disease is one of the causes of vitamin D deficiency.

In our study there was no statistically significant difference in 25 (OH) D levels between responders and non-responders (median = 15.5 ng/ml vs 19 ng/ml, respectively. $p = 0.327$). This result is in concordance with the results of a study by *Fattizzo et al.* [28] in which all ITP patients were treated with steroids, rituximab was administered in 1 case, cytotoxic immunosuppressives in 7, and splenectomy performed in 8; 5 cases unsuitable for or refractory to splenectomy were treated with thrombopoietin receptor agonists, and they found no significant association among the number of therapy lines and vitamin D levels. This observation may indicate that vitamin D deficiency has a role in ITP pathogenesis, but did not contribute in disease activity and treatment response. However, more studies with larger populations may be required to define the actual role of vitamin D in treatment response and the activity of disease in patients with ITP.

However, vitamin D was inversely correlated to disease activity in many other studies, in a study conducted by *Haga et al.* [30] on 302 patients with rheumatoid arthritis, they found that very low serum 25(OH) D levels (≤ 6 ng/ml) were associated with very high disease activity and with increase the requirement of treatment with at least 3 disease modifying anti-rheumatic drugs. Furthermore, a case-control study by *Ricceri et al.* [31] conducted on 68 patients with chronic plaque psoriasis and 60 healthy controls demonstrated a statistically significant inverse linear correlation between patient's vitamin D levels and their Psoriasis Area Severity Index (PASI) scores. Also *Mok et al.* [32] in their study on SLE patients found that 25(OH) D level correlated inversely and significantly with SLE activity. On the other hand *Ruiz-Irastorza et al.* [33] found no association between vitamin D levels and SLE disease activity. *D'Aurizio et al.* [34] found that low serum levels of vitamin D did not correlate with Hashimoto's thyroiditis or with Grave's disease activity. Also, no correlation between vitamin D levels and disease activity was observed in a cross-sectional study conducted by *Levin et al.* [35] on children with inflammatory bowel diseases.

In the present study there was no significant correlation between vitamin D levels and platelet counts at enrollment time. Our observations are consistent with the findings of *Fattizzo et al.* [28] who found no significant

association among vitamin D levels and hemoglobin, platelets, and absolute neutrophil counts values at enrollment for AIHA, ITP and chronic idiopathic neutropenic patients, respectively. However, 20 ITP patients with very low vitamin D levels (<1.3 ng/ml) displayed reduced platelets counts compared with the remaining 24. However, the majority of our patients were enrolled from the outpatient clinic and their platelet counts were not severely low at time of enrolment.

Despite corticosteroid use is thought to increase the catabolism of vitamin D by activating the expression of the Steroid and Xenobiotic nuclearReceptor (SXR), which induces the destruction of 25(OH)D and 1 α ,25(OH)₂D through the expression of CYP3A4^[36]. We found no correlation between use of steroids / steroid doses and vitamin D levels in our patients. Our observations are consistent with the findings of *Fattizzo et al.*^[28] where no relationship was found between serum 25(OH) D levels and steroid doses in ITP patients. Also, in a study conducted on SLE patients by *Schoindre et al.*^[37] they did not find an association between corticosteroid use or dose and vitamin D status. This finding may indicate that vitamin D deficiency in our patients was not due to steroid therapy itself. However, *Tolozza et al.*^[38] found that cumulative corticosteroid exposure in SLE patients was associated with low vitamin D levels.

Additional support for our results, two refractory thrombocytopenic patients with concomitant lupus-like syndrome responded to hydroxychloroquine and high-dose vitamin D replacement therapy^[39]. Also in a case study, 48-year-old female with refractory ITP, who was found to have an inadequate level of 25(OH) D, and significantly responded to high dose vitamin D^[4]

Thus, the present findings conclude that vitamin D deficiency may have an important role in the pathogenesis of primary immune thrombocytopenia and vitamin D supplementation might reduce the risk of the disease by modulating the immune system.

DECLARATION OF INTEREST:-

The authors report no conflicts of interest

Table (1):- Comparison between the three studied groups regarding clinical characteristics:

		Group I ITP patients	Group II AML,MDS, AA	Group III Healthy control	One Way ANOVA test		Post Hoc analysis		
		No. = 40	No. = 20	No. = 20	F/X ² *	P-value	P1	P2	P3
Age	Mean \pm SD	33.41 \pm 9.67	36.70 \pm 13.30	29.90 \pm 8.07	2.157	0.123	0.25 2	0.22 2	0.04 1
	Range	18 – 50 years	19 – 60 years	18 – 44 years					
Sex	Female	36 (90.0%)	16 (80.0%)	17 (85.0%)	1.159	0.560*	0.50 2	0.88 7	1.00 0
	Male	4 (10.0%)	4 (20.0%)	3 (15.0%)					
HB (g/dl)	Mean \pm SD	11.31 \pm 1.48	7.80 \pm 1.03	12.66 \pm 1.37	71.051	0.000	0.00 0	0.00 1	0.00 0
	Range	8.8 – 15	5.5 – 9.6	10.5 – 15.8					
PLT (x10⁹/l)	Median (IQR)	114 (64 – 200.5)	36.5 (19 – 56.5)	296 (230 – 318.5)	44.968	0.000•	0.00 0	0.00 0	0.00 0
	Range	15 – 352	2 – 98	176 – 442					
MPV (fl)	Mean \pm SD	11.16 \pm 2.14	9.10 \pm 3.40	9.79 \pm 1.01	5.846	0.004	0.00 2	0.03 5	0.35 2
	Range	6.8 – 19.1	0 – 14.3	8.1 – 11.5					

*: Chi-square test.

•: Kruskal-Wallis.

P1: Group I vs Group II.

P2: Group I vs control group.

P3: Group II vs control group.

(MPV=mean platelet volume).

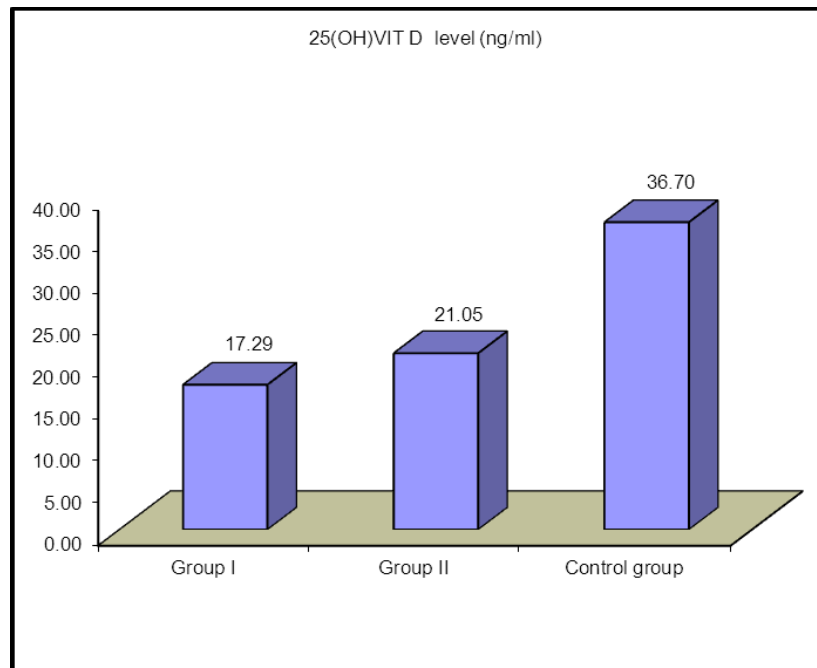


Figure (1):-Showing that 25(OH) D levels were significantly lower in ITP patients and patients with thrombocytopenia due to other causes than healthy control ($p=0.000$).

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RESEARCH ARTICLE

WRIST ARTHRODESIS IN RHEUMATOID ARTHRITIS USING RECONSTRUCTION PLAT.

Dr. Amit Lakhani, Dr. Aniketa Sharma, Dr. Uday and Dr. S M Bhatnagar.

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Abstract

This paper tries to find out the implications of trade liberalization on international trade tax revenues and its macroeconomic implication on Pakistan economy. The theoretical approach of trade reforms and its impact on the direction of change of revenue is ambiguous because of its dependence on the productivity of 'trade tax revenues'. By using the data of twenty five years, the paper first establishes the productivity of 'trade tax revenues' in Pakistan, then by an econometric analysis exploration of the relationship between trade revenues and trade liberalization in Pakistan is done. It was observed through our analysis that there is high productivity of trade tax revenues, further, our results show a positive and significant relationship between trade liberalization and collection of trade tax revenue. To support this relationship, a second test of Granger's causality is also performed which confirm the causal relationship between trade tax revenues and trade liberalization. It was suggested that supportive macroeconomic policies are prerequisite for successful trade liberalization.

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Introduction:-

Rheumatoid arthritis is a disease of chronic polyarticular inflammation that leads to joint swelling, joint deformity and loss of joint function. RA occurs in 1 to 3% of adult population with women affected slightly more than men¹. Rheumatoid hand deformities usually are bilateral and symmetrical. Wrist and metacarpophalangeal Joints are affected early². Management depends upon pain and loss of function. Operative treatment varies with the patient's age, handedness, occupation, needs, and expectations³. Five patients of RA from agricultural back ground were reviewed after wrist arthrodesis using 3.5mm reconstruction plate and iliac crest bone graft. All cases united well with marked improvement in wrist strength and diminished pain allowed improved function.

Material and Methods:-

Five patients(four female and one male) aged between 28 to 44 years underwent this procedure in our hospital. All conservative methods including wrist brace, intra articular steroid and PIN denervation⁴ were tried. Patients were explained about the procedure and limitations. Patients were admitted after all relevant investigations and preanaesthetic check up. wrist arthrodesis was done through dorsal approach and fusion was done in 10 to 20 degree of dorsiflexion and neutral to 5 degree of ulnar deviation. Pop slab was given from above elbow to mcp joint with finger in slight flexion. At 3 weeks stitches were removed and below elbow pop cast was given for further 6 weeks. Routine radiography at 6 and 12 weeks was done to check bony fusion.

Observation and Results:-

Patients were followed for a period of ten months. There were five wrist in four female and one male. Right side was in three and left in two. All procedures were done in dominant limbs. Finger motion was regained after 3 months of aggressive physiotherapy in four cases. One female aged 44 years took 5 months to regain full finger movements due to poor follow up and uncomplained physio. All cases united well between 12 to 16 weeks.



1. X ray showing wrist arthritis pre operative AP



2. X ray pre operative lateral view



3. Interosseus radio ulnar ligaments (intraoperative)



4. Xray at 3 weeks AP view



5. X ray at 3 weeks lat view



6. X ray 3 months lateral view



7. X ray 3 months lateral view

Discussion:-

Despite having trends towards many reconstructive procedures wrist arthrodesis remains an important procedure. Sorial et al. reported successful fusion in 18 patients with the use of a radial sliding graft fixed with a dynamic compression plate⁵. There are many studies in literature of wrist arthrodesis in painful wrist with syntheses fusion plate^{6,7} but due to poor patient affordability we used 3.5 mm reconstruction plate. Rheumatoid arthritis with painful destruction of the joint is a common indication for wrist fusion. Plate fixation provides great stability in all directions and delivers a high fusion rate. Common problems associated with wrist arthrodesis plates are plate discomfort and tendon irritation⁸. But we did not encounter any of these problems.

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RESEARCH ARTICLE

COMPRESSION DU NERF ULNAIRE AU POIGNET PAR UNE TUMEUR A CELLULES GEANTES : A PROPOS D'UN CAS ULNAR NERVE COMPRESSION IN GUYON'S CANAL BY GIANT CELL TUMOR: REPORT A CASE.

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Key words:-

Guyon's canal, giant cell tumor.

Abstract

A case of ulnar nerve compression in Guyon's canal is reported in 43-year-old woman. Magnetic resonance imaging demonstrated a giant cell tumor confirmed by histological examination. Surgical treatment had fully restored sensory and partial motor function of the right hand.

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Introduction:-

La compression du nerf ulnaire n'est pas rare, c'est la 2^e neuropathie compressive après celle du nerf médian au niveau du canal carpien. Si le site de compression le plus fréquent est le coude, la compression au niveau du canal de Guyon n'est pas exceptionnelle et souvent et souvent due à une cause locale. Nous rapportons le cas d'une patiente présentant un syndrome du canal de Guyon révélant une tumeur à cellules géantes.

Observation:-

Il s'agit d'une patiente âgée de 43ans, droitière, femme au foyer, ayant consulté pour des paresthésies avec fourmillements et picotements au niveau du territoire du nerf ulnaire. L'examen clinique a révélé une atrophie de l'éminence hypothénarienne, un signe de wartenberg positif [figure 1], une diminution de la sensibilité des deux derniers doigts et une absence de l'abduction et de l'adduction des deux derniers doigts. La radiographie standard a objectivé la masse en regard de l'éminence hypothénarienne et l'IRM a précisé son origine au niveau de la gaine des fléchisseurs comprimant et refoulant le pédicule ulnaire [figure 2]. L'exploration chirurgicale a montré une tuméfaction jaunâtre, encapsulée, se développant à partir de la gaine des fléchisseurs des quatres derniers doigts [figure 3]. L'intervention a consisté en une résection de la totalité de tumeur et la neurolyse du nerf ulnaire jusqu'à sa division dans le hiatus pisi-unciformien puis section du ligament pisi-unciformien et de l'arcade fibreuse des hypothénariens. Les suites postopératoires ont été simples, la patiente a démarré la rééducation fonctionnelle une semaine après l'intervention. L'examen anatomopathologique a confirmé le diagnostic de tumeur à cellules géantes. Un an après son intervention la patiente a récupéré complètement la sensibilité de la main et partiellement la force motrice.

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Discussion:-

La compression du nerf ulnaire dans le canal de Guyon n'est pas exceptionnelle. Décrite pour la première fois par l'urologue français Félix Guyon en 1861 [1]. Ses limites anatomiques bien précises conditionnant la symptomatologie clinique ont permis de diviser le canal de Guyon en trois zones [2,3].

La zone I, correspond à la zone où le nerf ulnaire va se diviser en ses deux branches terminales, superficielle sensitive et profonde motrice. La compression en cette zone va produire soit un déficit sensitivo-moteur, soit sensitif ou moteur.

La zone II, se situe autour de la branche motrice. La compression à ce niveau sera responsable d'une symptomatologie purement motrice.

La zone III, autour de la branche superficielle. Sa symptomatologie sera purement sensitive.

Plusieurs étiologies peuvent être responsables du syndrome du canal de Guyon [3]. Les tumeurs à cellules géantes viennent au 2^o rang après les kystes synoviaux. Ces tumeurs bénignes malgré leur grande fréquence au niveau de la main [4] sont mal diagnostiquées en préopératoire. Dans une revue de la littérature seulement quelques cas de tumeurs à cellules géantes responsables de syndrome du canal de Guyon ont été décrits [5-11]. Ces tumeurs bénignes se développent à partir de la gaine synoviale des tendons et des articulations du poignet et de la main, et plus précisément au niveau du dédoublement externe de la gaine ce qui les différencie des synovites villonodulaires à développement interne [12]. Les tumeurs à cellules géantes peuvent être localisées ou diffuses pouvant intéresser également le genou, la hanche...

Pour le diagnostic, l'électromyogramme (EMG) reste le maître examen complémentaire des syndromes compressifs. Dans notre cas, il a objectivé un allongement de la conduction sensitive du nerf ulnaire à sa partie distale et une atteinte de la branche profonde motrice. L'IRM a un rôle prépondérant dans le diagnostic étiologique, elle permet de faire le diagnostic différentiel avec le sarcome des parties molles qui produit un signal d'intensité égale ou supérieure à celle de la graisse en T2, alors que les tumeurs à cellules géantes ont un signal entre le muscle et la graisse en T2 [13]. De plus, les tumeurs à cellules géantes se rehaussent après injection de Gadolinium du à la prolifération des capillaires dans le stroma, ce qui les différencie des kystes synoviaux [14]. Le traitement des tumeurs à cellules géantes est essentiellement chirurgical et repose sur l'excision complète de la tumeur avec un risque de récurrence moyen de 26,5% durant les deux premières années [15]. Ces récurrences sont favorisées par l'excision incomplète, la localisation au niveau du pouce ou l'érosion de l'os [16]. L'irradiation, autrefois utilisée pour éliminer le risque des récurrences est actuellement dépassée vu ses effets secondaires, en particulier la contracture de la main [17]. À côté de l'exérèse de la tumeur, on procède à une neurolyse du nerf jusqu'à sa division dans le hiatus pisi-unciformien où la section de l'arcade fibreuse des hypothenariens et du ligament pisi-unciformien est indispensable [18].

Conclusion:-

Le syndrome du canal de Guyon est rare, souvent due à une cause locale. Les tumeurs à cellules géantes fréquentes au niveau de la main doivent faire partie des diagnostics différentiels dominés par les kystes synoviaux. L'IRM est très utile dans l'orientation étiologique.



Figure 1:- Atrophie de l'éminence hypothénarienne et signe de Wartenberg.

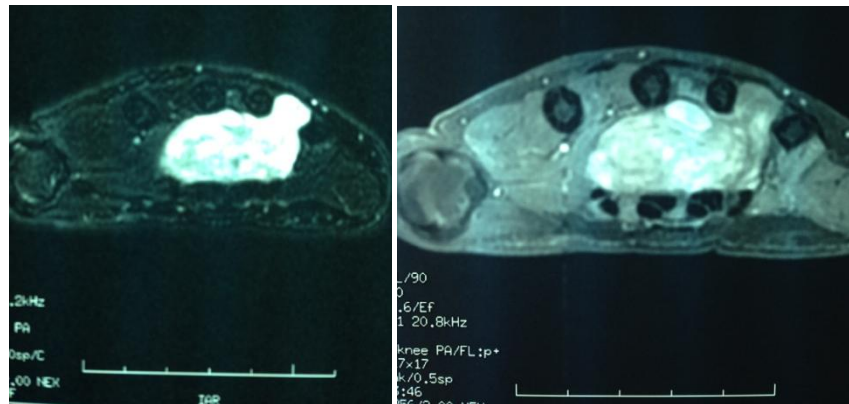


Figure 2:- IRM de la main objectivant la tumeur et son retentissement sur les structures avoisinantes.



Figure 3:- Excision de la tumeur.

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RESEARCH ARTICLE

TUMEUR GLOMIQUE DE L'HALLUX. NOUVEAU CAS ET REVUE DE LA LITTÉRATURE GLOMUS TUMOR OF HALLUX. NEW CASE AND REVIEW OF LITERATURE.

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Key words:-
 glomus tumor-hallux.

Abstract

- Glomus tumors are rare and benign, arising from a neuromyoarteriel glomus body. If the digital location is well known in surgery of the hand, the extradigitales locations suffer from a misunderstanding ending in diagnostic and therapeutic errors. We report a new case of tumor glomique of the hallux, and through a review of the literature, we would try to draw the attention towards these atypical location.

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Introduction:-

Les tumeurs glomiques sont des tumeurs bénignes et rares qui se développent aux-dépends du glomus neuromyoartériel des anastomoses artério-veineuses. Elles touchent surtout les extrémités digitales, mais les localisations extra digitales ne sont pas rares mais surtout méconnues ce qui est responsable du retard de leur diagnostic et de leur prise en charge. Le but de notre travail est de rapporter un nouveau cas de tumeur glomique de l'hallux et d'attirer l'attention vers cette localisation inhabituelle.

Cas Clinique:-

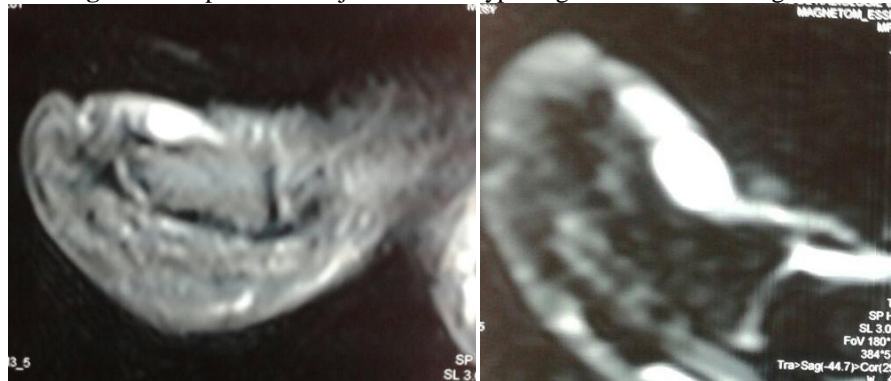
Il s'agit d'une patiente âgée de 35 ans, sans antécédents pathologiques particuliers qui présente depuis 4 ans, des douleurs intermittentes siégeant à la base du gros orteil droit, coïncidant avec un traumatisme du même orteil. Nous avons reçu la patiente qui a consulté à plusieurs reprises vu l'augmentation de l'intensité des douleurs et l'absence d'amélioration sous traitement symptomatique. L'examen clinique de cette patiente montrait une décoloration de la base de l'ongle qui était très douloureuse à la palpation. L'examen ostéo articulaire de l'hallux droit était sans particularités. La radiographie standard ainsi que le bilan biologique étaient normales. L'IRM a objectivé un hyper signal autour du lit unguéal suggérant la possibilité d'une tumeur glomique [figure 1]. Une excision par voie latérale sous unguéale a été réalisée et l'examen anatomopathologique a confirmé le diagnostic de tumeur glomique. Les suites opératoires ont été simples, la patiente a eu un soulagement immédiat des douleurs et l'examen clinique de la patiente après un recul de 3 ans n'a pas montré de récurrence de la symptomatologie.

Discussion:-

Les tumeurs glomiques sont rares et bénignes, elles représentent environ 1 à 5% de l'ensemble des tumeurs des tissus mous [1]. Ce sont des tumeurs qui touchent le sujet adulte, l'âge moyen est de 40 ans, elle est rare avant 20 ans [2]. Signalée pour la première fois par Wood en 1812 [3] qu'il a dénommé (nodule sous cutané douloureux), c'est Masson [4] en 1924 qu'il l'a défini en décrivant le glomus qu'il nomme ainsi à cause de sa similitude avec la glande vasculaire coccygienne de Luschka [5]. Si la localisation digitale est bien connue en chirurgie de la main [6], les localisations atypiques [7-13], comme notre cas au niveau de l'hallux, souffrent de méconnaissance, ce qui conduit à des erreurs diagnostiques et thérapeutiques. La symptomatologie clinique associe une triade classique

avec douleurs sévères, point sensible et intolérance au froid. Ce qui caractérise également la symptomatologie est le contraste entre l'intensité des signes subjectifs et la pauvreté des signes objectifs. Rarement sont les cas où il y a une histoire traumatique comme dans notre cas [14]. Il n'y a pas d'imageries spécifiques permettant la confirmation diagnostique, cependant l'échographie, malgré sa faible spécificité aide à localiser la lésion [15]. L'IRM reste le gold standard dans le diagnostic des tumeurs glomiques, elle précise le siège exact de la lésion et sa relation avec les structures avoisinantes [16-18]. Son traitement est toujours chirurgical, il consiste en une exérèse chirurgicale qui entraîne une disparition rapide des douleurs. Les récurrences sont rares mais possibles, elles sont toujours précoces et le fait d'exérèse incomplète, d'où la recommandation de certains auteurs [18-20] d'exciser plus que les limites apparentes de la tumeur.

Figure 1:- aspect IRM objectivant un hyper signal autour du lit unguéal.



Conclusion:-

Les tumeurs glomiques sont rares mais non exceptionnelles. Elles peuvent siéger partout ou existe les glomi. Devant toute douleur avec ou sans masse palpable, sans étiologie évidente, le diagnostic de tumeur glomique doit être évoqué.

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RESEARCH ARTICLE

Do Gamma Rays of Cancer Radiotherapy Effect on the Sequence of Collagenase Gene?

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Key words:-

Acute Leukemia, HLA Polymorphism.

Abstract

Background: expression of certain HLA alleles, considered susceptible factor to acute leukemia (AML, ALL), there were clear associations between resistance or susceptibility to cancer and (HLA) profile, this work **aims to** investigate role HLA-DRB1 and anti HLA-G in development of AML. **Patients and methods:** (40) patients with acute leukemia aged from 4 to 60 years (23 male and 17 female), and 20 healthy subjects as a control, all were tested for anti-HLA-G by four-color flow cytometer using FACS Canto (BD Biosciences: USA), HLA-DRB1 by Sequence Specific Oligonucleotide PCR (SSO-PCR), DNA purification by (Spin Protocol), Qiagen, Hilden, Germany, QIA amp DNA Mini, thermal cyclers BIO-RAD PTC-100 (USA), hybridization with DNA probes, alleles detection; INNO-LiPA HLA typing, Fuji Rebio Group, Belgium, with the INNO-LiPA HLA-DRB1 Plus kit, Cat. No. #58355. **Statistical analysis:** SPSS 19 edition was used, gene frequencies of HLA-DRB1 alleles were calculated by direct count. **Results:** there is an allelic association between HLA-DRB1 and leukemia, patients and control showed significant difference between DRB1*13 ($p = 0.006$ and $p = 0.016$ respectively) (protective nature), but there were moderate difference among DRB1*06 ($p = 0.098$), DRB1*14 ($p = 0.098$), (also, protective nature) and *15 ($p = 0.098$) (Risk factor). However, we did not observe any significant difference for the others HLA-DRB1. **Conclusion:** HLA-DRB1*13, and HLA-DRB1*15 alleles were protective while HLA-DRB*06 and DRB1*14 were predisposing factor for AML.

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Introduction:-

Leukemia is uncontrolled proliferation of hemopoietic cells (1). Acute leukemia is divided into a number of different subtypes based upon clinical, morphological, immunophenotyping, cytochemical, cytogenetic and molecular biology or by combinations of these characteristics (2). Although a study of HLA understanding of that relationship has been slow to emerge (3). Studies show that HLA genes are involved in various mechanisms of pathogenesis and immunoediting of hematological diseases (4). Leukemia was the first disease in which involvement of the major (MHC) was reported (5). Studies on the role of the HLA in leukemia susceptibility represented a lot of data with many questions were raised but few confirmatory answers were available (6). Although, not all studies of leukemia and HLA demonstrate associations. Further studies are required to support these conclusions (7). Studies of the

correlation between HLA polymorphisms and susceptibility or resistance to disease started soon after serological methods for HLA class I had been standardized (8).

There are clear associations between resistance or susceptibility to cancer and (HLA) profile of an individual (9). The association between HLA and various hematological malignancies has been studied extensively (10).

The rate of expression of certain HLA alleles is considered among the factors of susceptibility to develop leukemia. Many studies have been performed in humans investigating the role of HLA antigens in pathogenesis of hematological malignancies. Although, the association of HLA with the molecular features of leukemic cells is still unclear (11). HLA-G contributes to tumor escape host responses, and its potential clinical relevance in various malignancies was discussed (12). The part devoted to HLA-G in tumor escape remains to be more precisely determined in these acute pathologies (13).

Patients and methods:-

Design and population:-

This prospective study has been carried out in Hematology Unit in Clinical and Chemical Pathology department of Sohag University Hospital in the period from January, 2012 to August, 2015. Study was conducted on forty (40) patients; 23 males (57.5%) and 17 females (42.5%), suffering from acute leukemia of both lymphoid and myeloid lineage, they aged from 4 to 60 years old. The control group consisted of 20 unrelated healthy volunteers. The control panel reasonably corresponds to the same age, sex distribution, living in the same geographical area and with the same ethnic origin of the patients' panel.

Ethics:-

This study was conducted in accordance with the ethical standards of the Helsinki Declaration of 1964, as revised in 2000 and was approved by the Institutional Review Board of the Faculty of Medicine, Sohag University. Informed consent was obtained from all the study participants.

Sampling: Blood samples were collected from all patients and healthy subjects. 2 ml of EDTA anticoagulated blood were collected. Diagnosis of leukemia was based on complete blood count, blood film morphology, B.M examination cytochemistry and immunophenotyping.

Flowcytometry for anti HLA-G: Four-color flow cytometer; FACS Canto (BD Biosciences, USA) was used. Purified mouse anti-human HLA-G denatured: FITC goat anti-mouse IgG/IgM 0.5 mg polyclonal Cat. No. # 555988. Purified mouse IgG1, κ isotypic control 0.1 mg, MOPC-21 Cat. No. # 555746. FACS lysing solution 10X ; BD Cat. No. # 349202. Falcon disposable 12 x 75-mm capped polystyrene test tubes B.D Cat. No. #2058.B.D. Monoclonal antibodies to human cell surface antigens (anti-CD33, anti-CD13 and anti-CD19 stained with Fluorescein isothiocyanate (FITC) and anti-CD45 antibodies stained with phycoerythrin (PE)).

Flowcytometry for anti HLA-G was carried out in Hematology Unit in Clinical and Chemical Pathology Department of Sohag University Hospital.

HLA Typing:-

All patients and controls were subjected to molecular HLA-DRB1 typing. 2ml EDTA-anticoagulated blood samples were collected from patients and controls. Samples stored by freezing to -80°C then thawed in 37°C in the incubator in the day of the run. Samples were processed for detection of HLA-DRB1 alleles by Sequence Specific oligonucleotide PCR (SSO-PCR) in following steps : DNA extraction using the spin column technique (QIAamp DNA Blood Mini kits; Qiagen, Hilden, Germany) was performed according to the manufacturer's guidelines; polymerase chain reaction (PCR) amplification of (exon 2) of the HLA-DRB1 alleles (INNO-LiPA HLA-DRB1 Amplification Plus kit; Fuji Rebio Europe, Ghent, Belgium) were performed and the amplicons were chemically denatured to form single-stranded DNA. Reverse dot-blotting was performed on a nylon membrane (INNO-LiPA HLA-DRB1 Plus strips, Fuji Rebio Europe), which contains an array of immobilized, sequence-specific oligonucleotide (SSO) probes. The biotin-labeled amplicons were then bound (hybridized) to these SSO probes, which contain a complementary target sequence and, thus, were captured onto the membrane strip; 3) visualization of the results was achieved by incubating with an enzyme conjugate (streptavidin and alkaline phosphatase), which binds to the biotin of the PCR product, followed by the addition of a substrate. The bands with the captured PCR product turned blue. Interpretation was achieved by using standard tables with the help of specialized software:

LiRAST™ FOR LiPA HLA V6.0X.

HLA typing was carried out in Immunology Unit in Clinical and Chemical Pathology Department of Mansoura University Hospital.

Statistical Analysis:-

Statistical analyses were carried out using the SPSS® statistical package. Released 2010. IBMSPSS Statistics for Windows, Version 19.0. Armonk, NY: IBM Corp., Microsoft Office Excel 2013 for Windows and online analytical computer assisted sites.

Gene frequencies of the HLA-DRB1 alleles of both study groups were calculated by direct count. For comparison between the groups, we used χ^2 analysis by using 2×2 contingency tables and Fisher's exact test when appropriate; *P* values less than 0.05 were considered statistically significant. For estimating risks, we employed odds ratio (OR) with a 95% confidence interval (95% CI).

Results:-

The polymorphism of HLA-DRB1, genes was investigated in twenty normal individuals and forty leukemic patients. The clinical data of the examined subjects were presented in table (1)

Comparison of the results between the normal population and acute leukemia patients revealed that there is no difference between anti HLA-G antibodies in both patients and control groups as in table (2) figure (1,2 and), there is an allelic association between certain HLA-DRB1, and leukemia disease. Our results showed that in leukemic patients and in normal individuals, the difference between DRB1*13 was significant (*p* = 0.006 and *p* = 0.016 respectively) (protective nature), but there were moderate difference among DRB1*06 (*p* = 0.098), DRB1*14 (*p* = 0.098), (also, protective nature) and DRB1 *15 (*p* = 0.098) (Risk factor). However, we did not observe any significant difference for the others HLA-DRB1.

As regard gender the HLA-DRB1*10 allele was not detected in any of the male patients (*P* = 0.091). With no significance difference between males and females in other HLA-DRB1, illustrated in figure (3). According to the immunophenotyping of leukemia, the HLA-DRB1*01 allele was not detected in any of the AML patients in relation to ALL patients (*P* = 0.109). With no significance difference between AML and ALL in other HLA-DRB1. The HLA-DRB1*14 allele was not detected in any of the controls in relation to ALL patients with moderate significance (*P* = 0.057) (relatively risk factor). The HLA-DRB1*13 show high frequency in controls in relative to ALL patient with moderate significance (*P* = 0.054) (Relatively Protective). With no significance difference between ALL and controls in other HLA-DRB1 as demonstrated in table -3 and 4.

HLA-DRB1*06 allele was not detected in any of the controls in relation to AML patients that significant difference (*P* = 0.044) (risk factor). The HLA-DRB1*13 show high frequency in controls in relative to AML patient (*P* = 0.006) (Protective nature). With no significance difference between AML and controls in other HLA-DRB1.

Discussion:-

The first study on HLA in human leukemia demonstrated an increased frequency of HLA-A2 in ALL in 1967. From that date many previous studies have demonstrated some significant differences in HLA allele frequencies in leukemic patients and normal subjects which had reported by Diase et al (2015) and others (14, 15,16). In our study, significant positive association with the disease, in patients compared to controls, was found for two alleles: HLA-DRB1*013. Also there were moderate difference among DRB1*06 DRB1*14, and *15. It is suggested that HLA-DRB1*13, and HLA-DRB1*15 alleles may play a protective genetic factor against leukemia while the HLA-DRB1*06 and –DRB1*14 allele could be a presumptive predisposing factor. Regarding anti-HLA-G antibodies analysis, no significant association was found between patients and control groups. Also, our study shows that HLA-DRB1*10 allele was not detected in any of the male patients with moderate significance with no significance difference between males and females in other HLA-DRB1 or anti-HLA-G antibodies. Our results are consistent with Du et al (2013), that demonstrated that the allele frequencies of DRB1*15 obviously decreased in patients with leukemia so, it can be considered as genetic indicators for resistance of leukemia. But they demonstrated that the DRB1*07 have a protective role too (17). Diase et al (2015) had reported that HLA-DRB1*13, which showed a decrease in patients, should be protective against ALL (14). The findings that were demonstrated also by Dorak et al (2013), who reported that the frequency of the HLA-DRB1*13 allele was lower in male patients of leukemia (15).

Also Bosen et al (2013); reported that the frequency of the HLA-DRB1*13 allele was lower the female ALL patients compared to controls (7). On the other hand, Chaing et al (2012) had found that there is significant positive association with the disease, in patients compared to controls, was found for three alleles: HLA-DRB1*13 and HLA-DRB*01 (10). Also there were moderate difference among DRB1*04, *03 and *15. It is suggested that HLA-DRB1*13 (on contrast to our results), *03 and *04 alleles may play a presumptive predisposing factor while the HLA-DRB*01 and *15 (on contrast to our results) alleles could be a protective genetic factor against leukemia (16). A significant increase in the frequency of the HLA-DRB1*04 allele in the overall and ALL patients, was reported in many studies although there is no significant difference between patient and controls (17). We found that the HLA-DRB1*14 allele was not detected in any of the controls in relation to ALL patients with moderate significance (relatively risk factor). There is no significance difference between ALL and controls in other HLA-DRB1 or anti-HLA-G antibodies. Our results are consistent with Ehernberg et al (2014); who found, significant decrease in HLA-DRB1*04 (in contrast to be previously reported by Bosen et al (2013), and an increase in allelic distribution of HLA-DRB1*03 in ALL patients in relative to controls which show no significant difference in our study(7,18).

In our study, The HLA-DRB1*06 allele was not detected in any of the controls in relation to AML patients that significant difference) (risk factor). The HLA-DRB1*13 show high frequency in controls in relative to AML patient (protective nature). There is no significance difference between AML and controls in other HLA-DRB1 or anti-HLA-G antibodies.

Our findings are inconsistent with Elansary et al (2015), who reported that high significant increase allelic distribution in HLA-DRB1*11 and moderate decrease in HLADRB1*07 in AML patients in relative to controls which show no significant difference in our study (19).

Bosen et al (2013), observed that the frequencies of the HLADRB1*15 alleles in patients with AML were significantly higher in AML patients compared with healthy controls, Du et al (2013), observed significant increase in allelic distribution of HLA-DRB1*07 in AML patients and suggests that it is susceptible to the disease in contrast to Ehernberg et al (2014) observations. These results not consistent with Chaing et al (2012) that demonstrated that allelic distribution of the HLA-DRB1*13 is significantly high among AML and overall acute leukemia patients group in relation to controls (7, 10, 17 and 18). Few studies had been performed on HLA-G expression in different types of leukemia patients, however, data are limited and conclusions remain controversial and discordant (19). Regarding anti-HLA-G antibodies analysis, no significant association was found between patients and control groups. Also our results are inconsistent with the results of other authors who have not found membrane bound or cytoplasmic HLA-G expression (7). A group of previous studies addressed that no cell surface HLA-G was expressed in various types of hematopoietic diseases, such as AML, ALL, CLL and CML. In contrast to these previous studies, B-CLL samples express cell surface HLA-G antigen in a variable proportion of leukemic tumor cells. Methodology difference address this controversy (20). The discrepancy between the results of different studies, including our results, may be attributed to the differences in population race and geographical distribution. However, further larger studies are required to support our findings.

Conclusion:-

Some HLA alleles are associated with an increased susceptibility while others are protective. Although, some of the results from the present study support these earlier findings. The discrepancy between the results of different studies, including our results, may be attributed to the differences in population race and geographical distribution, environmental, occupational factors.

Conflicts of interest:-The authors had no conflicts of interest to declare in relation to this article.

Table (1):- Clinical data of the studied groups

Variables	Control	Patients
Age (years)		
Mean± SD	32.35 ± 19.07	33.08 ± 21.79
Male/Female	11/9	23/17
Diagnosis and Frequency	100 % healthy normal	AML (22) 55% ALL (18) 45%
Total	20	40

Table (2):- Anti-HLA G positivity between cases and controls

Item		Anti HLA-G		Total
		Negative	Positive	
Patients number		17	23	40
% within group		42.5%	57.5%	100.0%
Control number		6	14	20
% within group		30.0%	70.0%	100.0%
Total number		23	37	60
% within group		38.3%	61.7%	100.0%

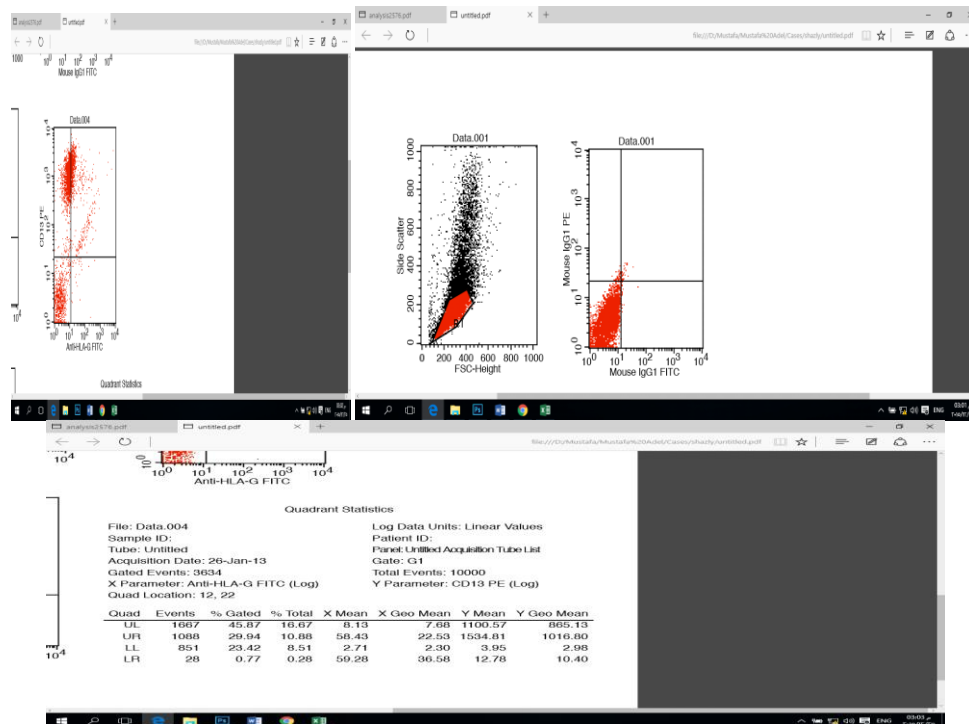
Table (3):- HLA-DRB1 alleles in acute leukemia patients compared with healthy control.

Allele	P value	Odd's ratio	Confidence interval of odd's	
			Minimum	Maximum
1	0.534 (NS)	2.662	0.122	58.122
2	-	-	-	-
3	0.686 (NS)	1.286	0.381	4.343
4	0.680 (NS)	0.778	0.236	2.568
5	0.383 (NS)	3.827	0.188	77.769
6	0.219 (NS)	6.352	0.334	120.845
7	0.638 (NS)	1.417	0.332	6.048
8	0.716 (NS)	1.541	0.150	15.83
9	-	-	-	-
10	0.534 (NS)	2.662	0.122	58.122
11	1.000 (NS)	1.000	0.290	3.454
12	0.789 (NS)	1.557	0.061	39.948
13	0.008 (S)	0.206	0.065	0.660
14	0.219 (NS)	6.352	0.334	120.845
15	0.108 (NS)	0.333	0.087	1.272
16	0.789 (NS)	1.557	0.061	39.948

S: Significant NS: Non significant

Table (4): Gene frequency of HLA-DRB1 alleles corresponding to ALL and AML patients

HLA-DRB1 alleles	Allele positivity among ALL (no. = 18)	Allele positivity among AML (no. = 22)
1	2(11.11%)	0
2	0	0
3	5(27.78%)	7(31.81%)
4	4(22.22%)	6(27.27%)
5	1(5.56%)	2(9.09)
6	1(5.56%)	4(18.18)
7	5(27.78%)	3(13.64)
8	1(5.56%)	2(9.09)
9	0	0
10	0	2(9.09)
11	4(22.22%)	6(27.27%)
12	1(5.56%)	0
13	7(38.89%)	6(27.27%)
14	3(16.67%)	2(9.09%)
15	2(11.11%)	3(13.64%)
16	0	1(4.55%)

**Fig (1):-** Case No. 27: AML Patient with HLA-G Positive

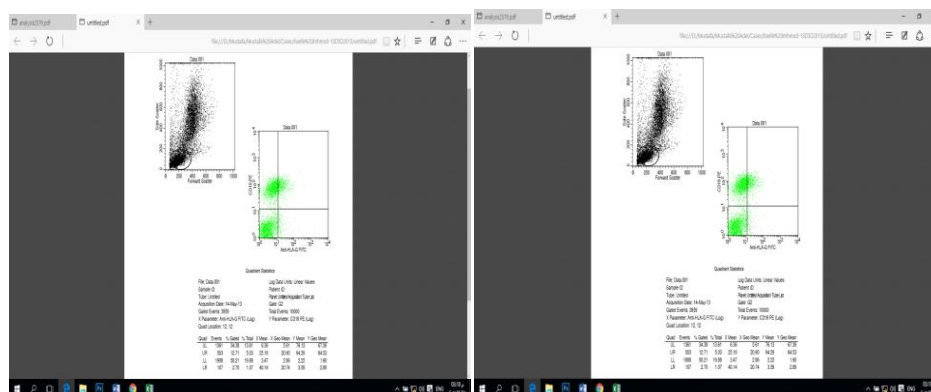


Fig (2):- Case No. 28: ALL Patient with HLA-G Positive.

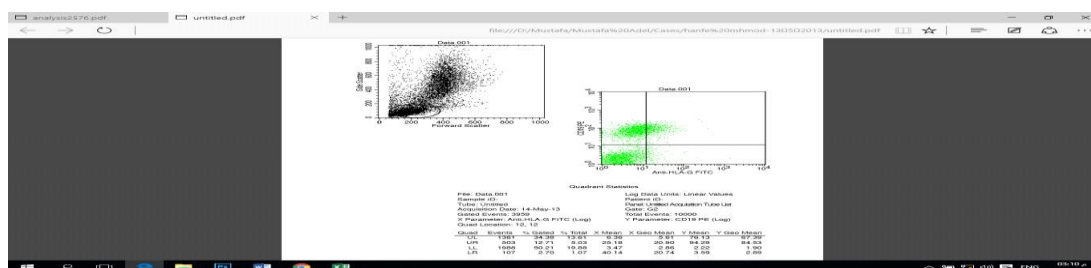


Fig (3): Odd ratio of HLA-DRB1 alleles corresponding to males and females frequency

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RESEARCH ARTICLE

UNDERSTANDING THE SOCIODEMOGRAPHIC PROFILE AND LONG TERM COMMUNITY PSYCHOSOCIAL NEEDS OF CHILDREN AND ADOLESCENT SURVIVORS OF A NATURAL DISASTER ONE DECADE AFTER 2005, 'SNOW-TSUNAMI' IN KASHMIR.

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Abstract

Objective: To explore the sociodemographic profile and long term psychosocial needs of children and adolescents survivors of snowstorm disaster who survived a natural disaster (snow storm) in Kashmir more than a decade back.

Methods: It is a cross sectional study done in a disaster affected population of children and adolescents in South Kashmir 10 years after the snow storm disaster. The study was designed as a survey of the sociodemographic and long term psychosocial correlates and mental health status of young adults living in the same vulnerable area today also.

Results: Participants were 139 young adult inhabitants of snow storm stricken area in south kashmir in 2005 (exposed group) and 71 residents of a geographically and ethnoculturally identical population in central Kashmir area. Rampant psychosocial problems among the young disaster survivors long after the disaster, when no one is around to help after the initial knee jerk response of relief agencies, is clearly reflected through the results of our study.

And measures to address them as clearly revealed by our study through an outreach psychosocial and clinical service programme long after the disaster when no one is around to help after the initial knee jerk response of relief agencies is greatly stressed.

Conclusion: The results indicate that disasters can have long-standing adverse effects on survivor's psychosocial well being and mental health even after 10 years later. Post trauma morbidity is more lasting in people with lesser support viz lower socio economic class, rural background and lower education.

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Introduction:-

Though disasters are inevitable part of human existence but increasingly frequent massive disasters and horrendous events over the past decade or so have not only directly impacted many regions of the world but indirectly affected humanity as a whole¹⁶.

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A disaster is the outcome of an extraordinary occurrence that destroys property, kills people, and produces physical or psychological damage as well as overwhelming the adaptive potential of the social group¹².

Disasters produce multiple patterns of outcome depending on a combination of risk and resilience factors.² As compared to manmade disasters; natural disasters occur suddenly, usually without warning and kill a large number of people in defined or limited geographical area over a limited period of time.

The consequences of natural disasters are worse and the death toll is greater in developing countries¹.

Children and adolescents are at a higher risk of becoming victims and suffering the losses resulting from disasters. Recent findings have shown that children's reactions to disasters can be severe and are not merely fleeting, transitory events that quickly dissipate⁴. Often an entire community is impacted, further undermining a child's sense of security and normalcy. These factors present a variety of unique issues and coping challenges, including issues associated with specific types of natural disasters, the need to relocate when home and/or community have been destroyed, the role of the family in lessening or exacerbating the trauma, emotional reactions, and coping techniques¹¹. Efforts to provide effective services and interventions for children and adolescents following a disaster therefore represent an important, but frequently overlooked, mental health need.

The northern most state of the country, Kashmir (Jammu and Kashmir) has been regarded as the heaven on earth. Its beauty and hospitable population is legendary. Unfortunately the state has been witnessing a continuous mass manmade disaster situation for more than 25 years, with thousands of people dead, maimed and mutilated, many missing or confined, thousands of children orphaned, and women widowed, a colossal damage to the property, and a damage to the cultural ethos including en-mass migration of a minority community¹³. As if this unrelenting manmade mass traumatic situation and its sequel was not enough, a small picturesque village "Waltengo Nard" at the foothills of Pir Panchal range of Himalayas about 90km from Srinagar, the summer capital of Kashmir, was struck by a devastating snowstorm following continuous heavy snowfall for five consecutive days.^{7,18}

The snowstorm was named "Snow Tsunami" by the Air Chief of India and the other rescue agencies. Almost 25% (n=164) population perished within minutes. Since dead bodies could not be moved out to graveyard, 5-10 bodies had to be buried together in mass graves in the same houses where some of the injured and surviving people were still trapped. One-year longitudinal study of the snow storm disaster survivors by Margoob et al. in 2006 had revealed a very high level of psychological problems and psychiatric morbidity among the surviving population¹⁸. Disasters can have detrimental consequences at the individual and community level, altering the infrastructure of societies and causing both physical and psychological suffering among those affected. Mental health research has increasingly focused on the adverse consequences of disasters on the health of survivors but most of the research available so far from natural disaster related literature is confined to only short term impact of such catastrophes. So to advance our understanding of the consequences of disasters on survivor's mental health, by examining the possible sociodemographic vulnerabilities and psychosocial outcomes 10 years after storm disaster was undertaken as the objective of this research endeavor.

Material and Methods:-

Study Design:-

This was a cohort cross sectional study, the study group comprised of the young adults of the whole surviving population of the disaster affected area (exposed) at the Waltengo Nard, Qazigund. The control group included the population of snow storm unaffected area (unexposed) about 100 km away from the disaster site at Inder Nard, Kangan, District Gandral with identical sociodemographic and socio cultural background

Sample:-

All the young adult snow storm disaster survivors of age between 19 years to 39 years were approached to be included in the study, after the households were enumerated with the help of key persons at the site. Similarly in the control population, households were included randomly and their corresponding age inmates were enrolled as study subjects. Informed consent was obtained from the from each subject included in the study. Those who refused to consent or could not be contacted were excluded.

Tools:-

A semi-structured interview schedule specifically prepared for the study was used to collect information on the socio-demographic variables in both groups and the extent of exposure to snow storm disaster in the study group. Kuppaswamy's Socioeconomic Status Scale ¹⁰ was used for determining socio-economic status. Oslo-3 Social Support Scale (OSS-3) was used to calculate social support ³. Appropriate statistical tools to analyse the results were used.

Results:-**Table 1:-** Present age wise make up (as on ending Oct.2016) of disaster survivors and Comparison population.

	Disaster survivors		Comparison group	
	N 49 4	(100%)	N 228	(100%)
Adult males	130	(26.31)	79	(34.64)
Adult females	131	(26.51)	65	(28.50)
children	233	(47.16)	84	(36.84)

Table 2:- Sociodemographic profile of the study group and control population-I.

Variable	Study group		Control group		Z value	P value
	N 139	% age	N 71	% age		
Age						
19-25 years	60	43.16	34	47.88	0.65	0.51
26-31 years	37	26.62	21	29.57	0.45	0.65
32-38 years	42	30.22	16	22.55	1.17	0.23
Sex						
Males	70	50.35	42	59.15	1.20	0.22
Females	69	49.65	29	40.85	1.20	0.22
Educational Status						
Primary School (1 st To 5 th Standard)	66	47.48	34	47.88	0.05	0.95
Middle School (6 th To 8 th Std.)	52	37.41	26	36.61	0.11	0.97
High School (9 th & 10 th Std.)	15	10.79	8	11.26	0.10	0.92
11 th class and above	6	4.31	3	4.22	0.03	0.97
Family Type						
Nuclear	30	21.58	21	29.57	1.27	0.20
Extended Nuclear	63	45.32	18	25.35	2.81	0.004
Joint	46	33.93	33	46.47	1.89	0.06

Table 3:- Socio demographic profile of study group and control population-II.

Variable	Study group		Control group		Z value	P value
	N 139	% age	N 71	% age		
Socio-Economic Status						
Upper						
Upper Middle			5	7.04	NA	NA
Lower Middle	5	3.59	7	9.85	1.84	0.06
Upper Lower	76	54.67	47	66.19	0.60	0.11
Lower	58	41.72	12	16.90	3.61	50.05
Social Support						
Good/Fair	40	28.77	28	39.43	1.56	0.11
Poor	99	71.22	43	60.56	1.56	0.11
Birth Order						
Eldest	20	14.38	15	21.12	1.23	0.21
Middle	70	50.35	34	47.88	0.33	0.72

Youngest	36	25.89	17	23.94	0.30	10.75
Only Child	13	9.35	5	7.04	0.56	0.56
Source Of Income						
Labourer Cattle/Farming	110	79.13	57	80.28	0.19	0.84
Semi-Government/private need based worker	20	14.38	10	14.08	0.05	0.95
Government Job	9	6.47	4	5.63	0.23	0.81

Discussion:-

The studied population of Gujars (semi-nomadic) tribe had a comparable age wise distribution between the two groups as reflected by Table 1. Gujar are a pastoral agricultural ethnic group with populations in India and Pakistan and a small number in north-eastern Afghanistan. According to the 2011 Census of India, Gujar is the most populous scheduled tribe in the state of Jammu and Kashmir having a population of 763,806. Though there were no statistically significant differences between the snow storm-exposed and comparison groups with regard to most of the demographic information, but as revealed by the result the study sample is mostly comprised of young adults belonging to the poorest strata of the society with lowest education level, families without any sustained or permanent source of income, poorest social support, (71.22% in study group and 60.56% among the control group.)

Undesirably except for the initial 1-2 months after disaster, the survivors' real immediate and future concerns were never secured. Almost every year the extremely poor and marginalised disaster hit community continues to be subjected to severe re-traumatisation in winters when following a snow fall they are evacuated and rescued to safer places by the government agencies which has continued till this year's snow fall on 18th January 2017. The importance of psycho-social consequences need to be recognized earnestly so as to develop appropriate policy with relevant plan of action and integrate it into the overall disaster management strategy to meet the mental health needs of the disaster effected populations. For children, adolescents, and young adults response to disaster involves a complex interplay of pre-existing psychological vulnerabilities, stressors and nature of support in the aftermath.

The present study is a far-sighted one which aims to determine long term psychological morbidity among the surviving children and adolescents of this disaster affected area ten years after the snowstorm disaster. Since, sociodemographic factors also play a role in trauma vulnerability and resilience, therefore, the age group studied in this research was obviously more vulnerable sub group of the disaster hit population and has faced the brunt of the storm as young children and adolescents. This is consistent with the research that has suggested that younger children possess fewer strategies for coping with both the immediate disaster impact and its aftermath, and thus may suffer more severe emotional and psychological problems¹⁷ and adolescents are also known to suffer significant morbidity under such conditions¹⁹

Our study shows that people with lower educational level have higher psychiatric morbidity. This was in accordance with an earlier study by Kar et al⁸. These findings are also in agreement with the findings of another study which found maximum morbidity (34.1 %) in less educated young population⁵, but nothing can be commented about the reported protective role of higher educational status in our studied group due to the harsh reality that more than 85% of the young adults studied by us were forced by unfavourable circumstances to abandon education before qualifying even before qualifying the 8th Std. As recently observed by Kousky(2016) not only are disasters themselves stressful and frightening, but children suffer psychological harm from the damage to their homes and possessions; from migration; from the grief of losing loved ones; from seeing parents or caregivers undergo stress; from neglect and abuse; and from breakdowns in social networks, neighbourhoods, and local economies⁹. Disasters can interrupt children's education by displacing families, destroying schools, and pushing children into the labour force to help their families make ends meet in strained times. Reinforcing coping and resilience among the disaster survivors is also imperative.

The coping strategies adopted by our subjects to tackle the psychological distress were surrender to the will of God, sharing with relatives and family members, getting busy, using tobacco and other medication, and some of them could not define it. This resulted in their rationalising the event and hence, possibly, prevented them from much psychological distress. This observation is in agreement with the observation reported by Margoob et al in the same population at one year post disaster period.¹⁴ Spiritual help from faith healers, clergy and respected elders from the community was of immense help in helping survivors to come to terms with and live down the trauma. Eastern

religions teach inevitability of fate, which helps survivors to accept and live beyond trauma.¹⁵ Spiritual involvement in the survivors of Waltengo also involved attachment to and obedience of local clergy (Imams) and spiritual leaders (Pirs). In many cases extreme avoidance or numbing of even desperate cases was significantly reduced, within a few sessions of advice and counselling by them. Integrating spiritual components as a part of the psychosocial interventions was rewarding in the management of many patients with severe post disaster psychopathology.¹⁴ Spiritual healers (Pirs) and clergy (Imams) have a central role in this process among socioeconomically deprived and educationally backward people, who in the absence of any mental health care service hold the pir in high reverence.⁶ Psychotherapeutic intervention through the medium of spirituality actually contains some essential elements of cognitive behaviour therapy including informal administration of elements of prolonged exposure, stress inoculation therapy and cognitive reframing.

Summary and Conclusion:-

The following conclusions can be drawn from the study:-

- The results indicate that disasters can have long standing effects on survivor's psychosocial well being and mental health even 10 years later.
- Post trauma morbidity is more lasting in people with lesser support viz lower socioeconomic class, rural background and low education level.
- Post trauma sequel need to be recognised in the study sample in view of having a high prevalence of trauma.
- The positive coping methods and socio- cultural bonds and spirituality need to be further strengthened.

Suggestions:-

As reflected by the study, disasters can have a long term negative impact on the psychosocial well being of the survivors, particularly the vulnerable groups such as children and adolescents. The interaction at the interface of society and environment in developing countries essentially determines the outcome of disasters. There is a need to frame a timely and well framed policy to solve disaster related problems. Raising the awareness regarding the coping mechanisms for disasters especially in the developing world should be given priority for measures of post disaster recovery. Attention needs to be paid to high risk groups and strong policy support and appropriate guidance mechanisms need to be put in place.

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RESEARCH ARTICLE

DATA SECURITY IN SMARTPHONES USING BIT-LOCKER TECHNOLOGY.

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 Operations.

Abstract

Objectives: Securing portable devices with the help of existing technology of Bit-Locker drive encryption, with more security of reset operations, if you lost your portable devices like memory cards, pen drive etc. The main objective is to make Data more secured and unauthorized access is denied when they try to access the personal information that are present in SD card after the User lost his/her Mobile/SD card.

Methods: a performance difference between Bit-Locker enabled device and Bit-Locker not enabled device with varying of read/write operation is more in non-enabled Bit-Locker device, so we will suggest to encrypt SD card using Existing Method Bit-Locker and also we suggest an Idea that User can Encrypt once in Windows Platform and Decrypt in Other Platform (Ex: Ubuntu) better way of protecting the SD card.

Findings: The issues of Bit-Locker device that is we enabled Bit-Locker drive encryption, even the drive is locked but the user can be able to format the entire drive without any interruption. so we have given better solution for these type of problem and also if you lost your portable device without encrypting the unknown user can easily misuse the data present in portable device, so we provided a solution if any unknown user wants to access your data it will ask for password, if unknown user enters 5 times wrong password the portable device will be locked and all the data is format the unknown user cannot perform any read/write operation on portable device. And also if we enabled bit-locker drive encrypted it will have decrypted in windows operating system so we introduced a concept of encrypt once and decrypt in any operating system [EODAS].

Improvements: The privacy data is more secured compared to Bit-Locker technology, with reset operation if unknown user enters five times wrong password the portable device will be format and lock the portable device with read/write operations. Performance is more improved.

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Introduction:-

Data security is the main challenges that prevents the wider acceptance of mobile devices especially within business¹. Mobile Technologies are rapidly replacing the traditional cellular phones because of its PC-like features which allow them to use many services like (browsing, money transaction etc.). all the smartphone as the feature to install the third-party application and if not enough storage is available in the internal memory the user can explicitly move all the installed third party application to SD card as the installed application are moved to SD card the user data stored on this SD card and the services that they provide becomes more and more vital. Mobile operating systems naturally become the target for security inspection as they are responsible for managing all the information and services.² studied on the analysis of security weakness in bit-locker technology and puts forward some measures to improve its security.

So, the existing problem in Bit-Locker technology we encrypt SD card and It cannot be decrypted in any other operating system and it takes lot of time for encrypting. Unfortunately, we discovered that Bit-Locker possesses some critical weaknesses. In this paper, we illustrate our discoveries about the vulnerabilities of the scheme. We also suggest a way to take advantage of the ideas introduced with Bit-Locker technology using standard tools in order to provide an efficient and strongly secure encryption scheme, if the Smartphone lost/stolen the content of SD card can easily misuse by any person like SD card consists of personal photos, videos, third party applications data (WhatsApp backup file, Account password text file etc.) for better way to eliminate this problem our paper explain the different way of encrypting the SD card that helps protect your files and folders from unauthorized access in case your SD card is lost or stolen. To Provide better security we introduce a concept called Encrypt once and Decrypt in any Operating system (EODAOS) usually if the user encrypts SD card in Android Operating System, the user can only decrypt in that particular operating system, our paper will explain about encrypt in one operating system and decrypt in any operating system with secure password/passcode authentication.

Literature Review:-

An algorithm of SE (self-encryption)^{1,2} is an efficient encryption for sensitive files requiring the user to enter password or PIN code, the author explains detailed analysis of the weaknesses of the SE scheme.

The algorithm takes better way when using AES encryption³ algorithms based on the high-performance computing of GPU, usually encrypting time of traditional AES algorithm is too long to meet fast encryption. The author come up with better way to encrypt with High performance computing with GPU Using AES Encryption.

A comparison between DES, BLOWFISH and AES, the author^{4,5,11} explains the different Cipher type, Possible keys, Key length and rounds, and time consumption for each algorithm with specific cipher type. Discusses an algorithm the increasing need for Secured data communication⁶ has led to development of several cryptographic algorithms, the author explains secured High throughput implementation of AES algorithm, author explains complete detailed explanation of AES algorithm and comparison between AES Versions, Key Length, Block Size and Number of Rounds.

The key-based security for document encryption⁷ system, utilizing the security key on the key usage of effective administration to guarantee that exclusively approved clients can decode the document and make the proper operation, and utilize the hash calculation for document honesty check, and in this way successfully enhance the security of electronic records, guaranteeing its respectability, proposed a new design to give a safe authentication⁸ such that the Hardware and security measures executed in the system vanquishes any endeavors of unauthorized access to retrieve the information and to keep up extreme control to secure information and prevent theft, by dealing with outline measurements also, proposed discusses Mobile security^{9,10,13} testing targets to detect vulnerabilities and malicious apps on a mobile devices explains various mobile security model like Android security model, IOS security model, Windows Phone security model and briefly explains about Application-based Mobile Threats in all mobile platforms^{12,14,15} explains about image encryption in mobile devices and explains elliptic curve cryptography (ECC) and apply the same for convenient encryption and decryption of an image^{16,17} author explains the securing applications in windows phone, To have an excellent security to the windows phone the developers and Microsoft will utilize the better methodology, By utilizing the sandbox idea, applications won't have the entrance to utilize the information of different applications.

Existing System:-

In order to encrypt the disk, we currently use Bit locker (or) other third party software (Ex: True crypt, File vault, McAfee Drive Encryption (safe boot), private disk etc.). So using Existing Method we are going to Explain How to Securing SD card with Bit Locker Drive encryption.

Bit Locker:-

BitLocker Drive Encryption¹⁹ is built into the Windows operating system and uses Advanced Encryption Standard (AES) with configurable key lengths of either 128 bit (default) or 256 bit (as to configure in Group Policy). The idea behind the BitLocker Drive Encryption¹⁸ is that once you secure your drive, only you, or someone who has your password and recovery key, will be able to get to your data. Explains the attack strategies²⁰ against Bit-Locker which goal the way Bit-Locker is using the TPM sealing device.

Bit Locker Drive encryption encrypts your entire drive.

Bit Locker Performance:-

Enabling Bit Locker drive encryption in PC/Mobile to protect the personal files/folders, but we have to think about the performance. If we configure Bit Locker drive encryption there is a performance degradation in both read and write operation of the disk. Performance depends on the combination of processor, RAM and hard disk type (SSD, HDD).

The Performance was tested the performance of read/write operation using a free tool called AS-SSD-Benchmark version number 1.9 with laptop configuration of 1.50 GHZ AMD A4-5000 APU with Radeon™ HD Graphics with 4 GB RAM and 16 GB SanDisk Ultra USB 3.0 pen drive with Bit Locker Drive Encryption and without Bit Locker Drive Encryption.

Tool Explanation:-

Article²¹ explains the tool as test the sequential or random read/write performance without using the cache. AS SSD Benchmark reads/writes a 1 GByte file as well as randomly select 4K blocks. Moreover, it performs the tests using 1 or 64 threads and it determines the SSD's access time.

Two extra benchmark tests inspect the drive's behavior when (1) copying a few big files, a lot of small files and a combination of file sizes by using cached copy functions of your OS as well as (2) Reading/writing data depending on the data's compressibility.

Read operation:-

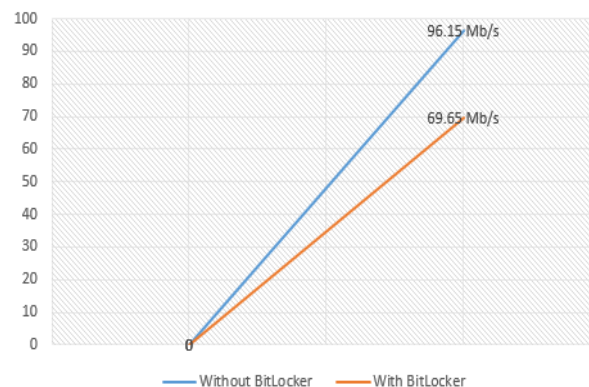
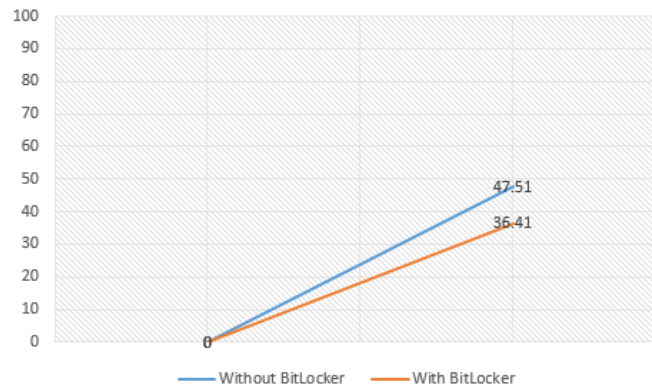


Figure 1:- Read operation.

Read operation takes data out of a specified address in the memory. In Figure¹ explains the performance of varying in Without Bit locker, the reading speed will be more, if we Enable Bit Locker Drive encryption then the speed will be less than 25 %. Taking more time to retrieve the address of specified block in Bit Locker drive encryption because of each block encrypted and taking more time to decryption, for increasing the speed of Bit Locker drive encryption the processing as done both using GPU and CPU to increase the efficiency of an encryption.

Write operation:-**Figure 2:-** Write operation.

In Figure ² explains the performance of varying in Write operation puts data into a specified address in the memory. If you are copying small file, the file copied very fast. But think if you are copying more than 10 – 20 GB in Bit Locker Drive Encryption disk it takes lot of time to get copied.

Proposed System:-

our algorithm and it's working, algorithm follows two fundamental operations. This include a phase of reset operations, increases the speed of encryption process in bit-locker. Since it undergoes two operations (phases) the overall complexity increases and algorithm becomes quite difficult to attack from hackers or attackers. The fixed length of key is generated which must be kept secret. Even through the complexity is very high, but time taken by the algorithm for encryption is less and

Procedure to encrypt the portable device is simple. Thus, algorithm can be used to any portable devices, such as camera SD card, PC, Mobile etc. The following section discusses the procedure to encrypt the SD card.

Algorithm for Encrypting: -

1. User has to Select Key length (64 bits, 128 bits, 256 bits).
Note: if we select lower bits for encryption it will take less time to complete encryption.
2. The key is generated and stored in Flash memory i.e. Root folder of SD card [System Volume Information]
3. The SD card blocks are divided based on the Key Size that user as selected.
4. Temporary master file is created in User selected local disk. By default, master file is secured by AES cipher encryption.
5. All the block address is stored in the master file.
Initial password has been written in master file.
 $M = \text{AES}(BA)$
Where,
M=master File
BA=Block Address
6. After successful store of master file, the shift rows or mix column Sub-programs will execute with all blocks.
7. Encrypt all the blocks with key size if not go to step 6.
8. After encrypting the master file consists of two separate section one is before encrypting block address and after encrypting block address.
9. Now master file, reset program and auto run programs are flashed to SD card.
10. Now SD card is encrypted.



Figure 3:- Encryption Process of SD card.

Algorithm for Decrypting: -

1. User Mounts SD card.
2. When User Opens the SD card Drive the Command prompt will be shown The user has to enter password, If password does not match the reset program will be executed will be formatted everything in SD card, after that the user still unable to save any content in SD card.
3. If password matches the block is divided with Key size.
4. After that the master file will assign the block with before encryption content.
5. Now the SD card is decrypted.

Results:-

Table 5.1:- Comparison between existing method.

	<u>Read</u>	<u>Write</u>	<u>Format</u>	<u>Platform support</u>	<u>Security</u>
Without Bitlocker	YES	YES	YES	All platform	Less
With Bitlocker and Password	YES	YES	YES	windows	More but not portable
With Bitlocker and without Password	NO	NO	YES	windows	Less
With Bitlocker and EODAS without password	NO	NO	NO	Windows, Ubuntu	More with portable
With Bitlocker and EODAS with password	YES	YES	YES	Windows, Ubuntu	More with portable

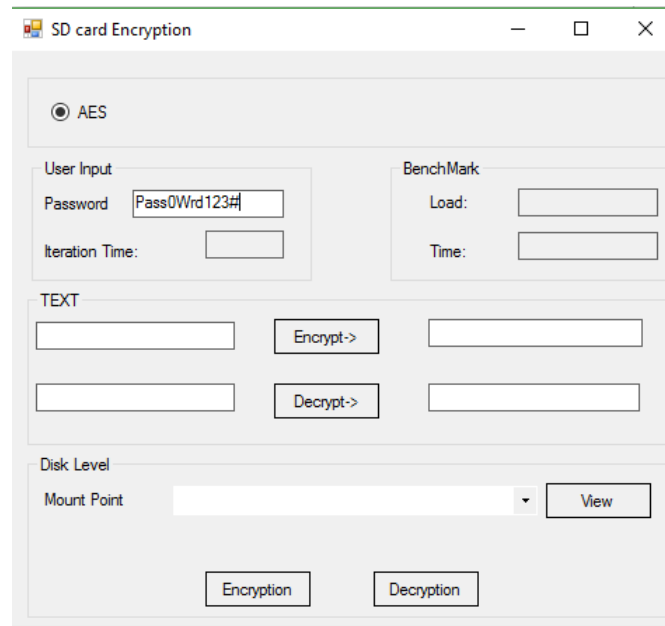


Figure 4:- GUI for Mount Point.

In figure ⁴ the user select Mount Point and Enter password based on password the Key length will be taken, if user has given less time it will take less time to encrypt, It will display how much time it has taken for loading and how much time it takes for encrypting complete disk using this Approach.

Select Disk Location:-

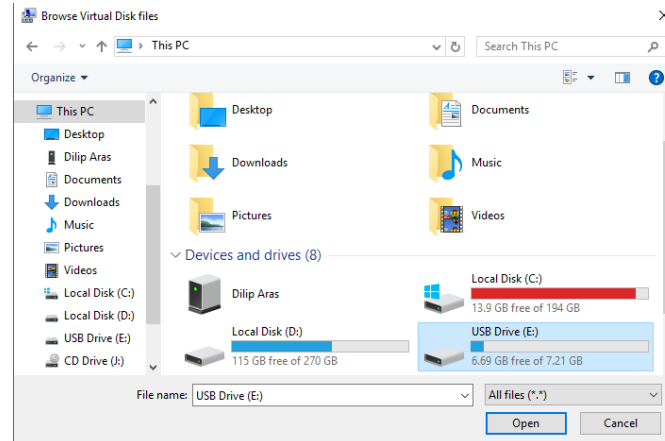


Figure 5:- Select Disk Location.

In figure ⁵ if the user wants to decrypt the disk he will first select disk location in my computer, If user decrypts SD card the SD card is no more secure it just clears the password and encryption information from Autorun.exe.

SD Card Reset Operations:-

When User Opens the SD card drive the Command prompt will show in which user has to enter password, if password does not match the reset program will be executed and will be formatted everything in SD card

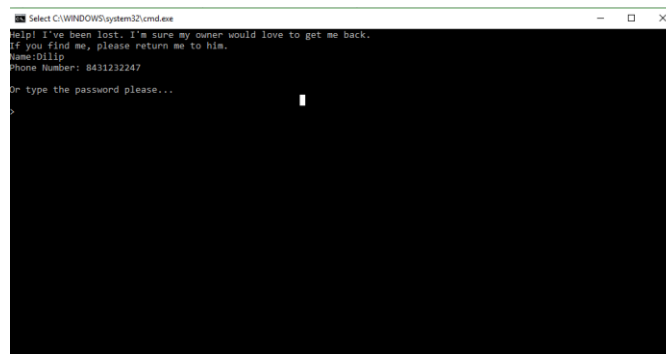


Figure 6:- Enter Password Screen.

If user enters Wrong Password the Disk will be formatted.

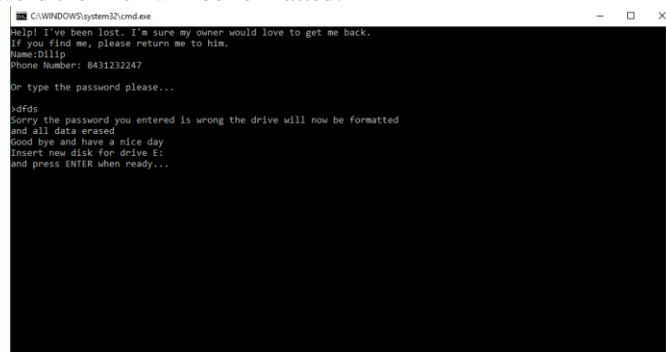


Figure 7:- Displays Error Message Screen.

The SD card data completely formatted and hence unknown user can not get any information.

Conclusions:-

The AS-SSD-Benchmark tool is used to help to determine the speed of the Bit Locker Disk encryption in both read/write operation. We suggest to encrypt the disk using Bit-Locker and new way of resetting the disk, if the password as entered wrong for 5 times the disk will be formatted.so far we tested in two platforms (windows, Ubuntu) if idea succeeded we will try to improve in all platforms.

The encryption is faster and more secure with reset option and disabling the format option under Context Menu which was the disadvantage in Bit locker drive encryption security. Then we proposed ideas in order to construct an encrypt once and decrypt in many devices, we verified the feasibility of our ideas by making a performance analysis which involves experiments on a popular mobile platform and popular desktop operating system.

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REVIEW ARTICLE

A COMPARATIVE STUDY BETWEEN 0.5% CENTBUCRIDINE HCL AND 2% LIGNOCAINE HCL WITH ADRENALINE (1:200,000): SYSTEMATIC REVIEW AND META-ANALYSIS.

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Dental extractions; local anesthesia;
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lidocaine; lignocaine.

Abstract

Purpose: The aim of the present study was to identify significant differences between centbucridine and lignocaine with regard to efficacy of local anesthesia, cardiovascular stability, and side effects of the local anesthetics.

Materials and methods: We performed a systematic and electronic search of several databases using specific keywords, and a manual search through February 2017. The inclusion criteria were clinical human studies, including randomized controlled trials (RCTs), controlled clinical trials (CCTs), and retrospective studies, with the aim of comparing the two drugs in dental/oral surgical practice. Meta-analysis was used to select a fixed or random effects model according to the heterogeneity of the studies. Heterogeneity was assessed using Cochran's Q - test and I^2 . The standard difference in means (SDM) was used as the effect measure for all variables. The results were graphically presented using Forest plot. Funnel plot was used to assess publication bias. The significance level was set at p -value ≤ 0.05 .

Results: Three studies met our inclusion criteria. No statistically significant difference was found between the 2 groups regarding onset of anesthesia ($P = 0.870$), duration of anesthesia ($p=0.327$), depth of anesthesia ($P=0.794$), and pulse rate after 10 minutes ($p=0.087$). A statistically significant difference was found between the two drugs regarding pulse rate after 30 minutes ($p = 0.017$).

Conclusion: The results of the meta-analysis have shown that Centbucridine significantly reduced pulse rate 30 minutes after dental extractions compared with the lignocaine.

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Introduction:-

The development of local anesthetics (LAs) has marked the beginning of new era in the field of dentistry. The use of local anesthetics in dentistry and other surgical procedures for adequate pain control with minimum systemic side effects is one of the major concerns all over the world. Control of pain has been one of the medical portents of twentieth century (Malamed, 2004; Gupta et al., 1989).

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Lignocaine is the 'Gold Standard' local anesthetic agent against which all new local anesthetics are compared. Although its properties resemble an ideal LA agent, it is not completely free from cardiovascular toxicity and has an inherent vasodilating property (Malamed, 2004; Gupta et al., 1982; Patnaik et al., 1982). As a result of the vasodilating characteristic, it has to be combined with a vasoconstrictor, such as adrenaline, to decrease its rate of absorption at the injection site and hence prolong the duration and depth of anesthesia for routine minor dental or oral surgical procedures. The use of adrenaline as a vasoconstrictor is sometimes contraindicated for medically compromised patients. To overcome these disadvantages, other local anesthetics have been developed over the past few years (Dugal et al., 2009; Mansuri et al., 2011 ; Goyal et al., 2013).

Search for a more effective local anesthetic led to the development of centbucridine (Mansuri et al., 2011) which was mainly developed in India (Muthu et al., 2007). Centbucridine, chemically known as 4-N-butylamino-1,2,3,4-tetrahydroacridine hydrochloride, is a new quinoline derivative with LA action. It was synthesized at the central drug research institute, Lucknow, India, by Patnaik, and has the advantage of having an inherent vasoconstrictor property (Patnaik & Dhawan, 1982). It has an inherent vasoconstrictor effect so it does not need adrenaline to be added. Its efficacy and safety has been proven with its use in subdural anaesthesia and ocular anaesthesia but it has not been widely used for dental anaesthesia (Muthu et al., 2007).

Many studies have been performed on centbucridine in the medical field including ophthalmic surgery (Gupta et al., 1985; Ghose et al., 2004), subarachnoid (Dasgupta et al., 1984), and spinal blocks (Samsi et al., 1983). Most of the clinical work on centbucridine has been published in Indian medical journals. Centbucridine does not affect the central nervous or central vascular system except when administered at very large doses (Gupta et al., 1982; Goyal et al., 2013). However, in dentistry, only a few studies have been published (Gupta et al., 1989; Vaccharajani et al., 1983; Dugal et al., 2009) and most of them were carried out in the early 1980s. All of the published literature has shown centbucridine to be a potent and reversible LA. One study reported it to be four to five times more potent than lignocaine (Dasgupta et al., 1984). The aim of this study is to focus on the question: "Is there a significant difference in the clinical outcomes between the two drugs in dental/oral surgical practice?"

Patients and Methods:-

Data sources and key words:-

An electronic search was performed without language and date restrictions in February 2017 in the following databases: Pub Med, Cochrane Database of Systematic Reviews, Cochrane Central Register of Controlled Trials (Central) and manual search

The key words and their combinations used in this search included

In PubMed: (((((((tooth extraction [MeSH Terms]) OR oral surgery [MeSH Terms]) OR oral surgical procedure [MeSH Terms]) OR tooth extraction [Title/ Abstract]) OR dental extractions [Title/Abstract]) OR dental extraction [Title/ Abstract]) OR tooth extractions [Title/Abstract]) OR minor oral surgery [Title /Abstract]) OR oral surgery [Title/Abstract]) OR dental surgery [Title/Abstract])) AND (((((local anesthesia [MeSH Terms]) OR local anesthetics [MeSH Terms]) OR centbucridine [Title/Abstract]) OR centbucridine HCl [Title/Abstract]) OR local anaesthesia [Title/Abstract]) OR local anesthesia [Title/Abstract]) OR local anesthetics [Title/ Abstract])) AND (((lidocaine [MeSH Terms]) OR lidocaine [Title/Abstract]) OR lignocaine [Title/Abstract]).

In Cochrane Database: (Tooth extraction [MeSH Terms]) OR (surgery, oral) [MeSH Terms]) OR (oral surgical procedure [MeSH Terms]) OR "tooth extraction" (Title, Abstract, keywords) OR dental extraction (Title, Abstract, keywords) OR tooth extractions (Title, Abstract, keywords) OR minor oral surgery (Title, Abstract, keywords) OR oral surgery (Title, Abstract, keywords) OR dental surgery (Title, Abstract, keywords) AND Anesthesia, local [MeSH Terms] OR anesthetics, local [MeSH Terms] OR "centbucridine" (Title, Abstract, keywords) OR local anaesthesia (Title, Abstract, keywords) OR local anesthesia (Title, Abstract, keywords) OR local anesthetics (Title, Abstract, keywords) AND lidocaine [MeSH Terms] OR lidocaine (Title, Abstract, keywords) OR lignocaine (Title, Abstract, keywords).

A manual search of oral and maxillofacial surgery related journals including British Journal of Oral and Maxillofacial Surgery, the International Journal of Oral and Maxillofacial Surgery, Journal of Maxillofacial and Oral Surgery, Journal of Craniofacial Surgery, Journal of Oral and Maxillofacial Surgery, Oral Surgery, Oral Medicine, Oral Pathology, Oral Radiology and Journal of CranioMaxillofacial Surgery was performed. Relevant reviews on the subject and the reference lists of the studies identified were scanned for possible additional studies.

Inclusion and exclusion criteria:-

Inclusion criteria were studies in humans including randomized controlled trials (RCTs), controlled clinical trials (CCTs), prospective studies (RS), and retrospective studies (RS). The aim of the studies had to be a comparison between centbucridine and lignocaine in physically fit (ASA Class I) adult patients of either sex scheduled for extraction of teeth using nerve blocks, with the outcome variables being efficacy of local anesthesia, cardiovascular stability and side effects of the anesthetics.

Exclusion criteria were: highly anxious patients, patients with a history of allergies to local anesthetic agents and known cardiovascular problems, cases with acute infection in the orofacial area, technical reports, case reports, in vitro studies, animal studies, and review papers.

Selection of relevant studies:-

The following data were extracted from the studies included in the final analysis: authors, year of publication, study design, number of participants, patient age range and/ or mean age, sex, site of extraction, types of local anesthesia, intradermal sensitivity test, informed written consent, efficacy of local anesthesia including onset of anesthesia, duration of anesthesia, the depth of anesthesia, cardiovascular stability (blood pressure and pulse rate), side effects to the anesthetic (if any).

Assessment of quality:-

A methodological quality analysis was performed by merging the proposed criteria of the Strobe statement (Von Elm et al., 2007), Moose statement (Stroup et al., 2000), and Prisma statement (Moher et al., 2009), to verify the force of scientific evidence in making clinical decisions. The classification of the risk of potential bias for every article was based on the following criteria: random selection in the participants, definition of inclusion and exclusion criteria, report of losses to follow up (attrition bias), validity of assessments, and statistical analysis. A study that comprised all the criteria mentioned above was categorized as having a low risk of bias, a study that did not comprise one of these criteria was categorized as having a moderate risk of bias. If two or more criteria were missed, the study was classified to have a high risk of bias.

Meta-Analysis:-

Meta-analysis of the present study was performed using Comprehensive Meta-analysis version 2.2.048 software (Biostat, Inc., Englewood, NJ, USA). The first step in meta-analysis is to select the model to be used whether fixed or random effects model according to heterogeneity of the studies. A variety of heterogeneity measures were provided to decide on the fixed or random effects approach. Cochran's Q provides a P-value for the test of homogeneity. I^2 was deemed to be more reliable in assessing inconsistency between studies, with values of 25%, 50% and 75% corresponding to low, moderate and high heterogeneity respectively (Higgins et al., 2003).

Whenever heterogeneity was identified with Cochran's Q - test and/or I^2 , a random effects model was preferred over the fixed effects. Heterogeneity was identified by rejecting the homogeneity hypothesis. Meta-analysis done for prevalence of post-operative sensitivity used the Odds Ratio (OR) as the effect measures while meta-analysis for pain scores (VAS) used the standard difference in means (SDM) as the effect measure.

The results were graphically presented using forest plot. In forest plot, each study is represented by a line. There is a box (or circle) in the line for each study. The mid-point of the box (or circle) represents the point effect estimate, that is, the mean effect estimate for each study. The area of the box (or circle) represents the weight given to the study. The diamond below the studies represents the overall effect. The width of the line showed the confidence interval (95% CI) of the effect estimate of individual studies. The

width of the diamond shows the confidence interval (95% CI) of the overall effect estimate. The vertical line in the middle of the Forest plot (Corresponding to the value 0) is the line of no effect. If the confidence interval overlaps this line (Overlaps 0 value), then there is no statistical significance at 0.05 significance levels. If 0 value is not included in the 95% CI, the results are statistically significant at 0.05 significance level. This is applicable for the effect estimates for the individual studies and the overall estimate.

Funnel plot was used to assess publication bias. The plot by precision is the traditional form. Large studies appear toward the top of the graph, and tend to cluster near the mean effect size. Smaller studies appear toward the bottom of the graph, and (since there is more random variation in the small studies) are dispersed across a range of values. This pattern tends to resemble a funnel, which is the basis for the plot's name. In the absence of publication bias the studies will be distributed symmetrically about the combined effect size. By contrast, in the presence of bias, the bottom of the plot would tend to show a higher concentration of studies on one side of the mean than the other.

Egger's test of the intercept was used to quantify the display of the funnel plot. This approach may offer a number of advantages over other approaches. Under some circumstances this may be a more powerful test. Additionally, this approach can be extended to include more than one predictor variable, which means that we can simultaneously assess the impact of several factors, including sample size, on the treatment effect. The significance level was set at $p\text{-value} \leq 0.05$.

Results:-

Summary of the study selection process is shown in Fig. 1. The electronic search resulted in 575 studies; three additional articles were added from hand-searching and other sources. After the initial screening of articles, 123 articles were excluded because of duplication. Of the remaining 455 articles assessed, 380 were excluded by title and abstract because they were not related to the topic. Seventy five studies were selected for full text analysis leading to the exclusion of 72 articles because they did not meet the inclusion and exclusion criteria. Thus, a total of 3 articles were included in this systematic review and meta-analysis.

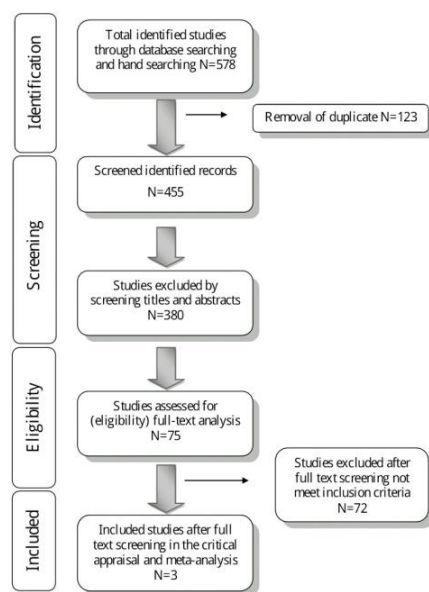


Fig.1:- Flow diagram of study selection process

Description of included studies:-

Extracted data of the included 3 studies are listed in Table 1. Three prospective studies were included in this study (Dugal et al., 2009; Mansuri et al., 2011; Goyal et al., 2013). A total of 760 patients were

enrolled in the three studies. 390 patients in interventions group (0.5% Centbucridine) and 370 patients in control group (Lignocaine HCl with Adrenaline 1:200,000). The ages ranged from 16-60 years. Intradermal sensitivity test was performed for all patients and informed written consent was obtained from all patients.

In first study the patient received 3 ml of anesthetic for nerve blocks using either 0.5% Centbucridine HCl or 2% Lignocaine HCl with adrenaline (1:200,000) (Dugal et al., 2009). In the second study patients randomly received a single anesthetic dose of either 0.5% Centbucridine HCl or 2% Lignocaine HCl with adrenaline (1:200,000) (Mansuri et al., 2011). In last study both drugs were supplied in equal amounts in identical vials labeled only with a code number (Goyal et al., 2013).

Table 1:- Studies comparing 0.5% Centbucridine HCl and 2% Lignocaine HCl with Adrenaline (1:2,00,000).

Authors, Publication year	Study design	P(n)	Patient age range (mean), years	Sex	Site of extraction	Type of local anesthesia	Groups types	Dose of local anesthesia
Dugal et al., 2009	PS	240	Adult either sex	Either sex	Extractions (surgical/ closed method) of teeth requiring nerve block (upper anterior or lower molar teeth)	(G1) 0.5% Centbucridine (n=120) (G2) 2% Lignocaine + Adrenaline (n = 120)	Intervention Control	3 ml of anesthetic was injected for nerve blocks
Mansuri et al., 2011	RCT	198	18 -60 (mean 37.7)	94M/ 104F	extraction of lower molars	(G1) 0.5% Centbucridine (n=100) (G2) 2% Lignocaine + Adrenaline (n = 98)	Intervention Control	They each randomly received a single anesthetic dose of either 0.5% Centbucridine HCl or 2% Lignocaine HCl with adrenaline
Goyal et al., 2013	RCT	322	18 -60 (mean 36.7)	182M /140F	Extraction of lower molars	(G1) 0.5% Centbucridine (n=170) (G2) 2% Lignocaine + Adrenaline (n = 152)	Intervention Control	Both drugs were supplied in equal amounts in identical vials labeled only with a code number

PS , Prospective study; RCT, randomized controlled trial; P, participants.

Assessment of quality:-

The risk of bias outcomes is summarized in Table 2. One study was considered to have high risk of bias (Dugal et al., 2009), one was considered to have moderate risk of bias (Goyal et al., 2013) and one was considered to have low risk of bias (Mansuri et al., 2011).

Table 2:- Results of the quality assessment.

Authors and year of Publication	Random selection of participants	Definition inclusion/exclusion criteria	Attrition bias (Loss of follow-up)	Validity of assessment	Statistical analysis	Reported potential risk of bias
Dugal et al., 2009	No	Yes	No	Yes	yes	high
Mansuri et al., 2011	yes	yes	yes	yes	yes	low
Goyal et al., 2013	yes	No	yes	yes	yes	Moderate

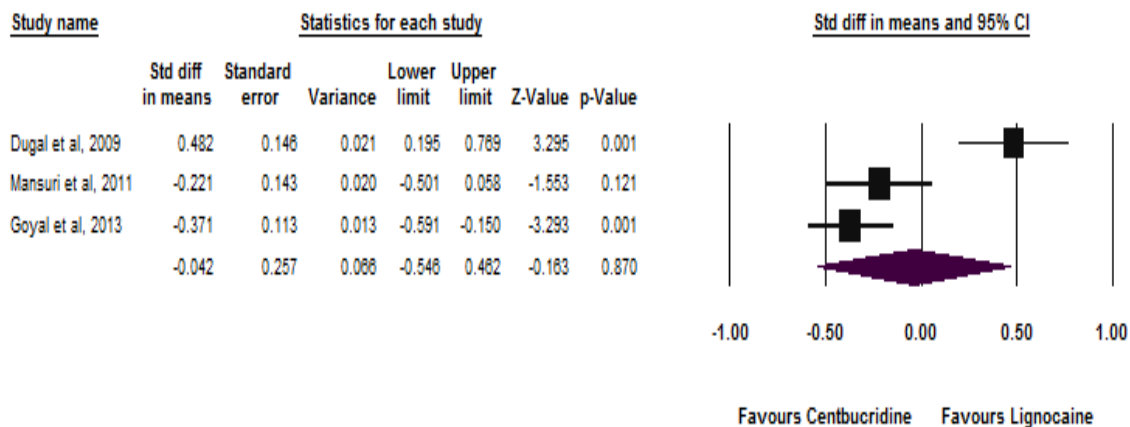
Effect of intervention:-**Onset of anesthesia:-**

Heterogeneity measures showed statistically significant Cochrane Q value (p -value <0.001). I^2 value was 91.0 % indicating high heterogeneity. So the homogeneity hypothesis was rejected and the random effects model was used.

The random effects model showed a standardized mean difference (effect size) of -0.042 with a 95% CI (-0.546 – 0.462). The effect size was not statistically significant with p -value = 0.870 Fig. 2. Thus, there was no statistically significant difference between onset of anesthesia in the two groups.

The relative weight of the included studies revealed that the Goyal et al., 2013 study had the highest weight (34.3%) while Mansuri et al., 2011 study showed the lowest weight (33.0%).

Funnel plot analysis for the included studies showed no publication bias Fig. 3. This was confirmed by Egger's regression intercept which showed a non-statistically significant result (p -value = 0.464). As the results were not statistically significant, we concluded that there was no publication bias.

**Fig. 2:-** Forest plot for onset of anesthesia.

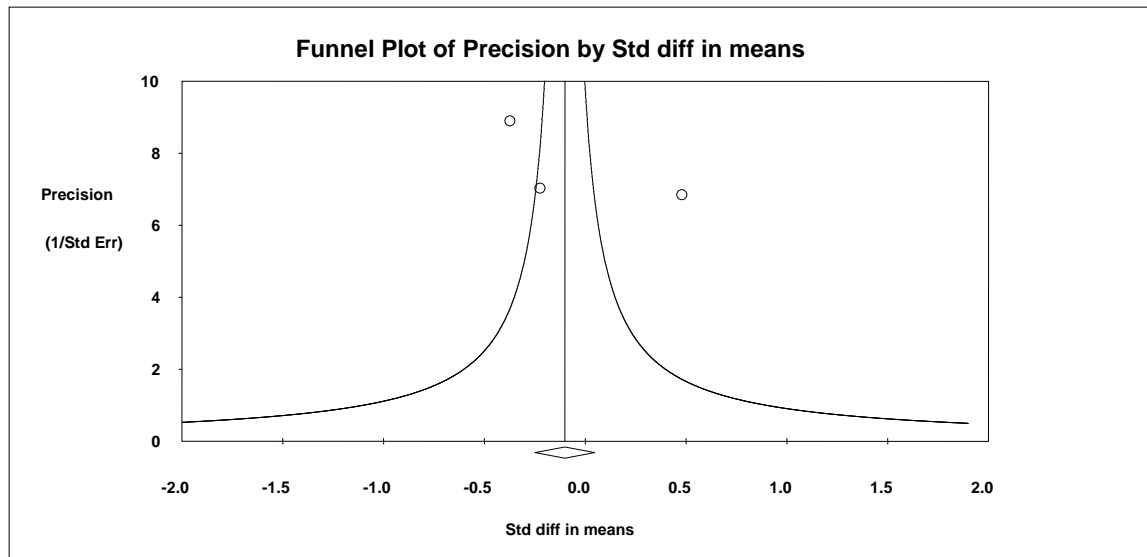


Fig . 3:- Funnel plot for onset of anesthesia.

Duration of Anesthesia:-

Heterogeneity measures showed statistically significant Cochrane Q value (P -value = 0.001). I^2 value was 86.6% indicating high heterogeneity. So the homogeneity hypothesis was rejected and the random effects model was used.

The random effects model showed a standardized mean difference (effect size) of 0.206 with a 95% CI (-0.206 – 0.618). The effect size was not statistically significant with p -value = 0.327 Fig. 4. Thus, there was no statistically significant difference between duration of anesthesia in the two groups.

The relative weight of the included studies revealed that Goyal et al., 2013 study had the highest weight (34.7%) while Dugal et al., 2009 study showed the lowest weight (32.6%).

Funnel plot analysis for the included studies showed no publication bias Fig. 5. This was confirmed by Egger's regression intercept which showed a non-statistically significant result (p -value = 0.709). As the results were not statistically significant, we concluded that there is no publication bias.

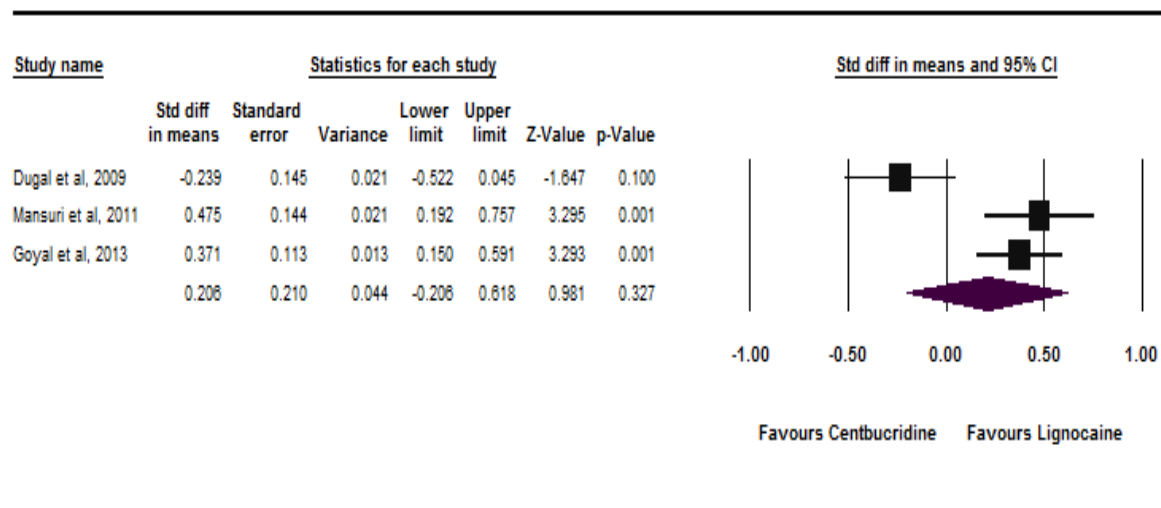


Fig. 4:- Forest plot for duration of anesthesia.

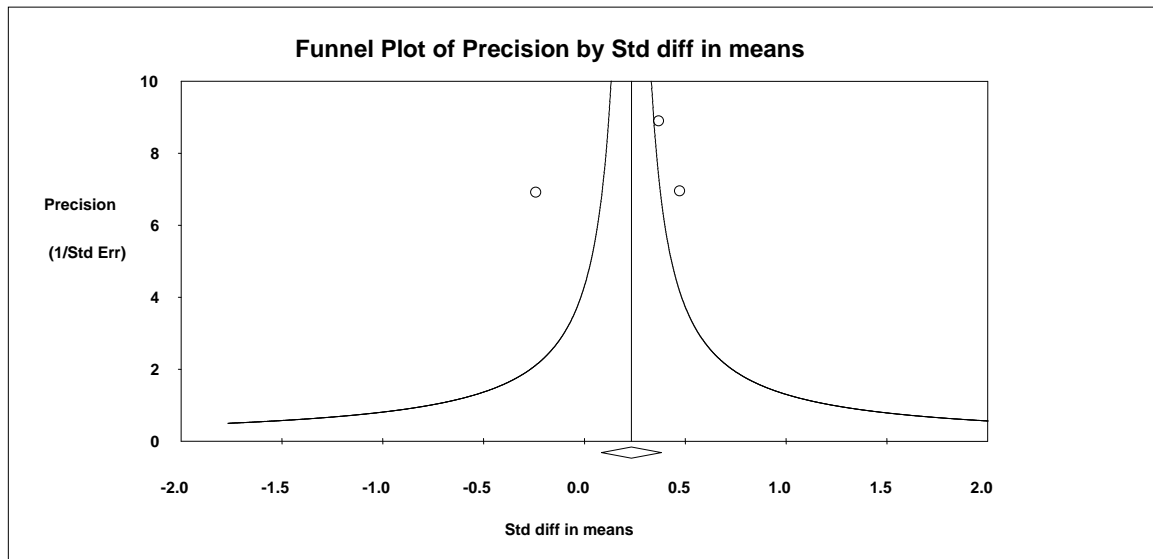


Fig. 5:- Funnel plot for duration of anesthesia.

Depth of anesthesia (VAS score):-

For the ease of calculation, the mean VAS score was computed according to the data presented in the study. However, the Dugal et al., 2009 study was not included in the meta-analysis because the standard deviation in Lignocaine group was zero (all cases had zero score on VAS).

Heterogeneity measures showed a non-statistically significant Cochrane Q value (p -value = 0.967). I^2 value was 0.0% indicating no heterogeneity. Thus, the homogeneity hypothesis was not rejected and the fixed effects model was used.

The fixed effects model showed a standardized mean difference (effect size) of 0.023 with a 95% CI (-0.149 – 0.195). The effect size was not statistically significant with p -value = 0.794 Fig. 6. Thus, there was no statistically significant difference between depth of anesthesia in the two groups.

The relative weight of the included studies revealed that the Goyal et al., 2013 study had the highest weight (61.8%) while the Mansuri et al., 2011 study showed the lowest weight (38.2%).

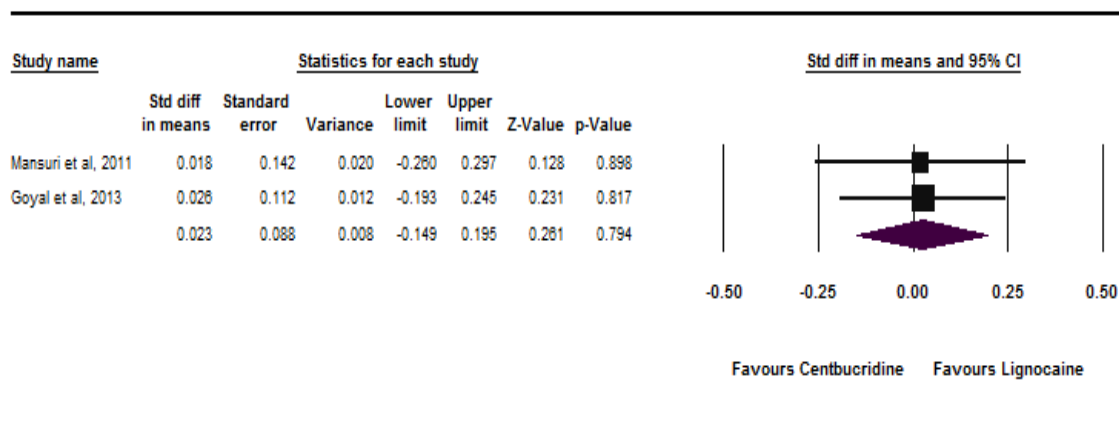


Fig. 6:- Forest plot for depth of anesthesia.

Pulse Rate:-**After 10 minutes:-**

Heterogeneity measures showed non-statistically significant Cochran Q value (P -value = 0.079). I^2 value was 60.6% indicating moderate heterogeneity. Thus, that the homogeneity hypothesis was not rejected and the fixed effects model was used.

The fixed effects model showed a standardized mean difference (effect size) of 0.129 with a 95% CI (-0.019 – 0.276). The effect size was not statistically significant with p -value = 0.087 Fig. 7. Thus there was no statistically significant difference between pulse rate after 10 minutes in the two groups.

The relative weight of the included studies revealed that the Goyal et al., 2013 study had the highest weight (45.4%) while the Dugal et al., 2009 study showed the lowest weight (26.6%).

Funnel plot analysis for the included studies showed no publication bias Fig. 8. This was confirmed by Egger's regression intercept which showed a non-statistically significant result (p -value = 0.473). As the results were not statistically significant, we concluded that there was no publication bias.

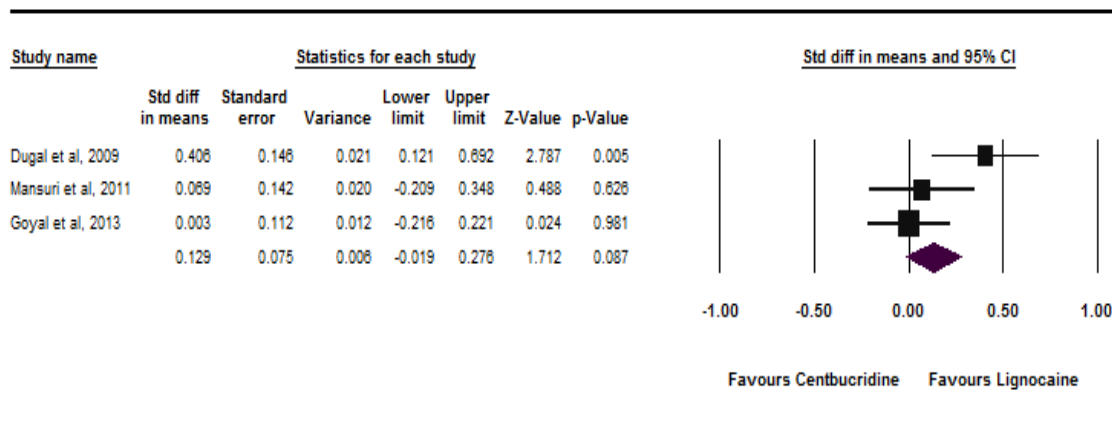


Fig.7:- Forest plot for pulse rate after 10 minutes.

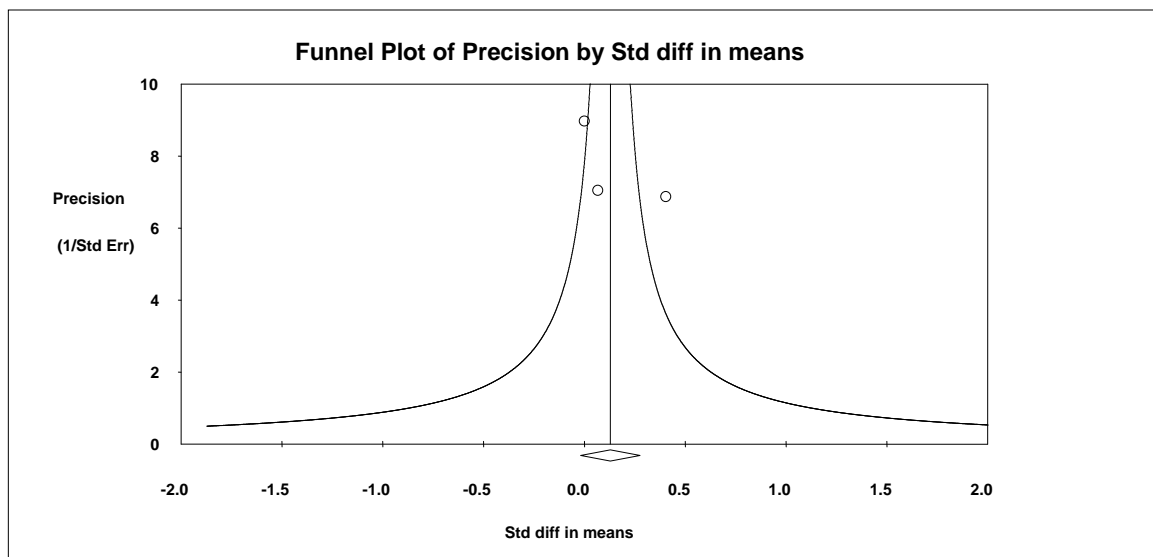


Fig. 8:- Funnel plot for pulse rate after 10 minutes.

After 20 Minutes:-

No meta-analysis could be performed because the Dugal et al., 2009 study didn't report the pulse rate after 20 minutes and the Mansuri et al., 2011 study reported that in Lignocaine group the pulse rate after 20 minutes was the same as pre-operative rate so no p -value was computed for the comparison. This left only the Goyal et al., 2013 study so no meta-analysis could be done with just one study.

After 30 Minutes:-

Heterogeneity measures showed non-statistically significant Cochran Q value (p -value = 0.810). I^2 value was 0.0% indicating no heterogeneity. Thus, the homogeneity hypothesis was not rejected and the fixed effects model was used.

The fixed effects model showed a standardized mean difference (effect size) of 0.212 with a 95% CI (0.038 – 0.385). The effect size was statistically significant with p -value = 0.017 Fig. 9. Thus, there was a statistically significant increase in pulse rate after 30 minutes and the direction of effect is in favor of Lignocaine indicating that Lignocaine causes higher increase in pulse rate after 30 minutes compared to Centbucridine.

The relative weight of the included studies revealed that the Goyal et al., 2013 study had the highest weight (62.6%) while the Dugal et al., 2009 study showed the lowest weight (37.4%).

Publication bias could not be assessed because there were only two studies.

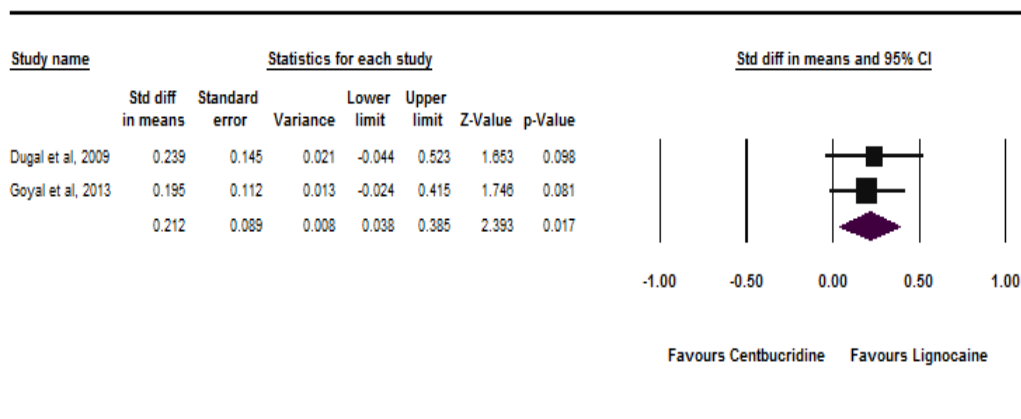


Fig. 9:- Forest plot for pulse rate after 30 minutes

After 60 minutes:-

No meta-analysis could be performed because the Dugal et al., 2009 study didn't report the pulse rate after 60 minutes and the Mansuri et al., 2011 study reported that in both groups the pulse rate after 60 minutes was the same as pre-operative rate so no p -values were computed for the comparison. This left only the Goyal et al., 2013 study and no meta-analysis could be done with just one study.

No meta-analysis could be performed for blood pressure because only one study reported it.

Regarding side effects, no meta-analysis could be performed because no study reported it.

Discussion:-

Although Centbucridine is relatively new in dentistry, it has demonstrated good clinical results in the literature (Vacharajani et al., 1983; Muthu et al., 2007). The aim of this review was to verify through a meta-analysis whether there is a significant difference in the clinical outcomes between centbucridine and lignocaine plus adrenaline. In general, the results of the present meta-analysis show statistically good results for centbucridine as a local anesthesia in dental extraction.

Regarding onset of local anesthesia, when the onset of both anesthetics is compared, there was not much difference. This is in accordance with the reports by (Gupta et al., 1982; Muthu et al., 2007). This could be due to the inherent vasoconstrictive effect of Centbucridine as compared to Lignocaine. On average, patients felt the anesthetic effect of Centbucridine a few seconds quicker than that of Lignocaine but this difference was not statistically significant and more importantly, is not clinically significant. The onset of action of Centbucridine was within the reported range of initiation of anesthesia as reported by others to be between 1 and 6 minutes (Vacharajani et al., 1983; Gupta et al., 1989).

Regarding duration of local anesthesia, our study found the duration of anesthetic action of Centbucridine compared favorably to lignocaine with adrenaline. There was no statistically significant difference between them $p=0.327$. This is similar to what has been reported previously (Gupta et al., 1982; Muthu et al., 2007). This duration is sufficient for both surgical and nonsurgical extractions. A possible reason could be the fact that since Centbucridine has a natural vasoconstrictive effect; the LA solution remained close to and around the nerve tissue for a longer period of time. The solution was prevented from being absorbed and dispersed, and this could have resulted in the sufficient duration of anesthetic time that was obtained (Gupta et al., 1989).

Regarding depth of local anesthesia, there was no statistically significant difference between the two groups ($p=0.794$). All patients were sufficiently anesthetized to carry out the procedures. Centbucridine showed sufficient efficacy as a local anesthetic agent. A possible reason could be the fact that since Centbucridine has a natural vasoconstrictive effect; the LA solution remains close to and around the nerve tissue for a longer period of time (Gupta et al., 1989).

Regarding pulse rate, there was no statistically significant difference between the two groups after 10 minutes ($p\text{-value} = 0.087$). Mild elevation of this parameter during this initial time was attributed to anxiety and fear. This has also been reported by other authors (Malamed, 2004; Gupta et al., 1989; Vacharajani et al., 1983). However, there was a statistically significant increase in pulse rate after 30 minutes ($p\text{-value} = 0.017$) and the direction of effect is in favor of Lignocaine indicating that with Lignocaine caused a higher increase in pulse rate after 30 minutes compared to Centbucridine. Lignocaine has an inherent vasodilating property, which in turn requires the addition of adrenaline. This has been shown to increase the blood pressure and heart rate in some studies (Malamed, 2004; Gupta et al., 1989); which may become more significant in cardiac cases (Dugal et al., 2009).

Regarding blood pressure, no meta-analysis could be done for blood pressure because only one study reported it. Three studies declared lignocaine has an inherent vasodilating property, which in turn requires the addition of adrenaline. This has been shown to increase the blood pressure and heart rate in some studies (Malamed, 2004; Gupta et al., 1989).

Regarding side effects, no meta-analysis could be done for side effects because no study reported it. One study reported side effects of Centbucridine like headache, dizziness and nausea, which gradually resolved within 10 minutes without any medication (Dugal et al., 2009). Other studies showed that there were no adverse, toxic, or allergic reactions to either of the LAs in their sample population and confirmed its safety (Mansuri et al., 2011; Goyal et al., 2013). It is not surprising that there were no patients who reported adverse reactions to Centbucridine. Centbucridine has demonstrated an antihistaminic activity by blocking the H1 histamine receptors which makes it an ideal LA agent in patients with known allergy to other conventional LAs (Gupta et al., 1985; Gupta et al., 1989; Mansuri et al., 2011; Goyal et al., 2013).

In conclusion, the overall results of meta-analysis showed no statistically significant difference between the 2 groups regarding onset of anesthesia ($p = 0.870$), duration of anesthesia ($p=0.327$), depth of anesthesia ($p=0.794$), and pulse rate after 10 minutes ($p=0.087$). But there was a statistically significant difference between the two drugs with regard to pulse rate after 30 minutes ($p = 0.017$) with lignocaine causing higher increases in pulse rate compared to Centbucridine. Hence Centbucridine had good CVS stability and was devoid of any cardiovascular effects. Centbucridine can be effectively used as an ideal substitute in place of

lignocaine in patients undergoing minor oral surgical procedures when adrenaline is absolutely contraindicated due to systemic problems.

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Competing interests:-

None declared.

Ethical approval:-

Not required.

Patient consent:-

Not required.

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RESEARCH ARTICLE

EFFECT OF GERMINATION AND FERMENTATION ON SPECIFIC NUTRIENTS IN MILLET KOOZH PREPARED USING SORGHUM BICOLOUR (*MONECH.L*), PEARL MILLET (*PENNISETUM GLAUCUM*) AND FINGER MILLET (*ELEUSINE CORACANA*).

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Key words:-

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Abstract

Koozh is a product of double fermentation of millet and rice caused by a mixture of micro flora involving lactic acid bacteria and yeast. The present study was undertaken to determine the effect of germination and fermentation on macro and certain micro nutrient contents in koozh prepared using finger millet, Pearl millet, Red sorghum and white sorghum. It was inferred from the study that fermentation of millet koozh enhanced the protein levels with a substantial reduction in carbohydrate and energy content. Germination and fermentation resulted in a concomitant raise in thiamine levels along with iron and vitamin C. A sensory evaluation of both germinated & fermented and non-germinated & fermented millet koozh revealed equal acceptance with no significant difference in their hedonic scale ratings. There was an increase in the yeast content of fermented koozh which explained the increase in B vitamin content. Thus this study concludes that millets can be used in different proportions to formulate various fermented nutritious food products for all age groups.

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Introduction:-

Millets are one of the oldest foods known to humans and possibly the first cereal grain to be used for domestic purposes^[1]. Millets are not only comparable to major cereals with respect to their nutritional features but are very good sources of carbohydrates, micronutrients and phytochemicals with nutraceutical properties. Millets contain 7-12 per cent protein, 2-5 per cent fat, 65-75 per cent carbohydrates and 15-20 per cent dietary fiber. These millets are a good source of both soluble and insoluble dietary fiber^[2]. A variety of foods from the millets are traditionally prepared and consumed, over the centuries, in the Indian subcontinent, Africa and Central America.

Several traditional household food processing and preparation methods are used to enhance the bioavailability of micronutrients from millets. Koozh is one such preparation that has strong connection with South Indian culture and tradition and has been consumed regularly for breakfast before rice became a common consumable. Koozh is a full millet based preparation along with rice. Millets like finger millet, pearl millet and sorghum are used singly or in combination for Koozh preparation. It is a soft porridge prepared from traditionally processed millet (germinated and fermented).

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Preparation of Koozh involves two fermentation processes, before (primary fermentation) and after cooking (secondary fermentation). During fermentation, the millet slurry would undergo major biochemical changes like starch hydrolysis, sugar transformation and softening. So the conventional process of fermentation is believed to increase the nutritive value of millets. The processing methodology is similar throughout south India with slight modifications in fortifying substances and raw materials. Unlike other cereal and millet based fermented foods in which fermenting microorganisms are killed by thermal treatment, secondary fermentation step in Koozh offers live microbes along with the product. This feature adds functional value to the traditional product and makes it to stand on par with probiotic dairy foods.^[3]

Aim of the study:-

To assess the influence of germination and fermentation on the levels of specific nutrients in millet koozh prepared using finger millet, pearl millet, Red sorghum & white sorghum and broken raw rice

Objectives of the study

To estimate the energy, carbohydrate, protein and fat, ash, fibre, moisture, vitamin C and iron content of millet koozh prepared in the following combinations of processing methods.

1. Non-Germinated and Non-fermented millet Koozh
2. Non- Germinated and fermented millet koozh.
3. Germinated and fermented millet koozh.
4. Germinated and Non-fermented millet Koozh

To assess the levels of Thiamine, Riboflavin and Niacin in

1. Non-germinated & fermented millet koozh and
2. Germinated & fermented millet koozh

To conduct sensory evaluation of

1. Non-germinated & fermented millet koozh and
2. Germinated & fermented millet koozh.

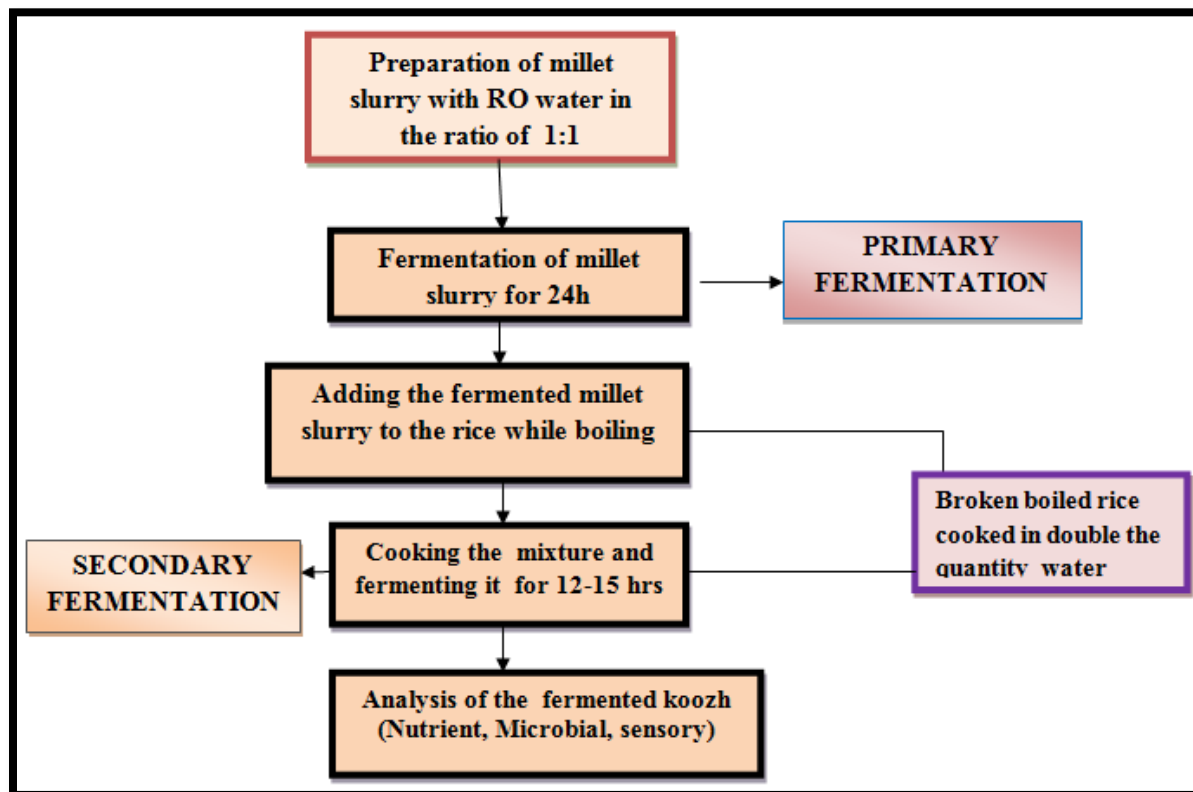
Methodology:-**Research design:-**

The research design of this study is experimental in nature.

Sample selection:-

As the study comprises of germination of millets, those millets which germinated well under optimum conditions were chosen after experimenting on all the major and minor millets available in the market. Red and white varieties of sorghum (*L.Monech*), pearl (*Pennisetum glaucum*) and Finger millet (*Eleusine coracana*) were selected for the study. The millets were obtained from the millet retailer supplying at the Tamil Nadu Agricultural University Centre at Chennai. Ponni raw rice was procured from the grocery.

Flow chart showing the preparation of millet koozh using Finger millet (*Eleusinecoracana*), pearl millet (*Pennisetumglaucum*) and Bi color sorghum (*L.Monech*)



Millet Koozh was prepared in four different combinations of processing methods as shown in table 1. The steps included and omitted in the preparation of millet koozh using the four processing methods are spelt out. This was done to find out the actual effect of germination and fermentation on the nutrient quality of the millet koozh.

Table 1:- Different combinations of Processing Methods adopted in the preparation of millet koozh

Processing methods	Steps adopted and omitted in the preparation of millet koozh
Non-germinated and Non-fermented millet koozh	No germination of millets and fermentation of millet koozh
Non- Germinated and fermented millet koozh	No germination of millets but only fermentation of millet koozh
Germinated and Fermented millet koozh	Germination and fermentation adopted in the preparation of millet koozh
Germinated and Non-fermented millet koozh	Only germination of millets but no fermentation of millet koozh

Results and Discussions:-

Comparison of the proximate principles (carbohydrate, protein and fat) and energy content of millet Koozh

A comparison of the proximate principles (carbohydrate, protein and fat) and energy content of millet Koozh prepared using the stated combinations of processing methods is presented in table 2.

Table 2:- Comparison of proximate composition of Millet Koozh prepared using varying combinations of processing methods.

Processing methods	ENERGY (kcal)			CARBOHYDRATE (g/100g)			PROTEIN (g/100g)			FAT (g/100g)		
	Mean \pm SD	t value	Level of sig	Mean \pm SD	t value	Level of sig	Mean \pm SD	t value	Level of sig	Mean \pm SD	t value	Level of sig
Non-germinated & Non-fermented Millet Koozh	215.94 \pm 0.616	125.09	0.000**	46.79 \pm 0.316	122.82	0.000*	4.03 \pm 0.015	46.80	0.000**	1.41 \pm 0.18	0.849	0.444 ^N _s
Non-germinated & fermented Millet Koozh	138.73 \pm 0.874			24.09 \pm 0.049			7.34 \pm 0.122			1.32 \pm 0.02		
Non-germinated & Non-fermented Millet Koozh	215.94 \pm 0.62	95.56	0.000*	46.79 \pm 0.32	62.03	0.000*	4.03 \pm 0.02	73.89	0.000**	1.41 \pm 0.18	0.652	0.550 ^N _s
Germinated & Non-fermented Millet Koozh	276.33 \pm 0.90			60.7 \pm 0.23			5.33 \pm 0.03			1.34 \pm 0.04		
Non-germinated & non-fermented Millet Koozh	215.9433 \pm 0.616	121.60	0.000*	46.786 \pm 0.316	81.51	0.000*	4.02 \pm 0.015	116.29	0.000**	1.41 \pm 0.18	4.363	0.012
Germinated & Fermented Millet Koozh	161.4333 \pm 0.473			31.090 \pm 0.106			7.27 \pm 0.046			0.94 \pm 0.02		
Germinated & non-fermented Millet Koozh	276.326 \pm 0.905	194.99	0.000*	60.720 \pm 0.227	205.28	0.000*	5.33 \pm 0.026	63.50	0.000*	1.34 \pm 0.03	15.911	0.000*
Germinated & Fermented Millet Koozh	161.433 \pm 0.473			31.090 \pm 0.106			7.27 \pm 0.046			0.94 \pm 0.02		
Non-germinated & Fermented Millet Koozh	138.73 \pm 0.874	39.58	0.000*	24.09 \pm 0.049	103.89	0.000*	7.34 \pm 0.122	0.93	0.404NS	1.32 \pm 0.02	21.16	0.000*
Germinated & Fermented Millet Koozh	161.43 \pm 0.473			31.09 \pm 0.106			7.27 \pm 0.046			0.95 \pm 0.02		

NS- Not significant * - statistically significant

From table 2, it is evident that fermentation of the millet koozh resulted in a substantial reduction in the energy and carbohydrate levels. The fermented samples had the lowest energy value which is due to the low fat and carbohydrate content. The reduction in carbohydrate content in fermented koozh could also be attributed to increased activity of alpha-amylase which hydrolyses starch to simple sugar. The sugar provides a source of energy for the fermenting micro-organisms.^[4] In the present study, fermented koozh was found to exhibit significant ($p < 0.01$) reduction in carbohydrate content. This observation confirms the utilization of the carbohydrate by the fermenting microbes. Carbohydrates are a major energy source for fermenting microbes.

The protein levels also have shown a considerable rise in the fermented millet koozh which reflects on the activation of the proteolytic enzymes during fermentation. Antony and Chandra (1998)^[5], concluded in their study that as fermentation is associated with the degradation of antinutrients, there is an increase in the bioavailability of minerals and an improvement in the digestibility of proteins in tannin-rich cereals.

There was a decrease in fat content after fermentation and a further statistically significant decrease in germinated and fermented millet koozh which may be attributed to the breakdown of fats to fatty acid and glycerol by lipolytic organisms present in the sample during fermentation and the utilization of fat for energy purposes by the fermenting microbes. A similar observation has been reported by Anthony and Babatunde (2014)^[6] where the break down resulted in an increase in aroma, taste, odour and texture of fermented food.

Comparison of Ash and Fiber content of Millet Koozh:-

A comparison of the Ash, fiber and moisture content of millet Koozh prepared in different combinations of processing methods is presented in table 3.

From table 3, it can be seen that the ash content of the millet koozh prepared using different processing methods ranged between 1.03 to 1.4 g/ 100g. The present study elucidates that in all the preparative methods of koozh, even though there was a marginal decrease in the overall ash content of the koozh on germination and fermentation, there were no significant changes. Arora, Jood and Khetarpaul (2011)^[7] found that germination of grains decreased ash content. In the present study, the least ash content was found in germinated and fermented millet koozh.

The fermented koozh samples showed a significant appreciation in the fiber content compared to the non-fermented samples. A similar trend was also observed in the germinated and fermented koozh. It is interesting to note that fermentation increased the crude fiber content.

Table 3:- Comparison of Ash and Fiber content of Millet Koozh prepared using varying combinations of processing methods.

Processing methods	ASH (g)			FIBER (g)		
	Mean ± SD	t value	Level of sig	Mean ±SD	t value	Level of sig
Non-germinated & Non-fermented Millet Koozh	1.13 ±0.058	2.079	0.106 ^{NS}	0.60 ±0.100	51.594	0.000*
Non-germinated & fermented Millet Koozh	1.06 ±0.020			3.61±0.015		
Non-germinated & Non-fermented Millet Koozh	1.13±0.06	2.219	0.091 ^{NS}	0.60±0.10	1.549	0.196 ^{NS}
Germinated & Non-fermented Millet Koozh	1.40±0.20			0.40±0.20		
Non-germinated & non-fermented Millet Koozh	1.133±0.058	2.929	0.043 ^{NS}	0.600±0.100	49.026	0.000*
Germinated & Fermented Millet Koozh	1.030±0.020			3.463±0.015		
Germinated & non-fermented Millet Koozh	1.400±0.200	3.188	0.033*	0.400±0.200	26.452	0.000*
Germinated & Fermented Millet Koozh	1.030±0.020			3.463±0.015		
Non-germinated & Fermented Millet Koozh	1.06±0.020	1.83	0.140 ^{NS}	3.61±0.015	12.02	0.000*
Germinated & Fermented Millet Koozh	1.03±0.020			3.46±0.015		

Vitamin C and Iron content of Millet koozh:-

The vitamin C and iron content of millet koozh subjected to four different processing methods are presented in table 4.

Table 4:- Vitamin C and Iron content of Millet koozh prepared using varying combinations of processing methods.

Processing methods	Vitamin C (mcg/100g)	Iron (mg/100g)
Non-germinated & Non-fermented millet koozh	16.63	2.24
Non-germinated & Fermented millet koozh	36.45	3.17
Germinated & Non-fermented millet koozh	28.83	1.06
Germinated & Fermented millet koozh	53.73	2.81

From table 4 it can be observed that the germinated & fermented millet koozh had the highest vitamin C content of 53.73 mcg/ 100g and the lowest content of 16.63 mcg / 100g was detected in the non-germinated & non-fermented millet koozh. Millets are generally not a source of vitamin C. However on germination, some amount of vitamin

C is synthesized in the grain and on fermentation there is a further rise in the vitamin content as also observed by Juana, Miranda and Doblado (2005)^[8].

It can also be observed from Table 4 that the Non-germinated and fermented millet koozh had the highest iron content of 3.71mg/ 100g and the lowest iron content of 1.06mg/100g was observed in the germinated but non-fermented millet koozh. Saleh et al., (2013)^[9] observed a marked reduction in phytic acid during germination and fermentation that helps release the iron bound to phytates which in turn increases the bioavailability of iron from foods.

Comparison of the vitamin B content in millet koozh:-

The thiamine, riboflavin and niacin content of non-germinated & fermented millet Koozh and germinated & fermented millet koozh are presented in Table 5.

Table 5:- Thiamine, Riboflavin and Niacin content in Non-germinated & fermented millet koozh and Germinated & fermented millet koozh.

Processing method	Thiamine B ₁ (mg/100g)	Riboflavin B ₂ (mg/100g)	Niacin B ₃ (mg/100g)
Non-germinated & fermented millet Koozh	0.2788	0.21	1.7714
Germinated & fermented millet Koozh	0.625	0.272	1.4486

It can be inferred from table 5 that there is an increase in vitamin B₁ and a negligible raise in B₂ in millet Koozh prepared using germinated millets and subjected to subsequent fermentation. The Niacin content of the Non-germinated & fermented millet Koozh was found to be slightly higher than the Germinated & fermented millet Koozh.

The marginal increase in B₁ (Thiamine) from 0.278 to 0.625 mg in the germinated and fermented sample could be due to the presence of yeast in the millet koozh. Khetarpaul et al., (1989)^[10] have stated that yeast fermentation raises the level of thiamine two-three fold times.

In the present study, the appreciation in thiamine and riboflavin levels places the millet koozh as a good complementary food as it meets the RDA of children 1-3 years of age. It can also be used as a nutritious food supplement during convalescence, a nourishing diet for the elderly and a healthy breakfast option for all age groups.

LAB and yeast content of millet koozh:-

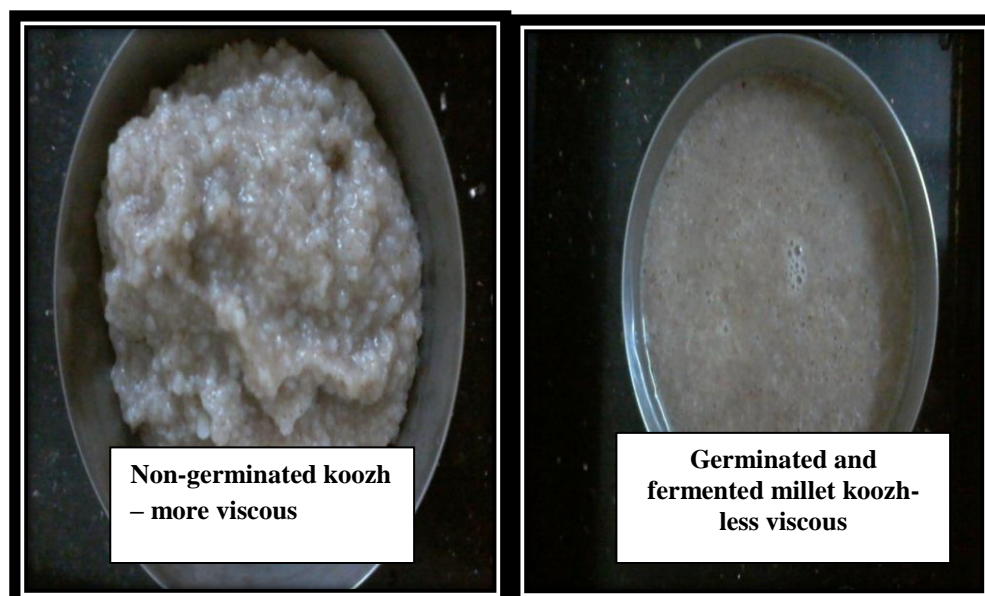
In this study, the presence of Lactic Acid Bacillus (LAB) was seen both in the non-germinated & fermented and the germinated & fermented millet koozh. LAB were generally reported to be the predominant bacteria in almost all fermented cereal and millet products (Khetarpaul et al., 1989)

The yeast content in the germinated & fermented millet koozh was significantly high which also substantiates the increase in B vitamins in the germinated and fermented millet koozh.

Sensory evaluation of millet koozh:-

Non-germinated & fermented millet koozh and Germinated & fermented millet koozh were evaluated for their colour, flavour, texture, taste, aroma and over all acceptability using the 9 point hedonic scale by a panel of twenty five respondents in the age group of 18 to 50 yrs

The germinated & fermented millets scored better on texture than the non-germinated & fermented millet koozh, as the germinated millet koozh was found to be less viscous than the non-germinated & fermented millet koozh (significance at $p < 0.01$). Due to enzymatic breakdown of starch to sugars during germination, the viscosity and bulk density of porridge made from sprouted grains were significantly lower.



It was observed that fermentation generally enhanced the taste, flavour, aroma and consistency of the food product. This may be due to softening of the substrate by starch hydrolysing microorganisms causing amylolytic activity (Ojijo&Shimoni, 2004). This emphasizes the importance of starch hydrolysing bacteria population in the koozh fermentation process.

Conclusion:-

This study reveals that germination and fermentation of millets are good processing methods that can be adopted to enhance not only the quantity of nutrients but also their bioavailability. They could serve as a less expensive source of nutrients for all strata of society across all age groups. It could also serve as an ideal food for convalescents and as a weaning food, compared to expensive commercially available formulas. The introduction of millets into the public distribution system and into government meal programmes like the mid-day meal scheme can help the poor and the marginalized households in meeting their nutrition needs.

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RESEARCH ARTICLE

PATIENTS' PRIMARY COMPLAINTS FOR SEEKING DENTAL HEALTHCARE SERVICES.

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Abstract

Background: Many patients visit dental hospital every day and the reasons for their visit can be varied. While few of them went for regular dental check-up, a majority of the patients visited dental hospital only when they have primary complaints most commonly being pain. Thus, this survey is done to find out the details about patients' dental check-up and primary complaints.

Materials and methods: The data was collected within three continuous working days of the Saveetha dental hospital in the outpatient hospital department. The study sample consisted of 110 people, from the age group of 18-65 years. A self-administered, anonymous questionnaire was prepared in both local language and English version used to accumulate data regarding demographic details, history of dental visit, patients' primary complaints and reasons for not visiting dental clinic.

Results: The study revealed that the patients' primary complaints were toothache which scored the highest about 39%. With the primary complaints, patient would proceed with invasive procedures such as tooth extraction and if possible root canal treatment. If stains were patients' primary complaints, procedures such as scaling can be carried out. Majority of the patients have received scaling as the treatment undergone so far with the least was prosthetic treatment. Patients' primary complaints can proceed with either preventive or corrective treatment.

Conclusion: Data from the study revealed that diverse patients of a private dental hospital had toothache as their primary presenting complaints. There is an essential to mobilize community health approach in order to increase awareness of importance of dental healthcare to encourage regular dental check-up by more preventive treatment rather than corrective treatment.

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Introduction:-

Nowadays, missionaries and nongovernmental organizations in addition to government dental healthcare have provided improved healthcare access especially dental healthcare though there is inequitable distribution. As there is increasing prevalence of oral diseases in government dental healthcare associated with prolonged patient physical waiting-time, thus patients are motivated to seek care in missionary and also private-owned dental health care. There is an evidence that missionary healthcare provides various dental healthcare services to various population as the cost of dental care is lesser in comparison to private clinics. As most of the available studies were conducted

primarily in government-owned dental healthcare, thus there is paucity of information regarding reason of patients for seeking dental care services in missionary hospital (1,2).

People visiting dental healthcare for various reasons. Most of them visited dentist only when there was toothache or follow-up treatment. The reasons can be varied depending upon the current situations. Patients' complaints could be toothache, fractured teeth, mobile teeth, routine dental check-up, follow-up appointment, decayed teeth, stains and others. People went to dentist to solve dental problems and relieve pain.

Therefore, this survey was conducted to study the patients' primary presenting complaints for visiting dental healthcare, dental history and reasons for not visiting dentist.

Materials and Methods:-

The data was collected within three continuous working days of the dental college hospital. The survey was done in the outpatient hospital department. The study sample were patients visiting the department during 3-days period.

The study sample consisted of 110 people, aged 18-65 years. Out of 110 people, 87 people had previously visited a dentist. A self-administered, anonymous questionnaire was prepared in both local language and English version used to accumulate data regarding demographic details, history of dental visit, primary patients' complaints and reasons for not visiting dental clinic.

For those who had previously visited dental hospital were requested to state the time from their last dental visit, reason for it and the treatment received during last dental visit. In the questionnaire, the participants were asked to choose from a list, the reason for not visiting dental hospital. The statistical analysis was collected and presented in percentages.

Results:-

The results from the study has shown that majority of people who had previously attended a dental hospital belonged to the age group of 20-40 years, approximately 73%, and majority of the participants who had never previously attended dental healthcare service was in age group of 41 and above.

More numbers of female had visited dental hospital, about 60% in comparison to male participants, 40%.

Table 1:- Sample description by age and gender.

Age	N	Gender	
		Male	Female
Below 20	19	10	9
21-30	47	17	30
31-40	33	15	18
41 and above	11	3	8
Total	110	45	65

Table 2:- Results of survey questionnaire

Questions	Number of patients	
	Male	Female
How many times have you visited dental clinics?		
○ First visit	19	11
○ Sometimes	29	36
○ Frequent	7	8
What is your age on your first dental visit?		
○ Below 20	12	20
○ 21-30	20	21
○ 31-40	17	10
○ 41 and above	3	7
What is the primary complaints of your first dental visit?		
○ Tooth pain	20	16

<ul style="list-style-type: none"> ○ Decayed teeth ○ Mobile teeth ○ Stains ○ Missing teeth ○ Others 	5 10 19 5 2	8 11 13 1 -
When is your last dental visit? <ul style="list-style-type: none"> ○ Last week ○ Last month ○ Last year 	10 9 25	14 4 18
What is the reason of your last dental visit (if have)? <ul style="list-style-type: none"> ○ Tooth pain ○ Decayed teeth ○ Mobile teeth ○ Stains ○ Missing teeth ○ Others 	15 3 1 4 9 8	16 9 3 9 2 1
What is the treatment that you received for your last dental visit? <ul style="list-style-type: none"> ○ Scaling ○ Tooth extraction ○ Restoration ○ Prosthetic treatment ○ Orthodontic treatment ○ Others 	10 7 9 3 - 2	18 13 6 1 5 6
What is the treatment that you have undergone so far? <ul style="list-style-type: none"> ○ Scaling ○ Tooth extraction ○ Restoration ○ Prosthetic treatment ○ Orthodontic treatment ○ Others 	10 7 9 3 - 2	18 13 6 1 5 6
What is the reason for not visiting dentist? <ul style="list-style-type: none"> ○ Problem is not severe enough ○ Neglect oral problems ○ No dental problem ○ Treatment is costly ○ Fear of dental treatment ○ Frequent visits are required 	11 11 3 5 7 17	9 4 12 4 18 9
What is your primary presenting complaint for dental visit? <ul style="list-style-type: none"> ○ Toothache ○ Follow up appointment ○ Fractured teeth ○ Routine dental check up ○ Others 	21 7 11 6 6	26 15 8 6 4

Discussion:-

In this study, more females than males seek dental healthcare services. This result is similar with likely studies regarding oral health seeking behaviour conducted in Southern Nigeria (3,4).

The study results showed that majority of the people visited dentist mostly sometimes, about 60% followed by first visit, 27% and frequent, 14% as shown in Figure 1.

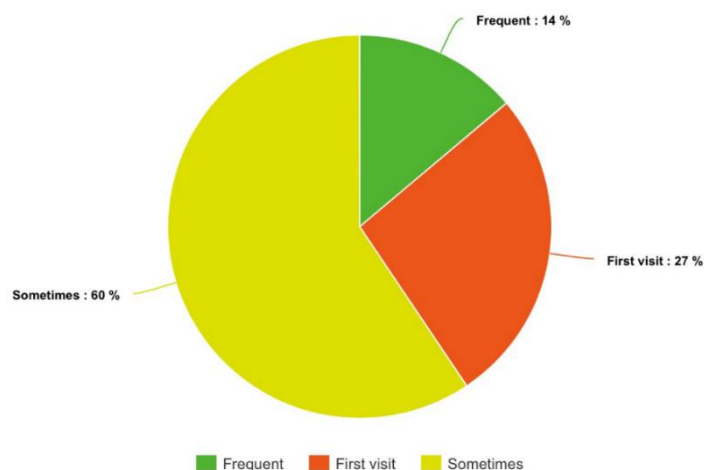


Figure 1:- Frequency of dental visit.

In figure 2, most of the patients visited dentist for the first time at the age of 21-30 and 31-40, altogether contribute approximately 62%. 41 and above scored for the least whose people visited dentist for the first time.

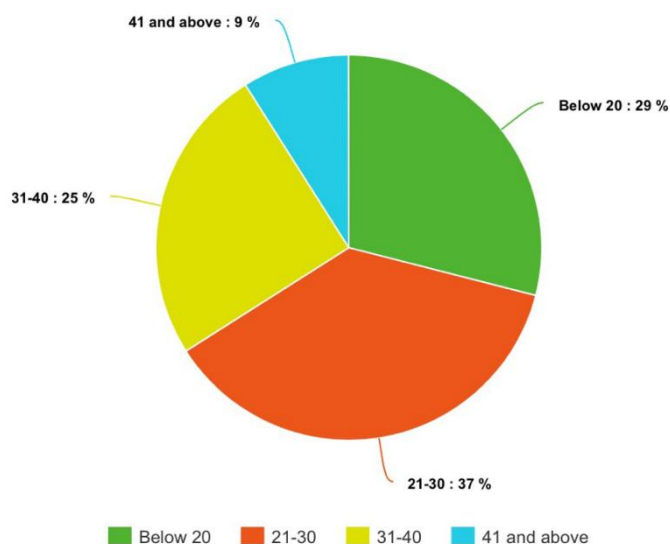


Figure 2:- Age on first dental visit.

The purposed of their first dental visit was tooth pain which scored the highest about 39%. This finding is similar with reports of others studies citing pain as the most likely reason stated by patients for seeking dental healthcare services (1,4). Stains scored the second highest, followed by mobile teeth, decayed teeth, missing teeth and others for the patients' primary complaints. Toothache complaints were primarily due to dental caries and its complications are gingivitis periodontitis, fractured teeth, impacted teeth, and oral ulcers.

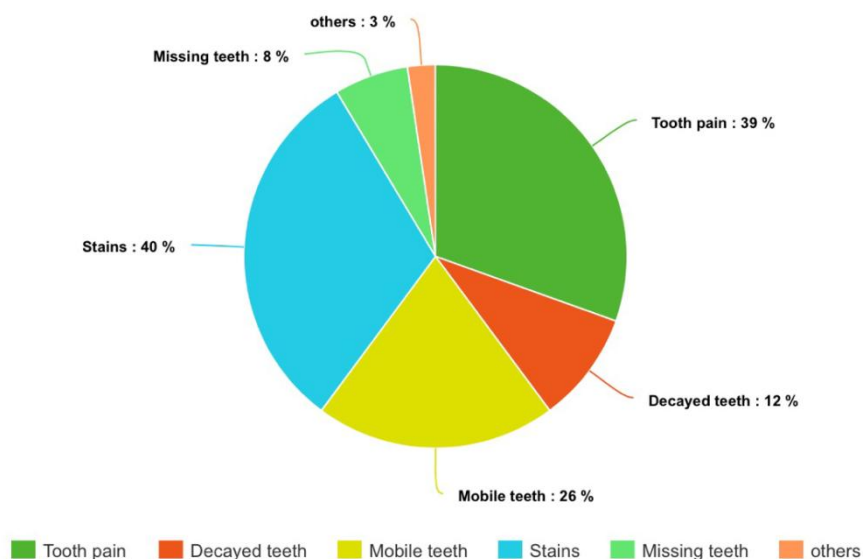


Figure 3:- Primary complaints of first dental visit

Almost 54% of the patients went for last dental visit last year, followed by last week and last month. The percentages recorded by each were 30% and 16% respectively.

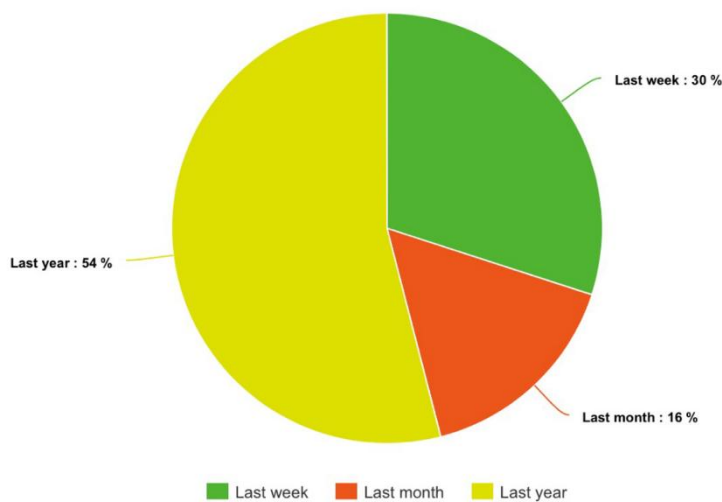


Figure 4:- Last dental visit.

The main reason of their last dental visit which scored highest and least were tooth pain and mobile teeth, 39% and 5% respectively. Other reasons were decayed teeth, stains and missing teeth as depicted in Figure 5. Each has corresponding percentages. Shaking teeth accounted for 5% of the last presenting complaints which is an indication of irreversible periodontal diseases, periodontitis.

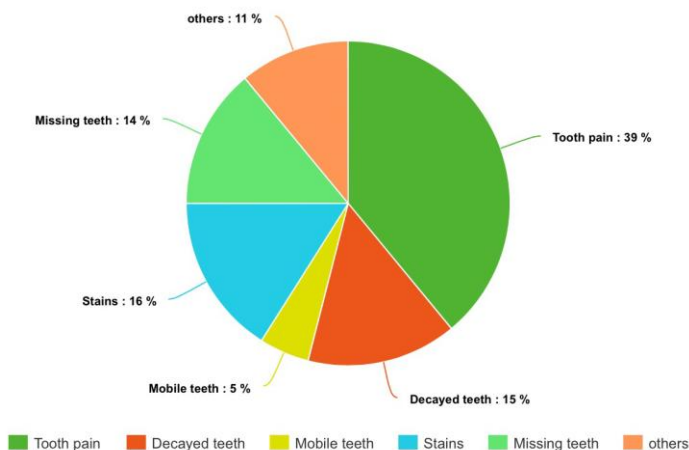


Figure 5:- Main reason of last dental visit.

Approximately, treatments received by 35% of the patients were scaling, followed by tooth extraction, 25%, restoration, 19%, others, 10%, orthodontic treatment, 6% and prosthetic treatment, about 5%.

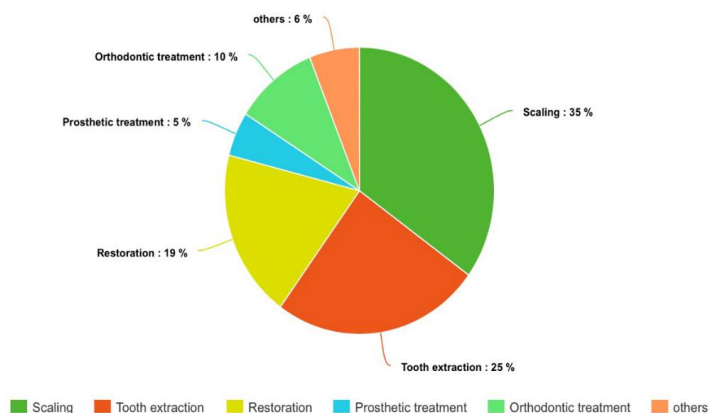


Figure 6:- Treatments undergone so far.

There were few reasons why people did not attend dental healthcare. They were frequent visits are required, fear of dental treatment, thinks problem is not severe enough, neglect oral problems, no dental problem and treatment is costly as shown in Figure 7. Percentages contributed by each were 24%, 23%, 18%, 14% and 8% respectively. Symptomatic and irregular dental visit has been reported to be more common than preventive dentist visits. This could be due to ignorance, poor oral health awareness, and poverty.

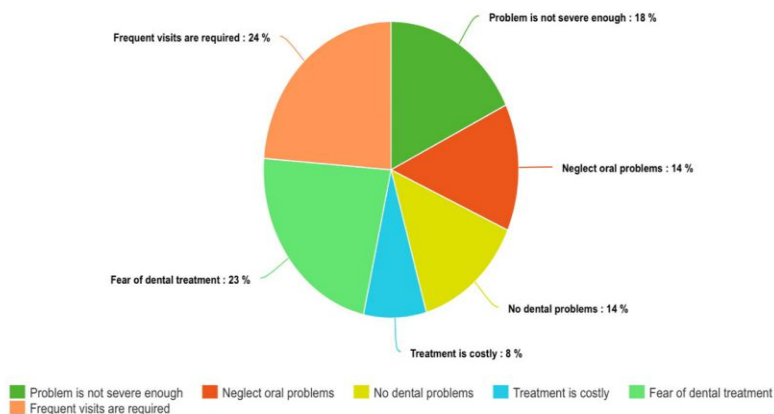


Figure 7:- Reasons for not attending dentist.

The toothache complaints and dental caries were highest among all range of patients visited dental hospital. Follow-up appointments, 20% being the second most common presenting complaints in this study reflects an aesthetic demand/need among dental patients. Fractured teeth, 17% were the third most common primary presenting complaints among the patients. The prevalence of fractured teeth as one of the major contributor to factors prompting the patient to seek dental healthcare services. Routine dental check-up and others scored the least, 11% and 9% respectively.

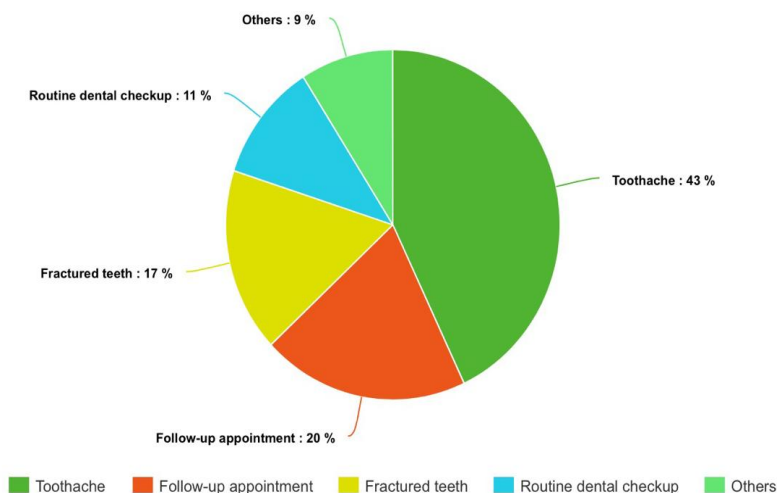


Figure 8:- Primary presenting complaints.

Conclusion:-

Data from the study revealed that diverse patients attended a private dental hospital had toothache as their primary presenting complaints. There is an essential to mobilize community health approach in order to increase awareness of important dental healthcare.

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RESEARCH ARTICLE

STUDIES ON COASTAL GEOMORPHOLOGICAL LANDFORMS IN THE SOUTHEAST COAST OF THIRUVALLUR COASTAL STRETCH, USING REMOTE SENSING AND GIS TECHNIQUES.

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Key words:-

Geomorphology, Thiruvallur, ArcGIS.

Abstract

Geomorphology is the study of the landforms of the earth and the process that form them especially those that lead to the erosion, transport, and deposition of sediments. The present study has been made to record the varied coastal geomorphic features along the Thiruvallur coastal stretch. The coastal zone of southeast coast have contained a lot of morphological changes due to the many natural hazards. The area of Thiruvallur is the part of the Southeast coast of India. The geomorphic features of this area resulted from various coastal landward process of erosion-accretion or due to other natural causes of sea level changes and sea water interactions. The details of the features such as strand lines, sandy beach, mudflat, lagoon, back swamps, barrier Island, salt marshes, estuary, tidal creeks, spit and shoal has been recorded as well as the significance of their occurrence is presented. The coastal geomorphology of the study area clearly established not only the sea level changes but also landward process and climatic conditions.

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Introduction:-

The geomorphology is the study of the physical features of the surface of the earth and their relation to its geological structures. Coastal geomorphology is a branch of geomorphology in- Geomorphology methodologies focused mainly on field observation, description and measurement but also included physical experimentation, there is growing recognition of the importance of geomorphology and geomorphologists in contributing to a range of environmental investigations and management issues. The coastal geomorphology was highly descriptive and much of it was influenced by concept related to the erosion cycle. Much of the research in coastal geomorphology in the past four decades has been concentrated in the zone influenced by wave and tides wave, generated currents, and tidal currents and on the erosional and depositional features associated with these. The coastal zone features have been developed due to the process of erosion-accretion or due to other natural causes. The features can be categorized as depositional features, erosional, and special features (Nair, 1990). The geomorphic unit under different heads namely features formed by present day wave action, features formed due to sea level oscillations and features formed by rock and sea water interaction have been presented. The influence of the sea level oscillations and land and sea interactions forms a diverse coastal geomorphology along the coast (Jaganadha Rao, 2012). The response of coastal geomorphic types and landforms to sea level rise is complex, coastal forms being a product of interactions between energy and material, with stepwise adaptations external forcing sea level rise as a driver of coastal change needs to be seen within the context of other process and determinant particularly in relation to wave tides, storms and

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sediment supply (Flemming, 2011). Geomorphic hazards are regarded as related to landscape changes that affect human systems. The process that produces the changes are rarely geomorphic in nature but are better regarded as atmospheric or hydrologic (Paul, 1994). Geomorphology is also concerned with the study of earth surface forms and their evaluation in time and space due to the physicochemical and biological factors acting on them most of the evolution is the product of a cyclic process based on erosion transport deposition of sediment particle (Gerardo, 1995). The geomorphology mapping it involves the partitioning of the terrain into conceptual spatial entities based upon criteria that include morphology, genetics compositions, and structure, chronology environmental system (Michael, 2012). Extreme rainfall and flood events in steep upland catchments leave geomorphological traces of their occurrence in the form of boulder berms, debris and alluvial fans (Foulds, 2014).

The Geoinformatics system provides multiple applications for studying different coastal aspects. With the rapid development and change of GIS software package in the late 1980's their utility for coastal management was quickly recognized. The satellite remote sensing due to its repetitive multispectral and synoptic nature provides a unique view to recognize various features on land and sea (Siddiqui, 2003). This technique can be a great help in the collection of continuous wave current, wind, tides, suspended matter, shallow water area, wetlands, mangroves mudflats and other coastal features and coastal change like accretion or erosion.

Study Area:-

The study area lies in the coastal stretch of Thiruvallur district, Tamil Nadu, India it is bounded to the north by Pulicat lake south by Chennai east by the bay of Bengal and west by Minjur area. It lies between $80^{\circ}02'30''\text{E}$ and $80^{\circ}22'30''\text{E}$ longitude $13^{\circ}15'00''\text{S}$ and $13^{\circ}35'00''\text{S}$ latitude. The coastal stretch of the study area has a total length of 28km along the Bay of Bengal. The Thiruvallur coastal stretch is known for diverse coastal geomorphic features. The environment namely as strand lines, sandy beach, mudflat, lagoon, back swamps, Barrier Island, salt marshes, estuary, tidal creeks, spit and shoal resulted in characteristic and significant coastal geomorphology because of land-sea interaction. This coast has a very vast coastal plain, which extends from north of toppalapalayam to south of sattangadu. The physiography of the coast consists of sandy beach sands and extends over a width of 500m from the coastline. The main geological formation occurring in the coast are upper Gondwana sand silt, quaternary sand and clay undulation by Archean crystalline rocks Charnockite.

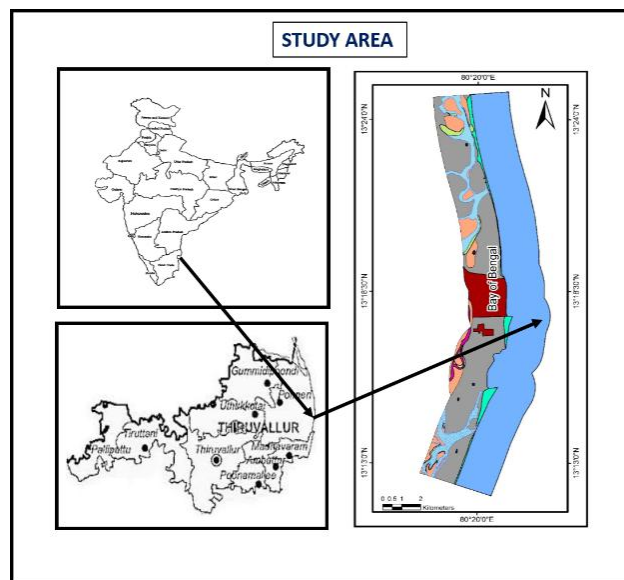


Figure 1:-study area map.

Methodology:-

In this study deals with the description, mapping, and field analysis of various coastal geomorphological assemblages along the southeast coast of Thiruvallur. Coastal geomorphological studies involve mapping of different types of landforms. Mapping of coastal is primary to the understanding of the evolution of any coastal area. The integrated remote sensing and GIS techniques are employed for extracting the coastal geomorphological landforms at high resolution. The success met with the geomorphology mapping from imagery for obtaining accurate results and imageries are mainly utilized for mapping. The various types of spatial data source such as

topographical map (scale of 1:50,000) published by Survey of India the year of 1970-1971, satellite data Landsat- 8 OLI image interpreted based on the visual interpretation keys and identified the geomorphology of the study area using ArcGIS 10.2 software. The Trimble GPS was used for ground truth validation. The geomorphic features formed by present day coastal process, features formed due to sea level oscillation and most of the coastal features has been developed due to the process of erosion and accretion or due to the natural causes have been identified at different places and description of individual features have been recorded. The features were confirmed using field data.

Results and Discussion:-

The area of Thiruvallur coastal stretch between Pulicut to Tiruvottiyur a length of the shoreline 28km the entire area is marked by varied and geologically significant geomorphological features the geomorphic units under different domes namely features formed by present day wave action features formed due to sea level changes and land and sea water interaction have been presented. The entire area is marked by varied geomorphological features. The extensive structure-activity due to rapid urbanization is resulting in the damage of these beautiful features day by day. As such these features are verified for the help for the feature researchers. All the significant features have been studied for scientific documentation. Geomorphological map of this area is prepared fig:2. The study area occurs different land forms that detailed description of individual features is presented below. For the ease of arrangement, the features have been classified into two types that are marine landforms and fluvial landforms.

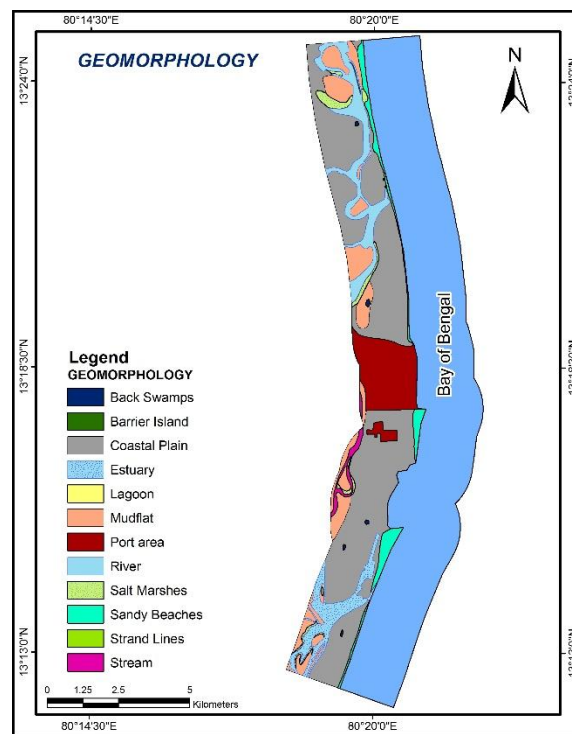


Figure 2:-Geomorphology Map.

Depositional features:-

Coastal deposition is the arranging depressed of material on the coast by the sea it occurs when waves lose energy or when large inputs of sediments are made into the coastal system. Here, the study area occurs different type of depositional features that is a sandy beach, coastal dunes, Barrier Island, coastal plain, Back swamps, mud flats, strand line, salt marshes, and tidal creek. In this study area, the coastal depositional features are categorized by marine landforms and fluvial landforms.

Marine landforms:-

The sea implements the function of erosion and deposition through sea waves, supported by currents, tides, and storms in coastal areas. The study area falls under different marine landforms such as the sandy beach, Sand dunes, Coastal plains, Barrier Island, Strandlines, Lagoons, Mudflat, Salt marshes.

Sandy Beach:-

The sandy beaches are extensively developed along the entire coast and its occupied nearly 3 sq.km of the study area. A gently sloping zone and it is composed of particles. Beach typically occur in areas along the coast where wave or current action deposit and rework sediments. Coastal sediments are affected by wave courses, but commonly the deposition of sediments along the shoreline are referred to as the beaches. In this study area beaches are present for about a length of 28 km in a linear from Tiruvottiyur to Pulicut. A marked landward ridge which forms the landward limit of wave swash is referred to as the berm. The berm is predominantly visible in the Kattupalli and Tiruvottiyur location also it has been observed all along the beaches between Tiruvottiyur to Pulicut. Wave cut face of the berm reveals the alternative layering of black sand and white sands thickness of the individual layers varies from a few mm to few cm. beach morphology of the study area is highly dynamic and changes with wave energy, sediment supply, long shore current and substrate physiography. The surface structure like ripple marks also occur. The ripple formation can be ascribed in due to the oscillatory flow of wind.

Sand dunes:-

In this study area, coastal morphological features of dunes are presently more active. the feature is met all along the south-eastern side of the study area. Coastal is the formation of coastal and dunes the material for the formation these dunes is essentially derived from the present day beach covering from foreshore to backshore. The furthestmost characteristic of this features are dunes is the vegetative cover the vegetation is a good effect in helping to bind together the sand by their complex root system and those help to protect the dunes from erosion. Coastal dunes sediments from the intertidal area get regularly and periodically exposed to the onshore winds to be reworked in the form of coastal dunes. Dunes from where constructive wave encourage the accumulation of sand and where preventing onshore winds blow this sand inland. The wind starts to affect the mound of sand by eroding sand particles from the windward side and depositing them on the leeward side gradually this action causes the dune to migrate inland as it does so it accumulate more and more sand.

Coastal plains:-

The coastal plain is the relatively flat landforms created by the deposition of sediment over a long period of time by one or more rivers coming highland regions. The study area occurred 25 sq.km of flat coastal plain along the Tiruvottiyur to Pulicut region. The coastal plain is a flat, low laying piece of land next to the ocean are separated from the rest of the interior by nearby landforms such as mountains coastal plains can also develop when river current carry rock soil and sedimentary material into the ocean (Nsengmuuremayi 2016). Level or gently sloping region or a slightly undulating land surface produced by extensive deposition of alluvium usually adjacent to the river that periodically overflows its banks.

Barrier Island:-

These islands are long, narrow, offshore deposits of sand or sediment that run parallel to the coastline. They are separated from the mainland and it's a type of dune system that is exceptionally flat or lumpy areas of sand that form by a wave and tidal action parallel to the mainland coast. The area of Pulicut Lake occurred one barrier island north part of the study area. The length and width of the barrier and overall morphology of barrier coast are related to parameters containing tidal range, wave energy, sediment supply, sea-level trends and basement controls. Barrier islands are very important for reducing the divesting effect of wind and waves and for absorbing storm energy. They are also imported marine habitats that support commercially important fish species as well as birds, sea turtles, and other wildlife species.

Strandlines:-

The study area contains two strand lines it is a broad belt of sand along a shoreline with a surface exhibiting well defined parallel sand ridges separated by shallow swales. A strand plain differs from a barrier island in that it lacks either the lagoons or tidal marshes that isolated barrier islands from the shoreline. The strand plain is directly attached strand plain typically are created by the redistribution by waves and longshore current of coarse sediment on either side of a river mouth. Beach ridges include all relict strand plain ridges, whether dominated by wave/swash-built or by Aeolian lithesome. A strand line refers collectively to the assemblages of various features characteristics of the former coastal area the strand lines here represent the transgression and regression process.

Lagoons:-

A lagoon is the shallow body of water separated from a larger body of water by Barrier Island of reefs. The inside of Ennore estuary has lagoon they have also been identified as occurring on mixed sand and gravel coastline. Coastal

lagoons from along gently sloping coasts where barrier islands or reefs can develop off-shore, and the sea level rise is rising relative to the land along the shore, coastal lagoons do not form from a long steep or rocky coast or if the range of tides is more than 4 meters due to the gentle slope of the coast the coastal lagoons are shallow. The lagoons are sensitive to change in sea level due to global warming. Coastal lagoons are young and dynamic and many be short lived in the geological term. Coastal lagoons are usually connected to the open ocean.

Mudflat:-

Mudflat is a flat area containing 6 sq.km of the study area. Mudflats are fluid to a mixture of finely divided particles and solid materials mainly silt, clay and water they are associated with the sheltered environment like lagoons, estuaries and another embayment the barrier beaches occurring on the seaward side of the backwaters create a sheltered environment which has favored the development of mud flat. Mudflats are developed where enough fine sediments are available and the wave action is not strong enough to disturb the sediments. This area is very suitable sites for the deposition of mud flats because of the availability of sediments monthly suspended very fine grained. Are wide expanses of fine grained soft mud along the shore they generally consist of deposit of clay, silt, oozes etc. they are further classified into high tidal, subtidal and supratidal mudflats, intertidal slopes and subtidal zones. High tide tidal flats are more or less flat and are near the high – water line deposition is provided by the highest tides. The intertidal slope is the area of instability and is affected by daily tides.

Salt marshes:-

Low laying Athipettu area that is stabilized by cordgrasses and flooded by daily tidal activity. Salt marshes are salty because they are flooded by seawater every day. The study area contained 2 sq.km marshy because their ground is composed of grass and peat. The more often area is flooded, the more saline it is planted living in salt marshes have different tolerance to salt. Those with higher tolerance and found in the low marsh, and those with lower tolerance to salt are found in the high marsh zones. Plants from one marsh zone are never found the other. Salt marshes serve as the transition from the ocean to the land, where fresh and salt water mix. Salt marsh plants are salt tolerant and adapted to water levels that fluctuate with the tide. Tide carry in nutrients that stimulate plant growth in the marsh and carry out organic material

Fluvial landforms:-

The fluvial landform process associated with rivers and streams and the deposits and landform created by them. Coastal lowlands contain a variety of landforms most of which are geologically young. Today coastal landforms are quite recent their formation and properties depend in part on whether fluvial process interacted with the marine process. The combined actions of fluvial and marine process determine whether or not a depositional body can form at the mouth of a river and what kind of body will form.

Estuary:-

An estuary is a partially enclosed coastal body of brackish water with one or more rivers or streams flowing into it and with a free connection to the open sea. The estuary is otherwise known as where the salt water and fresh water mix. The study area have included Ennore estuary and it is very vast and extent from Tazhamkuppam to Tiruvottiyur. Estuaries and their surroundings wetlands are bodies of water usually found where rivers meet the sea. Estuaries are home to single plant and animal communities that have adapted to brackish water a blend of fresh water draining from the land and salty seawater. Estuaries are among the most productive ecosystems in the world many animals rely on estuaries for food. Estuarine environments are among the most productive on earth, creating more organic matter each year than comparably-sized areas of forest, grassland or agricultural land. The sheltered waters of estuaries also support unique communities of plants and animals specially adapted to life at the margin of the sea.



Figure 3:-Sand Dune.



Figure 4:-Sand Berm.



Figure 5:-Sandy Beach.



Figure 6:-Sand Ripple.



Figure 7:-Ennore Estuary.



Figure 8:-Coastal Plain Area.



Figure 9:-Salt Marshes.

Back swamps:-

Back swamps are the section of floodplain where a deposit of fine silts and clays settle after a flood. These are usually lying behind a stream's natural levees are located some distance away from the stream channel on the flood plain when water spills over onto the flood plain. The fine-grained alluvium holds much water and drains rather slowly creating altered areas back swamps are retained water that might cause severe flooding downstream. Some area has identified with back swamps and that contains the very small area.

Tidal creek:-

Any inlet through which water flows alternatively towards landward with the rising tide and seaward with the falling tide is called as tidal creeks or inlets. Inlet may be permanent or temporary. Kalikuppam location south one permanent inlet through which water flows alternatively towards land with the rising tide and seaward with the falling tide throughout the year is seen. Is a tidal water channel in the coast creeks may often dry to a muddy channel with little or no flow at low tide but often with significant depth of water at high tide, creek are seen mostly in the southern region and some in the northern region of the study area, which includes the Ennore creek. The mouth of certain paleochannels is also seen as present day creeks in the study area.

River and stream:-

The river is a natural flowing watercourse, the river flowing over gently sloping ground begin to curve back and forth across the landscape the moving water washes away the sediment like sand and mud. The stream is a narrow river and it also a body of water. Rivers in the study area covers nearly 4 Sq. km. all rivers are carrying flood during monsoons.

Conclusion:-

The attempt to determine the coastal geomorphology from Tiruvottiyur to Pulicat by using Landsat- 8 OLI data. The present study shows the satellite remote sensing based coastal geomorphological features mapping is very useful and very effective for getting the result of coastal geomorphology. The study has been conducted with Landsat- 8 OLI data; the study area falling the coastal area Tamil Nadu shows major geomorphological landforms under the influence of the coastal parameters.

The geomorphological features are classified by two that is marine landforms and fluvial landforms. The study area is marked by diverse geomorphic features of these two landforms with the help of the field validation. The coastal geomorphic features clearly established the sea level oscillations and variations in climatic conditions along this study area.

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RESEARCH ARTICLE

PHARMACOVIGILANCE STUDY OF ANTI-ASTHMATIC AGENTS IN PATIENTS OF BRONCHIAL ASTHMA AT A TERTIARY CARE CENTRE.

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Pharmacovigilance, antiasthmatic agents, WHO-UMC causality categories, Hartwig and Siegel scale

Abstract

Background: Bronchial asthma is one of the most common chronic disease in the world affecting around 334 million people. Management of bronchial asthma includes multidrug therapy for long duration, which leads its association with adverse drug reactions (ADRs). Pharmacovigilance studies of antiasthmatic agents are scarce in India. Therefore, the present study was planned to monitor and evaluate adverse drug reactions associated with antiasthmatic agents.

Objectives: The present study was conducted to evaluate the pattern, causality and severity analysis of adverse drug reactions associated with antiasthmatic agents in a tertiary care centre.

Materials and Methods: 150 patients of either gender, ageing above 18 years with established bronchial asthma attending outpatient and inpatient department of medicine at a tertiary care centre interviewed during the time period of October 2016 to February 2017. Central Drugs Standard Control Organisation (CDSCO) ADR forms were filled. World Health Organisation- Upasala Monitoring Centre (WHO-UMC) causality categories were used for assessment of causality. Severity of ADRs was assessed by using Hartwig and Siegel scale.

Results: A total 33 ADRs were reported in 23 patients out of 150 bronchial asthma patients. Among the 23 patients reported with ADRs 10 (43.47%) were male while 13 (56.52%) were female. Oral thrush was most common ADR (33.33%) followed by palpitation (15.15%), sore throat (12.12%), running nose, tremors (each 9.09%), dry mouth, GI distress, bitter taste (each 6.06%) and headache (3.03%) among the patients of bronchial asthma receiving antiasthmatic agents. Most ADRs were associated with inhalational Beclomethasone (58.33%) followed by inhalational budesonide (25%), montelukast (23.07%), salbutamol (18.75%), theophylline (14.29%), ipratropium (7.4%) and salmeterol (02.22%).

According to WHO-UMC categories 48.57% ADRs were found to be probable while 51.42% were possible. Highest percentage of ADRs 75.79% were classified as mild and 24.24% were moderate on Hartwig and Siegel scale.

Conclusion: Results of our study highlighted the need for ADR monitoring of antiasthmatics in asthma patient. Patients receiving inhalational steroids need to be properly counselling and also written

advice about cleansing mouth after steroid inhalation to reduce the risk of oral thrush.

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..... **Introduction:-**

Bronchial Asthma is defined as reversible obstruction of airways of lungs due to its hyper-responsiveness to external or internal allergen or nonspecific stimulus like exercise, cold and characterized pathologically by chronic airway inflammation and clinically by cough, wheeze, chest tightness and dyspnoea.¹ The global prevalence of asthma is anticipated to be approximately 4.5 per cent.^{2,3} There are about 334 million patients with asthma affecting all age groups, across the world. The prevalence of asthma has increased over the time and an additional 100 million people worldwide are expected to develop asthma by the year 2025.⁴

In the Indian study on epidemiology of asthma, respiratory symptoms and chronic bronchitis in adults (INSEARCH), the prevalence of asthma in adults was 2.05 per cent, with an estimated burden of 17.23 million.⁵ A recent analysis using three different estimate models (INSEARCH, GINA and WHO survey) suggests that the prevalence of asthma in India varies between 2.05 to 3.5 per cent (17-30 million patients).⁶ India is projected to become the world's most populous nation by the year 2050. As a result, further predicted increase in the prevalence of asthma will result in a marked increase in the number of asthmatics.⁷

Adverse drug reaction (ADR) is associated with almost every drug ranging from mild to serious and life threatening. WHO defines ADR as "any noxious, unintended & undesired effect to a drug that is administered in standard doses by the proper route for the purpose of prophylaxis, diagnosis or treatment".⁸ WHO, 2004 pioneered the concept of pharmacovigilance i.e. "Science and activities relating to the detection, assessment, understanding and prevention of ADR or any other medicine related problems".⁹

ADRs constitute a considerable burden of society both financially and in terms of human suffering, and systemized ADR monitoring and reporting may sensitize physicians to rational prescribing.¹⁰

Pharmacovigilance studies for monitoring ADRs related to antiasthmatic agents have been performed by various workers around the globe. Reports on monitoring of ADRs in India are scarce.¹¹ Oral thrush, tremor, palpitations, throat irritation and cough are common adverse effects of antiasthmatic agents.¹² However causality and severity of these adverse effects remains undetected due to lack of pharmacovigilance studies in Indian context. The monitoring and evaluation of ADRs associated with antiasthmatic agents would provide the necessary information on adverse effect of different antiasthmatic agents. Therefore, the present study is planned to study the pattern of ADRs associated with antiasthmatic agents and their analysis for causality and severity in a tertiary care centre.

Materials and Methods:-

It was a non-comparative and hospital based cross-sectional questionnaire based pharmacovigilance study. The study protocol was approved by Institutional Ethics Committee. The study was carried out in outpatient and inpatient medicine department of tertiary care teaching hospital. Total 150 patients were interviewed from October 2016 to February 2017 and informed consent form was taken. All the patients of bronchial asthma, who were receiving antiasthmatics not less than 1 month were included in this study. Patients with other co-morbid conditions like hypertension, diabetes mellitus, arthritis, respiratory infections (such as COPD, bronchitis, lung diseases) and immunocompromised state were excluded from study.

Based on an ADR monitoring form which is drafted according to Central Drug Standard Control Organisation (CDSCO) monitoring guidelines. Demographic details (age, sex, weight) relevant medical history, present drug therapy, adverse event description, adverse event outcome and assessment information were collected. World Health Organisation-Uppsala Monitoring Centre (WHO-UMC) causality categories¹³ was used for assessment of causality. Severity of ADRs was assessed using Hartwig and Siegel scale. The obtained data was analyzed by using percentage method and chi square test to conclude the study results.

Results:-

A total 33 ADRs were reported in 23 patients out of 150 bronchial asthma patients. Among the 23 patients reported with ADRs 10 (43.48%) were male while 13 (56.52%) were female. 8 (34.78%) patients associated with ADRs observed in the age group of 41-50 years, followed by 5 (21.74%) in age group 21-30 years, 4 (17.40%) in age group 31-40 years, 3 (13.04%) in age group 51- 60 and 61-70 each were observed. Distribution of ADRs among various age groups ([Table 1](#)).

Oral thrush was most common adverse drug reaction (33.33%) followed by palpitation (15.15%), sore throat (12.13 %), running nose, tremors (each 9.09%), dry mouth, GI distress, bitter taste (each 6.06%) and headache (3.03%) among the patients of bronchial asthma receiving anti asthmatic agents ([Table 2](#)).

Most ADR was associated with inhalational Beclomethasone (58.33%) followed by inhalational budesonide (25%), montelukast (23.07%), salbutamol (18.75%), theophylline (14.29%), ipratropium (7.4%) and salmeterol (02.22%) ([Table 3](#)).

Among the total 23 patients, 11 were on monotherapy while 12 were on combination therapy. There was no significant difference in ADRs associated with monotherapy and combination therapy [Chi- square test, $p > 0.05$] ([Table 4](#)).

According to WHO-UMC categories, 48.49% ADRs were found to be probable while 51.51% were possible. ([Table 5](#)).

Highest percentage of ADRs (75.76%) were classified as mild on Hartwig and Siegel scale which included oral thrush, sore throat, running nose, dry mouth, GI distress, bitter taste, headache and were well tolerated by patients. While 24.24% were moderate which included palpitations and tremors, no severe reaction was observed ([Table 6](#)).

Discussion:-

A total 33 ADRs were reported in 23 patients out of 150 bronchial asthma patients. Among the 23 patients 10 (43.48%) were male while 13 (56.52%) were female.

Oral thrush was most commonly observed adverse drug reaction and was observed in 11 patients out of 23 (33.33 %) who received inhalational Beclomethasone and Budesonide corticosteroids, Most ADR (oral thrush) was associated with inhalational Beclomethasone in 7 out of 11 patients (58.33%). Oral thrush is an infection in the mouth caused by a yeast germ called, candida. Inhalational steroids may cause an overgrowth of candida which can lead to a bout of oral thrush. Cleansing mouth and brushing teeth after using the steroid inhaler may reduce the risk of oral thrush.^{14,15,16,17,18} All 7 patients were not rinsing mouth or brushing teeth after use of inhalation. No prior advice was given to patients regarding cleansing mouth after beclomethasone inhalation. High incidence of oral thrush in patients receiving inhalational beclomethasone is suggestive of need of counselling and advice to reduce the risk of oral thrush. There was no significant difference in ADRs associated with monotherapy and combination therapy. Highest percentage of ADRs (75.79%) were classified as mild ADR on Hartwig and Siegel scale which included oral thrush, sore throat, running nose, dry mouth, GI distress, bitter taste, headache and were well tolerated by patients. While 24.24% were moderate which included palpitations and tremors.

Conclusion:-

Results of our study highlighted the need of ADR monitoring of antiasthmatics in asthma patient. Patients receiving inhalational steroids needs to be proper counselling and also written advice about cleansing mouth after steroid inhalation to reduce the risk of oral thrush.

Table 1:- ADRs among various age groups of asthmatic patients.

AGE RANGE	MALE	FEMALE	Total (%)
21-30	2	3	5 (21.74)
31-40	2	2	4 (17.40)
41-50	3	5	8 (34.78)
51-60	2	1	3 (13.04)
61-70	1	2	3 (13.04)

Table 2:- Types and number of Adverse Drug Reactions.

Type of Adverse Drug Reaction	Number of ADRs (Percentage)
Oral thrush	11 (33.33%)
Palpitations	5 (15.15%)
Sore throat	4 (12.13%)
Running nose	3 (9.09%)
Tremors	3 (9.09%)
Dry mouth	2 (6.06%)
GI distress, nausea	2 (6.06%)
Bitter taste	2 (6.06%)
Headache	1 (3.03%)

Table 3:- Suspected drugs and their associated type of ADRs and number.

Antiasthmatic class	Drugs	No. of ADRs/ No of prescriptions (percentage)	ADRs (No.)
β_2 Agonist	Salbutamol	6/32 (18.75%)	Palpitations (3) Bitter taste (1) Tremors (1) Headache (1)
	Salmeterol	1/45 (02.22%)	Bitter taste (1)
Methyl Xanthines	Theophylline	4/28 (14.29%)	Palpitations (2) Tremors (2)
Corticosteroids	Beclomethasone	7/12 (58.33%)	Oral thrush (5) Sore throat (2)
	Budesonide	8/32 (25%)	Oral thrush (6) Sore throat (2)
Anti cholinergic	Ipratropium	4/54 (7.40%)	G I Distress (2) Dry mouth (2)
Leukotrine Antagonist	Montelukast	3/13 (23.07%)	Running nose (3)

Table 4:- Number of ADRs in patients receiving mono therapy and combination therapy.

Therapy	No. of Patients	No. of ADR	P > 0.05
Mono therapy	11	15	
Combination therapy	12	18	
Total	23	33	

Table 5:- Causality assessment of ADRs according to WHO-UMC categories.

Assessment	No. of ADRs	Percentage of ADRs
Certain	0	0
Probable	16	48.49
Possible	17	51.51
Unlikely	0	0
Conditional	0	0
Unconditional	0	0

Table 6:- ADR classification on basis of severity by Hartwig and Siegel scale

Severity	No of ADRs	Percentage of ADRs
Mild	25	75.76
Moderate	8	24.24
Severe	0	00.00

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RESEARCH ARTICLE

CLINICAL PRESENTATION OF POLYCYSTIC OVARY SYNDROME AMONG SAUDI ARABIAN WOMEN – JEDDAH – SAUDI ARABIA.

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Abstract

Background: Polycystic ovary is a highly variable condition with a wide array of presentations. The Polycystic ovary syndrome should meet at least two of the following three criteria: oligo- or anovulation; clinical and/or biochemical signs of hyperandrogenism; polycystic ovaries on ultrasound. The prevalence of PCO is largely unknown in Saudi Arabia.

Objectives: The aim of this study was to investigate the reproductive hormones levels in patients with PCOS in addition to the effect of age and body mass index (BMI) on the hormonal findings and ultrasound & to determine the clinical, biochemical and etiologic features of hirsutism in Saudi females.

Methodology: A cross sectional study has been conducted among total of 183 patients diagnosed with PCOS have been assessed clinically along with measuring the level of reproductive hormones in Obstetrics and Gynecology Clinic at King Abdul-Aziz hospital and oncology center in Jeddah, Saudi Arabia, between June 2015 and June 2016.

Conclusion: Elevated levels of LH/FSH and testosterone and reduced FSH, SHBG, and progesterone were predictors of PCOS. This was independent of BMI or age. Future studies with larger sample size and data on insulin levels are needed for greater understanding of the manifestation of PCOS in the Saudi population.

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Introduction:-

Polycystic ovary (PCOS/PCOD) is the most common endocrinopathy in women of reproductive age, with a prevalence of up to 10%. It is a complex condition that was first described in women who had PCOS as the underlying cause of hirsutism and chronic anovulation ^[1]. PCOS is a growing epidemic among Saudi Arabian women, and women around the world although is a highly variable condition with a wide array of presentations. In study conducted on Saudi girls, the estimated prevalence of PCOS was observed to be 53.7% which is strikingly higher ^[2].

With the new guidelines for diagnosis of PCOS, The European Society of Human Reproduction and Embryology/American Society for Reproductive Medicine criteria, often called Rotterdam, includes various phenotypes based on a combination of any two of the three findings of hyperandrogenism, menstrual irregularity, and polycystic ovaries on ultrasound^[3]. However, polycystic ovary as a phenomenon is very common in women with idiopathic hirsutism and oligomenorrhea^[4]. Also a wealth of literature agrees that women may present an appearance of PCOS at ultrasound without any sign of androgen excess, although with indicators of ovarian dysfunction^[5]. Similarly, PCOS is diagnosed by exclusion, and disorders having a phenotype related to that of PCOS must be ruled out; such as congenital adrenal hyperplasia, Cushing syndrome and virilising tumors^[6].

The principal findings in patients with PCOS include irregular menstruation, acne, and excessive amounts of androgenic hormones. Obesity is a common finding of women with PCOS^[7-8-9]. PCOS is not merely a disease of the reproductive system, since type 2 diabetes, metabolic syndrome, and sometimes cardiovascular disease have been associated with this condition [9].

Ultrasound evaluation of patients with suspected PCOS in study has been confirmed that presence of 12 or more 2–9 mm follicles appears more sensitive than either ovarian volume or stromal brightness to diagnose PCOS^[2].

In the literature, several groups have investigated the reproductive hormones of women with PCOS in comparison to healthy controls. Follicle stimulating hormone (FSH) and sex hormone-binding globulin (SHBG) were found to be lower in several studies^[10-11-12-13]. In addition, mean concentration of testosterone was higher in patients with PCOS compared to controls^[14]. Furthermore, it has been shown that luteinizing hormone (LH) and LH/FSH ratio were elevated in PCOS patients compared to normal controls^[15-16-17]. The results, however, depended on the day of the cycle on which the hormones are measured, wherein LH was significantly elevated in PCOS patients only late in the menstrual cycle but not earlier^[12].

In Study has been done in 2006 explained that PCOS has three major pathophysiologic hypotheses have been proposed to explain the clinical findings^[18]: the LH hypothesis, the insulin hypothesis and the ovarian hypothesis although the fact that the pathogenesis of PCOS has not clear.

Methodology:-

This study was conducted on women who attended the Obstetrics and Gynecology Clinic at King Abdul-Aziz hospital and oncology center in Jeddah, Saudi Arabia, between June 2015 and June 2016. Women aged between 18 and 45 years were screened for the presence of PCOS.

Study subjects:-

A total of **183** Saudi patients diagnosed with PCOS based on the Revised 2003 Rotterdam Criteria during this study period were included in the study. According to the Rotterdam ESHRE/ASRM-sponsored PCOS consensus workshop group, two out of three of the following must be present to establish diagnosis: Oligo- and/or anovulation, clinical and/or biochemical signs of hyperandrogenism, and polycystic ovaries. Exclusion of other etiologies (congenital adrenal hyperplasias, androgen-secreting tumors, Cushing's syndrome) was conducted. The agreed ultrasound features of PCOS was confirmed by presence of 12 or more 2–9 mm ovarian follicles; a peripheral distribution of ovarian follicles; an ovarian volume of more than 10 cm³ and a highly echogenic ovarian stroma^[2]. For the purpose of this study, all diagnoses were made by a consultant gynecologist in association with a senior ultrasonographer. All participants signed an informed consent form.

Hormonal investigations:-

Blood samples were withdrawn between days 1 and 5 of the period. LH, FSH, estradiol (E2), dehydroepiandrosterone sulfate (DHEA-SO₄), SHBG, total testosterone, prolactin, and progesterone were determined by immunoassay. Serum levels of thyroid stimulating hormone (TSH) and 24-h urinary cortisol were not measured.

Other information:-

Regular menstruation was defined as 9–16 cycles of 21–35 days duration within a year, and no more than a 4-day difference in duration between cycles. The subjects were checked for the presence of acne and/or blackheads on the face, neck, upper arm, chest, and back. Using the modified Ferriman and Gallwey score^[18], the subjects were checked for hirsutism. A Ferriman and Gallwey score higher than 7 indicated hirsutism.

Results:-

The main demographics of cases and controls are presented in Table 1. A total of 183 participants; 77 (42.08%) aged 38 to 45 years old, 71 (19.13%) aged 18 – 27 years old, and 35 (19.13%) aged 28 – 37 years old. In relation to body mass index (BMI) 0 (00%) were under weight, 45 (24.59%) were normal weight, 15 (8.20%) were overweight, 58 (8.20%) were class I obesity, 62 (33.88%) were Class II obesity, and 3 (1.64%) class III obesity. Regarding the clinical presentation (80.33%, 93.44 %, 68.31%, and 20.77%) of the participants were had acne, hirsutism, oligomenorrhea, and infertility respectively. In addition to that 97.81% of the participant has 12 or more follicles measuring 2–9 mm.

Table 1:- Main Demographics Of Cases.

VARIABLES		NUMBER	PERCENTAGE
AGE (YEAR)	18 - 27	71	38.80%
	28 - 37	35	19.13%
	38 - 45	77	42.08%
BMI	< 18.5	0	0.00%
	18.5 - 24.9	45	24.59%
	25 - 29.9	15	8.20%
	30 - 34.9	58	31.69%
	35 - 39.9	62	33.88%
	> 40	3	1.64%
HISTORY OF ACNE	yes	147	80.33%
	No	36	19.67%
FERRIMAN–GALLWEY SCORE	< 7	12	6.56%
	> 7	171	93.44%
HISTORY OF OLIGOMENORRHEA	yes	125	68.31%
	No	58	31.69%
HISTORY OF INFERTILITY	Yes	38	20.77%
	No	145	79.23%
Positive finding OVARIAN FOLLICLE	≥ 12 follicles or more follicles measuring 2–9 mm	179	97.81%
	Ovarian volume of more than 9 cm ³	4	2.18%

To eliminate the potentially confounding bias of age and BMI, we carried out multivariate analyses in Table 2. Indeed, LH/FSH and total testosterone were positively correlated with the disease (regression coefficient = 0.2 and 0.06, respectively, with $P = 0.03$ and 0.02 , respectively). However, FSH, SHBG, and progesterone were negatively correlated with the disease (regression coefficient = -0.02, -0.004, and -0.2, respectively, with $P = 0.05$, 0.02 , and 0.002 , respectively).

Table 2: Age and BMI adjusted comparison of reproductive hormones

VARIABLE	Regression coefficient	P value
LH	0.04	0.18
FSH	-0.02	0.05
LH/FSH	0.2	0.03
E2	0.001	0.6
DHEA - SO4	-0.002	0.92
SHBG	-0.004	0.02
TOTAL TESTOSTERONE	0.06	0.02
PROLACTIN	0.003	0.5
PROGESTERONE	-0.2	0.002
CORTISOL	0.002	0.8

Discussion:-

In this study, we have assessed the clinical presentation along with measuring the level of reproductive hormones of Saudi women diagnosed with PCOS according to the Rotterdam criteria. The importance of our study lies in the fact that studies describing PCOS in the Saudi population are very scarce.

In our studied population most of the participants were 38-45 years old (42.08%) although PCOS can occur throughout reproductive age ^[19]. Other study show that PCOS can occur in younger age group up to 9 years old ^[20]. In addition to that (33.88%) of them were class obesity with BMI between 35 - 39.9 which reflects the fact that obesity is a common finding in PCOS and also in the general Saudi population although obesity is not part of the diagnostic criteria. In the literature, BMI has been suggested to influence the levels of reproductive hormones with some contradictory results. Indeed, some studies found that higher BMI was associated with lower LH ^[21-22-23-24], but others described that BMI had no influence on LH ^[25-26]. On the other hand, recent research has indicated that age can also influence both the clinical presentation and metabolic manifestations of PCOS ^[27-28].

In fact, the results of the multivariate regression analyses confirmed that regardless of the age and weight factor, cases had higher levels of LH/FSH and total testosterone, and that cases had lower level of FSH, SHBG, and progesterone. Even in the adjusted comparison, we could not find a significant increase in LH levels. Unlike the previous published studies ^[15-16], we have failed to find a significant increase in LH in patients with PCOS. A major factor that tends to affect the results is the variation of hormonal level with the menstrual cycle. According to a published study ^[12], elevated LH levels are not very reproducible in the early menstrual cycle, which was the time at which we measured LH in this study. Furthermore, it was previously demonstrated that LH is more elevated in lean PCOS patients compared to obese PCOS patients ^[17] and having most of our patients with BMI >25 (75.41%) could have been a major influence of the results. Several studies in the literature have reported that not all PCOS patients have elevated LH ^[15-16-29], and this could be very well the reason why the average increase in LH observed in our study was not statistically significant. The absence of data on insulin levels has impacted some constraints on the interpretation of the hormonal findings.

Recommendations:-

Future studies with a larger number are required to further clarify the hormonal variations in PCOS. Another limitation is the unavailability of a measurement for insulin level for the study subjects.

Conclusion:-

Elevated levels of LH/FSH and testosterone and reduced FSH, SHBG, and progesterone were predictors of PCOS. This was independent of BMI or age. Future studies with larger sample size and data on insulin levels are needed for greater understanding of the manifestation of PCOS in the Saudi population.

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RESEARCH ARTICLE

THE EFFECTIVENESS OF USING VIRTUAL MATHEMATICS LABORATORIES (VML) IN UBIQUITOUS LEARNING ENVIRONMENTS.

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Abstract

The purpose of this study was measuring the effectiveness of using Virtual Mathematics Lab (VML) to improve teaching and learning of calculus. For learning calculus in VML mathematics classroom is proposed that describes the interactions among the students, VML, and the learning tasks, and how they lead to the learning of calculus. A design experiment was conducted in a class of 50 students from a secondary school. Use of VML was integrated into the teaching and learning of calculus concepts in the classroom with the aid of Acadox environment Navigator, a wireless classroom network system that enables instant and active interaction between students and teachers. It was found that the appropriate use of graphical, numerical and algebraic representations of calculus concepts using VML enabled students to better visualize the concepts and make generalizations about relevant mathematical properties. In addition, the students were able to link multiple representations, especially algebraic and graphical representations, to improve their conceptual understanding and problem-solving skills. VML was used as an exploratory tool, graphing tool, confirmatory tool, problem-solving tool, visualization tool and calculation tool. This suggests that VML is a multi-dimensional tool that supports mathematics learning. Overall, the findings of the study indicate that VML is an effective tool to develop mathematical concepts and promote learning and problem solving.

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Introduction:-

The graphing calculator has been widely used as a technological tool in teaching and learning mathematics. There is a growing body of research which shows that the pedagogical affordances of the graphing calculator have a close relationship with improving learning of mathematics. Indeed, mathematics can be taught in a more coherent way by providing students with the opportunities to connect mathematical concepts within and between topics, and here the graphing calculator is well-positioned to offer a learning environment that enables mathematics to be experienced through multiple representations [1].

The curriculum standards of the National Council of Teachers of Mathematics[2] Suggest the use of graphing calculators to provide students with multiple representations to explore and investigate mathematical concepts. Extensive research has been conducted to answer questions such as whether graphing calculators significantly

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improve mathematical learning and problem solving [3]. However, many such studies have compared the use of graphing calculators to traditional chalk-and-talk learning, and little research has examined how students use graphing calculators in learning and the relationship between pedagogical strategies for learning and the use of graphing calculators.

Chu & et al. Out an extensive review of research on graphing calculators, they found that certain teaching styles, such as interactive and inquiry oriented, are more compatible with the use of graphing calculators than others. However, they also point out that many studies fail to address the use of graphing calculators in an instruction context, which leads to inconsistent findings on their effectiveness in teaching and learning. This review suggests a need for studies that address the use of graphing calculators in an instruction context within a particular learning environment [4].

Accordingly, the purpose of this study was to investigate how the use of VML, an advanced graphing calculator, could enhance teaching and learning of calculus. This article provides the details and results of a designed experiment, which involved the design and conduct of a VML for a class of 50 Secondary students. Use of VML was integrated into the teaching and learning of calculus concepts with the Acadox environment Navigator

Conceptual Frameworks:-

In the literature of mathematics reform, learning mathematics is viewed as a social endeavor, with the mathematics classroom functioning as a community in which thinking, presenting, talking, agreeing and disagreeing are encouraged [5]. Mathematical tasks are given to students to develop and extend their mathematical thinking and problem-solving ability. Student interaction with mathematical tasks and tools encourages them to construct mathematical ideas. Hence, create meaning for their learning.

Hwang Et Al. [6] observed that students should construct meaning from the tools that they use and the mathematical tasks in which they are engaged. They elaborated that learners construct mathematical meanings from tools as they use them. This leads to meaningful learning, as the tools become intertwined with the mathematical tasks. Bruce [7] pointed out that the quality of mathematics tasks is important in determining whether a task engages students and whether they attain higher order learning by being given opportunities to explain and justify their reasoning.

It is widely acknowledged that calculus concepts are abstract and complex for students and that teaching and learning these concepts can be challenging and even exasperating at times. Research has shown that many students have difficulties in learning the key concepts of calculus. It does not help that traditional calculus courses tend to focus on algebraic drills and the practicing of calculus problems without generating an understanding of the underlying concepts.

Specifically, in terms of calculus, many researchers have advocated that teaching and learning should focus on concepts, rather than simply techniques to improve student performance. Tall emphasized the need to help students move flexibly from one representation to another in learning calculus. Using a graphing calculator extends the teaching and learning of calculus concepts from being based solely on algebraic methods to the incorporation of numeric, algebraic and graphical representations. The idea of using the graphing calculator to provide a better connection between algebraic and graphical representations of calculus has been reported in several studies.

It is believed that the use of technology in the teaching of calculus can be beneficial if it is accompanied with appropriately designed activities. Lazonderet al.[8] highlighted that mathematical tools are important in helping students to make connections to build their mathematical understanding. Activities such as guided discovery and inquiry learning have been used in the design of worksheets to encourage interaction with graphing calculators to help students develop an understanding of calculus concepts.

In this study, many activities using Acadox environment were designed by the author, with inputs from the teacher of the participating class, hereafter the participating teacher, to provide students with opportunities to explore, generalize mathematical concepts, to help them develop confidence and ability in communicating mathematically. The preliminary framework for the use of VML in the intervention programme is an adaptation.

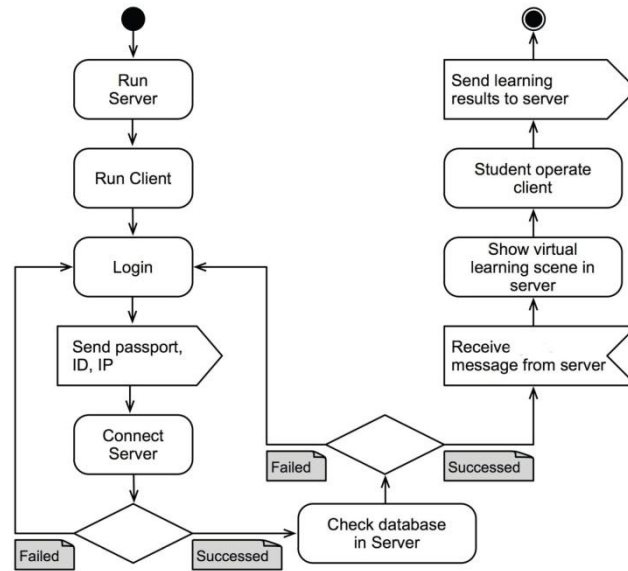


Fig. 1:- the steps to enter the student for Acadox environment.

A conceptual framework for calculus learning and teaching using Acadox environment was devised based on the foregoing literature review, and is illustrated in Fig. 2 This study seeks to the interactions among the students, VML, and the learning tasks lead to the learning of calculus. It describes Student Guided discovery and inquiry-based design learning tasks VML with acadox environment

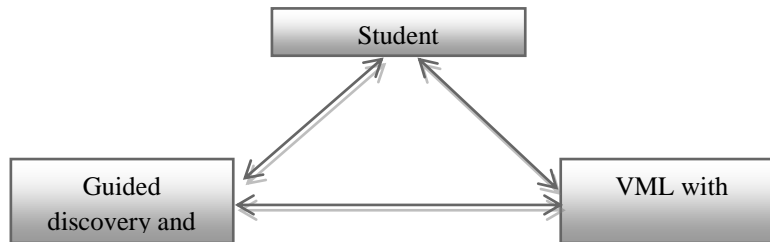


Fig. 2:- Conceptual framework.

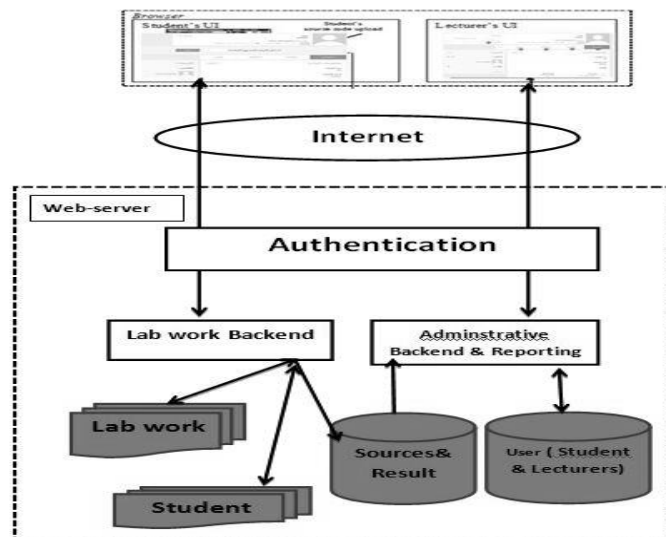


Figure 3:-Architecture of the setting up the VCL in acadox environment.

Methodology:-

This study employed a designed experiment methodology that involved the preparation, design, implementation and retrospective analysis of a teaching experiment. A designed experiment is an iterative process that features cycles of invention and revision. The participating teacher worked in close partnership with the author to incorporate the use of VML in lessons, examining the strengths and limitations of existing pedagogy and practice and acknowledging areas for future development with the VML.

Research Design:-

The subjects of the study were 50 students from a secondary school. Each student was trained before the start of the VML. The purpose of the training sessions was primarily to introduce the interface design and to train the participating students in the use of the algebraic, graphical and numerical functions of VML while engaging the students in inspiring activities involving mathematical problem solving and exploration with the acadox environment.

To determine more precisely, the role of VML in teaching and learning calculus in the participating class, a calculus package focusing on differentiation and integration that integrated VML was designed. The topics covered were Differentiation by First Principles, Equations of the Tangent and the Normal, Rate of Change, Stationary Points, Derivatives of Trigonometric Functions, Derivatives of Exponential and Logarithmic Functions, Integrations and Indefinite Integrals, Definite Integrals, Integration of Trigonometric Functions, Integration of Exponential Functions and $1/x$, and Area of a Region. It starts with a known pattern: a parabolic, sine or circular. The curves will be shaped by the graphical effect that corresponds to the presence of the parameter k in the algebraic expression. Different forms and graphical effects are included on considering the mathematical functions listed in Table 1.

Table 1:- Functions and graphical effects considered in the application.

Graphical effect	Parabola	Sine	Circle
Original	$y = x^2$	$y = \sin x$	$y = \sqrt{1 - x^2}$
Vertical scroll	$y = x^2 + k$	$y = \sin x + k$	$y = \sqrt{1 - x^2} + k$
Horizontal scroll	$y = (x + k)^2$	$y = \sin(x + k)$	$y = \sqrt{1 - (x + k)^2}$
Contraction	$y = kx^2$	$y = k \sin x$	$y = k\sqrt{1 - x^2}$
Expansion			
Other		$y = \sin kx$	$y = \sqrt{R^2 - x^2}$

The VML learning activities focused on introducing mathematical concepts through exploration using VML. Students were encouraged to examine the setting of a given problem or to manipulate the parameters of a simulation in a VML document that was

d

$$dx (\sin x) = \cos x \quad (1)$$

Notes:-

- Use VML to verify the above results. The gradient of the tangent line to $y = \sin(x)$ at a given point gives the value of the derivative of $y = \sin(x)$ at that point.
- Use the tangent line to trace an approximate graph of the derivative of $y = \sin(x)$. The slope of the tangent is shown at the top right of the screen. Your goal is to move the tangent line by grabbing and dragging the point of tangency such that it is positioned directly above each of the 13 points, beginning with the leftmost point.
- Inspect the resulting scatter plot, which represents the approximate graph of the derivative of the sine function. Write down your observations.
- Use the Graphs and Geometry application to verify your answer.

They were asked to formulate conjectures and examine and confirm those conjectures, culminating in the derivation of a formula, the generalization of the results or the summarization of the underlying concepts. A worksheet was designed to accompany these activities to facilitate guided discovery and inquiry learning.

In the second category, the students were assigned tasks to reinforce their conceptual understanding using VML. In a typical activity, an exercise consisting problems related to the concept being taught was assigned and students were required to complete the exercise with the aid of VML.

Occasionally, a formative assessment using the 'Quick Poll' function of ACADOX Navigator, which allows the teacher to gather instant feedback from students through the network, was conducted to assess student understanding. Where time permitted, students were invited by the participating teacher to share their solutions or ideas through the use of the 'Presentation' function of ACADOX Navigator. VML Navigator was also used by the participating teacher to monitor the progress of the students in the in-class tasks by periodically viewing their ACADOX screens.

Acadox presents the following functionalities:-

- **Web-intercom:-** This functionality allows the user take part in chats within the 3D environment. The records of such chats can be viewed both in the virtual environment and in Acadox server. These records are stored in Acadox's database.
- **Registration booth:** This functionality allows relating student's avatars to their corresponding Acadox's accounts. It is required because students frequently have different names for their accounts and their avatars which can pose some problems when recording their activities.
- **Quiz tool and 3D Drop Box:** This functionality allows the students to execute tasks involving the creation of objects in the virtual environment. The objects created are then transferred to Acadox similar to the way a file is uploaded.
- **Multi-function Toolbar:** This functionality allows the user to activate the blog development function within the virtual environment. Such blogs can be viewed both in the virtual environment and in Acadox.
- **Presenter:** This functionality allows the users to post in the virtual environment, slideshows or videos that can be viewed or downloaded from the Acadox server.
- **Quiz Chair:** This functionality allows the students to answer prepared questions in Acadox, which also stores the answers.

All of the activities and accompanying worksheets were designed after thorough discussion with the participating teacher who then co-taught the calculus package with a research assistant of the project who assisted the participating teacher with the use of VML and its Navigator, Activities such as guided discovery.

Data Collection Instruments:-

A questionnaire was used to measure the quality of the suggested system in a simple way. To make sure of clarity of statements, they were adjusted by some professional professors.

Data Analysis:-

To analyze the questionnaire statements, SPSS19 was used to extract mean M, standard deviation SD and T-test to regard the studied statements as qualitative data for knowing if there is a relationship between science laboratories and their support to the educational process.

Expert's attitudes:-

The use of SPSS software to make a statistical analysis of the terms of the views of expert's form towards the use of VML system to conduct ,Experiments and the results were as follows:

Table 2:- Descriptive statistics of Expert's attitudes toward the VML system.

NO	statements	N	Minimum	Maximum	Mean	Std. Deviation
1-	The application provides an interactive environment for students.	30	1	2	1.20	0.407
2-	Application to embody the abstract scientific concepts helps.	30	1	2	1.07	0.254
3-	Application to improve the level of understanding among students help,	30	1	3	1.13	0.434
4-	This application was designed in an interesting and attractive.	30	1	2	1.10	0.305
5-	Application of self-learning skills of the learner grown.	30	1	3	1.20	0.484
6-	This application helps in achieving educational goals.	30	1	2	1.30	0.596
7-	Application to get good educational outcomes able to use developments in educational technology helps.	30	1	3	1.17	0.461
8-	The application software provides simulated reality.	30	1	2	1.13	0.346
9-	The application meets the needs of the learner.	30	1	3	1.27	0.583
10-	Application to increase the motivation of Hamas helps the learner to learn.	30	1	2	1.17	0.379
11-	The application provides an atmosphere of privacy for students.	30	1	3	1.17	.0461
12-	The application makes an active and positive student.	30	1	2	1.03	0.183
13-	Application on the continuous training of student helps which helps workmanship.	30	1	2	1.03	0.183
14-	Easy to use for student application.	30	1	2	1.07	.0254
15-	The application provides interactive features with the students.	30	1	2	1.03	0.183

"The application provides an interactive environment for students" (M=1.20, SD=0.407), "Application to embody the abstract scientific concepts helps." (M=1.07, SD=0.254), "Application to improve the level of understanding among students help" (M=1.13, SD=0.434), "This application was designed in an interesting and attractive." (M=1.10, SD=0.305), "Application of self-learning skills of the learner grown." (M=1.20, SD=0.484), "This application helps in achieving educational goals." (M=1.30, SD=0.596), "Application to get a good educational outcomes able to use developments in educational technology helps." (M=1.17, SD= 0.461), "The application software provides simulated reality." (M=1.13, SD= 0.346), "The application meets the needs of the learner." (M=1.27, SD= 0.583), "Application to increase the motivation of Hamas helps the learner to learn." (M= 1.17, SD= 0.379), "The application provides an atmosphere of privacy for students." (M=1.17, SD=0.461), "The application makes an active and positive student." (M=1.03, SD= 0.183), "Application on the continuous training of student help which helps workmanship." (M= 1.03, SD=0.183), "Easy to use for student application." (M=1.07, SD= 0.254), "The application provides interactive features with the students." (M=1.03, SD=0.183).

● **The views of students' attitudes:** A field study focused on the importance of VML system in the laboratory of traditional real to the stage of a secondary school where the number of students involved in this experiment are 50 students of a secondary school simulation was conducted. Data were collected by the views of a questionnaire to gauge the views of students about the use of the proposed VML system includes wage trial on four steps:

The first step: Students have been creating content for laboratory experiments by having competent to rule clarification and guidance to the students' action steps prescribed by the laboratory tests that were true through most of the meeting between the competent and students.

The second step: The students were trained on how to use the VML and guide students in dealing with the system pragmatic system pragmatics, by training students on how to enter the system, and keep track of work experience measures and how to move from a chemical experiment to another.

The third step: The students to re-do the experiments using the proposed application. The role of the researcher and competent to rule at this stage, is to follow the progress of students in work experience, and providing assistance to them, and answering questions raised by students during the experiment At the end of each experiment, there was a discussion with the students about what is implementation.

Step Four: A questionnaire was distributed to measure the views towards the use of VML system to conduct experiments.

B. Results:

The use of SPSS software to make a statistical analysis of the terms of the views of students form towards the use of VML system to conduct experiments and the results were as follows:

Table 3:- Descriptive statistics of student's attitudes toward the VML system.

NO	statements	N	Minimum	Maximum	Mean	Std. Deviation
1-	VML is a new teaching method	50	1	2	2.62	0.490
2-	Help install information.	50	1	2	2.48	0.505
3-	Easy to use.	50	1	2	2.60	0.495
4-	Makes the learner's active and positive.	50	1	2	2.48	0.677
5-	It provides the use of a suitable environment.	50	1	2	2.38	0.667
6-	Provide interactive experiences.	50	1	2	2.56	0.611
7-	Offer to extract information.	50	1	2	2.56	0.577
8-	Take into account individual differences.	50	1	2	2.44	0.611
9-	It provides an opportunity to work outside the formal learning tools.	50	1	2	2.60	0.571
10-	It helps to learn many skills in a short time.	50	1	2	2.72	0.454
11-	It provides an opportunity to end the learning process according to your own pace and his ability to learn.	50	1	3	2.56	0.577
12-	Grown self-learning skills.	50	1	2	2.62	0.490
13-	This application is an advanced technical system and is important to face the challenges of the times.	50	1	2	2.72	0.454
14-	The application provides the privacy of student.	50	1	2	2.34	0.626
15-	Application to understand the information deeper help.	50	1	2	2.38	0.753
16-	It helps to learn and absorb mathematics.	50	1	2	2.64	0.485
17-	Encouraged to overcome the negative reception of information to active participation in education.	50	1	2	2.54	0.579
18-	Encourages the application on the stimulus and preview the information and control.	50	1	2	2.60	0.571

The Alpha physical coefficient was calculated about student attitudes towards VCL system, It gave the highest score of acceptance ($\alpha=0.898$). It was also found that students showed their acceptance that VML is a new teaching method ($M=2.62$, $SD=0.490$) and that Help install information. ($M=2.49$, $SD=0.505$) and that easy to use. ($M=2.60$,

SD=0.495). Makes the learner's active and positive. (M=2.48, SD=0.677) and It provides the use of a suitable environment. (M=2.38, SD=0.667). Provide interactive experiences. (M=2.56, SD= 0.611).

Students also agreed that Offer to extract information. (M=2.56, SD=0.577). There was also taking into account individual differences. (M=2.44, SD= 0.611), It provides an opportunity to work outside the formal learning tools. (M=2.60, SD=0.571) and It helps to learn many skills in a short time. (M=2.72, SD=0.454).

It also provides an opportunity to end the learning process according to your own pace and his ability to learn. (M=2.56, SD=0.577), It also Grown self-learning skills. (M=2.62, SD=0.490). This application is an advanced technical system and is important to face the challenges of the times. (M=2.72, SD=0.454). The application provides the privacy of student (M=2.34, SD=0.626). Application to understand the information deeper help (M=2.38, SD=0.753). It helps to learn and absorb mathematics. (M=2.64, SD=0.485). Encouraged to overcome the negative reception of information to active participation in education. (M=2.54, SD= 0.579). Encourages the application on the stimulus and preview the information and control (M=2.60, SD= 0.571).

Comparison between RML and VML:-

The comparison of Real Mathematic Lab (RBL) and Virtual Mathematic Lab (VBL) the terms of learning performance and the factor of learning time.

Comparison of learning performance:-

In this selection, we want to understand whether or not there is a difference between learning methods. This research employs a T-test to analyze the relation between students' "pre-examination achievement", "post-examination achievement" and "learning model (VML)".

Table 4:- Results of the t-test between pre-test and post-test regarding Comparison of learning performance.

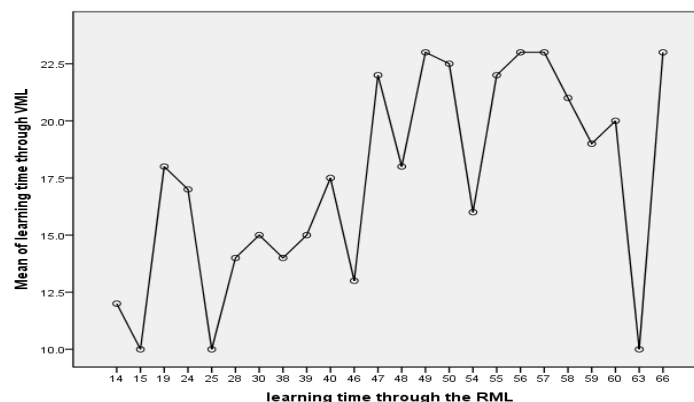
Group	Mean	Standard deviation	t-test
Pre-test	11.87	3.09	5.685
Pro-test	14.91	2.27	

Table 4. Shows that the results of the t-test shows there was a statistically significant difference between the pre-test (M= 11. 87) and the post-test (M = 14. 91). In terms of VML application, where we find T-TEST = 5.685

Learning Time:

In this selection, we want to understand whether or not there is a difference learning time between in RML and VML, This research employs an ANOVA to analyze the relation between VML, RML

ANOVA					
Table 4. ANOVA result for assessment learning time between in RML and VML					
	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	568.367	23	24.712	.982	.562
Within Groups	151.000	6	25.167		
Total	719.367	29			



On average, there was a significant difference in the percentage learning time, where we find that the average learning time through the RML (Mean= 44.47), while we find learning time through VML (Mean= 17.03), from indicates the effectiveness of the system in reducing the learning time.

Discussions:-

The results of the study support the research hypothesis that the new mathematical experiences afforded by VML offer students a rich setting to explore calculus concepts. The dynamic features of graphing calculators, such as multiple representations of mathematical concepts, if used appropriately, can improve students' ability to make the connection between graphical and algebraic representations. This feature was found to be particularly useful in teaching and learning calculus.

Many ways of using VML as a tool in the mathematics classroom were identified in this study. In the Simulation programme, VML was used as an exploratory tool, graphing tool, confirmatory tool, problem-solving tool, visualization tool and calculation tool. Underlying these uses was the pedagogy of the design of the mathematical tasks and the interaction between the students and the tasks designed for use with VML.

Zacharia et al. [9] theorized four roles for technology in relation to teaching and learning interactions: master, servant, partner and extension of self. In the current study, uses of VML as a confirmatory tool, a calculation tool or a graphing tool by the students certainly have characteristics of the servant role while uses of VML as an exploratory tool, a problem-solving tool or a visualization tool illustrate using the technology as a partner. It was evident that students in this study were able to use VML beyond the level of using it as a master as it was clear that the students were not 'subservient to the technology'. However, there was insufficient evidence to suggest that they could use the technology as an extension of self, the highest level of functioning which involves 'users were incorporating technological expertise as an integral part of their mathematical repertoire'. On the other hand, Lampi [10] found five patterns and modes of graphing calculator tool use: computational tool, transformational tool, data collection and analysis tool, visualizing tool and checking tool. While students in this study used VML as a computational tool in evaluating numerical expressions, and as a checking tool in confirming conjectures.

Analysis of the interviews with the students from the participating class on the Concept of the derivative at a point revealed that the use of VML, and particularly its emphasis on visual and numerical representations of the derivative, affected the students' conceptual development. Students interviewed were able to find the derivative of a given function using a visual conception of the derivative that they had learned in the intervention programme. This finding suggests that the students were able to form connections between the different representations of the derivative in calculus using VML, which helped them to develop a conceptual understanding of the derivative. The conventional teaching and learning of mathematics over-emphasizes algebraic methods, and the students had been trained to use such methods to solve most mathematics problems. However, the use of a graphing calculator can change the teaching and learning of calculus concepts from relying solely on algebraic method to encompassing numeric, algebraic and graphical methods, as indicated by Elizondo [11]. Several studies have shown that the connection of International Journal of Mathematical Education in Science and Technology different representations leads to the development of conceptual understanding and the improvement of students' problem-solving skills [12].

It was found that some of the students claimed that they have been taught mainly using algebraic methods since they were young. Thus, the predominant approach that they used in problem solving remained algebraic even after using VML, although they saw the usefulness of the graphical approach in solving problems and understanding concepts better. Some even claimed that they only realized the accuracy and efficiency of the graphical method in problem solving after the intervention programme. These findings suggest that the use of a graphing calculator should be integrated into mathematics teaching and learning at an early stage, such as Secondary Stage.

Teachers play an important role in conventional mathematics teaching and learning in the classroom. Most of the time, they are in full control of the teaching and learning situation and act as the knowledge provider. However, the teacher's role changes when VML is used in the classroom from an instructor to a learning facilitator. Students play an active role in the learning process, and construct their learning through guided discovery and inquiry-based learning tasks that they can then share using VML Navigator.

The visualization aspect of VML engendered a better conceptual understanding through demonstrating graphs and simulations. In addition, the mathematical tasks were designed in a way that facilitated students' learning. The

conventional mode of learning that focuses on the regurgitation of what teachers have taught does not necessarily allow students to gain a full understanding of concepts, which are of utmost important in calculus.

Conclusions:-

The findings from this research demonstrate the potential of VML as a tool to promote learning in mathematics. The encouraging results recommend the use of the graphing calculator to provide multiple representations for mathematics learning and problem solving. A framework is also proposed that shows the importance of the interactions among the students, VML, and the learning tasks in the learning of calculus. In the future, VML could be used to stimulate students to think mathematically so that they will engage strongly with mathematical structures and concepts in ways that are not possible with traditional paper and pencil approaches. However, before it is used for this purpose, training should be provided so that teachers and students can become familiar with the features and functions of VML as a pedagogical tool.

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RESEARCH ARTICLE

STRUCTURAL AND PHOTOLUMINESCENCE CHARACTERIZATION OF $\text{MgAl}_2\text{O}_4\text{:Eu}$ PHOSPHOR SYNTHESIZED BY COMBUSTION METHOD.

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Abstract

The MgAl_2O_4 spinel doped with Eu^{2+} ions powder phosphor was prepared at temperature as low as 500 °C using the combustion route. A structural property of the powder was characterized by X-ray diffraction (XRD). This XRD pattern shows the well crystallized cubic phase, Fd-3m space group of MgAl_2O_4 . The estimated average crystalline size is about 36 and 30 nm for MgAl_2O_4 and Eu doped MgAl_2O_4 particles respectively. The photoluminescent property of prepared powder was investigated using excitation and emission spectroscopy at room temperatures. Energy levels scheme is proposed for emission from $\text{MgAl}_2\text{O}_4\text{:Eu}^{2+}$.

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Introduction:-

In recent years, rare earth ion doped inorganic nanomaterials have received significant attention in the field of science and engineering [1, 2]. Inorganic phosphors doped with rare earth ions have been used in lighting systems and for multicolor displays to overcome the drawback such as poor color rendition, high toxicity and complicated synthesizing process [3,4]. Among them, magnesium-aluminum oxide (MgAl_2O_4) spinel crystals have received a great deal of attention due to its mechanical strength, high resistance to chemical attacks, and its outstanding dielectric and optical properties [5]. Europium (Eu) doped in host can improve the photoluminescence properties of $\text{MgAl}_2\text{O}_4\text{:Eu}^{3+}$ phosphors [6]. $\text{MgAl}_2\text{O}_4\text{:Eu}^{3+}$ phosphors have been prepared by various methods such as solid-state reaction, sol-gel, spray-drying, co-precipitation, microwave assisted hydrothermal method and combustion synthesis. Among the methods combustion is a simple method which requires low temperature and less time as compare to other one. In the combustion process, the phosphors can be synthesized within 5 minutes at low temperatures. In the present investigation we have reported the synthesis of Eu^{2+} activated MgAl_2O_4 ($\text{MgAl}_2\text{O}_4\text{:Eu}^{2+}$) and it was characterized by using techniques such as powder X-ray diffraction (XRD), and photoluminescence (PL).

Experimental:-

In the present study analytical grade reagents were used without any further purification. $\text{MgAl}_2\text{O}_4\text{:Eu}^{2+}$ was prepared by combustion technique [7] using precursors Magnesium Nitrate ($\text{Mg}(\text{NO}_3)_2$), Aluminium Nitrate, ($\text{Al}(\text{NO}_3)_3 \cdot 9\text{H}_2\text{O}$) and Urea ($\text{NH}_2\text{CO.NH}_2$). Metal nitrates are used as oxidizers, and urea is employed as fuel. Starting materials ($\text{Al}(\text{NO}_3)_3 \cdot 9\text{H}_2\text{O}$) and $\text{NH}_2\text{CO.NH}_2$ were taken in the stoichiometric ratios to crushed and ground in an agate mortar to form gel-like paste with the help of distilled water. The china dish containing the mixture was inserted in a vertical furnace preheated at 500 °C. Initially, the paste melts and undergoes dehydration followed by decomposition with the evolution of large amounts of gases. The mixture then foamed and finally

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ignited, yielding a white fluffy powder, which was collected after removing the dish from the furnace. The structural characterization of the product was performed by an X-ray diffractometer (Rigaku rotating anode H-3R). Photoluminescence spectra at room temperature were recorded in the range 200-750 nm on Spectrofluorophotometer (Shimadzu RF-5301 pc).

Results and Discussions:-

Structural Characterizations:-

The phase composition of the synthesized powder was analyzed by XRD using Cu-K α radiation (1.54184 \AA) in the 2θ range of 10° to 80° . Fig. 1 shows the XRD pattern of the as-prepared MgAl_2O_4 sample and Eu doped MgAl_2O_4 sample. The observed diffraction peaks correspond to those of the standard patterns for cubic MgAl_2O_4 (JCPDS, No.77-0435). The XRD data of sample is very similar to standard pattern and there are no additional peaks observed for the doped MgAl_2O_4 materials. This pattern indicates that the well crystallized cubic phase, Fd-3m space group of MgAl_2O_4 was obtained by the urea-assisted combustion method even at furnace temperature 500°C . The XRD pattern of this phosphor contains three phases: the main spinel type MgAl_2O_4 phase (Fd3m space group) and some additives of MgO (Fm3m space group) and $\alpha\text{-Al}_2\text{O}_3$ (space group R3c). Eight diffraction peaks can be indexed as the (111), (2 2 0), (3 1 1), (4 0 0), (4 2 2), (5 1 1), (4 4 0) and (5 3 3) diffraction lines of high intensities. A stronger peak (3 1 1) at 2θ of around 36° is used to calculate the average crystalline size (D) of the MgAl_2O_4 and Eu doped MgAl_2O_4 particles.

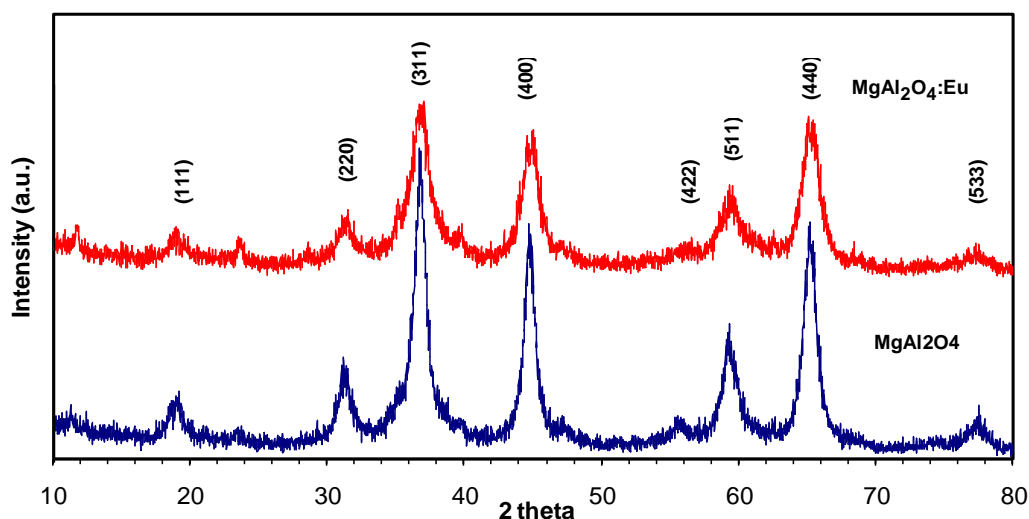


Fig. 1:- X-ray diffraction of MgAl_2O_4 and $\text{MgAl}_2\text{O}_4:\text{Eu}^{2+}$ phosphor powder.

The average crystallite size was estimated from the full width at half maximum (FWHM) of the diffraction peaks by using the Scherrer formula [8]

$$D = \frac{0.9\lambda}{\beta \cos\theta}$$

The estimated average crystalline size is about 36 and 30 nm. It has been reported that the particle size of MgAl_2O_4 spinel synthesized by various methods have been in the range of 43–55 nm [9].

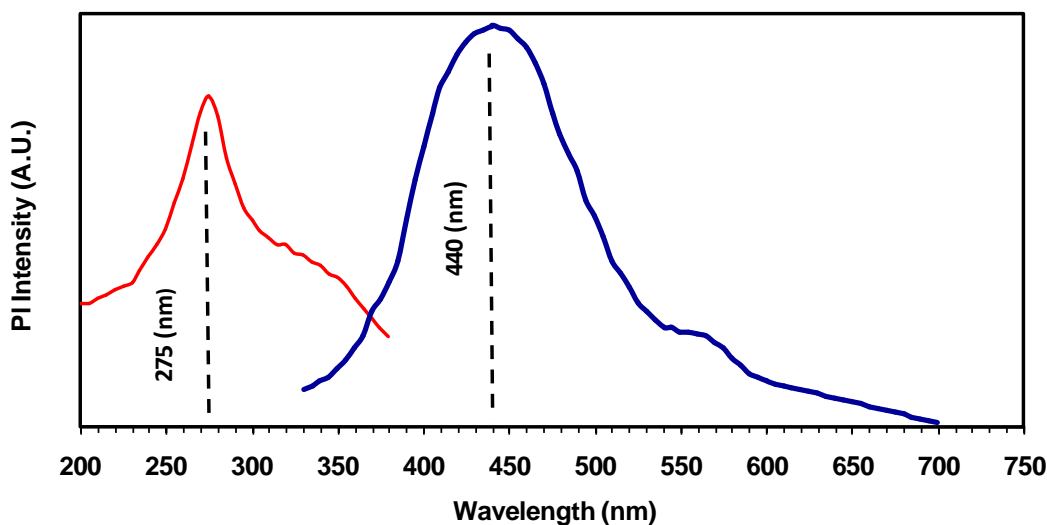
Photoluminescence studies:-

Fig. 2:- shows the photoluminescence spectrum.

Fig. 2 shows the photoluminescence spectrum (excitation and emission spectra) of Eu ions incorporated in MgAl_2O_4 ranging from 200 to 750 nm. Excitation spectra of $\text{MgAl}_2\text{O}_4:\text{Eu}$ materials prepared at 500°C were recorded under emission monitored at 440 nm. The excitation spectra display intense peaks at 275 nm would be due to the defects within the host material [5].

From the spectra it can also be noted that the broad emission peak is located at 440 nm, which could be attributed to the emission of Eu^{2+} ion. The short-wavelength part of the spectrum, bluish green broad band emission at 430–570 nm, is attributed to the $4f^65d^1 \rightarrow 4f^7(^8S_{7/2})$ transitions of Eu^{2+} [10,11]. Europium is the most versatile among all lanthanides. It occurs more often in the divalent than trivalent state, which is in the contrast to all the other lanthanides. The high stability of the divalent state originates from the stability of the half filled shell configuration ($4f^7$) for Eu^{2+} . The valence state of europium has a great influence on its luminescence properties. The 5d-4f transitions of Eu^{2+} emissions are parity allowed. The luminescence of divalent europium consists mostly of broad intense band emission in the visible spectral range. The ground state of the $4f^7$ configuration of Eu^{2+} is $^8S_{7/2}$, while the lowest $4f^7$ excited state is the 6P_J state in the UV region. In most Eu^{2+} activated phosphors, the lowest level of the $4f^65d$ configuration lies below the 6P_J state, leading to broadband luminescence due to the inter-configurational electronic $4f^65d - 4f^7 (^8S_{7/2})$ transition [12].

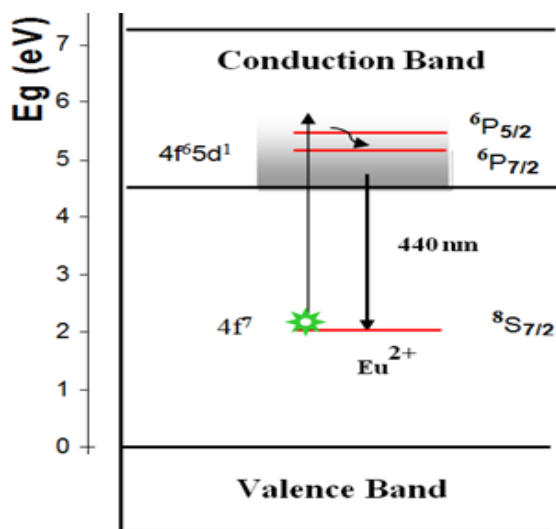


Fig.3:- Proposed energy levels scheme for emission from $\text{MgAl}_2\text{O}_4:\text{Eu}^{2+}$

On excitation with UV, two types of lowest excited states are possible, namely 6P_j (f-f) or $4f^65d$ (f-f), depending on the host matrix [13]. In case of Eu^{2+} ion in MgAl_2O_4 the possible lowest excited states is $4f^65d$ (f-f) [5]. The optical band gap energy value of magnesium aluminate is estimated as 4.5 eV [14]. Based on above discussion the possible emission mechanisms from the Eu^{2+} activated MgAl_2O_4 is illustrated in Fig 3.

Conclusions:-

The nanocrystals of the $\text{MgAl}_2\text{O}_4:\text{Eu}^{2+}$ spinel was prepared via combustion method. Present investigation revealed that urea-nitrate combustion synthesis has outstanding potential for producing Eu-doped MgAl_2O_4 phosphors in a single step. Nanopowder demonstrated a strong and quite short lived luminescence, characteristic for the doped Eu^{2+} ions. The broad emission peak at 440 nm is attributed to the usual near UV emission of Eu^{2+} ion from the $4f^65d^1 \rightarrow 4f_7({}^8S_{7/2})$ transition.

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RESEARCH ARTICLE

DECLINED MORPHOLOGY IN TOBACCO (*Nicotiana tabacum* L.) DUE TO CADMIUM STRESS.

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Cadmium, tobacco and morphological parameters.

Abstract

Cadmium is one of the most toxic heavy metal in the environment. Cadmium presence in soil can cause serious problem to all organisms through food chain. The present study was conducted through pot culture experiment to estimate the effect of cadmium (10, 25, 50, 75, 100, & 200mg kg⁻¹ soil) on the morphological parameters (root length, shoot length, number of leaves, total leaf area, fresh and dry weight of root, stem, leaf) of tobacco. The values were recorded on 30th, 60th, 90th, 120th, 150th and 180th days of tobacco. For all the morphological parameters, the highest values were recorded in control. Due to increase in cadmium concentrations all the morphological parameters were decreased from 10mg kg⁻¹ to 200mg kg⁻¹ of soil.

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Introduction:-

Due to globalization and industrialization, heavy metal (HM) pollution is one of the most important and widespread environmental problem of the modern world, due to its hazardous and toxic effects on all living organisms. Heavy metal pollution is a serious environmental problem because heavy metals are non-biodegradable and persistent in nature [10,11].

Heavy metals are defined as a group of metals and metalloids with atomic density higher than 5 g cm⁻³. The remediation of heavy metal contaminated soil requires the removal of toxic metal from contaminated areas. Heavy metals were important environmental pollutants particularly in areas where there was a high anthropogenic pressure, but they also occur naturally (Sanità and Gabbrielli, 1999). Anthropogenic cadmium contamination often results from mining or smelting of metal ores, but cadmium is also released into the environment by power stations, heating systems, waste incinerators, urban traffic, and cement factories and as a by-product of phosphate fertilizers. Use of sewage sludges as fertilizers has further contributed to a significant contamination of agricultural soils. Cadmium presence in the soil can cause serious problems to all organisms. Although non-essential for plant growth, when bio available, cadmium is readily taken up by roots and translocated into aerial organs where it can accumulate to high levels. The most apparent visible symptoms of cadmium toxicity in plants are retardation of plant growth, chlorosis and stunting (Das *et al.*, 1997). The present investigations was carried out to analyse the changes in growth parameters such as, root and shoot length, number of leaves and total leaf area, fresh and dry weight of root, stem, leaf of tobacco plants due to cadmium toxicity.

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Materials and Methods:-

Seed Materials:-

The seeds of tobacco were procured from Central Tobacco Research Institute, Research Station, Vadasandur, Dindugal Dt, Tamilnadu. The duration of the crop was 180 days. Seeds of uniform size, colour and weight were chosen for the experiments. Cadmium and other chemicals used in this investigation were analytical grade. Cadmium was used in the form of chloride.

Pot Culture Experiments:-

The experiments were performed during the months of December 2011 to May 2012. Tobacco plants were grown in pots in untreated soil (control) and in soil to which cadmium had been applied (0, 10, 25, 50, 75, 100 and 200 mg kg⁻¹ of soil). The inner surface of pots was lined with a polythene sheet. Each pot contained 3.00 kg of air dried soil. Cadmium as chloride (CdCl₂ 2½ H₂O) were finely powdered and applied to the surface soil. This was then thoroughly mixed with the soil and 1 gram of seeds was sown in each pot. All pots were watered to field capacity twice a day. Plants were thinned to a maximum of three per pot after a week of germination. The treatments were replicated five times in a completely randomized block design.

Sampling:-

Plant samples were collected at random, at regular intervals (30, 60, 90, 120, 150 and 180th days) and used for morphological growth parameters. Three plants from each replicate of a pot were analysed for its various parameters and the average was calculated. These mean values of the replicates were used for statistical analysis.

Growth:-

Root and Shoot Length:-

The root and shoot length and elongation rate are essential for plants exploring for water and mineral nutrients. In the pot culture experiments there was a gradual decrease in the root and shoot length with an increase in cadmium level 10, 25, 50, 75, 100 and 200 mg kg⁻¹ in the soil in all the sampling days. The inhibitory action of excess of cadmium in root and shoot length might be due to reduction in cell division, toxic effect of heavy metals on photosynthesis, respiration and protein synthesis. These obviously contributed to the retardation of normal growth Kupper, et al. (1996). The above results were in agreement with the findings of Chen, et al. (2003) in soybean, Rai, et al. (2005) in *Phyllanthus amarus* and Xu, et al. (2008) in garlic. Hagemeyer et al. (2002.) and Marciano et al. (2002) also suggested that the morphological and structural effects caused by metal toxicity in plants was due to decrease in root elongation, root tip damage, decrease in root formation, suppression of elongation growth rate of cells, affecting the ultra cellular structure of meristematic cells and inhibition of the size of plant cells and inter cellular spaces.

Number of Leaves and Total Leaf Area:-

Number of leaves and total leaf area were decreased due to the increase of cadmium concentrations in all the sampling days. Number of leaves, branches and leaf area decreased as the concentration of metal increased. Strong decrease in leaf area was correlated to accumulation of chlorophyll pigments as disturb integration of chlorophyll molecules into stable complex (Skkorzynska Polit and Baszynski, 1997). Similar findings have been reported by Mehendirata et al. (2000)

Fresh and Dry weight:-

Fresh and dry weight of various parts of the tobacco plants showed a marked decline with the increase in cadmium level in the soil in all the sampling days. Hédiji et al. (2010) reported fresh weight reduction of *Solanum lycopersicum* under high Cd levels. There was a progressive fall in the dry matter yield of root, shoot and leaves with the corresponding increase in cadmium (10-200 mg kg⁻¹) level in the soil, in all the sampling days. Similar results were obtained by several authors in a number of plants such as Vitoria et al., (2001) in radish, Kim et al., (2002) in cabbage and lettuce, Zhang et al., (2002) in wheat, Shukla et al., (2003) in wheat, Rai et al., (2005) in *Phyllanthus amarus*. Abdo, Fatma (2008) in soybean.

Table 1:- Effect of various concentration of cadmium on root length (cm plant⁻¹) of tobacco (*Nicotiana tabacum* (L.).

Cadmium added in the soil (mg kg ⁻¹)	Sampling days					
	30	60	90	120	150	180
Control	3.20	49.30	65.72	76.34	82.54	85.69
10	3.00 (-6.25)	46.32 (-6.04)	62.35 (-5.13)	72.58 (-4.93)	77.25 (-6.41)	80.65 (-5.88)
25	2.80 (-12.50)	43.67 (-11.44)	58.46 (-11.05)	70.32 (-7.89)	74.36 (-9.87)	76.39 (-10.85)
50	2.40 (-25.00)	35.24 (-28.52)	52.40 (-20.27)	67.42 (-11.68)	72.32 (-12.38)	75.42 (-11.99)
75	2.10 (-34.38)	32.45 (-34.18)	49.72 (-24.35)	62.35 (-18.33)	66.54 (-19.38)	69.24 (-19.20)
100	2.00 (-37.50)	24.65 (-50.00)	42.65 (-35.10)	56.49 (-23.00)	59.22 (-28.25)	63.25 (-26.19)
200	1.80 (-43.75)	21.44 (-56.51)	36.75 (-44.08)	52.75 (-30.90)	56.33 (-31.75)	59.35 (-30.74)

Table 2:- Effect of different treatments of cadmium on shoot length (cm plant⁻¹) of tobacco (*Nicotiana tabacum* (L.).

Cadmium added in the soil (mg kg ⁻¹)	Sampling days					
	30	60	90	120	150	180
Control	5.40	28.00	39.67	54.23	58.13	62.56
10	5.10 (-5.56)	27.78 (-0.79)	37.63 (-5.14)	50.28 (-7.28)	56.73 (-2.41)	58.84 (-5.95)
25	4.80 (-11.11)	25.18 (-10.07)	34.56 (-12.88)	46.80 (-13.70)	52.19 (-10.22)	55.80 (-10.81)
50	4.20 (-22.22)	22.65 (-19.11)	31.23 (-21.28)	43.39 (-19.99)	48.49 (-19.58)	52.50 (-16.08)
75	4.00 (-25.93)	17.00 (-39.29)	26.54 (-33.10)	34.69 (-36.03)	38.67 (-33.48)	45.39 (-27.45)
100	2.60 (-51.85)	13.32 (-52.43)	23.45 (-40.89)	31.36 (-42.17)	36.52 (-37.18)	39.24 (-37.28)
200	2.80 (-48.15)	11.34 (-59.50)	19.32 (-51.30)	28.65 (-47.17)	33.56 (-42.27)	34.69 (-44.55)

Table 3:- Effect of various cadmium concentration of leaf number (plant⁻¹) of tobacco (*Nicotiana tabacum* (L.).

Cadmium added in the soil (mg kg ⁻¹)	Sampling days					
	30	60	90	120	150	180
Control	5.00	9.32	13.58	19.34	18.44	17.45
10	4.00 (-20.00)	8.65 (-7.19)	12.62 (-7.07)	18.66 (-3.52)	17.24 (-6.51)	16.58 (-4.99)
25	4.00 (-20.00)	8.00 (-14.19)	12.24 (-9.87)	18.00 (-6.93)	16.63 (-9.82)	16.30 (-6.59)
50	3.00 (-40.00)	7.34 (-21.24)	11.24 (-17.23)	16.45 (-14.94)	15.84 (-14.10)	15.08 (-13.58)
75	2.00 (-60.00)	7.00 (-34.89)	9.52 (-29.90)	15.76 (-18.51)	15.22 (-17.46)	13.34 (-23.55)
100	2.00 (-60.00)	6.33 (-32.08)	8.80 (-35.20)	15.49 (-19.91)	13.39 (-27.39)	12.66 (-27.45)
200	2.00 (-60.00)	5.82 (-37.55)	8.24 (-39.32)	13.62 (-29.58)	12.54 (-32.00)	12.09 (-30.72)

Table 4:- Effect of cadmium on total leaf area (cm²) of tobacco (*Nicotiana tabacum* (L.)).

Cadmium added in the soil (mg kg ⁻¹)	Sampling days					
	30	60	90	120	150	180
Control	62.14	657.49	936.05	1056.45	865.32	788.90
10	60.01 (-3.43)	624.52 (-5.01)	924.69 (-1.21)	988.40 (-6.44)	846.56 (-2.17)	727.13 (-7.83)
25	57.57 (-7.35)	595.43 (-9.44)	861.13 (-8.00)	934.25 (-11.57)	812.30 (-6.13)	677.29 (-14.15)
50	57.38 (-7.66)	546.80 (-16.84)	836.72 (-10.61)	908.57 (-14.00)	768.27 (-11.22)	643.15 (-18.48)
75	43.82 (-29.48)	510.87 (-22.30)	741.98 (-20.73)	897.23 (-15.07)	754.35 (-12.82)	609.24 (-22.77)
100	37.11 (-40.28)	489.64 (-25.53)	683.76 (-26.95)	765.27 (-27.56)	723.40 (-16.40)	570.12 (-27.73)
200	28.72 (-53.78)	453.16 (-31.08)	548.97 (-41.35)	738.67 (-30.08)	698.24 (-19.31)	534.25 (-32.28)

Table 5:- Effect of cadmium on root fresh weight (g plant⁻¹) of tobacco (*Nicotiana tabacum* (L.)).

Cadmium added in the soil (mg kg ⁻¹)	Sampling days					
	30	60	90	120	150	180
Control	3.380	5.628	8.212	10.504	12.720	14.792
10	3.093 (-8.49)	5.231 (-7.05)	8.120 (-1.12)	10.345 (-1.51)	12.546 (-1.37)	14.025 (-5.19)
25	2.856 (-15.50)	4.674 (-16.95)	7.459 (-9.17)	10.256 (-2.36)	12.190 (-4.17)	13.564 (-8.30)
50	2.530 (-25.15)	4.267 (-24.18)	7.235 (-11.90)	10.178 (-3.10)	11.589 (-8.89)	12.785 (-13.57)
75	2.324 (-31.24)	4.067 (-27.74)	7.189 (-12.46)	9.467 (-9.87)	11.280 (-11.32)	11.568 (-21.80)
100	2.102 (-37.81)	3.780 (-32.84)	6.984 (-14.95)	8.432 (-19.73)	10.453 (-17.82)	10.482 (-29.17)
200	1.345 (-60.21)	3.455 (-38.61)	6.745 (-17.86)	8.345 (-20.55)	10.260 (-19.34)	9.680 (-34.56)

Table 6:- Impact of cadmium on stem fresh weight (g plant⁻¹) of tobacco (*Nicotiana tabacum* (L.)).

Cadmium added in the soil (mg kg ⁻¹)	Sampling days					
	30	60	90	120	150	180
Control	5.435	17.028	35.635	66.849	73.760	80.534
10	5.178 (-4.73)	16.324 (-4.13)	35.097 (-1.51)	63.765 (-4.61)	71.005 (-3.74)	78.546 (-2.47)
25	4.325 (-20.42)	14.455 (-15.11)	32.678 (-8.30)	60.324 (-9.76)	69.425 (-5.88)	74.750 (-7.18)
50	3.754 (-30.93)	13.769 (-19.14)	30.537 (-14.31)	57.920 (-13.36)	66.439 (-9.93)	72.435 (-10.06)
75	3.432 (-36.85)	13.769 (-19.14)	28.980 (-18.68)	55.345 (-17.21)	63.761 (-13.56)	70.523 (-12.43)
100	3.098 (-43.00)	11.560 (-32.11)	25.008 (-29.82)	52.894 (-20.88)	62.835 (-14.81)	69.425 (-13.79)
200	2.270 (-58.23)	10.426 (-38.77)	23.349 (-34.48)	49.670 (-25.70)	60.033 (-18.61)	67.213 (-16.54)

Table 7:- Influence of different treatments of cadmium on leaf fresh weight (g plant⁻¹) of tobacco (*Nicotiana tabacum* (L.)).

Cadmium added in the soil (mg kg ⁻¹)	Sampling days					
	30	60	90	120	150	180
Control	7.692	19.296	34.938	53.342	44.890	40.453
10	7.420 (-3.54)	17.650 (-8.53)	32.540 (-6.86)	49.430 (-7.33)	40.563 (-9.64)	38.546 (-4.71)
25	6.765 (-12.05)	16.548 (-14.24)	30.123 (-13.78)	45.367 (-14.95)	38.425 (-14.40)	35.638 (-11.90)
50	6.324 (-17.78)	14.670 (-23.97)	28.324 (-18.93)	44.215 (-17.11)	36.430 (18.85)	32.879 (-18.72)
75	5.798 (-24.62)	13.567 (-29.69)	26.365 (-24.54)	40.652 (-23.79)	34.630 (-22.86)	30.098 (-25.60)
100	4.430 (-42.41)	12.646 (-34.46)	24.310 (-30.44)	37.638 (-29.44)	30.768 (-31.46)	27.432 (-32.19)
200	3.544 (-53.93)	10.780 (-44.13)	22.645 (-35.19)	35.319 (-33.79)	28.429 (-36.67)	25.560 (-36.82)

Table 8:- Various treatment of cadmium on root dry weight (g plant⁻¹) of tobacco (*Nicotiana tabacum* (L.)).

Cadmium added in the soil (mg kg ⁻¹)	Sampling days					
	30	60	90	120	150	180
Control	0.845	1.657	2.053	2.876	3.180	3.698
10	0.765 (-9.47)	1.465 (-11.59)	1.596 (-22.26)	2.563 (-10.88)	2.897 (-8.90)	3.182 (-13.95)
25	0.709 (-16.09)	1.089 (-34.28)	1.256 (-38.82)	2.239 (-22.15)	2.569 (-19.21)	2.845 (-23.07)
50	0.675 (-20.12)	0.960 (-42.06)	1.189 (-42.08)	1.980 (-31.15)	2.165 (-31.92)	2.654 (-28.23)
75	0.496 (-41.30)	0.827 (-50.09)	1.025 (-50.07)	1.635 (-43.15)	1.988 (-37.48)	2.267 (-38.70)
100	0.463 (-45.21)	0.724 (-56.31)	0.898 (-56.26)	1.298 (-54.87)	1.760 (-44.65)	1.918 (-48.13)
200	0.375 (-55.62)	0.549 (-66.87)	0.617 (-69.95)	1.056 (-63.28)	1.562 (-50.88)	1.875 (-49.30)

Table 9:- Different treatment of cadmium on stem dry weight (g plant⁻¹) of tobacco (*Nicotiana tabacum* (L.)).

Cadmium added in the soil (mg kg ⁻¹)	Sampling days					
	30	60	90	120	150	180
Control	2.860	7.684	12.456	15.675	18.467	21.019
10	2.389 (-16.47)	7.398 (-3.72)	12.180 (-2.22)	15.270 (-2.58)	18.167 (-1.62)	20.867 (-0.72)
25	2.160 (-24.48)	7.212 (-6.14)	11.678 (-6.25)	15.055 (-3.96)	17.985 (-2.61)	19.539 (-7.04)
50	1.989 (-30.45)	6.389 (-16.85)	11.398 (-8.49)	14.784 (-5.68)	17.580 (-4.80)	18.480 (-12.08)
75	1.670 (-41.61)	6.318 (-17.78)	11.267 (-9.55)	14.470 (-7.69)	17.360 (-5.99)	18.384 (-12.54)
100	1.467 (-48.71)	6.265 (-18.47)	11.098 (-10.90)	14.354 (-8.43)	16.936 (-8.29)	17.934 (-14.68)
200	1.082 (-62.17)	5.545 (-27.84)	10.865 (-12.77)	13.088 (-16.50)	16.680 (-9.68)	17.685 (-15.86)

Table 10:- Efficacy of cadmium treatment on leaf dry weight (g plant⁻¹) of tobacco (*Nicotiana tabacum* (L.)).

Cadmium added in the soil (mg kg ⁻¹)	Sampling days					
	30	60	90	120	150	180
Control	2.564	6.435	11.546	17.756	15.762	13.453
10	2.380 (-7.18)	6.319 (-1.80)	11.345 (-1.74)	17.534 (-1.25)	15.564 (-1.26)	13.235 (-1.62)
25	2.245 (-12.44)	6.212 (-3.47)	10.689 (-7.42)	17.231 (-2.96)	14.432 (-8.44)	13.154 (-2.22)
50	1.987 (-22.50)	5.289 (-17.81)	10.453 (-9.47)	16.564 (-6.71)	14.245 (-9.62)	13.058 (-2.94)
75	1.589 (-38.03)	5.167 (-19.70)	10.156 (-12.04)	15.780 (-11.13)	14.180 (-10.04)	11.890 (-11.62)
100	1.420 (-44.62)	5.098 (-20.78)	10.098 (-12.54)	15.324 (-13.70)	13.657 (-13.35)	11.134 (-17.24)
200	0.982 (-61.70)	4.545 (-29.37)	09.437 (-18.27)	15.160 (-14.62)	13.423 (-14.84)	10.985 (-18.35)

Conclusion:-

In the present study tobacco plants were grown in control and different levels of cadmium were assessed. Results showed that all the morphological parameters of tobacco were decreased as cadmium level increased. The results indicated that all the morphological parameters dependent on both the different levels of cadmium accumulation and biomass of the plants, when the tobacco plants were grown in cadmium contaminated soil.

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RESEARCH ARTICLE

ASSESS THE EFFICIENCY OF MARJORAM ON WEIGHT CONTROL AND BODY COMPOSITION OF MALE AND FEMALE ADULT STUDENTS.

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Abstract

Background: Obesity is a global epidemic and consequently many individuals are seeking strategies to reduce their body weight. These strategies may include weight-loss food supplements, including plant food supplements PFS, such as appetite suppressants or those increasing resting metabolism. PFS that claim to contribute to weight loss are marketed worldwide and readily available over the Internet. This increased usage has coincided with a resurgence of interest in nutritional therapy and complementary and alternative medicine. Marjoram is one of the most familiar kitchen herbs. Marjoram is gastrointestinal tract stimulant, tonic, carminative, hypoglycemic and diuretic as well as antibacterial and as antioxidant.

Objectives: The present work was designed to study the effects of marjoram on weight reduction in obese and overweight humans.

Methodology: The study was conducted on 64 healthy students, female students were from Faculty of Applied Medical Sciences, and male from College of Medicine, Umm Al-Qura University, Makkah, Saudi Arabia. 40 students consume marjoram one month. Anthropometric measurements were done before and after marjoram consumption. Statistical analysis was performed using SPSS software version 20.

Results: The obtained data showed most of student's preferred strategy of physical activity and diet to reduce their weight. A significant reduction on body weight after one month of consuming marjoram was observed.

Conclusion and Recommendation: The present study suggested that marjoram has effective herb on weight reduction.

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Introduction:-

Obesity is a public health problem, which is increasing in KSA with an overall obesity prevalence of 35.5% ⁽¹⁾. It is strongly associated with diabetes, hypercholesterolemia, and hypertension in the KSA, although the epidemic's characteristics differ between men and women. Therefore, the reduction in overweight and obesity are of considerable importance to public health ⁽²⁾.

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Many people who are obese or overweight try to lose their weight by following different strategies, such as increasing physical activities, diets prescribed by health professionals or from official dietary recommendations, or by using medications if the lifestyle modification is not sufficient ^(3,4).

A cross-sectional study was done in Australia in normal, overweight, and obese subjects showed that participants tried losing their weight by using one or more of strategies, which include weight loss center programs, prescription medication, over the counter supplements, changed diet, increased exercise or consulted a specialist. This study showed that changing diet and increasing exercise are the most common strategies used by participants ⁽⁵⁾.

Another group of people tends to use herbs as a safest, costless, and effortless way to lose weight. Nowadays, herbal medical is a growing area of alternative medicines. Many active ingredients in manufactured drugs are derived from plant compounds and have a wide range of use. Plants and plant extracts considered as safer as and more popular than chemical products ⁽⁶⁾.

In recent years, many studies have focused on the bioavailability of phenolic compounds in the prevention and treatment of obesity. Phenolic compounds and flavonoids have pharmacological properties such as antioxidant, antimutagenic, antithrombotic, anti-inflammatory, anticancer and antihyperlipidemic. They are widely distributed in plants and form part of the human diet ⁽⁷⁾.

Marjoram (*Origanum majorana*) is one of the herbal plants that contains phenolic compounds. It is found all over Asia, Arabian Peninsula, Africa, America, and Europe ⁽⁶⁾.

Ethanol extract of *Origanum majorana* has been shown to possess anti-inflammatory, antioxidative, hypolipidemic and hepatoprotective effects against LPS-induced endotoxemia. So it can be used as an effective and safe alternative complementary treatment ⁽⁸⁾.

In addition, medicinal effects of marjoram also include gastrointestinal tract stimulant, tonic, carminative, diaphoretic, hypoglycemic, diuretic as well as antibacterial ⁽⁷⁾.

One of the previous study was done on rats to see the effect of natural herbs of marjoram and ginger on weight gain and level of cholesterol, this study found that the hypercholesterolemic rats which fed on hypercholesterolemic diet with marjoram, ginger or mixture of them demonstrated lower values of body weight gain compared to control group which fed also on hypercholesterolemic diet without taken marjoram or ginger ⁽⁶⁾.

Subjects And Methods:-

Subjects:-

Study was conducted on 64 healthy students (36 female – 28 male), female students was study on Faculty of Applied Medical Sciences, and male from College of Medicine, Umm Al-Qura University, Makkah, Saudi Arabia. The students were Saudis and have been collected in variation of different stages and different medical specialties.

Methods:-

Study design:-

40 students were agreeing to consume marjoram tell one month all these who participated were met the following inclusion criteria:

1. Consent to participate in the study.
2. Healthy
3. Not use any medications or nutritional supplements.
4. Not pregnant or lactating women.
5. Not following any regimen for weight loss
6. Not allergic.
7. Not suffering from any chronic illness like Diabetes Mellitus, Chronic renal failure, asthma etc.

Data collection:-

The data were collected by direct interview. The questionnaire was completed by the researchers.

The study questionnaire: The questionnaire covering three sections as follows:

Demographics-Socio- Economic Data:-

Include data about the general characteristics of the study sample such as gender, age, and income.

Dietary and Health Assessment Questionnaire:-

Include dietary habit and intake, interest of weight management, and health status of the participants.

Anthropometric Measurements:-

Anthropometric measurements are reliable and valid measurements that indicate nutritional status of the subjects. Bioelectrical Impedance Analysis (BIA) device was used to measure weight, BMI, visceral fat, body fat percentage, body muscle percentage. Regarding to weight, participants were asked to remove their heavy outer garments and their shoes.

Height was measured by using a measuring tape, with the shoulders in relaxed position and arms hanging freely.

Body Mass Index (BMI):-

Body mass index (BMI) is a simple index of weight-for-height that is commonly used to classify overweight and obesity in adults (table A). It is defined as a person's weight in kilograms divided by the square of his height in meters (kg/m²)⁽⁹⁾.

Table (A):-Categories of BMI (WHO, 2016) (9).

BMI Kg/m ²	Classification
≤18.5	Underweight
18.5to24.9	Desirable weight
25to29.9	Overweight
≥30	Obese

Herbal Material:-

Organic marjoram herb (dried leaves) was purchased from local markets. It was placed in small storage bags, each containing 1.5 g of herb (equivalent to one heaped teaspoon). Preparation of marjoram tea was performed by participants. They were taught to prepare this in accordance with the traditional method, which steeping the bag of marjoram tea in 250 mL of boiled water until the color changed, followed by straining. Participants were instructed to drink one cup (250 mL) of the assigned tea, unsweetened, daily after their main meal by 2 hours.

Statistical Analysis:-

All results were tabled with SPSS software 21th edition. Descriptive statistics were run to summarize the collected data, and the results were displayed in frequencies and percentages. Chi square analysis was performed to test the differences between variables. Obtained results were tabulated and expressed as (mean ± SD). The differences among the participants were analyzed by T, test. All statistical analyses were performed at significant level of 95% (P. value ≤ 0.05).

Results:-

The demographic profile of participants presents in table (1) which shows that out of 36 subjects, 56.2 % were females and 43.8 % were males. Income vary between them, nearly half of them has >8000 SR/month .All of the participants were falling in the age group of 19- 25 years old.

Table 1:-Demographic, social, and economical characteristics of male and female participants including both intervention and control group.

Variable	Frequency	%
Age (year)	40	62.5
19-22	24	37.5
22-25	64	100.0
Total		
Gender		
Female	36	56.2
Male	28	43.8
Total	64	100.0
Income level		
≤ 3000	18	28.1
~5000	6	9.4
~8000	10	15.6
>8000	30	46.9
Total	64	100.0

As showed In table (2) the number of females have regular weight monitoring and looking for ideal weight is more than males which represents (83.3% Vs.72.4%) respectively. Most of females showed that they measure their weight once in a month or less (44.4%), while most males showed to measure it twice a month (35.7%). Females have interest to loss their weight to improve confidence and appearance and health status with a significant difference with males ($P<0.05$). Most participants prefer using diet on weight reduction than other methods.

Table 2:-Weight control and monitoring awareness among males and females participants.

Variables	Male		Female		P.V
	No.	%	No.	%	
Regular weight monitoring					0.373
Yes	16	57.1	26	72.2	
No	12	42.9	10	27.8	
Total	28	100.0	36	100.0	
Looking for ideal weight					0.419
Yes	20	71.4	30	83.3	
No	8	28.6	6	16.7	
Total	28	100.0	36	100.0	
How many times you measure your weight?					0.429
≤ 1 per month					
2 per month	6	21.4	16	44.4	
1-2 per week	10	35.7	10	27.8	
Every day	6	21.4	6	16.7	
Don't remember	0	0	2	5.6	
	6	21.4	2	5.6	
Total	28	100.0	36	100.0	
First reason to lose weight					0.048
Health	10	35.7	9	38.9	
Improve confidence and appearance	10	35.7	55.6		
Society influences	0	0	25	5.6	
Other	0	28.6	2	0	
Total	8	0	0		
Common way to lose weight					0.747
Herbs	2	7.1	16.7		
Medications	0	0	8	5.6	
Diet	0	57.1	2	38.9	
Physical activity	16	21.4	14	22.2	
	6	14.3	6	16.7	

Nothing	4		6		
Total	28	100.0	36	100.0	

As showed in the table (3) the appetite of males was significantly ($P<0.05$) better than females (50%) of males have excellent appetite, while most of females (44.4%) have good appetite. Both of males and females consider lunch as the main meal and preferred home foods as their favorite food.

Table 3:-Distribution of the participants regarding to dietary habits and intake.

	Male		Female		P.V
	No.	%	No.	%	
Appetite					0.042
Weak	0	0.0	8	22.2	
Good	6	21.4	16	44.4	
Excellent	14	50.0	10	27.8	
Over	8	28.6	2	5.6	
Total	28	100.0	36	100.0	
Main meal					0.154
Breakfast	0	0.0	8	22.2	
Lunch	20	71.4	14	38.9	
Dinner	6	21.4	8	22.2	
None, But frequent snacks	2	7.1	6	16.7	
Total	28	100.0	36	100.0	
Favorite Food					0.118
Fast food	14	50.0	8	22.2	
Fruits & Vegetables	0	0	6	16.7	
Home meals	14	50.0	22	61.1	
Total	28	100.0	36	100.0	

In table (4) data shows that 28.6% of males have heard about marjoram herb, while 44.4% of female heard about it, 71.4% of males and 55.6% of females did not have knowledge and background about marjoram herb. 7.1 % of males known about health benefits of marjoram herb while the same percentage (7.1%) of them answered that marjoram herb doesn't has health benefits, and the majority of males (85.7%) don't know whether the herb has health benefits or not. The same trends were found on females.

Table 4:-Knowledge and attitude towards marjoram herb.

Questions	Male		Female		P.V
	No.	%	No.	%	
Have you ever heard about marjoram herb?					0.358
Yes		28.6		44.4	
No	8	71.4	16	55.6	
	20		20		
Total	28	100.0	36	100.0	
Does it have a health benefits?					0.394
Yes	2	7.1	6	16.7	
No	2	7.1	0	0	
Don't know	24	85.7	30	83.3	
Total	28	100.0	36	100.0	

In Table (5) data and results shows Post-consumption feedback by asking six questions, then make a comparisons between males and females. 33.3% of males noticed effect of marjoram herb, compared to 66.7% did not. On the other hand, 37.5% of females noticed effect of marjoram herb, while 62.5% did not. Half of males said that they have experienced a side effect while the other half did not. 9.4% of females experienced a side effect while the majority (90.6%) did not with a significant difference ($P<0.05$). 16.7% of males have nausea and abdominal pain accompanied with using herb while there's no reported case of associated diarrhea. In females, results show that 93.8% have no side effect, 10% have nausea and no reported case of them associated with diarrhea or abdominal pain. Data show a significant difference ($P<0.05$) between meals and females who decide to use marjoram, 71.9% of females would like to continue drink it compared to 66.7% of males decided to not continue drink marjoram tea. Regarding to the role of

marjoram in suppress appetite, 33.3 % of males and 31.3% of females answered (yes), while 66.7% of males and 68.7% of females answered (no). Most of them noticed suppression effect of marjoram toward fat foods with insignificant difference between them.

Table 5:-Demonstration of the Post-consumption feedback.

Questions	Male		Female		P.V
	No.	%	No.	%	
Did you note any effect?	8	33.3	14	37.5	0.790
Yes	16	66.7	18	62.5	
No					
Total	24	100.0	32	100.0	
Side effect?	12	50.0	3	9.4	0.044
Yes	12	50.0	29	90.6	
No					
Total	24	100.0	32	100.0	
There is any problem accompanied with herb....	14	58.3	30	93.8	0.039
No	4	16.7	2	6.2	
Nausea	0	0	0	0	
Diarrhea	4	16.7	0	0	
Abdominal pain	2	8.3	0	0	
Other					
Total	24	100.0	32	100.0	
Would you like to continue drink it?	8	33.3	23	71.9	0.043
Yes	16	66.7	9	28.1	
No					
Total	24	100.0	32	100.0	
Is there any suppression effect on your appetite?	8	33.3	10	31.3	0.551
Yes	16	66.7	22	68.7	
No					
Total	24	100.0	32	100.0	
If yes, towards which food?	4	50	8	80.0	0.411
Fats	2	25	2	20.0	
All	2	25	0	0	
Fast food					
Total	8	100.0	10	100.0	

Data presented in table (6) shows that the mean value of weight was significantly ($P < 0.005$) decreased after one month of consuming marjoram than before represented (78.88 ± 5.06 vs. 77.95 ± 5.01) respectively. Percent of fat and Visceral fat was decreased but with insignificant different.

Table 6:-Anthropometric measurements of participants before and after one month of marjoram consumption

Variable	Before	After	P.V
weight	78.88 ± 5.06	77.95 ± 5.01	0.006
BMI	27.10 ± 1.39	26.74 ± 1.44	0.086
Fat %	33.66 ± 1.41	33.34 ± 1.72	0.727
Muscle %	27.71 ± 1.02	27.65 ± 0.84	0.942
Visceral fat	8.62 ± 1.11	8.16 ± 1.07	0.314

Discussion:-

Obesity becomes more prevalent, weight-loss practices grow increasingly popular in developed and developing countries. More than two-thirds of the adult population of the United States has attempted to lose or maintain weight⁽¹⁰⁾, and between 15 and 35% of adults in Canada, Netherlands and the European Union are trying to lose weight at any given time⁽¹¹⁾. The proportion of adolescents and university students attempting to lose weight is even greater: 30% in Lebanon⁽¹²⁾, 23% in Canada⁽¹³⁾, 37% in Japan⁽¹⁴⁾.

Obtained data in the present study showed that 57.1%, 72.2% of male and female respectively interest to monitor their weight and 71.4% of male and 83.3% of female take care of IBW, to feel satisfied with their body shape

appearance and to prevent diseases related to obesity. These finding agree with **Lynch *et al.*, (2007)**⁽¹⁵⁾ they reported that, thin body is the most preferred body shape in Western countries. Data showed that the first priority of females to loss their weight is to improve confidence and appearance. In agreement with this result **Najat *et al.*, 2011**⁽¹⁶⁾ they found that females were more worried about their body shape than males as 89% of the worried students were females, whereas, the majority of the “not worried” group were male students.

Most of students preferred diet and physical activity to reduce their weight, in the study of dieting practices, weight perceptions, and body composition: a comparison of normal weight, overweight, and obese college females, **Malinauskas *et al.*, 2006**⁽¹⁷⁾ found that 80% of their participated students used physical activity as a weight control strategy. Another study done by **King *et al.*, 2007**⁽¹⁸⁾ among 204 students, recruited from Midwestern University, reported that students did exercise to lose weight and improve their body appearances.

Less of participants use herbs for weight control (7.1% of male and 16.7% of female). In disagreement with this results **Mamtaniet *et al.*, (2005)**⁽¹⁹⁾ they found that most participants favored herbal supplements for reducing weight and they reported that all health-care practitioners and students should be educated about benefits and risks of herbal supplements. In the study of Herbal Medicine among Saudis: Awareness, Uses, Reasons and Common Herbs, **Abdulaziz Alanzi *et al.*, 2016**⁽²⁰⁾ found that (80%) out of their sample have used herbs as a medications and herbs used for acute conditions account for (70%) while chronic conditions only (20%).

Data presented in table (6) showed significant reduction on body weight after one month of marjoram consumption. In agreement with this result **Lobna *et al.*, 2014**⁽²¹⁾ they found that food weight gain, food intake and food efficiency ratio showed significant decreases in experimental group of rats consumed marjoram compared to normal control group. **Soltan and Abdel Wahab, 2006**⁽²²⁾ reported that the hypolipidaemic activity of marjoram in rats could be attributed to the presence of valuable polyphenolic compounds, terpenoids, flavonoids, tannins, hydroquinone, phenolic glycosides and sabinene. **Amarowicz *et al.*, 2008**⁽²³⁾ found that marjoram ethanolic extract contain considerable amounts of total phenolics compounds and have antioxidant activity and free radical-scavenging capacity. **Rang and Dale, 1991**⁽²⁴⁾ reported that the hypocholesterolemic effect of marjoram could be attributed to presence of isoflavones which prevent intestinal absorption of cholesterol by competition for its absorption sites. These results are also in accordance with **Negm, 2002**⁽²⁵⁾ who found that marjoram extract lead to significantly lowering in TG ($p < 0.01$) than may be due to lower fatty acids synthase. In the study of Biochemical and histopathological studies on the water extracts of marjoram and chicory herbs and their mixture in obese rats **Lamiaa *et al.*, (2009)**⁽²⁶⁾ found a significant weight reduction in rats group take 5% marjoram than negative control group.

Conclusion:-

The present study suggested that marjoram has a good effect on weight control and percent of body fats.

Recommendations:-

- More scientific studies on the effects of marjoram on weight reduction on the human are needed.
- Make other study emphasis on side effect of mega dose of marjoram.

Limitations:-

The limitation in this study may be related to the small sample size and limited time.

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RESEARCH ARTICLE

A CASE STUDY ON THE EFFECT OF PUNARNAVA (*Boerhavia diffusa*.Linn) IN GALACTORRHOEA.

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Ayurveda, case study, galactorrhoea , hyperprolactenemia, *srotodusti*.

Abstract

Galactorrhoea is one of the common complain found in adolescence to post menopausal women. Though it has varied etiology but most common cause is hyperprolactenemia. Long term use of hormonal therapy is the only way of management in modern medical science. This gives scope to find alternative treatment measures to relieve the patient from physical, mental, social discomfort and also harmful effect of long term hormonal therapy.

In *Ayurveda*, galactorrhoea can be compared to a symptom manifesting due to *Rasavaha sroto dusti*. Here type of *srotodusti* is *Atipravritti*. *Stanya*(milk) is the *upadhatu* of *rasa dhatu*. Galactorrhoea is nothing but *Stanyavridhi*. The objective of the treatment includes finding out exact culprit for galactorrhoea and symptomatic relief.

The present study has been conducted on a patient suffering from galactorrhoea for fifteen days which is due to mild elevation of serum prolactine level. The treatment plan includes *Langhan* in term of dietary restriction, life style modification and capsule Punarnava and capsule Trikatu orally. After ten days galactorrhoea stop and patient was symptomatically fine. The pathogenesis, treatment details, mode of action, result are discussed in detail in full paper.

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Introduction:-

Galactorrhoea is a discharge of milk or a milk-like secretion from the breast in the absence of parturition or beyond six months' post-partum in a non breastfeeding woman. The secretion may be intermittent or persistent, scant or abundant, free-flowing or expressible, and unilateral or bilateral. The condition is more common in women who are 20 to 35 years of age and in previously parous women; it is less common in children and nulligravid women. Galactorrhea also can occur in men. In children, galactorrhea is more common in infants and teenage girls. The condition may result in patient anxiety and can signify a serious underlying disorder. Galactorrhea is commonly associated with hyperprolactinemia. [1] Prolactin plays an important role in growth of mammary gland. It causes proliferation of epithelial cell of alveoli thereby inducing and maintaining milk production. Prolactin is responsible for lactogenesis. During pregnancy, particularly in latter months; large quantity of prolactin is secreted. But the activity of this hormone is suppressed by estrogen and progesterone secreted by placenta. Because of this, lactation is prevented during pregnancy. Immediately after the delivery of the baby and expulsion of placenta, there is sudden lissos of estrogen and progesterone. Now, the prolactin is free to exert its action on breast and to promote lactogenesis.[2]

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Galactorrhea itself isn't a disease, but it could be a sign of an underlying problem. This condition occurs in less than 1% of the general population and in 5-14% of patients presenting with secondary amenorrhea. Approximately 75% of patients presenting with galactorrhea and amenorrhea have hyperprolactinemia. Of these patients, approximately 30% have prolactin-secreting tumors.[3]

Mortality/Morbidity:-

Mortality is unlikely; however, in cases where the condition is due to a large prolactin-secreting tumor, local mass effect can lead to significant morbidity. [4]

Causes of Galactorrhea:-[5]

Some Medications Associated with Galactorrhea:-

Dopamine-receptor blocker:- Butyrophenones, Metoclopramide (Reglan), Phenothiazines, Risperidone (Risperdal).

Selective serotonin reuptake inhibitor:- Sulpiride, Thioxanthenes, Tricyclic antidepressants.

Dopamine-depleting agents:- Methyldopa (Aldomet), Reserpine (Serpasil).

Inhibition of dopamine release:- Codeine, Heroin, Morphine.

Histamine-receptor blocker:- Cimetidine (Tagmet) etc.

Other causes of galactorrhea include:-

Herbal supplements, such as fennel, anise or fenugreek seed, Birth control pills, Noncancerous pituitary tumor (prolactinoma) or other disorder of the pituitary gland, Underactive thyroid (hypothyroidism), Chronic kidney disease, Excessive breast stimulation, which may be associated with sexual activity, frequent breast self-exams with nipple manipulation or prolonged clothing friction, Nerve damage to the chest wall from chest surgery, burns or other chest injuries, Spinal cord surgery, injury or tumors

Idiopathic galactorrhea:-

Sometimes doctors can't find a cause for galactorrhea. This is called idiopathic galactorrhea, and it may just mean that your breast tissue is particularly sensitive to the milk-producing hormone prolactin in blood.

Galactorrhea in Men:-

In males, galactorrhea may be associated with testosterone deficiency (male hypogonadism) and usually occurs with breast enlargement or tenderness (gynecomastia). Erectile dysfunction and a lack of sexual desire also are associated with testosterone deficiency.

Galactorrhea in newborns:-

Galactorrhea sometimes occurs in newborns. High maternal estrogen levels cross the placenta into the baby's blood. This can cause enlargement of the baby's breast tissue, which may be associated with a milky nipple discharge.

In *Ayurveda*, galactorrhea can be compared to a symptom manifesting due to *Rasavaha sroto dusti*. As *Stanya* is the *upadhatu* of *rasa dhatu*. [6] Normal function of *stanya* are breast development and maintenance of life. [7] Galactorrhea is nothing but *Stanyavridhi*. Feature of *stanyavridhi* increased size of breast (*stanya-pinatva*), frequent flow of milk (*muhur-muhur pravritti*) and breast pain (*toda*). [8] *Rasavahasrota* get vitiated due to excessive intake of heavy (*guru*), cold (*shita*), unctuous (*snigdha*) food and over-worry. [9]

B. diffusa L. (*Punarnava*) occurs abundantly as a weed throughout India. It is a creeping and spreading perennial herb. It is an important medicinal plant much used in traditional medicine in many parts of the world. The plant dies during hot summers and puts forth fresh shoots after rains and is believed to be a rejuvenator. This plant was named in the honor of Hermann Boerhaave a famous 18th century Dutch physician commonly known as *tarvine*. It is an herbal species of flowering plant in the four-o'clock family. *Punarnava* means which rejuvenates the body or renews the body. It is taken in herbal medicine and often used as a green vegetable in many parts of India. [10] The properties and action of *punarnava* are –

Rasa – Tikta, Kasaya, Modhur, Guna – Laghu, Ruksha, Veerya – Ushna, Vipak – Katu, Probhava – Not signify. Dosh-karma-Vata-kapha Shamak. [11]

Worldwide Ethno medical uses of Punarnava are- In India B.diffusa has a long history of medicinal use in Ayurveda and Unani. Different plant parts used as an appetizer, eye tonic, flushing out the renal system, to treat seminal weakness and blood pressure. Its roots are used in treating Jaundice Ascites, Anasarca, scanty urine, internal inflammation, Asthma, Piles. The plain juice used as an antidote for rat poisoning. The seed are use as expectorant, carminative, muscular pain, lambago, scabis, scorpion sting, in Yunani medicine. Seed powder mixed with dried ginger, it is given in Urticaria. In Punjab, the drug is considered useful for eye. In Mumbai, it uses for dropsical swelling. In Goa, it uses as a diuretic in Gonorrhoea. In West Indies, use for gonorrhoea. In Arab country, it uses for the treatment of diabetes, stress, dyspepsia, inflammation, congestive heart failure, anti convulsant, anti fibrinolytic, night blindness, corneal ulcer etc. In Brazil the plant as a whole or, its extracts is used for albuminuria, Beri-beri, bile insufficiency, cystitis, edema, gall stone, gonorrhea, guinea worms, hepatitis, Hypertension, Jaundice, Kidney disorders, Renal stone, Liver sclerosis, snake bite, splenomegaly, urinary retention. In Guatemala, it is used for erysipelas, and guinea worms (Taylor, 2005). In Iran, it is used for abdominal pain, amaemia, ascites, asthma, blood purifier, cancer cararacts, child birth, cholera, constipation, cough, debility, digestive sluggishness, dropsy, dyspepsia, edema, eye problem, fever, heart ailments child birth hemorrhage, thoracic hemorrhage, internal parasites, lactation aid, menstrual disorder, rheumatism, as a diuretic, expectorant. In Iraq, it is used for edema, gonorrhea, liver, intestinal gas, joint pain, lumbago, nephritis, an appetite stimulant. In Nigeria, it is used for abscesses, asthma, boils, convulsions, epilepsy, fever, laxative, In West Africa it is used for abortion, guinea worms, and menstrual irregularities and as an aphrodisiac. In Tropical Africa the boiled roots are applied to ulcer abscess: The boiled leaves are considered expectorant and febrifuge and in large doses emetic. A decoction of aerial parts is also taken to treat gastro intestinal pains, intestinal worms and to regulate menstruation. In Mauritania, the seeds are ground and made into cakes which are coocked and eaten as a remedy for dysentery.[12]

Pharmacological Activities of B. diffusa L.(Punarnava) as per recent research are Diuretic, Hepatoprotective, Cholorectic activity, Anti inflammatory activity, Haematinic Effect, Ca^{+2} channel antagonistic Effect, Corticosteroids like effect Emetic and cathartic Effect, Anti convulsant activity, Anti cancer activity, Anti Diabetic/Hypoglycaemic effect, Anti Amoebic, Anti Fibrinolytic activity, Anti fungal activity, Restorative activity against stress induced changes in adrenal cortisol level, Anti oxidant property (It produced 50% inhibition of lipid peroxidation), Anti viral activity, Anti microbial activity specially Gram negative bacteria, Negative chronotropic activity. etc[13]

Details of the case:-

A 19 year old female patient, reported to IPD of Raghunath Ayurved Mahavidyalaya and Hospital, Contai, West Bengal with complain of galactorrhoea of both breast predominantly left side, mild tenderness of both breast for last fifteen days. Associated complaints were dysmenorrhoea, general weakness, nausea and occasional vomiting, headaches. There was no history of serious systemic illness, any positive past and family history.

History:-

Mode of Onset:- Sudden.

Duration:- For fifteen days.

Precipitating Factors:- No such

Past History:- No such

Family History:- patient's mother suffer from psychological disturbance probably schizophrenia for last fifteen years and she is under treatment.

Drug History:- A detailed drug history is crucial; galactorrhoea is associated with a wide variety of drugs that raise serum prolactin levels. Frequent use of Domperidon for nausea and vomiting is positive here.

Gynecologic History:- Irregular menstrual cycle in 3 to 5 days of duration. Patient is suffering from dysmenorrhoea, nausea, occasional vomiting during first day of menstruation. Her LMP was 12th May, 2015.

Associated Symptoms:- Nausea, occasional vomiting, and headaches, loss of appetite, Constipation, cold intolerance, lethargy. etc

Physical Examination:-

General:- On examination patient is thin built, weight 45kg, non pallor, non icteric, no lymph gland enlarged and other vital signs are normal. Pulse rate 72 bpm and blood pressure 110/70 mm of Hg. Visual field within normal limit.

Breast Examination:- No sign of local irritation, infection, or trauma in the both breast.

Nipple Discharge:- A milky, thin, intermittent discharge from bilateral breasts, the discharge is scanty from right side and abundant from left one, on expressed and sometimes spontaneous. There was mild tenderness and no nodules.

Laboratory Evaluation:-

1. Pregnancy test –negative. (Dated on 31th July, 2015).
2. Levels of serum prolactin(fasting)-53.94ng/ml (Dated on 4th Aug,2015).
3. Level of serum T3-0.801ng/ml. T4-6.74mcg\100ml, TSH-4.41microIU\ml. (Dated on 4th Aug, 2015).
4. Levels of serum prolactin (fasting)-0.48ng/ml (Dated on 5th Nov,2015).
5. CT of brain and pituitary-nothing abnormality detected. (Dated on 14th Nov, 2015).
6. Levels of serum prolactin(fasting)-0.377ng/ml (Dated on 16th Aug,2016).

Materials and Methods:-

1. Capsule Punarnava 2cap thrice daily (Himalaya Drug Company) for 3month.
2. Capsule Trikatu 2cap twice daily after meal (Himalaya Drug Company) for 30days.

Patient was advised to avoid day sleep, night awakening, guru and Kapha aggravating substance like heavy meal ,fast food, fatty food, curd,etc.. Breast stimulation by clothing, suckling, self-manipulation, or stimulation during sexual activity should be avoided.

Observation:-

The sign and symptoms observed like discharge and tenderness were subsiding after 10 days. Occasional scanty discharge was found on expressed. After 30 days discharge was completely absent.

Discussion and Conclusion:-

In *Ayurveda*, galactorrhoea can be compared to a symptom manifesting due to *Rasavaha sroto dusti*. As *Stanya* is the *upadhatu* of *rasa dhatu*. Normal function of stanya is breast development and maintenance of life. Galactorrhoea is nothing but *Stanyavridhhi*. Feature of stanyavridhhi increased size of breast (*stanya-pinatva*), frequent flow of milk (*muhur-muhur pravritti*) and breast pain (*toda*). *Rasavahasrota* get vitiated due to excessive intake of heavy (*guru*), cold (*shita*), unctuous(*snighdha*) food and over-worry(*ati-cinta*). Feature of *Rasavahasrota dusti* is similar to feature of *Rasa dhatuvridhhi*, that is feature of increased *Kapha dosha*. *Kapha* when increased produces poor digestion (*Agni sadan*), excess salivation (*Proseka*), laziness(*alasya*), feeling heaviness of the body(*gourava*), *paleness(sveta)*, coldness(*saita*),feeling looseness of the body parts (*slathangata*) i.e, weakness ,dyspnoea (*swas*),cough(*kasa*), excess of sleep(*ati nidra*).These all features are very much similar with Hyperporlactenemia. On the other hand, Punarnava being a *ushna virya* and *katu vipak dravya*, it alleviate *Kapha dosha* as well as *Vata dosha*. Persons suffer from glactorrhoea must be avoid diet and regime which increases kapha dosha like heavy meal, curd, dairy product, day sleep etc and use fat free, little in quantity, warm meal possessing katu, tikta kashaya taste, honey, drugs which reduce fat, fasting ,different kind of exercisese etc . Extensive study and research is necessary to find out the role of punarnava in hyperprolactenemia as well as others endocrinal disorder.

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RESEARCH ARTICLE

EFFICACY OF COMBINED DRUG THERAPY ON CARPAL TUNNEL SYNDROME.

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Key words:-

Carpal tunnel syndrome,
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Abstract

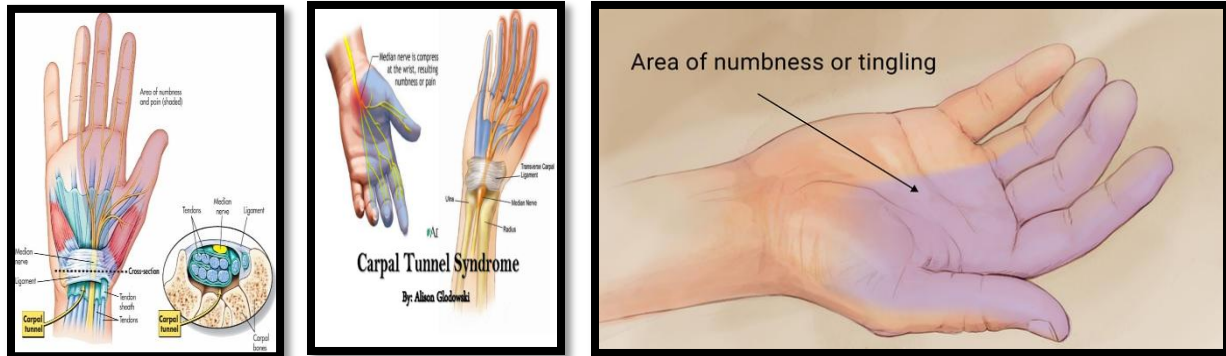
Carpal tunnel syndrome (CTS) is characterized by pain, numbness and tingling sensation in the hand caused by compression of median nerve at the wrist. Ammukkarachooranam(AMC), as internal drug and Pinder oil(PT) as external drug along with thokkamam(massage) were included in this combined strategy of therapy. The Aim of study is to evaluate reduction of signs and symptoms of CTS as per 5 point- Likert scale. The study was conducted at Teaching Hospital of Siddha medicine, Konesapuri, Trincomalee, the period of study was two years and four months. The Hospital drug is given for the CTS patients who were selected for the documentation of this research study. A patient should receive the drugs for three months. The base line and follow up symptoms were recorded with the global assessment of five point likert scale. The evaluation made to observe the reduction of significant symptoms such as pain, numbness and tingling. Amukurachooranam(AMC) 1 gram twice a day given orally with warm water and Pinder oil(PT) applied externally to the affected area. The clinical trial showed remarkable reduction of symptoms of pain and numbness. Because experimental phase shows the trial drugs have reducing effect in pain, numbness and tingling sensation from "always"(5) and "often"(4) to "never"(1) as per Likert scale. This was correlated with SPSS version 16, T Test paired sample statistics, trial combined drug showed significant reduction of symptoms of pain and numbness and tingling sensation compared to base line symptoms, because mean value of pain in base line 3.72 ± 0.54 , final visit 1.24 ± 0.52 ; mean value of numbness in base line 4.12 ± 0.6 , final visit 1.4 ± 0.64 , mean value of tingling sensation in base line 3.32 ± 0.62 , final visit 1.04 ± 0.2 with. It has been concluded that the strategy of this combination of therapy of tested drugs along with thokkanam is effective in the management of Carpal tunnel syndrome.

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Introduction:-

The carpal tunnel is located at the middle third of the base of the palm, is a narrow rigid passage way along in the wrist, about an inch wide. The floor and sides of the tunnel are formed by small carpal bones namely scaphoid tubercle and trapezoid, the roof of the tunnel is a strong band of connective tissue called the transverse carpal

ligament. Nine tendons joint together called as flexor retinaculum, which bend the fingers and thump also travel through the carpal tunnel. The median nerve is the one of the main nerve in the hand. It originates as group of nerve root in neck. The roots come together and form a single nerve in the arm. The median nerve goes down the arm and forearm, passes through the carpal tunnel at the wrist and goes to the hand. The nerve provide sensation in the thump, index, middle and ring finger and also controls muscle around the base of the thump as well as impulses to some small muscles in the hand for the movements of fingers. Sometimes thickening from irritated tendons or other swellings narrows the tunnel and causes compression of the median nerve as it runs deep to the transverse carpal ligament causes atrophy of the thenar eminence, weakness of the flexor pollicis, abductor pollicisbrevis as well as sensory lost in the digits supported by the Median nerve. Carpal tunnel syndrome is neurological condition commonly occurs in female, they are three times more likely than men. It is characterized by pain, numbness or tingling sensation in the wrist. Symptoms most often occur in thump, index finger, middle finger and half of the ring finger.



The patient with Carpal Tunnel Syndrome is frequent burning sensation, tingling, itching, and numbness in the palm of the hand and the fingers, especially in thumb, index and middle finger. These Symptoms usually start gradually. The symptoms often first appear in one or both hands during the night .As symptoms worsen, patient may feel tingling during the day. Decreased grip strength .Muscle wasting noticed in the base of the thump in chronic patients.

Aim and objective:-

The feedback of some of the CTS patients regarding this therapeutic effect made me to document this therapy as a research study. At the same time, no previous studies were conducted to focus the scientific view for this type of condition with combined drug therapy. Therefore the aim of the researcher is to design this study to explore the combined efficacy of AMC and PT oil along with Thokkanam on carpal tunnel syndrome.

Materials and Methods:-

The study was conducted at Teaching Hospital of Siddha medicine, Konesapuri, Trincomalee. The Hospital drug is given for CTS patients and randomly selected for the documentation of this research study. A patient should receive the drugs for three months. The period of study was two years and four months since this type of patients rare to attend hospital. All the patients were screened after history taking and physical examination along with differential diagnosis. The consent was obtained while the patients were made to aware this therapy will be documented and used for research purposes. Among 32 only 25 patients (15 female and 10 male) records were selected for this research paper. Other 07 were excluded because they were Pregnant, lactating mothers and some of them having Diabetes mellitus, Hypertension, Asthma etc .The base line and the follow up symptoms were correlated with the global assessment of five point Likert scale. The evaluation made to observe the reduction of significant symptoms such as pain, numbness and tingling. The treatment given in OPD and advice guideline also was given to patients to follow strictly. The internal drug AMC1 gm three times daily with warm water was given , the PT oil and thokkanam (massage therapy) given to apply externally in the affected area for ninety days.

The diagnosis of the CTS is initially suspected based on symptoms supported by history and physical examination and also confirmed by following nerve conduction testing.

Tinel's sign test:-

It is performed by gently tapping skin over the flexor retinaculum to elicit a tingling sensation or "pins and needles".

Phalen's sign test:-

It is performed by flexing the wrist gently as far as possible, then holding this position for up to 60 sec, positive results is feeling of numbness along median nerve distribution.

Two point discrimination:-

Ask the patient to close the eye and then use small tips of opened paper clips, to touch two points nearly 0.5 cm apart on patient's hand or finger. The positive test is patient can't able to feel difference between the two touches in severe CTS, so it may feel as though only one place is being touched.

Results and Discussion:-

		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Patient	13.00	25	7.360	1.472
	PainFirstVisit	3.72	25	.542	.108
Pair 2	Patient	13.00	25	7.360	1.472
	PainEightVisit	1.24	25	.523	.105
Pair 3	Patient	13.00	25	7.360	1.472
	NumbnessFirstVisit	4.12	25	.600	.120
Pair 4	Patient	13.00	25	7.360	1.472
	NumbnessEightVisit	1.40	25	.645	.129
Pair 5	Patient	13.00	25	7.360	1.472
	TinglingSensationFirstVisit	3.32	25	.627	.125
Pair 6	Patient	13.00	25	7.360	1.472
	TinglingSensationEightVisit	1.04	25	.200	.040

Point-Likert scale:-

1. Never
2. Rarely
3. Some times
4. 4.Often
5. Always

Frequency chart -- Base line & Eighth visit:-**Pain:-****Pain First Visit**

	Frequency	Percent	Valid Percent	Cumulative Percent
Valid	Sometimes	8	32.0	32.0
	Often	16	64.0	96.0
	Always	1	4.0	100.0
	Total	25	100.0	

Pain Eight Visit

	Frequency	Percent	Valid Percent	Cumulative Percent
Valid	Never	20	80.0	80.0
	Rarely	4	16.0	96.0
	Sometimes	1	4.0	100.0
	Total	25	100.0	

Numbness First Visit

	Frequency	Percent	Valid Percent	Cumulative Percent
Valid	Sometimes	3	12.0	12.0
	Often	16	64.0	76.0
	Always	6	24.0	100.0
	Total	25	100.0	

Numbness:-**Numbness Eight Visit**

	Frequency	Percent	Valid Percent	Cumulative Percent
Valid	Never	17	68.0	68.0
	Rarely	6	24.0	92.0
	Sometimes	2	8.0	100.0
	Total	25	100.0	

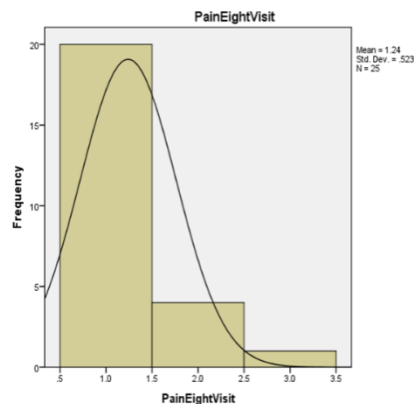
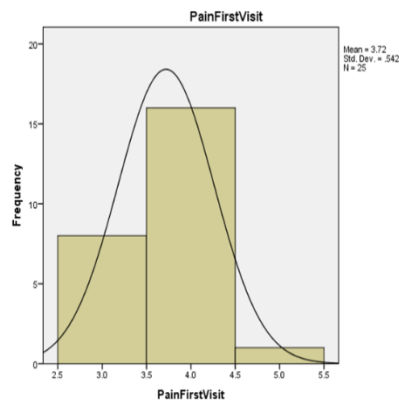
Tingling Sensation First Visit

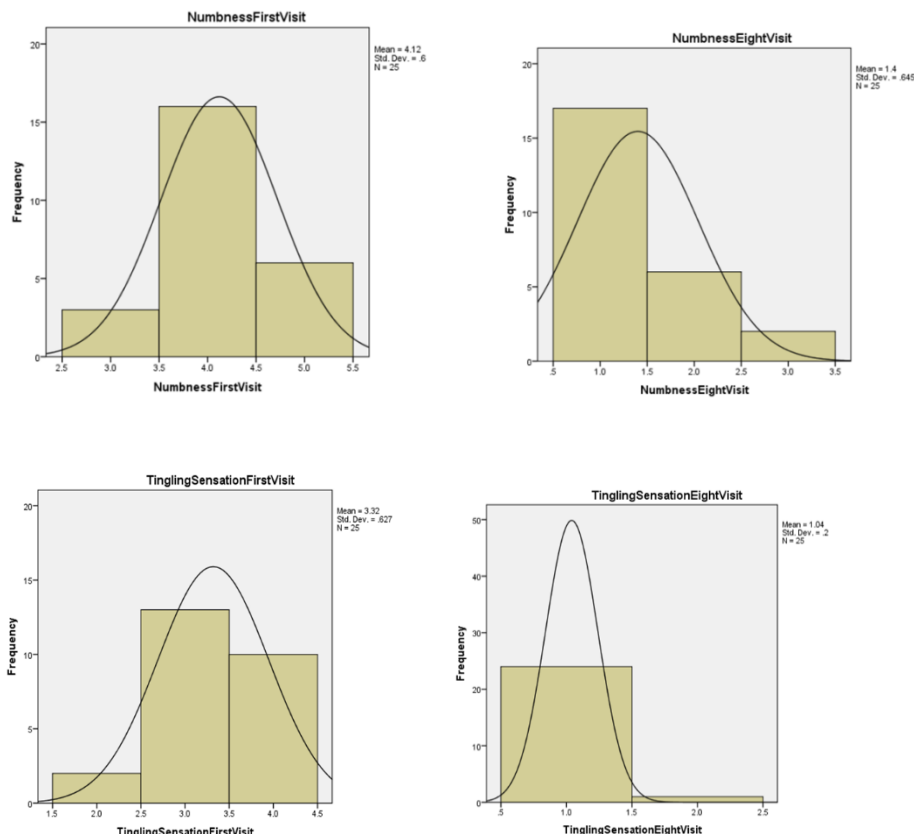
		Frequency	Percent	Valid Percent	Cumulative Percent
Valid	Rarely	2	8.0	8.0	8.0
	Sometimes	13	52.0	52.0	60.0
	Often	10	40.0	40.0	100.0
	Total	25	100.0	100.0	

Tingling Sensation Eighth Visit

		Frequency	Percent	Valid Percent	Cumulative Percent
Valid	Never	24	96.0	96.0	96.0
	Rarely	1	4.0	4.0	100.0
	Total	25	100.0	100.0	

The results showed remarkable reduction of symptoms of pain , numbness and tingling sensation compared to base line symptoms, because mean value of pain in base line 3.72 ± 0.54 final visit 1.24 ± 0.52 ,mean value of numbness in base line 4.12 ± 0.6 final visit 1.4 ± 0.64 ,mean value of tingling sensation in base line 3.32 ± 0.62 , final visit 1.04 ± 0.2 with SPSS version 16, T Test paired sample statistics .





As per the siddha medical reputed literatures, the root of Amukira can be taken internally with warm water, honey, milk or ghee. It enhances the function of nervous system and improves memory and promotes the functions of reproductive system. Amukira root has been used as a Rejuvenator, tonic, and aphrodisiac, narcotic. Anti-inflammatory, analgesic, diuretic anthelmintic, astringent and stimulant. It is commonly used in children for emaciation; in adults for improve sexual function, in elders' debility, rheumatism, nerves damage, and insomnia. as well as the pinda oil also mentioned in siddha pharmacopeia and siddha pharmaceutical text for rheumatic or traumatic conditions as 'pain killer'. This present study shows positive results in the management of CTS since the Amukirachooram and Pinda oil having analgesic, anti-inflammatory and soothing effect.

The various phytochemicals isolated from *Withaniasomnifera*(Amukira/Aswakantha) such as Alkaloids-Isopelletierine, anaferine, cuseohygrine, anahygrine ,Steroidal lactone, thanolides, withaferins. Saponins-Sitaindosides and acylsterylglucosidesSitaindosides vii-x and The aerial part of withaniasomnifera yielded 5-dehydroxywithanolite -R , withasomniferin- A (Rahman et al 1991) (Mishra .et al 2000)

Withaferin A and 3-b- hydroxyl-2, 3-dihydro withanolide isolated from withaniasomnifera show antibacterial, antitumourimmune modulating and anti-inflammatory properties (Budhiraja and Sudhired al., 1987). Withaferine found to be effective in anti-stress activity (Bhattacharya et al., 1989) Many constituents show immune modulatory actions (Ghosal et al., 1989). *Withaniasomnifera*(Amukira) improves the body's defense against diseases by improving the cell mediated immunity and also possesses potent antioxidant properties that protect against cellular damage caused by free radicals.

The scientific researchers were reported the findings of the ingredients of this trial drugs as follows; Amukra 1000 mg/kg oral produced significant analgesic activity for a rat experiencing heat analgesia induced by hot plate method. (Mazen et al., 1990). Amukira is an analgesic that soothing nervous system from pain response (Twair et al., 1989). The powerful anti-arthritis properties of Amukira are now widely accepted and documented by several researchers. Ethanolic extract of *Withaniasomnifera* elicited significant dose dependent acute anti-inflammatory activity in

carrageen an induced paw edema comparable to hydrocortisone and also elicited significant chronic anti-inflammatory activity is found adjuvant induced arthritis conferrable to hydrocortisone. (KiranGiri et al., 2015). Withaferin has remarkable analgesic and antipyretic properties accompanied with absence of significant gastric damage. (Sabina et al 2008)

The external medicine Pinder oil consist of Commiphoramukul (Guggul), Hemidesmusindicus (Nanari or Indian sarsaparilla), Rubiacordifolia (manjisti), beeswax, Sesamumindicum (Gingelly oil). Pider oil known as Sivappukkukilennai endorsed in siddha literature for pain reliever and anti-rheumatic healer as external application.

Guggul plant consists of number of phytochemicals such as terpenoids, steroids, flavanoids, guggultertols, lignans, and aminoacids. Guggulsterones are chief constituents. It has anti-inflammatory and analgesic activity, and also has hypoglycemic and hypolipidaemic effect on type II diabetic patients. (PrernaSarup et al 2015). Commiphora .mukul showed significant decrease in volume thickness in paw edema in rats, and also produces significant analgesic activity as measured by hot plate and writhing test in mice; moreover the thermal hot plate method was more sensitive than chemical writhing. (Shalaby, Hammouda et al., 2014)

Root of Indian sarsaparilla has various phytoconstituents like essential oil, glycosides, resins saponin steroids and tannins which all endowed with many pharmacological activities. Root of the plant exhibits significant anti-inflammatory activity in both acute and sub-acute condition in experimental rats.

The *Rubiocordifolia* showed significant Neuroprotective activity and reduce memory dysfunction. (Joy, Nair CKF et al 2008.) Anthroquinones rich fraction of ethanol extract and aqueous root extract showed significant anti-inflammatory activity in edema inhibition induced arthritic model in experimental rat (Jaijesh et al; Sirinivasan et al., 2008)..

Sesame oil used as food supplement and also in medicinal preparations. It is highly nutritious oil and enriching with protein, essential vitamin like, E, D K, B, and Niacin complex and mineral such calcium, phosphorus, Manganese, Magnesium etc. This oil has high viscosity, lubricating and penetrating power. Thus, helps in quick penetration, thereby regulating and enhancing blood circulation of affected area. Sesame oil is rich in essential fatty acids like linoleic acid and palmitic acids. Linoleic acid shows anti-bacterial and anti-inflammatory properties. The oil also rich in anti-oxidants, hence they nourish the deep tissues and improve the blood circulation. Beeswax used as surface protector of the skin and glazing agent. The previous research findings indicated the typical pharmacological activity such as anti-inflammatory, analgesic, Neuro-protective and soothing activity present in the research dugs AMC and PT oil. The thokkanam (massage) therapy enhance further quick relieve from Carpal tunnel syndrome. This study shows the trial drugs have reducing effect in pain, numbness and tingling sensation from “always”(5) and “often”(4), sometimes(3) rarely(2) to “never”(1) as per 5 point Likert scale concordance with statistical analysis.

Conclusion:-

It is concluded that statistical analysis with SPSS Version 16, T-test paired sample showed significant reduction of symptoms like pain, tingling and numbness of CTS, trial combined drug showed significant reduction of symptoms of pain and numbness and tingling sensation compared to base line symptoms, because mean value of pain in base line 3.72 ± 0.54 , final visit 1.24 ± 0.52 ; mean value of numbness in base line 4.12 ± 0.6 final visit 1.4 ± 0.64 , mean value of tingling sensation in base line 3.32 ± 0.62 , final visit 1.04 ± 0.2 . The therapeutic strategy of this combination of tested drugs along with thokkanam is effective in the management of Carpal tunnel syndrome with Amuukirachooram (AMC) as internal and Pinder oil (PT) as external application along with thokkanam as enhancer.

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RESEARCH ARTICLE

TO EVALUATE THE EFFICACY OF SUB GINGIVAL IRRIGATION WITH OCTENIDOL AND CHLORHEXIDINE ON PERIODONTAL INFLAMMATION.

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Key words:-

sub gingival irrigation, Chlorhexidine, octenidol, scaling, oral hygiene methods.

Abstract

Background: To evaluate the efficacy of octenidol and Chlorhexidine in periodontal pockets and the practicability of patient self therapy by using sub gingival irrigation.

Methodology: 40 subjects with periodontal pockets (4-6mm) were enrolled and 30 were responded to treatment, divided randomly into 2 groups (Group 1 octenidol & Group 2 Chlorhexidine). Prior to scaling all clinical parameters (SBI, PI, PD & CAL) were recorded. Sub gingival irrigation done in both groups at 0,7,14 days on premolars and molars. Data were analyzed by SPSS (p<0.05).

Results: A significant reduction in periodontal inflammation and clinical parameters from base line to 14days were observed and maintained for 21 days without irrigation. When intergroup comparisons were made no statistical significance was observed in all clinical parameters except for sulcus bleeding index (SBI) showed highly significant difference in octenidol group.

Conclusion: sub gingival irrigation with octenidol and Chlorhexidine are effective in reducing periodontal inflammation and in controlling sub gingival plaque. Intermittent treatment of this kind by the patient at home might reduce to more manageable levels, the frequency of hygiene visits and the need for rigorous interdental oral hygiene.

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Introduction:-

The primary etiological factor for inflammatory periodontal disease is bacterial plaque. So the principal goal in the prevention of inflammatory periodontal disease is removal of bacterial plaque.¹ Various Mechanical and chemical

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methods are available presently for treating periodontal diseases. However, antibiotics appear to reduce the severity of gingivitis and may be useful in managing some form of periodontitis. Based on potential problems associated with the prolonged use of systemic antibiotics for controlling plaque, a wide variety of studies have been directed to topical or sub gingival debridement of antimicrobial agents. These agents can control colonization of the teeth by oral microorganisms which will prevent the development of gingivitis and further progression of periodontitis.²

Chlorhexidine has long been used as an antiplaque agent on the basis of the non-specific plaque hypothesis.³ Sub gingival application of Chlorhexidine may lead to changes in the sub gingival flora which selectively decreases the re-establishment of a pathogenic flora.⁴ So that Chlorhexidine may be used as a chemical agent adjunct to mechanical therapy.

Octenidine (N,N'-(1,10-Decanediyldi-1 (4H)- pyridinyl-4-ylidene) bis-[1-octanamine] dihydrochloride), a new bispyridine compound developed by the Sterling-Winthrop Research Institute.² It appears to possess substantial antiplaque activity. In monkeys, daily topical administration of octenidine resulted in a significant reduction of plaque accumulation over a seven day period (**Emilson et al 1980**).⁵ Octenidol is a highly effective bactericide against the leading germ group of infectious diseases of the periodontitis and gingivitis types, otherwise known as odontogenic infections. It is used for the regeneration of inflammatory diseases in the oral cavity and for daily application at home with restricted oral hygiene capacity. With its active ingredient Octenidine, it inhibits bacteria rapidly and persistently, prevents them from forming again and ensures healthy oral flora.⁶

Octenidol is superior over Chlorhexidine in Supportive treatment of Multi-resistant *Staphylococcus aureus* (**MRSA**). It does not discolor the teeth and it is alcohol free. So the present study is undertaken to evaluate the efficacy of sub gingival irrigation with octenidol and Chlorhexidine on periodontal inflammation.⁷

Methodology:-

A total sample of 30 patients with periodontal pockets (4-6mm) was selected from the Out patients, Department of Periodontics, Lenora institute of dental sciences, Rajahmundry, Andhrapradesh. The sample of the study was randomly divided into 2 groups, one of which received octenidol and other received Chlorhexidine. All of them were instructed to brush their teeth once daily and none had received any instructions in interdental cleaning aids. The study was approved by Ethical Committee, Lenora Institute of Dental Sciences.

Prior to scaling all clinical parameters including: Sulcus bleeding Index (SBI) (Muhlemann & Son 1971), Pocket depth (PD), Plaque Index (PI) (Silness & Loe 1964) and Clinical attachment level (CAL) were recorded. Test sites were in relation to approximal surfaces of premolars and molars. The present study was based on the following inclusion and exclusion criteria after obtaining informed consent from the patients.

Inclusion criteria:-

- Patients with age group of 20-50 years.
- Patients with at least 26 natural teeth with diagnosis of chronic gingivitis with localised periodontitis are enrolled in the study.
- Patients with pocket depth 4-6mm in premolars and molars were included in this study.

Exclusion criteria:-

- Patients with systemic diseases and orthodontic bands.
- Patients who were sensitivity to mouth wash.
- Patients who were receiving systemic antibiotics in last six months.

The irrigation device consisted of 10ml disposable plastic syringe and blunt plastic tips as showed in figure 1. The irrigation solution used is either 0.2% Chlorhexidine gluconate solution or 0.1% octenidol solution. These were supplied in identical bottles, distinguishable only by a random code number. The key to the code remained unbroken until all the data had been collected. Irrigation device

The teeth to be treated were shown and periodontal pocketing explained briefly to the patient. They were shown how to assemble and use the irrigation device. With 10ml of solution in the syringe, the investigator inserted the needle gently into the pocket until resistance was felt, without causing discomfort to the patient. About 1ml of solution was deposited into each pocket by the patient themselves without any injury to the tissues⁸ as shown in figure 2. An

individual irrigation technique was checked on day 7 and if any fault should be corrected. Except on days 0 and 7, pockets were irrigated by the patient once only at night for 14 days. At day 21, irrigation devices and solutions were taken from the patient. Clinical parameters were assessed at 0,7,14 and 21 days.

Analyses were performed using commercially available software (SPSS). Mann Whitney U test were carried out for inter group comparisons⁷ and Paired t test used for analyzing intra group comparisons⁷. The null hypothesis was rejected at the 0.05 level of significance.

Figure 1:- Irrigation Devices and Blunt Plastic Tips for Sub gingival irrigation.



Figure 2:- Technique of Sub Gingival Irrigation



Results:-

The study was conducted with a sample of 40 patients, in that 30 were responded to treatment. The study sample was randomly divided into two Groups for sub gingival irrigation (Group1- octenidol and Group 2- Chlorhexidine).

When comparison of means of Sulcus Bleeding Index (SBI) in both Groups: Octenidol group has higher reduction of mean SBI than Chlorhexidine group and continued until day 21 ($P=0.000$) as shown in (Table 1). When comparing means of SBI within the groups, there was significant reduction from baseline to 7 days and this reduction was greatest by day 21 ($P<0.001$) as shown in (Table 1a&1b).

When comparison of means of Plaque Index (PII) in both Groups: octenidol group showed significant improvement by day 7 ($P<0.001$). However, day 14 and 21 no significant difference was observed in both the groups as shown in

(Table 2). When comparing means of PII within the groups, there was significant reduction in PII scores were observed by day 21 ($P < 0.001$) as shown in (Table 2a & 2b).

When mean Probing depth (PD) values were compared in between the groups there was no significant difference was observed as shown in (Table 3). When mean PD was compared within the groups, both groups showed significant PD reduction from baseline to 21 days ($P < 0.001$) as shown in (Table 3a & 3b).

When comparison of means of Clinical attachment levels (CAL) in between the groups there was no significant difference was observed by day 7 and day 21, but the difference was significant by day 14 in octenidol group as shown in (Table 4). When Comparing means of CAL within the groups: highly significant difference was observed from day 0 to day 21 ($P < 0.001$) as shown in (Table 4a & 4b).

Table 1:- Comparison of Means of Sulcus bleeding index (SBI) between Group I and Group II

Sulcus bleeding index (SBI)					
DAY	GROUP I	GROUP II	Difference	Z value	P value
	MEAN \pm SD	MEAN \pm SD	MEAN \pm SD		
0	2.75 \pm 0.45	2.53 \pm 0.52	0.22 \pm 0.07	-1.240	0.215 NS
7	1.13 \pm 0.34	1.87 \pm 0.35	0.74 \pm 0.01	-4.062	0.000 S
14	0.63 \pm 0.50	1.53 \pm 0.52	0.90 \pm 0.02	-3.730	0.000 S
21	0.13 \pm 0.34	0.73 \pm 0.59	0.60 \pm 0.25	-3.062	0.002 S

Statistical Analysis: Mann Whitney U test.

Statistically significant if $P < 0.05$

Table 1a:- Comparison of Means of Sulcus Bleeding Index (SBI) In Group I with Different Time Periods.

sulcus bleeding index (SBI)					
DAY	GROUP I : Day 0	GROUP I	Difference	t value	P value
	MEAN \pm SD	MEAN \pm SD	MEAN \pm SD		
7	2.75 \pm 0.45	1.13 \pm 0.34	1.63 \pm 0.11	13.000	<0.001 S
14		0.63 \pm 0.50	2.13 \pm 0.05	13.729	<0.001 S
21		0.13 \pm 0.34	2.63 \pm 0.11	21.000	<0.001 S

Statistical Analysis: Paired t test.

Statistically significant if $P < 0.05$

Table 1b:- Comparison of Means of Sulcus Bleeding Index (SBI) In Group II with Different Time Periods.

Sulcus bleeding index (SBI)					
DAY	GROUP II: Day 0	GROUP II	Difference	t value	P value
	MEAN \pm SD	MEAN \pm SD	MEAN \pm SD		
7	2.53 \pm 0.52	1.87 \pm 0.35	0.66 \pm 0.17	5.292	<0.001 S
14		1.53 \pm 0.52	1.00 \pm 0.00	10.247	<0.001 S
21		0.73 \pm 0.59	1.80 \pm 0.17	16.837	<0.001 S

Statistical Analysis: Paired t test.

Statistically significant if $P < 0.05$

Table 2:- Comparison of Means of Plaque index (PII) between Group I and Group II.

Plaque index (PII)					
DAY	GROUP I	GROUP II	Difference	Z value	P value
	MEAN \pm SD	MEAN \pm SD	MEAN \pm SD		
0	2.31 \pm 0.48	2.60 \pm 0.51	0.29 \pm 0.03	-1.581	0.114 NS
7	1.13 \pm 0.34	1.73 \pm 0.59	0.60 \pm 0.25	-3.331	0.001 S
14	1.00 \pm 0.00	1.20 \pm 0.41	0.20 \pm 0.41	-1.852	0.064 NS
21	1.00 \pm 0.00	1.00 \pm 0.00	0.00 \pm 0.00	0.000	1.000 NS

Statistical Analysis: Mann Whitney U test.

Statistically significant if $P < 0.05$

Table 2a:- Comparison of Means of Plaque index (PII) in Group I with different time periods.

Plaque index (PII)					
DAY	GROUP I : Day 0	GROUP I	Difference	t value	P value
	MEAN±SD	MEAN±SD	MEAN±SD		
7	2.31±0.48	1.13±0.34	1.19±0.14	11.783	<0.001 S
14		1.00±0.00	1.31±0.48	10.967	<0.001 S
21		1.00±0.00	1.31±0.48	10.967	<0.001 S

Statistical Analysis: Paired t test.

Statistically significant if P<0.05

Table 2b:- Comparison of Means of Plaque index (PII) in Group II with different time periods.

Plaque index					
DAY	GROUP II: Day 0	GROUP II	Difference	t value	P value
	MEAN±SD	MEAN±SD	MEAN±SD		
7	2.60±0.51	1.73±0.59	0.87±0.00	4.516	<0.001 S
14		1.20±0.41	1.40±0.00	10.693	<0.001 S
21		1.00±0.00	1.60±0.00	12.220	<0.001 S

Statistical Analysis: Paired t test.

Statistically significant if P<0.05

Table 3:- Comparison of Means of Probing depth (PD) between Group I and Group II.

Probing depth (PD)					
DAY	GROUP I	GROUP II	Difference	Z value	P value
	MEAN±SD	MEAN±SD	MEAN±SD		
0	6.19±0.75	5.73±0.59	0.46±0.16	-1.774	0.076 NS
7	4.25±0.58	4.07±0.46	0.18±0.12	-1.020	0.308 NS
14	3.31±0.48	3.40±0.51	0.09±0.03	-.501	0.617 NS
21	3.19±0.40	3.00±0.38	0.19±0.02	-1.299	0.194 NS

Statistical Analysis: Mann Whitney U test.

Statistically significant if P<0.05

Table 3a:- Comparison of Means of Probing depth (PD) in Group I with different time periods.

Probing depth (PD)					
DAY	GROUP I : Day 0	GROUP I	Difference	t value	P value
	MEAN±SD	MEAN±SD	MEAN±SD		
7	6.19±0.75	4.25±0.58	1.94±0.17	13.508	<0.001 S
14		3.31±0.48	2.88±0.27	15.999	<0.001 S
21		3.19±0.40	3.00±0.35	16.432	<0.001 S

Statistical Analysis: Paired t test.

Statistically significant if P<0.05

Table 3b:- Comparison of Means of Probing depth (PD) in Group II with different time periods.

Probing depth (PD)					
DAY	GROUP II: Day 0	GROUP II	Difference	t value	P value
	MEAN±SD	MEAN±SD	MEAN±SD		
7	5.73±0.59	4.07±0.46	1.66±0.00	13.229	<0.001 S
14		3.40±0.51	2.33±0.00	12.486	<0.001 S
21		3.00±0.38	2.73±0.00	23.127	<0.001 S

Statistical Analysis: Paired t test.

Statistically significant if P<0.05

Table 4:- Comparison of Means of Clinical attachment level (CAL) between Group I and Group II.

Clinical attachment level (CAL)					
DAY	GROUP I	GROUP II	Difference	Z value	P value
	MEAN±SD	MEAN±SD	MEAN±SD		
0	6.63±0.62	6.13±0.64	0.50±0.02	-1.960	0.050 NS
7	4.38±0.89	4.33±0.62	0.05±0.27	-.592	0.554 NS
14	3.25±0.45	3.67±0.62	0.42±0.17	-2.012	0.044 S
21	3.19±0.66	3.47±0.52	0.28±0.14	-1.188	0.235 NS

Statistical Analysis: Mann Whitney U test.

Statistically significant if P<0.05

Table 4a:- Comparison of Means of Clinical attachment level (CAL) in Group I with different time periods.

Clinical attachment level (CAL)					
DAY	GROUP I : Day 0	GROUP I	Difference	t value	P value
	MEAN±SD	MEAN±SD	MEAN±SD		
7	6.63±0.62	4.38±0.89	2.26±0.27	9.668	<0.001 S
14		3.25±0.45	3.38±0.17	18.781	<0.001 S
21		3.19±0.66	3.44±0.04	15.413	<0.001 S

Statistical Analysis: Paired t test.

Statistically significant if P<0.0

Table 4b:- Comparison of Means of Clinical attachment level (CAL) in Group II with different time periods.

clinical attachment level (CAL)					
DAY	GROUP II: Day 0	GROUP II	Difference	t value	P value
	MEAN±SD	MEAN±SD	MEAN±SD		
7	6.13±0.64	4.33±0.62	1.80±0.02	16.837	<0.001 S
14		3.67±0.62	2.46±0.02	12.854	<0.001 S
21		3.47±0.52	2.66±0.12	21.166	<0.001 S

Statistical Analysis: Paired t test.

Statistically significant if P<0.05

Discussion:-

Sub gingival plaque control and removal is a fundamental objective of periodontal therapy. Conventional close debridement of sub gingival plaque from root surfaces can be effective, but is time consuming and technically demanding. It has been shown that the chance of failing to remove all of the sub gingival plaque increases in pocket depths greater than 4 mm. As sub gingival plaque accumulates and matures, periodontal breakdown can occur. Therefore, professional maintenance therapy may not always achieve complete or adequate debridement. This has led to the adjunctive use of antibacterial agents, usually in the form of irrigants or systemic antibiotics, to overcome the limited efficacy of the conventional treatment. The primary purpose of irrigation is to non-specifically reduce the bacteria and their by-products which prevent the initiation or progression of periodontal diseases.

In any case, insertion of an identical irrigation needle 3 mm inside a deep periodontal pocket is sufficient to irrigate the apical plaque border (**Hardy et al. 1982**).⁹ In Our study patients were advised to carry out the irrigation procedure once daily at night for 14days. Blunt irrigation tips were given to prevent the injury and supervising the patients until they were able to irrigate the involved pockets without problems.

The results of the present study showed that sub gingival irrigation with octenidol and Chlorhexidine are effective in reducing periodontal inflammation and in controlling sub gingival plaque. A significant reduction in periodontal inflammation and clinical parameters from base line to 14days were observed and maintained for 21 days without irrigation.

On evaluation of mean sulcus bleeding index between the two groups were done Octenidol group has significant reduction of mean SBI than Chlorhexidine group. This reduction in clinical parameter up to 21st day in octenidol

group may be attributed to well known antimicrobial effects of octenidol and resolution of gingival inflammation. This reduction in bleeding and improvement in gingival health of patients using 0.1% octenidine formulation was suggested by decreased gingival fluid. These results were in accordance with **Patters M R et al (1983)**² stated that Octenidol is a highly effective bactericide against the ten tested representatives in the leading germ group of infectious diseases of the periodontitis and gingivitis types.

When comparison of means of Plaque Index (PII) in both Groups: octenidol group showed significant improvement by day 7 than Chlorhexidine group. This is due to antiplaque activity of octenidol. At a 0.1% concentration, Octenidine appeared to completely inhibit plaque formation over a 7 day period. These results were in accordance with **Patters M R (1983)**² and **Emilson et al (1980)**⁵. However by day 14 and 21 no significant difference was observed in both the groups.

When mean probing depths and clinical attachment levels were compared in between the groups there was no significant differences was observed, where as when compared within the groups both groups had showed significant pocket depth reduction and clinical attachment level gain. This may be due to reduction in tissue inflammation by the decrease in plaque and sulcus bleeding index scores. This result goes in concurrence with **David L et al (1990)**.¹⁰

Limitations of the present study include: short sample size, short duration. Microbiological evaluation after sub gingival irrigation was not considered.

Conclusion:-

0.1% octenidol and 0.2% Chlorhexidine applied directly to periodontal pockets reduces periodontal inflammation, even in the absence of effective interdental aids. The level of inflammation is maintained below baseline values for 3 weeks after the two week irrigation period in both the groups. In the light of our results it was stated that sub gingival irrigation with octenidol and Chlorhexidine are effective in reducing periodontal inflammation and in controlling sub gingival plaque. There was no difference found in all the parameters with both groups except for SBI which showed slightly superior effect for octenidol than Chlorhexidine, yet long term studies are to be conducted to know the efficacy and Substantivity of octenidol mouth rinse.

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RESEARCH ARTICLE

THE ESSENCE OF JUSTICE FOR THE LIVING LAW IN THE ALTERNATIVE DISPUTE RESOLUTION OF INDONESIA LEGAL SYSTEM.

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Abstract

The Alternative Dispute Resolution (ADR) In Indonesia Legal System, is a model of dispute resolution which based on living law in Indonesia society. This model is often used other than the settlement of dispute through the court by Indonesia people. Whereas, the settlement of dispute through the court can guarantee legal certainty, because as guided by the statutory law. Based on that statement, this research describes essence of justice for the living law In the ADR In Indonesia Legal System. This research uses the type of normative legal research. It based on conceptual and regulation approach. This research is analysed with qualitative descriptive in using. The result of research shows that essence of justice which based on the living law in ADR, in principle, it focuses on the substantive justice for the sake of harmonization and peacefulness which based on cohesive living value in existence. It still appreciates the procedure of applicable in the Indonesia Society, such as colloquy based on kinship. So that, it yields for society's legitimated justice.

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Introduction:-

Dispute sometimes comes up in our life as a consequence of social relations between individuals or groups to each other. Folberg and Taylor (I Made Widnyana, 2007), argue that there are two types of disputes, among others: (1). intrapersonal conflict or conflict within the individual; and (2) interpersonal conflict or intergroup conflict. According to Folberg and Taylor's opinion, the second type can be considered as a conflict in the scope of legal object. The dispute contains conflict of law in relationship between community members, basically, it could lead disharmony because there are those whose neglected rights. The injured party, usually, will express dissatisfaction to the other. Since, they feel to get in loss. Based on that, in Indonesia society context, there is alternative legal instrument and legal mechanism which can be used as an attempt to resolve the dispute in order to restore such rights, namely the the settlement of disputes through the courts (litigation) and the the settlement of disputes through the outside court (non litigation).

Settlement of dispute by using a model of ADR, is also an integral part of the effort to implement progressive law as conceived by Professor Satjipto Rahardjo. It is more accommodating legal values which lives in Indonesian society context, which accentuates the legal sense for Indonesian people than the statutory law which emphasizes the ruling class' political will. This is often not accordance with the values of justice Indonesian society itself (Muhammad Jufri and Deity Yuningsih, 2016).

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In Indonesia, Legal effort through the court (litigation) is usually taken by people. But this effort does always depletes vigor, long time, and high cost. However, it requires the ability of law proceeding in the court. Settling of dispute through the court would produce a legal certainty since the judge's decision in winner and loser. The winner will take all of the loser, while the loser will feel dissapointed. For the people of Indonesia, it can terminate relationship and it will cause the new conflict.

There legal effort which does performance in Indonesia's legal system, namely the Alternative Dispute Resolution (ADR). The parties in dispute can choose it. This is an alternative dispute which not through the court. It is set out in Regulation of Number of 12 of 1999 on Arbitration and the Alternative Dispute Resolution. The ADR is one of the institutions for dispute resolution which is agreed by the parties in conflict. It is done which not involved the court by performance of negotiation, consultation, mediation, conciliation, or the involvement of legal expert (see Indonesia Regulation of Number of 12 of 1999).

The ADR, in principle, especially in consultation, negotiation, and mediation. It is recognized by Indonesia people as a form of dispute resolution in outside court through deliberation. This is known as a living law in Indonesia people since old time. In this context, the author does to research on ADR of Indonesia people as a living law in an effort to obtain justice. Actually, the living law in the context of Indonesia people, has been integrating into customary law as it is known as *adat* law. It is an unwritten law which does accommodate the values of justice in the legal sense of Indonesia people. Court decision, vice versa, it will be guided by statutory law which does glorify legal certainty.

Method of Research and Materials:-

This research uses normative legal research which is to examine basic norms in Indonesia's positive legal (Johny Ibrahim, 2008). This research, also uses regulation and conceptual approach (Peter Mahmud Marzuki, 2007). Regulation conceptual is useful for Indonesia regulation which concerned with legal issues of ADR, while the conceptual approach is used to analyze ADR in living law based Indonesia's legal system. Therefore, the conceptual approach is based on jurists' opinion and legal experts' view. This research uses written legal materials as analysis instrument, containing primer legal materials such as Indonesia Regulations on ADR, and secondary legal materials such as text books, law journal, law article, and scientific paper on law. Author analyses this topic research using interpretation of legal material substance based on legal theory and legal principle, in order to explain the nature of justice for ADR in Indonesia Living Law.

Result and Discussion:-

The result of research shows that living law in the Indonesia society of context, it becomes the main runway as a guidelines for dispute resolution of outside the court. The living law usually does include customary law (*adat* law). In Indonesia, dispute resolution of outside the court or ADR, it can be held if the parties in quarrel to choose it. There is a reason that dispute resolution of outside the court is preferable than through court. Because the settlement of dispute through the court has long procedure and convoluted. For example, Sasak Tribe located in Lombok, Nusa Tenggara Barat, is one of provinces of Indonesia. Sasak Tribe usually uses ADR to resolve disputes occurring among each other. Sasak people argue that settlement of dispute in ADR using, is preferred. ADR accommodates legal value contained in customary law (*adat* law). In Sasa Tribe, there is an institution in charge of maintaining the *adat* law. It is known as "Krama". It seems that this institutotion performing the role for dispute resolution of outside the court such as ADR. "Krama" has authority to hear the case based on *adat* law, and it imposes sanction of *adat* law. This institution is unique. It resolve disputes in Sasak Tribe not to issue a decision in lose or win, but its win-win solution. The leaders of "Kara" who act as judge attempts to give advice for parties in obedience of *adat* law as the living law. They suggest to their people for improving the quality of understanding on *adat* law values based on local wisdom. It is intended to achive a harmonious common life in worthy and well becoming (Syapruddin, 2014).

According to Hadikusuma (2003), dispute resolution of outside the court, in Sasak Tribe, among heirs is resolved based on five of *adat* law's principles. Those include in Indonesia tribes, as follows:

- a. Principle of divine and principle of self-control (*betel leq reden neneq* in Sasak Language);
- b. Principle of equal rihts and togetherness (*doe sopog, bareng ngepe* in Sasak Language);
- c. Principle of harmony and kinship (*awak sopog, saling peririq, saling angkat, saling ajinin, saling sedok* in Sasak Language);
- d. Principle of deliberation and consensus (*Soloh*);

e. Principle of Justice and *Parimirma* (*endeq naraq bine kire, tarik nyacap*)

Another example, Tolaki Tribe, is one of tribes living in Southeast Province of Indonesia. Rahmat Saini (2015) wrote that dispute on family law, such as inheritance problem, divorce, and quarrel. It was solved by using of Tolaki's *Adat* Law. Parties in dispute would complain to the chairman of village, known as *Kepala Desa*. In Rahmat Saini's writing that *Kepala Desa* with along prominent leaders of *Adat* Law, would do summoning for parties, they collected evidences and informations for unpleasantness among the parties. It would be resolved according to Tolakinese of *Adat* Law.

Until now, Tolakinese views that Tolaki's *Adat* Law still in keeping of justice, peacefulness, and welfare. Decision of Tolaki's *Adat* Law ties up Tolaki Tribe without differentiating of social status. Everyone who breaks *Adat* Law, it does consider as a person in saucy deed, so that he or she will be ostracized.

The ADR concept, it does provide creation for a good relationship among disputed parties. In author's view, basically, it is one way for dispute solution or alternative solution in settlement dispute without court procedure. In society's Indonesia legal culture, especially, legal conflict is solved through amicable way based on values in harmony and recognition of living law. However, the ADR concept, it does accomodate the living law as values recognized by society itself. The ADR concept does consider dynamic dispute resolution. This is very simpel because it can be done through co-operative solution. Also, it can be the model of dispute resolution which generates win-win solution.

In Von Benda Beckman's view that dispute resolution or dispute of legal conflict in simple condition of society where kinship or relationship is still very tight, it is usually there institution as a choice which settles conflict based on folk institution. This institution is the traditional institution which acts as guard regularity and magical balance in society itself (Sahnan, 2010).

Indonesia Legal System admits two kinds of legal format, as follows: Written Legal such as statutory law and Unwritten Legal such as customary law and *adat* law. Later, there are legal values which do provide living law as legal obeyed by Indonesia people. *Adat* Law contains them. *Adat* Law, actually, is unwritten legal which does not require a specific procedure. Unlike the written legal which requires a specific procedure. *Adat* law and customary law for Indonesia people is an actual law. Since it usually obeyed duly by Indonesia people.

In Professor Soepomo's view that *adat* law in Indonesia is living law since it is the embodiment of real legal sense of Indonesia people. *Adat* law will continue which follows headway of society itself. This opinion, also, is followed by Frederich Carl Von Savigny (1779-1861). He said that there was a legal embodied into people's soul (*volkgeist*) where law would flourish to expand with society itself. This opinion is known as Historical Jurisprudence. One of schools of law is followed by scholars of *adat* law in Indonesia, including Dr Guswan Hakim and Dr Ruliah, both lectures of Law Faculty, Halu Oleo University, located in Southeast Sulawesi Province of Indonesia (Jufrina Rizal, 2008).

There is same opinion expressed by Eugen Erlich. He is one of the pioneers of sociology of law. He said that Savigny's view on *volkgeist* was fact of law as a living law in the society. It was a living law of the people. Eugen Erlich said that the centre of gravity of legal development was not placed in legislation nor in juristic science, nor in judicial decision, but in society itself. Eugen Erlich, actually, is the first who used the term of "living law" in his popular book titled *Grundlegung der Sociologie des Recht*. This book has been translated into English "*Fundamental Principles of Sociology of Law*".

Professor Satjipto Rahardjo is one of Indonesia experts for sociology of law. He also agrees towards existence of *adat* law as a living law which reflects legal idea and legal sense of Indonesia people. On the other hand, Professor Djojodiguno explains *adat* law as the living law which in human creation on legal values. It can be flourished in evolution with its elasticity, so it provides different dispute resolution for the case in different legal event. Especially, the problem of rights and obligations in dispute (Deity Yuningsih, 2016).

The scholars of *adat* law in Indonesia have argument that *adat* law is simple and dynamic. It is not rigid which will receive the change and be responsive towards values in Indonesia society. Changing values is realistic in society. *Adat* law, in principle, stresses on substantive justice in order to reflect harmony and peacefulness based on compact

values in existence. This reason why *adat* law in frame work of dispute resolution, it based on living law. To reflect realistic substantive justice, then *adat* law has self-mechanism for dispute resolution. This mechanism does use deliberation in kinship which provides realistic justice as values legitimated by society itself.

In view of Robert Seidman, in his theory on “the law of the non transferability of law”, says that law could not be transferred away from society to another. Because law does not always permanently attached in a particular community. Also, not to be considered as traditional law in legal retardation (Deity Yuningsih, 2011). It means that different law can be effective for such society, since it can guarantee legal certainty and justice. However, such legal can provide justice in accordance with the needs of law for society itself.

According to author’s point, law as living values, it should be kept in order to guarantee social justice. In the Indonesia legal system context, *adat* law still does provide legal justice in accordance with legal awareness of Indonesia people. Therefore, effort to be positive legal in statutory law whereas legal values containing in it, not in accordance with needs of law for society itself. It will effect new conflict in society, including rejection of society.

Closing:-

Living law is a main principle used as the guidance in dispute resolution for Indonesia people, it does resolve disputes in outside court. ADR, basically, it can become a solution in dispute without the court. In Indonesia society context, ADR is very important because it accommodates the living law based on kinship in legitimated harmony. ADR for dynamic or simple people is suitable since it prioritizes co-operative solution and win-win solution.

The nature of justice in ADR based on living law, in principle, it more emphasizes on substantive justice in order to reflect harmonization and justice in compact legal values. The meaning of living law in this context, it based on *adat* law which recognizes exist procedure, such as colloquy in kinship. So that, it can achieve the justice as societal expectations in legitimacy. However, enforcement for living law depends on the professionalism of traditional leaders. Based on that, it is very important to give legal understanding and legal training on legal procedure, scope, including legal force in ADR based on living law in Indonesia society context.

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RESEARCH ARTICLE

STUDY TO ANALYSE AND COMPARE THE QUALITY OF LIFE IN PATIENTS OF ADVANCED STAGE LUNG CANCER UNDERGOING TWO DIFFERENT MODALITIES OF TREATMENT.

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Abstract

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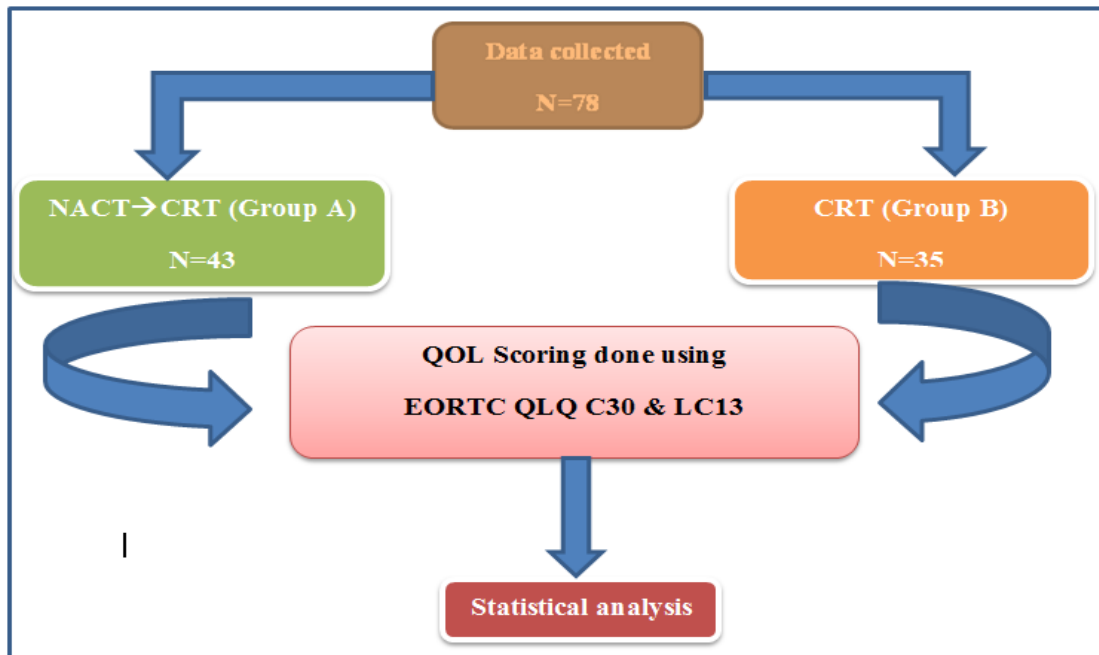
Introduction:-

Lung cancer is one of the leading causes of mortality and morbidity worldwide. In view of high morbidity and symptom burden, quality of life (QOL) is an important parameter to measure. Incorporation of the assessment of QOL as part of daily oncology practice may enhance current treatment strategies for cancer patients [1]. In addition, enhanced understanding of the common QOL of patients enables to improve the clinical care of patients [2]. We in this study have tried to analyse and compare the quality of life in patients of advanced stage lung cancer undergoing two different modalities of treatment.

Material and Method:-

Data of 78 patients of advanced staged (stage III A & B) carcinoma lung was collected and was randomised according to the treatment received followed by QOL scoring using Quality of life Questionnaire Core 30 (QLQC30) and Lung Cancer Module (LC13), version 3.0. Results were subsequently analysed statistically using SPSS software version 20. Flow chart illustrating the work flow is shown in Fig-1.

Fig-1.Flow chart depicting work flow.

**Result:-****Table 1:-** Baseline characteristics of 78 lung cancer patients.

Characteristic	Categories	Number	Percent
Age at presentation (years)	Mean	56	
	Median	58	
	Range	42-74	
Patients	Group A	43	55.12
	Group B	35	44.88
	Total	78	100
Stage	Stage III A	47	60.25
	Stage III B	31	39.75
Treatment History	NACT→CRT	43	55.12
	CRT	35	44.88

Table 2:- Baseline QoL (C30) scores of 78 lung cancer patients.

QLQ-C30 Scale	Mean	Median	SD	Range
Global	53.6	58.3	26.3	0-100
Physical	68.7	73.3	25.5	0-100
Role	58.9	66.7	34.3	0-100
Emotional	64.1	66.7	25.8	0-100
Cognitive	74.9	83.3	25.9	0-100
Social	61.2	66.7	32.9	0-100
Fatigue	46.8	44.4	28.4	0-100
Nausea/Vomiting	14.5	0	22.2	0-100
Pain	37.1	33.3	32.8	0-100
Dyspnea	37.6	33.3	33.6	0-100
Insomnia	39.9	33.3	33.5	0-100
Appetite Loss	31.3	33.3	34.1	0-100
Constipation	24.2	0	31.3	0-100
Diarrhea	9.9	0	19.7	0-100

Table 3:- Baseline QoL (LC13) scores of 78 lung cancer patients.

LC13	Mean	Median	SD	Range
Cough&Hemoptysis	58.9	66.7	34.3	0-100
Shortness Of Breath	64.1	66.7	25.8	0-100
Swallowing,Tingling	74.9	83.3	25.9	0-100
Pain & Pain relief	61.2	66.7	32.9	0-100

Table 4:- Multivariate Cox regression analysis for QoL function and symptom scales.

QoL Domain	HR	95% CI	P-value
Physical	0.99	0.986 to 0.994	< 0.001
Role	0.999	0.995 to 1.002	0.5
Emotional	1.001	0.997 to 1.005	0.58
Cognitive	1	0.996 to 1.003	0.8
Social	1	0.996 to 1.003	0.85
Fatigue	0.996	0.992 to 1.001	0.16
Nausea/vomiting	0.997	0.993 to 1.001	0.2
Pain	1.002	0.999 to 1.005	0.16
Dyspnea	1.001	0.999 to 1.004	0.34
Appetite Loss	1.003	1.000 to 1.005	0.07
Constipation	1	0.998 to 1.003	0.73
Diarrhea	1.003	0.999 to 1.006	0.16

Discussion:-

In our study we tried to evaluate and compare the effect on QOL in patients receiving either modality of treatment i.e NACT→CRT or CRT alone in patients of advanced stage carcinoma lung and found that QOL is better off in patients receiving CRT alone, this probably could be due to more toxicity profile with chemotherapy received by patients in both neoadjuvant as well as in concurrent setting in group A. Study done by J. A. Langendijk et al showed chemoradiation was superior to chemotherapy alone with respect to survival and HRQoL at the expense of more hospital admissions due to toxicity[4]. In another study done by Shintaro Traumani et al, they found that pulmonary rehabilitation program for patients with non-small cell lung cancer undergoing induction chemoradiotherapy seems to improve respiratory function. It is particularly recommended for smokers and patients with respiratory impairment [5]. In a study done by Bottomley A, radical radiotherapy offers palliation of respiratory symptoms and improved QoL in a substantial proportion of patients with NSCLC who have relatively good prognostic features [6].

Conclusion:-

Quality of life is better off with single modality treatment when compared to double modality of treatment.

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RESEARCH ARTICLE

AN ETHNOGRAPHIC ACCOUNT ON THE MEANING, STRATEGIES, AND DIMENSIONS OF STREET VENDING IN AN URBAN SETTING OF KOLKATA, WEST BENGAL.

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Key words:-

Ethnography, Hawker, Street Vending, Informal Sectors, Crisis, Strategies, Dimensions, Kolkata.

Abstract

In most of the third world countries like the Indian cities, informal economic livelihood activities have grown immensely over the past decade due to lack of employment opportunities in formal sectors of the urban areas in the spaces like-Kolkata, West Bengal. Specifically the urban 'poor' through such activities have gained employment and provide low cost services to a large section of the middle class residents of the area under study.

Several studies have already shown that street vendors are one of the most 'marginalised' and 'vulnerable' sections of the urban and sub-urban Indian context. To perceive the meaning, strategies, livelihood, and other dimensions of this sector, the present authors purposively selects "street vendors" including both perishable and non-perishable commodity sellers of the area under present research.

The mode and approach of one year long research is grounded on the ethnographic approach and methods from designing, framing, collection of data, triangulation, transcription, data analysis and interpretation of the findings. The data were collected through semi structured and in-depth interviews and intensive case studies.

The current effort reflects a detailed understanding of the processes and networking of informal sector that could provide an open scope for future research and innovative regulatory programmes, rather than opposing the informal workers, in part of the Government, initiatives should take into consideration in the process of implementing policies for the hawkers. The reflection of the present and in-depth ethnographic study represents a particular section of informal earning groups, but the present authors expects the research as a case and instance for better policy and actions of the area under study.

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Introduction:-

Street vending or selling goods, commodities, and services in the public space is one of the most common, accessible, and momentous occupational categories in the context of globalized informal economy, extending its

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horizons in the developing countries due to limited scope of employment in the 'organized' and/or 'formal' sectors; concurrently stimulates the opportunities among the academicians, researchers, socio-developmental activists, administrative personnel, and policy makers for conducting in-depth and effective researches in the domain of informal sectors (Vargas and Urinboyev, 2015; Saha, 2011; Timalisina, 2011; ILO, 2004). The concept of informal sector was first introduced by Hart in 1971 (Bekkers and Stoffers, 1995), but the essence of the field is embedded with the earlier impression of 'unorganised sector' that indicates 'small' size production units (Kabra, 1995). The 'unorganised' and 'domestic' nature of informal economy may also be a part of 'non-monetary' sector (GOI, 1951). Kabra (1995) indicates that the terminological diversities (including the term-'survival sector', 'non-structured sector', 'transitional activities' and so on) and debates to contextualise and localise the practices of informal economy in classical and contemporary researches, reflects the dynamic and complex nature of the issues concerned. Thus came, the priorities, angles and frames of research in informal economic practices are multilayered and manifold, is undergoing through major shifts and changes in its discussion and interpretation of living in informal mode of economy as a phenomenon, types of labour employed, productivity and contribution of the agencies associated to Gross Domestic Product, and the issues of policy intervention (Williams and Tumusiime-Mutebile, 1978; Tokman, 1990; ILO, 1972; Kabra, 1995; Charmes, 1990; Arye, 1981; Bromley, 1978).

Street vending as a significant manifestation of informal economy varies in terms of scale, timing, location, remuneration, workforce, types of goods sold and services provided, and it can be a full-time occupation, a part-time occupation, seasonal, and/or occasional dealings (Rosales, 2013; Bhowmik, 2005). The process of 'global-restructuring' instigates, accelerates, and expands the scope for individuals to incline towards street vending as a profession to serve the specialized classes in the global cities (DeLuca, 2012; Stoller, 2002). The profession of street vending can be carried out by one person but can also be a franchise of a larger street business. It can be a survival strategy or part of a big business. The incomes of street vendors consequently vary widely (Ray and Mishra, 2011).

In urban India, street vending is the largest informal sector serving mainly the 'urban poor' faces great difficulties and challenges since the era of 'economic reform' in this country (Srivastava et al. 2012; Ray and Mishra, 2011). The 'old Indian cities' are going through the rapid pace of globalization and urbanization that promotes the restructuring of the cities with different developmental strategies, including the formation of new markets, but, these reforming strategies does not effectively attempts to develop the 'old' markets, and/or rarely implements any strategy related to the alternative space for the street vendors, even in the context of a rapid increasing of participation in street vending among the immigrant population (Avigna AS, 2010). Moreover, because they sell cheaper products, the street vendors often cater for the urban poor (Ray and Mishra, 2011). There are no labour standards at the international level dealing directly with street vendors. The manner in which street vending is approached at the domestic level varies dramatically from one country to another. Many countries specifically regulate this activity, providing a clear legal framework and jurisdictional mandates, while others have an overlapping jurisdictional mandate, which creates confusion and conflict, and in others still, street hawking is simply considered illegal (Ray and Mishra, 2011).

The Objectives:-

The Present study is set to explore-

- a) The perception and lived experiences of hawking and street vending and the crisis associated with the profession from an insider's perspective.
- b) The meaning, strategies and other related dimensions of street vending to find out the meaningful alternatives for better policy and action.

Methodology:-

The current study has been conducted in the Sealdah region of Kolkata, West Bengal. The area has been selected for study due to three major reasons- i) the area is surrounded by almost two hundred (200) hawkers and multiple kinds of perishable and non perishable commodities; ii) the consumers are frequent to the region and they are from both urban and suburban premises of the state, and iii) the authors are well aware of the concerned market place and their communication with the principal agency (the hawkers) of the research were convenient to the area.

Fifty (50) respondents were selected through a primary informative schedule that included name, age, sex, the migratory information, location of the stall, selling time and duration, types of items or commodities, and perception about target consumers. After having the baseline data from the schedule, the researchers have gone through the data minutely and prioritized twenty five (25) respondents for depth interviews.

The core nature of the research is inclined to ethnographic approach and methods. The data related to the issues of socio-economic details, the process of vending, interactive pattern with the consumers, the crisis of the profession, emotional orientations towards the space, the maintenance of the local network, and the broker and mediator issues have been collected by using semi-structured and depth interviews. Field notes were collected during and after the fieldwork that facilitated data analysis and to endorse meaningful insights to the findings.

The collected data have been transcribed verbatim to include the insider's perspective of the issues concerned. After the phase of transcription, the data have been sorted out and significant findings have been marked to proceed the data to the next stage of analysis. The primary data have been analyzed by using narrative data analysis method. After analysing the data, the findings were contextualised with the research frame and question, and interpreted accordingly.

The Findings:-

A) "Street Vending"- getting into the Profession:-

The degree of choices of the participants to the street vending as profession varies from independent selection to situational inconveniences. Smt. Pratima Basak has opined that- *after my husband's sudden death there was a single option left to me i.e. to continue my husband's occupation...before his death I was a house-wife and we have no child...at the very outset the profession seemed to me very difficult...I had no idea how to manage the consumers....but gradually things became habituated...one of his friends (is a fruit seller like my husband) helped me a lot....now I can estimate who will buy and who not...how to behave with a regular customer and how to handle those who is just bargaining.*

Mr. Bhola Baidya (a vegetable seller) chose his profession independently. He is from a family who depends on cultivation as their main earning source. He opined that- *I have chosen the profession independently...we have our own land for cultivation....at first I used to sell vegetables at my local area...I am here for last six years...the consumers here are of different kinds-some are almost regular and known to me very well who buy on the way return from their offices...sometimes they ordered many vegetables which I am not used to sell...but I bring that for them...after all they are my fixed customers...*

B) Communicating with the Consumers and dealing with the Chaos:-

Street market gives a larger section of the urban people specially the middle class fresh products and traditional food in relatively lower price in compare to the super markets or malls. As a profession street vending always needs several strategies to gain profit in such a congested crowded place competing with others by sensing consumer's mentality from their choices and preferences of items and sometimes influences the shopping behaviour of the customers. Md. Sipahi, aged 40 years, fruit seller in the road side stated- *I have some fixed customers who visited everyday evening while returning from office.... I generally prefer to make them wait for few minutes if there is rush... I sell them fresh fruits in low rate... even I sense the mentality of a unknown customer by seeing his/her face that whether he/she came to buy or just to check and compare rates with other shops. Sometimes they also give intra-personal signals to their known customers in presence of others to not to take fruits or vegetables if they are not in good condition but they sell them with smiling face to others. To earn profit I need to sell fruits in higher rates and even the rotten ones from unsold stock of previous day to unknown customers... ummm... if my fixed customers are there at that time I just simply "blinking my eye" to signal them to keep quiet and wait....soon after the customer left off I come out with fresh fruits to the known ones....sometimes I also give credit if the wont have change – as said by another fruit seller Mr. Sambhunath Saha.*

C) The Problems, Crisis, and the Major Challenges of Street Vending:-

A number of daily felt problems were reported by the hawkers which are presented here categorically -

i) Lack of Drinking water and Toilet facilities:-

The place is devoid of facilities like drinking water as well as toilets. To respond to the nature's call they even have to walk for more than five minutes. In voice of Gorabini Seal (50yrs), 'vegetable seller' - *there is neither toilet nearby nor any drinking water...we have to walk to either Sealdha station or to Shradhananda Park... According to Nirapada Bhounik, a 'readymade garment seller', 50 yrs old - the principal trouble is felt by all hawkers that only toilet is there at Sradhananda Park...a walking distance of five minutes from hawking spot...which is not convenient when there is a pressure of customers.*

ii) Threats and Troubles from the Local Mediators/Brokers:-

In response to the commonly felt need to organize them from within, initiatives were taken long time back in the 70s and soon after it was termed as 'Kolkata Street Hawkers Union', affiliated to a political party. But problems arose when it comes to the issue of money lending and the lenders were the local political party leaders as well as the members of the union. Mr. Dharmendar Prasad, a 40 yrs middle aged 'jhal-mudi-wala' stated that - *problem is mainly financial...there is no saving, but the family is large...if any member of the family is sick then we have to borrow the money from our union leaders...the amount of interest is 10% monthly which is quite high for me...due to the fear of losing selling-space, we become silent....our struggle for social security purposes.* Mr. Mujafar Khan (54 yrs old), a 'vegetable seller' for the last 25 years, stated that - *the rate of interest for hawkers like us (perishable commodities) is much higher than other kind of hawkers of these place...even 20% per month...* As they are not recognized formally by the Government so they have no other alternatives but to borrow money from these private money lenders at a very high rate of interest which varies depends on their selling commodities.

iii) The Crisis and troubles associated with the Space:-

Very common problems faced by most of the hawkers are space they occupied for selling their commodities. The local union fixed 5'×3' for every hawker but sometimes it become 3'×3'. According to Mr. Chandan Mallik, who is regular in this place as 'sandal wood- rudraksha seller' for last 30 yrs stated that - *the space demarked by the union is not equal for all...in most cases, they are allotted accordance to personal choices of the union leaders...it is problematic for me to display all variety of items within this little place of merely 3'×3'.* In addition to that, according to Mr. Tapan Seal, 'cup-dish seller'...*we are always in a fear that if any day I will absent due to any reason, I may lose my selling place, there is always an additional pressure of some other hawkers who have no fixed place allotted by the local union.* Usually they occupy the space when a union recognized hawker is absent.

iv) Scarcity of Storage:-

There is no arrangement of preservation of fruits or vegetables, so unsold items become rotten, for which the prices are fixed after taking into consideration of all cost. According to 55 yrs old 'fruit seller' Mr. Swapan Roy - *I can't sell all the fruits which I purchase daily...I have to sell 3 kgs of mangoes at the price equal to the price of 5 kgs of mangoes...in this way I try to raise the purchasing price...if all fruits are not selling out daily...it is hard to earn profit.* Mr. Sadanada Sil (43 yrs of age), a regular 'vegetable seller' opined that - *the major problems for perishable commodity sellers like us is the absentee of storage room...apart from the burden of not sold vegetables storage...we also faced seasonal problems like coping with rain.*

v) Lack of Organized policies and Schemes:-

As there is no as such particular policy or scheme for legalising street vending in almost all cities of our country, corruption, in the form of bribery and extortion from all possible means as well as from police, eats the earnings of these people and reduces their income further. Mr. Krishna Das, a boot polisher was there for almost long 40 yrs said- *well my equipment values a small amount of Rs. 500/ but for doing business even in this foot path with this little amount I need to pay bribe to the local political leaders to sustain here... even police also did the same.... I won't tell you the amount... we all (indicating other vendors) are living in anxiety and struggling for long.* If Government will take initiatives then this gives a lever to the municipal authorities to earn more through license fees and street vending can also be regulated.

The Interpretation of the Findings: A Brief Overview:-

Each state government may have own planning and policies for development of its undertaken cities like- allocating space for hospitals, parks, markets, bus and rail terminuses etc. The present study intends to mirror that instead of implementing any comprehensive plans, they could take into account the alternative livelihood for hawkers for the successful and actual development of the city.

The present research are purposively restricted to portray in street vending to unearth a detailed account on the life, living and crises of the concerned respondents in the studied area. The qualitative parameters and themes developed on the basis of the narrative data analysis method from the findings of the study clearly suggests the perception, living the profession, the crises of the profession, and an hazardous and/or hectic situations to sustain the profession for a long period of time.

The present authors through their analytical findings with a very brief interpretation further instigates to intensive future and detailed research and recommendations to the governmental and other non-governmental agencies to

avoid these limitations on the multiple and different informal market oriented spaces and settings of the concerned city, as well as on the other cities in India for the issues of wellbeing and sustainable living of informal sectors professionals.

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RESEARCH ARTICLE

HYDROTHERMALISM ASSOCIATED TO THE UPPER VISEAN BASIC MAGMATIC ROCKS OF MACHRAA BEN ABBOU BASIN. (NORTHERN REHAMNA-WESTERN MESETA- MOROCCO).

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Abstract

Le bassin de Mechraa Ben Abbou (MBA) (Rehamna-Maroc) a connu au Viséen supérieur une intense activité magmatique basique.

L'analyse structurale par des coupes réalisées sur le terrain a montré que ces roches se mettent en place sous forme de sills. Ces sills suivent en concordance les ondulations dessinées par le plissement principal cartographique du synclinal de la Gada Jennabia. La minéralogie des différents facies identifiés reste comparable avec les minéraux principaux, plagioclases et clinopyroxènes. Ce sont des gabbros à texture grenue, des dolérites à textures optique, sub optique ou intersertale et des basaltes à texture microlitique porphyrique vacuolaire.

Dans ces roches l'altération se manifeste à l'échelle de l'affleurement par des fillonnets de calcite ou de quartz et au niveau des lames minces par des processus de séricitisation, de chloritisation, de carbonatation, de silicification, d'ouraltisation et d'oxydation. Ces processus d'altération peuvent être liés à la circulation de fluides hydrothermaux syn ou post-magmatique.

Les diagrammes géochimiques en éléments majeurs en fonction des pertes au feu ont permis de qualifier et de quantifier les effets de l'altération. La confrontation des données de terrain de point de vue structural avec les données microscopiques et géochimiques permettent de caler dans le temps les phénomènes d'altérations observés qui correspondent aux phases tectoniques hercyniennes.

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Introduction:-

Le magmatisme lié à l'orogénèse hercynienne au niveau de la Meseta marocaine a fait l'objet de plusieurs travaux de recherche (Jenny, 1974 ; Huvelin, 1977 ; Lagarde, 1987 ; Kharbouch, 1982 ; Kharbouch, 1994 ; Aarab, 1995 ; Essaifi et al., 1995 ; Mrini et al., 1992 ; Remmal, 2000 ; Remmal et al., 1997 ; Roddaz, 2000 ; Ben Abbou, 2001 ; Ben Abbou et al., 2001 ; Kholaiq et al., 2015b). Ce magmatisme est associé aux séries sédimentaires et métamorphiques du Carbonifère terminal.

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Dans le massif des Rehamna septentrionaux, le magmatisme préorogénique est impliqué dans le plissement hercynien d'âge Namuro- Westphalien selon Gigout, (1951). Il s'agit de dolérites et de gabbros sous forme de sills dans la série viséenne et de dykes dans la série du Dévonien inférieur.

Jenny, (1974) et Michard, (1976), signalent l'existence d'ophites et de dolérites en sills et dykes à Mechraa Ben Abbou. C'est ainsi que Jenny, (1974) à travers une coupe réalisée à Bled El Mekrech reprise par Saber, (1989) montre l'existence de sills de puissance métrique séparés par des minces couches métriques de sédiments sur le flanc Est de la Gada Jennabia d'âge Viséen supérieur.

Kharbouch, (1994) a distingué dans les Rehamna septentrionaux deux ensembles magmatiques :

- Un ensemble préorogénique formé par des roches hypovolcaniques et volcaniques ;
- Un ensemble post orogénique formé par des coulées et par des filons microdioritiques.

Elle conclue que les laves sont basiques et que les sills sont constitués de gabbros et de dolérites et que les intrusions sont plus importantes que les faciès effusifs. Les seuls pyroxènes analysés dans l'ensemble magmatique de barrage El Massira révèlent une tendance alcaline. Alors que Remmal et al., (1997) attribuent un caractère tholéitique continental au magmatisme des Rehamna occidentales. Ils signalent que les roches magmatiques sont surtout des dolérites et des gabbros sous forme de laccolites ou de filons sous-marins présentant des pillow-lavas.

Pour El Kamel, (2002) à Mechraa Ben Abbou les intrusions magmatiques sont effusives et intrusives de types alcalin à tholéitique dont leur mise en place est régie par une tectonique distensive attestée par la présence de failles normales syn-sédimentaires.

Les travaux récents Kholaiq et al, (2015b) distiguent au niveau du synclinal de la Gada Janabia des gabbros, des dolérites et de laves basaltiques dont les analyses géochimiques les attribuent à une série tholeitique orogénique continentale (Kholaiq et al., 2016).

Cadre géographique et géologique:-

Le massif hercynien des Rehamna se situe à 80 Km au Nord de Marrakech et à environ 150km au Sud de Casablanca (fig.1). Le bassin de Mechraa Ben Abbou (MBA), objet de notre étude, constitue la partie septentrionale de ce massif.

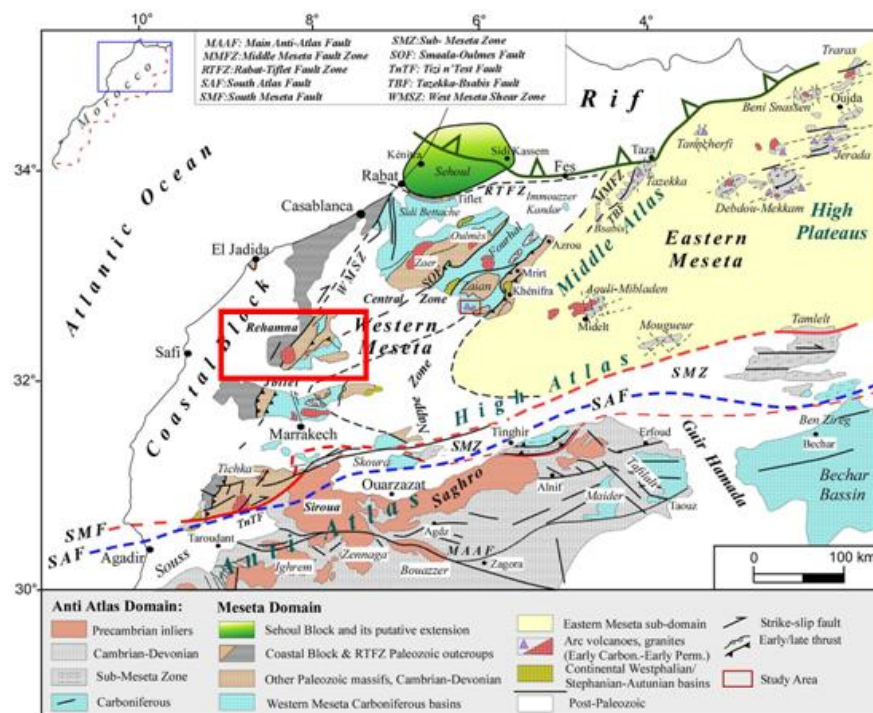


Fig. 1:- Situation du massif des Rehamna dans la Meseta occidentale marocaine (Ntarmouchant et al., 2016).

Le bassin de Mechraa Ben Abou montre des terrains paléozoïques allant du Dévonien inférieur au Viséen supérieur. Ces terrains présentent une diversification des faciès allant des détritiques, aux carbonatés et aux pélagiques, contemporains de la mise en place des roches magmatiques basiques (gabbros, dolérites et basaltes).

L'ensemble est structuré par la phase paroxysmale hercynienne sous forme d'un synclinal (Fig.2) (synclinal de la Gada Jennabia).

Les secteurs de Bled El Mekrech (coupe AB) et Cheikh El Basri (coupe CD) situés sur le flanc Est, le secteur de Lala El Gara (coupe EF) qui constitue la terminaison péri-synclinal et le secteur de Leffaid (coupe GH) situé sur le flanc Ouest, sont les quatre sites qui permettront d'approcher le magmatisme dans le bassin (Fig.2).

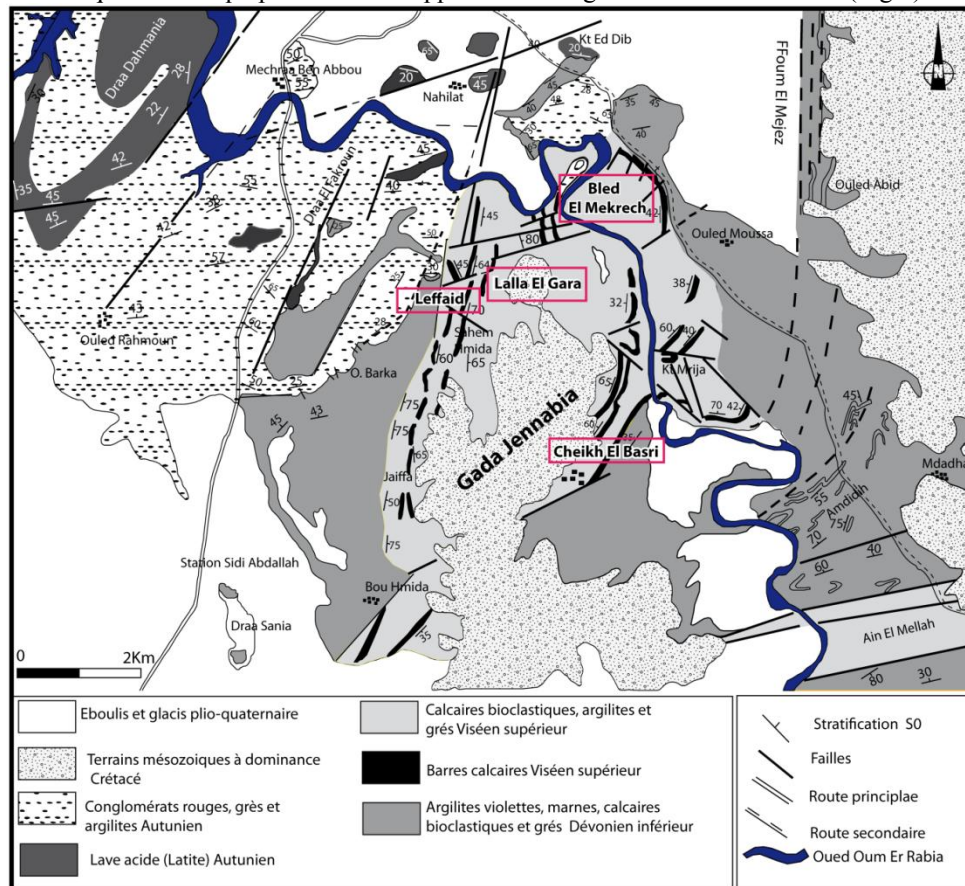


Fig. 2:- Répartition des affleurements de roches magmatiques basiques dans le synclinal de la Gada Jennabia (Kholaiq et al, 2015a)

Caractérisation structurale et pétrographique des roches magmatiques:-

Les données pétrographiques ont été obtenues à partir de l'étude d'une vingtaine d'échantillons considérés comme représentatifs. Des lames minces ont été observées au microscope pour déterminer les textures et les associations minéralogiques de ces roches. De point de vue mode de gisement, l'ensemble des roches magmatiques forment des sills et les coulées de laves parallèles à l'encaissant immédiat d'âge Viséen supérieur probable. Ainsi, leurs caractères relativement résistant à l'érosion et à l'altération mettent les gabbros et les dolérites en relief au sein des schistes. Les roches sont généralement de couleur vert olive quand elles sont fraîches, et prennent une couleur sombre par altération.

Mais avant d'étudier ses roches, et pour mieux cerner le mode de la mise en place des roches magmatiques nous avons réalisé plusieurs coupes sur le terrain.

Coupe de Bled el Mekrech:-**Relation roche magmatique – encaissant:-**

Une coupe réalisée sur le flanc Est de la Gada Jennabia, orientée ESE-WNW (fig.3), révèle la présence des plis dans les terrains d'âge Dévonien inférieur. Ces plis de taille métriques sont tous de type isopaques dont l'orientation des axes varie de N90 à N125 avec un prolongement axial de 25° à 30° vers l'Ouest.

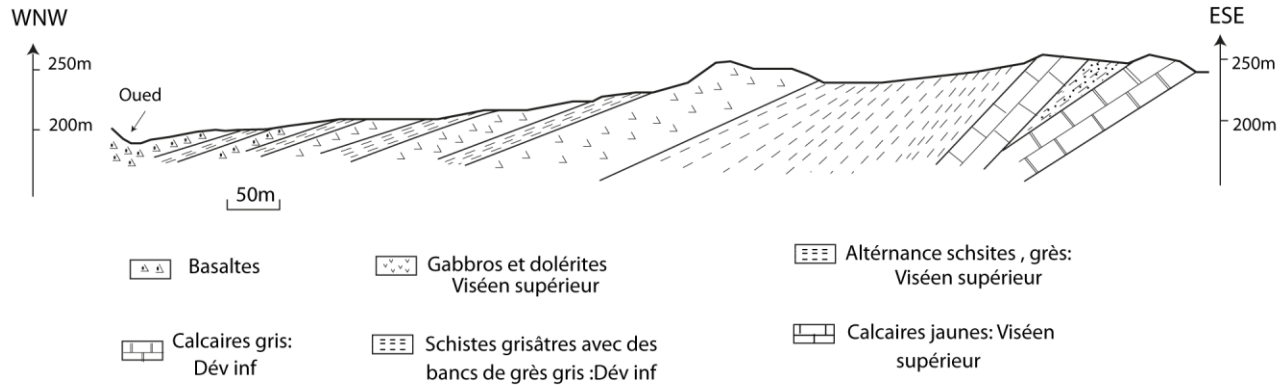


Fig.3:- Coupe géologique dans les terrains Dévono-Viséens de Bled El Mekrech.

Sur le flanc Sud, la direction des couches varie de N100 à N105 avec un pendage de 45° à 40° vers le Sud. Sur l'autre flanc la direction est de N75 avec un pendage de 25° vers le Nord. Au niveau de la charnière la stratification est orientée N145 avec un pendage de 30° vers l'Ouest. La schistosité est surtout visible dans les bancs incompetents schisteux. Elle est sub-verticale avec une orientation N15 à N20. Cette schistosité parfois subit des virgations illustrant des zones de cisaillement à l'échelle de l'affleurement orientées N120-N130 senestres.

La deuxième barre calcaire d'âge Viséen supérieur montre une stratification orientée N160 avec un pendage de 30° dans laquelle on observe des plis SE. Au niveau de la surface structurale de cette barre on rencontre des fentes de tension remplis par la calcite de direction N55 et N160 et un pendage de 55° vers l'Est parfois en échelons matérialisant des zones de cisaillement.

La barre de Viséen supérieur est suivie par une alternance de schistes et grès dont l'orientation est la même que la barre calcaire de Viséen supérieur avec un pendage de 55° vers l'Ouest.

Dans les gabbros et les dolérites, on trouve des miroirs de failles N170 55° vers l'Est ainsi que des cassures de même direction remplis par la calcite. Ces cassures sont parfois plissées. On trouve aussi des filons pluri-métriques N 130 sub-verticaux, remplis par du quartz. Le contact encaissant - gabbros et dolérites est souvent accompagné d'une schistosité de fracture sub verticale N110 à N120. (Fig. 4). Par endroits ce contact prend un aspect bréchifié. Plus on se dirige vers l'Oued c'est-à-dire vers le cœur du synclinal le pendage des couches devient de plus en plus faible pour atteindre des valeurs très faibles (20° vers l'Ouest).

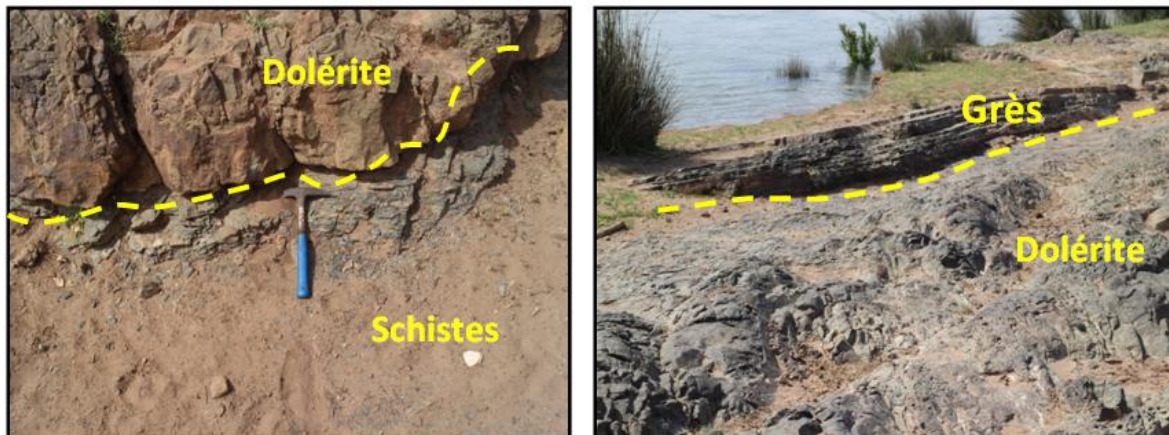


Fig. 4:- Photos de terrain montrant la relation entre les dolérites et les roches encaissantes

Pétrographie:-

L'étude microscopique des roches magmatiques à Bled El Mekrech (Fig.5), nous a permis de mettre en évidence un faciès gabbroïque. Les gabbros de couleur vert clair à vert foncé développent une variété de textures, grenue, microgrenue, et doléritique. Dans les gabbros à texture grenue, les associations minéralogiques montrent un gabbro à pyroxène et un gabbro ordinaire sans pyroxène. Les gabbros à pyroxène (Fig.5,A) montrent une composition minéralogique à plagioclases (30 à 40%), de type labrador ou les macles polysynthétiques sont assez fréquentes. La taille des lattes est comprise entre 2 et 5 mm. Ces cristaux développent une altération en calcite et en chlorite. Le clinopyroxène 40 à 50%, est le minéral le plus abondant après le plagioclase, et forme des plages xénomorphes très fissurées. Il montre des transformations en granules d'oxydes noirâtres, en chlorite, et/ou en calcite.

Vers les bordures figées et à l'approche du contact avec l'encaissant, les gabbros ordinaires (Fig.5, B) montrent une texture microgrenue ou les phénocristaux sont représentés par des plagioclases millimétriques et des transformations surtout en calcite. Dans le faciès à texture doléritique (Fig.5, C et D), les plagioclases et les clinopyroxènes constituent la paragenèse primaire (60% à 70%) et la calcite, la chlorite et les oxydes de fer constituent la paragenèse secondaire. Le plagioclase de type labrador, cristallise en lattes maclées sub-tabulaires de 0,4 à 4mm de long. Ces dernières sont en association intersertale triangulaire renfermant des clinopyroxènes. Parfois, les plagioclases se présentent sous forme de baguettes à l'intérieur des clinopyroxènes, conférant à la roche une texture ophitique.

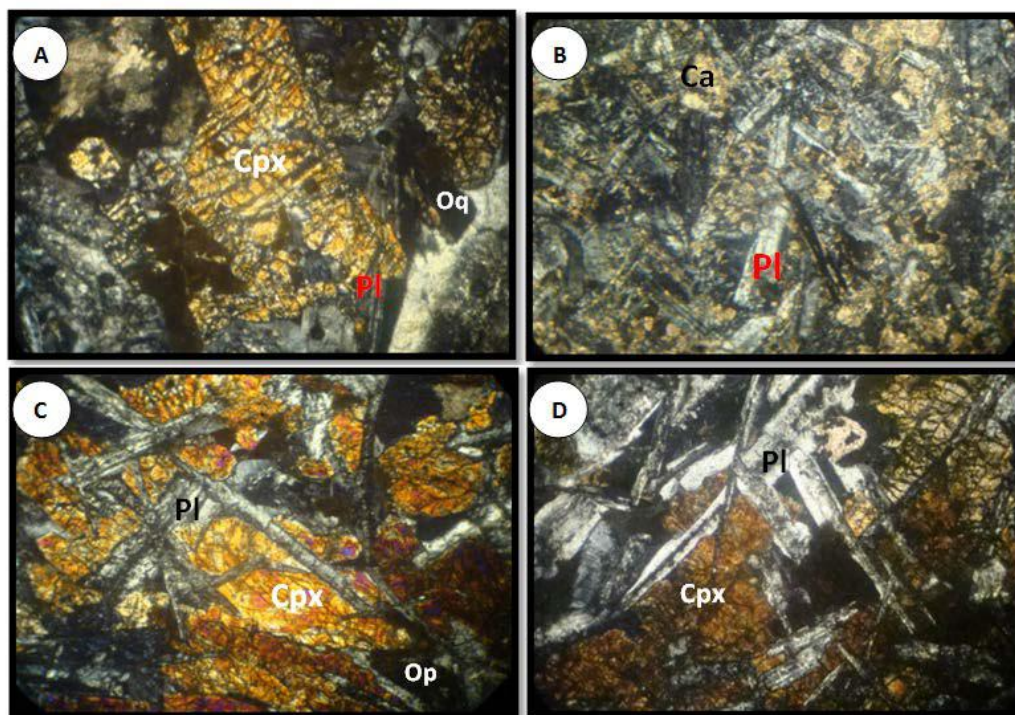


Fig .5:- Photographie des associations minérales des roches magmatiques basiques de Bled El Mekrech.

A : gabbro à pyroxène au coeur du sill. B : gabbro ordinaire des bordures figées. C et D: dolérite à texture intersertale et sub-ophitique. Pl : Plagioclase. Cpx : Clinopyroxène. Ca : Calcite. Op : Opaque (Gr*100)

Coupe de Cheikh El Basri :**Relation roches magmatique – encaissant:-**

C'est une coupe E-W réalisée dans les terrains géologiques de Cheikh El Basri (fig.6), les formations détritiques et carbonatées recoupées par des sills des roches magmatiques basiques d'âge Viséen supérieur, forment une structure monoclinale à pendage vers l'Ouest. Cette structure est surmontée par une couverture tabulaire d'âge Mésozoïque en discordance angulaire.

A la base, on trouve des schistes caractérisés par une schistosité de fracture. Ces schistes sont intercalés par des bancs gréseux, l'ensemble est orienté N40 avec un pendage vers l'Ouest. Les filons gabbroïques sont intercalés d'une façon concordante avec une barre calcaire orientée N50 avec un pendage de 70° vers le Nord. Au dessus de cette barre on trouve un deuxième affleurement gabbroïque qui a toujours la même direction que l'encaissant. Par endroit, le contact immédiat est souvent caché par des éboulis qui rendent difficile de toucher et préciser la nature de la limite encaissant- gabbro. Les schistes qui viennent après sont perturbés (aspect chaotique) assimilables à des brèches tectoniques, ce qui confirme un passage de faille. Le haut de ces schistes est caractérisé par des passés gréseux de couleur grisâtre à noirâtres dite cornés orienté N40 avec un pendage vers l'Ouest.

Sur les schistes un filon gabbroïque est de couleur rougeâtre parfaitement parallèle avec la stratification témoigne une autre fois une mise en place sous forme de sill stratiforme.

Sur ces gabbros repose une barre calcaire N50 avec un pendage de 70 à 60° vers l'Ouest. Avec cette barre s'affine le Viséen supérieur et commence le continentale intercalaire. Il est formé essentiellement par des grès, calcaires et des argiles rouges dont la stratification est orientée N40. Dans la partie sommitale, on trouve une alternance des barres calcaires et des marnes jaunâtres constituent une couverture sub-tabulaire.

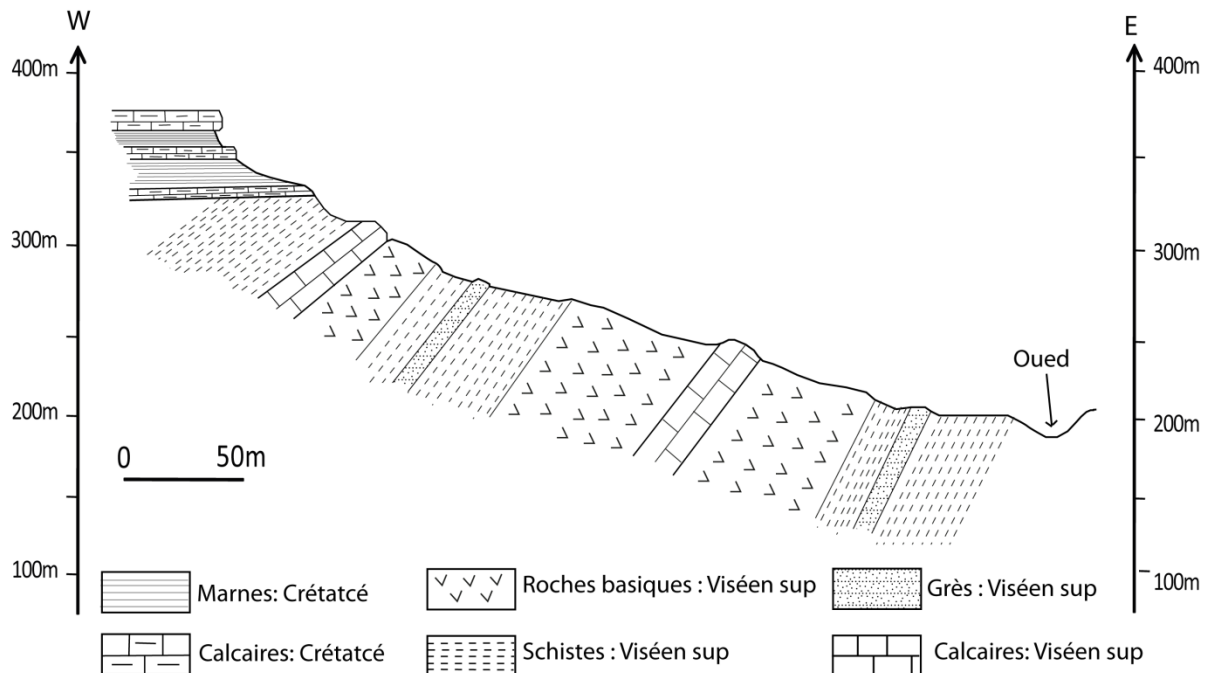


Fig. 6:-Coupe géologique E-W à travers les terrains Viséens supérieurs à Cheikh El Basri

Pétrographie:-

Ce massif constitue l'un des principaux corps magmatiques des Rehamna septentrionaux. Dans les travaux antérieurs, il n'a jamais fait l'objet d'étude pétrographique. Il s'agit de dolérite de couleur rouge surpatine et sur cassure fraîche. Au microscope la texture est intersertale ± ophitique et/ou sub-ophitique (Fig.7). La composition minéralogique est marquée par une proportion en plagioclase quasi égale à celle des minéraux ferromagnésiens représentés par des clinopyroxènes. Le plagioclase en phénocristaux cristallise en premier. Il est de type labrador avec des macles polysynthétiques. Il est fréquemment fracturé mais rarement altéré. Le clinopyroxène qui cristallise en plages xénomorphes dans une texture de type ophitique à sub-ophitique où les lattes de plagioclases sont incluses totalement ou partiellement dans les plages de clinopyroxène.

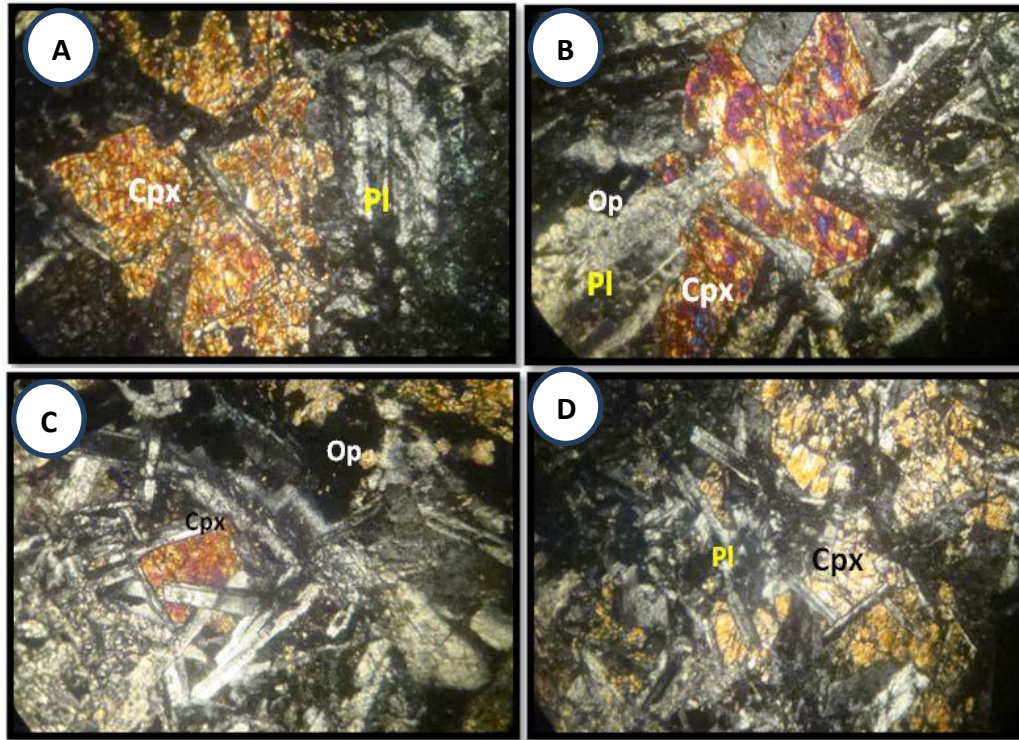


Fig. 7:- Photographie des associations minérales des roches magmatiques basiques de Sheikh El Basri.
A et B : Dolérite à texture inter tale, ophitique et sub-ohitique. C : Dolérite à texture inter tale et sub-ophitique. D: Dolérite à texture inter tale, ophitique et subohitique.
*Pl : Plagioclase. Cpx : Clinopyroxène. Ca : Calcite. Op : Opaque (Gr*100)*

La coupe de lala El Gara:-

Relation roches magmatique – encaissant:-

La coupe W-E réalisée au niveau de la terminaison périsynclinale de la Gada Jennabia (Lala El Gara) révèle (fig.8) : Une série schisto- gréseuse où s'intercale des gabbros sous forme de sills. La stratification aux limites des gabbros est orienté N150 avec un pendage de 70° vers le Nord.

Dans cette formation on a pu mettre en évidence des plis dont le cœur présente une schistosité de plan axial N20 - N30 sub- vertical donnant un aspect en frites aux schistes lorsqu'elle coupe la stratification.

En allant vers l'Ouest, les sills stratiformes suivent le plissement des schistes pour former des synclinaux et des anticlinaux de direction N20 - N30, le pendage sur les flancs est généralement de l'ordre de 50° vers l'Est et de 30° vers l'Ouest. Ces plis apparaissent dans des couloirs limités par des failles verticales.

Le plissement des sills suivant les ondulations observées surtout au niveau de la terminaison périsynclinale de Lala El Gara confirme une mise en place syn phase paroxysmale (fig 9 et 10).

Lala El Gara au sommet présente une structure synclinale dont les pendages aux flancs sont faibles, soit vers l'Est ou soit vers l'Ouest. Elle est bien marquée par les coulées basaltiques interstratifiées dans une formation schisteuse.

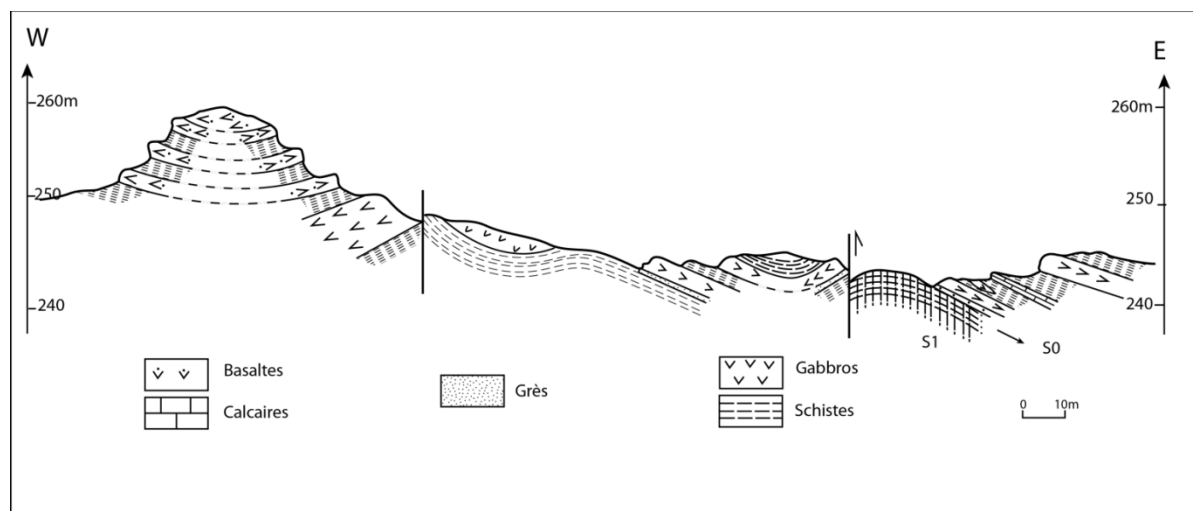


Fig.8:- Coupe géologique dans les terrains Viséens de Lala El Gara.

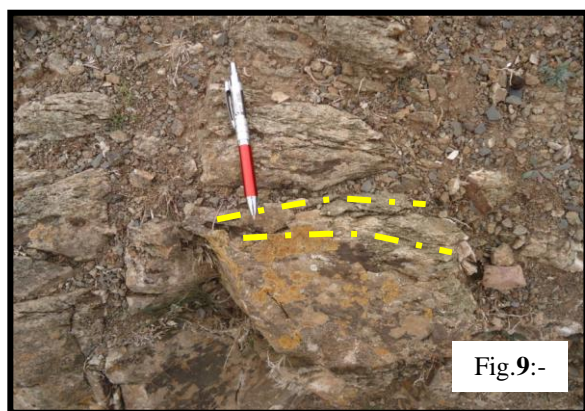


Fig.9:-

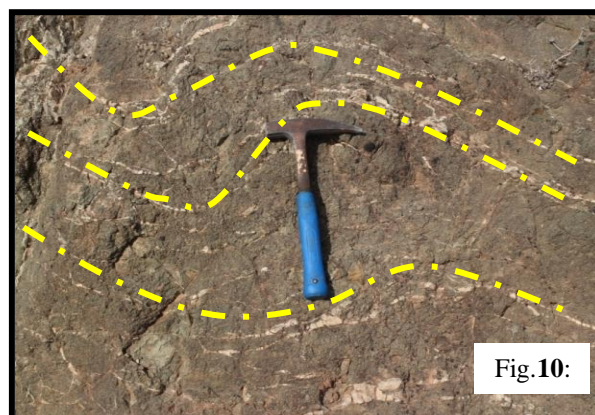


Fig.10:-

Fig.9:- Photo montrant un débit schisteux dans les roches magmatiques basiques.

Fig.10:- Photo montrant le plissement des veines calcitiques au sein des gabbros.

Pétrographie:-

L'étude pétrographique des roches magmatiques de Lala El Gara montre qu'il s'agit de deux ensembles : un ensemble doléritique et un ensemble basaltique. L'ensemble doléritique (Fig.11, A et B) a une composition minéralogique semblable à celle de Bled El Mekrech et de Cheikh El Basri, mais s'en diffère par l'abondance des plagioclases. Le plagioclase constitue le minéral cardinal, de type labrador sous forme des cristaux automorphes avec des macles polysynthétiques. Le clinopyroxène cristallise en plages qui peuvent englober totalement ou partiellement des cristaux de plagioclases formant une texture ophitique à sub-ophitique.

L'ensemble basaltique est reconnaissable grâce à la présence de vacuoles centimétriques, la roche est de couleur grise à verte olive. A l'échelle de la lame mince, la roche a une texture microlitique porphyrique vacuolaire (Fig.11, C et D). La paragenèse primaire est constituée essentiellement par des cristaux de plagioclases de type labrador, sous forme de baguettes allongées de taille variant de 0,05 à 4mm, qui constituent 60% à 70% du volume total de la roche. La macle polysynthétique est bien distincte. Ces cristaux sont emballés dans un fond vitreux (mésostase), qui représente 30% à 40% de la roche. Celle-ci renferme des vacuoles à remplissage calcitique, dont certaines montrent un remplissage séquentiel avec de la chlorite au centre et la calcite en bordures. Ce caractère traduit, soit la présence de deux phases d'hydrothermalisme liées respectivement à deux phases orogéniques, soit l'existence de deux stades d'hydrothermalisme tardi à post-magmatiques d'une même phase tectonique dont l'un est précoce et l'autre est tardif. Les vacuoles peuvent être isolées ou communicantes avec deux ou plusieurs individus.

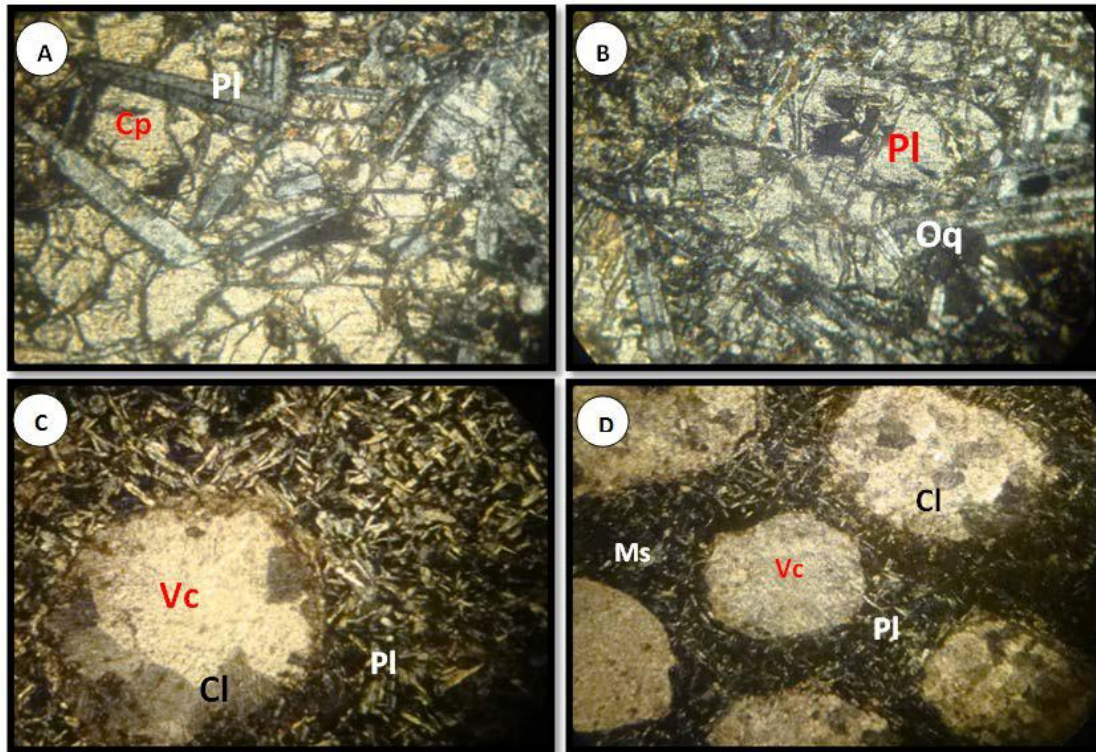


Fig.11:- Photographies des dolérites et des basaltes de Lala El Gara.

*A : Dolérite à texture ophitique et sub-ophitique (centre du sill). B : Dolérite des bordures figées. C et D : Basaltes porphyriques microlitiques vacuolaire Pl : Plagioclases, Cpx : Clinopyroxènes, Ca : Calcites, Oq : Opaques, Vc : Vacuoles Ms : Mésostase (Gr*100)*

A Leffaid:-

Relation roches magmatique – encaissant:-

Il s'agit d'une coupe E-W (Fig. 12), réalisée dans les terrains viséens de Leffaid (flanc Ouest du synclinal de la Gada Jennabia. En se basant sur les relations entre les différents faciès, une succession lithostratigraphique a pu être reconstituée, cette dernière comprend de bas en haut (de l'Ouest à l'Est) :

Des schistes grisâtres orientés N30 avec un pendage de 40 à 30° vers l'Est, suivi par une barre calcaire orientée N160 avec un pendage de 40° vers l'Est. Au niveau de la surface structurale on trouve des gradins matérialisés par des failles N80 portant des stries à aspect boueux, cette barre est suivie par une alternance de schistes et de grès dont l'orientation est N30 avec un pendage de 30° vers Est.

Après une deuxième barre calcaire un peu pentée que la première mais qui garde la même direction et qui est toujours vers l'Est, des filons gabbroïques sont intercalés d'une façon concordante avec des schistes et des grès (fig 13), sur ces roches magmatiques on remarque un débit orienté N30 et des fractures orientées N80 remplies de calcites et de quart. Une alternance de coulées basaltiques et des schistes coiffent la série viséenne.

Les terrains du Viséen supérieur sont couverts en partie par une série tabulaire d'âge mésozoïque sur lesquels elle repose en discordance.

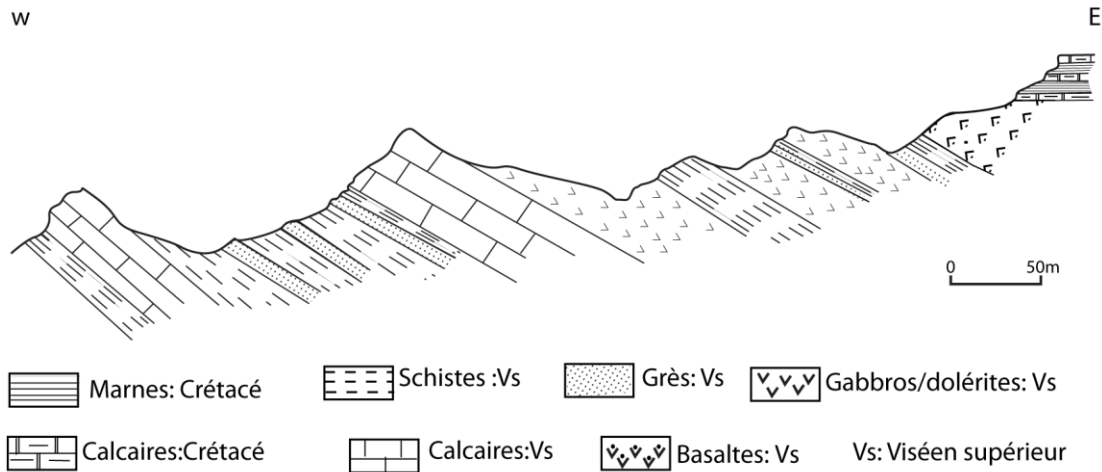


Fig. 12:- Coupe géologique à travers les terrains Viséens supérieurs de Laffaid



Fig. 13:- Photo montrant la relation entre les dolérites et les roches encaissantes

Pétrographie:-

L'étude microscopique des roches magmatiques dans la région de Laffaid nous a permis de mettre en évidence deux ensembles : un ensemble doléritique (Fig 14, A et B) et un autre basaltique (Fig 14, C et D).

L'ensemble doléritique est caractérisé par une couleur vert olive sous forme de sills. Il montre à l'oeil nu des plagioclases et des clinopyroxènes. Microscopiquement, les épontes de ces sill sont caractérisées par une texture microcristalline. Vers le coeur, la texture évolue et devient intersertale, ophitique et/ou sub-ophitique. La paragenèse minérale est constituée essentiellement par des plagioclases, des clinopyroxènes, des opaques et accessoirement de la calcite, de la séricite et de la chlorite.

Le plagioclase est en général séricitisé. Il représente 40% à 50% des minéraux de la roche. Il s'agit en général de labrador sous forme des cristaux automorphes avec des macles polysynthétiques. Le clinopyroxène, le plus souvent transformé en chlorite et/ou en calcite, représente 30% à 50% des minéraux de la roche. Il est de type clinopyroxène formant avec les plagioclases une texture doléritique de type ophitique et sub-ophitique. Le clinopyroxène cristallise sous forme des plages xéno-morphes très fissuré où on observe des oxydes de fer.

Les transformations secondaires sont, en plus de la chlorite, les oxydes de fer et la calcite. Les minéraux opaques sont souvent automorphes et abondants dans les roches.

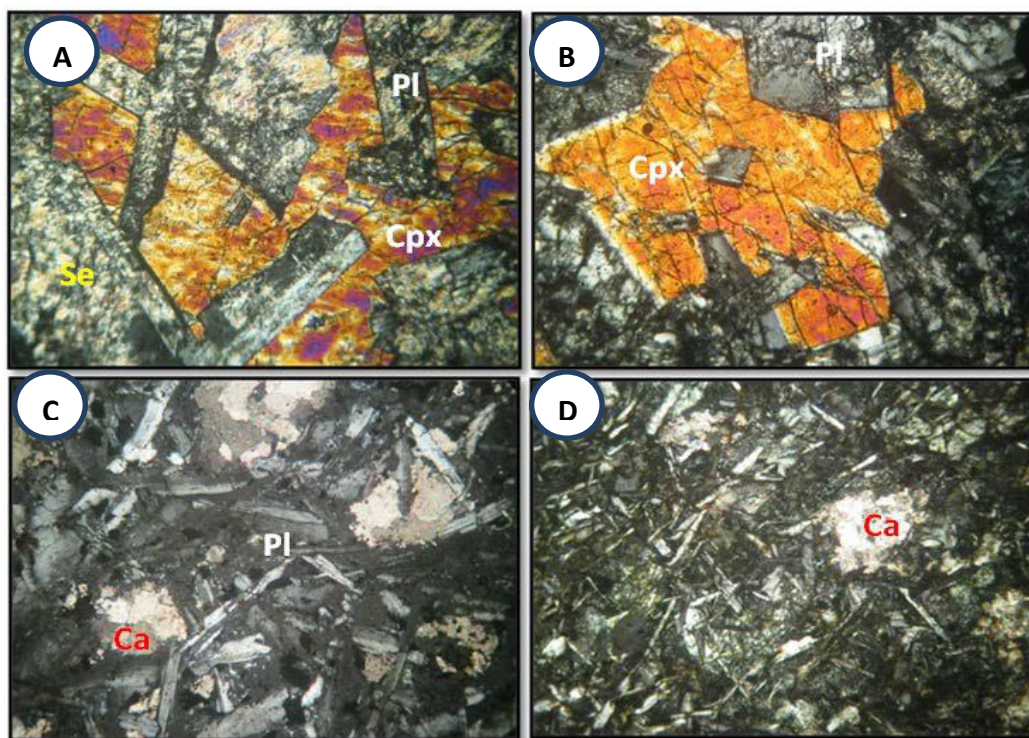


Fig 14:- A, B : Dolérites à texture inter tale, ophitique et sub-ohitique. : Dolérite à texture intertale et sub-ophitique.
C et D : Basaltes porphyriques microlitiques vacuolaire.
Pl : *Plagioclase*. Cpx : *Clinopyroxène*. Ce : *Séricite*, Ca : *Calcite*. Op : *Opaque* (Gr*100)

Il paraît que les roches magmatiques des quatre affleurements (Bled El Mekrech, Cheikh El Basri, Lala El Gara, et Leffaid), présentent des minéralogies comparables (Tableau I) a paragenèse primaire des gabrros et des dolérites est caractérisée par une abondance du plagioclase suivi par le clinopyroxène formant des textures ophitiques, sub-ophitiques et intersertales dont la paragenèse secondaire est surtout représentée par des minéraux d'altérations tels que la calcite, la chlorite, et les oxydes de fer.

Les basaltes microlitiques porphyriques vacuolaires de Lala El Gara et Leffaid sont caractérisés par des plagioclases. Les minéraux d'altération sont représentés par la Calcite, la Chlorite et la Séricite. Concernant l'ordre de cristallisation et d'après la relation mutuelle entre les minéraux, on constate la cristallisation d'abord d'opagues, de plagioclase, puis de clinopyroxène (Fig.15).

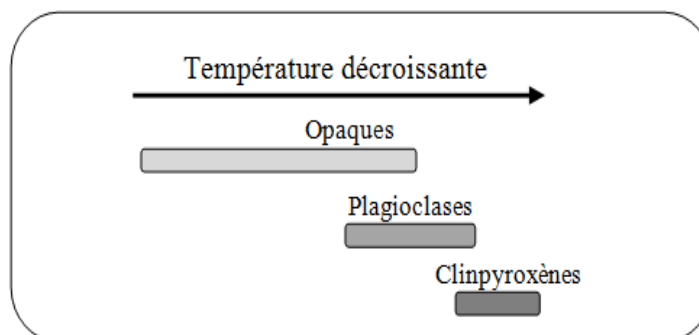


Fig.15:- Schématisation de l'ordre de cristallisation

Altération des roches magmatiques étudiées:-

A l'échelle d'affleurement et du banc, Les phénomènes d'altération sont manifestés par des filons de calcites et de quartz. En effet, les roches magmatiques sont affectées par une fracturation plus ou moins intense. Les fractures (0,5 à 4 cm d'ouverture) sont remplies selon les cas de calcite ou d'ankérite.

A Bled El Mekrech, les roches magmatiques sont affectées par deux familles de fractures remplies par la calcite. La première famille est sub verticale orientée N140, alors que la deuxième famille tardive orientée N20. Parfois, l'aspect réticulé rend difficile l'établissement d'une chronologie relative de leur apparition. Par endroit, les fractures deviennent très serrées et sont remplies par la calcite, qui illustre des amygdales de cisaillement.

Parfois, ces fractures sont plissées indiquant un remplissage anté-déformation. Certaines fractures montrent un remplissage polyphasé (séquentiel), avec cristallisation de la calcite au centre et de l'ankérite en bordures. (fig.16).

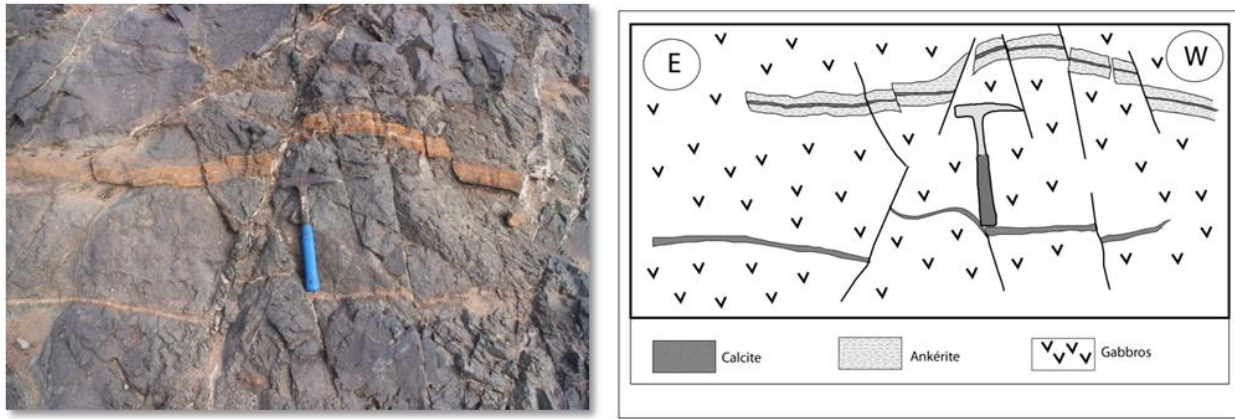


Fig.16:- fractures montrent un remplissage polyphasé

A **cheikh El Bassri**, les roches magmatiques sont traversées par des fractures N 70 remplies aussi par la calcite, alors que à **Lala El Gara**, les roches sont affectées par deux familles de fractures, dont la première est orientée N150 sub- verticale, et la deuxième famille est orientée N90 avec un pendage de 50° vers le Nord.

Microscopiquement, les roches magmatiques des trois zones d'études montrent des transformations en calcites, chlorites, opaques et en quartz rarement au centre des sills très intensément aux bordures figées.

Calcitisation:-

La calcite présente en quantité appréciable dans la majorité des échantillons. Elle représente un produit de transformation et d'altération des minéraux primaires. Elle se rencontre presque dans les roches altérées qui se situent aux bordures des sills. Ainsi Certaines roches sont complètement transformées en calcite. Cette dernière cristallise soit en petites plages, soit sous forme des paillettes et / ou des petites veines qui recoupent les lattes de plagioclases (fig.18.a). Elle cristallise dans les cassures des cristaux de clinopyroxènes et de plagioclases aussi bien qu'à la limite plagioclase-clinopyroxène. Dans les gabbros de Cheikh El Basri, la calcite est associée à l'ankérite ou au chlorite. (fig.17et 18.b).

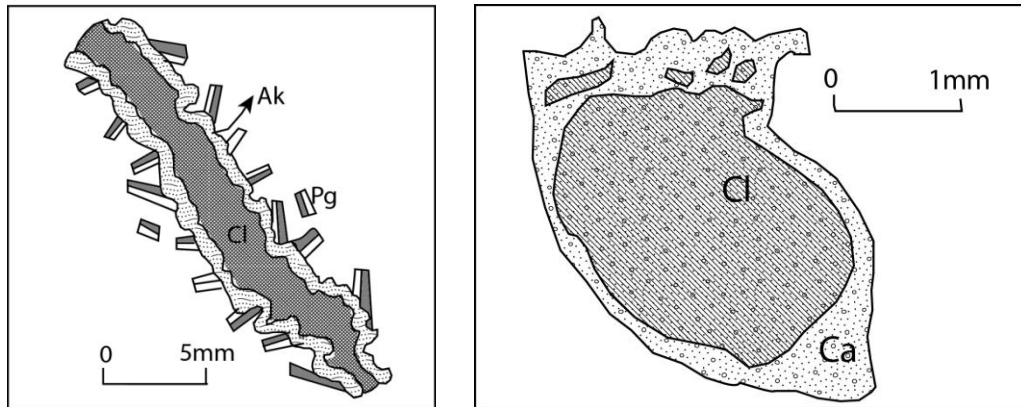


fig. 17:-Altération polyphasée dans les gabbros de Cheikh El Basri

Ak : Ankérite, **Cl** : Chlorite, **Ca** : Calcite, **Pg** : Plagioclase

Chloritisation :-

La chlorite est un minéral secondaire qui se développe aussi bien dans gabbros que les dolérites. Elle cristallise en petits amas qui se développent aux dépens du clinopyroxène et du plagioclase. Grâce à sa couleur verdâtre, elle se reconnaît facilement en lame mince. Elle se rencontre associée à d'autres produits de transformation telle que la calcite (fig.18.c).

Oxydation :-

L'oxyde de fer fait partie des produits d'altération les plus répandus dans toutes les lames altérées. Il se développe au dépend des fractures des minéraux surtout les pyroxènes (fig.18.d).

Ouralitisation:-

Ouralitisation des pyroxènes est la formation d'amphibole (Trimolites ?) qui cristallise en amas et en plage au dépend des pyroxènes (fig.18.e).

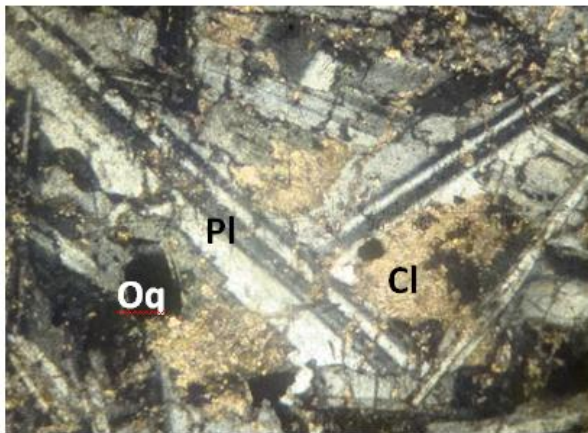


Fig.18.a : Calcite au dépend des plagioclases

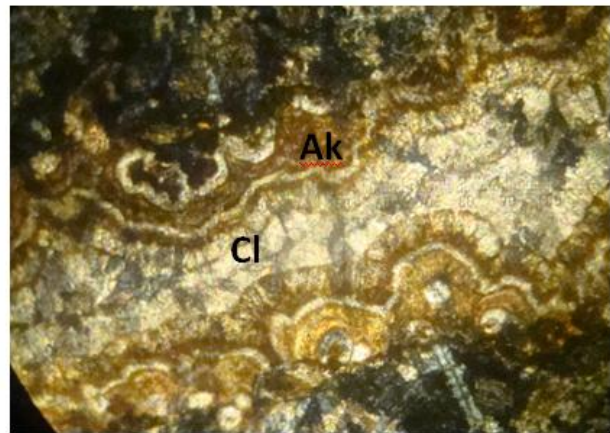
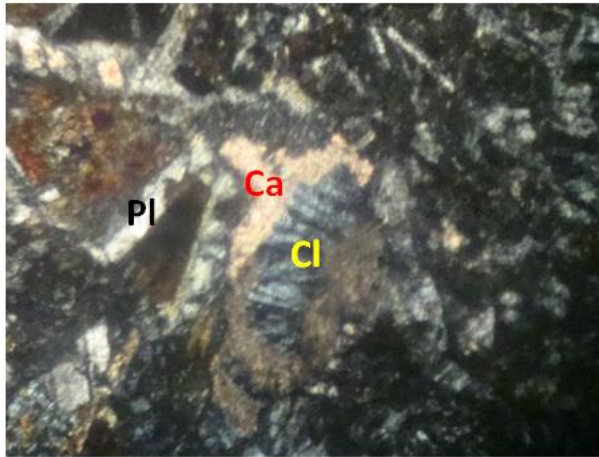


Fig.18.b : Calcite au milieu de l'ankérite



Pl : Plagioclases
 Cp : Clinopyroxènes
 Ca : Calcites
 Cl : Chlorites
 Oq : Opaques
 Ak : Ankérites

Fig.18.c : Chlorite dans la calcite

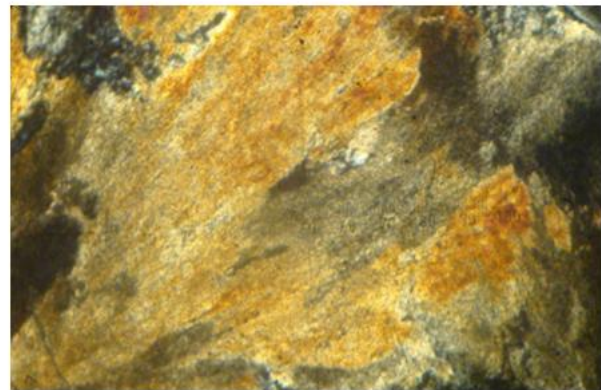
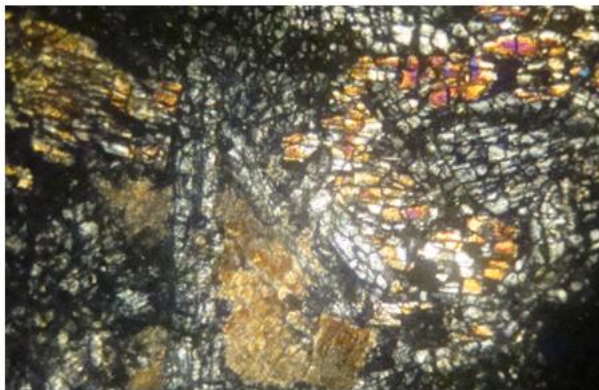


Fig.18.d : Oxyde de fer dans les fractures de pyroxènes Fig.18.e : Amphibole

Fig.18:- lames minces des différents produits d'altérations (LPA) Gr*100

Approche géochimique des effets de l'altération:-

Les compositions chimiques des roches analysées peuvent être affectées par une altération post-magmatique. En effet, elles montrent des transformations hydrothermales liées aux événements tectono-métamorphiques hercyniens. D'après une lecture directe des compositions chimiques et en particulier les pertes au feu qui traduisent le plus souvent la transformation des phases minéralogiques primaires en phases secondaires hydratées. L'examen des analyses chimiques (tableau) montre que les roches étudiées présentent des pertes au feu variable, de 4,16 à 9,84%. Les valeurs élevées traduisent une altération importante des minéraux primaires.

Dans ce cas, le diagramme (K_2O+Na_2O) en fonction de $K_2O/(K_2O+Na_2O)$ de Hughers (1973) (Fig19) nous semble très important du fait qu'il définit le champ des roches de composition magmatique initiale, le champ des roches enrichies en potassium et celui des roches enrichies en sodium

L'examen de ce diagramme montre que les points représentatifs des quatre affleurements du bassin de MBA gardent l'empreinte des caractères magmatiques originaux comme l'indique leur emplacement dans le spectre des roches ignées

Donc même avec des valeurs élevées des pertes au feu, les roches étudiées gardent leur caractère initiale, on peut dire donc qu'elles ont subi une altération post formation

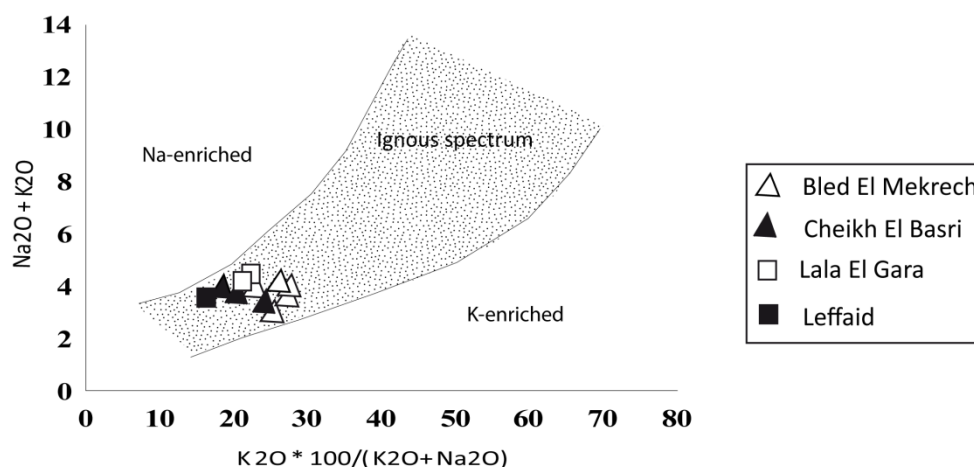


Fig 19:- projection des roches magmatiques basiques étudiées dans le diagramme de Hughes (1973) montrant que ces roches ont une composition magmatique originelle comparable

Diagrammes oxydes pertes au feu:-

A partir de la lecture des analyses chimiques brutes des roches magmatiques, nous constatons que les pertes au feu sont relativement élevées, les roches magmatiques peuvent en contenir jusqu'à 9,84%

Les fluctuations des pertes au feu des roches étudiées semblent être liées à la proportion des minéraux hydratés (chlorites, séricites) et la néoformation de la calcite et le quartz dans les cassures, les veines et les vacuoles dans les basaltes. Il est évident que les teneurs en pertes au feu soient proportionnelles au degré d'hydratation que le magma et les minéraux peuvent subir. Ce phénomène qui traduit évidemment l'effet conjugué de l'altération phylliteuse et carbonatée, peut s'accompagner d'un apport ou d'un lessivage d'éléments chimiques.

Ainsi, les variations des principaux oxydes en fonctions des pertes au feu (fig 20) permettent de rendre compte du comportement des éléments chimiques au cours de l'altération. Dans ce sens, le diagramme SiO₂-PF montre une diminution des teneurs en SiO₂ corrélativement à l'augmentation de celle de pertes au feu, cette distribution des points représentatifs des échantillons analysés est sans doute une conséquence d'un hydrothermalisme responsable de la mobilité de la silice comme en témoigne la présence de filons, veines, et vacuoles remplis de quartz secondaire.

Le diagramme CaO – PF montre une corrélation négative marqué par une augmentation des teneurs en CaO pour des pertes au feu de plus en plus élevées, ceci peut être expliqué par la mobilité du calcium comme témoigne d'une part la calcitisation des plagioclases et de la mésostase et d'autre part, le remplissage des vacuoles, des veines et des filons de la calcite

Dans le diagramme de Fe₂O₃- PF une légère corrélation négative indique un lessivage du fer, comme le laisse prévoir l'oxydation des microfractures, l'hématisation et l'oxyde de fer dans cassures et le clivage dans les clinopyroxènes

Le diagramme de MgO- PF met en évidence une corrélation positive des représentatifs des roches basiques qui traduit un apport du magnésium, comme l'atteste la formation de la chlorite magnésienne au dépend des minéraux ferromagnésiens (chloritisation des clinopyroxène, dans les vacuoles et dans la mésostase

Le diagramme de K₂O vs PF met en évidence une corrélation négative qui indique que les phénomènes d'altération s'accompagnent d'un lessivage de K₂O.

Le Na₂O vs PF présente une très faible corrélation positive et pourrait s'expliquer par une contribution du Na marquée par l'albitisation des plagioclases calciques.

En consultant le diagramme $K_2O - Na_2O$ nous constatons que ces deux éléments sont liés par une corrélation négative si bien que les échantillons les plus potassiques étant les plus sodiques

Certes, cette évolution ne peut trouver une explication que dans les processus de différenciation magmatique, nous envisageons une métasomatose potassique couplée à un lessivage de la soude pour interpréter le comportement des alcalins dans roches magmatiques basiques du bassin de MBA

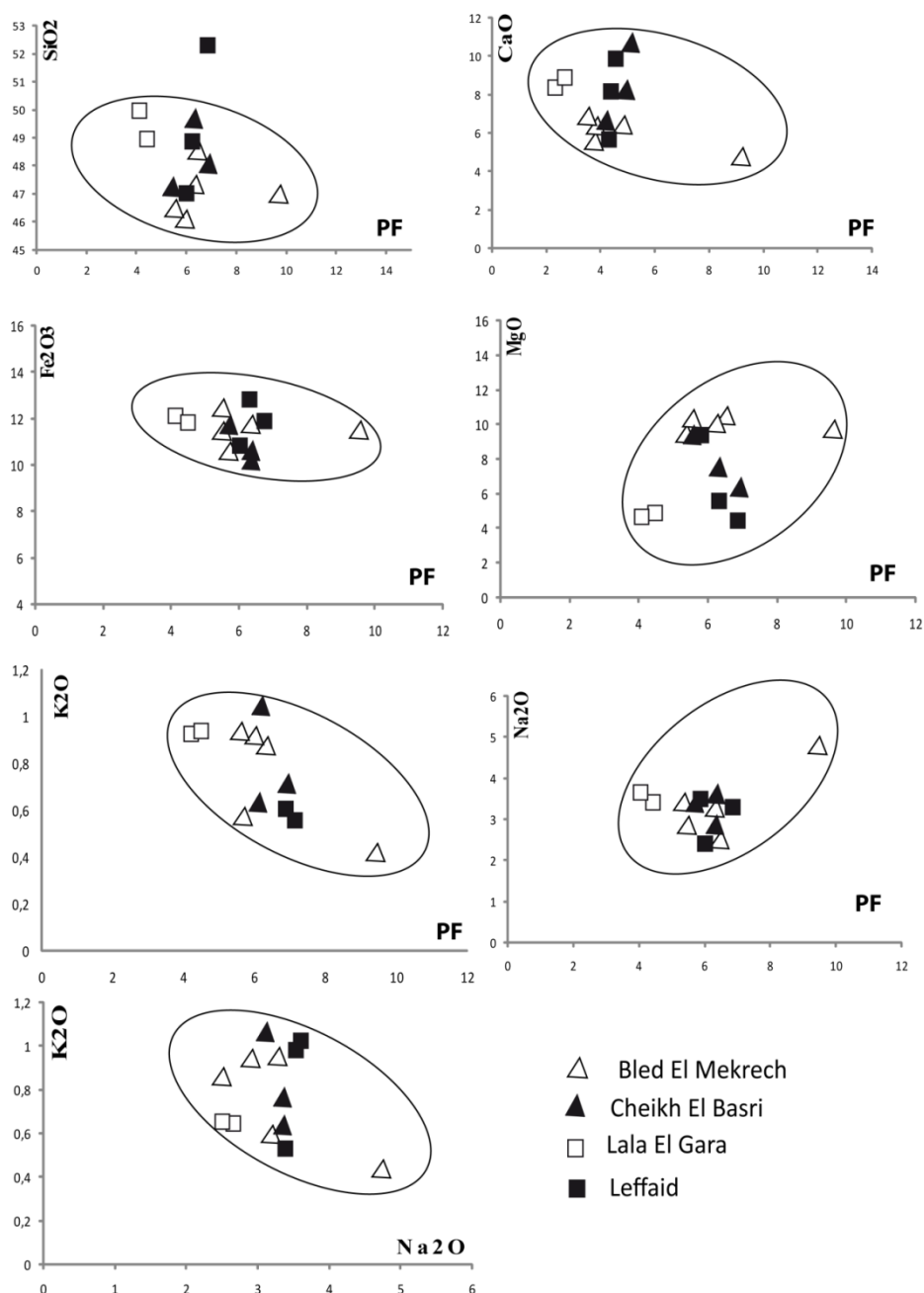


Fig. 20:- Variation de la composition chimique des éléments majeurs en fonction de la perte au feu

Conclusion générale:-

Dans le but d'approcher les phénomènes d'altération dans les roches magmatiques basiques de Mechraa Ben Abbou nous étions amenés à suivre une démarche multiple structurale, pétrographique et géochimique. Ainsi, l'analyse structurale de quatre affleurements représentatifs a permis à travers des coupes réalisées sur le terrain que les contacts entre les différentes roches magmatiques et l'encaissant immédiat sont toujours parallèles à la stratification ce qui témoigne d'une mise en place de ces roches sous forme de sills.

L'étude pétrographique des roches magmatiques des quatre affleurements (Bled El Mekrech, Cheikh El Basri, Lala El Gara et Leffaid) a permis de remarquer que ces roches présentent des minéralogies comparables dominées par une paragenèse primaire constituée par une abondance de plagioclase suivie par le clinopyroxène et une paragenèse secondaire qui est surtout représentée par des minéraux d'altérations tels que la calcite, la chlorite, l'amphibole, et l'oxyde de fer.

Ces phénomènes d'altérations qui peuvent être liés aux circulations des fluides hydrothermaux se manifestent à l'échelle de l'affleurement et du banc par des filons de calcites, de quartz et d'ankérite et à l'échelle de la lame mince les roches magmatiques des quatre zones d'études montrent des transformations en calcites, en chlorites, en oxydes de fer et en quartz rarement au centre des sills, mais très intensément aux bordures figées. Les altérations se développent, aussi bien au niveau de la mésostase qui inclut localement des vacuoles millimétriques colmatées par la calcite et la chlorite.

Les analyses géochimiques montrent que les altérations se traduisent par un changement dans le comportement des éléments chimiques qui témoigne de l'interaction entre les fluides hydrothermaux et les roches magmatiques étudiées. Ainsi, les diagrammes de variation des oxydes en fonction des pertes au feu indiquent un lessivage de SiO_2 , CaO , Fe_2O_3 et de K_2O et d'un apport en MgO et en Na_2O .

La combinaison des données structurales (filons plissés et filons cassés), la nature du remplissage minéral séquentiel avec la cristallisation de la chlorite (haute T°) et tardive de la calcite (basse T°) plaident pour deux phases d'altérations hydrothermales concordantes avec les deux phases tectoniques hercyniennes paroxysmale et tardive :

- Une phase I, caractérisée par un plissement P1 à l'échelle cartographique dont l'axe est orienté N20– N 30 avec un plongement de 10 à 20° vers le Sud. C'est le synclinal plurikilométrique dit de la Gada Jennabia ;
- Une phase II, caractérisée par un plissement P2 détectable à l'échelle de l'affleurement métrique à décimétrique accompagnant les failles transverses N60-N80.

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RESEARCH ARTICLE

UNDERSTANDING STUDENTS' VIEWS ON THE NATURE OF SCIENCE.

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Abstract

This study sheds some light on the commonly held views of senior secondary students about the nature of science. The study is framed around the philosophical positions on the nature of science and elaborate treatment of its various aspects as given by Lederman (2002). The study is purely qualitative and emergent and has been conducted in the Indian context. Samples of the study are school children who have opted science and the college students who are in the first year of graduation course with chemistry as one of their major subjects. In this study, we have tried to describe how students perceive science to be and how it works along with trying to present an emerging relationship between their understanding of the nature of science and their interest in and appreciation of science. Furthermore, taking insights from many studies (Mc Comas, Almazora and Clough (1998)) that chemistry textbooks have an important role in shaping ideas and understanding of concepts in science and the nature of science, we reviewed the NCERT chemistry textbook looking for representation of some aspects of the nature of science. This book is chosen because of been relied upon heavily by students and teachers as the main source of information in Indian schools. Also, looking at the current trend worldwide (Nuffield Projects, PISA documents) there was a felt need how NCERT textbook fare on the yardstick of parameters set to decipher the effectiveness of chemistry concepts taught to the children. We also have used in this study the Niaz and Fernandez (2008) framework to review a chapter on 'structure of atom' looking for the representation of Historical and Philosophical treatment of the chapter.

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Major findings reveal that college and school students had almost the same views about the nature of science. Textbook writing needs to incorporate the style of presentation which doesn't distort the real science, its nature and how it develops. The textbook reviewed for the chapter 'structure of atom' was not written as per the historical and philosophical framework. Students didn't appreciate the need to study older theories. Several such findings are enlisted in this study. Thus, this study presents how actually this study was conceptualized, took its course and finally manifested in the form of results, findings and discussions which we would like to share with you.

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Introduction:-

Understanding the Nature of Science:-

Science is so much part of our everyday life that we find it impossible to stand back and examine its social context. Our policy documents NCF 2005 and Position Paper on the teaching of science (NCERT, 2006) talk about the development of 'scientific literacy' in all citizens. 'Scientific literacy' means the ability to use scientific knowledge to make informed personal and societal decisions. This further implies that people need to be able to distinguish between good science, bad science and non-science and make critical judgments about what to believe in. They also need to be critical consumers of science. For example, in order to understand and evaluate the scientific advice on food safety, an adult would need to not only know some basic facts about the composition of nutrients, but also to be able to apply that information. The term "literacy" is used to encapsulate this broader conception of knowledge and skills (OECD/PISA, 2003; Ramanna, 1981).

There are different researches which emphasize on different dimensions of scientific literacy or some even look at it under the rubric of public understanding of science. However, an adequate understanding of the nature of science is underlined as a central component of scientific literacy (AAAS, 1993).

The nature of science is characteristic of science. It reflects the values and assumptions that demarcate scientific knowledge and science from other disciplines. It is referred to as knowledge about science. The nature of science is directly related to the epistemology of science as distinct from scientific process and scientific content. The phrase 'nature of science' is used to describe the intersection of issues addressed by the philosophy, psychology, sociology and history of science.

Despite the acknowledged importance of the nature of science, a number of studies document students' misconceptions concerning the nature of science (Lederman, 1992; Meyling, 1997;) on some aspects which include tentativeness of scientific knowledge, social and cultural aspects

of scientific knowledge, and the theory-laden nature of observation. Textbooks on which a majority of students depend for their understanding of scientific knowledge are often devoid of presentation of issues dealing with the nature of science. Despite the advocacy at the policy level, they continue to present scientific knowledge as final, in a 'rhetoric of conclusion' (Schwab, 1962).

In continuation of our interest to explore the understanding about the nature of science (Dabas, 2010; Rampal, 1993; 1992) and to relate to the readings undertaken as part of the master's course in science education, this study was carried out to assess students understanding about the nature of science at school and college levels.

Philosophical Positions on the Nature of Science:-

The construct of the nature of science has a history of more than sixty years, which has been influenced by various philosophical debates. There are different philosophical positions on the nature of science, such as, positivism, inductivism, hypothetico-deductivism, contextualism and relativism. It often becomes difficult and complex for people working in the area of science education to define it. However, the way NOS is viewed by McComas, Clough and Almozroa, (1998, p.4) presents a comprehensive perspective "A fertile hybrid arena, which blends aspects of various social studies of science, including the history, sociology, and philosophy of science combined with research from the cognitive sciences such as psychology into a rich description of what science is, how it works, how scientists operate as a social group and how society itself both directs and reacts to scientific endeavors".

However, according to Lederman (1992) the nature of science typically has been used to refer to the epistemology of science, science as a way of knowing, or the values and beliefs inherent in the development and validation of scientific knowledge cited in abed-El-Khalick et al. (2008, p. 837). These philosophical positions on the NOS generally cannot provide a simple framework within which school science could look at issues related to the nature of science.

In the 1960s the nature of science used to be viewed mainly as observing, hypothesizing, interpreting data and designing experiments i.e. primarily science as process skills. It was believed that accurate observations lead to scientific knowledge and thus a good observer was taken as 'objective' and 'theory free'. In school, students were expected to spend more time in the laboratory making observations, even if they were not necessarily constructing knowledge. Objective observations coupled with inductive reasoning gave a greater focus to the so called "scientific method" (Dabas, 2010).

As with the shift in the trend from positivism to constructivism, educationists too made a shift from a 'science centered' to 'student centered' view of school science. The humanistic perspective in science was promoted by Aikenhead (1987) and intended to prepare students to critically and rationally assess science and technology. Stress on the STS approach began to increase because science was no longer seen as happening in isolation. Thus, the nature of science, its social aspects and its human character were sought to be described through the study of the history, philosophy and sociology of science.

Osborne et.al (2003) conducted an empirical study on experts drawn from among science educators, scientists, historians, philosophers and sociologists, as well as science teachers and then determined the aspects of the nature of science which must be featured in the school science curriculum. These themes, which emerged from the study and from the themes identified by McComas and Olson created a framework of the essential components of the nature of science to be included in school science curricula. Also, it was highlighted that these ideas and aspects of the nature of science have to be taught in an integrated manner and not in isolation and should be fully contextualized. It was also emphasized that the nature of science should be the core of science education and should not be merely confined to margins.

The Nature of Science and ‘Models’ of the Atom:-

The developments in the area like structure of the atom have immense scope to develop aspects of the nature of science in students. Students in an attempt to understand the models of microscopic entities like ‘atom’ which cannot be observed directly, need to know the nature and role of observations and inference in reaching out to these conceptions.

They may acknowledge the role of unique ideas which initiated the development of the atomic models in earlier times if the historical/developmental aspect of the nature of science could be developed in them. The structure of the atom traces the history from Thomson to quantum mechanical model; enabling the students to understand the tentative nature of science, the existence of rival theories, creativity of scientists and science as a socio-cultural human enterprise.

These aspects of the nature of science like- tentativeness in science, social and cultural association of scientific knowledge, personal beliefs and theory laden nature of observation if developed in students may lead them to have a more informed understanding about the use of ‘models’, such as, for the structure of the atom. Moreover, what is observed today, especially in the context of the microscopic world, will change tomorrow due to advancement in technology and knowledge as it has changed since Robert Hooke discovered the cell. So, students might not develop an absolutist view about science, i.e. scientific knowledge is final and that atomic models are exact replica of the ‘atom’ if these aspects of the nature of science are developed in them.

Science relies on inference and interpretation as all phenomena are not directly observable; for example, inference is used to establish the existence and properties of atoms. The stage at which this concept is introduced, students are normally expected to have developed critical ability to understand the epistemological status of facts, theory, model, and the law that they encounter in science (NCF, 2005). By not properly incorporating the aspects of nature of science, either in teaching learning processes or in the textbooks explicitly, may lead to alternative conceptions among students which gradually keep on being entrenched in their minds. This may create doubt with them about the relevance of the so called ‘redundant’ theories or models in the syllabus. As was seen during FGD and through the questionnaires, there are students who question the inclusion of Thomson’s and Rutherford’s models in the syllabus. They might appreciate their work when an understanding about the tentative nature of scientific knowledge is developed among them. They must also appreciate that scientific progress is an evolutionary process and

that earlier theories pave way for the newer researches, and that very often new theories might build upon the earlier ones, unless there is a paradigm shift.

Rationale to include the NOS in the Textbook:-

Several scholars had (Driver, 1988; West and Pines, 1985) stressed that until now, attention has mainly focused on traditional science content, though in the last decade content related to the nature of science received attention because of interest in teaching science through the history and philosophy of science (Aikenhead and Ryan, 1992, p.881).

Driver et al (1996) suggested some additional arguments supporting the inclusion of the nature of science as a goal of science education. The arguments include the utilitarian view that “an understanding of the nature of science is necessary if people are to make sense of science and manage the technological processes and objects they encounter...” (p.16).

Theoretical Framework:-

This section will lay out the framework of Lederman et al. (2002), used in the current study to understand the students' views on the nature of science.

Study of Students' Views on the Nature of Science

The present study makes use of the framework developed by Lederman using characteristics about the nature of science to analyze the views of high school and undergraduate students and subsequently reviews the chemistry textbook to see how these aspects are dealt with.

The dimensions of the nature of science in Lederman's theoretical framework are defined as:

The tentative nature of scientific knowledge:-

Scientific knowledge, including facts, theories, and law changes as new evidences are collected or gathered through advances in thinking and technology. It is reinterpreted in the light of new theoretical advances (inferential and creative), or changes in the social and cultural spheres. Contrary to common belief, scientific hypotheses, theories and laws can never be absolutely proven irrespective of the amount of the supporting empirical evidence (Popper, 1959). To be proven, a law should account for every instance of the phenomenon it purports to describe. It can logically be argued that one such future instance, of which we have no knowledge whatsoever, may behave in a manner contrary to what the law states. Thus, laws and theories can never acquire an absolutely proven status. Thus, scientific knowledge, although reliable and durable, is never absolute and certain and always tentative.

The empirical nature of scientific knowledge:-

The empirical nature of scientific knowledge addresses that science is at least partially based on observations of the natural world, and sooner or later, the validity, scientific claims is settled by referring to observations of phenomena (AAAS, 1990, P4). However, scientists do not have direct access to most natural phenomena and observations of nature are always filtered through our perceptions and interpreted from within elaborate theoretical frameworks.

The theory laden nature of scientific knowledge:-

Scientific knowledge is theory laden because it is governed by various factors associated with scientists' theoretical and disciplinary commitments, beliefs, prior knowledge, training,

experiences and expectations actually influence their work and what they investigate, how they conduct their investigation, how they interpret their observations. Observations are always guided and acquire meaning in reference to questions or problems, which are derived from certain theoretical perspectives.

The creativity and imaginative nature of scientific knowledge:-

Science, contrary to common beliefs is not entirely rational and orderly activity. Ernest Rutherford conducted an experiment of scattering of alpha particles by gold foil, in 1911. He established the nuclear model of an atom. After he has done an experiment every laboratory assistant could understand it and repeat it. To quote Hospers (1988) “when one reads his work one is always saying, ‘why did not someone else think of that?’ ” cited in Dabas (2010, p.13). It is the imagination of Rutherford which had led him to the gold foil experiment. Thus, science involves the invention of explanations and theoretical entities, which require a great deal of creativity on the part of scientists.

The social and cultural embeddedness of scientific knowledge:-

In an attempt to answer the question- ‘whether we should grow crops like corn or jatropha to produce biofuels, we need to know the social and cultural context. Developed countries depend on their industry will favor fuel, whereas developing countries will favor food. Thus, scientific knowledge is socially and culturally embedded.

Observation, Inference, and theoretical entities in science:-

Observations are different from inferences. Observations are directly accessible to the senses or extension of the senses, for example, objects released above ground level tend to fall to the ground. An inference drawn from this can be: ‘objects tend to fall to the ground because of gravity’. The notion of gravity is inferential as it cannot be accessed or measured only manifested through its effects.

Myth of the scientific method:-

Discussion on students’ responses from the questionnaire shows that they believe ‘experiments are the chief route to scientific knowledge’. Of course, experimentation is a useful tool in science, but it is not the sole route to knowledge. Many scientists had used non-experimental techniques to advance knowledge. In a number of situations, true experimentation was not possible because of inability to control variables. Discovery in astronomy is based on extensive observations rather than experiment. For instance, Copernicus theory and Kepler laws used evidence derived from lengthy and detailed observations. Charles Darwin in his discoveries used qualitative techniques, recorded his observations, speculations and thoughts and then reached at conclusions. Sometimes, theoretical advance can also suggest what to look for in an experiment for instance; the concept of antiparticle was first introduced theoretically by Paul Dirac in 1930 and confirmed two years later by the experimental discovery of positron by Carl Anderson (Ejlin, 1999). Scientific knowledge is gained in a variety of ways, including observation, analysis, speculation, library investigation and experimentation.

Scientific theories and laws:-

Within the given framework, the laws are seen as empirical propositions, which describe certain uniformities that exist in the universe, for instance Kepler's laws of planetary motion describe motion of planets. Laws can be verified by experimentation based on observations or based on inferences (Newton's laws). However, in some cases, laws describe what would happen under ideal conditions such as Boyle's, Charles's law for ideal gases, Galileo's laws of falling bodies describe the velocities with which the bodies would fall in a vacuum. Generally, laws are descriptive and discovered.

Theories are inferred explanation for observable phenomena and are tested by experimentation. Scientific theories are well established, highly sustained, internally consistent systems of explanations. A good theory makes predictions, which can be tested for its validity. However, some theories in natural sciences cannot have definite predictions, for example, Darwin's theory of natural selection and survival of the fittest which is a probabilistic theory. Scientific theories are human constructions, but have passed the guessing stage: they have substantive evidence that supports them, but it doesn't become the law, even if very strong support is there to substantiate the theory as many students tend to believe (Meyling, 1997; Ryan and Aikenhead, 1992), but it becomes a more credible theory. Once a theory become established, a large amount of disconfirming evidence is required before it can be challenged. Also, as seen from the students' responses from VOSTS and VOSE questionnaire, the majority of the students held the misconceptions about scientific theories and laws.

Students seemed to have a simplistic, hierarchal view of the relationship between theories and laws whereby theories become laws depending upon supporting evidence. Laws have higher status than theories.

Students' Views about 'Models' of the Atom:-

The framework remained the same as above for understanding students' conceptions of the structure of the atom and atomic models as part of the nature of science. The major emphasis was on the historical as it indicates evolution of scientific truths, philosophical as it emphasizes the importance of a blend of imaginative and logical thinking in the generation of new ideas and epistemological dimensions of the nature of science which is implicit in the knowledge about the structure of the atom and hence possess wide scope to develop these aspects in students.

Review of the NCERT Textbook:-

In this study, a brief analysis of the chapter "structure of the atom" present in the NCERT textbook in India is discussed on the basis of the framework developed by Niaz (1998) and Niaz et al. (2008). A slight modification was done as per the requirement of my research –An additional criterion related to use of thought experiments (Heisenberg's microscope) in understanding the quantum mechanical model was incorporated which already has been used in the study by Malhotra (2013).

The framework is based on the understanding that history, philosophy and epistemology are must to be included in the chemistry textbook particularly in the chosen chapter i.e. structure of the atom. The analysis looks if the NCERT textbook involves these aspects and to what extent. For this analysis, Niaz(1998, p541-542) developed a framework based on the rational

recommendations of developments that led to the formulation of atomic models suggesting that the framework will help teachers develop insight as to how a theory or model develops.

The framework was developed by analyzing primary sources like original papers of Thomson, Rutherford, Bohr and some secondary sources. This framework consists of two criteria to evaluate Thomson's model and three criteria to evaluate the Rutherford's and Bohr's models. In addition, five criteria developed by Niaz and Fernandez (2008, p881-883) to evaluate quantum mechanical model were added to the framework. The criteria are discussed in brief:

1. Thomson conducted his cathode ray experiment in the background of controversy against ethereal theory. His experiment aimed at finding out whether the cathode rays were charged particles or waves. The experiment was conducted to find out the mass by charge ratio to investigate whether the cathode rays were fundamental charged particles or ions.
2. Rutherford and his colleagues while working on radioactivity observed something extraordinary which diverted them from their original work towards the study of atom. They had observed that on bombarding the gold foil with alpha particles; 1 in 2000 rays deflected back through large angles and to explain this they proposed a nuclear model of atom. This model had to compete to already existing Thomson's model. Both Rutherford and Thomson tried to defend their theories by giving possible explanations for the results of alpha particle scattering experiment.
3. Bohr's mathematical calculations proved that Rutherford's model was unstable and Bohr decided to address the problem. Bohr thus, proposed a new model of the atom. His main objective was to tackle the instability of the Rutherford's atom. His proposed model was able to explain the line spectrum of the Hydrogen atom, but Bohr himself was unaware of this line spectrum when he proposed his model. The beauty of his model was that it could incorporate Planck's quantum mechanical theory and Maxwell's electromagnetic theory, which are two rival frameworks. Bohr used these two theories to formulate his model and the salient features of his model were quantization of energy and angular momentum.
4. The developments that led to the quantum mechanical model were Planck's and Einstein's contributions to which quantum mechanics provided a better answer. The comparison of the classical and quantum mechanics addresses many misconceptions about the structure of the atom. The quantum mechanical model is analyzed on the basis of success to address a major obstacle for students between orbital and electron density. The last criterion for evaluation of quantum mechanics is related to the concept of quantum numbers, how these are introduced in the textbooks, what is the basis of the formulation of quantum numbers.

On the basis of this framework, many chemistry and physics textbooks have been analyzed.

The above-mentioned studies illustrate that most of the textbooks emphasize on experimental details and observations. There is a dearth of mentioning about how the scientists make decisions, solve problem creatively with incomplete information, propose many hypotheses and judge which fits best and use mental formulation to create theories. The curtailed historical and philosophical aspects in the textbook might give rise to the misconceptions students may possess.

The suggested influence of historical treatment in the topic structure of the atom on the students' understanding of the topic prompted the researcher to look into the NCERT textbook which is accessed by a large number of students.

The Nature of the Study and Methodology:-

The present study has a qualitative research model and requires an understanding of the processes, events and relationships in the context of the social & the cultural situation (Creswell, 2012).

In this study, we wanted to explore how students understand the nature and process of science viz. how science progress, how scientists work as a social group and not in isolation. How the NCERT book (which is the major course book in science curriculum in Delhi) developing essential components of the nature of science in students?

Essential themes of the nature of science in 'structure of the atom' were selected which include historical, epistemological and philosophical aspects of science, tentativeness of theory, observation and inference (conception of atomic models), theoretical embeddedness of scientific knowledge and personal values of scientists affecting their research. The sample size of 30 senior secondary students and 15 undergraduate students were taken and their views were deciphered with the help of a questionnaire. The questionnaire had two sections A and B. Through Section A, we intended to elicit the general understanding about the NOS amongst 11th grade and college students. Whereas the section B was theme specific through which understanding of 'structure of the atom' was probed because it further helped in analyzing students' understanding of the nature of science. The questions were purposefully drawn and modified from VOSTS questionnaire to suit social and cognitive contexts of our learners.

This was followed up by a teaching module which students were asked to read and jot down those points which made better understanding of the topic in them and wanted to be in the chapter 'structure of the atom' in the NCERT. They were also encouraged to write down about those topics which they do not understand by giving reasons (could be pedagogic reasons or language of the book, not interesting illustrations etc.).

The next step was to deeply probe them for what they had answered in questionnaires through FGD (Focus Group Discussion).

Data Analysis:-

Analysis of items in the questionnaire (adapted from VOSE and VOSTS) are done using the frameworks adopted by their developers.

Section A (I) consisted of VOSE items. The framework for analysis is based on a suggestion in the VOSE questionnaire (Chen, 2006). Analysis of the VOSE items is done using a convention that except few statements in the items, the rest of the NOS items may be considered acceptable as diverse views among students and these are not wrong or naive. This convention implies that NOS is not fixed set of beliefs and so students need not conform to certain views. All students cannot have similar views about the nature of science and may perceive science differently and hence its major aspects like nature of science, process, development, scientific activities may be comprehended differently by them.

Thus, students are permitted to have diverse viewpoints about the nature of science. The developers of the test suggest that emphasis should be not on making students possess particular

views about the nature of science, but rather urge that provision should be given to students' justifications they give in support of the stance they choose for their views and beliefs about science. The need is pointed towards allowing students to develop their own conception of the nature of science with the scope of discussion in the classroom so that if there is misperceived or alternative conception about science, then that should be brought to an informed level by proper discussion and arguments which should fully satisfy students' curiosity and area of confusion. There is mention that legitimate/ official and accepted knowledge of 'Scientists' and philosophers' should be made known to students, but not taught in a way as to stamp it as if there could be no other valid knowledge than those which are documented in the texts.

The given below are those items which the developers of the questionnaire considered as the partial or incomplete views about the nature of science and considered these views as naïve (immature, raw, inexperienced) views. The items written here are from the questionnaire used for this present study. The numeral and alphabet denotes the item number and option arrangement in the item respectively.

6A, 6B, 6F- Concern with universal scientific method.

6A: The scientific method ensures valid, clear, logical and accurate results. Thus, most scientists follow the universal method in research.

6B: Most scientists use the scientific method because it is a logical procedure.

6F: No matter how the results obtained, scientists use the scientific method to verify it.

9A, 9B- Laws are more certain than theories

9A: Yes, theories are not as definite as laws.

9B: Yes, if a theory stands up too many tests it will eventually become a law; therefore a law has more supporting evidence.

2C, 2D, 2E- Using no imagination in science

2C: No, imagination is not consistent with the logical principles of science.

2D: No, imagination may become a means for a scientist to prove his point at all costs.

2E: No, imagination lacks reliability.

4C, 4D- No influence of socio-cultural milieu on the scientific investigations.

4C: No, scientists with good training will remain value free when carrying out research.

4D: No, because science requires objectivity, which is contrary to the subjective socio-cultural values.

5C, 5D-Highlight no influence of personal beliefs on scientific investigation and observations are considered to be independent of theories.

5C: Observations will be same, because through scientific training scientists can keep aside personal values to conduct objective observations.

5D: Observations will be the same, because observations are exactly what we see and nothing more. Facts are facts. Interpretations may be different from one person to another, but observations should be the same.

We present here, the pattern of responses to VOSE items.

The tabular details of students' responses to all VOSE items are given in Appendix 4. In the parenthesis, I have kept the total number of responses for particular stance i.e. strongly disagree(SD), disagree(D), neutral(U), agree(A) and strongly agree(SA) against the given options and in another line the respective breakup of responses are given for college (C) and

school(S) students. As I have explained in detail in chapter 3 about the sample size of both the groups which is: College students =15(F; 7: M; 8) initially and FGD was done with 7 college students (F; 4: M; 3). School students = 30(F; 12: M; 18) initially and FGD was done with 10 school students (F; 4: M; 6).

The following is the analysis of VOSE items:

Item no.1 ‘when two different theories arise to explain the same phenomenon, scientists will accept the two theories at the same time.’ deals with the progress of scientific knowledge which is often characterized by competition between rival theories. Textbooks do not include examples of such nature where controversies and conflicts between theories are shown. The product aspect of science is presented in the textbook as if some theory, model, law, discovery etc. in science is made without actually understanding the kind of situations these scientists had to undergo i.e. no mention about their ecstasy on finding something, fear of financial uncertainty, frenzied stages in the process of discovering something etc. in the textbook which makes science appear smooth and struggle free. So, students might not develop the understanding about science in the making.

The observations which the researcher has made fall on similar lines. The majority of the college (53%) and school (63%) students have similar views on this aspect when they think that two theories which come up to explain the same phenomenon or concept at the same time are accepted tentatively because scientists cannot objectively tell which one is better. 73% college and 92% school students agree with the position that the two theories are accepted because they explain the phenomena with different perspectives, no one being right or wrong.

For another position in this item where, options imply that only that theory is accepted with which the scientists are more familiar and that scientists tend to accept the simpler theories and avoid complex theory; substantial section of students (66% college and around 70% of school students) disagreed with these stances. 26% of college students and 50% of school students strongly disagree with this position that academic status of the person who proposed the theory will influence other scientists’ acceptance of the theory. Very few students (13% college and 6% school students) take up the stance in favor of it. This might reflect the view of science they have. They might perceive science as secluded, objective, free from human subjective interferences and therefore, they take up those options where science is seen as unaffected by power, prestige and other influences.

There are an equal proportion of sample students who agree and disagree towards the position which says that the theory which deviate less from the current core scientific theory is accepted and the one with more deviation is not accepted.

30% of school students and 26% of college students strongly agree with the position that there is only one truth, and scientists will not accept any theory before distinguishing which is best. This position taken up by the students points towards the general notion that truth is objective and is independent of subject which comes from the strong positivist framework of scientific method. The dichotomy between how science is actually practiced and depicted shows the need to teach science as actually practiced for a better understanding of the dynamics of scientific progress (Niaz, 2011, p.7).

Item no. 2 deals with the importance of imagination in scientific research. It requires creativity to imagine a pattern to fit into the existing framework or go beyond to explain the problem. It helps in solving the big jigsaw puzzle. The findings reveal that majority of students (54% college and 84% school) acknowledges the use of imagination in the scientific research.

But there are substantial numbers of (53%) school students who think that imagination lacks reliability, whereas only 13% of college students think so. Also, there is ambiguity in the thought of school students as they take the position that imagination is the main source of innovation and is used more or less in scientific research and also agree with the position that imagination might become the tool for scientists to prove his point at all costs and also it lacks reliability (S9, S5; FGD). As per the suggestion to infer the views as explained above; these students possess naïve views about the role of imagination. The students were followed up in FGD.

The larger number of college students, recognizing the importance of imagination in science may come because of longer exposure to science and scientific practices as they study similar topics like atomic models, rules and principles to explain the nature of electron in much detail in graduation than in class XI. But this needs to be explored because even at this level majority of students think that imagination cannot be used in science; “it is the thing to be used in literature” (C6, C5: FGD). Around (26%) students are going for uncertain and neutral stance. This might reveal that they do not know the topic well and hence, playing safe by not being on either side of extremes to avoid being asked further. However, the researcher could not follow this pattern up afterwards to elicit the reasons from these students for being uncertain in many items due to time constraint.

Item no.3 deals with the tentative nature of scientific knowledge and starts with the statement- ‘Even if the scientific investigations are carried out correctly, the theory proposed can still be disproved in the future.’ There are 66% of college students, who disagree with the position that scientific research will face revolutionary change, and the old theory will be replaced; whereas 50% of school students agree with this position. Though the response cannot be considered naïve in either case; but it may point toward college students looking at the scientific progress as building up on the earlier theories and thus, looks in terms of evolutionary nature of scientific progress whereas school students seem to look at them as sudden fall of a ‘well established theory’ because of some anomalies and rise of a new theory to substitute the earlier one. Substantial section of college and school students goes for the position which talks about cumulative nature of scientific advances and hence the old theory is preserved. There are 53% of college and 56% of school students who think that with the accumulation of research data and information, the theory will evolve, finding additional and useful piece and clue about the problem and thus explaining it even better. The responses of students show that they have the idea about the tentative nature of scientific knowledge as in their textbook; they find the representation of various discoveries and scientific advances in a chronological manner, giving them the idea that scientific research does not remain static but constantly undergo certain changes and thus are tentative in nature. However, linkages among various theories and history of its development are not presented in the textbook (Chapter 3, section 3.1.2).

Item no.4 concerns with socio-cultural embeddedness of scientific knowledge and the statement is- Scientific investigations are influenced by socio-cultural beliefs and values present at a given

time. The response of both the sample students indicates that they have held naïve views about this aspect which has significant role to play in the scientific knowledge. 60% of college students disagree and 33% of school students strongly disagree with the position that socio-cultural values influence the direction and topics of scientific investigations. They possess similar ideas for the position which says scientists are influenced by socio-cultural values and they disagree with this stance. 40% college and 33% school students strongly agree with the option that scientists with good training remain value free and thus do not get influenced by socio-cultural values and hence carry out neutral and objective research. 26% college and 72% school students think that science is an objective sphere and is contrary to the subjective socio-cultural values; so in scientific research, socio-cultural values and beliefs have no role to play. These are clearly the naïve views students have upheld. Only 6% of college and school students acknowledge that even scientists are social beings and that they cannot be context free and subjectivity will come into place while carrying out research. The root to these notions was required to be elicited from the students to know why and how they think about scientific activities, so the theme of the item (socio-cultural beliefs and values influence scientific investigations) was kept for FGD.

Item no.5 deals with theory laden nature of scientific knowledge and the statement is- Scientists' observations are influenced by their personal beliefs therefore; they may not make the same observations for the same experiment. 40% of college students disagree with the position that 'observations' will be different, because different beliefs lead to different expectations influencing the observation', however, only 16% of school students think the same. There are 63% of school students and only 26% of college students who agree with this position. This may appear that college students are more steeped into the thinking about science as objective reality only and no scope of any context and subjectivity remains on the part of scientists but good number of school students has informed view on this position. Again, this difference in the opinion of two groups needed to be explored so was kept for FGD with both the groups separately. 46% college and 33% school students hold naïve views in the position which says that observations will be the same, because scientists with training can keep aside personal values to conduct objective observations. 40% college and 57% school students have naïve views for the position as they agree with it which implies that observations are considered to be independent from theories. Also, Karl Popper puts it "all observations have a frame of reference and horizon of expectations" with necessary selectivity.

Item no. 6 deals with the myth of 'scientific method'. 53% of college students and 93% of school students hold naïve views as they agree with the position which states that the scientific method ensures valid, clear, logical and accurate results. Thus, most scientists follow the universal method in research. Again, 80% of college students and 93% of school students have naïve views as they think that most scientists use scientific method because it is a logical procedure; as if undermining other methods of research too as logical procedure. 57% school and 26% college students hold the naïve views as they think that the only method to verify the obtained results is scientific method.

Item no. 7 and 8 are not discussed here because we could not get consistent responses of students for these items. These items are related to the epistemological status of scientific knowledge. The items intended to understand students' position for invention and discovery and what they think of laws and theories in terms of these.

Item no. 9 deals with the understanding about law and theory amongst students. 66% school students and 20% college students hold the naïve view as they agree with this statement that theories are not as definite as laws. However, 40% of college students have informed views in relation to 20% of school students. Substantial section of the sample students hold naïve views about the law and theory where they look upon a theory which can become laws on more supporting evidence and after standing to many tests. There were 93% of school students and 60% of college school students who think in this manner. Understanding about theory and laws in students was followed up in FGD.

The findings from VOSE are summarized in the following table:

Dimensions of NOS	Likert scale	Strongly Disagree	Disagree	% of disagreement	Neutral	Agree	Strongly Agree	% of Agree.
Tentativeness of scientific theory	2	4	13	5	25	9	76	
Observation is theory laden	14	15	64	4	4	8	27	
Diverse scientific methods	1	8	20	15	16	5	47	
Theories and laws are invented	3	11	31	9	20	3	51	
Theories and laws are different ideas	2	17	42	14	5	7	27	
Use of imagination in science	2	4	13	11	12	16	62	
Empirical evidences validate scientific knowledge	3	4	16	7	20	11	69	
Subjectivity in science	4	11	33	4	6	20	61	

The table reflects the thought profile of the sample group (combined college and school students). As indicated, majority of students (seventy six percent) recognized the tentativeness of scientific knowledge. However, few students (twenty seven percent) opted for theory laden nature of observation and hence influence in the research. Considerable number of students (sixty two percent) appreciates the use of imagination in scientific research. Forty six percent of students chose for the positions of diverse scientific methods.

Around twenty six percent held the position of laws and theories as different ideas and majority of them look at these as related hierarchically. Around half of the students think that theory and laws are invented. From the findings, it appears that some aspects of nature of science like tentativeness in science, imagination in science, need of diverse scientific methods in science are known to most of the students. But the aspects like subjectivity, theory laden nature of observation are not understood to them as appeared from their responses. They lay more emphasis on the empirical method of gaining scientific knowledge and thus undermining other means of generating scientific knowledge like observation, thought experiments. It was found in

the NCERT that there is dearth of mention about the varied methods of scientific investigations used by different scientists to give various atomic models.

Students' Conception about 'Model' of the Atom:-

This part (Section B) of the questionnaire had used the concept of 'structure of the atom' to focus on the tentative, inferential, and developmental nature of the scientific knowledge.

The items in this section of the questionnaire were adapted from the teaching module developed by Dabas (2010) in her thesis to develop understanding about the NOS in the students who are studying these topics. The items in this section provided an opportunity to understand the students' conception of the atom which reflects their understanding about the nature of science. The ten item questionnaire (open-ended) helped elicit their intuitive ideas which showed that:

- About thirty three percent of school students believed that atoms were solid particles in response to item no-1. They listed different reasons like- atoms are always depicted as sphere and circles, having perfect shape in the science textbooks; they say "if they were not solid then how this structure would have been maintained and matter would have been compressible then how orbits and orbital electrons would remain in the atom" (S1: FGD). Another group of students was saying that everything in the universe is made up of atoms and these are building blocks of everything; if the atoms were not solid, then how the matter around us would have got fixed shape. One student wrote very differently that "light passes through liquids and gases, but we see that at a shortest and smallest place which is negligible the light cannot pass through which depicts its solid nature" (S6: FGD). What the student meant by this was followed up with and is discussed in section (4.3). Others gave reasons like-atoms have definite mass and occupy space; atoms are made up of neutron, proton, and electron with a nucleus at the centre so it should be solid. Ben-Zvi et al (cited in Driver et al (1994)) working with Israeli children found that pupils often regard atoms as 'small bits of solid' and it is widely accepted that this misconception persists even when they go to University. Around 20% college students held the similar notions that atom were solid particles because everything ultimately is solid and that is why after some extent they are incompressible and cannot be broken further.
- About forty six percent school students said that atoms were not solid particles as a liquid, and gases also have atoms. These students tend to think that 'solid particles' related only to the solid state of the matter. Some had the views like atoms appear like cloud which is not solid and it has lots of spaces in it. One student wrote, "Combination of atoms can be judged into solid, liquid and gases. A single atom cannot be judged into solid, liquid and gas. So, the classification of this sort is baseless" (S2: FGD). Similar types of views in students are reported in the research of Dabas (2010) like students think that if atoms were solid, then they could not have existed freely in the nature. Clearly, there is much more that needs to be understood about students' conception related to atoms and states of matter.
- 70% of college students say they do not agree with atoms as solid particles and they also listed the similar reasons as that of school students for explaining why they think so. Some say, because we see penetrable material around us which means atoms are not only solid particles. Again, what do they understand about the nature of constituents of solid, liquid and gas need to be understood. One student wrote "atoms appear wavelike so they must be made up of wave like things" (C4: FGD). This view might have come from the representation they find in the books without explaining in an engaging manner.

- Scientists now have a complete understanding of the atom (statement 2). 28% of school students held the view that we have complete understanding of the atom as we have well developed quantum mechanical models which explain most of the properties, when probed further, they could not explain what type of properties are those which earlier theories could not explain but the quantum mechanical model can explain. It appears that they may have crammed what heard from teachers and read in the books only as information without understanding it. But most of the students disagree with it and give reasons like, what we know today changes tomorrow with advancement in technology, researches are still going on to know more about the atom and its structure. Still conclusively we cannot say that we know accurately and completely about atoms. Many said that we still doubt whether entity called atom exists or not. We know more than our earlier times but should not become complacent about it. College students also had similar views to share, but the difference in the reasons they were giving like- one student said “we have complete understanding about the atom because then only we are able to study the properties of elements as they are made up of atoms so we have to know them before” (C1: FGD). Another group of college students says that knowledge construction and development is an ongoing and dynamic process.
- Atoms are mainly composed of empty spaces. Around 45% school students agreed with this statement. They related this to the Rutherford’s experiment which showed lots of alpha rays passing through gold foil straight. Some also related this to Bohr model which talks about discrete orbits and shells like-K, L, M, N etc. and the rest being the empty spaces in between. College students also believed that atoms have empty spaces and gave reasons because we know about the Rutherford’s alpha scattering experiment.
- In the response to the question where they were asked to draw the structure of the atom; most of the school students (80 percent) used the analogy to the solar system and drew accordingly nucleus at centre and electrons revolving around it as they understood of Rutherford’s nuclear model. In spite of having studied more advanced models to elucidate the structure of atom viz. Quantum mechanical model; they were using this model. On probing further, it was found that students could retain it because of its simple appearance and easiness. However, most of the college students tried to depict the atom according to the quantum model which gives a probabilistic picture of finding the electrons.
- In response to item no.5 which says that since scientists cannot see inside of an atom, it is not possible to believe any of these models. About 45% of school and 30% of college students indicated that scientists had seen the inside of the atom. Some thought that with the help of modern technology like STM (Scanning Tunneling Microscope) etc. we can see the interior of the atoms. They had no clue that the models need not be always representing the physical reality, but could also be based on interpretations. They also had no idea about the difference between inference and observation though they use these terms in their practical classes. Some were not sure about imagination explaining the real phenomena. Similar types of views have also been expressed in other researches (Bent, 1984; Dabas, 2010).

Such students appeared to possess a naïve epistemological view of scientific knowledge (Nadeau and Desautels, 1984). They viewed the structure of the atom as copy of reality, rather than as the constructed representations that embodied certain theoretical perspectives. The most common rationale for students’ explanations of the change in science was attributed to better equipment or technology.

When asked about examples of a model; about seventy percent listed the models like that of a heart, skeleton, windmill, eye, etc. Majority of the students were having the idea about models as a working model as they are in the habit of using the term without knowing the difference between mental and physical models. All these examples indicated that students had understood model as a three dimensional picture and a proven fact (Lederman and O' Malley, 1990).

Discussion:-

The primary aim of this study was to understand the students' views on the nature of science and the role played by the NCERT chemistry textbook in developing this understanding in them. In this study, it was attempted to understand what are students' understandings and perception towards the nature of science and science processes.

Scientific content, processes and the nature of science overlap and interact in significant ways and all are important for students to learn. For example, observing and inferring is a scientific process. On the other hand, an understanding that observations are constrained by our perceptions and are inherently theory-laden is part of an understanding of the nature of science. The tentativeness of scientific knowledge is not limited to the recognition that scientific knowledge has changed through history. Rather, science draws its tentative and revisionary characteristics of the development of theories and the limitations imposed on ways of knowing. We present the conclusions of the current research in this section as follows:

The review of the NCERT chemistry textbook reflects that historical and philosophical treatment which is required for better understanding of the topic 'structure of the atom' is not included in the text (Appendix 1 and section 3.2.2). Also this affects students' content knowledge of the topic as was found in this study that majority of participants had weak content knowledge.

Students' views on various dimensions of the nature of science were identified using the VOSTS and VOSE questionnaires. Also, students' understanding about models of the 'structure of the atom' was probed as part of the nature of science by using the teaching module designed by Dabas (2010).

Over one third of the students held the naïve view that scientific knowledge, including theories and laws does not change but is final and fixed and static as seen from the VOSE questionnaire (Item no.3). The FGD with college and school students revealed that they understood that the scientific concept like the 'structure of the atom' changes in the light of new experiments. About two thirds of students recognized that scientific knowledge is not absolute and final but tentative in nature. It was also observed that an understanding of the role of imagination in science blend of inference and observation required to reach a scientific concept could be developed in students provided the scope for discussion with them is there. As was seen in this study during focus group discussions the students who earlier said that scientific knowledge is more reliable if it is proven empirically later on came to the point that the role of inference in scientific results are also very important because not everything in the universe can be seen or observed. We have to do thought experiments and infer on the basis of some premises (Thomson's plum pudding model) as in the case of the 'structure of the atom'. This modification in their perception came about after the 'tricky track activity' developed by Lederman was performed with students.

Students could make out that some events are explained on the basis of inference because there may be the possibility that no one was present at the time when the event had happened.

Students had some understanding about the tentative nature of scientific knowledge as they gave reasons like better equipment lead to improved observations which may modify or add to the existing knowledge thus scientific knowledge is never final. Fifty five percent of school and thirty percent of college students held the naïve view about 'atom and its structure'. As they seemed to believe that for scientists to know and be sure about the structure of the atom they have to see them. Scientists cannot be certain unless they 'see' the atoms or anything else they want to investigate.

Concerning the cultural and social aspects of scientific knowledge students initially felt that scientists and their research remain uninfluenced by this aspect, but during FGD they could understand that same phenomena like that of biofuels or GM foods can be investigated differently by different scientists belonging to different social and cultural contexts. They recognized that research priorities are determined by the need of the country so developed and developing countries would look upon the problems differently.

In the VOSTS over fifty percent of college and sixty percent of school students held the naïve view that models are copies of reality (section 4.3 statement 1). While discussing with students during FGD this was revealed that students could recognize that models which explain 'structure of the atom' are the mental representation and a guess based on experimental results (Section 5.2; FGD). However, in the theme of 'scientific method' there were very few students at both the levels who think that scientific knowledge not necessarily develop by following 'scientific method' only but it may arise by chance imagination etc. also. Others thought that in any situation scientific knowledge to develop; firstly, they have to encounter the problem and then other steps have to be followed which we call scientific method. They seem to undermine the role of creativity, imagination and the open mindedness of scientists. This persistence may be attributed to the strong influence of school textbooks and other materials which conjecture events of chance and serendipity as part of the process of discovery.

Students at both the levels held misconceptions about theories, laws and hypotheses. For them all three concepts are related hierarchically on the basis of supporting evidence in a progressive manner, i.e. hypothesis becomes theory and theory becomes law on accumulation of more supporting evidence.

It was observed during interaction with students of grade 11 that those students who had availed congenial environment at home i.e. where discussion about science content about scientists, etc. (with the help of family members like parents, elder brother etc.) takes place had comparatively better understanding about the nature of science. These students held informed views on certain dimensions of nature of science pertaining to theory laden nature of the observation role of imagination in science, social and cultural embeddedness of scientific knowledge tentativeness in science and so on in comparison with other students who had no one at home or outside to discuss with about such issues.

During FGD it was observed that students were able to better relate to the discovery of atom various events which had taken place in explaining about the structure of atom on being provided with a teaching module (Dabas 2010) to read. This step was not the primary focus of the research, but was done in a cursory way to understand how some enriching sources could help them develop a better understanding of the nature of science. Positive remarks from students at both the levels came. They found it interesting because the development of 'atom and its structure' was written with the mention about the contexts in which the theories or models were given with links among various discoveries in this particular area. The narrative was written with the inclusion of history and philosophy of science which got students interested in understanding the journey 'atom' travelled to come in its present form.

Then school students were asked to express how they would want this topic 'structure of the atom' to be written in the NCERT chemistry textbook many had written language that should be made simpler and commonly used. In a way, their writings implied that technical and jargonized language in the textbook complicates the flow of understanding of the scientific concepts. However, we did not analyse the textbook on these lines. This step was taken up as a suggestion from those for whom we want the textbooks to be written. They had also pointed that coherence should be there throughout the chapter because new topic starts without any link with the earlier topics which create confusion and understanding is not built and we are forced to cram this information to pass examination.

Implications:-

The science education and scientific researches call for special attention on science education right from earlier classes. So, that students may be able to relate to scientific concepts better in its entirety from beginning. The textbook has a great role to play because students and teachers use it as the main book and rely on them heavily. Bodies like the NCERT could make an effort to write the science textbook at senior secondary level in such a way that students better relate to the concepts taught. It may help them to develop interests to continue further in scientific research. This could be possible only when their understanding about the NOS is strong. FGD gives a ray of hope that if discussion in the classroom includes the aspects of the nature of science may enhance knowledge of the science and about the science.

Science education aims at preparing individuals to have a better understanding of the world. This may enable them to take responsible decisions to appreciate the natural world help in its protection and respond towards changing technology sensibly. This may also encourage them to learn the science processes to understand and analyze situations. Therefore, it is necessary to design textbooks with more explicit representation of aspects which develop understanding about the nature of science in students. This study also recommends the need to design textbook according to the frameworks of HPS for 'structure of the atom' and Lederman pertaining to the NOS in the whole textbook.

It is noteworthy that the NCERT could aid in the proper incorporation of the nature of science in curriculum. It could also train the teachers to support pedagogical methods where they provide learners to engage in actual processes of science. The understanding of the nature of science is important in creating and reviving interest in science in addition to the work of scientists. Since

knowing about the ways the theories are developed may better help them understand process of science.

The relevant and fruitful activities and situations should be provided to students to allow them to challenge their conception about various aspects of the nature of science. Projects and home assignments may be given to students, which require the use of aspects of the nature of science and science processes. For example, types of questions that are raised in the teaching modules developed by Dabas (2010) such as - Controversies in science and the media over genetically modified food Is there one scientific method scientists and the language they use, etc. If students are exposed to these types of debates requiring their own explanation and ideas may likely improve their understanding about science.

The topic 'structure of the atom' has wide scope to convey the philosophical, historical epistemological and psychological aspects of the nature of science to the students. The curriculum designers and textbook writers could think about incorporating chance and serendipity. These could be emphasized as one of the processes of discovery of scientific knowledge with more elaboration. It has been seen that one or two brief examples in the textbooks may not be effective in steering the naïve views about discovery in students away. As other researches also claim that the change in students' views on the nature of science is a difficult process and take place gradually and progressively (Driver et al 1996).

Conclusion:-

This study is exploratory in nature and has found that there is a need to develop interesting and engaging reading materials for better content development of students as well as for developing an understanding about the nature of science in them. Also, the current examination system may need a redress to revisit what it purports to test amongst students. What does it emphasize on (understanding about the process of science, nature of science or merely seeking/ justifying mugging up some concepts to pass the exam)? Is the current practice of examination system able to create scientifically literate citizens? All these questions arise because they serve a better guide to those who are science educationists, policy makers, researchers so that immediate and collective steps may be raised with the collective benefit of our young learners of science, to make our citizens scientifically literate and most of it can be achieved when there is an emphasis from all sources of education to develop an understanding of the nature of science amongst students.

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Annexure 1

Criteria Chosen for a Review of the NCERT textbook

Thomson Model

T1- cathode rays are charged particles or waves in the ether.

T2- Determination of m/e ratio to decide whether the cathode rays were ions or a universal charged particle.

Rutherford Model

R1- Nuclear atom

R2- Probability of large deflections is exceedingly small, as the atom is the seat of an intense electric field.

R3- single/ compound scattering of alpha particles.

Bohr's Model

B1- Paradoxical stability of the Rutherford model of the atom.

B2- explanation of the hydrogen line spectrum

B3- Deep philosophical chasm. Bohr's incorporation of Planck's 'quantum of action' to the classical electrodynamics of Maxwell represented a strange 'mixture' for many of Bohr's contemporaries and philosophers of science. This episode illustrates how scientists, when faced with difficulties, often resort to such contradictory 'grafts.'

Quantum Mechanical Model

Q- Heisenberg's gamma ray microscope: It emphasizes on the significance of thought experiment in scientific research in situations where laboratory setup is not feasible. This criteria was developed by Malhotra (2010).

Q1- Origin of the quantum hypothesis

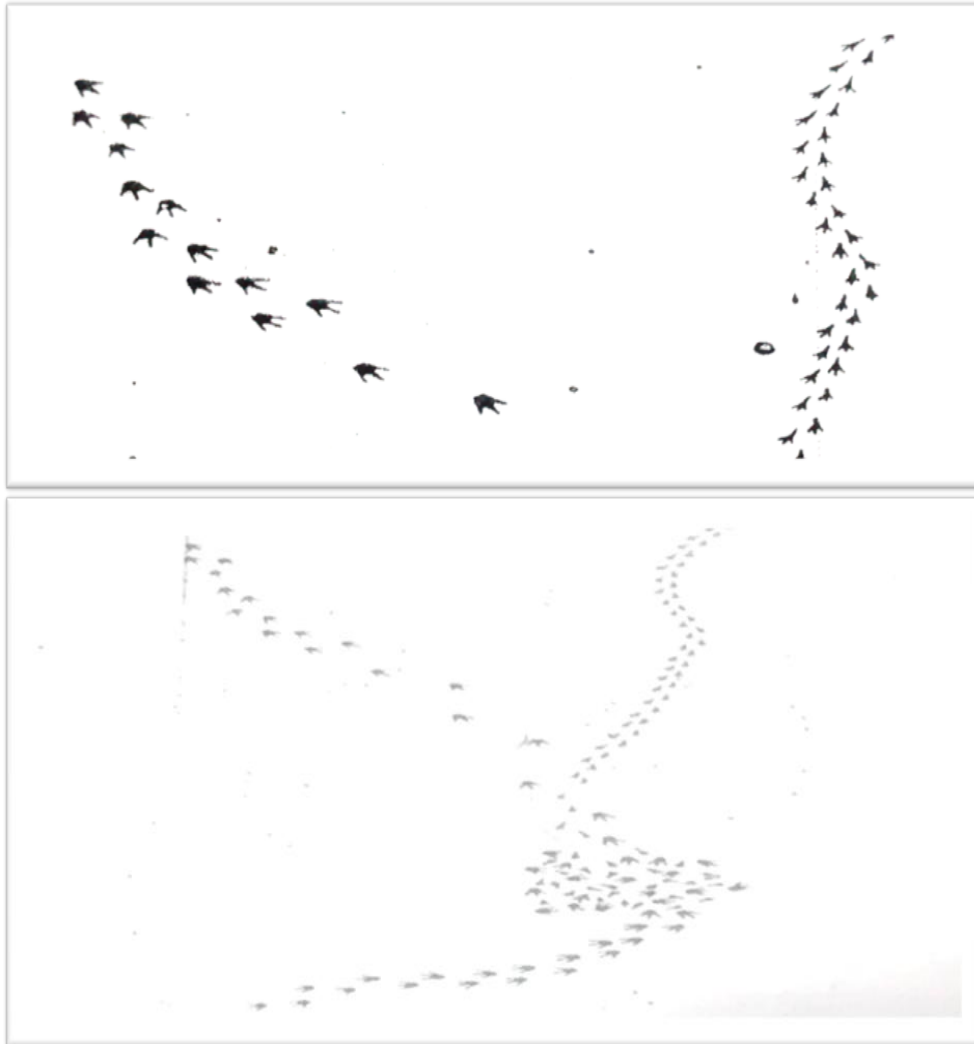
Q3- Differentiation between an orbital & electron density.

Q4- differentiation and comparison between classical and quantum mechanics.

Q5- Introduction of quantum numbers based on electron density.

Annexure 2

Pictures shown to participants to perform 'Tricky Track Activity'



Picture 1:- (above) and 2 (below)

Annexure 3

Questionnaire used for Understanding Students' Views on the Nature of Science

Section–A (I)

Instructions to participants: Each item of this questionnaire starts with a statement about the nature of science. You may strongly agree (SA) with it, strongly disagree (SD) with it, or have other thoughts about it. Each statement is followed by several responses. Please read all the responses first, then circle your opinion on the right side (SD, D, U, A, SA) of each response according to *your* knowledge of how scientists work and science develops. There is no right or wrong answer. Thank you.

Note:

SD= Strongly Disagree

D = Disagree

U = Uncertain or No Comment

A = Agree

SA= Strongly Agree

1. When two different theories arise to explain the same phenomenon (e.g. the nature of electron), scientists will accept the two theories at the same time.

A. Yes, because scientists still cannot objectively tell which one is better; therefore, they will accept both tentatively.

SD D U A SA

B. Yes, because the two theories may provide explanations from different perspectives, there is no right or wrong.

SD D U A SA

C. No, because scientists tend to accept the theory they are more familiar with. SD D

U A SA

D. No, because scientists tend to accept the simpler theories and avoid complex theories. SD D

U A SA

E. No, the academic status of the person who proposed the theory will influence other scientists' acceptance of the theory. SD D

U A SA

F. No, scientists tend to accept new theories which deviate less from the current core scientific theory.

SD

D U A SA

G. No, scientists use intuition to make judgments. SD D

U A SA

H. No, because there is only one truth, scientists will not accept any theory before distinguishing which is best.

SD D U A SA

2. While scientists are conducting scientific research, they also use their imagination.

A. Yes, imagination is the main source of innovation. SD D

U A SA

B. Yes, scientists use their imagination more or less in scientific research. SD D

U A SA

C. No, imagination is not consistent with the logical principles of science. SD

D U A SA

D. No, imagination may become a means for a scientist to prove his point at all costs. SD D

U A SA

E. No, imagination lacks reliability. SD

D U A SA

3. Even if the scientific investigations are carried out correctly, the theory proposed can still be disproved in the future.

A. Scientific research will face revolutionary change, and the old theory will be replaced. SD D

U A SA

B. Scientific advances cannot be made in a short time. It is through a cumulative process; therefore, the old theory is preserved.

SD D U A SA

C. With the accumulation of research data and information, the theory will evolve more accurately and completely, not being disproved.

SD D U A SA

4. Scientific investigations are influenced by socio-cultural beliefs and values present at a given time (e.g., religious beliefs, social bias about gender, caste, class etc.).

A. Yes, socio-cultural values influence the direction and topics of scientific investigations. SD D U A SA

B. Yes, because scientists participating in scientific investigations are influenced by socio-cultural values.

SD

D U A SA

C. No, scientists with good training will remain value-free (neutral) when carrying out research.

SD

D U A SA

D. No, because science requires objectivity, which is contrary to the subjective socio-cultural values.

SD D U A SA

5. Scientists' observations are influenced by their personal beliefs (e.g., personal experiences, presumptions) therefore; they may not make the same observations for the same experiment.

A. Observations will be different, because different beliefs lead to different expectations influencing the observation.

SD D U A SA

B. Observations will be the same, because the scientists trained in the same field hold similar ideas.

SD

D U A SA

C. Observations will be the same, because through scientific training scientists can keep aside personal values to conduct objective observations.

SD D U A SA

D. Observations will be the same, because observations are exactly what we see and nothing more. Facts are facts. Interpretations may be different from one person to another, but observations should be the same.

SD D U A SA

E. Observations will be the same. Although subjectivity cannot be completely avoided in observation, scientists use different methods to verify the results and improve objectivity.

SD D U A SA

6. Most scientists follow the universal scientific method, step-by-step, to do their research (i.e., state a hypothesis, design an experiment, collect data, and draw conclusions).

A. The scientific method ensures valid, clear, logical and accurate results. Thus, most scientists follow the universal method in research.

SD D U A SA

B. Most scientists use the scientific method because it is a logical procedure. SD D
U A SA

C. The scientific method is useful in most instances, but it does not ensure results; therefore, scientists invent new methods.

SD D U A SA

D. There is no so-called the scientific method. Scientists use any methods to obtain results. SD D
U A SA

E. There is no fixed scientific method; scientific knowledge could be accidentally discovered.

SD

D U A SA

F. No matter how the results are obtained, scientists use the scientific method to verify it. SD D
U A SA

7. Is scientific theory (e.g. atomic theory) “discovered” or “invented” by scientists from the natural world?

A. Discovered, because the idea was there all the time to be uncovered. SD

D U A SA

B. Discovered, because it is based on experimental facts. SD D
U A SA

C. Some scientists discover a theory accidentally, but other scientists may invent a theory from their known facts.

SD D U A SA

D. Invented, because a theory is an interpretation of experimental facts, and experimental facts are discovered by scientists.

SD D U A SA

E. Invented, because a theory is created or worked out by scientists. SD D

U A SA

F. Invented, because a theory can be disproved.

SD D U A SA

8. Is scientific law (e.g., gravitational law) “discovered” or “invented” by scientists from the natural world?

A. Discovered, because scientific laws are out there in nature, and scientists just have to find them.

SD

D U A SA

B. Discovered, because scientific laws are based on experimental facts. SD D
U A SA

C. Some scientists discover a law accidentally, but other scientists may invent a law from their known facts.

SD D U A SA

D. Invented, because scientists invent scientific laws to interpret discovered experimental facts.

SD

D U A SA

E. Invented, since there are no absolutes in nature, therefore, the law is invented by scientists.

SD

D U A SA

9. In comparison to laws, theories have less evidence to support them.

A. Yes, theories are not as definite as laws.

SD D

U A SA

B. Yes, if a theory stands up to many tests it will eventually become a law; therefore a law has more supporting evidence.

SD D U A SA

C. Not quite, some theories have more supporting evidence than some laws.

SD D

U A SA

D. No, theories and laws are different types of ideas. They cannot be compared.

SD D

U A SA

Section-A (II)

Instruction to participants:

Read the statement carefully-Think to yourself whether you agree or disagree with the statement or can't make up your mind. Then read the list of different positions on the topic. Pick the one that comes closest to your own positions.

Every statement ends with the same three positions. Here is how you can use them if you wish:

X. "I don't understand." This choice is included in case there is a key word or phrase that you just don't understand.

Y. "I don't know enough about this subject to make a choice".

Z. "None of these choices fit my basic viewpoint." This choice can be used when none of the other positions comes close to your own belief, or when you want to combine two or more choices into one position.

There are no "right" answers; this is not a test. We simply want to understand what your view or position is on a number of issues about science and how scientists work. Thank You.

1. Many scientific models used in research laboratories (such as the model of heat, the neuron, DNA, or the atom) are copies of reality.

Your position basically is :(**Please read from A to J, and then choose one.**)

Scientific models ARE copies of reality:

A. Because scientists say they are true, so they must be true.

B. Because much scientific evidence has proven them true.

C. Because they are true to life. Their purpose is to show us reality or teach us something about it.

D. Scientific models come close to being copies of reality, because they are based on scientific observations and research.

Scientific models are NOT copies of reality:

E. Because they are simply helpful for learning and explaining, within their limitations.

F. Because they change with time and with the state of our knowledge, like theories do.

- G. Because these models must be ideas or educated guesses, since you can't actually see the real thing.
- H. I don't understand.
- I. I don't know enough about this subject to make a choice.
- J. None of these choices fits my basic viewpoint.

2. When scientists classify something (for example, a plant according to its species, an element according to the periodic table, energy according to its source, or a star according to its size), scientists are classifying nature according to the way nature really is; any other way would simply be wrong.

Your position basically is. **(Please read from A to I, and then choose one.)**

- A. Classifications match the way nature really is, since scientists have proven them over many years of work.
- B. Classifications match the way nature really is, since scientists use observable characteristics when they classify.
- C. Scientists classify nature in the most simple and logical way, but their way isn't necessarily the only way.
- D. There are many ways to classify nature, but agreeing on one universal system allows scientists to avoid confusion in their work.
- E. There could be other correct ways to classify nature, because science is liable to change and new discoveries may lead to different classifications.
- F. Nobody knows the way nature really is. Scientists classify nature according to their perceptions or theories. Science is never exact, and nature is so diverse. Thus, scientists could correctly use more than one classification scheme.
- G. I don't understand.
- H. I don't know enough about this subject to make a choice.
- I. None of these choices fits my basic viewpoint.

3. When developing new theories or laws, scientists need to make certain assumptions about nature (for example, matter is made up of atoms). These assumptions must be true in order for science to progress properly.

Your position basically is: **(Please read from A to I, and then choose one.)**

Assumptions MUST be true in order for science to progress:

- A. Because correct assumptions are needed for correct theories and laws. Otherwise scientists would waste a lot of time and effort using wrong theories and laws.
- B. Otherwise society would have serious problems, such as inadequate technology and dangerous chemicals.
- C. Because scientists do research to prove their assumptions true before going on with their work.
- D. It depends. Sometimes science needs true assumptions in order to progress. But sometimes history has shown that great discoveries have been made by disproving a theory and learning from its false assumptions.

- E. It doesn't matter. Scientists have to make assumptions, true or not, in order to get started on a project.
- F. Scientists do not make assumptions. They research an idea to find out if the idea is true. They don't assume it is true.
- G. I don't understand.
- H. I don't know enough about this subject to make a choice.
- I. None of these choices fits my basic viewpoint.

4. When scientists investigate, it is said that they follow the scientific method. The scientific method is:

Your position basically is: **(Please read from A to M, and then choose one.)**

- A. The lab procedures or techniques; often written in a book or journal, and usually by a scientist.
- B. Recording your results carefully.
- C. Controlling experimental variables carefully, leaving no room for interpretation.
- D. Getting facts, theories or hypotheses efficiently.
- E. Testing and retesting proving something true or false in a valid way.
- F. Postulating a theory then creating an experiment to prove it.
- G. Questioning, hypothesizing, collecting data and concluding.
- H. A logical and widely accepted approach to problem solving.
- I. An attitude that guides scientists in their work.
- J. Considering what scientists actually do, there really is no such thing as the scientific method.
- K. I don't understand.
- L. I don't know enough about this subject to make a choice.
- M. None of these choices fits my basic viewpoint.

5. The best scientists are those who follow the steps of the scientific method.

Your position basically is: **(Please read from A to H, and then choose one.)**

- A. The scientific method ensures valid, clear, logical and accurate results. Thus, most scientists will follow the steps of the scientific method.
- B. The scientific method should work well for most scientists; based on what we learned in school.
- C. The scientific method is useful in many instances, but it does not ensure results. Thus, the best scientists will also use originality and creativity.
- D. The best scientists are those who use any method that might get favorable results (including the method of imagination and creativity).
- E. Many scientific discoveries were made by accident, and not by sticking to the scientific method.
- F. I don't understand.
- G. I don't know enough about this subject to make a choice.
- H. None of these choices fits my basic viewpoint.

6. Scientific discoveries occur as a result of a series of investigations, each one building on an earlier one, and each one leading logically to the next one, until the discovery is made.

Your position basically is: **(Please read from A to J, and then choose one.)**

Scientific discoveries result from a logical series of investigations:

- A. Because experiments (for example, the experiments that led to the model of the atom, or discoveries about cancer) are like laying bricks onto a wall.
- B. Because research begins by checking the results of an earlier experiment to see if it is true. A new experiment will be checked by the people who come afterwards.
- C. Usually scientific discoveries result from a logical series of investigations. But science is not completely logical. There is an element of trial and error, hit and miss, in the process.
- D. Some scientific discoveries are accidental, or they are the unpredicted product of the actual intention of the scientist. However, more discoveries result from a series of investigations building logically one upon the other.
- E. Most scientific discoveries are accidental, or they are the unpredicted product of the actual intention of the scientist. Some discoveries result from a series of investigations building logically one upon the other.

Scientific discoveries do not occur as a result of a logical series of investigations:

- F. Because discoveries often result from the piecing together of previously unrelated bits of information.
- G. Because discoveries occur as a result of a wide variety of studies which originally had nothing to do with each other, but which turned out to relate to each other in unpredictable ways.
- H. I don't understand.
- I. I don't know enough about this subject to make a choice.
- J. None of these choices fits my basic viewpoint.

7. Scientists publish the results of their work in scientific journals. When scientists write an article for a journal, they organize their report in a very logical orderly way. However, scientists actually do the work in a much less logical way.

Your position basically is : **(Please read from A to J, and then choose one.)**

Articles are written in a more logical way than the actual work:

- A. Because scientists can think and work without following a set plan. Consequently, if you read the actual order of their thoughts and procedures, it would be confusing. Therefore, scientists write logically so other scientists will understand the results.
- B. Because scientific hypotheses are personal views or guesses and thus are not logical. Scientists, therefore, write logically so other scientists will understand the results.
- C. Scientists usually don't want to give away "the recipe" but they do want to tell the world about their results. So they write it up logically but in a way that does not reveal how it was actually done.
- D. It depends. Sometimes scientific discoveries happen by accident. But other times discoveries happen in a logical orderly way, just like the articles are written.

Articles are written in a logical way showing how the actual work was done:

- E. Because a scientist's work is conducted logically; otherwise, it would not be useful to science and technology.

- F. Because scientists do work in a logical way so that their published report will be easier to write in a logical way.
- G. Articles are not necessarily written in a logical way. They're written the way the work was done. This can be complicated or straightforward.
- H. I don't understand.
- I. I don't know enough about this subject to make a choice.
- J. None of these choices fits my basic viewpoint.

8. Scientists should NOT make errors in their work because these errors slow the advance of science.

Your position basically is :(**Please read from A to H, and then choose one.**)

- A. Errors slow the advance of science. Misleading information can lead to false conclusions. If scientists don't immediately correct the errors in their results, then science is not advancing.
- B. Errors slow the advance of science. New technology and equipment reduce errors by improving accuracy and so science will advance faster.

Errors CANNOT be avoided:

- C. So scientists reduce errors by checking each others' results until agreement is reached.
- D. Some errors can slow the advance of science, but other errors can lead to a new discovery or breakthrough. If scientists learn from their errors and correct them, science will advance.
- E. Errors most often help the advance of science. Science advances by detecting and correcting the errors of the past.
- F. I don't understand.
- G. I don't know enough about this subject to make a choice.
- H. None of these choices fits my basic viewpoint.

9. Even when making predictions based on accurate knowledge, scientists can tell us only what probably might happen. They cannot tell what will happen for certain.

Your position basically is: (**Please read from A to H, and then choose one.**)

Predictions are NEVER certain:

- A. Because there is always room for error and unforeseen events which will affect a result. No one can predict the future for certain.
- B. Because accurate knowledge changes as new discoveries are made, and therefore predictions will always change.
- C. Because a prediction is not a statement of fact. It is an educated guess.
- D. Because scientists never have all the facts. Some data are always missing.
- E. It depends. Predictions are certain, only as long as there is accurate knowledge and enough information.
- F. I don't understand.
- G. I don't know enough about this subject to make a choice.
- H. None of these choices fits my basic viewpoint.

10. Scientists in different fields look at the same thing from very different points of view (for example, H^+ causes chemists to think of acidity and physicists to think of protons). This makes it difficult for scientists in different fields to understand each others' work.

Your position basically is: **(Please read from A to H, and then choose one.)**

It is difficult for scientists in different fields to understand each other:

- A. Because scientific ideas depend on the scientist's viewpoint or on what the scientist is used to.
- B. Because scientists must make an effort to understand the language of other fields which overlap with their own field. It is fairly easy for scientists in different fields to understand each other:
- C. Because scientists are intelligent and so they can find ways to learn the different languages and points of view of another field.
- D. Because they have likely studied the various fields at one time.
- E. Because scientific ideas overlap from field to field. Facts are facts no matter what the scientific field is.
- F. I don't understand.
- G. I don't know enough about this topic to make a choice.
- H. None of these choices fits my basic viewpoint

Section-B

Guidelines for students: There are statements given about the structure of the atom. Please go through these statements carefully and give your response. Remember, this is to know about your views and understanding; **it is not a test.**

Do you agree or disagree with the following statements (1-3, 5-6). Explain giving reasons why you think so.

1. Atoms are solid particles.

2. Scientists now have a complete understanding of the atom.

3. Atoms are composed mainly of empty space.

4. You have heard about different scientists giving a model of the atomic structure. What do you think a model is?

5. Since scientists cannot actually see inside the atom, it is not possible to believe any model of the atom that has been given.

6. Early models of the atom were useless theories, which were eventually discarded.

7. What does an atom look like? Describe in your words.

The atoms looks like_____

8. How do you know that atoms look like what you have described?

Because, _____

9. Why do you need to study different models of atomic structure?

10. Which model according to you can best explain the structure of the atom? Why do you think so?

Annexure 4

Responses of Students (College and School) on the Nature of Science Questionnaire: VOSE
Represented in a tabular form separately for Both the Participating Groups

Item no. 1

1. When two different theories arise to explain the same phenomenon (e.g. the nature of electron), scientists will accept the two theories at the same time.					
A. Yes, because scientists still cannot objectively tell which one is better; therefore, they will accept both tentatively.	SD (5)	D (9)	U (4)	A (22)	SA (5)
	C S 1 4	C S 4 5	C S 2 2	C S 6 16	C S 2 3
B. Yes, because the two theories may provide explanations from different perspectives, there is no right or wrong.	SD (4)	D (4)	U (3)	A (31)	SA (3)
	C S 1 3	C S 1 3	C S 1 2	C S 10 21	C S 1 2
C. No, because scientists tend to accept the theory they are more familiar with.	SD (19)	D (12)	U (8)	A (4)	SA (2)
	C S 6 13	C S 4 8	C S 1 7	C S 2 2	C S 2 0
D. No, because scientists tend to accept the simpler theories and avoid complex theories	SD (21)	D (13)	U (5)	A (4)	SA (2)
	C S 10 11	C S 1 12	C S 2 3	C S 2 2	C S 0 2
E. No, the academic status of the person who proposed the theory will influence other scientists' acceptance of the theory.	SD (19)	D (15)	U (5)	A (4)	SA (2)
	C S 4 15	C S 6 9	C S 2 3	C S 2 2	C S 1 1
F. No, scientists tend to accept new theories which deviate less from the current core scientific theory.	SD (5)	D (13)	U (11)	A (12)	SA (4)
	C S 2 3	C S 2 11	C S 4 7	C S 2 10	C S 4 0
G. No, scientists use intuition to make judgments.	SD (6)	D (14)	U (11)	A (9)	SA (5)
	C S 2 4	C S 4 10	C S 3 8	C S 2 7	C S 4 1
H. No, because there is only one truth, scientists will not accept any theory before distinguishing which is best.	SD (7)	D (7)	U (4)	A (14)	SA (13)
	C S 3 4	C S 2 5	C S 2 2	C S 4 10	C S 4 9

Item no. 2

2. While scientists are conducting scientific research, they also use their imagination.					
A. Yes, imagination is the main source of innovation.	SD (0)	D (1)	U (3)	S (16)	SA (25)
	C S 0 0	C S 0 1	C S 3 0	C S 8 8	C S 4 21

B. Yes, scientists use their imagination more or less in scientific research.	SD (2) C S 0 2	D (4) C S 0 4	U (11) C S 3 8	S (13) C S 1 12	SA (6) C S 2 4
C. No, imagination is not consistent with the logical principles of science.	SD (2) C S 1 1	D (13) C S 2 11	U (12) C S 3 9	S (6) C S 0 6	SA (3) C S 0 3
D. No, imagination may become a means for a scientist to prove his point at all costs.	SD (13) C S 10 3	D (11) C S 1 10	U (6) C S 2 4	S (15) C S 2 13	SA (0) C S 0 0
E. No, imagination lacks reliability.	SD (8) C S 6 2	D (7) C S 1 6	U (12) C S 6 6	S (16) C S 2 14	SA (2) C S 0 2

Item no. 3

3. Even if the scientific investigations are carried out correctly, the theory proposed can still be disproved in the future.					
A. Scientific research will face revolutionary change, and the old theory will be replaced.	SD (3) C S 1 2	D (14) C S 10 4	U (7) C S 6 1	S (17) C S 2 15	SA (4) C S 2 2
B. Scientific advances cannot be made in a short time. It is through a cumulative process; therefore, the old theory is preserved	SD (4) C S 2 2	D (4) C S 1 3	U (12) C S 4 8	S (19) C S 2 17	SA (6) C S 2 4
C. With the accumulation of research data and information, the theory will evolve more accurately and completely, not being disproved.	SD (2) C S 1 1	D (4) C S 2 2	U (5) C S 2 3	S (25) C S 8 17	SA (9) C S 2 7

Item no. 4

4. Scientific investigations are influenced by socio-cultural beliefs and values present at a given time (e.g. religious beliefs, social bias about gender, caste, class etc.).					
A. Yes, socio-cultural values influence the direction and topics of scientific investigations.	SD (14) C S 4 10	D (15) C S 9 6	U (4) C S 2 2	S (4) C S 2 2	SA (8) C S 1 7
B. Yes, because scientists participating in scientific investigations are influenced by socio-cultural values.	SD (11) C S 6 5	D (18) C S 4 14	U (8) C S 2 6	S (5) C S 2 3	SA (3) C S 1 2
C. No, scientists with good training will remain value free (neutral) when carrying out research.	SD (2) C S 1 1	D (6) C S 2 4	U (6) C S 2 4	S (15) C S 4 11	SA (16) C S 6 10
D. No, because science requires objectivity, which is contrary to the subjective socio-cultural values.	SD (1) C S 1 0	D (4) C S 2 2	U (14) C S 8 6	S (22) C S 4 18	SA (4) C S 0 4

Item no. 5

5. Scientists' observations are influenced by their personal beliefs (e.g. personal experiences, presumptions) therefore; they may not make the same observations for the same experiment.						
A. Observations will be different, because different beliefs lead to different expectations influencing the observation	SD	(4)	D	(11)	U	(4)
	A					
	(23)					
B. Observations will be the same, because the scientists trained in the same field hold similar ideas.	SD	(5)	D	(23)	U	(8)
	A					
	(8)					
C. Observations will be the same, because through scientific training scientists can keep aside personal values to conduct objective observations	SD	(4)	D	(12)	U	(12)
	A					
	(12)					
D. Observations will be the same, because observations are exactly what we see and nothing more. Facts are facts. Interpretations may be different from one person to another, but observations should be the same.	SD	(3)	D	(4)	U	(15)
	A					
	(19)					
E. Observations will be the same. Although subjectivity cannot be completely avoided in observation, scientists use different methods to verify the results and improve objectivity.	SD	(2)	D	(4)	U	(5)
	A					
	(23)					

Item no. 6

6. Most scientists follow the universal scientific method, step by step, to do their research (i.e., state a hypothesis, design an experiment, collect data, and draw conclusions).						
A. The scientific method ensures valid, clear, logical and accurate results. Thus, most scientists follow the universal method in research.	SD	(1)	D	(4)	U	(4)
	A					
	(26)					
B. Most scientists use the scientific method because it is a logical procedure.	SD	(0)	D	(1)	U	(4)
	A					
	(32)					
C. The scientific method is useful in most instances, but it does not ensure results; therefore, scientists invent new methods.	SD	(1)	D	(8)	U	(15)
	A					
	(16)					

D. There is no so-called the scientific method. Scientists use any methods to obtain results.	SD (5) C S 1 4	D (12) C S 8 4	U (8) C S 2 6	A (15) C S 2 13	SA (5) C S 2 3
E. There is no fixed scientific method; scientific knowledge could be accidentally discovered.	SD (0) C S 0 0	D (6) C S 6 0	U (3) C S 2 1	A (29) C S 6 23	SA (7) C S 1 6
F. No matter how the results obtained, scientists use the scientific method to verify it.	SD (3) C S 2 1	D (10) C S 4 6	U (9) C S 3 6	A (17) C S 2 15	SA (4) C S 2 2

Item no. 7

7. Is scientific theory (e.g. atomic theory) “discovered” or “invented” by scientists from the natural world?					
A. Discovered, because the idea was there all the time to be uncovered.	SD (3) C S 2 1	D (13) C S 4 9	U (7) C S 3 4	A (20) C S 4 16	SA (2) C S 2 0
B. Discovered, because it is based on experimental facts.	SD (3) C S 3 0	D (8) C S 2 6	U (5) C S 2 3	A (26) C S 6 20	SA (2) C S 1 1
C. Some scientists discover a theory accidentally, but other scientists may invent a theory from their known facts.	SD (2) C S 1 1	D (3) C S 2 1	U (4) C S 2 2	A (28) C S 8 20	SA (6) C S 2 4
D. Invented, because a theory is an interpretation of experimental facts, and experimental facts are discovered by scientists.	SD (2) C S 2 0	D (9) C S 2 7	U (8) C S 5 3	A (18) C S 2 16	SA (5) C S 4 1
E. Invented, because a theory is created or worked out by scientists.	SD (3) C S 1 2	D (8) C S 2 6	U (16) C S 6 10	A (17) C S 4 13	SA (4) C S 2 2
F. Invented, because a theory can be disproved.	SD (1) C S 1 0	D (5) C S 4 1	U (15) C S 6 9	A (15) C S 2 13	SA (9) C S 2 7

Item no. 8

8. Is scientific law (e.g. gravitational law) “discovered” or “invented” by scientists from the natural world?					
A. Discovered, because scientific laws are out there in nature, and scientists just have to find them.	SD (1) C S 1 0	D (6) C S 2 4	U (5) C S 2 3	A (20) C S 6 14	SA (13) C S 4 9
B. Discovered, because scientific laws are based on experimental facts.	SD (1) C S 0 1	D (6) C S 1 4	U (5) C S 2 3	A (29) C S 10 19	SA (4) C S 2 2

C. Some scientists discover a law accidentally, but other scientists may invent a law from their known facts.	SD (1) C S 1 0	D (5) C S 4 1	U (15) C S 2 13	A (18) C S 6 12	SA (6) C S 2 4
D. Invented, because scientists invent scientific laws to interpret discovered experimental facts.	SD (0) C S 0 0	D (17) C S 2 15	U (8) C S 3 5	A (18) C S 10 8	SA (2) C S 0 2
E. Invented, since there are no absolutes in nature, therefore, the law is invented by scientists.	SD (2) C S 0 2	D (24) C S 6 18	U (6) C S 4 2	A (9) C S 2 7	SA (4) C S 3 1

Item no. 9

9. In comparison to laws, theories have less evidence to support them.						
A. Yes, theories are not as definite as laws.	SD (3) C S 2 1	D (9) C S 4	U (10) C S 6	A (20) C S 2	SA (3) C S 1	
B. Yes, if a theory stands up to many tests it will eventually become a law; therefore a law has more supporting evidence.	SD (2) C S 2 0	D (3) C S 2 1	U (3) C S 2 1	A (24) C S 4 20	SA (13) C S 5 8	
C. Not quite, some theories have more supporting evidence than some laws.	SD (0) C S 0 0	D (11) C S 4 7	U (11) C S 6 5	A (19) C S 2 17	SA (4) C S 3 1	
D. No, theories and laws are different types of ideas. They cannot be compared	SD (4) C S 0 4	D (15) C S 6 9	U (14) C S 6 8	A (5) C S 2 3	SA (4) C S 1 3	



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RESEARCH ARTICLE

SUSTAINABLE MANAGEMENT OF FOREST ECOSYSTEMS IN E. AFRICA FOR IMPROVED LIVELIHOODS AND ENVIRONMENTAL RESILIENCE.

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Abstract

Forest ecosystems play a pivotal role in socio-economic development and ecological stability. Critical role played by forests ecosystem cannot be over-emphasized noting the current commitment by UN Member States under COP 21 of 2015 on climate change adaptation and mitigation. Forests provide a pool of genetic resources, timber, fruits, medicine, fodder, regulating services such as flood and climate regulation; cultural services, aesthetic, recreational values; and supporting services including primary production, carbon sequestration, nutrient cycling, soil formation and biodiversity conservation. They are critical in provision of renewable energy that majority of African households still depend on. Trees are important in integrated soil, water and biodiversity conservation and in mitigating climate change and variability. Forestry and agroforestry management principles are inherently geared towards harnessing the benefits of natural resources and biodiversity for livelihoods and sustainable productive environment. The study presents results of the existing challenges and opportunities facing forests ecosystem management, agricultural production and biodiversity conservation owing to human pressure on land-use and natural resources in E. Africa. It is concentrated on the analysis of three major pathways for sustainable forests ecosystem: (i) reducing pressure on natural forests; (ii) providing habitat for native plant and animal species; and (iii) forests and agroforestry systems for integrated land use management and fragmented landscapes. The review proposes a paradigm shift to accelerate the current low investment in these resources while promoting strategies that facilitate the mainstreaming of forestry and agroforestry management, including biodiversity conservation in the sub-region's development agenda. It identified the following action plans to be fast-tracked in the sub-region: Design, develop and disseminate improved technologies and innovations, management plans for sustainable and equitable management of forests and agroforestry resources; Enhance productivity and conservation of agro-ecosystems using trees and agroforestry practices /technologies for socio-economic benefits and environmental services; Facilitate the development of policy options and institutional innovations for enhanced investment in forestry and agroforestry and allied natural resources based enterprises; Strengthen the capacity and governance framework for effective and sustainable

management of forestry and agroforestry resources; and Promote awareness and access to knowledge and information for improved and adaptive management and innovations, including utilization.

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..... **Introduction:-**

The General Assembly of the United Nations adopted the definition of Sustainable Forest Management in December 2007 as a dynamic and evolving concept aims to maintain and enhance the economic, social and environmental value of all types of forests, for the benefit of present and future generations. It is characterized by seven elements, including: (i) extent of forest resources; (ii) forest biological diversity; (iii) forest health and vitality; (iv) productive functions of forest resources; (v) protective functions of forest resources; (vi) socio-economic functions of forests; and (vii) legal, policy and institutional framework (International Union for Conservation of Nature, 2012).

Developments in forest management over the past decades have focused on progress towards Sustainable Forest Management (SFM), an approach that balances environmental, socio-cultural and economic objectives of management in line with the "Forest Principles" adopted at the United Nations Conference on Environment and Development (UNCED) in 1992.

Parallel efforts in environmental conservation, particularly within the framework of the Conservation on Biodiversity, have led to the development of the Ecosystem Approach (EA) as a framework and holistic approach for the conservation and sustainable use of biological diversity and its components in all types of ecosystems (Metteet *al.*, 2003).

Africa as a continent is rich in natural resources. Its land, water, and forests underpin the sustained productivity of food crops and livestock on which millions of Africans depend directly for their livelihood and survival (Global Environment Facility, 2011). These resources are major assets on which most countries depend for economic growth and sustainable development (GEF, 2011).

Undoubtedly, the risk of expanding land degradation and desertification, increased deforestation, and water scarcity threatens to hasten a major global environmental crisis. In Kenya, it is not an entirely surprising statistic that about 12 % of the land areas which was originally covered by closed canopy forests has been reduced to a mere 1.7% of its original size. This is attributed to population pressure for settlements, infra-structure, demand for wood products and conversion to agriculture Government of Kenya (GOK, 2013).

Forests harbour two thirds of all terrestrial animal and plant species (IUCN, 2012). They provide food, oxygen, shelter, recreation, and spiritual sustenance. They are the source for over 5,000 commercially-traded products, ranging from pharmaceuticals to timber and clothing. The biodiversity of forests, variety of genes, species, and forest ecosystems underpins these goods and services, and is the basis for long-term forest health and stability. The Convention on Biological Diversity (CBD) addresses the conservation and sustainable use of forest biodiversity through a comprehensive programme of work, adopted in 2002 and revised in 2008. Many of the activities of the programme of work promote poverty reduction and creation of sustainable livelihoods (IUCN, 2012).

Despite the major roles played by forests such as provision of goods and ecological services, waste assimilation and aestheticism, it is threatened by habitat modification and fragmentation, a reduction in vegetation cover and species diversity, and over-exploitation and competition from invasive species, mainly as a consequence of human population growth. The high level of deforestation is attributed to the need for woodfuel for home consumption but more so for commercial purposes where the charcoal is sold to urban dwellers in major towns. Further, there is limited information and knowledge on appropriate management regimes of the forests and also non-availability of suitable germplasm for propagation of biological species (GEF, 2011). This scenario is compounded by limited focus and priority on the forests by implementing agencies both in the government and NGO sector. Community participation is also limited in the management and utilization of such resources.

The review involved identification and assembling of relevant documents; assessment of the documents using review checklist developed in line with the emphasis on the sustainable forest management and focus areas. Past GIS database was also reviewed and key sites of major environmental significance identified.

Challenges facing forest ecosystem:-

It is widely acknowledged that forests are faced with a myriad of political, socio-economic, and environmental challenges with profound implications for their sustainable management and development. Forests and associated biodiversity, which underpins the livelihoods of the majority of the rural poor, is being destroyed irreversibly by human activities. The situation is exacerbated by climate variability and change, whose effects are already being witnessed in the region in the form of variable water supplies, lengths of growing season, extreme weather events (floods, droughts), and changing habitats. How to deal with these trends and challenges to sustainably manage these resources so as to conserve, utilize and improve livelihoods and the environment in East African forest ecosystems remains a great question.

The review findings reveal that degradation and loss of forests and associated habitats and the species that they support is a result of a wide range of natural and man-made causes interacting at different levels and intensities on the forest ecosystems. The Millennium Ecosystem Assessment (MEA) report (2005) for instance asserted that an estimated 70% of natural habitats in the Mara River Basin have been converted over time to farmland and other forms of settlement. Table (1) shows the areas in Kenya and rate of change in forest cover from 1990 to 2010

Table 1:- Areas of Forest in Kenya and Rate of Change since 1990.

Category of forest resource (using FAO definitions)	Area ('000 Ha)				Annual change 1990 to 2010 ('000 Ha)
	1990	2000	2005	2010	
Indigenous closed Canopy Forest	1240	1190	1,165	1,140	-5
Indigenous Mangroves	80	80	80	80	0
Open woodlands	2150	2100	2075	2,050	-5
Open woodlands	170	134	119	107	-3.15
Private Plantation forests	68	78	83	90	+1.1
<i>Sub-total Forest land (total of above categories)</i>	3708	3582	2357	3, 467	-12.05
Bush-land	24800	24635	24570	24, 510	-14.5
Farms with Trees	9420	10020	10320	10, 385	+48.25
Total Area of Kenya	58037	58037	58037	58, 037	0

Source: (GoK, 2013).

Over the past three decades, large areas of forest reserves have been officially “de-gazetted” and unofficially converted to other uses, mainly agriculture, and the remaining protected indigenous forests managed by for example Kenya Forest Service (KFS) in Kenya. The review established that logging, both legal and illegal, of valuable timber trees results in reduced carbon stocks and degraded biodiversity values. Forests on community trust lands under the control of local authorities continue to be degraded and destroyed through over-exploitation for timber, poles, charcoal and fuel wood, and through unregulated grazing and clearance for agriculture; depicting what the previous studies termed as “tragedy of the commons”.

Matiet *et al.*, 2005 asserted that significant changes on land cover have taken place. According to their study, it is clear that forests, shrub land, grassland, and savannah reduced by 32%, 34%, 45%, and 26% respectively as shown in table 2. Despite efforts made to protect forest biodiversity in the region pressure on forest resources is still on the rise due to population increase and hence need for more land for settlement and farming.

Table 2:- Extent of land use/cover changes in the Mara river basin between 1973 and 2000

Land cover type	1973 (km ²)	1986 (km ²)	2000 (km ²)	Change	
				Km ²	%
Forest	1008	893	689	-319	-32
Tea/ open forest	621	1073	1948	+1327	+214
Agricultural land	826	1617	2504	+1678	+203
Shrubland	5361	5105	3546	-1815	-34
Grassland	2465	1621	1345	-1120	-45
Savannah	3163	2867	2354	-809	-26
Wetlands	286	604	1394	+1109	+387
Water bodies	104	54	55	-49	-47

Source: Mutiet *et al.* 2005

Key challenges to forest ecosystems:-

Expanding agriculture: The review established general soil fertility of forest ecosystems which has attracted settled agriculture, including commercial food and horticultural plantations. It is noted that the soils in upland and lowland Riverine forests patches are more fertile than those of surrounding farmlands and woodlands and hence face pressure to be converted for agriculture.

Charcoal production and firewood harvesting: Although not well quantified, the business of charcoal production has heavily impacted the forests ecosystems. Away from towns and roads this threat is much less important as local people use firewood for cooking. Most firewood is collected from trees and bushes outside the protected forest areas.

Uncontrolled fires: Widespread pastoralism for example in Mara River Basin forests ecosystem particularly with Maasai community in both Kenya and Tanzania threaten woodland and shrub land forest vegetation of being cleared for pastures. Although forest fires are a natural phenomenon, majority that occur are started by pastoralists and people who use the fire to clear farmland, drive animals for hunting, collect honey, and remove tsetse flies from an area. Some other fires are started accidentally, for example from cigarettes thrown from passing vehicles or by pedestrians. These occasional fires destroy forest and other biodiversity. Over time and with frequent and intense burning it is believed that lowland forests ecosystems and thicket vegetation is converted to more fire-adapted vegetation types. This results in a loss of the narrowly endemic forests ecosystem species and their replacement by wide-ranging invasive species.

Illegal and unsustainable logging: The commercial logging of forests ecosystem tree species occurs mainly for example in Mau Forest, Kenya and in the middle and lower Riverine forests of the Mara river Basin (MRB) forests ecosystem. Logging using pit-sawing techniques occurs in forests ecosystem where timber trees remain. Many forests have already been logged to exhaustion, particularly heavy exploitation for round wood. Logging of the valuable trees is often the first major disturbance to a forest, which then progresses to fire wood collection and charcoal burning, and in the worse cases to clearance for agricultural use.

Unplanned settlement: Over much of forests ecosystem the expansion of settlements is only partly planned. For example, in the Mau forests, illegal settlers have interfered with the catchment vegetation which is the main sources of water to the Mara River. In some of the protected forests ecosystem of Kenya and Tanzania (both Forest Reserves and traditionally protected forest areas), settlements have been established within the boundaries of the reserve. When this happens, farming activities also start and there can be much damage to the habitats. Similarly, the expansion of tourism facilities into some forests ecosystem is also an issue for concern.

Minor threats: Some threats, such as invasive species, are considered to be relatively minor. However these threats need to be carefully monitored to ensure that action is taken so that they do not become more significant.

Causes of Forest Biodiversity loss:-

Limited alternative economic activities: A significant proportion of the population in Kenya and Tanzania, particularly the communities relying on agriculture and pastoralism for their livelihoods. Direct threats from agricultural expansion and associated fires, in particular, are linked to poverty and limited alternative economic activities in rural areas. As subsistence agriculture is the primary livelihood strategy for most people in rural areas, the need for land puts negative pressure on forests, even though many forest products are used and valued by local people. A high level of direct reliance on forest products to sustain livelihoods contributes to their overuse in some areas. The lack of affordable energy alternatives also drives the urban demand for charcoal.

Demographic changes: Demographic changes combined with uncontrolled settlement increases overall direct pressures on land, timber and non-timber forest resources. Demand for additional farmland for subsistence agriculture also rises. Population increases are also linked to habitat degradation associated with increased demand for firewood, charcoal, timber, building materials and bush meat.

Declining respect for traditional forest protection systems: Small areas of forests have traditionally been conserved as sacred sites. These areas have been protected by community elders but are now threatened and some cases being destroyed by recent migrants who do not necessarily owe allegiance to traditional authorities. A decline

in the respect for elders among other members of the local population is causing further pressures to convert these forests to farmland.

Limited institutional capacities and financial support: While decentralization efforts are focusing more attention and responsibilities at local levels, the local natural resource management agencies are often understaffed, poorly funded, and have limited capacity to adopt new management approaches such as 'participatory forest management'. Financial support for capacity building and management activities, including management of protected areas, and enforcement of regulations on commercial resource extraction, is also limited.

Limited Consideration of Environmental Impacts of Economic Development Policies: National and Local Governments tend to give priority to economic development activities and will permit the clearance of natural habitats to establish mining, tourism facilities, roads, and agricultural projects. Given the poverty levels in Kenya and Tanzania, such a development focus is understandable. However, there remains considerable scope for undertaking development that is less environmentally damaging through enhanced integration of conservation objectives in development planning and consideration of environmental impacts.

Inadequate Capacity to Enforce Regulations on Commercial Extraction: Forest ecosystems suffers inadequate capacity to enforce forest rules and regulations, such as those on charcoal and timber production. For instance, the MRB Ecoregion Illegal activities are both depleting natural resources and limiting the economic value of these activities that is captured by the national economies.

Legal Issues and Land Tenure: The existence of different land tenure systems in East Africa contributes to loss of forest biodiversity. In Kenya for example, the current land tenure system leaves a large percentage of rural population without secure title, especially those on the Trust Lands that make up a majority of the land in the country. This limits incentives for people's long-term stewardship of these lands. In Tanzania, recent changes in the Land Law (1999) mean that it is now possible for villages to gain title deeds for their village lands, and also for individuals to obtain deeds for land. This is a change from the past system where land was controlled by the government, and may offer better possibilities for the management of forest resources at the village level.

Inadequate Institutional Coordination and Integration of Sectoral Policies: At the national level, conflicting objectives and policies among government ministries and departments have also contributed to the loss of habitats across the region. In both Tanzania and Kenya there are over 20 sectoral policies that either directly or indirectly affect forest management and conservation practices, with no clear mechanism for ensuring linkages between these different policies and Acts. However, the governments of Kenya and Tanzania have started putting mechanisms in place to address this issue.

Sustainability Management Issues:-

Despite the abundance and huge potential of the sub-region's forest and tree resources for sustainable development and provision of environmental services, the Partner States have not invested adequately in these resources in pursuit of effective and sustainable management to bring about the required transformation for improvement in livelihoods. To change this trend, there is a urgent need to increase investment in strategies to facilitate the mainstreaming of forestry and agroforestry management, including biodiversity conservation in the sub-region's development agenda. Unprecedented degradation pressure arising from human activities and climate change and variability put forests and agroforestry systems at risk. Principal threats identified include: land use and land cover changes, mainly through conversion of natural forest ecosystems to agricultural land and settlement and loss of flora due to adverse effects of climate change and variability. It is also feared that with increasing population pressure, land clearing and deforestation will continue which is a threat to genetic diversity due to loss of species.

Conclusion:-

The importance accruing from forest ecosystems cannot be over-emphasized. The riparian communities appreciate what forest ecosystems do to them. The only challenge is sustainable management of this natural resource. There is therefore need to collectively strive by partner states to ensure that their utilization, activities and management of the forest resources are geared toward sustainability.

Recommendations for sustainable forest management:-

- Designing, developing and disseminating improved technologies and innovations, management plans for sustainable and equitable management of forests and agroforestry resources;

- Enhancing productivity and conservation of agro-ecosystems using trees and agroforestry practices / technologies for socio-economic benefits and environmental services;
- Facilitating the development of policy options and institutional innovations for enhanced investment in forestry and agroforestry and allied natural resources based enterprises;
- Strengthening the capacity and governance framework for effective and sustainable management of forestry and agroforestry resources; and
- Promoting awareness and access to knowledge and information for improved and adaptive management and innovations, including utilization;

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RESEARCH ARTICLE

ROAD TRAFFIC ACCIDENTS AS A CONSEQUENCE OF “DRIFTING” IN THE KSA.

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Abstract

Car drifting is a serious problem as it may lead to damage of the body, loss of some organs, loss of public and private materials as well suffering borne by the parents of drifters, as they suffer because of the bad reputation of their son actions. Causes of popularity of this phenomenon are the desire to be famous, weak control of parents, media, websites and weakness of religious faith

The purpose of this study was to analyze road traffic accident data from drifters, investigate the lifestyle of drifters, and suggest lowering the risk of accidents caused by drifting. We will also recommend lifestyle modifications in drifting to improve quality of life and may suggest safe practices.

Our main finding in this study was that the majority of drifters are teenager students and they learned it and started it at an early age, with their friends being the most encouraging factor and their family the most discouraging factor. Most of them damaged cars and many knew a friend who was injured or killed by drifting. Camry & Hilux users seemed to have more dangerous behaviours compared to other car users.

Drifting has a negative effect on the socioeconomic and health state of the country. We think it is difficult to stop, but recommend it should be regulated so it is a legal activity in special areas, so to keep drifters busy but in a safe environment. This will save lives and reduce the socioeconomic and health cost burden.

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Introduction:-

Drifting requires skill, and is growing in popularity. There is potential for harm if the driver loses control of the car. While it wasn't up until recently that drifting became as popular as it is today, the history of drifting dates back to many years ago. Drivers have been using this technique as early as the 1930's in Grand Prix races. People didn't call it drifting back then, but throughout the 20th century it started as a racing technique used in motorsports.

Drifting which better known locally as “hajwalah” or “tafheet”, where the car is intentionally spun several times at speed of 180 kph or more, this form of reckless stunt-driving is strongly condemned in the Kingdom of Saudi Arabia, with strict sentences for drivers involved in fatal accidents.

Car drifting is a serious problem which may lead to damage of the body, loss of some organs, and loss of public and private property. It also affects the parents of the persons who drift; they suffer if their child is injured or killed and

also due to the bad reputation of their child's actions. Causes of popularity of this phenomenon are the desire to be famous, weak control of parents, media and websites popularizing this and weakness of religious faith.

A published definition is "Drifting refers to a specific driving technique, as well as a type of motor sport based on that technique. It occurs when the wheels are pointing in a different direction to the direction the car is sliding overall. Drifting requires skill, and is growing in popularity. There is potential for harm if the driver loses control of the car." [1]

In the process, racers often drive dangerously close to traffic, barriers and spectators, who are watching from the sides without any protection. Drifters usually drive and damage local rental vehicles. "Tafheet" practice and events occur with little to no concern for any vehicle occupants, other drivers or any spectator safety.

In videos posted to YouTube, large crowds are seen gathering by the side of the road to watch drivers skid their cars – often typical family sedans like Honda Accords and Hyundai Sonatas – through traffic at high speeds, sometimes heading in the wrong direction. These uploaded video give drivers' kudos within the culture even after vehicle damage, major injury or fatality and naturally after successful high speed skill displays. Tragically it terrorizes other highway users who are unaware of the event until they are in the middle of it and at risk of death or major injury. Several of the clips show out of control cars crashing into spectators causing serious injury, or even death. One particularly graphic video that surfaced in the internet in May 2013 showed a car that rolled through a crowd as several passengers were thrown from vehicle. A middle-aged man in Saudi Arabia was been sentenced to execution by public beheading after killing two people while 'drifting' his car.

Causes of Drifting:-

Drifting is gaining popularity as it spreads all over the internet via social networking sites like MySpace and YouTube. There is a cult following for many drifters, who have earned such titles as 'The Drift King' or 'Drift Master' from their outrageous drifting antics. However, despite all the glory from enthusiasts, drifting is quickly becoming a public safety hazard and a nuisance for Saudi authorities.

"The driving problems are with young people," Ali Abdul-Rahman Al-Mazyad, a Saudi columnist in Riyadh told The Media Line. "There are very little outlets for young people to enjoy themselves and kids basically do what they want."

"There is also not such great education in schools about driving and respecting the road," he said. "Drug use is also a contributing factor. These are the central problems." [2]

During a New York Times investigation, their reported found that between drag races and the "drifting" shown in the videos, the Saudi streets came to resemble a cross between the movies *Death Race* and *The Fast and The Furious*. It seems that a great national boredom, or "tufush," has seized the young men of Saudi, and with no public entertainment and few jobs, an underground car culture has flourished. Wealthier and middle-class men drag race Corvettes and Imprezas all night, but for the poorer, more desperately in need of excitement, only drifting cars through and around traffic will do. And the scene has created one crazy melting pot of young, angry desperation. "The idea behind drifting is, the economy and society don't need you," says Pascal Ménoret, an anthropologist with four years of field work in Riyadh. "They are mostly young Bedouins who recently moved to the city, and whose lives are marked by suffering and self-destructive behavior." [3]

Literature Review

Drifting is gaining popularity as it spreads all over the internet via social networking sites like MySpace and YouTube. There is a cult following for many drifters, who have earned such titles as 'The Drift King' or 'Drift Master' from their outrageous drifting antics. However, despite all the glory from enthusiasts, drifting is quickly becoming a public safety hazard and a nuisance for Saudi authorities.

There have been several drifting accidents which have resulted in fatalities over the years. The most infamous 'Abu Kab' drifting accident occurred in Riyadh in 2005 when a Navy officer decided to try his hand at performing the Arab Drift. He rented a Honda Accord and allowed 4 young boys to ride with him at a speed of 120 kph. The officer crashed and 3 of his passengers were killed. He was found guilty of negligence and sentenced to death by a Jeddah court. [4]

The Saudi government adopted the tougher law this past spring to deter Arab drifting and joyriding. Previously, fatalities would have been classified as “accidental” but now they are classified as “criminal negligence” which carries a much heavier punishment than the prior. Government officials hope that such a serious punishment will deter future Arab drifters from setting foot, or tire for that matter, on Saudi roads with the intention of performing daredevil stunts.

Jeddah's traffic department has recorded a whopping 2,000 cases of motorists running red lights over a two-week period. Lt. Col. Zaid Al-Hamzi, spokesman of the city's traffic department, said 60 percent of the violators were detected by the Saher system and the rest by security patrols. He said teenagers were responsible for most of the violations. A total of 200 violators were arrested, detained for 48 hours and fined SR500 each. Al-Hamzi said that during the same period, about 1,500 offenses were detected for drifting, passenger overloading, hiding of license plates, driving without seat belts, speeding, and talking on mobile phones while driving. Al-Hamzi urged motorists to abide by the country's traffic laws for the safety of all road users. [5].

The World Health Organization found Saudi Arabia to have the world's highest number of deaths from road accidents, which now make up the country's principal cause of death in adult males aged 16 to 36. First reported by the Saudi daily Arab News, the study found that 6,485 people had died and more than 36,000 were injured in over 485,000 traffic accidents during 2008 and 2009. [2].

An average of 19 Saudi Arabian residents die on the country's roads each day, a report by the Kingdom's General Directorate of Traffic has revealed. [7] In an interview with *Arab News* in September, the associate vice president and transportation systems director of Middle East Operations at traffic management consultancy Iteris Inc., Glenn N. Havinovski, said infrastructure wasn't an issue, but "when you see people turning left out of the far right lane and traffic cutting through parking lots and frontage roads, there are clearly some issues with discipline." [6]

Study Aim and Objectives:-

There are very few publications on the drifting and none were found in the on the Middle Eastern population. The aim of the study is to document and analyze road traffic accident data from drifters.

Objectives:-

- Analyze road traffic accident data from drifters.
- Investigate the lifestyle of drifters.
- Suggest lowering the risk of accidents caused by drifting.
- Recommend designated areas for drifting and attracting drifters to these areas.
- Suggest safe practices for drifting.
- Investigate risk factors for drifting.
- Suggest lifestyle modifications in drifting to improve quality of life and better future.
- The benefit of this work is that this research will help in raising the level of awareness the society by explaining the side effects and harms of drifting.

Methods:-

1. Study design: Random, cross-sectional study
 2. Study population and sample size: This will be a random cross-sectional study of 100 male subjects, aged 16 to 40 years, who will be recruited by personal contacts and online forums of the drifting community. Exclusion criteria will be subjects who do not drift. All subjects will complete a questionnaire.
 3. Data collection: A questionnaire has been prepared and will be validated to make sure it is reliable. It will pilot tested in AGU before being made available to drifters via an online questionnaire.
- Variables: The research is based on questionnaire (see Appendix) results recording age, marital status, occupation, educational level. Other questions are age of drifting initiation, what factors encourage you to drift, what factors discourage you to drift, car model, presence or absence of a driving license, drift frequency, seat belt use, frequency of car damage, frequency of being caught or arrested by police, whether any friends died or were injured by drifting and which areas do you prefer to drift.

Data Analysis:-

Method of analysis: All data will be presented as mean \pm standard deviation unless otherwise stated. Package used: IBM SPSS Statistics 19. Statistical tests used: Paired t-test, linear and multiple regression analyses.

The data will be analysed by SPSS version 19. The results will be expressed as means (\pm SD). Odds ratios (OR) with 95% confidence intervals (CI) will be calculated.

The Pearson's Chi squared (χ^2) and Fisher's exact tests were used to determine statistically significant associations of categorical variables. For quantitative variables, an unpaired t-test was used.

Results:-**A. Descriptive results**

All 105 subjects in this study were male and all were from the Kingdom of Saudi Arabia.

The mean age (\pm SD) of all our subjects was 18.44 ± 3.33 years. Their age range was 14 – 36 years. Their age of initiation i.e. the age they started drifting, was 15.10 ± 1.86 years. Their age of initiation range was 12 – 26 years. The mean number of times they drifted in one week was 2.63 ± 1.77 times.

B. Frequency

94 % of the drifters in this study were single (Figure 1). The remaining 6 % of drifters were equally either married or divorced. In terms of age, most subjects in our study were young. 54 % of drifters were aged 14 to 17 years, 29 % were aged 18 to 21 years and only 17 % were aged 22 years and above (Figure 2).

In terms of occupation, 71.4 % of the drifters in this study were students, while 21.9 % were unemployed and only 7 % were other i.e. employed (Figure 3). 91.4 % of our drifters started at a young age (12 – 17 years), 8.5 started from 18 to 21 and only 1 % of people started to drift after 22 years of age (Figure 4).

In our population, friends and free time were considered as the major encouraging factors for drifting with percentages of 35.2 % and 23.8 %, respectively (Figure 5). The remaining 41 % of encouraging factors included after exams, after football matches, stress and other factors. The greatest factors which helped discourage drifters in our study to drift were family (34.3 %). Other discouraging factors included police, religion, teachers, fear and other factors (Figure 6).

69.5 % of drifters in our study revealed that there was damage to their own vehicle or to others vehicles during the time they were drifting (Figure 7). A minority of only 30.5 % reported that there no car damage during the time they were drifting. 38.1 % of our drifters admitted they had been arrested for drifting on at least on one occasion (Figure 8). The majority were not arrested.

72.4 % of our drifters would participate in drifting 1-3 time/week, while only 27.6 % would get involved 4-7 times per week.

Figure 1:- Bar chart showing the frequency of marital status of drifters in our study (n=105).

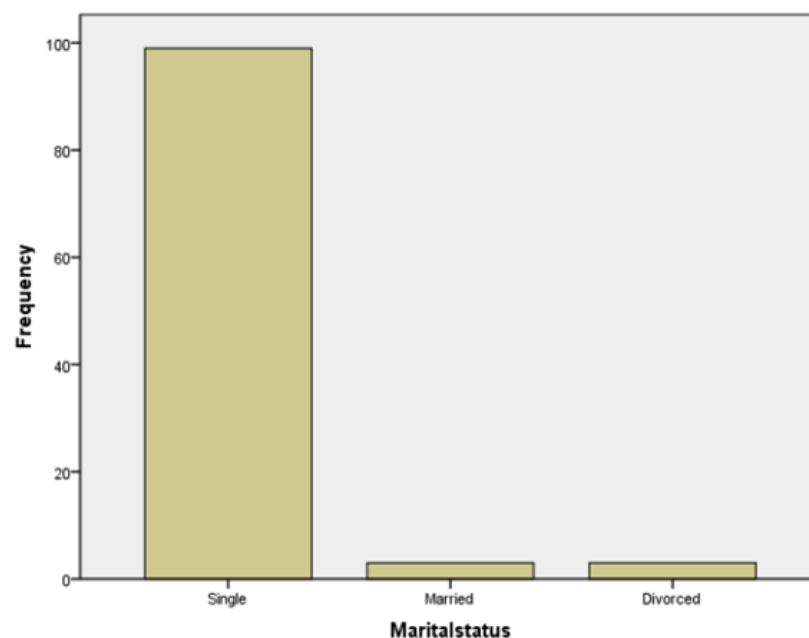


Figure 2:- Bar chart showing the frequency of various ages of drifters in our study (n=105).

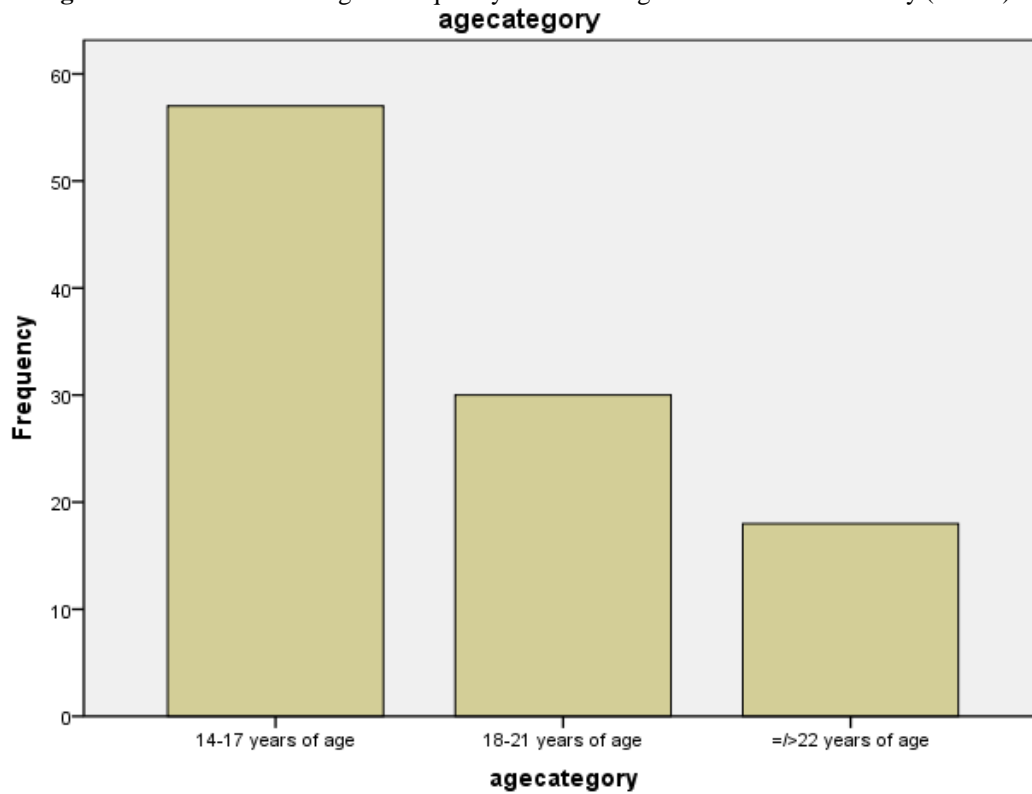


Figure 3:- Bar chart showing the frequency of various occupations of drifters in our study (n=105).

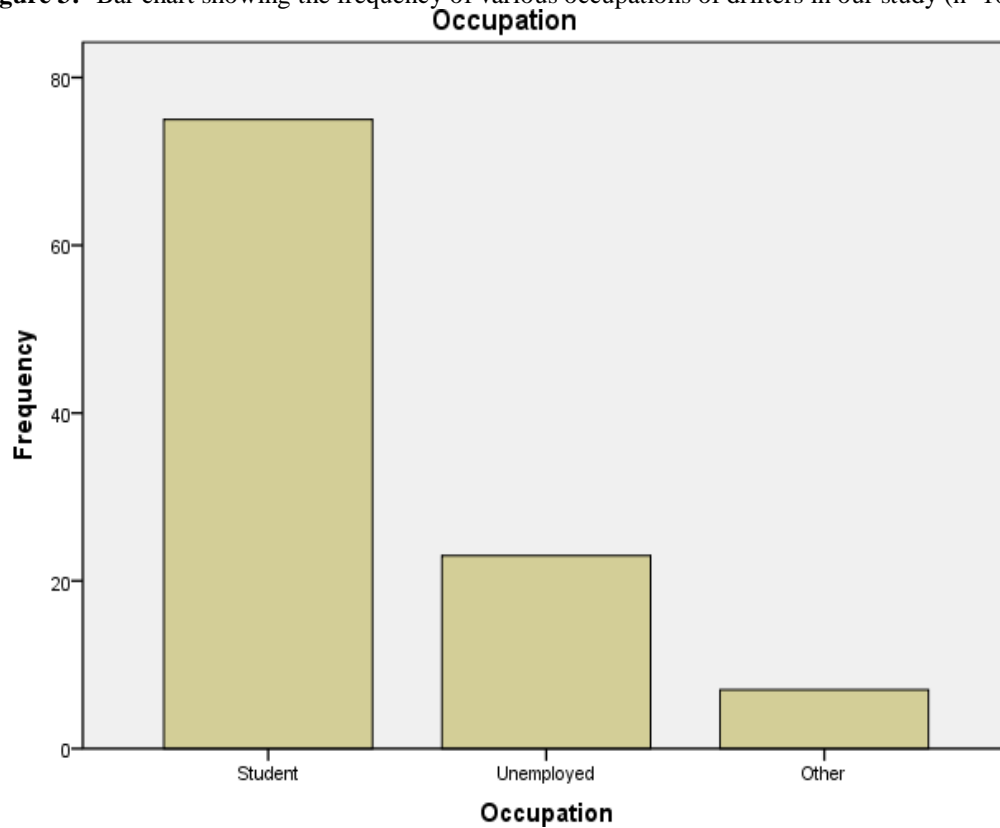


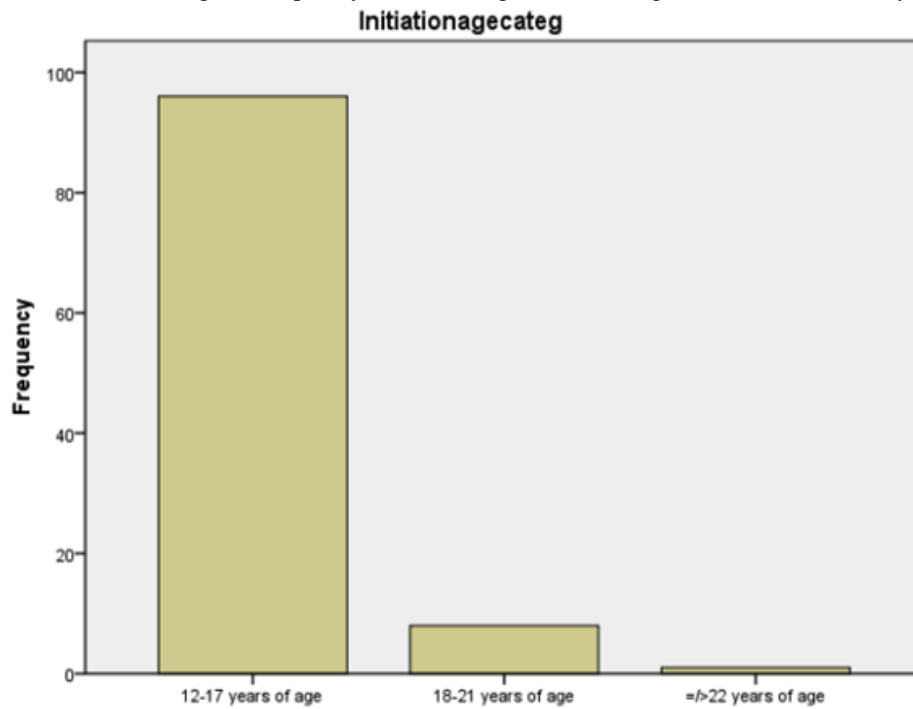
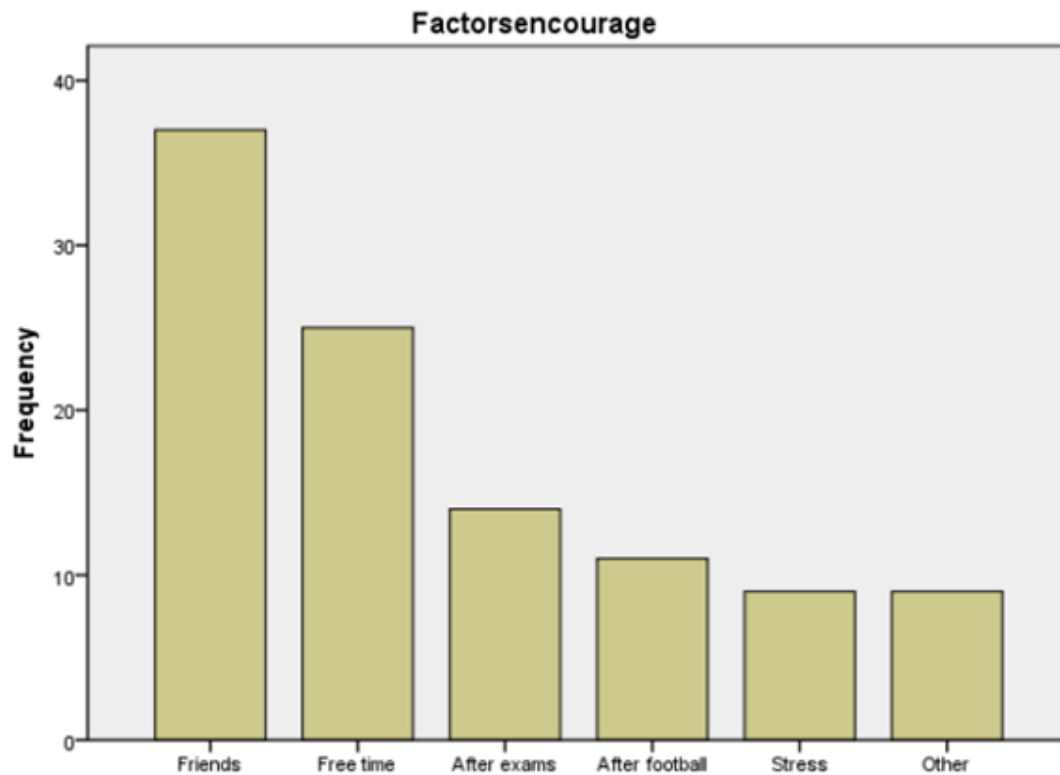
Figure 4:- Bar chart showing the frequency of various ages of initiating in drifters in our study (n=105).**Figure 5:-** Bar chart showing the frequency of various factors which encouraged our subjects to drift (n=105).

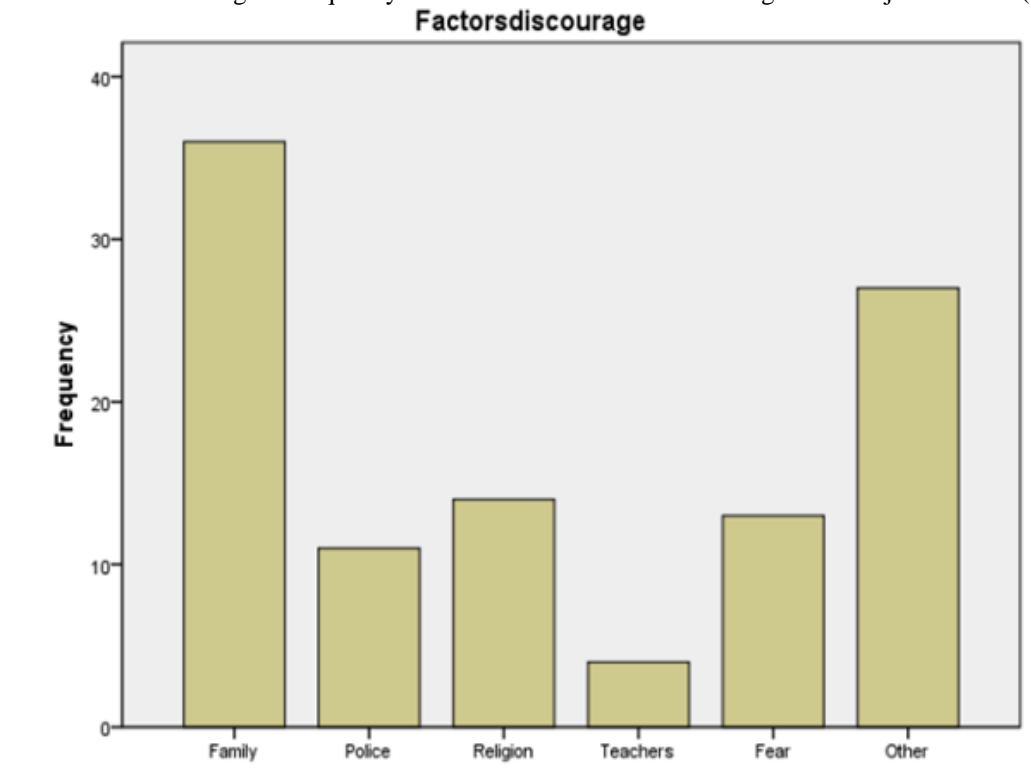
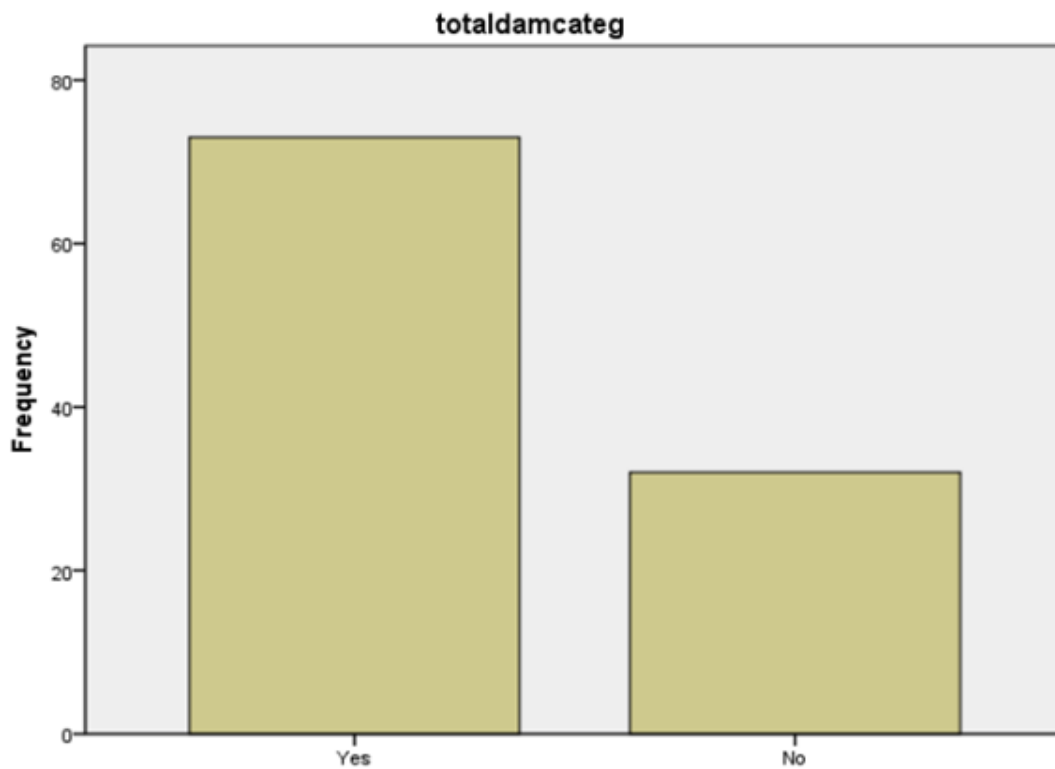
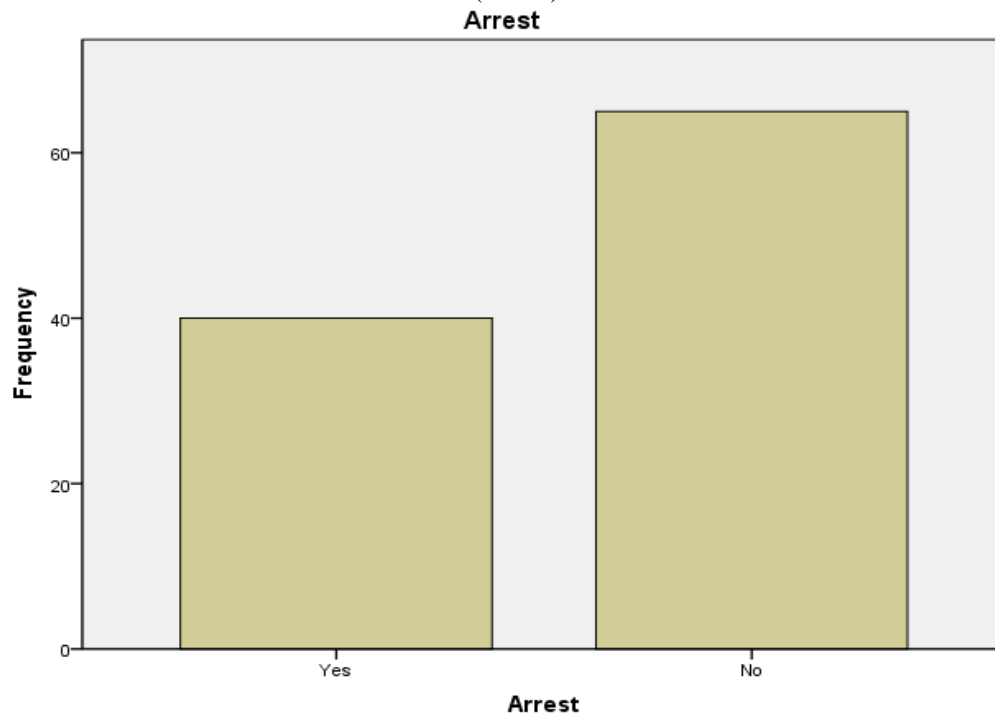
Figure 6:- Bar chart showing the frequency of various factors which discouraged our subjects to drift (n=105).**Figure 7:-** Bar chart showing the frequency of total vehicle damage (own and others vehicles) during drifting (n=105).

Figure 8:- Bar chart showing the frequency of whether our study participants were ever arrested during drifting or not (n=105).



Chi-square analysis

I. Effect of age on results state ...

- ❖ Age did not have a significant effect on whether drifters were arrested or not ($\chi^2 = 0.288$, $p = 0.591$). See Table 1.
- ❖ There were also no significant effect of the age on whether police caught drifters or not ($\chi^2 = 0.338$, $p = 0.561$).
- ❖ All the following relationships were not significant when relating age to the following:
 1. death of friend during drifting ($\chi^2 = 2.627$, $p = 0.105$)
 2. frequency of drifting ($\chi^2 = 0.450$, $p = 0.502$)
 3. injury of friend during drifting ($\chi^2 = 2.702$, $p = 0.100$)
 4. seatbelt use during drifting ($\chi^2 = 0.586$, $p = 0.444$)
 5. total damage to the car ($\chi^2 = 2.794$, $p = 0.095$)
- ❖ There was also no significant effect of the age category on the frequency of drifting (Table 2; $\chi^2 = 0.338$, $p = 0.561$).

Table 1:- χ^2 analysis showing relationship between arrest and age of drifters (n=105).

		Arrest		Total
		Yes	No	
agecategory	14-17 years of age	20	36	56
	=/>18 years of age	20	29	49
Total		40	65	105

Table 2:- χ^2 analysis showing relationship between frequency of drifting and their age category (n=105).**agecategory * Driftfreqcateg Crosstabulation**

Count		Driftfreqcateg		Total
		1-3 times/week	4-7 times/week	
agecategory	14-17 years of age	39	17	56
	=/ > 18 years of age	37	12	49
Total		76	29	105

II. Age of initiation of drifting:

There were no significant effects of age of drifting initiation on the following:

- ❖ Death ($\chi^2 = 0.861$, $p = 0.353$).
- ❖ Injury ($\chi^2 = 0.047$, $p = 0.674$).
- ❖ Seat belt use ($\chi^2 = 0.008$, $p = 0.927$).
- ❖ Total vehicle damage ($\chi^2 = 0.585$, $p = 0.444$).

III. Occupation:

Unemployed drifters had significantly higher prevalence of injuries of a friend during drifting than students drifters ($\chi^2 = 8.111$, $p = 0.004$). See Table 3.

Table 3:- χ^2 analysis showing relationship between injury and occupation of drifters (n=105).

		Injury		Total
		Yes	No	
Occupation	Student	30	45	75
	Unemployed	17	6	23
Total		47	51	98

There were no significant effects of occupation of drifters on:

- ❖ Death ($\chi^2 = 3.099$, $p = 0.078$).
- ❖ Seatbelt use ($\chi^2 = 1.527$, $p = 0.217$).
- ❖ Total vehicle damage ($\chi^2 = 2.473$, $p = 0.116$).
- ❖ Arrest during drifting ($\chi^2 = 0.074$, $p = 0.785$).
- ❖ Police catching drifters ($\chi^2 = 0.008$, $p = 0.767$).

Car model vs. seatbelt

Users of Camry and Hilux drifting vehicles had significantly lower prevalence of seat belt use during drifting (Table 4). As the χ^2 analysis had an expected count lower than 5, Fisher's Exact Test was used. This showed that when Camry cars were compared with Other cars, Camry users had significantly lower prevalence of seat belt use during drifting ($p=0.05$). Similarly, when Hilux and Other cars were compared, Hilux users had significantly lower prevalence of seat belt use during drifting ($p=0.04$).

Lastly, using a χ^2 analysis Camry and Hilux was combined and compared to Others cars. Results showed Camry + Hilux users had significantly lower prevalence of seat belt use during drifting ($\chi^2 = 5.13$, $p=0.023$). The odds ratio of wearing a seatbelt during drifting (to not of wearing a seatbelt during drifting) in Camry + Hilux users, compared to Other car users was 0.286 (95 % CI, 0.091 - 0.864).

Table 4:- χ^2 analysis and results showing the relationship between seat belt use and car model used by drifters (n=105).

		Seatbeltuse		Total
		Yes	No	
Car model	Camry	4	24	28
	Hilux	2	18	20
	Other	19	38	57
Total		25	80	105

	Value	df	Asymp. Sig. (2-sided)
Pearson Chi-Square	6.353 ^a	2	.042
Likelihood Ratio	6.731	2	.035
Linear-by-Linear Association	4.647	1	.031
N of Valid Cases	105		

a. 1 cells (16.7%) have expected count less than 5. The minimum expected count is 4.76.

Discussion:-

Our main finding in this study was that the majority of drifters are teenager students and they learned it and started it at an early age, with their friends being the most encouraging factor and their family the most discouraging factor. Most of them damaged cars and many knew a friend who was injured or killed by drifting. Camry & Hilux users seemed to have more dangerous behaviours (not using their seatbelts) compared to other car users.

There are no deeper reasons apart from excitement and fun making that makes people in Saudi Arabia take part in drifting. In truth, drifting is mostly a reflection of other social issues including drug abuse, selfishness and deviance. Many people who attempt or actually drift their cars might be under the influence of drugs. What moves one to drifting is purely a selfish desire to satisfy one's curiosity without put into considerations other road users and relatives. The fact that drifting is illegal in Saudi Arabia, participating in it is an outward show of the levels of deviance among Saudi drivers. It is expected that once the government declares an act illegal people should desist from it.

Major concerns noted in our study were that the average age of 105 drifters was 18 years of age and some were as young as 14 years. Also, we found that the most of them initiated drifting at a very young average age of only 15 years, and some had started by even 12 years of age. It is difficult to imagine how the parents of these children allow this, but it is happening. At the time of injuries or death, parents have called for the death penalty of the responsible drifting drivers, but parents shoulder the full responsibility of their children's actions and should know where they are and what they are doing. In assessing factors encouraging our drifters, we noticed that friends and free time were considered the most important numerically, while their family was one of several factors discouraging them from drifting the most. In order to limit the level of drifting we have to deal with the most discouraging factor starting with their family. The family and parents should make their relationship stronger with their children, talk to them, let them know that they are an important member in their family, that they love them and that they are afraid from losing them. This can be exploited where by the Ministry of the Interior and Transport Departments can advertise on television and through brochures/pamphlets and target drifters and their families. Information can be given to them about the dangers of drifting with statistics to prove their point.

The majority of drifters in our study had either damaged their car or other cars while drifting. This is a large amount. If these figures were only halved, lots of resources including court, police, hospital, car insurance and other costs will be saved. No one has estimated the financial burden of drifting in Saudi Arabia, but an intelligent guess would put it in the billions of Riyals annually.

Approximately one third of our drifters' claim they had been arrested by the police. Video footage of drifters often shows they are ready and planning to escape. They choose a specific area e.g. empty space out of the city; this gives them the opportunity to escape easily. It was interesting to note that drivers who used a Camry or Hilux to drift had significantly less seatbelt usage than other car drifters. Therefore, it seems these two brands were popular and maybe drivers of these cars had more dangerous behaviours compared to other car users. The number of drifters who knew someone who had died or was injured during drifting was almost equal. It is unclear whether the factors

discouraging them from drifting would be different between these two groups, but we did not analyse this data as our sample size was only 105 and we had several discouraging factors.

With regard to the age and gender of the drifters, all are male and most are young. They are at a sensitive age with their hormones changes, in which their testosterone levels are increasing. They have a greater desire to try new things and get involved in different activities. If they are not given opportunities for outings, sports, etc then they might be attracted to drifting and other dangerous behaviours such as substance abuse. We noticed that their friends are the most common factor for them encouraging them to drift.

We had no way to compare our data with other GCC countries, such as Kuwait, Qatar, Bahrain or the UAE, as we could not find any published data on this topic. However, anecdotal evidence suggests this problem is not very common in these other neighbouring countries. One possible reason for this could be that in these other GCC countries, their governments have provided avenues for young men to release their energy, be it by providing special spaces for these activities, other sports activities, competitions, greater opportunities for higher education, allowing them drift or drag race during competitions, so to allow it in a legal way. One such example is the Bahrain International Circuit (Formula1 Track), where people can drift or drag legally and safely using their own cars and also they have opportunities to use go-karts.

In coming to the outcome, every year the government loses billion of riyals to treat the drifters and the people who get injured from drifting. A large proportion of hospital beds have patients paralyzed by road traffic accidents including drifters or their spectators. In addition to all of that they waste the time of policemen who try to stop and catch them. It's a very serious issue which has also reached the social media. A famous Saudi television programme called 'Al Tamanna with Dawood Shriya', contacted some drifters for one of their episodes. They revealed two shocking facts: firstly, some of drifters steal others cars in order to use it in drifting and then they burn it after they have finished drifting. Secondly: they hide their faces and refuse giving their real name, but use nicknames in their activity. This is obviously so they cannot be identified by the authorities.

A limitation of our study was that data was only collected from one GCC country.

Drifting has a negative effect on the socioeconomic and health state of the country. We think it is difficult to stop, but recommend it should be regulated so it is a legal activity in special areas, so to keep drifters busy but in a safe environment. This will save lives and reduce the socioeconomic and health cost burden. They harm themselves and they should be aware of what Allah says in the Quran (وَلَا تُلْقُوا بِأَيْدِيكُمْ إِلَى التَّهْلُكَةِ) 'do not throw [yourselves] with your [own] hands into destruction' البقرة آية 195

Conclusion and Recommendation:-

There is no doubt that drifting is a now major problem in Saudi Arabia. The fact that road accidents have been rated the number one killer shows how a menace drifting is. What makes it more problematic is that fact that many commercial organizations have funded advertisements for drifting or dangerous driving in order to make their products acceptable and popular among the youths. In addition, the increase in internet access as seen by the availability of free drifting videos in YouTube has continued to encourage many youths to take part in the act. The best way to decrease its frequency should be stripping away the participants driving licenses' besides charging them in a court of law. In this case they will be unwilling to participate since their freedom to drive will be taken away from them once their driving licenses are impounded.

It is really unfortunate that commercial enterprises have found a platform to advertise their commodities in such a social menace as drifting. Youth subcultures especially dangerous sports like drifting should be discouraged by all stakeholders. The idea of commercial enterprises to fund dangerous behaviours is very frustrating and in bad faith. Such firms include manufacturers of cars, tires and soft drinks. They are contributing to a course that will see many youths engage in dangerous driving hence high speed road accident cases and ultimately raising death tolls. This is the case where commercial enterprises run product adverts that show product acceptance among street youth gangs that are suspected of perpetuating crime and other social evils.

We don't want to have our youth living fast and dying young but rather living the normal pace and fully completing the metamorphosis of phases that life has laid before them. Until next time, please slow the pace down. From this it is obvious that to discourage drifting, ministries of health, interior, sports, culture and education need to work together to rid ourselves of this evil.

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Appendix

'Survey on road traffic accidents as a consequence of "drifting" in Saudi Arabia'

1. Age: _____ years
2. Nationality: _____
3. Marital Status: ☐ single ☐ married ☐ divorced
4. Occupation: _____
5. Education level: ☐ High school ☐ University
6. What was your age when you started drifting? _____ years
7. What encourages you to drift? (can be more than one box)
 - ☐ Friends
 - ☐ Too much free time
 - ☐ After exams
 - ☐ After football game
 - ☐ Stress
 - ☐ Other (_____)
8. What model is your car? _____
9. Do you have a driving license? ☐ Yes ☐ No
10. How often do you drift? _____ week
11. Do you use a seat belt while you drift? ☐ Yes ☐ No
12. How many times did you damage your own car during drifting? _____
13. How many times did you damage others cars during drifting? _____
14. How many times did the police catch you drifting? _____
15. Did you ever get arrested by the police for drifting? ☐ Yes ☐ No
16. What discourages you to drift?
 - ☐ Family
 - ☐ Police
 - ☐ Religion
 - ☐ Teachers
 - ☐ Fear of injury/death
 - ☐ Other (_____)

17. Did any of your friends die by drifting? ☐ Yes
☐ No
18. Did any of your friends/spectators get seriously injured by drifting?
☐ Yes
☐ No
19. Where do you prefer to drift?
☐ Main roads
☐ Open areas
☐ Other (_____)



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RESEARCH ARTICLE

APPLICATION OF DESIGN EXPERT IN THE ANALYSIS OF RESPONSE TRANSFORMATION OF PROCESSES – A CASE STUDY OF BIOETHANOL PRODUCTION PROCESS FROM CORN-STOVER.

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Abstract

Statistical tools and features of the Design Expert software (7.0.0 Trial version) were used in experimental design and analyses of bioethanol production process from corn stover. The Box-Behnken design (BBD) was used in the design of the experiments and it yielded a total of 54 experimental runs. The response variable, Bioethanol (mg/l) was represented with Y, while the six independent variables; Sulphuric acid concentration, Hydrolysis time, Fermentation time, Concentration of yeast, Fermentation temperature and pH of Hydrolysate samples were represented by A, B, C, D, E & F respectively. A model equation was developed from the experimental results using the analysis of variance (ANOVA) feature of the Design Expert software. From the analyses of variance, only the terms A, F, AF, CD, A² were significant model terms (having P-value ≤ 0.05), which resulted in a reduced Model Equation (i.e. Adjusted) in terms of actual factors, as follows:

$$Y = 552.60398 - 0.31052*A + 3.87830*F - 5.76556*A*F - 0.26625*C*D + 5.38883*A^2.$$

The optimum process parameters for the predicted optimum bioethanol yield of 149.41 mg/l were 1.08%, 3.32hrs, 14.32hrs, 6.43g/l, 39.34°C and 7.64 for A, B, C, D, E & F respectively. However, an experimental bioethanol yield of 143.15 mg/l was obtained from three experimental replicates with the optimum process parameters. The experimental bioethanol yield obtained was 95.81% close to the predicted optimum bioethanol yield. The values of R, R², adjusted R² and Adeq Precision were 0.8747, 0.7651, 0.5212 and 8.309 respectively. The value of R² indicates a good degree of correlation between the experimental and predicted bioethanol yields. The model is adjudged to be adequate since the "Adeq Precision" is greater than 4.

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Introduction:-

The bioethanol production from a lignocellulosic material, corn-stover using the separate hydrolysis and fermentation (SHF) process where hydrolysis (saccharification) of the cellulose to glucose occurs first, separate

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from the fermentation of the resulting glucose and other sugars (from the pretreatment of hemicellulose,) to ethanol. The separate saccharification step enables operation of the saccharification step at an elevated temperature. For fermentation, the *Saccharomyces cerevisiae* is used as the biocatalyst which will ferment glucose and other sugars to ethanol. Some important factors that affect the performance of separate hydrolysis and fermentation (SHF) and thus bioethanol yield are: concentration of the hydrolysis solution, hydrolysis time, pH of hydrolysate, concentration of yeast, fermentation temperature and time. The feasibility of these two operations is determined by the effects of each factor as well as the interactions between factors at varying values of all the factors.

Thus to design the experiment involving multivariable factors, a statistical software package like the Design-Expert, that is specifically dedicated to performing comparative tests, screening, characterization, optimization, robust parameter design, mixture designs and combined designs is highly recommended (Tanco *et al.*, 2008). Design-Expert offers test matrices for screening up to 50 factors. To keep pace with this, therefore, a power calculator which helps to establish the number of test runs needed is required. Statistical significance of these factors is established with analysis of variance (ANOVA). Based on the validated predictive models, a numerical optimizer helps the user to determine the ideal values for each of the factors in the experiment. Graphical tools help identify the impact of each factor on the desired outcomes and reveal abnormalities in the data (Cornley, 2009). Design-Expert provides 11 graphs in addition to text output to analyze the residuals (Plant, 2013). The software determines the main effects of each factor as well as the interactions between factors by varying the values of all factors in parallel (Black, 2013). Design-Expert provides the user with a broad range of possible response transformations. The appropriate choice depends on subject matter and/or statistical considerations. The software provides extensive diagnostic capabilities to validate statistical assumptions. This feature of the Design-Expert offers a helpful plot, called the Box-Cox, which recommends the appropriate power transformation (including the no transformation option). Design-Expert also has an option of plotting the responses in terms of the original response data by calculating the surface matrix of data points and then applying the inverse transform before making the plot.

In the present study, the application of statistical tools of the design expert software to determine statistical significance of the various process factors in bioethanol production from corn-stover is presented as a case study. The impact of these factors on the response (i.e. bioethanol yield), diagnostic capacities to validate statistical assumptions, possible response transformations and the criteria for the choices of transformation were clarified.

Materials and Methods:-

2.1 Design of Experiment:-

Box-Behnken design (BBD), a statistical tool, was used in the design of the experiment in this study. In BBD, the six independent variables; sulphuric acid concentration X_1 (A), hydrolysis time X_2 (B), fermentation time X_3 (C), concentration of yeast X_4 (D), fermentation temperature X_5 (E), and pH of hydrolysate X_6 (F), were all set at three levels; minimum (-1), centre (0) and maximum (1). The BBD was used to determine the number of runs or sets of experiments that were needed to be carried out. Each experiment was carried out in triplicates and the mean values outcome, taken as response, Y, which is bioethanol yield (mg/l).

For statistical calculations, the variables $X_1, X_2, X_3, \dots, X_n$ were coded as $x_1, x_2, x_3, \dots, x_n$ respectively, according to Equation 1 Nuran (2007):

$$x_i = \frac{(X_i - \bar{x}_i)}{\Delta x_i} \quad i = 1, 2, 3, \dots, n \quad (1)$$

Where x_i is a dimensionless value of the independent variable, \bar{x}_i is the mean value of the independent variable at the center point and Δx_i is the step change.

The design matrix used for the six independent variables showing the real values of the variables in terms of the three levels is presented in Table 1.

Table 1:- Design matrix employed for the six independent variables

Factors	Code	Range		
		-1	0	+1
Sulphuric Acid Concentration (%)	A	1.00	2.50	4.00
Hydrolysis Time (Hrs)	B	2.00	4.00	6.00
Fermentation Time (Hrs)	C	12.00	30.00	48.00
Concentration of Yeast (g/l)	D	3.00	6.00	9.00
Fermentation Temperature (°C)	E	30.00	35.00	40.00
pH of Hydrolysate	F	5.00	6.50	8.00

The outcome of the BBD experimental design of the process (Separate Hydrolysis and Fermentation, SHF) using the Design Expert software, resulted in a total of 54 experimental runs as sets of coded variables.

Pretreatment, Hydrolysis and Fermentation of corn stover:-

The pretreatment, hydrolysis and fermentation of the corn stover were carried out in line with standard procedure (Ohimor *et al.*, 2016). The milled corn stover was alkaline pretreated with dilute sodium hydroxide (2% w/w NaOH) summing to a solid to liquid weight ratio of 1:8. The hydrolysis of the alkaline pretreated corn stover was with dilute-acid (H₂SO₄) of varying concentrations (1, 2.5 and 4%), such that a solid to liquid ratio of 1:10 is maintained in a 250ml round bottom flask and then refluxed. Hydrolysate samples were retained at intervals of 2, 4, and 6 hours for subsequent fermentation. The hydrolysate sample for fermentation was adjusted to the various pH of 5, 6.5 and 8 by adding concentrated sulphuric acid and 2N sodium hydroxide as may be appropriate.

Varying quantities of yeast (*Saccharomyces cerevisiae*) equivalent to 3, 6 and 9 g/l respectively where added to each hydrolysate samples contained in 250 ml Erlenmeyer flask, then incubated at various temperatures (30, 35 and 40 °C) for fermentation. The bioethanol content was determined by gas chromatography at various fermentation times of 12, 30, and 48 hours.

Statistical Analyses:-

Upon completion of the laboratory analyses, the results (bioethanol yield) of all 54 experimental runs were entered into a table in the file already created in the Design Expert software for subsequent analysis. The results were statistically analyzed in order to have an understanding of the interactions between variables and the bioethanol yield. These results were also used for the optimization of the process.

A second order polynomial model, similar to that of Dasgupta *et al.* (2013), was obtained from the Design Expert software indicating linear, interaction and quadratic effect of variables on the response variable (Y).

$$Y = b_0 + b_1X_1 + b_2X_2 + b_3X_3 + b_{11}X_1 + b_{22}X_2 + b_{33}X_3 + b_{12}X_1X_2 + b_{23}X_2X_3 + b_{13}X_1X_3 \quad (2.2)$$

Where Y is the predicted response; X_1, X_2, X_3 are the independent variables; b_0 is the offset term; b_1, b_2, b_3 are the linear effects; b_{11}, b_{22}, b_{33} are the square effects; and b_{12}, b_{23}, b_{13} are the cross effects of the interaction terms.

Using regression analysis, the significance of the coefficients of the terms in the model equation was determined by computing the standard errors, T-values and P-values. Analysis of variance (ANOVA) was also used to determine the level of confidence (α) as well as linear, interaction and quadratic effects. The quality of fit of the BBD model was estimated by comparing the variance (R^2) of the adjusted model and that of the predicted Model.

Finally, the experiment was repeated in triplicate using the variables that gave the optimum bioethanol yield and the average of the results was compared to the optimum value. The validity of the model was adjudged to be good since the result is comparable to the optimum bioethanol yield to an extent. Point Prediction was carried out which allows the entering of each factor or component into the current model at different levels. The software then calculates the expected responses and associated confidence intervals based on the model equation that is shown in the ANOVA output. The predicted values are updated as the levels are changed. The **95%** confidence interval (CI) is the range in which the process average is expected to fall into 95% of the time. The 95% prediction interval (PI) is the range in which any individual value is expected to fall into 95% of the time. The prediction interval will normally have a wider spread than the confidence interval because of the random nature of the individual values when compared to the averages.

Results and Discussion:-

Experimental Design of the Bioethanol Production Process and Bioethanol Yield:-

The results of the design of experiments and experimental bioethanol yields (mg/l) for each runs are presented in Table 2; it shows the experimental design of the six (6) independent variables for the bioethanol production process in terms of their coded levels (i.e. -1, 0, +1).

Table 2:- Experimental design and bioethanol yield

Run Order	Coded Variables						Experimental	Std
	A	B	C	D	E	F	Bioethanol Yield (mg/l) (Y)	
1	0	0	0	0	0	0	58.14	51
2	+1	0	+1	0	0	-1	52.77	44
3	+1	0	-1	0	0	+1	53.28	46
4	0	0	-1	+1	0	+1	87.44	23
5	-1	+1	0	+1	0	0	63.89	7
6	0	+1	-1	0	+1	0	65.74	14
7	-1	-1	0	+1	0	0	86.19	5
8	+1	0	0	+1	+1	0	54.52	32
9	0	0	0	0	0	0	66.89	52
10	0	+1	0	0	+1	-1	81.88	36
11	0	-1	0	0	+1	-1	53.36	35
12	0	-1	0	0	-1	+1	37.48	37
13	+1	0	+1	0	0	+1	87.21	48
14	-1	0	0	-1	+1	0	88.32	29
15	0	0	+1	-1	0	-1	69.56	18
16	0	0	+1	+1	0	+1	39.34	24
17	0	0	0	0	0	0	67.33	49
18	0	0	+1	-1	0	+1	86.64	22
19	0	+1	-1	0	-1	0	72.86	10
20	-1	0	-1	0	0	-1	79.25	41
21	+1	-1	0	-1	0	0	61.26	2
22	+1	0	0	-1	-1	0	68.29	26
23	0	-1	0	0	+1	+1	109.23	39
24	0	-1	+1	0	-1	0	66.77	11
25	-1	0	+1	0	0	-1	68.14	43
26	0	0	+1	+1	0	-1	57.33	20
27	0	0	0	0	0	0	88.76	50
28	0	-1	-1	0	+1	0	88.68	13
29	+1	+1	0	-1	0	0	41.44	4
30	+1	-1	0	+1	0	0	39.88	6
31	0	0	-1	+1	0	-1	66.73	19
32	-1	0	0	+1	+1	0	134.88	31
33	-1	0	0	-1	-1	0	111.87	25
34	-1	0	-1	0	0	+1	139.56	45
35	0	0	-1	-1	0	+1	65.56	21
36	0	-1	+1	0	+1	0	68.22	15
37	0	0	0	0	0	0	67.33	53
38	-1	-1	0	-1	0	0	87.93	1
39	-1	0	+1	0	0	+1	147.64	47
40	+1	0	-1	0	0	-1	51.69	42
41	0	+1	0	0	+1	+1	63.42	40
42	0	+1	+1	0	-1	0	83.02	12
43	+1	+1	0	+1	0	0	62.24	8
44	-1	+1	0	-1	0	0	76.38	3

45	+1	0	0	-1	+1	0	28.62	30
46	+1	0	0	+1	-1	0	39.74	28
47	0	-1	0	0	-1	-1	66.13	33
48	-1	0	0	+1	-1	0	114.23	27
49	0	-1	-1	0	-1	0	68.01	9
50	0	+1	0	0	-1	+1	68.06	38
51	0	+1	0	0	-1	-1	73.76	34
52	0	0	-1	-1	0	-1	33.12	17
53	0	0	0	0	0	0	67.33	54
54	0	+1	+1	0	+1	0	91.45	16

The analytical experiments were then carried out according to the assignment of variables in Table 2. The experimental runs were performed randomly to avoid systemic error.

Transformation Equation:-

The response (i.e Bioethanol Yield) ranged from 28.62 mg/l to 147.64 mg/l. The ratio of the maximum to minimum response determines whether a transformation equation is needed or not for the statistical analysis. A ratio greater than 10 usually indicates a transformation is required. For ratios less than 3, the power transform will have little effect. For ratios ranging from 3 to 10, a transform is not required.

Transformation types can either be Square Root, Natural Logarithm, Base 10 Logarithm, Inverse Square Root, Inverse, Power, Arcsin Square Root or None.

From the experimental results, the ratio of maximum to minimum bioethanol response is 1 : 5.15863, hence a transform was not required before we proceeded with the statistical analysis.

Model type selection:-

From the statistical analysis using Design Expert, the information on *Sequential Model Sum of Squares [Type I]* and *Lack of Fit Tests*, shown in Tables 3 and 4 respectively, were obtained.

Table 3:- Sequential model sum of squares [Type I]

Source	Sum of Squares	df	Mean Square	F- Value	p-value Prob > F	Comment
Mean vs Total	2.844E+005	1	2.844E+005			
Linear vs Mean	15434.74	6	2572.46	6.82	< 0.0001	Suggested
2FI vs Linear	6864.30	15	457.62	1.35	0.2321	
Quadratic vs 2FI	3074.07	6	512.35	1.71	0.1584	
Cubic vs Quadratic	6223.58	18	345.75	1.77	0.2080	Aliased
Residual	1565.33	8	195.67			
Total	3.175E+005	54	5880.54			

Table 4:- Lack of fit tests

Source	Sum of Squares	Df	Mean Square	F- Value	p-value Prob > F	Comment
Linear	17206.59	42	409.68	3.93	0.0647	Suggested
2FI	10342.29	27	383.05	3.68	0.0755	
Quadratic	7268.22	21	346.11	3.32	0.0932	
Cubic	1044.64	3	348.21	3.34	0.1134	Aliased
Pure Error	520.69	5	104.14			

For the "Sequential Model Sum of Squares [Type I]", it is expected that the model equation that was not aliased is to be selected provided the model is shown to be significant in terms of its "Model F-value". In this case the Cubic vs Quadratic Model was aliased, meaning that the Cubic Model cannot be resolved. The test suggested Linear vs Mean, meaning that a Linear Model is preferred compared to a Mean. Nevertheless, the Quadratic Model was preferred over 2FI (i.e two – factor interaction) because it was not Aliased. The choice of the Quadratic Model was to account for interactions between two or more variables.

For the "Lack of Fit Tests", the selected model is to have an insignificant lack-of-fit, which is a "P-value " that is > 0.05. In this case the software suggested Linear, however, the Quadratic model is preferable in as much as it was not aliased, with a P-value of 0.0932. The choice of the quadratic over linear model is to be able to account for interactions between two or more variables.

The "Lack of Fit F-value" of 3.32 implies there is a 3.32% chance that a "Lack of Fit F- value" as much as this could occur due to noise. Lack of fit is bad -- we want the model to fit. This relatively low probabiity (<10%) is worrisome. What this means is that there are many insignificant model terms and as such, there is need to consider model reduction in order to improve the model.

Regression analysis:-

The result of the regression analysis is shown in Table 5 below. If the T-value is above threshold and P-value is lower than 0.05 for any given term, then that term is said to have a meaningful contribution to bioethanol production. Terms that have no contribution were removed from the predicted model and a new model referred to as adjusted model was obtained.

Table 5:- Regression analysis

Source	Coefficient Estimate	Sum of Squares	DF	Mean Square	F-value	P-value (Prob. >F)	
Model	69.30	25373.11	27	939.74	3.14	0.0023	Significant
A-H ₂ SO ₄	23.22	12942.83	1	12942.83	43.20	< 0.0001	
B-Hydrolysis Time	0.46	5.04	1	5.04	0.017	0.8978	
C-Ferm. Time	1.92	88.82	1	88.82	0.30	0.5907	
D-Conc. of Yeast	1.14	31.33	1	31.33	0.10	0.7490	
E-Ferm. Temp	2.42	140.65	1	140.65	0.47	0.4993	
F-pH	9.63	2226.07	1	2226.07	7.43	0.0113	
AB	4.55	165.53	1	165.53	0.55	0.4639	
AC	4.76	180.88	1	180.88	0.60	0.4441	
AD	-2.37	89.87	1	89.87	0.30	0.5886	
AE	-2.75	60.45	1	60.45	0.20	0.6570	
AF	-12.97	1346.29	1	1346.29	4.49	0.0437	
BC	7.20	414.29	1	414.29	1.38	0.2503	
BD	3.93	123.48	1	123.48	0.41	0.5265	
BE	-4.77	363.95	1	363.95	1.21	0.2805	
BF	-6.42	329.99	1	329.99	1.10	0.3036	
CD	-14.38	1653.70	1	1653.70	5.52	0.0267	
CE	-0.46	1.68	1	1.68	5.620E-03	0.9408	
CF	-0.13	0.26	1	0.26	8.513E-04	0.9769	
DE	12.33	1216.48	1	1216.48	4.06	0.0543	
DF	-5.85	273.78	1	273.78	0.91	0.3479	
EF	8.97	643.69	1	643.69	2.15	0.1547	
A ²	12.12	1512.13	1	1512.13	5.05	0.0334	
B ²	-6.92	492.21	1	492.21	1.64	0.2112	
C ²	4.97	254.56	1	254.56	0.85	0.3651	
D ²	-9.60	948.45	1	948.45	3.17	0.0869	
E ²	8.24	698.35	1	698.35	2.33	0.1389	
F ²	-1.45	21.74	1	21.74	0.073	0.7897	
Residual		7788.91	26	299.57			
Lack of Fit		7268.22	21	346.11	3.32	0.0932	Not significant
Pure Error		520.69	5	104.14			
Cor Total		33162.02	53				

Model summary statistics:-

The summary of the statistical analyses are shown in Tables 6 and 7 below.

Table 6:- Model summary statistics

Source	Std. Dev.	R-Squared	Adjusted R-Squared	Predicted R-Squared	PRESS	
Linear	19.42	0.4654	0.3972	0.2770	23975.89	Suggested
2FI	18.42	0.6724	0.4575	-0.0657	35340.10	
Quadratic	17.31	0.7651	0.5212	-0.1677	38722.52	
Cubic	13.99	0.9528	0.6873	-7.0869	2.682×10^5	Aliased

From Table 6, the choice of model was based on the Predicted R-Squared value. A positive value of Predicted R-Squared is more preferable to a negative term, hence the Cubic model was aliased (i.e ruled out) while the Linear model was suggested. However, the Two-Factor Interaction (2FI) and Quadratic models were not aliased because their Predicted R-Squared values were not too negative, hence the Quadratic model was selected.

Table 7:- Adeq Precision and various R– Squared values for quadratic model

Std. Dev.	17.31	R-Squared	0.7651
Mean	72.57	Adj R-Squared	0.5212
C.V. %	23.85	Pred R-Squared	-0.1677
PRESS	38722.52	Adeq Precision	8.309

The adequate precision and various R–Squared values for the quadratic model are given in Table 8. The "Adeq Precision" is a measure of the signal to noise ratio and a ratio greater than 4 is desirable. For the quadratic model, the "Adeq Precision" is 8.309. Since it is greater than 4, the model can be used to navigate the design space, but there is need for improvement, through the removal of insignificant model terms.

Confidence interval:-

The confidence interval (CI) depicted in Table 8 is another statistical data that can be used to determine whether a model term has effect (i.e. significant) or not on the model). Hence, the terms in the columns “**95% CI Low and 95% CI High**” represent the range that the true coefficient should be found in 95% of the time. If the range spans zero (i.e. one limit is positive and the other negative) then the coefficient would not be true, indicating that the corresponding term or factor has no effect. However, if the range does not span zero (i.e. both limits having the same sign, either positive or negative) then the coefficient would be true, meaning that the corresponding term or factor has effect. For example, the term B has a confidence interval which spans 0 since it is between -6.80 and 7.72, hence term B has no effect. Whereas term A has a confidence interval which does not span 0 since it is between -30.48 and -15.96, as such it has significant effect on the model equation. Similarly, the confidence intervals of the following terms A, F, AF, CD, A^2 as well as the intercept do not span 0 and therefore have significant effects on the model equation.

Table 8:- Confidence interval, degree of freedom and variance inflation factor

Factor	Coefficient Estimate	Degree of Freedom	Standard Error	95% CI Low	95% CI High	VIF
Intercept	69.30	1	7.07	54.77	83.82	
A-H ₂ SO ₄	-23.22	1	3.53	-30.48	-15.96	1.00
B-Hydrolysis Time	0.46	1	3.53	-6.80	7.72	1.00
C-Fermentation Time	1.92	1	3.53	-5.34	9.19	1.00
D-Conc. of Yeast	1.14	1	3.53	-6.12	8.40	1.00
E-Fermentation Temp	2.42	1	3.53	-4.84	9.68	1.00
F-pH	9.63	1	3.53	2.37	16.89	1.00
AB	4.55	1	6.12	-8.03	17.13	1.00
AC	4.76	1	6.12	-7.82	17.33	1.00
AD	-2.37	1	4.33	-11.26	6.52	1.00
AE	-2.75	1	6.12	-15.33	9.83	1.00
AF	-12.97	1	6.12	-25.55	-0.39	1.00

BC	7.20	1	6.12	-5.38	19.77	1.00
BD	3.93	1	6.12	-8.65	16.31	1.00
BE	-4.77	1	4.33	-13.66	4.12	1.00
BF	-6.42	1	6.12	-19.00	6.16	1.00
CD	-14.38	1	6.12	-26.96	-1.80	1.00
CE	-0.46	1	6.12	-13.04	12.12	1.00
CF	-0.13	1	4.33	-9.02	8.77	1.00
DE	12.33	1	6.12	-0.25	24.91	1.00
DF	-5.85	1	6.12	-18.43	6.73	1.00
EF	8.97	1	6.12	-3.61	21.55	1.00
A ²	12.12	1	5.40	1.03	23.22	1.30
B ²	-6.92	1	5.40	-18.01	4.18	1.30
C ²	4.97	1	5.40	-6.12	16.07	1.30
D ²	-9.60	1	5.40	-20.70	1.49	1.30
E ²	8.24	1	5.40	-2.85	19.33	1.30
F ²	-1.45	1	5.40	-12.55	9.64	1.30

VIF: Variance Inflation Factor in Table 8, measures how much the variance of the model is inflated by the lack of orthogonality in the design. If the factor is orthogonal to all other factors in the model, the VIF is one. Values greater than 10 indicate that the factors are too correlated together (they are not independent.). Thus, since the values of Variance Inflation Factors range from 1.00 to 1.30, it indicates that the factors or terms are independent.

Standard Error: The Standard Error in Table 8 is associated with the calculation of the mean. It comes from the standard deviation of the data divided by the square root of the number of repetitions in a sample. The larger the value of the Standard Error of a term the more insignificant is the term.

Diagnostics Case Statistics:-

The actual value, predicted value of bioethanol and their residuals are given in Table 9, which informed the plots in Figures 1-5.

Table 9:- Internally and Externally Studentized Residual and Influence on Fitted Value

Std Order	Actual Value	Predicted Value	Residual	Leverage	Internally Studentized Residual	Externally Studentized Residual	Influence on Fitted Value	Cook's Distance	Run Order
1	87.93	92.63	-4.70	0.563	-0.411	-0.404	-0.458	0.008	38
2	61.26	41.83	19.43	0.563	1.697	1.765	* 2.00	0.132	21
3	76.38	76.59	-0.21	0.563	-0.019	-0.018	-0.021	0.000	44
4	41.44	43.98	-2.54	0.563	-0.222	-0.218	-0.247	0.002	29
5	86.19	91.80	-5.61	0.563	-0.490	-0.483	-0.547	0.01	17
6	39.88	31.52	8.36	0.563	0.731	0.724	0.821	0.025	30
7	63.89	91.47	-27.58	0.563	-2.409	-2.681	* -3.04	0.267	5
8	62.24	49.39	12.85	0.563	1.123	1.129	1.280	0.058	43
9	68.01	72.76	-4.75	0.562	-0.415	-0.408	-0.463	0.008	49
10	72.86	68.82	4.04	0.562	0.353	0.347	0.393	0.006	19
11	66.77	63.13	3.64	0.562	0.318	0.312	0.354	0.005	24
12	83.02	87.98	-4.96	0.562	0.433	-0.426	-0.483	0.009	42
13	88.68	88.06	0.62	0.562	0.054	0.053	0.061	0.000	28
14	65.74	65.04	0.70	0.562	0.061	0.060	0.068	0.000	6
15	68.22	76.59	-8.37	0.562	-0.731	-0.725	-0.822	0.025	36
16	91.45	82.36	9.09	0.562	0.794	0.788	0.893	0.029	54
17	33.12	30.16	2.96	0.563	0.258	0.254	0.287	0.003	52
18	69.56	63.02	6.54	0.563	0.571	0.564	0.639	0.015	15
19	66.73	72.90	-6.17	0.563	-0.539	-0.532	-0.603	0.013	31

20	57.33	48.25	9.08	0.563	0.793	0.787	0.893	0.029	26
21	65.56	61.38	4.18	0.563	0.365	0.359	0.407	0.006	35
22	86.64	93.73	-7.09	0.563	-0.619	-0.612	-0.694	0.018	18
23	87.44	80.72	6.72	0.563	0.587	0.580	0.657	0.016	4
24	39.34	55.56	-16.22	0.563	-1.417	-1.446	-1.640	0.092	16
25	111.87	106.93	4.94	0.563	0.431	0.425	0.481	0.009	33
26	68.29	70.72	-2.43	0.563	-0.213	-0.209	-0.236	0.002	22
27	114.23	89.29	24.94	0.563	2.178	2.362	* 2.68	0.218	48
28	39.74	43.61	-3.87	0.563	-0.338	-0.332	-0.376	0.005	46
29	88.32	92.61	-4.29	0.563	-0.374	-0.368	-0.417	0.006	14
30	28.62	45.40	-16.78	0.563	-1.466	-1.501	-1.702	0.099	45
31	134.88	124.29	10.59	0.563	0.925	0.922	1.045	0.039	32
32	54.52	67.61	-13.09	0.563	-1.144	-1.151	-1.305	0.060	8
33	66.13	54.43	11.70	0.563	1.022	1.023	1.160	0.048	47
34	73.76	77.73	-3.97	0.563	-0.347	-0.341	-0.387	0.006	51
35	53.36	50.87	2.49	0.563	0.217	0.213	0.242	0.002	11
36	81.88	55.10	26.78	0.563	2.340	2.582	* 2.93	0.251	10
37	37.48	68.60	-31.12	0.563	-2.718	-3.151	* -3.57	0.339	12
38	68.06	66.21	1.85	0.563	0.162	0.159	0.180	0.001	50
39	109.23	100.92	8.31	0.563	0.726	0.719	0.815	0.024	23
40	63.42	79.45	-16.03	0.563	-1.400	-1.428	-1.619	0.090	41
41	79.25	88.27	-9.02	0.563	-0.788	-0.782	-0.886	0.028	20
42	51.69	58.26	-6.57	0.563	-0.574	-0.566	-0.642	0.015	40
43	68.14	82.86	-14.72	0.563	-1.285	-1.303	-1.477	0.076	25
44	52.77	71.87	-19.10	0.563	-1.668	-1.731	-1.963	0.128	2
45	139.56	133.73	5.83	0.563	0.510	0.502	0.569	0.012	34
46	53.28	51.83	1.45	0.563	0.127	0.125	0.141	0.001	3
47	147.64	127.81	19.83	0.563	1.732	1.806	* 2.05	0.138	39
48	87.21	64.93	22.28	0.563	1.946	2.065	* 2.34	0.174	13
49	67.33	69.30	-1.97	0.167	-0.124	-0.122	-0.055	0.000	17
50	88.76	69.30	19.46	0.167	1.232	1.245	0.557	0.011	27
51	58.14	69.30	-11.16	0.167	-0.706	-0.699	-0.313	0.004	1
52	66.89	69.30	-2.41	0.167	-0.152	-0.149	-0.067	0.000	9
53	67.33	69.30	-1.97	0.167	-0.124	-0.122	-0.055	0.000	37
54	67.33	69.30	-1.97	0.167	-0.124	-0.122	-0.055	0.000	53

* Exceeds limits

Diagnostic plots:-

Important statistical plots derived from the analyses of the data are shown in Figures 1-5 below:

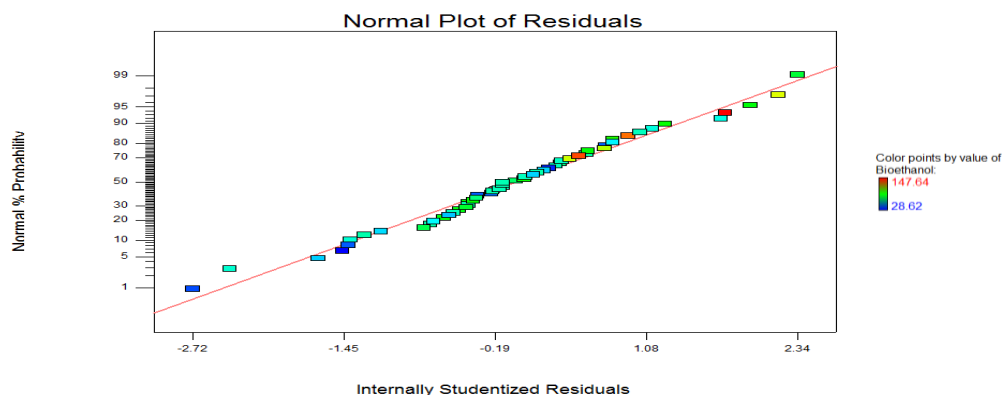


Figure 1:- Normal probability plot of the studentized residuals.

The normal probability plot of the studentized residuals in Figure 1, follows a linear pattern, which indicates a normal distribution of the residuals, hence a transformation is not required. This is in line with Hothorn, and Everitt (2014), who reported that a non-linear pattern indicates that the residuals did not follow a normal distribution, in which case a transformation of the response may provide a better analysis.

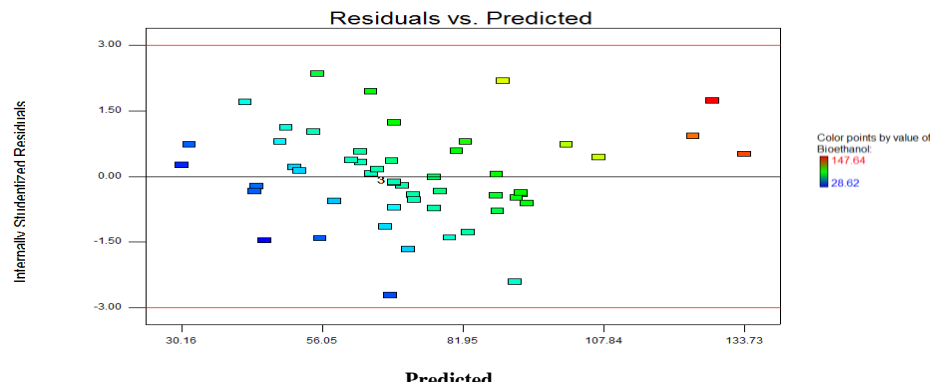


Figure 2:- Studentized residuals versus predicted values plot.

The studentized residuals versus predicted values plot, Figure 2, is a plot of the residuals versus the ascending predicted response values. It tests the assumption of constant variance. A random scatter (constant range of residuals across the graph.) is acceptable while an expanding variance ("megaphone pattern <") indicates the need for a transformation (Engle and Sheppard 2001). Since the plot showed a random scatter, it indicates that there was no assumption of a constant variance, thus, transformation is not required in the data so analyzed.

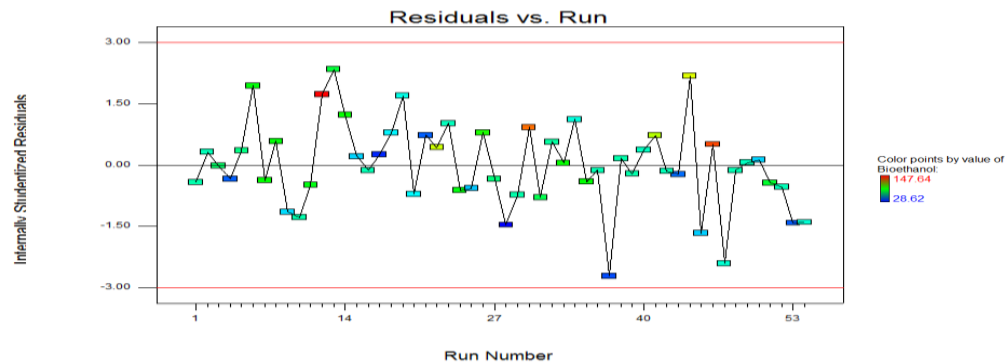


Figure 3:- Internally studentized residuals versus run number

The internally studentized residuals versus run plot, Figure 3, show that there were no lurking variables that may have influenced the bioethanol responses, since it followed a random scatter. According to Engle and Sheppard (2001), internally studentized residuals versus run plot which results in a trend will normally indicate the presence of lurking variables which may have influenced the responses and as such a transformation would be required.

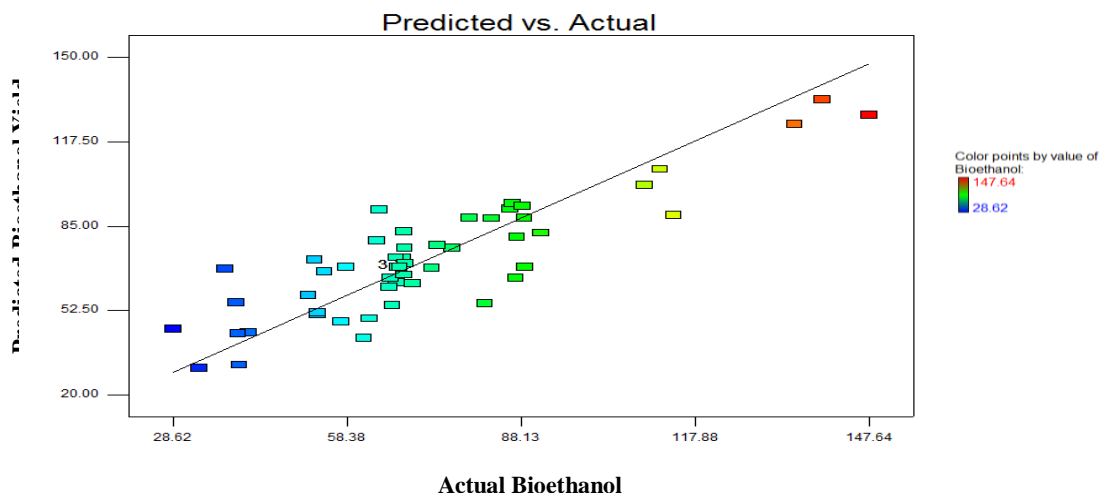


Figure 4:- Predicted bioethanol yield versus actual bioethanol yield plot.

The predicted bioethanol yield versus actual bioethanol yield plot, Figure 4, shows points clustering along a linear graph which indicates that there are no values that would not be easily predicted by the model. A predicted response versus actual response plot helps detect a value, or group of values, that are not easily predictable by a model. Thus, the clustering of the points around a line indicates parity between the predicted and actual data being analyzed; hence the model can easily be used to predict a value of a response when the actual value is known (Cao *et al.*, 2009).

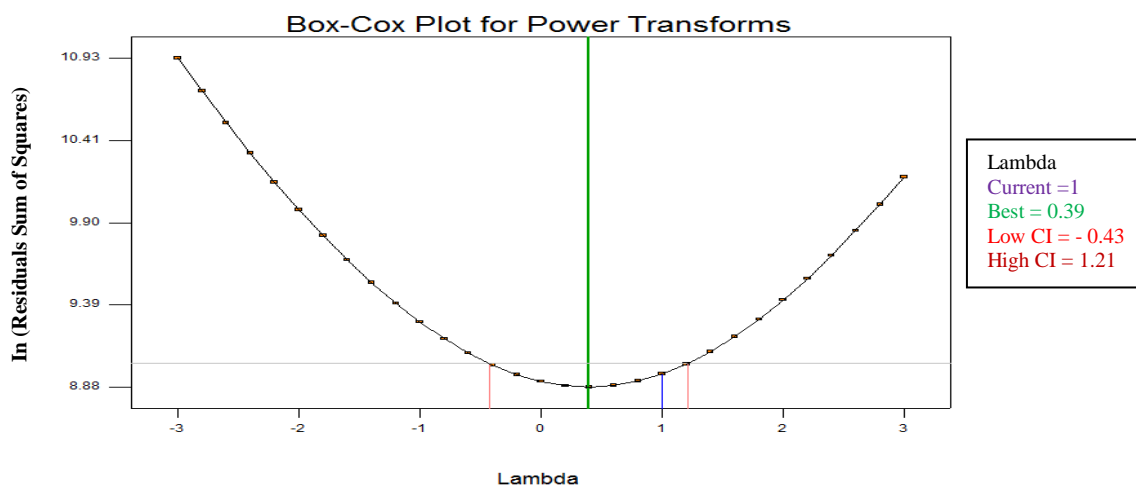


Figure 5:- Box-Cox plot for power transformations.

The Box Cox Plot, Figure 5, gave the best lambda value of 0.39 corresponding to the minimum point of the curve generated by the natural log of the sum of squares of the residuals. It was also observed that the 95% confidence interval around this best lambda value, includes 1, therefore no specific transformation is recommended for the data that was analyzed, based on the recommended transformation list in Box and Cox, (1964).

Conclusion:-

The Design Expert software and its several statistical tools are dedicated to designing experiments involving multivariable factors and analyzing the main effects of each factor as well as the interactions between factors. Thus in this paper it has been employed to determine statistical significance of the various process factors in bioethanol production from corn-stover, their impact on the response variable (bioethanol yield), diagnostic capacities to validate statistical assumptions, models and possible response transformations.

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RESEARCH ARTICLE

EFFECT OF TEMPERATURE AND IRRADIANCE ON THE ELECTRICAL PERFORMANCE OF A PV MODULE.

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Abstract

Solar cells are the devices, which convert the solar radiation into the electrical energy without effecting the environment. To predict the real behavior of solar cells like changes in maximum power, short circuit current and open circuit voltage by changing the temperature and irradiance can be studied by using the MATLAB (simulation model). In this paper, a single diode MATLAB model is used to study the changes in solar PV module by varying temperature (100C, 200C, 300C) and by varying the irradiance (400W/m², 600W/m², 800W/m²). To study the changes in electrical parameters of a solar PV modules KD330GX-LFB and KD325GX-LFB experimental data sheet are taken and the results are discussed in the conclusion.

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Introduction:-

The continuous use of fossil fuels for industrial purpose leads to huge damage to eco system and living conditions. To sustain our earth, the mankind has to decrease the green house gases like CO₂, Methane.., by choosing alternatives to fossil fuels. In order to conserve natural living spaces and not to harm the next generation utilization of renewable energy should increase. Among all the renewable energy sources, solar energy is having the large potential and small amount of it is utilizing for various purposes. Photovoltaic Effect converts solar energy to the electrical energy. Widely used applications of PV system are water heaters, street lights, electric vehicles, military and space applications(1,2). Solar panels and modules are works on the principle of Photo voltaic effect, and generates the electric power. These PV modules and panels are largely effected by different weather conditions. In this paper, mainly modeling parameters are explained. By changing the temperature of a simulink PV module, the changes in output parameters are observed. The simulated results are compared with the KD330GX-LFB, KD325GX-LFB experimental results.

State of Art: Swapnil Dubey.et.al, investigated the temperature Effect on the solar panel. they concluded that numerical parameters or equations are material dependent and also system dependent (3). Ike and C.U. investigated the effect of ambient temperature on performance. They observed the behavior of solar PV system under dry and rainy season in the year 2013. Conclusions came out as, the installation conditions shows the effect on the performance of PV panel(4). Khaled Matter, Hala.J.E-Khozondar.et.al. studied the shading effects on the maximum power point and remarkable reduction in efficiency of the solar PV system. Conclusions are there is a reduction in the maximum power point, by increasing temperature. To study the shading effects on the performance, they used the KC200GT panel(5).

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Tarak Salmi , Mounir Bouzguend, Adel Gastli, Ahmed Masmoudi developed solar PV module using basic semiconductor equations and they studied the performance of solar PV module under various atmospheric conditions(6). Jay Patel and Gaurang Sharma studied the characteristics of a module, they observed the solar cell performance under various temperature and irradiance conditions(7).

Characteristic I-V equation:- The I-V equation of a solar cell, which represents the output of a solar cell is described by few researchers from past four decades(8). The solar cells is generally represented by the single diode due to its simplicity in computational time. The Single diode solar cell can be modeled using the electrical components current source, parallel resistor, series resistor.

The single diode photovoltaic I-V equation (8):

$$I = I_{ph} - I_0 \left\{ \exp \left(\frac{q(V + IR_s)}{A.K.T_c} \right) \right\}$$

$$I_{ph} = G [I_{sc} + K_1 (T_c + T_{ref})]$$

Where

I_{ph} =Photo current

I_0 =Saturation Current

R_s =Series resistance

A =Ideality factor

$K=1.38 \times 10^{-23} \text{ W/m}^2 \cdot \text{K}$ (or) $8.617 \times 10^{-5} \text{ eV/K}$.

I_{sc} =Short circuit current.

Photo current (I_{ph}): The photo current is the electricity generated by the solar radiation and I_{ph} , explicitly depends upon cell temperature(T). Habbati. Bellia.et.al. Presented the detailed I_{ph} equation as (9).

$$I_{ph} = \frac{G}{G_{ref}} [I_{ph} + \mu_{sc} \cdot \Delta T].$$

$$\Delta T = T_c - T_{c,ref}$$

Here,

G = Irradiance(W/m^2)

G_{ref} =Irradiance at STC =1000w/m²

$T_{c,ref}$ = cell temperature at STC:25+273

μ_{sc} =Coefficient of temperature of short circuit current (A/K).

Reverse Saturation Current: In solar cell, the reverse saturation current is produced due to the diffusion of charges from n-side to p-side and p-side to n-side. The equation for I_0 is given from the literature as

$$I_0 = DT_c 3 \exp \left(\frac{-q\epsilon_g}{A.K} \right)$$

T_c = Standard temperature.

Ideality factor: The ideality factor gives us how closely the diode follows the ideal diode equation, the ideality factor is material dependent Some of the researchers like Bashahu and Nkunbabakura(12) used different methods to identify, ideality factor and suggested that it lies between 1.26 to 1.5 and 'n' is assumed to be independent of temperature and solar radiation(10).

Series Resistance(R_s):The movement of charges through the emitter and base, the contact resistances are the main reason for series resistance.

Series resistance is defined as:

$$R_s = -\left(\frac{dv}{di}\right) - \frac{1}{X}$$

Shunt Resistance: Manufacturing defects and poor solar cell design are the main reason for shunt resistance (R_{sho}), leads to power losses by flowing the light generated current in an alternative path. The deviation of current flow through the junction leads to decrease in the voltage of the cell.

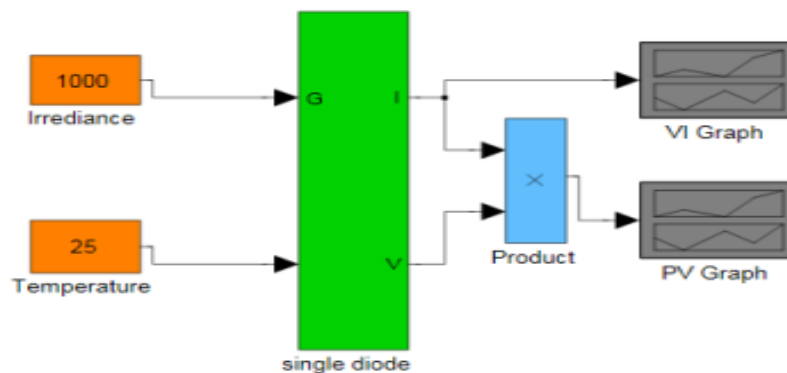
The shunt resistance is defined as :

$$R_s = R_{sho}$$

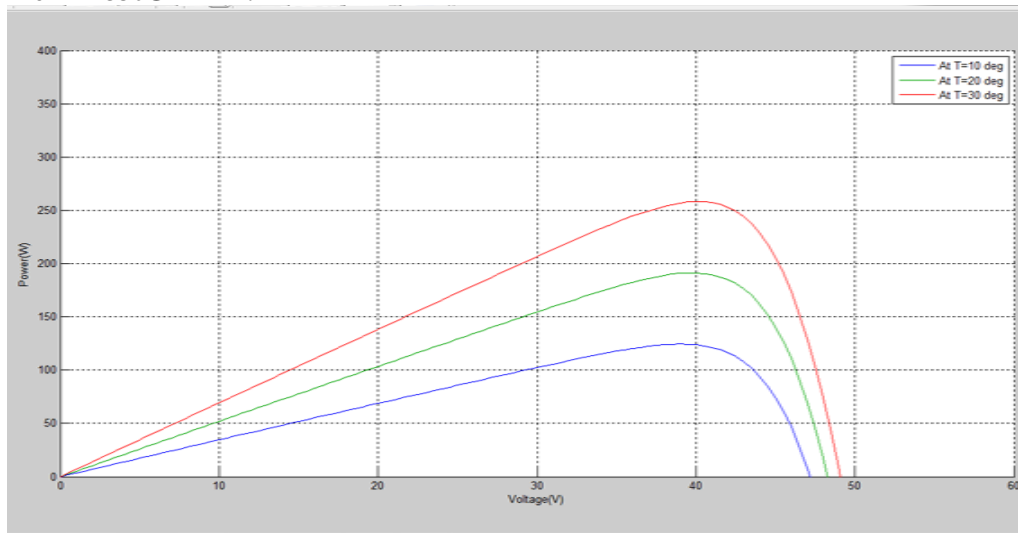
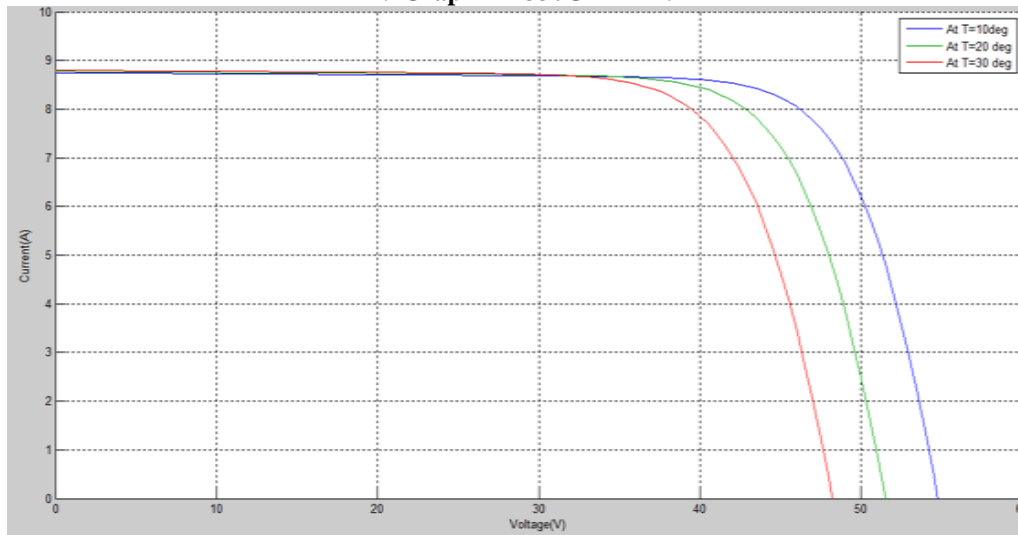
$$R_{sho} = -\left(\frac{dv}{di}\right) \quad (13,14)$$

Simulink single diode model: To understand the effect of atmospheric variations on the performance of solar PV module, we need a computer based model. For this purpose, by using the semi conductor basic equations, a simulated single diode model is developed. By using that model, and changing the input parameters those are temperature and irradiance, we can observe the changes in the like P_{max} , V_{mp} , I_{mp} , V_{oc} , I_{sc} from I-V, P-V graphs and the following observations are tabulated.

Single Diode Model: Simulink model consists of few subsystems inter connected and the output is linked to the multi plot graphs (I-V and P-V graphs). The following figure represents the generalized model of PV module can be compared with the any practical device.



By varying the temperature: One of the input parameter Temperature change shows the major influence on the Performance of the module. The effect of temperature on KYOCERA KD330GX-LFB is observed by varying the temperature from 10°C to 30°C , by an increment of 10°C . The corresponding changes are tabulated and compared with the practical values. The resistances are identified for the simulink model by the Newton_Raphson numerical method. The resistances are $R_s=0.2500\text{ohm}$ and $R_p=493.206\text{ohm}$

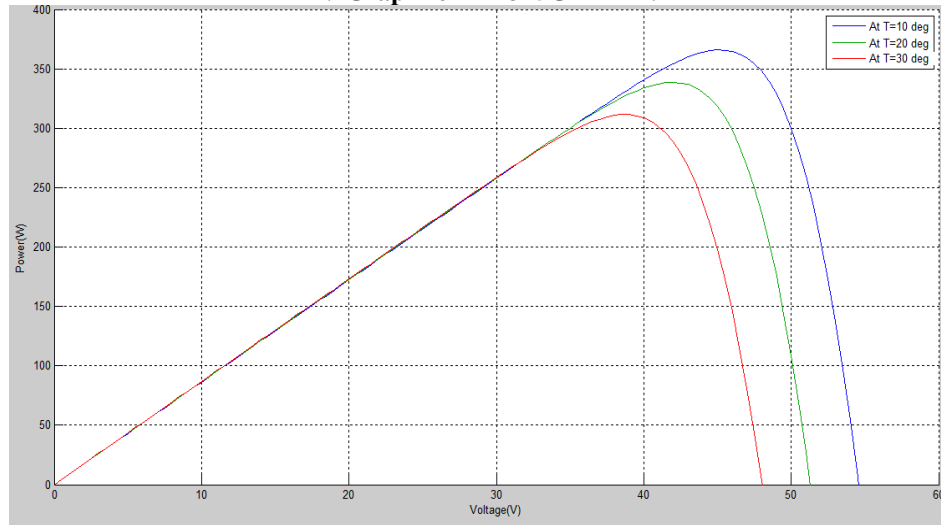
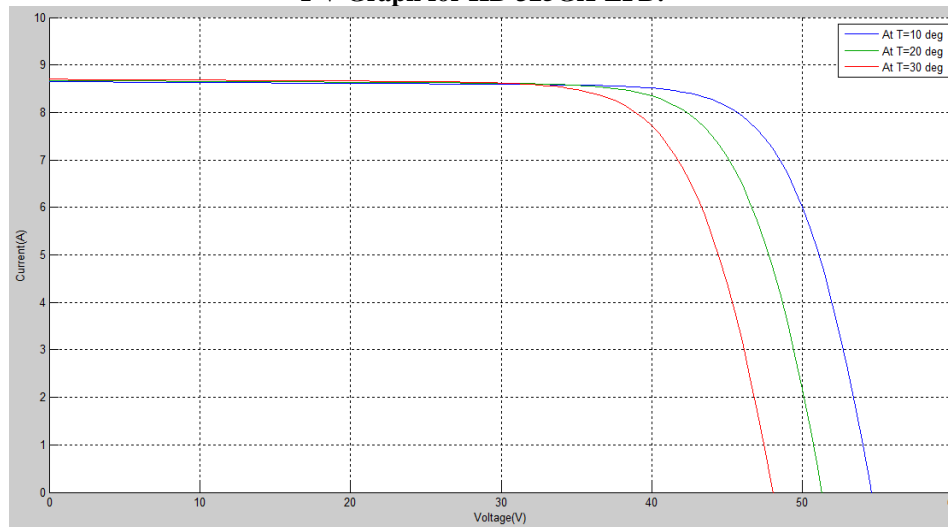
P-V Graph for KD330GX-LFB:-**I-V Graph KD330GX-LFB:****Table-1:-**

Parameters of KD330GX-LFB (At $G=1000\text{W/m}^2$ and $T=25^\circ\text{C}$)	Simulink model Parameters at Different Temperatures		
	$T=10^\circ\text{C}$	$T=20^\circ\text{C}$	$T=30^\circ\text{C}$
$P_{\text{max}}=330(\text{W})$	371	343.4	316.2
$V_{\text{mp}}=40.5(\text{V})$	45.5	42.5	39
$I_{\text{mp}}=8.15(\text{A})$	8.15	8.08	8.11
$V_{\text{oc}}=49.9(\text{V})$	54.8	51.6	48.2
$I_{\text{sc}}=8.79(\text{A})$	8.75	8.77	8.80

Change of Efficiency with temperature:

Table-2:-

Temperature ($^\circ\text{C}$)	Fill Factor (%)	Efficiency (%)
T=10	77.33	17.57
T=20	75.85	16.27
T=30	74.56	14.99

P-V Graph for KD325GX-LFB:**I-V Graph for KD 325GX-LFB:****Table-3:-**

Parameters of KD 325GX-LFB At $G=1000\text{W/m}^2$ and $T=25^\circ\text{C}$	Simulink model Parameters at different Temperatures		
	$T=10^\circ\text{C}$	$T=20^\circ\text{C}$	$T=30^\circ\text{C}$
Pmax=325(W)	365.8	338.7	311.4
Vmp=40.3(V)	45	42	39
Imp=8.07(A)	8.13	8.06	7.98
Voc= 49.7(V)	54.6	51.3	48
Isc=8.69(A)	8.65	8.68	8.70

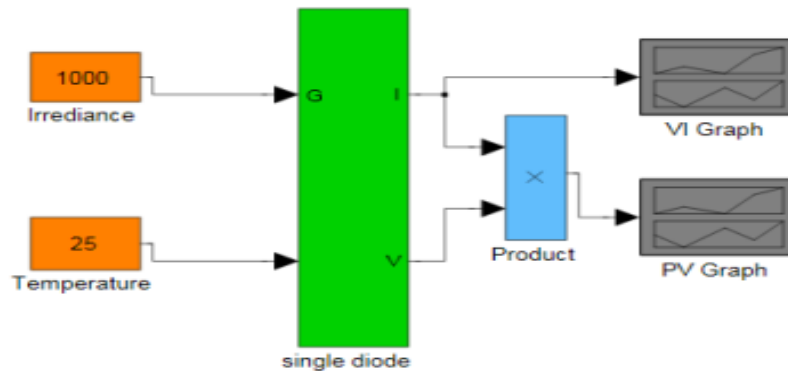
Table-4:- Change of Efficiency with Temperature:

Temperature ($^\circ\text{C}$)	Fill Factor(%)	Efficiency (%)
T=10	77.46	17.33
T=20	75.02	15.05
T=30	74.52	14.75

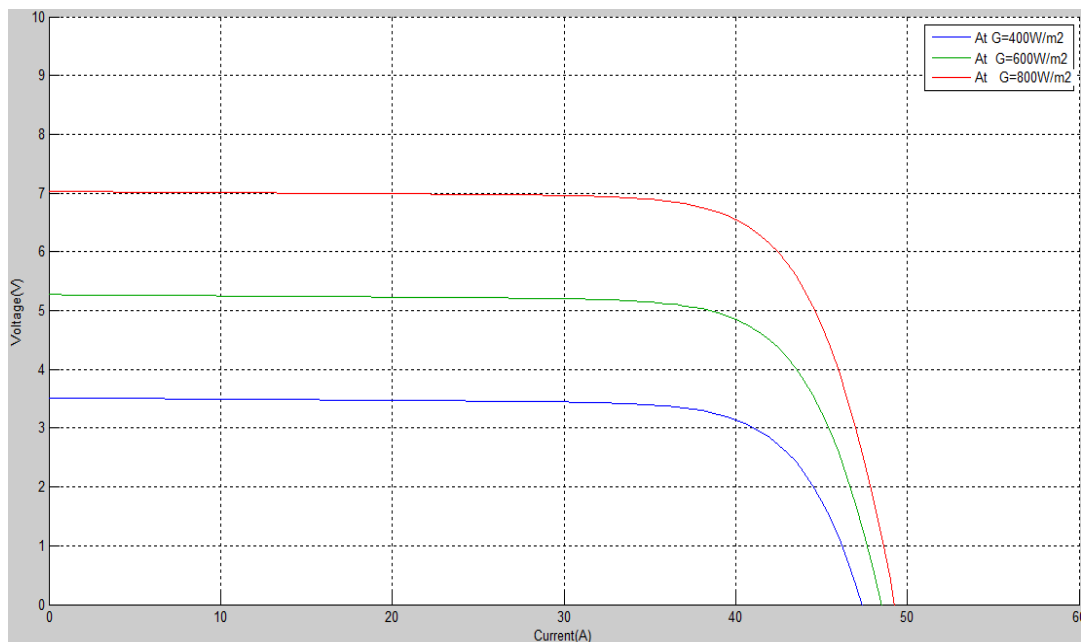
The temperature change of KYOCERA KD330GX-LFB is observed for the temperatures 10°C , 20°C , 30°C and the changes in P_{max} , V_{mp} , I_{mp} , V_{oc} , I_{sc} are taken from the I-V, P-V graphs and those values are tabulated. The resistances for KD330GX-LFB simulink model are identified using the Newton_Rapshon method those values are $R_p=493.206\ \Omega$ and $R_s=0.2500$.

By Varying Irradiance: Solar module efficiency depends upon the input variable irradiance, as the irradiance is changed from 400W/m^2 to 800W/m^2 the output efficiency of a module changes. To study the changes in the performance of a solar PV module, KD330GX-LFB and KD325GX-LFB are studied and the results are tabulated.

Simulink model of Single Diode model:



I-V Graph for KD330GX-LFB:-



P-V Graph for KD330GX-LFB:

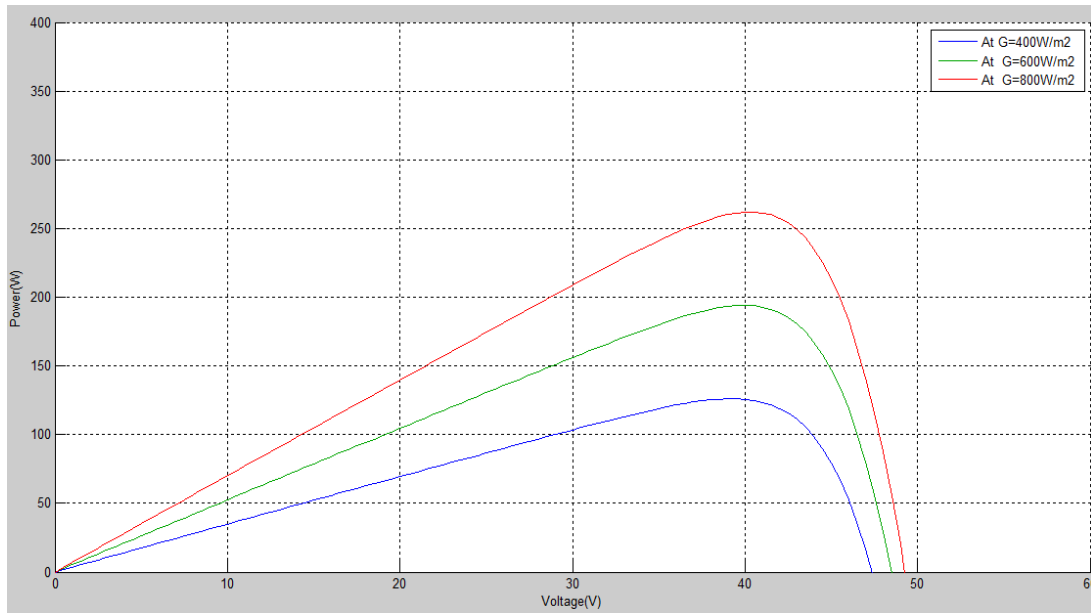


Table:5:-

Parameters of KD330GX-LFB At $G=1000\text{W/m}^2$ and $T=25^\circ\text{C}$	Simulink model Parameters Under varying irradiance(W/m^2)		
	G=400	G=600	G=800
Pmax=330W	125.7	194	262
Vmp=40.5V	40	40	40.5
Imp=8.15A	3.14	4.85	6.47
Voc=49.9V	47.3	48.5	49.3
Isc=8.79A	3.51	5.27	7.02

Table-6:- Change of Efficiency with Irradiance:

Irradiance (W/m^2)	Fill Factor (%)	Efficiency (%)
G=400	75.65	14.69
G=600	75.90	15.1
G=800	75.71	15.5

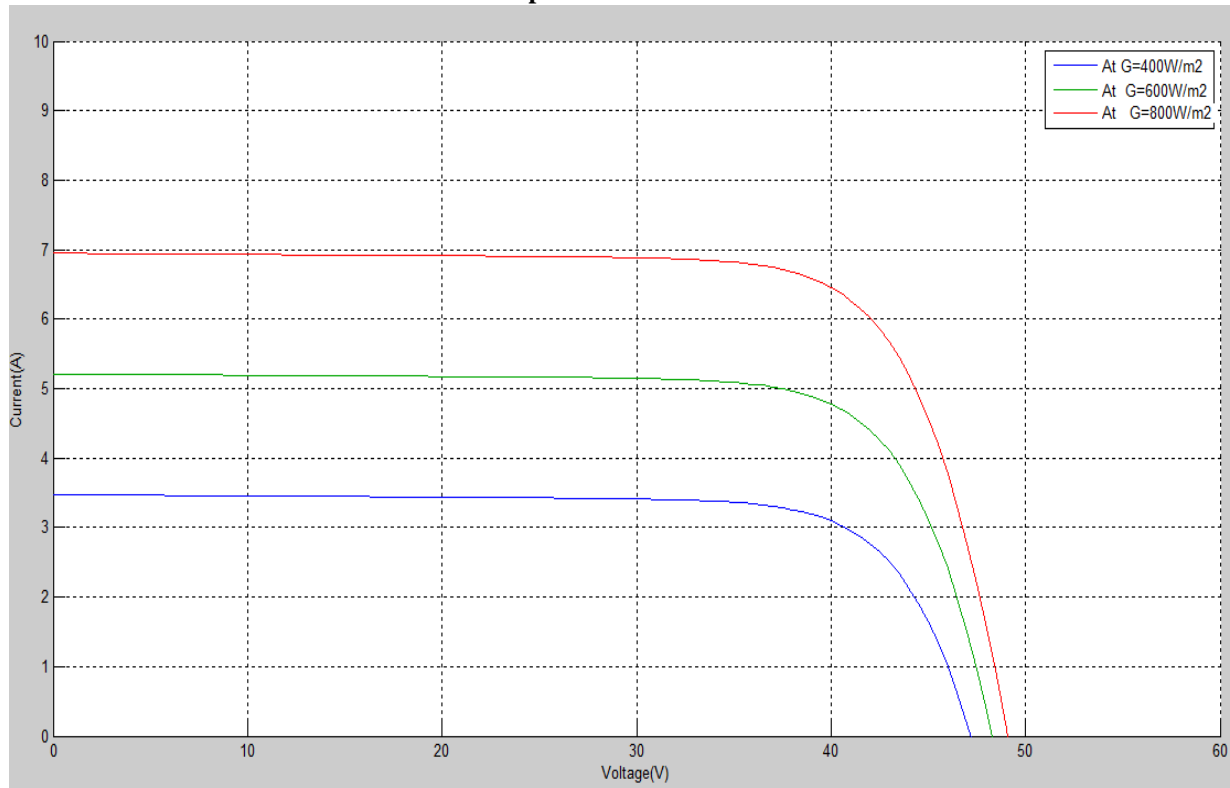
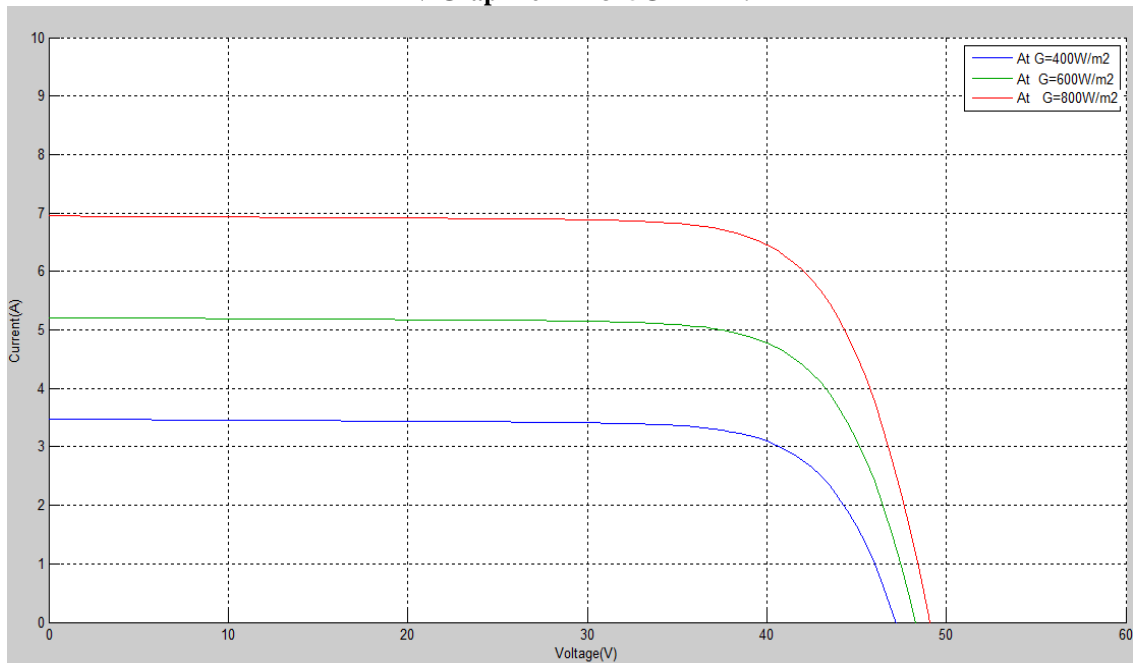
I-V Graph for KD325GX-LFB:**P-V Graph for KD325GX-LFB:**

Table-7:

Parameters of KD325GX-LFB	Simulink model parameters Under varying irradiance(W/m ²)		
	G=400	G=600	G=800
Pmax=325W	123.8	191.2	258.1
Vmp=40.3V	40	40	40.5
Imp=8.07A	3.1	4.78	6.38
Voc=49.7V	47.2	48.3	49.1
Isc=8.69A	3.47	5.21	6.95

Change of Efficiency with Irradiance:

Table 8:-

Irradiance(W/m ²)	Fill Factor(%)	Efficiency(%)
G=400	75.65	14.88
G=600	75.90	15.32
G=800	75.71	15.54

Constant Parameters used for Simulation:

Ideality factor (a)=1.3

Number of cells(Ns)=80

Boltzmann constant(K)= 1.38×10^{-23} W/m².K

Energy gap(E.g.)=1.12.

Electron charge(q)= 1.602×10^{-19} C**Conclusion:-**

From Table-1, Table-3 as the temperature increases maximum power, open circuit voltage decreases and short circuit current increases ultimately efficiency decreases. Table -2 and Table-4 represents the change in efficiency and fill factor, from those we can conclude that as the temperature increases the efficiency decreases, this is because of increased intrinsic recombination. By the increase of heat energy, band gap of semi conductor decreases the open circuit voltage which is a function of band gap, decreases faster than the increase in the short circuit current so it leads to the decrease in the efficiency. The negative temperature coefficient of Silicon material reveals that as the temperature increases, the maximum power output decreases. The Table-5 and Table-7 represents the changes in the parameters of solar cell with the change in the irradiance. The Table-6 and Table-8 represents the changes in the efficiency and fill factor with the changes in the irradiance. As the irradiance increases, the number of photons incidenting on the module produces maximum power and leads to increase in the efficiency. This generalized simulink model is used to study the changes in the output parameters of any photovoltaic module by using Newton-Raphson numerical method to identify the suitable resistances.

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RESEARCH ARTICLE

BACTERIAL SPECTRUM IN EXACERBATION COPD PATIENTS.

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Key words:-

COPD, exacerbation, bacteria.

Abstract

The precise role of bacterial infection in the course of exacerbation COPD has been a source of controversy for decades. Chronic bacterial colonization of the lower airways contributes to airway inflammation; The course of COPD is characterized by intermittent exacerbations of the disease. Approximately 40-50% acute exacerbations are caused by bacteria, by viruses in 25%, and both viruses and bacteria in another 25%. Airway inflammation is increased during the exacerbation resulting in increased hyperinflation, reduced expiratory air flow and decreased gas exchange. Expectored sputum is the most commonly used sample for diagnosis of lower respiratory tract infections (LRTI), which can be obtained easily and non-invasively.

The bacteria are very common in the lower respiratory tract of patients with COPD and cause inflammation. We have shown that among microorganisms causing acute exacerbations of COPD in our patients prevailed gram negative Enterobacteriaceae family and comparatively high resistance against cephalosporins and fluoroquinolones related to wide usage of antibiotics of this group for chemoprophylaxis.

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Introduction:-

Chronic obstructive pulmonary disease (COPD) is a major cause of chronic morbidity and mortality worldwide. The World Health Organization predicts that COPD will become the third leading cause of death worldwide by 2030. (1,2). Exacerbation of COPD is redefined as a sustained worsening of a patient's condition from a stable state. The precise role of bacterial infection in the course of exacerbation COPD has been a source of controversy for decades. Chronic bacterial colonization of the lower airways contributes to airway inflammation; The course of COPD is characterized by intermittent exacerbations of the disease. Approximately 40-50% acute exacerbations are caused by bacteria, by viruses in 25%, and both viruses and bacteria in another 25%. Airway inflammation is increased during the exacerbation resulting in increased hyperinflation, reduced expiratory air flow and decreased gas exchange. (3,4,5).

Bacteria have established niches in the human body where they exist in symbiosis with their host. The term microbiome is used to refer to complex communities of microorganisms, termed the microbiota, that inhabit the body surfaces in symbiosis with the host. The most clearly defined microbiome occurs in the gastrointestinal tract in

which there are far more bacterial cells than in the whole human body (as bacteria are much smaller than human cells). Bacteria which form part of the normal flora of the microbiome, are tolerated by the local mucosal immune response with minimal activation of inflammatory cells, in particular T cells. The lower respiratory tract is directly connected to the upper respiratory tract with frequent micro-aspiration of secretions.(5,6,7).

Expectorated sputum is the most commonly used sample for diagnosis of lower respiratory tract infections(LRTI),which can be obtained easily and non-invasively.However, sputum may show a heavy growth of commensal organisms because of the necessity to traverse a highly contaminated oropharynx which may prevent the determination of true epidemiologic agent. A potentially-photogenic microorganism(PPM) had to grow in significant counts to be considered a potential causative agent of an exacerbation. (8,9)

Major challenges remain in accurately defining the potential role of bacteria in the inflammatory process and how best to optimize the use of antibiotics without the overuse of this limited resource. The thresholds for positive cultures used in most of the recently published studies are as follows: $\geq 10^2$ or $\geq 10^5$ colony-forming units/millilitre (CFU/ml) for sputum, $\geq 10^2$ CFU/ml for bronchial lavage (BL) and $\geq 10^2$ or $\geq 10^3$ CFU/ml for bronchoscopic protected specimen brush (PSB) and bronchoalveolar lavage (BAL) samples. Considering bacterial exacerbations, Gram-negative bacteria such as *P. aeruginosa*, *Stenotrophomonas maltophilia* and members of the *Enterobacteriaceae* family are more often present in patients with a greater degree of functional impairment, recent antibiotic or systemic steroid therapy, and in those with severe exacerbations. The rates of positive cultures in COPD vary depending on the sampling technique .(10)

The aim of the study was the bacteriological examination of expectorated sputum taken from COPD patients during exacerbation, identification of microbes and studying their sensitivity to antibiotics for the purpose of optimization of antibiotic therapy.

Material and Methods:-

We retrospectively have studied the medical records of the 17 patients of the University Clinic at TSMU Department of Therapy and Cardiology from 2015 - until September 2016. Acute exacerbation was defined as the presence of an increase in at least two of the three following symptoms: dyspnea, cough and sputum production. At least one sample of spontaneously expectorated sputum for microbiological evaluation was obtained from all patients during admission for aerobic gram-positive and gram-negative bacteria. An early morning sample was preferred. Specimens were collected according to standard guidelines(Baron and Thomas, 2012). Sputum samples were plated on 5% Sheep blood agar, Chocolate agar, Endo agar and Sabouraud dextrose agar and incubated at 37 °C for both 24 h and 48 h. All microorganisms isolated were identified through standard laboratory methods : isolation of pure culture, identifying microbes with rapid identification system(API20E, APIStaph, API haemo, API Strep, API 20NE, biomérieux). Sensitivity of microorganisms to antibiotics was defined with disc-diffusion method using standard discs(EUCAST guidelines).Rapid tests for identification of oxidase and catalase.

Results:-

In 12 patients monomicrobial growth has been documented by bacteriological investigation, among which gram-negative bacteria prevailed, namely *Klebsiella pneumoniae* $\geq 10^5$ colony-forming units/millilitre (CFU/ml) in 5 patients, *Escherichia coli* $\geq 10^5$ colony-forming units/millilitre (CFU/ml)– 2, *Proteus mirabilis* 10^8 CFU/ml-1. Among gram positive bacteria *Enterococcus* spp $\geq 10^5$ colony-forming units/millilitre (CFU/ml) was in 2 patients, *Staphylococcus aureus* -1, *Streptococcus pneumoniae* -1. Polymicrobial growth were observed in 5 cases: *Klebsiella pneumoniae* 10^5 CFU/ml and *Candida albicans* 10^8 CFU/ml-3, *Escherichia coli* 10^4 CFU/ml and *Enterococcus* spp 10^7 CFU/ml-1, *Staphylococcus aureus* 10^8 CFU/ml and *Enterococcus* spp 10^7 CFU/ml-1. By defining sensitivity on local antibiotics on strains isolate by us only 12% of gram-negative bacteria was resistant on quinolones, 19% to the third generation of cephalosporins, and among gram-positive bacteria methicillin-resistant *Staphylococcus aureus* and vancomycin-resistant *Enterococci* was not found. Comparatively high resistance against cephalosporins and quinolones related to wide usage of antibiotics of this group for chemoprophylaxis .

Conclusion:-

The bacteria are very common in the lower respiratory tract of patients with COPD and cause inflammation. We have shown that among microorganisms causing acute exacerbations of COPD in our patients prevailed gram

negative Enterobacteriaceae family and comparatively high resistance against cephalosporins and fluoroquinolones related to wide usage of antibiotics of this group for chemoprophylaxis.

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RESEARCH ARTICLE

SCORPION HEMOCYTE- GRANULOCYTE.

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Scorpions, Hemocytes, Granulocyte

Abstract

The study of hemocytes in arthropoda is the most fascinating area of research. The hemocytes are having similarity with that of blood cells in vertebrates. The study of hemocytes is mostly concentrated around insects and the scorpion hemocytes are having limited attention by scientists. In the present investigation granulocytes (GRs) of five different scorpions- *Mesobuthus tamulus tamulus*, *Mesobuthus tamulus concanensis*, *Orthochirus bicolor*, *Heterometrus xanthopus* and *Heterometrus phipsoni*, were studied qualitatively as well as quantitatively.

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Introduction:-

The scorpions are the arachnids with open type of circulatory system with hemocoel filled by hemolymph. The hemolymph consists of plasma in which numerous blood cells- hemocytes are suspended and playing vital role in maintains of immunity against various pathogens (Bauchau, 1981). The defense role played by hemocytes is through phagocytosis, encapsulation and by lyses of foreign cells (Soderhall and Cerenius, 1992). The hemocytes are also important for transport of nutrients and hormones at the site of growth and wound healing (Lavine and Strand, 2002). According to Al-Robai et al. (2002), there are three well defined types of hemocytes in arthropoda- prohemocyte (PRs), plasmocytes (PLs) and granulocytes (GRs). Some more types of hemocytes are recorded in arthropods are- coagulocytes (COs), spherulocytes (SPs), adipohemocytes (ADs) and oenocytes (OEs). In the present investigation the GRs are studied in scorpions- *Mesobuthus tumulus tumulus*, *Mesobuthus tumulus concanensis*, *Orthochirus bicolor*, *Herterometrus xanthopus* and *Heterometrus phipsoni*.

Material And Methods:-

All five species of scorpions were collected from their natural habitat from different localities of Maharashtra (India). These are placed in perforated jars containing hibiscus leaves, water and cockroaches as their food. The males and females were identified according to sexual dimorphic characters as described by Tikader and Bastawade (1983) and placed in separate jars.

- Collection of hemolymph: The hemolymph was collected from the live animals as explained by Padmanabha (1967). It is collected by aspiration through arthroidal membrane at the third joint of one of the pedipalp. 1 to 3 ml of hemolymph was collected from single animal.
- Techniques employed to study hemocytes: The hemocytes were studied qualitatively and quantitatively. The methods used to study hemocytes were same that was followed to study vertebrate blood cells. For qualitative study none of the single method was satisfactory, hence combination of different methods were employed. For qualitative study different staining methods used are- Sudan Black B, Giemsa, Janus Green B and Leishman's stain. The hemocytes were observed under light as well phase contrast microscope. The transmission electron

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microscopic (TEM) observation was made for only one sample of *H. xanthopus*. For identifying the granulocytes qualitative methods employed are differential hemocyte count (DHC) and total hemocyte count (THC). For THC Neubauer's hemocytometer was used (Witting, 1966).

Result and Discussion:-

During the present investigation Jones's classification (Jones, 1962) method was used. According to this method seven types of hemocytes- PRs, PLs, GRs, Ads, OEs and COs are present in the scorpions. These are also reported by Shah and Patil have studied the hemocytes in *M. tumulus tumulus* (2011) and in *O. bicolor* (2013).

The GRs are characterized by presence of cytoplasmic granules in it, which are stainable by different stains like Sudan Black B, Giemsa and Leishman's stain. These are generally round to oval in shape. The shapes of the GRs are round in *O. bicolor*, oval in *H. xanthopus*, round/ oval in *M. tumulus tumulus*. The size of GRs is smallest in *H. concanesis* (6- 12 μm), while largest in *H. phipsoni* (11-15 μm) in diameter. In *M. tumulus tumulus* the cell is 9- 15 μm , 7- 14 μm in *O. bicolor* while it is 10- 15 μm in diameter in *H. xanthopus*.

The nucleus of all GRs in above five types of scorpions is round in shape. The size of the nucleus is minimum in *M. concanesis* (3- 5 μm) while largest in *H. xanthopus* and *H. phipsoni* (5- 8 μm) in diameter. The size of the nucleus is 4- 6 μm in *M. tumulus tumulus*. The nucleocytoplasmic ratio was ranging from 30-70%. In all the GRs the nucleus was mostly central in position. The cytoplasm and nucleus of all above five GRs are basophilic in nature.

An attempt has been made to study ultra structure of one of the scorpions- *H. xanthopus*, in which the GRs are with following characteristics-

1. The GRs are almost spherical in shape.
2. The plasma membrane appears smooth.
3. The nucleus is eccentric in position due to presence of granules. The chromatin is scattered with dense clumps. The nucleolus with two zones.
4. The cytoplasmic granules are concentrated near nucleus and measures about 1- 3 μm in diameter and are of three types- structure less and electron dense, structure less and with small tiny granules, structured granules.
5. The nuclear envelope is perforated by nuclear pores.

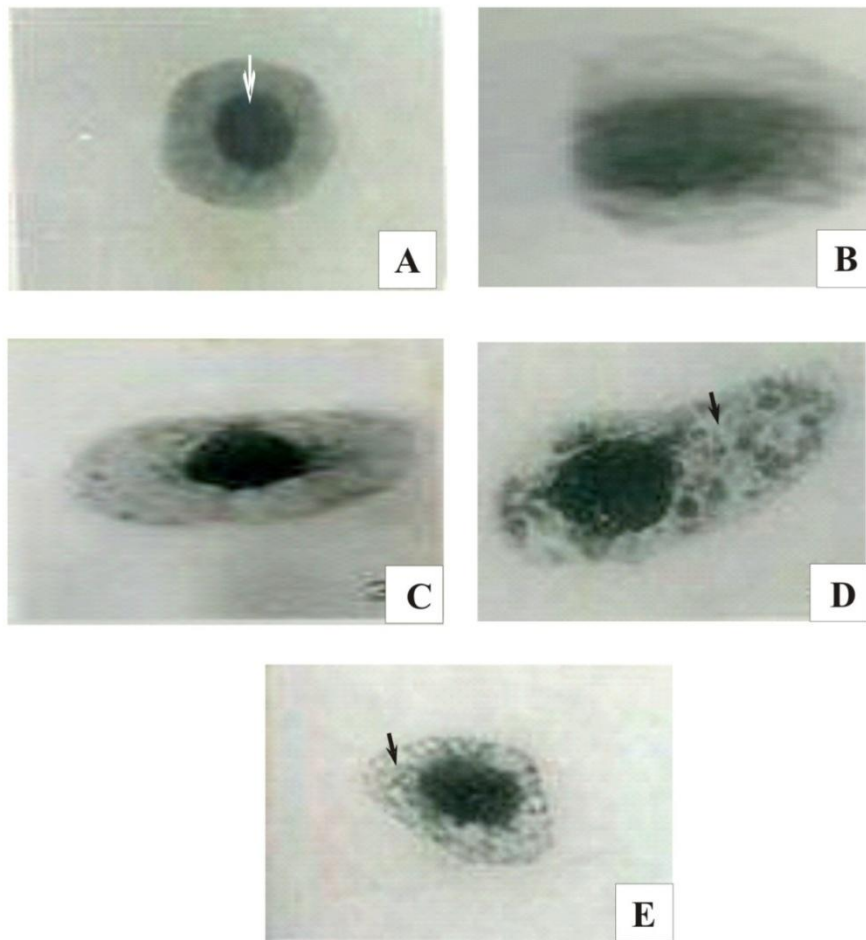
According to Gupta (1985) the structured granules by gradual transformed into structure less during maturation and discharged the contents in the hemolymph. In the present investigation during TEM study both types of granules were observed. The microtubules were associated with granules, which were also reported by Akai and Sato (1973) but there are no such structural microtubules are observed. The structured granules become structure less during discharge of its contents in hemolymph as shown in the Plate II. The same findings are reported by Gupta (1985). The chemical nature of the granules was identified by PAS (McManus, 1946) and Sudan Black B (Humason, 1979) methods. These granules are with mucopolysaccharides and lipid in it.

GRs are reported almost in all arthropoda including onychopora (Gupta, 1985a), hence are plesiomorphic hemocytes. Many researchers have reported phagocytic role of GRs (Gupta, 1985a). In the present investigation there were no any pseudopodia like structures are notified. The same findings were reported by Silva et al (2000) in GRs of larva *Anastrepha oblique*. There is conflicting views about the origin of GRs in arthropoda. Arnold (1974) has suggested that these arise from PRs and PLs. It was also supported by Takada and Kitano (1971). According to Gupta and Sutherland (1966) GRs are capable of giving rise to SPs, Ads and Cos. Arnold (1974) has suggested that "the GRs, might be considered as basic unit from which more precisely structural and functional classes of cells have been developed."

THC was performed in both male and female (Pregnant and non-pregnant) scorpions. The values of THC are lowest in male *H. xanthopus* (9750 per mm^3) and highest in *H. concanesis* (46850 per mm^3). The values of others are reported in the Table 1. The same findings were observed in females. The number of GRs are more in pregnant than non-pregnant females. The % of GRs was performed by DHC. Higher % of GRs was reported in *M. tamulus concanesis* (15%) while minimum (10%) in *H. xanthopus*. The granules of *M. tamulus tamulus*, *M. tamulus concanesis* and *O. bicolor* are smaller in size and number of GRs are more while the granules are larger in size in *H. xanthopus* and *H. concanesis* and number of GRs are less in number. It indicates that there is inverse relation between size of granules and number of GRs.

The hematology of scorpion can be done with ease as large volume of hemolymph can be obtained from single animal. Although Gupta (1986) has pointed out several functional analogies between arthropod immunocytes and vertebrate B- and T- lymphocytes, there is great scope to study it in future. In general the hemocytes of scorpions are similar to those in other arthropods. There are few reports about the study of hemocytes in scorpions. *M. tamulus concanensis* is one of the most venomous species found in India, whose poison fatal especially in children. Whether this venomous nature is related to high THC or to number of GRs is the matter of further research.

PLATE I



The granulocytes in the scorpions:

A: *Mesobuthus tamulus tamulus* (X940)

B: *Mesobuthus tamulus concanensis* (X940)

C: *Orthochirus bicolor* (X940)

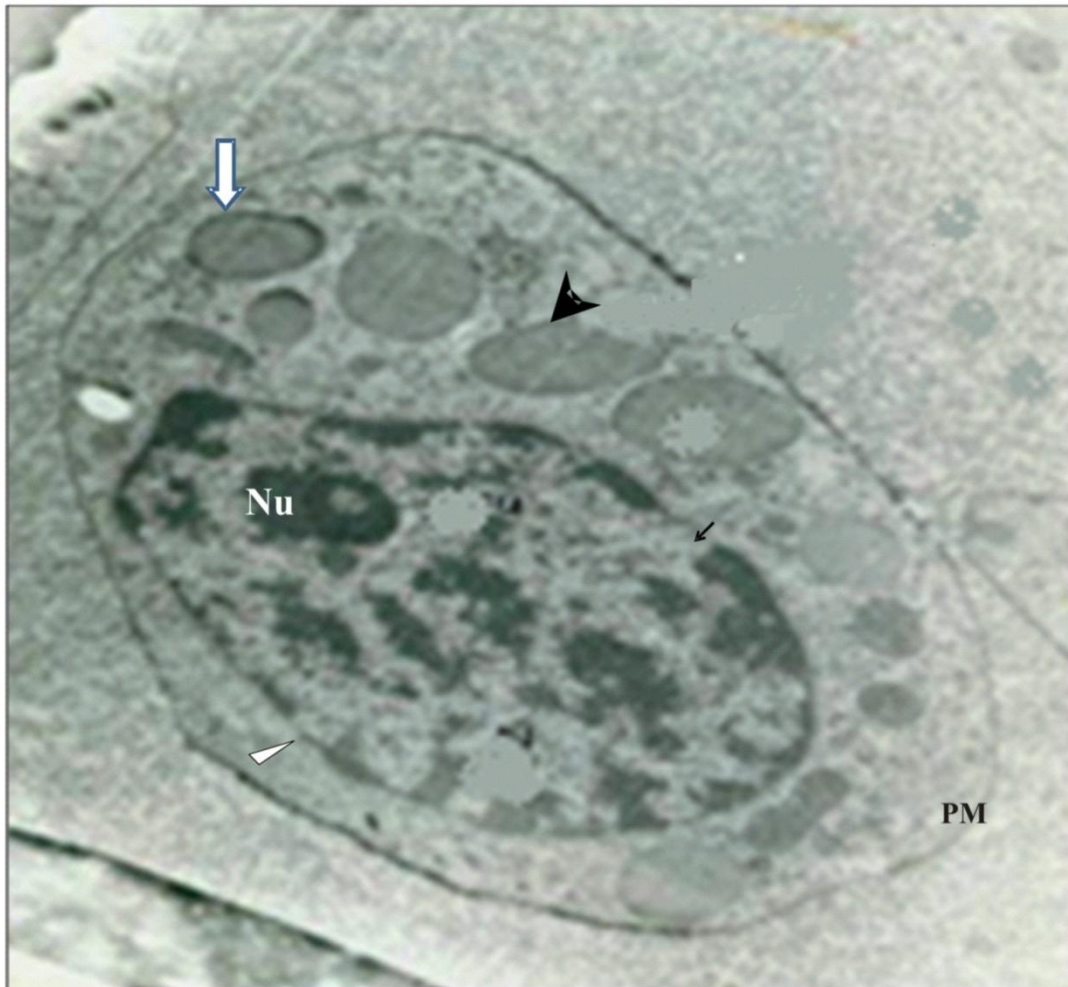
D: *Heterometrus xanthopus* (X940)

E: *Heterometrus phipsoni* (X940)

Black arrow- Granules

White arrow- Nucleus

PLATE II



TEM of Granulocyte in *Heterometrus xanthopus* (X15000)

Nu: nucleolus **PM:** Plasma membrane

Black arrow: Nuclear pore

White solid arrow: Membrane bound granules

Black arrow head: Bursting granule

White arrow head: Nucleus

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RESEARCH ARTICLE

ATYPICAL CLUB FOOT: EARLY DIAGNOSIS & TREATMENT BY MODIFIED PONSETI TECHNIQUE GOVERNMENT MEDICAL COLLEGE JAMMU.

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Abstract

Atypical clubfoot is a clinical type of idiopathic clubfoot that requires early detection and modification in the Ponseti treatment. The goal of treatment is to reduce or eliminate its 4 components so that the patient has a near functional foot and lives a normal life: We had 20 patients with 20 idiopathic atypical clubfoot deformity. Clinically, atypical clubfeet is defined as having rigid equinus, severe plantar flexion of all metatarsals, a deep crease above the heel, a transverse crease in the sole of the foot, short foot and a short and hyperextended first toe. Correction was achieved in all patients by modifying the Ponseti manipulation and casting technique. The mean number of casts that were applied to obtain correction was 8 (range four to nine casts). Tenotomy was done in all feet. Finally all the feet were considered clinically cured. Modified Ponseti method is an effective treatment for congenital idiopathic atypical clubfoot and reduces the need for corrective surgery.

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Introduction:-

During the course of treatment of idiopathic clubfoot by ponseti technique all feet show resistance to treatment. These feet have typical features like short foot, smaller great toe, deep planter crease. These feet are not usually identified initially but over the course of treatment these features become more prominent. Turco (1994) advised against surgery in these feet. He classified them as atypical clubfeet. Ponseti et al.(2006) advised about change in his standard protocol in these feet. We performed these modified Ponseti technique of manipulations and casting to see if this modified treatment would result in correction and maintenance of the correction for these atypical clubfeet.

Materials and methods:-

20 patients with 20 clubfeet were chosen. They were atypical clubfeet as they showed resistance to standard casting technique and had following features: feet were short and stubby, deep crease present over sole of foot, deep crease above the heel, severe equinus and supination, forefoot in adduction and planterflexion of all metatarsals.(figure 1)

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There were 9 boys and 7 girls. Average age of patients was 3 months (range 1- 5 months). Treatment had been started in our hospital and from outside by applying plaster casts. We excluded syndromic clubfoot & clubfoot associated with neuromuscular disorders. The average follow-up was for 1 & half years (range 1 to 3 years). In all these patients plaster casts were applied . In all these cases standard ponseti technique was modified. Correct identification of talar head is essential. It is done by moving index finger and thumb down lateral malleoli and then grasping the head of talus. With the other hand forefoot is abducted which causes movement at Talo-calcaneo-navicular joint and thus in the correct identification of talar head.

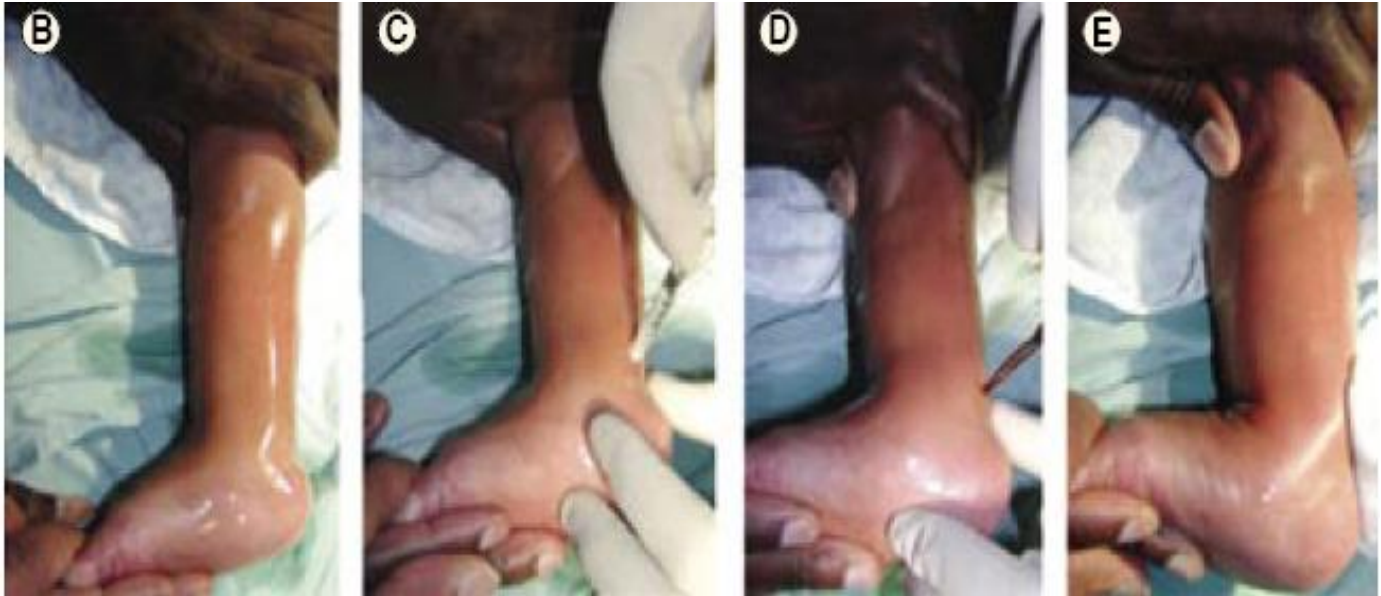
Abduction is done by putting thumb on lateral aspect of head of talus. Index finger of same hand is put behind lateral malleolus. Using thumb and index finger of other hand abduction is done.(figure 2)

Abduction is done upto 40 degrees only. Standard abduction of 70 degrees is not done. Abduction is always kept less than 40 degrees. Abduction beyond 40 leads to abduction at tarsometatarsal joints and thus disruption of this joint. While abduction is being done , the foot usually come s into alignment with the long axis of the leg. This usually leads to slipping of cast which may cause foot edema and skin excoriation. To prevent this we applied the cast with the knee bent to 110 degrees . To prevent excessive cast behind the knee, we applied slab of cast anteriorly on the knee to strengthen the cast. A similar slab was applied behind ankle. When desired abduction was obtained , the metatarsals were still severely planterflexed. Here another modification was performed. Thumbs of both hands were placed under the heads of 1st and 5th metatarsals. Index fingers of both hands were placed dorsally over the talar head and dorsiflexion of metatarsals and ankle was performed. (Figure 3) Tenotomy was performed in all cases.

(Figure 4)







Post tenotomy dorsiflexion of 10 degrees was usually obtained. Post tenotomy cast was applied for 3 weeks. If ankle dorsiflexion of less than 5 degrees was obtained post tenotomy then cast was applied for 1 week only and a new cast was applied weekly with increasing dorsiflexion till 10 degrees of dorsiflexion was obtained. Following cast removal Dennis Brown splint was applied. (Figure 5) Feet were kept at 40 degree abduction on both normal and affected sides unlike standard Ponsetti technique, where feet are kept at 70 degree abduction on affected side and 40 degree abduction on normal side.



Brace was applied for 23 hours for first 3 months. Following initial 3 months in brace, weaning from brace was done with 2 hours of bracing time reduced every month until night time bracing (12 hours) was started and was advised to be kept for 3-4 years.

Results:-

The average age of the patients at the start of treatment was 3 months (range, 1-5 months). All the twenty patients had unilateral clubfoot. The mean number of casts that were applied to obtain correction was 8 (range four to nine casts). Percutaneous tendo Achilles tenotomy to correct the residual equinus deformity was done in all patients. The average follow-up was for 1 and half years (range 1 to 3 years). Finally, all feet were corrected (figure 5) with mean ankle dorsiflexion of 15° (range, 10°–20°).

Discussion:-

Orthopaedicians have been treating clubfoot successfully using Ponseti technique (Chotel et al., 2002; Colburn and Williams, 2003). However there is a small group of clubfoot which does not correct or shows resistance to correction by standard Ponseti method. These feet are short and stubby. The hindfoot is in severe equinus and varus. The calf muscles are small and the tendo achilles is long, wide, and tight. The forefoot is adducted and all metatarsals are in severe plantar flexion. There is a deep crease across the sole of the foot and another above the heel. The great toe is short and in dorsiflexion. Applying casts by standard Ponseti methods leads to complex deformities in these cases. There develops hyperabduction of forefoot at tarso-metatarsal joint but cavus and equinus do not get corrected. There occurs slippage of cast causing sores over bony prominences and edema of the foot. This type of clubfoot is difficult to treat and requires modified Ponseti technique. Adduction of forefeet should not be corrected beyond 40 degrees to prevent hyperabduction at tarso-metatarsal joint. Adduction should be corrected by keeping thumb on lateral aspect of talar head and index finger of same hand behind lateral malleolus. This also corrects heel varus. Once the heel varus is corrected, the planter-flexed forefoot and the equinus are corrected simultaneously by forcefully dorsiflexing the metatarsals with both thumbs while keeping index fingers of both thumbs on dorsal aspect of talar head. The cast is strengthened by applying a posterior slab behind the ankle. To prevent the cast from slipping, knee is immobilized at 110 degrees of flexion and it is strengthened with a slab applied anteriorly over the knee. A tenotomy is performed before applying the last plaster cast to facilitate correction of any unyielding equinus.

Thus our study shows that an orthopaedic surgeon needs to be careful enough to identify an atypical clubfoot. There is need of modified Ponseti technique in case of atypical clubfeet. This modification corrects the deformity thus decreasing the need for surgery in most children. Once an atypical clubfoot is treated adequately, the foot develops normally.

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RESEARCH ARTICLE

A CERTIFICATE-DRIVEN APPROACH TO ACCESS CONTROL IN FUTURE INTERNET.

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Key words:-

Future Internet, mutual authentication, collusion attack, privacy, content integrity, normal distribution, bell curve, recurrence relation, time complexity

Abstract

The growing impact of the Internet in all walks of human life has also introduced us to newer problems related to security and hence the increasing demands to address such threats that prevail. The Future Internet (FI) being a confluence of various technologies, and the cloud being one such component technology in the FI that provides storage as a prominent service amongst the other services offered by it, it follows that the public cloud cannot be trusted at all times and therefore cannot be completely relied upon to regulate access to the stored sensitive data that it contains. This creates an avenue for collusion attack by the cloud and a malicious user. This has been addressed in our proposed system by the separation of access policy and access structure. Further considering the insecure communications and the vulnerability of the transmitted data, a mutual identity establishment scheme has been proposed to secure the channel and therefore the contents in transit. A recurrence relation for the established scheme has been derived to calculate its time complexity and the time taken to achieve mutual authentication has been administered for repeated iterations and the values thus obtained have been plotted and verified under the normal distribution bell curve to suggest the model's consistency and adaptability.

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Introduction:-

Internet has become an essential technology that impacts billions of people in every aspect of our lives today. Internet of today has billions of connected devices which are still predicted to evolve/increase over the years to come. It is facing multidimensional challenges ranging from attacks to performance issues, while at the same time should never cease upon its responsibility of maintaining organizations' and personal integrity and privacy. Hence there are ever increasing research activities in progress on having the Future Internet (FI) to address its current shortcomings. The future Internet being a confluence of devices, technologies and areas, and access management being one such area of concern in association with its component technology the FI, an attempt has been made in this paper to address a few security issues namely, collusion attack, replay attack resistance, mutual authentication establishment and content integrity.

Motivation:-

The evolution of Internet and its advancements that press towards the establishment of a futuristic era that involves a confluence of various devices/things intermittently connected online to exchange data/information and provide

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services, has led to the growth of demands in terms of security, privacy and trust which are the basic elements that need to be satisfied from an end user's point of view. The lack of a proper authentication mechanism and secured ways of exchanging data makes it easier for an intruder to steal/sniff data illegally thereby affecting the security and privacy of the users. Also, considering the usage of a public cloud, the cloud cannot be trusted all the time, and hence is capable of exposing sensitive data maliciously (collusion attack). This necessitates us to develop suitable architectures and algorithms that helps address the security, authentication (for secure channel establishment) and content integrity issues that occur in a communication between participating entities.

Related work and evaluation:-

Table 1:- Related work and Evaluation

Paper No	Mutual Authentication	Content Integrity	Forward Secrecy maintenance	Trust
[4]	No-one way	Not addressed	Not addressed	One way (server is assured trust on the requesting device)
[5]	No-one way	Not addressed	Not addressed	Not addressed
[6]	No-one way	Yes-hiding of data using one way hash function	Yes- session parameters and pseudorandom numbers	One way (server is assumed to trust on the requesting device)
[7]	No-one way	Not addressed	Not addressed	Authorization function delegated to third party, requires an assumed trust level.
[8]	Yes-Chbyshev chaotic maps	Yes – aggregated proofs	Yes- session parameters and pseudorandom numbers	Yes
[9]	No-one way	Partial – privacy on data stored not on user private information	Not addressed	Yes, Trusted third party
[10]	No-one way	Yes , OAuth	Not addressed	Pre-requisite for authentication

The need to regulate access to critical and sensitive information has led to the proposal of numerous access control schemes like the Dynamic broadcast encryption technique [1], role based access control (RBAC) [2] and Attribute based encryption (ABE) [3] which have addressed the issues such as scalability and anonymity. However, the assumption that the cloud is honest (trusted) but curious, does not hold good at all times since the cloud, controlled by the cloud service provider (CSP) may not belong within the trusted domain of the involved association/group thereby providing greater avenue for the cloud to expose the access policy (rules that regulate access to the sensitive data and provides protection) to malicious attackers and collude with them posing threat to the stored sensitive data.

Even though SLAs address this as a legal issue, preventing such attacks by providing technical solutions makes the system resilient and more reliable. Also, several schemes have been introduced to address the security issues that prevail in the various areas that make up the Future Internet. A few solutions include the challenge-response protocol coupled with a novel two factor authentication protocol [4], Kurento and Nubomedia [5], a Shared Authority based Privacy Preserving Authentication protocol (SAPA) to address the privacy issues using the Attribute based Access Control (ABAC) and proxy re-encryption scheme [6], a HTTP/CoAP (Constrained Application Protocol) service based architecture to provide an authorization framework [7], an aggregated proof based Hierarchical Authorization scheme in U2IoT [8], a novel privacy preserving authenticated Access Control scheme [9] and an optimization work on the OAuth 2.0 protocol [10] to solve the associated performance challenges while still ensuring privacy of user sensitive credentials.

However in all such cases the necessity to establish a trusted communication channel via mutual authentication and ensuring the integrity of the user sensitive data that is exchanged between devices has not been considered. Also, forward secrecy, a phenomenon wherein an attacker should be incapable of tracing back to the data transmitted previously with the information that he has managed to fetch at that instance is not considered in most cases. Table 1

summarizes the above discussions and provides a comparative analysis of a few security issues discussed in this section.

Proposed Architecture:-

Fig 1 shows the proposed architecture [11] that addresses the security issues such as maintaining user privacy (by providing a user controlled environment) and collusion attack caused due to the exposure of access policy that contains the sensitive user information to the cloud. The sequence of steps that occur in the system are labeled [1-10] in the figure below. As shown in fig 1. The system consists of three entities namely the Attribute Authority ((AA) owner of the data), the public cloud that provides services in terms of storage and processing power, and the users/devices with their sensors. The communication between the entities follows a model wherein the access policy lies with the sole owner AA and the AAs within the trusted domain of the owner.

The AA requires that the cloud and the devices register themselves initially depicted as channel 1 in fig 1. The AA provides certificates to the entities upon registration, which is to be used later to prove their identities to establish trust amongst the participating entities. The AA uploads the encrypted data and documents along with the access structure to the public cloud. The cloud only possesses the access structure (contains key and ID of values in the access policy) that facilitates it to map the user/device attribute values by matching the associated IDs of attributes. The cloud is therefore left unclear about the actual attribute values that facilitate access to the data. Its job is therefore limited to storing the data and collecting the required attributes, and forwarding it to the appropriate AA which ultimately makes the decision about allowing/denying access after comparison with the respective access policy. The cloud is thereby prevented from participating and executing a collusion attack.

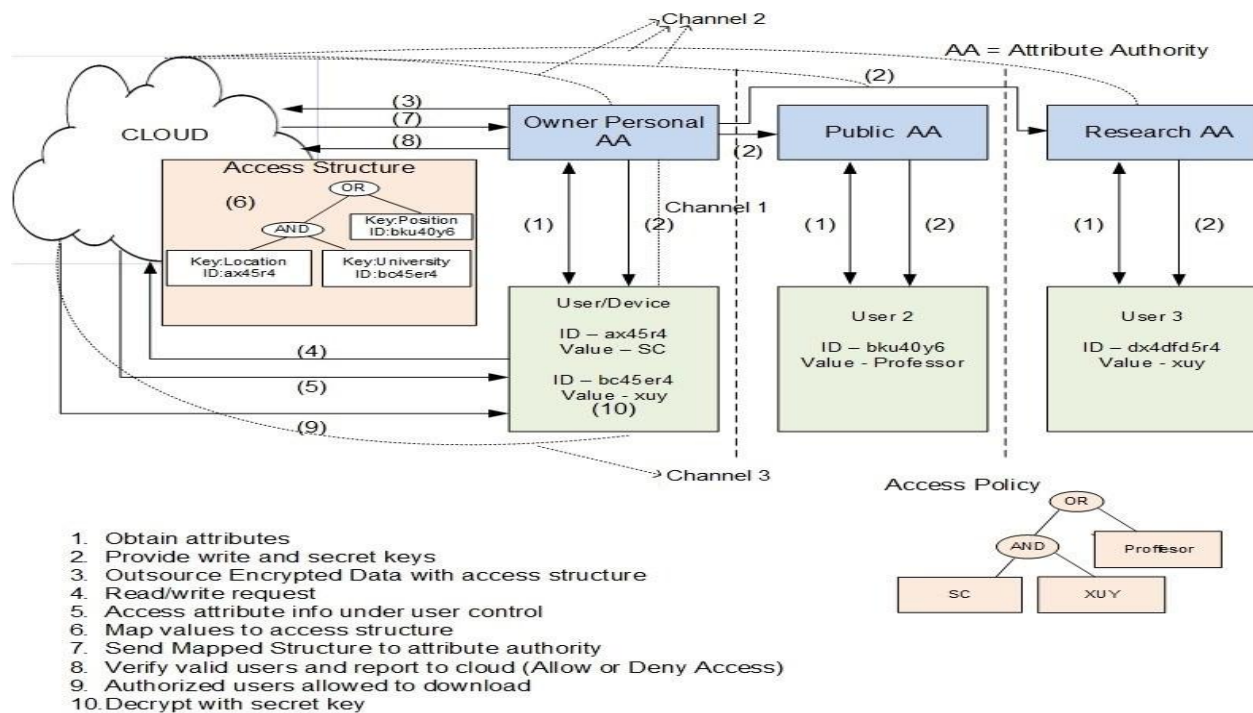
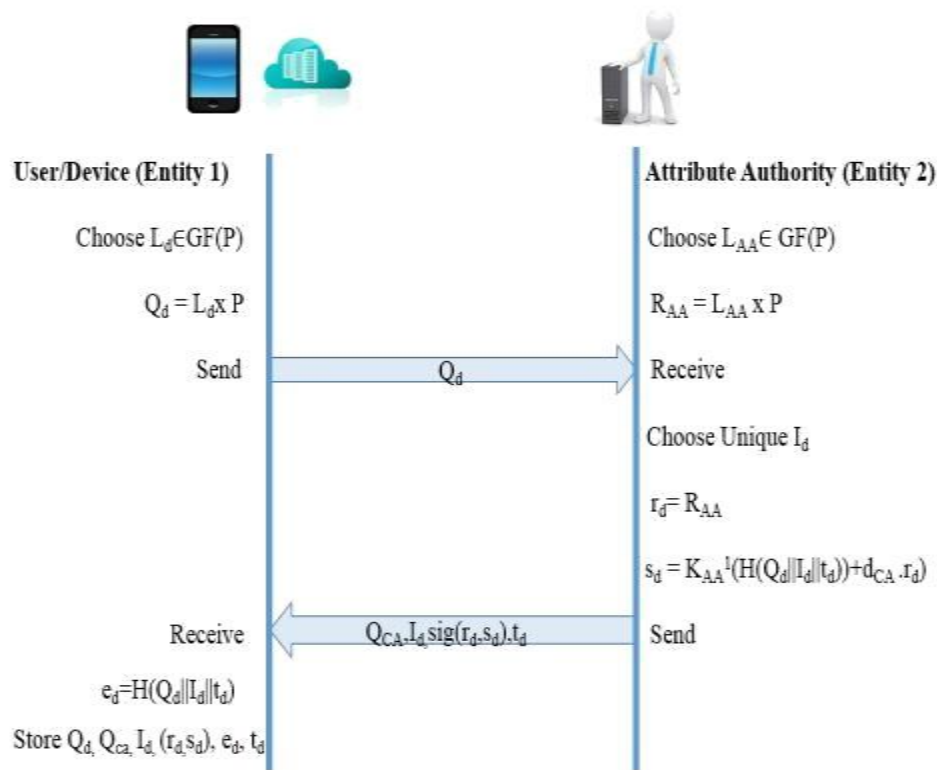


Fig 1:- Proposed Architecture

Algorithmic Details:-**Fig2:-** Certificate Distribution.

The communication between the entities namely the AA, user/device and cloud are classified into three parts namely,

1. AA (server) \rightarrow User/device/cloud (client) [user/devices register with the AA and obtain keys] represented as channel 1a and 1b in fig 2.
2. Cloud (server) \leftrightarrow user/device (client) [user/devices request access to data on the cloud] represented as channel 2 in fig 2.
3. AA (server) \leftrightarrow Cloud (client) [cloud sends the collected and mapped data for verification to the AA] represented as channel 3 in fig 2 [12][13].

In all the above cases the proposed mutual identity establishment scheme is executed in order to ensure the establishment of a secure communication channel and integrity of transmitted message. This scheme adopts the ECCDH (Elliptical curve diffie Hellman) mechanism for the establishment of a shared secret key (Q_{sk}) between the participating entities shown in fig 2. The execution of the scheme is threefold. First, as mentioned earlier, the AA issues certificates to the devices and cloud shown in fig 2. This certificate consists of a unique temporary ID (I_d) chosen by the AA for the cloud/device which in turn performs a hash on the chosen Identity and the certificate expiration time, signs it and passes it over to the cloud/device which is then utilized for authentication.

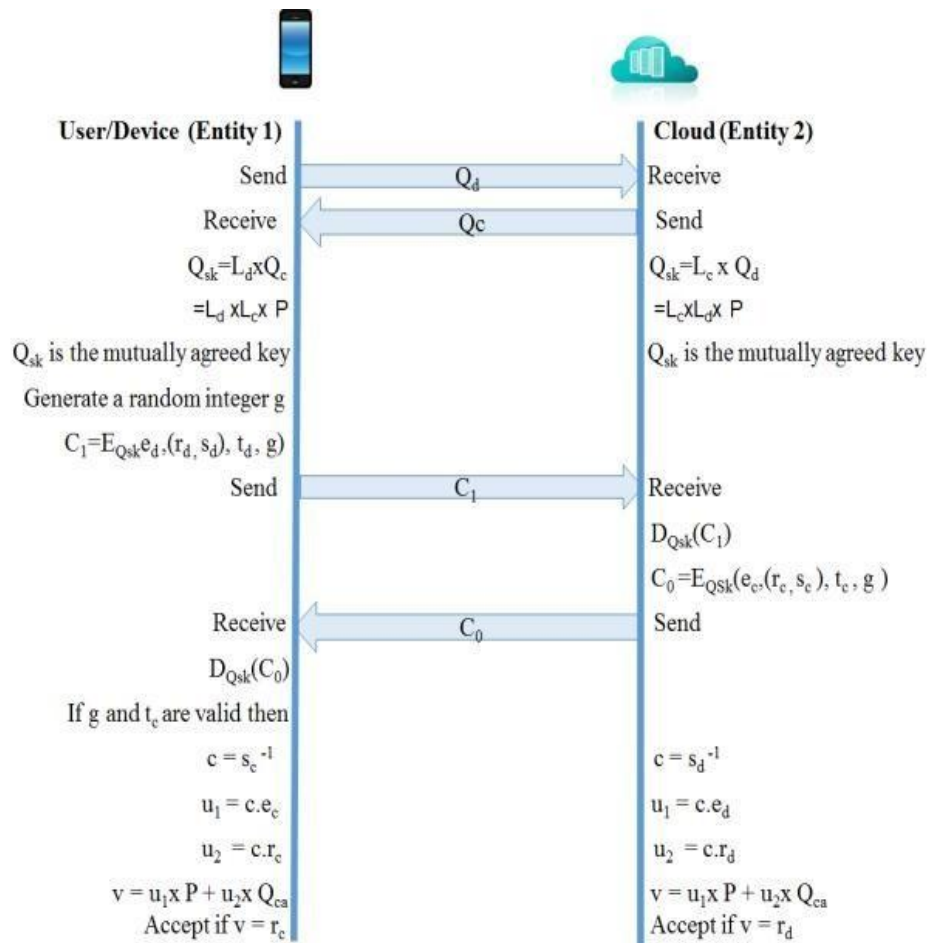


Fig 3:- Mutual authentication

Further, in order to ensure the establishment of a secure channel for exchange of data, mutual authentication has been implemented via exchange of the participating entities' certificates and a random number g that is appended to the certificate and encrypted with the shared secret key obtained via ECCDH to maintain the freshness of the exchanged information. The certificates are then verified on either side following the ECCDSA's (Elliptical curve Cryptography Digital Signature Algorithm) verification mechanism. This establishment of mutual authentication provides a secured communication channel to ensure trust, and prevents the privacy theft/tampering of the exchanged information (user attributes/requests and request approval from AA).

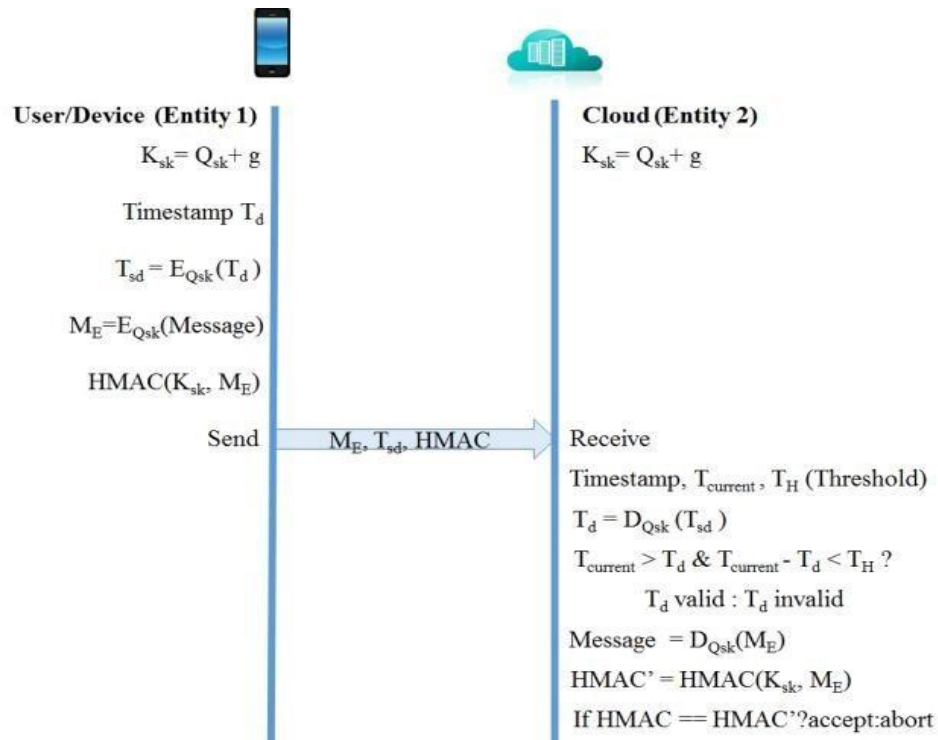


Fig 4:- Content Integrity

Also, the integrity of the transmitted message is achieved by computing a HMAC of the encrypted message (ME, encrypted by using the shared secret key (Qsk)) and the attribute structure is flushed off from the cloud by using the session management techniques, transported to the other side where the encrypted value of timestamp is decrypted and verified against a predefined threshold time (TH) thereby enabling the detection of replay attack. A session key Ksk is generated by adding the random number g with the shared secret key to help maintain sessions by which we can ensure that the mapped attributes are not retained at the cloud after transfer to the AA. This shared secret key is used to compute the HMAC of the encrypted message ME.

The computed HMAC, encrypted time-stamp (using shared secret key (Qsk)) and encrypted message (ME) are transmitted to receiver (Entity2) and any attacks/tampering of data is identified by verifying the received HMAC against the recomputed HMAC' on the receiver (Entity2) side. If any discrepancy is detected the received content is discarded, otherwise accepted for further processing. Fig 3 shows the mutual authentication establishment scheme for the communication between a device and the cloud and fig 4 shows the content integrity maintenance scheme in the communication between a device and the cloud.

A similar scheme that follows the same sequence of operations is defined for the communication between the cloud (client) and the AA (server) to achieve mutual authentication and content integrity.

Time Complexity Analysis and Performance Evaluation of the System:-

A recurrence relation for the proposed mutual identity establishment scheme presented in the previous section is given below as

$T(n) = aT(n/a) + 3n$ Where, n represents the number of requests that are incident at the cloud/AA/server via any device/user (client).

a, represents the number of domain attribute authorities(AA) which in the proposed system amounts to three AAs . Here the requests that arrive at the cloud are assigned to the appropriate attribute authorities which is given by n/a or $n/3$ and is handled by each of the AAs individually at each domain, therefore multiplied by the total number of attribute authorities i.e., $aT(n/a)$ or $3T(n/3)$.

$3n$, is a summation of the time required to distribute certificates for n entities that register with the AA (n units of time), time required to establish mutual authentication for n entities/devices and their associated requests (n units of time) Upon solving the recurrence relation by master's theorem to obtain the running time complexity we get,

Iteration frequency	2	1	2	3	4	1	1	2	1	1	1	2	2	1	2	1	1	1	1
MA in ms	4	4	4	4	4	4	4	4	4	4	4	4	4	4	4	4	4	4	4
	1	2	5	7	4	2	6	5	3	5	4	2	4	4	3	3	3	6	8
	0	2	0	0	0	3	7	5	4	0	7	0	5	2	0	2	3	0	3

Table 2:- Mutual Authentication (MA) Time Dataset in Milliseconds.

$T(n) = O(n \log n)$ [since $=n$ and $f(n) = 3n \approx n$ upon ignoring the constant term associated with n] and time required for integrity check of the transmitted contents (n units of time).

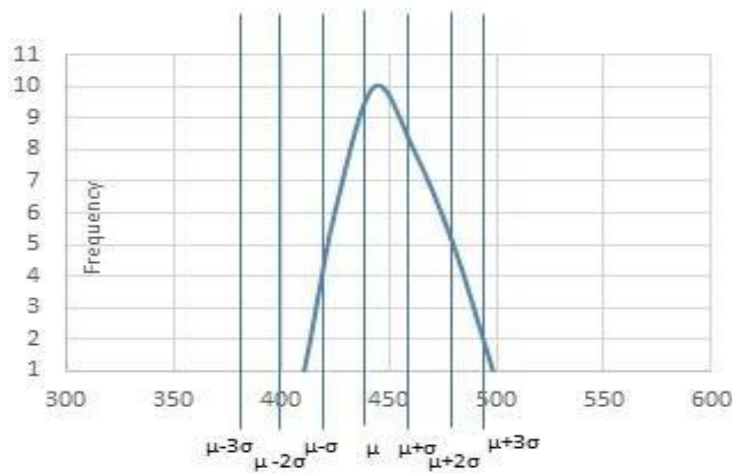


Fig 4:- Content Integrity

Further, the implementation of the proposed algorithm in the system has been verified against 30 iterations and the amount of time taken to mutually authenticate the devices was noted. For the data given in table 2 we calculate the average to be 443.1667ms and standard Deviation 18.41753. In statistics, the so-called 68–95–99.7 rule indicates the percentage of values that lie within a band around the mean in a normal distribution with a width of one, two and three standard deviations, respectively. With our data set of 30 samples we find that all the values fall within the width of three standard deviations i.e. 99.73% as depicted in the bell curve in fig 5, which suggests that the proposed model is predictable with no outliers and ideal for use.

Conclusion and Future Work

This paper has established the need for Future Internet to address privacy and integrity of exchanges/transmitted data that will determine its successful adoption universally. It has provided an insight into the proposed system and its resistance to collusion attack. Also, the reader has been introduced to the mutual identity establishment scheme that ensures secure transfer of data through the establishment of a secure channel via mutual authentication establishment and assurance of content integrity. We have successfully derived a recurrence relation for the proposed scheme and calculated the running time complexity for the same. Also, a plot (normal distribution bell curve) of the different times taken for mutual authentication for numerous iterations has revealed the system's consistency in performance and its adaptability. In future we propose to extend the scheme to implement the auto-delegation mechanism along with the interoperability of the devised protocol with the existing ones.

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RESEARCH ARTICLE

IN VITRO EFFICACY OF *TRICHODERMA* ISOLATES AGAINST SOME FUNGI CAUSING FUNGAL ROT DISEASE OF TOMATO.

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Abstract

Biological control has been found efficient eco-friendly approach for the management of several fungal diseases caused by pathogenic fungi. Therefore, present study was carried out to evaluate the *in vitro* efficacy of some *Trichoderma* isolates against the pathogenic fungi, *Alternaria alternata* (Fr.) Keissl, *Penicillium* sp and *Aspergillus niger* Van Tiegh known to infect tomato fruits in Kashmir. It was observed from the results that all the *Trichoderma* isolates brought about significant inhibition in the mycelial growth. The inhibition in mycelial growth of *Alternaria alternata*, by PPT3 isolate of *Trichoderma* was 26.79%, followed by isolate PPT1 (19.47%) and isolate PPT2 (14.64%) of *Trichoderma* respectively. Likewise, the highest inhibition in mycelial growth of *Penicillium* sp. was shown by isolate PPT3 (34.36%) followed by isolate PPT2 (23.44%) and by isolate PPT1 (9.37%) respectively. In case of *Aspergillus niger*, the inhibition in mycelial growth was highest by isolate PPT3 (42.5%) followed by isolate PPT1 (36.67%) and by isolate PPT2 (31.67) respectively. The results indicate that different local isolates of *Trichoderma* proved effective against the tested pathogenic fungi and can be evaluated further for their antifungal activity against other fungi.

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Introduction:-

Tomato (*Lycopersicon esculentum* Mill.) is a member of the family Solanaceae which comprises short-lived perennial herbaceous plants. It is one of the most popular vegetable crops widely grown for its edible fruits, high nutritive values and also for its diversified uses (Afroz *et al.*, 2008; Ewulo *et al.*, 2008). They are important source of vitamins and important cash crop for both small holders and medium scale commercial farmers (Ana *et al.*, 2003). The fruit also contains plenty of antioxidant carotenoid lycopene that has recently attracted interest because of its role in preventing cancer heart disease and muscular degeneration (Wener, 2008). Several pathogenic fungi are known to cause damage to tomato especially under storage conditions and also on the aerial parts of the plants. The main postharvest disease of tomato caused by various pathogenic fungi are *Rhizopus stolonifer* (Stevens *et al.*, 1997; Schena *et al.*, 1999), *Botrytis cinerea* (Lee *et al.*, 2006) and *Alternaria alternata* (Feng and Zheng, 2007). *A. alternata* is a saprophytic pathogen of tomato causing postharvest black rot at high frequency (Akhtar *et al.* 1994). Harvested fruits and vegetables decay due to fungal infestation is the main cause of major losses in food production. Postharvest decay of fruits and vegetables can be traced to infections, occur either between flowering and fruit maturity or during harvesting and subsequent handling and storage (Eckert and Ogawa 1988). The loss resulting

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from these infections have been reduced primarily by applying fungicides in the field or after harvest (Eckert and Ogawa 1988).

Trichoderma spp. is the most widely studied biocontrol agents (BCAs) against plant pathogens because of their ability to reduce the population of soil borne plant pathogens (Papavizas, 1985). They are soil borne fungi and show significant activity against a wide range of plant pathogenic fungi (Elad *et al.*, 1982). Mechanism used by *Trichoderma* spp. for control of plant pathogen includes competition, mycoparasitism, antibiosis and induced resistance of the plant host (Chet, 1987; Schirmböck *et al.*, 1994).

Trichoderma is one of the common fungal biocontrol agent is being used worldwide for suitable management of various foliar and soil borne plant pathogens. (Dominguesa *et al.*, 2000). Therefore, the objective of the present investigation was to assess the efficacy of local isolates of *Trichoderma* under in vitro condition against some fungal pathogens isolated from tomato fruits.

Materials and Methods:-

Tomato (*Lycopersicon esculentum* Mill.) fruits were obtained from different vegetable local markets of Kashmir Valley. Samples of infected fruits were brought to the laboratory in clean polythene bags and these samples were either used immediately or stored at 10°C in the laboratory for different pathological studies. Small portions of rotted tissues were isolated aseptically from the diseased tomato fruits and transferred to Potato Dextrose Agar (PDA) medium. Pure colony cultures were obtained by sub-culturing the isolated fungi such as *Alternaria alternata* (Fr.) Keissl., *Penicillium* sp and *Aspergillus niger* Van Tiegh, in separate Petri plates containing the same medium. The pathogen was identified by their morphological, reproductive and cultural characteristics (Ellis, 1971; Barnett and Hunter, 1972; Watanabe, 2002; Gilman, 2008). For pathogenicity, pathogens were re-inoculated after isolation onto the healthy pear fruits (Tomkin and Trout, 1931). Then all the fruits were kept in clean polythene bags and incubated at 25±2°C for ten days. These pathogenicity tests were used for the identification of plant pathogens and to confirm the detection of a particular disease. Identification of the disease and the pathogen was done following Koch's postulates.

Isolates of *Trichoderma* were isolated from rhizospheric soil of the tomato plant following serial dilution method (Brown 2004). These isolates were identified on the basis of morphological and cultural characteristics and named as PPT1, PPT2 and PPT3. The antimycotic effect of these pure local isolates of *Trichoderma* against isolated fungi such as *Alternaria alternata* (Fr.) Keissl., *Penicillium* sp and *Aspergillus niger* Van Tiegh was assessed by dual culture method (Bashar and Rai 1994, Prince *et al.* 2011). To assess the effect of *Trichoderma* isolates against fungi, known quantity of mycelia of both was inoculated onto petriplates containing PDA. Then the inhibition in mycelial growth of fungi such as *Alternaria alternata* (Fr.) Keissl., *Penicillium* sp and *Aspergillus niger* Van Tiegh by isolates of *Trichoderma* was observed after incubation. The percent inhibition in mycelial growth of tested fungal pathogens in presence of *Trichoderma* isolates was calculated as per the formula given by Skidmore and Dickinson 1976.

$$\text{Mycelial growth inhibition (\%)} = \frac{dc - dt}{dc} \times 100$$

Where dc = average diameter of fungal colony in control, and dt= average diameter of fungal colony in treatment group.

Results:-

It was observed from the results (Table 1) that isolate PPT-3 of *Trichoderma* inhibited the mycelial growth of *Alternaria alternata* to an extent of 26.79 per cent. This was followed by *Trichoderma* isolates PPT-1 (19.47%) and PPT-2 (14.64%) respectively. Likewise, growth inhibition of *Penicillium* sp by *Trichoderma* isolates PPT-1 was 9.37%, by PPT-2 was 23.44% and by PPT-3 was 34.36% respectively and inhibition in mycelial growth of *Aspergillus niger* due to *Trichoderma* isolates, PPT1, PPT2 and PPT3 were 36.67%, 31.67% and 42.5% respectively.

The maximum growth inhibition of *Alternaria alternata* and *Aspergillus niger* was due to *Trichoderma* isolate, PPT3 followed by isolate PPT1 and isolate PPT2 respectively. Whereas, isolate PPT3 caused maximum growth inhibition of *Penicillium* sp. However, the least inhibition in mycelial growth of pathogenic fungi was observed by *Trichoderma* isolate, PPT1.

Table 1:- Effect of *Trichoderma* isolates on the mycelial growth of fungi causing rot disease of tomato by dual culture plate method

Treatment	Mycelial growth (mm)		
	<i>Alternaria alternata</i>	<i>Penicillium sp</i>	<i>Aspergillus niger</i>
PPT1	11.00±1.00 (19.47%)	19.33±1.15 (9.37%)	25.33±1.52 (36.67%)
PPT2	11.66±1.52 (14.64%)	16.33±1.52 (23.44%)	27.33±2.08 (31.67%)
PPT3	10.00±1.00 (26.79%)	14.00±1.00 (34.36%)	23.00± 2.00 (42.5%)
Control	13.66 ± 1.52	21.33± 1.52	40.00±1.00

Each value is mean of 3 replicates ± SD

Figures in parenthesis is the mycelial growth inhibition (%)

Discussion:-

It is clear from the above results that different local isolates of *Trichoderma* caused significant inhibition in mycelial growth of *Alternaria alternata* (Fr.) Keissl., *Penicillium* sps and *Aspergillus niger* van Tiegh.

In a similar studies, the antagonistic activities of *Trichoderma harzianum* against several pathogenic fungi have been reported by many workers (Henis and Chet, (1975); Backman and Rodrigues-Kabana, 1974; Hadar *et al.*, 1979 and Elad *et al.*, 1980). Kakde and Chavan (2011) studied the antagonistic activity of *Trichoderma viride* and *Trichoderma harzianum* against storage fungi and found that growth of *Curvularia lunata*, *Rhizopus stolonifer*, *Fusarium oxysporum*, *Macrophomina phaseolina* and *Penicillium chrysogenum* was retarded due to *Trichoderma* species. Dharmaputra *et al.* (1994) also tested two isolates of *T. harzianum* and one isolates of *T. viride* against *Ganoderma* and found that all isolates inhibited the mycelial growth of the pathogen, but *T. harzianum* (isolates B 10-1) showed the best performance. Etabarrian (2006) reported that *T. viridi* (MO) reduced the colony area of *Macrophomina phaseoli* by 19.2 and 34.9% using the dual culture and cellophane methods, respectively. Intana *et al.* (2007) reported the efficacy of three mutant and two wild type strains of *T. harzianum* against *Colletotrichum capsicis*, causal agent of anthracnose of chili. The present study suggests that *Trichoderma* sp. effectively inhibits the growth of fungi causing rot of tomato, hence can be used as a biocontrol agent against rot diseases of vegetables. Bashar and Rai (1994) studied the three isolates of *Trichoderma* viz., *Trichoderma harzianum* Refai, *Trichoderma hamatum* and *T. viride* Pers. were selected to test their antagonistic potential against the pathogens following dual culture technique. However, further investigation is needed for the efficacy of these local isolates of *Trichoderma* against fungal pathogens.

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RESEARCH ARTICLE

A NEW SPECIES OF THE *PHYLLANTHUS VASUKII* SP. NOV. (PHYLLANTHACEAE) - FROM SOUTHERN EASTERN GHATS OF INDIA.

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Abstract

A new taxaon, *Phyllanthus vasukii* sp. nov., is described from the Southern Eastern Ghats. A detailed description, Phenology, Anatomical Characters, Micromorphology of seeds, Distribution and Ecology notes and IUCN status are provided for easy identification.

Key words:-

Phyllanthus vasukii, New species,
Namakkal, Southern Eastern Ghats.

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Introduction:-

Phyllanthaceae is the second largest segregate of Euphorbiaceae *sensu lato*, comprising c. 2000 species in 49 genera. Phyllanthaceae includes two subfamilies, Phyllanthoideae and Antidesmatoideae. Phyllanthoideae consists of four tribes and *Phyllanthus* belongs to the tribe Phyllanthae. Members of Phyllanthaceae are pantropical and include trees, shrubs, phyllocladous taxa, semi – succulents, annual herbs, and even a free floating species. The recent classification of angiosperms recognises five lineages of *Euphorbiaceae sensulato* at family rank: *Euphorbiaceae sensu stricto*, *Pandaceae* Engl. & Gilg, *Phyllanthaceae* Martynov, *Picrodendraceae* Small and *Putranjivaceae* (APG III, 2009).

Most Phyllanthaceae, are uniform with pinnate venation, entire margins and simple indumentums. Flowers are small and actinomorphic but display great diversity in shape, size and number of floral organs (Hoffmann *et al.*, 2006; Kathriarachchi *et al.*, 2006). The genus *Phyllanthus* L. was first described by Linnaeus in 1753 and consists of ca.833 species (Govaerts *et al.*, 2000) in the world and is chiefly distributed in moist humid tropics. In India, it is represented by ca. 40 species (Henry & Santapau, 1973), although Hooker f. (1887) has recorded 56 species from the then British India. In total, 12 species of herbaceous *Phyllanthus* have been identified in India.

Webster (1955, 1956a, b, 1957, 1958, 1967, 1970 and 1994; Webster and AiryShaw, 1971) has worked exhaustively on *Phyllanthus* and has provided detailed taxonomy. In course of botanical exploration of local region of Southern Eastern Ghats some exciting specimens of *Phyllanthus* were collected. After critical examination and perusal of literature, these specimens were determined as belong to a distinct new taxon of the genus *Phyllanthus*, which is being described here.

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Materials and Method:-

The plant specimens for collected from various parts of South India and the variations among these two plants were studied using fresh materials as well as herbarium specimens. The morphological and anatomical characters were recorded by examining several specimens of each species with the help of stereomicroscope and inverted phase-contrast microscope, respectively. Details about the distribution, habitat, local name and uses were taken from literature, herbarium data bank and field studies. Anatomical studies were conducted by hand sectioning and stained with safranin stain and glycerine.

Morphological study:-

The morphological characteristics of the specimens described herein as *Phyllanthus vasugii* sp.nov. were compared with those of the allied species, *P. urinaria* L.

Table.1:- The difference between of *Phyllanthus vasukii* sp. nov. and *Phyllanthus urinaria*.

Attributes	<i>Phyllanthus vasukii</i> sp. nov.	<i>Phyllanthus urinaria</i> L.
Habitat	Shrub	Herb
Leaf size and shape	5-20m long, 2-7mm broad, linear to oblong, When touched, the leaves not fold.	6-15m long, 3-15mm, broad oblong, When touched, the leaves fold in automatically.
Leaf apex	Mucronate	Mucronate
Stem colour and form	Dark green, round, not smooth, Branches and branchlets are stout and erect, tetra gon, very rarely branched, glabrous and woody at base.	Reddish-green, not round, smooth, Branches and branchlets are mostly weak and drooping, pentagonal, branched, glabrous and woody at base.
Fruit and flower colour	Greenish fruits and whitish green tepals. 6-capillary fruit. 6 dehiscent cocci each fruit.	Reddish fruits and reddish green tepals. 3-carpellary fruit. 3 dehiscent cocci each fruit.
Number of tepals	Hexatepalous (6)	Hexatepalous (6)
Nature of stipules	Greenish and closely attached	Reddish and laterally free
Plant height	Up to 150 cm high	Up to 60 cm high

Micromorphology study:-

Table.2:- Difference between anatomical Characters of the *Phyllanthus urinaria* and *Phyllanthus vasukii* species (Fig.2.).

Part of the plant species	Characters	<i>Phyllanthus urinaria</i>	<i>Phyllanthus vasukii</i>
LEAF	Number of palisade parenchyma in the mesophyll.	2 layer thick	1 layer thick
	Nature of intercellular spaces	Large	Small
	Nature of xylem cells within the mid rib	Clustered cells 2 layers thick	Clustered cells 3 layers thick
STEM	Epidermal circumference	Regular angular	Irregular angular
	Ridges and Furrows	Present	Present
	Hypodermis (Collenchyma)	Rectangular 2-5 layers thick on ridges 1-2 layers thick on furrows	Rectangular 2-4 layers thick on ridges 2 layers thick on furrows
	Oil ducts	Absent	Present
	Cortex (Parenchyma)	Oval & Polygonal 2-5 layers thick	Polygonal 1-9 layers thick
	Pericycle (Sclerenchyma)	2-4 layers thick	3 layers thick

ROOT	Xylem	2-5 elongated cells	2-8 elongated cells
	Phloem	3-5 elongated cells	2-8 elongated cells
	Ground tissue	Small	Very large
	Cork cells	8-11 cell thick	2-7 cell thick
	Cortex	2-4 cell thick	6-9 cell thick
	Xylem	11-43µm	8-25 µm
	Phloem	4-8 cell thick	8-11 cell thick

To distinguish *Phyllanthus vasukii* and *Phyllanthus urinaria* between lateral, dorsal and ventral ornamentation of the seed surfaces was analyzed by microscopy. Three types of surface ornamentation patterns were presented in the Table-3.

Table.3:- Micromorphology of seeds of features between of *Phyllanthus vasukii* and *Phyllanthus urinaria*.

Taxa Seed ornamentation <i>Phyllanthus L.</i>	Seed ornamentation		
	Dorsal	lateral	Ventral
<i>Phyllanthus vasukii</i>	Transversal ribs, aligned on a irregular microverrucose on ribs, with valleys	Stellate verrucose regular concentric C-shape alignment, with crusts	Regular or irregular concentric C-shape ribs, regular longitudinal finger-shape rodlets
<i>Phyllanthus urinaria.</i>	Transversal ribs, microverrucose on ribs, crusts on valleys	Asymmetrical longitudinal ribs valleys, microverrucose on ribs, crusts on valleys	Asymmetrical longitudinal ribs, microverrucose on ribs sometimes branched ribs, crusts on valleys

***Phyllanthus vasukii* parthipan et al., sp. nov.**

Shrub woody, erect, up to 150 cm high. Leaf sessile, bipinnate, opposite, entire, leaf let linear to oblong, pale green, 5-20 x 2-7 m, mucronate apex, the base slightly oblique, unequal; stipules linear, very finely puberulous on lower sides leaf margin; lateral veins in 3–5 pairs; reticulation inconspicuous on both sides. Stem green, suffrutescent, sticky angled, rarely branched, glabrous herb, 50-120 cm high, Stem diameter 2.0 – 2.9 mm. Rarely branchlets up to 35 cm long, with up to 50 leaves, Flowers, mostly bisexual fascicles, 6 petals, very small, 6-merous, axillary, ca. 1 mm in diameter. Higher leaf axils bear only solitary male flowers no paired, with female flowers in the lower axils zig- zag arrangement, single or sometimes paired. Male flowers : Pedicels 0.2–0.5 mm long; sepals 6, ovate or elliptic, 0.2–0.5 by 0.5–0.3 mm, white transparent green middle strip; disc glands 6, star-shaped or orbicular; stamens 3, staminal column 0.1–0.2 mm long, anthers 0.1–0.2 mm long. Female flowers : pedicels 1.0 – 1.5 mm long; sepals 6, obovate-oblong or lanceolate, 0.5–0.9 by 0.3–0.6 mm, glabrous, apex acute with broad red pinkish scarious margin with greenish sepals in the middle; disc glands 6; ovary covered with scurfy-tuberculate ; styles 0.2–0.9 mm long; stigma c. 0.5 mm long. Fruit green, pedicel 1.0–1.5 mm long, 0.5–1.5 mm diam, capsular, depressed globose, densely verrucous, cocci 3 , not spiny. Seeds are transversely ribbed on the back, and sides. Seed 2 in each cocci, 0.8–1.9 by c. 1 mm wide, 2-angled, curved on dorsal side, smooth sand in colour. (**Fig.1.**)

Flr. & Fr.: Throughout the year.

Etymology: The new species is named in honour of **DR.A.VASUKI**, Secretary, Kongunadu Arts & Sciences College, Coimbatore for her significant contribution in the field of Science.

Distribution: Endemic to Tamil Nadu part of South India.

Habitat and ecology: Growing along the cultivated land, especially wetland field, in moist grassy wetland. Namakal District (11°14'N 78°10'E / 11.23°N 78.17°E), Tamil Nadu, India.

Conservation measures:

The significance of field research is the detection of novel additions to a floristic region, which subsequently improve our understanding of plant biogeography, species diversity. This species is extremely very rare and known only from type locality with limited population density. This should be included in the Red Data Book of Indian plants. The known localities have already covered by Eastern Ghats. However, the new plant should be propagated by using biotechnology method and also to conserve under *ex-situ* conditions.

Conclusion:-

In present study, we observed that *Phyllanthus urinaria* differ from *Phyllanthus vasukii* and by their height, and leaves numbers, stem, When touched, the leaves not in fold in automatically, Six capillary fruit. 6 dehiscent cocci in each fruit. Anatomical studies also confirmed the new species *Phyllanthus vasukii* by Nature of intercellular spaces large in leaf, Epidermal circumference in regular angular stem and oil ducts present in stem. Entirely different of number of root arrangement cells, *etc.* Micromorphology of seeds of features very different between of *Phyllanthus vasukii* and *Phyllanthus urinaria*. By this, we confirmed that *Phyllanthus vasukii* can be treated as a new species due to the presence of variation in both morphological, Micromorphology of seeds and anatomical characteristic features.

Acknowledgement:

We extend our sincere thanks to Professor and Head, Dept. of Botany, Bharathiar University, Coimbatore.

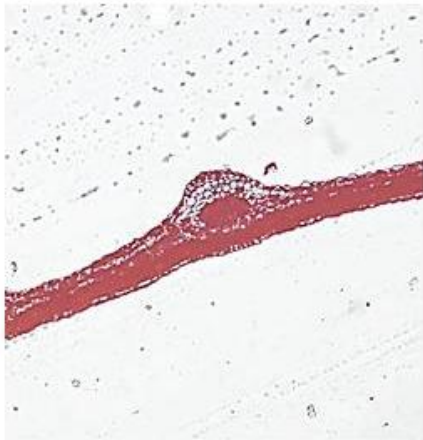


Fig.1:- *Phyllanthus vasukii* Parthipan et al.,

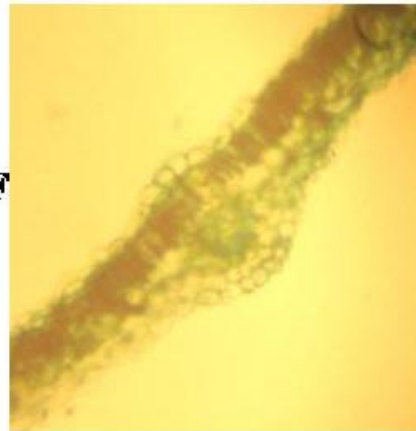
Fig.1: Difference between anatomical Characters of the *Phyllanthus urinaria* (Daniel *et al*, 2015) and *Phyllanthus vasukii* species .

***Phyllanthus vasukii* sp. nov.**

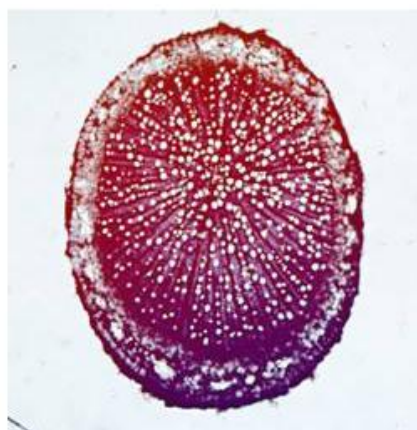
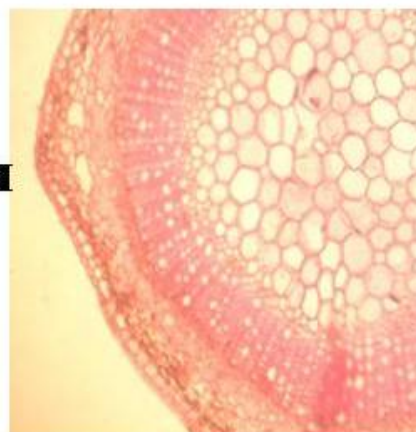
***Phyllanthus urinaria* L.**



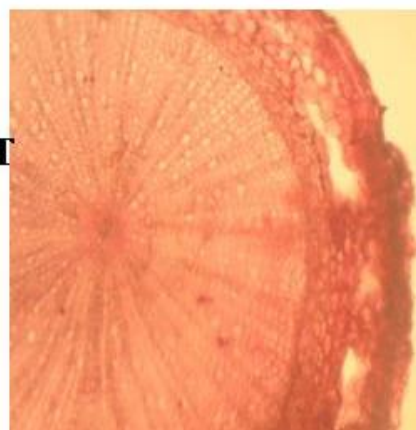
LEAF



STEM



ROOT



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RESEARCH ARTICLE

PRSE - CYCLICAL LEARNING STYLES AND STAGES BASED ON THE RESEARCH OF KURT LEWIN, DAVID KOLB, JOHN DEWEY, JEAN PIAGET.

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Abstract

The research thesis of this study is that matching students' learning style preferences with the complementary course syllabus and instruction improved academic achievement and student attitudes toward learning based on the research work of Kurt Lewin, David Kolb, John Dewey, Jean Piaget. The composition of this study method is both descriptive and exploratory. In the first part of the study, the qualitative research method was used to overview the literature background of the study. In the empirical part of the study, the factor analysis, using the Principle Axes Factoring method – PAF, was used to extract learning styles.

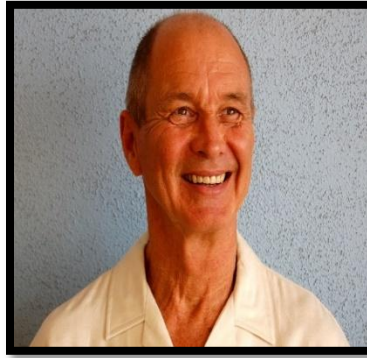
This study has four aspects. First, it outlines the literature review, summarizing learning styles taxonomy. Then it covers research framework and methodology, including data collection, sample characteristics, variables description and data analysis and tests the learning styles theories using factor analysis. Finally, it discusses the results by recognizing some limitations and by providing pedagogical implications and further research.

This article explains the cyclical learning styles like participating, reflecting, structuring and experimenting, based on the combined research work of many notable researchers. The concept of learning styles is embedded in different academic literature and researched from different approaches, including intelligent learning systems (Laureano-Cruces et al., 2006), a genetic algorithm approach to students' learning styles (Yannibelli et al., 2006), a web-based education perspective on learning styles (Garcia et al., 2007), learning about and through aesthetic experience (Welsh et al., 2007), use of business case studies in the learning process (Duff et al., 2008), problem-solving strategies within learning styles (Metallidou & Platsidou, 2008), preferred learning styles (Peters et al., 2008) and an adaptive learning system perspective of learning styles (Tseng et al., 2008).

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Introduction:-

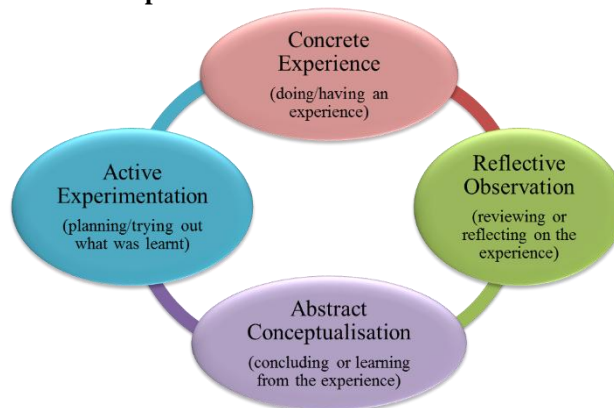


David Kolb.

Born 1939, he is an American educational theorist. He is the founder-chairman of Experience Based Learning Systems, Inc. (EBLS) whose interests and publications focus on:

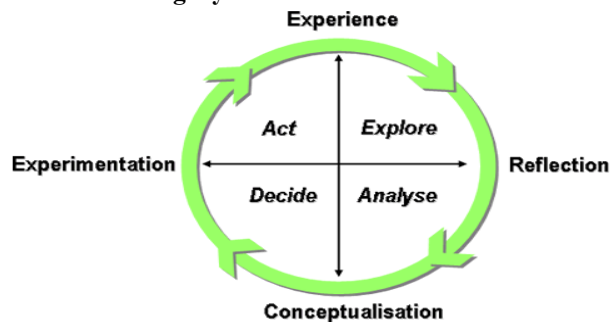
- experiential learning,
- the individual and social change, career development
- executive and professional education

David Kolb (1984) said that (deeper) learning runs through a cycle of **concrete experience, reflective observation, abstract conceptualization** and **active experimentation**.



Applying lessons learned into future actions provides the basis for another cycle of learning.

Kolb's Learning Cycle:-



- The left side of the vertical arrow represents **doing tasks**, the right side **observing tasks**.
- The upper half represents **feeling** (being creative and emotional), the bottom (logical) **thinking**.

- People usually have their own preferences in one of the four learning styles: they are more **exploring**, **analyzing**, **decision-making** or **acting** types.

A well-balanced team consists of people with different learning styles.

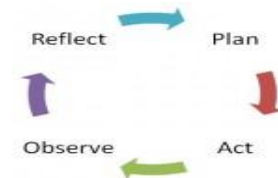


Kurt Lewin

Kurt Lewin, whom many consider to be the **father of modern social psychology** and the **study of organisational behaviour**, developed a four-stage model of action-research.

This model has been adapted by many others, the most notable of whom is David Kolb. The cycle starts when an individual encounters a problem within the environment. Then,

1. **Reflect** on what they know about situations like this.
2. **Plan** how they intend to proceed.
3. **Act** out their plan.
4. **Observe** the results their actions bring.

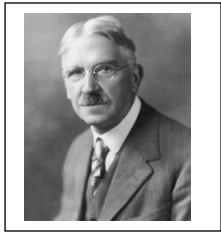


Jean Piaget

Swiss biologist and psychologist **Jean Piaget (1896-1980)** is renowned for constructing a highly influential model of child development and learning.

The concept of **cognitive structure** is central to his theory. Cognitive structures are patterns of physical or mental action that underlie specific acts of **intelligence** and correspond to stages of child development. There are 4 **primary cognitive structures** (i.e., development stages) according to Piaget:





John Dewey

John Dewey (October 20, 1859 – June 1, 1952) was an American philosopher, psychologist and educational reformer whose ideas have been influential in education and social reform.

Dewey was an important early developer of the philosophy of pragmatism and one of the **founders of functional psychology**. He proposed that education be designed on the basis of a TOE - **theory of experience**.

Central Tenets Of Dewey's Theory:-

Dewey's theory of experience rested on two central tenets:-

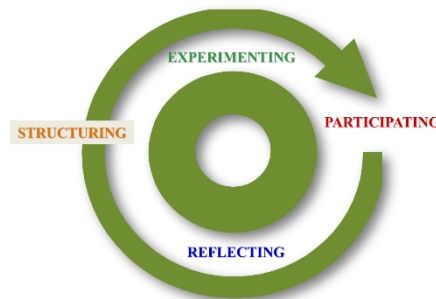


Continuity refers to the notion that humans are sensitive to (or are affected by) experience.

Humans survive more by learning from experience after they are born than do many other animals who rely primarily on pre-wired instinct.

Interaction builds upon the notion of continuity and explains how past experience interacts with the present situation, to create one's present experience.

Cyclical Learning Styles:-



Learning Is Cyclical In Nature - Kurt Lewin, David Kolb, John Dewey, Jean Piaget:-

Learners are **actively involved** (participating) in some form of learning event. They **reflect** back on the activity - either alone or in consultation with others, to develop an understanding of what they've **experienced**. They understand based on their **Participation** and **Reflection**. This helps them develop a generalized set of theories or rules (**Structuring**) of how things should work.

They **experiment** the theories to assess their validity that may help identify the **most practical approach** to achieving the desired results. Now, learners **participate** in a new learning 'event', repeating the experiential learning cycle once again.

The learning styles literature has had a revival during the past years, especially in the first decade of the 21st century (Alban & Metcalfe 2002; Duff & Duffy, 2002; Dunn & Griggs, 2003; Loo, 2004). Since 2007 and 2008, there has been an increasing interest in the potential of experiential learning (Reynolds & Vince, 2007; Argyris, 2007; Welsh

et al., 2007; Hornyak et al., 2007; Herbert & Stenfors, 2007; Hyde, 2007; Kayes, 2007 and Armstrong & Mahmud, 2008).

The Learning Style Questionnaire (PRSE) has been proposed as an alternative for Kolb's Experiential Learning Style Model (ELM). The PRSE is designed to probe the relative strengths of four different learning styles: Participator, Reflector, Structure and Experimenter. The authors' intention is that learners should become proficient in all four stages of the learning cycle. The authors are keen to emphasize that 'no single style has an overwhelming advantage over any other. Each has strengths and weaknesses, but the strengths may be especially important in one situation, but not in another'. These four styles correspond approximately to those suggested by Kolb's (1999) Experiential Learning Model (ELM): active experimentation (Participator), reflective observation (Reflector), abstract conceptualization (Structure), and concrete experience (Experimenter).

Participators involve themselves fully and without bias in new experiences. They are open-minded, not skeptical, and this tends to make them enthusiastic about anything new. Their philosophy is "I'll try anything once". They tend to act first and consider the consequences afterwards. Their days are filled with action. They tackle problems by brainstorming. As soon as the excitement from one action has died down, they are busy looking for the next. They tend to thrive on the challenge of new experiences but are bored with implementation and longer term consolidation.

- Flexible
- Gets bored with consolidation
- Happy to give things a try
- Open-minded
- Optimistic about change
- Rushes into action without preparation
- Takes immediate obvious action
- Takes unnecessary risks

Reflectors like to stand back to ponder experiences and observe them from many different perspectives. They collect data, both first hand and from others, and prefer to think about it thoroughly before coming to any conclusion. The thorough collection and analysis of data about experiences and events is what counts so they tend to postpone reaching definitive conclusions for as long as possible. Their philosophy is to be cautious. They are thoughtful people who like to consider all possible angles and implications before making a move.

- Careful
- Good listener
- Holds back from participation
- Methodical
- Does not jump to conclusions
- Slow to decide
- Thorough and thoughtful

Structures adapt and integrate observations into complex but logically sound theories. They think problems through in a vertical, step-by-step logical way. They assimilate disparate facts into coherent theories. They tend to be perfectionists who won't rest easy until things are tidy and fit into a rational scheme. They like to analyze and synthesize. They are keen on basic assumptions, principles, theories models and systems thinking. Their philosophy poses rationality and logic. "If it's logical, it's good". Questions they frequently ask are: "Does it make sense?" "How does this fit with that?" "What are the basic assumptions?" They tend to be analytical.

- Disciplined
- Intolerant of subjective, intuitive ideas
- Logical
- Low tolerance of uncertainty, ambiguity
- Objective
- Parental in approach
- Rational

Experimenters are keen on trying out ideas, theories and techniques to see if they work in practice. They positively search out new ideas and take the first opportunity to experiment with applications. They are the sort of people who come back from learning experiences brimming with new ideas that they want to try out in practice. They like to get on with things and act quickly and confidently on ideas that attract them. They are essentially practical, down-to-earth people who like making practical decisions and solving problems.

- Business-like – gets to the point
- Does not like theory
- Impatient with waffle
- Keen to test things out in practice
- Practical, down to earth, realistic
- Rejects ideas without clear application
- Task and technique focused

Research Methodology:-

In this research, data were analyzed using the Statistical Package for the Social Sciences (SPSS 16.0). An alpha level of 0.05 was used as a margin of statistical significance (Coakes & Steed, 2003). The factor analysis using the Principle Axes Factoring method - PAF was used to extract learning approaches (Miller et al., 2002; Coakes & Steed, 2003). The essential purpose of factor analysis is to describe the variation among many variables in terms of a few underlying, but unobservable, random variables called factors. The underlying assumption of factor analysis is that there exists a number of unobserved latent variables (or "factors") that account for the correlations among observed variables, such that if the latent variables are partially out or held constant, the partial correlations among observed variables all become zero. In other words, the latent factors determine the values of the observed variables. One of the most frequently used techniques for factor extraction is the Principal Factor Method, where factors are extracted in such a way that each factor accounts for the maximum possible amount of the variance contained in the set of variables being factored (Miller et al., 2002).

Nature Of The Items:-

All the items in the questionnaire belong to the content domain which facilitate in assessing an individual's personality type. The 20 items in the questionnaire describe multiple situations in which an individual is most likely to respond in various environments through which his personality characteristics are displayed. The items are repetitive in a way so as to track the individual's response to a certain-stimuli which best predicts his personality type across situations. The items are standardized as they are the same for every respondent with respect to the content, form and order

Data Collection And Sample Characteristics:-

The study sample included 527 students chosen by random sampling. Students were anonymously interviewed using online questionnaires. This was to a certain level an exploratory research, setting a base for further research in this field.

Data Analysis – Factor Analysis:-

The applicability of factor analysis was tested using the Kaiser-Meyer-Olkin Measure of Sampling Adequacy (KMO measure) and Bartlett's Test of Sphericity. The KMO measures the sampling adequacy which should be greater than 0.5 for a satisfactory factor analysis to proceed. The applicability criteria were the KMO measure being > 0.6 and χ^2 test statistically significant (Miller et al., 2002). Another indicator of the strength of the relationship among variables is Bartlett's test of sphericity. Bartlett's test of sphericity is used to test the null hypothesis that the variables in the population correlation matrix are uncorrelated. The observed significance level is .0000. It is small enough to reject the hypothesis. It is concluded that the strength of the relationship among variables is strong.

Kmo And Bartlett's Test:-

Measure of factor analysis applicability PRSE learning style theory:-

Kaiser-Meyer-Olkin measure of sampling adequacy	0.675
Approx. Chi- Square (Bartlett's test of sphericity)	259,572
Df - (Bartlett's test of sphericity)	78000
Sig. (Bartlett's test of sphericity)	0.000

Analyses And Findings:-

The factors extracted from the group of variables were labeled reflectors (best explaining four variables Vs3, Vs6, Vs11,17), Structure (best explaining five variables Vs10, Vs4, Vs1, Vs16, Vs18), Participators (best explaining four variables Vs5, Vs9, Vs19, Vs20), and pragmatists (best explaining five variables Vs7, Vs12, Vs13, Vs14, Vs15), confirming the applicability of PRSE Learning style questionnaire.

	Raw Factors					Rescaled Factors			
	1	2	3	4		Participator	Reflector	Structure	Experimenter
Vs3	.053	.766	-.078	-.227		.053	.773	-.079	-.229
Vs6	-.193	.621	.073	.060		-.200	.645	.076	.063
Vs11	.136	.694	.110	.126		.143	.732	.116	.133
Vs10	.242	.162	.510	.072		.260	.174	.548	.077
Vs4	.087	.029	.737	.299		.080	.027	.677	.274
Vs1	.271	.145	.306	.258		.346	-.185	.390	.329
Vs5	.816	-.067	.272	.069		.806	.066	.268	.068
Vs9	.673	.117	.029	.031		.632	.110	.027	.029
Vs7	.415	-.120	.200	.296		.430	-.124	.208	.307
Vs12	.102	-.180	.011	.592		.113	-.200	.012	.636
Vs13	.336	-.054	.185	.545		.394	-.063	.217	.640
Vs15	.020	.069	.218	.611		.022	.077	.245	.686
Vs14	-.012	.195	.131	.542		.014	.223	.149	.616
Vs19	.241	.127	.031	.214		.765	.114	.291	.331
Vs17	.132	.068	.263	.365		.047	.625	.034	.035
Vs20	.085	.172	.521	.431		.753	-.052	.231	.627
Vs18	.339	-.062	.035	.131		.357	.123	.612	.271
Vs16	.427	.165	.343	.071		.211	.043	.614	.072

This rule of thumb is cited in Schneider (2003). He indicated that factor loadings greater than 0.30 or less than -0.30 are considered significant, loadings greater than 0.40 or less than -0.40 are considered more important and loadings greater than 0.50 or less than -0.50 are considered very significant.

In order to assess the reliability of compound scales (the extracted factors) measuring applied learning styles concepts, the Cronbach Alpha Coefficient was calculated for the sample as a whole.

<u>Factors</u>	<u>Cronbach Alpha</u>
Participator	0.688
Reflector	0.743
Structure	0.630
Experimenter	0.744

Cronbach's Alpha measures how well a set of items (or variables) measures a single unidimensional latent construct. Cronbach's Alpha is not a statistical test, rather it is a coefficient of reliability (or consistency) the reliability coefficient α of 0.7 or higher is considered "acceptable" in most social science research situations (Coakes & Steed, 2003). As indicated, the results of factor analyses are close to satisfactory: Factors for the analysis have Cronbach Alpha values from 0.630 to 0.744. These results indicate that the extracted factors appropriately characterize the dimensionality of the data.

The aim of this study is to explore the widely used cyclical learning styles theory in Participator, Reflector, Structure, and Experimenter among the teenage student population. By analyzing the teenage student's learning approaches, typical patterns have been discovered. The research confirmed the results through qualitative meta-analysis and quantitative factor analysis. The study resulted in a clear extraction of four theoretically expected learning styles dimensions (factors – Participators, Reflectors, Structures, and Experimenters).

Recent thinking in this area suggests that unlike cognitive personality styles, learning styles can be modified to a degree through learning and training strategies. Instead of matching training to the styles of the learners, it could be

more rewarding to expose learners to a mismatched learning environment in order to help them develop a wider repertoire of coping behaviours and learning strategies. Those that can learn to use a variety of problem-solving and learning strategies, and apply them in situations that do not match with their natural learning style, may be more able to perform effectively across a wider range of situations than those who have limited stylistic versatility (Hayes & Allinson, 1996).

According to the research thesis of this study, we can summarize that matching students' learning-style preferences with the complementary course syllabus and instruction improved academic achievement and student attitudes toward learning. The mission of education is to create and disseminate knowledge to enable students' successful entry into the adult world. Teaching community need an awareness of the learning style preferences of students in order to develop and utilize effective and efficient teaching and pedagogical strategies and methods.

A significant number of researchers (Honey & Mumford, 1992; Armstrong & Mahmud, 2008) have argued that learning styles are not determined by inherited characteristics, but are developed through experience. Styles are therefore not necessarily fixed, but can change over time, even from one situation to the next. The implications regarding the learning strategies implementation in education suggest that students who are aware of a range of learning strategies are more likely to select the correct one for a particular task. The approach of the flexible learning style strategy is best suited to the case-study method of teaching. For the educators in educational institution, the challenge is to provide meta-cognitive support for students, enabling them to reflect not just on what they learn but also how and why.

The development of these new skills and knowledge requires a variety of teaching methods and learning strategies in order to match students' learning style preferences. Therefore, teachers/lecturers need an awareness of the learning style preferences of students in order to develop and utilize effective and efficient teaching and pedagogical strategies and methods. Recognizing students' learning styles allows educators to effectively lecture to a diverse population of students with different learning style preferences. Being an effective teacher implies matching individual learning style preferences among students with a collective course syllabus in teaching strategies.

Limitations:-

The most prominent deficiency of the research is that it does not recognize the dimension of time. Namely, the concept of this research is inherently static. Therefore, further analysis should focus on determining those developments - styles are not necessarily fixed, but can change over time. As well, from the methodological perspective of the research process, regarding the employment of the construct reliability, the average variance extracted and composite reliability index should be engaged too.

Conclusions:-

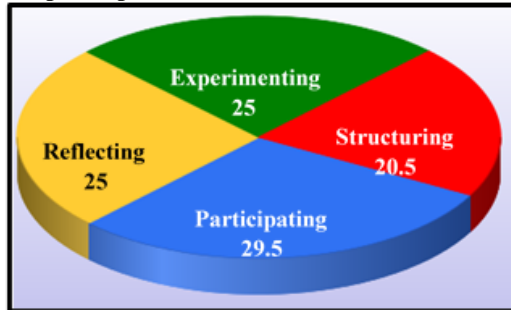
Researchers have pointed out that students learn effectively in a harmonic environment and by using teaching aids which match the students' learning style preferences (Li et al., 2008). The concept of the learning style has a broad meaning. In this research, it is proposed and defined as an individual's preferential focus on different types of information, the different ways of perceiving the information, and understanding the information (Li et al., 2008). The learning styles literature has had a revival during the past years, especially in the first decade of the 21st century (Alban & Metcalfe 2002; Duff & Duffy, 2002; Dunn & Griggs, 2003; Loo, 2004). Upon reviewing the literature on learning styles, the intense rate and growing interest is involved.

The implications for pedagogy indicate that instead of fixed learning styles strategies, adapting content to the learner, educators should rather implement flexible learning strategies. The implications regarding the learning strategies implementation in education suggests that students who are aware of a range of learning strategies are more likely to select the correct one for a particular task. The ultimate goal for the educators in educational institution is to provide meta-cognitive support for students, enabling them to reflect not just on what they learn but also how and why, thereby helping them to 'learn how to learn'.

The logic of lifelong learning suggests that students will become more motivated to learn by knowing more about their own strengths and weaknesses as learners. Consequently, if teachers can respond to individuals' learning style preferences, then the achievement rate is likely to rise and "learning to learn" skills of students may provide the foundation for the lifelong learning concept.

Sample Report:-

PRSE - Participating, Reflecting, Structuring and Experimenting: PRSE assesses the approach (Participating, Reflecting, Structuring and Experimenting) an individual takes to organize and internalize information. The individual's scores provide an objective analysis to understand the means by which he/she organizes new learning.

Sample Report Of "A":-**Analysis:-**

- Being naturally intuitive, the subject tends to form connections between concepts easily. This allows him/her to form ideas and enhances his/her ability to address problems effectively
- The subject tends to enjoy a non-restrictive learning environment that nurtures his/her creativity and gives the opportunity to express his/her ideas/opinions/views without bias
- Often, he/she tends to use fantasy and imagination as a ready resource to facilitate learning
- Taking a holistic approach, he/she likes to gather information from various sources and seek a well- rounded understanding of a topic
- Being people oriented, he/she would work well in a team; and would benefit from learning from or teaching others. Hence, he/she would tend to reflect on his/her learning alongside others
- Seeking intellectual stimulation, he/she may tend to ask lots of 'why' questions in order to gain better clarity on the topic
- Placing importance on practical application of his/her ideas, he/she also tend to focus on honing his/her technical skills
- Being energetic and enthusiastic, he/she would be open to volunteering in an activity and learning by doing
- The use of examples would trigger his/her ability to visualize the concept and connect it with his/her previous experiences. Hence, he/she would benefit greatly with real or hypothetical examples
- He/she prefers relating learning to a personal and emotional experience, thereby, enjoy personalized attention and emotional support during a learning event
- With a need to personally experience things, he/he may enjoy learning things through self-discovery
- He/she comes across as a person who is open to new ideas, and would reflect on the same through open discussions, debates and brainstorming sessions
- He/she would be willing to take risks in order to achieve objectives and complete tasks
- He/she seeks to understand "why" things happen the way they do; hence, he/she is likely to benefit by asking questions and seeking additional information
- Driven by his/her emotions and relationships with others, he/she would be influenced by his/her teacher to a large extent. Hence, he/she would exhibit a particular interest in subjects that are taught by his/her favorite teachers

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RESEARCH ARTICLE

CANTILEVER BEAM-LIKE DESIGN OF RC RETAINING WALL WITH MULTIPLE PRESSURE RELIEF SHELVES AND ELASTIC FOUNDATION.

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Design, Rankine 's Lateral Active Earth
Pressure

Abstract

In this study the magnitude of reduction in total active earth pressure, overturning moments at the bottom of the wall and its distribution due to the response of a relief shelf in a retaining wall is presented with cantilever type retaining wall on cohesionless soils with increase of additional resisting moments at the bottom of the wall. Retaining wall is considered as cantilever beam-like, multi-rows and stepwise reinforced concrete, multi-shelves on vertical wall and elastic foundation. A numerical study is conducted to investigate the effect of the number of shelves; shelf and wall stem rigidity and shelf horizontal location on the resulted lateral earth pressure distribution. Pressure quantity, the maximum acting bending moment and shear force on the wall are also discussed to perform the retaining design. Currently, numerical analysis is one of the easiest and the fastest method to examine the effect of each factor on the dynamic behavior of a retaining wall. According to the analysis it was found that the shelves have a significant effect on the distribution of the earth pressure. The numerical results indicate that the presence of a relief shelf behind the wall would result in a reduction of the earth pressure and also results show that shelf inclusions have positive role as pressure detractive for cantilever retaining walls in earthquake areas.

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Introduction:-

A retaining wall will be subjected to an additional load under the seismic and wave conditions and caused not only by the dynamic lateral earth pressure and wave force but also by the inertial force due to the structural mass reduced by pressure relief shelves. The lateral force acting between the retaining structure and the backfill mass is termed as lateral earth pressure. Cantilever retaining walls with pressure relief shelves are considered one special subset of retaining walls (Farouk,2015). The concept of providing pressure relief shelves towards the active soil mass side of a retaining wall reduces the total earth pressure on the wall, which results in reducing the thickness of the wall and ultimately to get an economic design by use of less reinforcement on wall horizontal cross section on the level of contraction joints (Farouk,2015). Over the decades, several types of methods have been developed to calculate the complex dynamic soil interactions between a retaining wall and retained earth mass. These methods can be categorized into limit state analyses, closed form solutions, numerical analyses, and experimental methods (Padhye,2011). Jumikis (1964) presented the provision of one or more relief shelves to increase the stability of retaining wall. The relief shelves have an advantage of decreasing the overall lateral earth pressure on the structure.

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An economical design is reached because of less material use in construction mechanism as compared to massive structure of cantilever retaining walls without shelves (Jumukis,1964). Chaudhury (1973) proposed Coulomb's theory for earth pressure computation for cohesionless soil. He presented the charts for various locations and widths of relief shelf. and compared cantilever and counterfort type retaining wall in reinforced concrete with and without relief shelf to show the economy in providing a relief shelf (Chaudhury,1964). According to Phatak (1975) the effect of relief shelves can increase the stability of retaining wall by using the Rankine's theory to evaluate the lateral pressures (Phatak,1975). He presented experimental study on flexible cantilever wall with relief shelf to show substantial reduction in earth pressure (Phatak,1975). In order to facilitate the reduction of earth pressure and increase stability against sliding and overturning, construction of relief shelves behind retaining wall was used (Banerjee,1977). Padhye and Ullagaddi (2011) presented the procurement of one or more relief shelves to increase and to provide the stability of retaining wall. They considerably applied Coulomb's theory for analysis and design of cantilever type of wall for cohesionless soil (Padhye,2011).

In this study, RC retaining wall to provide both of lower overturning moments caused by decreasing lateral active earth pressure forces and lower opposing bending moments originated from gravity weights of shelves are analytically analyzed on elastic foundation. Special retaining wall is designed and analyzed holistically under the conditions such as cantilever beamlike vertical section, doubly reinforced rectangular base section on elastic foundations and vertical wall with multi-shelf, stepwise and three-rows reinforcement.

Methodology:-

The retaining wall is designed against sliding and overturning using Rankin's earth pressure theory based on stress equilibrium of a soil mass element. The method of Rankine is still the most widely used to compute earth active thrusts on walls. For the wall geometry and ground conditions considered on this study, wall movements are reduced more effectively by providing multiple relief shelf than increasing the wall's base slab, at least if the wall's bending moment and stability remains unchanged. This paper deals mainly with seismic considerations. The stability of retaining wall checked to ensure that it is capable of supporting the design lateral forces. The stability regulations require that the selected retaining wall cast on the case study satisfy requirements for sliding, overturning and bearing capacity. The relationship between principal stresses when the soil reaches a state of plastic equilibrium can be derived from Mohr circle with a failure plane at an angle of $45 + \phi'/2$.

The active pressure coefficient is has been proposed by Rankine's theory as (Kip et al.1999)

$$K_a = \cos\alpha \frac{\cos\alpha - \sqrt{\cos^2\alpha - \cos^2\phi'}}{\cos\alpha + \sqrt{\cos^2\alpha - \cos^2\phi'}} \quad (1)$$

Where α is an inclination angle with the horizontal surface and ϕ' is internal friction angle of soil. The horizontal stress for the above condition is defined as Rankine's active pressure (P_a):

$$P_a = K_a \gamma z - 2c'\sqrt{K_a} \quad (2)$$

Here, c' is the cohesion. A reinforcement computation and arrangement is designed to satisfy the bending moment values and shear force magnitude of the system. The comparison of the calculated bending moments with the required moments is carried out to prove the accuracy of the reinforcement. Length of the reinforcement is adjusted in order to satisfy the stability of the junction points of the shelf configuration. To prevent failure at the joints of shelves, stability of reinforced concrete demand an assessment of the process that control the behavior of the shear forces and bending moments. The retaining wall is an important part of the transportation and geotechnical systems. The damages of retaining wall in an earthquake create difficulties to the traffic flow with huge economic loss of rescue work (Li et al,2011).

Case Study:-

This study deals with the application of a new retaining system in order to reduce the weight of soil layers on the shelves more than a single shelf. A novel approach to providing external stability on retaining wall had come into existence to minimize earth pressure with the multiple pressure relief shelves. An application of multiple shelves is performed to eliminate the necessity of enlargement of the stem cross section. Instead of construct vertical wall

terracing, the integrity of the retaining system was conserved and the problems due to the geometric constraints were eliminated.

A reinforced concrete cantilever wall with relief shelves is designed to retain fill to a height of 14.3 meters. The backfill behind the retaining wall has a dry unit weight of about 18 kN/m^3 and shear strength parameters of $c' = 5$ and $\phi' = 40^\circ$. Additional features, such as the ground water height H_w and surcharge load q are shown in Figure 1 as a typical cross-section with the maximum design wall height. A surcharge load of 17 kN/m^2 was applied for traffic loading.

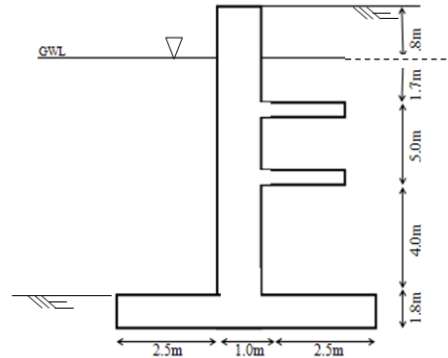


Fig. 1:- Retaining wall with multiple shelves.

The calculations of forces (total horizontal forces, R_H , total vertical forces, R_V) and moments (overturning moment, M_H , restoring moment, M_V) are set out in Table 1.

Table 1 Factors of safety against sliding and overturning with forces and moments.

Forces (t)		Moments (tm)	
R_H	21.75	M_H	109.39
R_V	59.72	M_V	226.48
Factor of safety			
Sliding	2.30		
Overturning	2.07		
Eccentricity	1.04		

The required number of stem reinforcement until the first shelf was 3 whereas number of stem reinforcement was 2 for second shelf above the base slab. Figure 2 shows the elevation and bending moments of retaining wall divisions with shear forces.

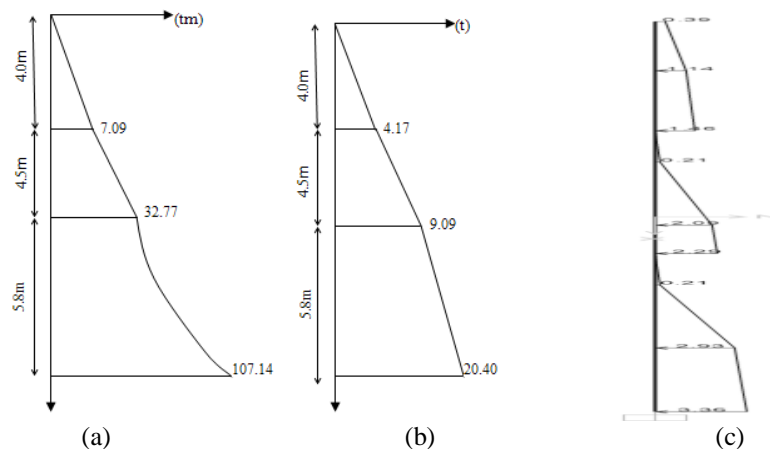


Fig. 2:- (a) Bending moments, (b) shear forces, (c) lateral pressures of stem part of wall

The cross sections and reinforcement details of wall are summarized in Figure 3a, 3b and 3c. Within three vertical sections of wall (base section-5.8m long, middle section-4.5m and top section-4.0m), the steel bars are placed in stepwise and three rows. Accordingly, number of steel bars and rows for unit-width wall (100cm) are indicated as in Fig.3.

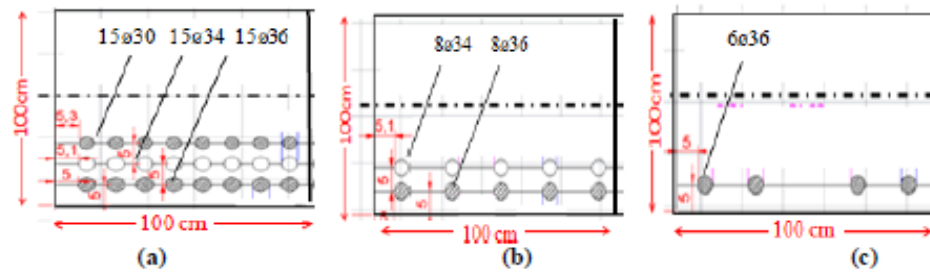


Fig. 3:- (a) Reinforcement of base section at 5.80 m-long, (b) Reinforcement of middle section at 10.30 m, (c) Reinforcement of top section at 14.30 m-long

First row reinforcement continues through the entire retaining wall. In each transfer region of reinforcement was taken as a quarter of reinforcement length. Figure 4 summarizes the theory of beam on elastic foundation.

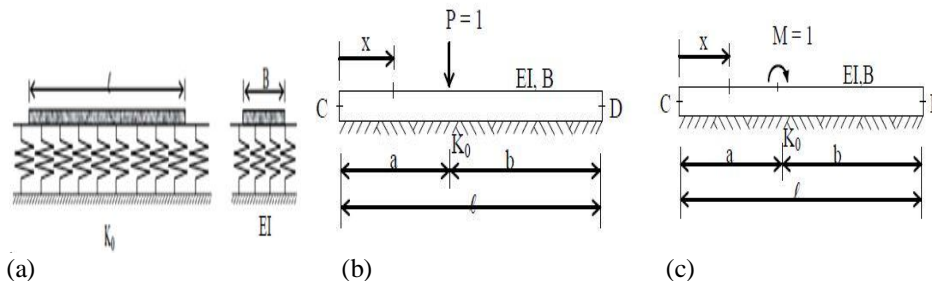


Fig. 4:- (a) Beam on elastic foundation, (b) Unit-concentrated loading on elastic foundation (c) Unit-bending moment loading on elastic foundation

Winkler assumes that reaction forces of the foundation are proportional at every point to the deflection of the beam at this point under applied external loads (Winkler, 1876). The elasticity of the foundation which follows the Hooke's law characterized by the force called the modulus of the foundation K_0 . Winkler's constant, K which includes the effect of the width of the beam and the characteristic of the system, λ are described by the following equations:

$$K = K_0 B \quad (3)$$

$$\lambda = \sqrt[4]{\frac{K}{4EI}} \quad (4)$$

The equations of shear force and moment in accordance with particular integral of beam on elastic foundation equations of concentrated moment and concentrated load can be expressed respectively as in Equations 5-8 (Kameswara Rao, 2011):

$$M(x)_M = -M \delta_{MM}^{(x)} \quad (5)$$

$$Q(x)_M = -\lambda \beta_{QM}^{(x)} = -\frac{\lambda l}{l} M \beta_{QM}^{(x)} = -M \delta_{QM}^{(x)} \quad (6)$$

$$M(x)_P = PL \frac{1}{\lambda l} \beta_{MP}^{(x)} = Pl \delta_{MP}^{(x)} \quad (7)$$

$$Q(x)_P = P \beta_{QP}^{(x)} = P \delta_{QP}^{(x)} \quad (8)$$

Here, $\delta_{MM}^{(x)}$, $\delta_{QM}^{(x)}$, $\delta_{MP}^{(x)}$, $\delta_{QP}^{(x)}$ values can be taken from charts and tables in practice (Kameswara Rao, 2011). For using easy-use of functions and integral terms including complex terms, the influence lines are generally generated as seen in Figure 4b and 4c. Figure 5 depicted the beam on elastic foundation mechanism in this study.

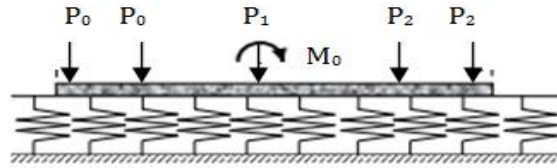


Fig. 5:- Beam on elastic foundation system of the retaining wall.

The computational model of a beam on an elastic foundation is often used to describe a lot of engineering problems and has application in geotechnics. In this study, an analytical solution of the problem of beam on elastic foundation was investigated bending moments and shear diagram were shown in Figure 6 and Figure 7. P_0 and P_2 represent respectively the water and water-soil concentrated loads on both cantilever parts of concrete base. P_1 is denoted as concentrated load originated from vertical part of concrete wall. M_0 is the bending moment at fixed support of the vertical part of RC wall which is assumed as cantilever beam and exposed to lateral active earth pressure. Sectional bending moments and shear forces of concrete base considered as beam on elastic soil are calculated by superposition of concentrated loads spaced at base and bending moment of vertical wall support placed at central point of base. Therefore, the bending moment and shear force diagrams required especially in RC design are plotted for both sides of concrete base of the vertical wall. RC design is carried out in accordance with Turkish Standards on Reinforced Concrete Design of Building (Turkish Standards-500,2000).

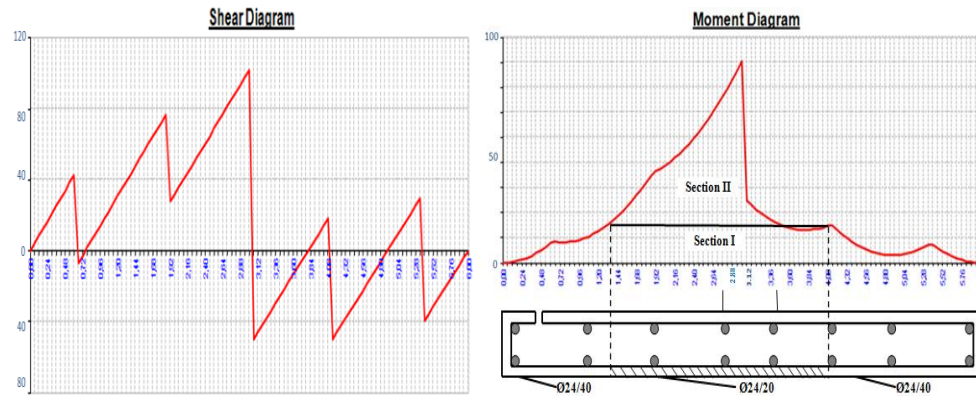


Fig. 6:- Shear and moment diagram of retaining wall as beam on elastic foundation.

In Figure 6, Section 1 considered as doubly reinforced rectangular section covers the 40cm-spaced steel bars 3#24 with the 600cm-long for the 100cm-width of base. In order to respond the maximum bending moment, the additional steel bars (more 3#24 with the 270cm-long) are placed only at Section 2. At the intersection length of base, the steel bars to be applied are 6#24. Base section where the support of vertical wall is located is therefore strengthened.

Conclusion:-

In this study, it is aimed that the lateral active earth pressure forces and their overturning moments can be decreased, the weight of double shelves and their protective moments can be increased, double reinforced rectangular section of base on elastic foundation can be equipped well and vertical steel bars in vertical retaining wall designed as cantilever can be placed by stepped and multiple rows reinforcement. Under these conditions, the required earth pressures, the overturning-protective moments and shear forces they causes, effect of elastic soil basement and reinforcement of vertical and horizontal rectangular wall sections are achieved. Results show that the proposed method can capture the displacements and bending moments of retaining wall more accurately than the retaining wall with single shelf. In this study, efforts were made to develop a multiple shelves system of retaining wall to satisfy successfully external and internal stability of the wall. The proposed method on special-designed RC retaining wall maximizes the friction capacity of the retaining system in presence of the multiple shelves. The purpose of this study is therefore realized by demonstrating the usage of multiple and elevated shelf on an elastically-founded retaining system with high backfill mass.

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RESEARCH ARTICLE

ANTIBACTERIAL ACTIVITY OF ENDOPHYTES FROM SELECTED MEDICINAL PLANTS

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Anti-bacterial activity, MIC, TLC.

Abstract

Bioactive compounds of endophytic fungi exhibit antimicrobial, antioxidant, anticancerous, antiviral and insecticidal properties. They harbor inter or intra-cellular in the plants epidermal layer. They are the most potent microorganisms in establishing interrelationship with plant without harming them or asymptotically. They have the mechanism which enables them to establish association with the plant and produce similar bioactive compound as plant produce. This research paper focused on the isolation, production, screening and separation of exploitable bioactive compounds from the plants Tulsi and Aloe vera. Total 5 fungi were isolated from segments of the plants. And the organisms taken for the study are both Gram-positive and Gram-negative bacteria i.e. *Staphylococcus aureus*, *Streptococcus pyogenes*, *Escherichia coli*, *Pseudomonas aeruginosa* respectively which were isolated from the clinical samples (like wound, blood and urine) and the results of antibacterial screening showed that (BCEF-01*), (BCEF-02*), (BCEF-03*), (BCEF-04*) and (BCEF-05*) inhibited all the test organisms. The inhibitory effect was promising to all the test organisms showing sufficient zone of inhibition with respect to all the fungal extracts.

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Introduction:-

Endophytic fungi are ubiquitous fungi that reside inter or intracellular in plant parts for at least a portion of their lives without causing apparent symptoms of infection and represent a large portion of fungal species (Petrini, 1991^[1]). Endophyte by definition is one which in the tissue beneath the epidermal cell layers and causes no apparent harm to the host. Endophytic fungi can be biotrophic, mutualists, benign commensals, decomposers or latent pathogens. According to Rodriguez *et al.*, (2009)^[2], all plants in the natural environment can shelter endophytic fungi, including algae, mosses, ferns, conifers and angiosperms. This fungal group appears to significantly influence the lifestyle of its host. Taxonomically most of the endophytic fungi belong to the phylum Ascomycota and its associated anamorphs, while some species belong to the phyla Basidiomycota and Zygomycota (Huang *et al.*, 2001 and 2008^[3, 4]). Dreyfuss & Chapela (1994)^[5] have estimated that approximately 1.3 million species of endophytic fungi remain to be discovered. This diverse fungal group could impact the ecology, fitness and shape of plant communities, conferring resistance to abiotic (temperature, pH, osmotic pressure) and biotic (bacteria, fungi, nematodes and insects) stresses. Endophytic fungi are an important source of bioactive molecules. These bioactive metabolites have a broad range of biological activities and could be the starting materials for

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pharmaceuticals or lead structures for the development of pharmaceutical or agrochemical products. These endophytes protect their hosts from infectious agents and adverse conditions by secreting bioactive secondary metabolites (Carroll and Carroll, 1978^[6]; Azevedo *et al.*, 2000^[7]; Strobel, 2003^[8]). The substances produced by endophytic fungi originate from different biosynthetic pathways, including isoprenoid, polyketide and amino acid and belong to diverse structural groups, such as terpenoids, steroids, xanthenes, quinones, phenols, isocoumarins, benzopyranones, tetralones, cytochalasins and enniatins (Schulz *et al.*, 2002^[9]). Indeed, these bioactive molecules represent a chemical reservoir for discovering new compounds, such as antibiotic, antioxidant, immunomodulating, anticancer, antiparasitic and antidiabetic compounds, for use in the pharmaceutical and agrochemical industries.

Ocimum species belongs to the family Lamiaceae are very important for their therapeutic potentials. Tulsi leaves contain bright yellow volatile oil reported to possess the antimicrobial properties and also act as insecticide. Tulsi is a fragrant bushy perennial growing up to 1.5 m in height with profusions of white blooms and slightly purple tinted foliage. This herb has been known from as early as the Vedic period and is held sacred by the Hindus and is often planted around temples and used in rosaries. It is native to India, reached Western Europe in 16th century. Tulsi leaves are traditionally used for getting relief from common cold, bronchitis, cough, mild indigestion, diminished appetite and malaise. *Ocimum* (tulsi) are also known for its antimicrobial, immunomodulatory, anti-inflammatory, anti-pyretic, anti-asthmatic, hypoglycemic, hypotensive and analgesic activities. The only side effect reported is constipation.

Aloe vera is a perennial, drought-resisting, succulent plant belonging to the Asphodelaceae family. The name, aloe, is derived from the Arabic "alloe" or Hebrew "halal" meaning bitter shiny substance. It has a vast traditional role in indigenous system of medicine like ayurveda, siddha, Unani and homoeopathy. *Aloe barbadensis* miller or *Aloe vera*, a semi tropical plant is one of the 250 species of Aloe. There are more than 200 compounds found in *Aloe barbadensis*, about 75 of which have biological activity, *Aloe vera* leaves contain a diverse array of compounds, including anthraquinones (e.g. aloe-emodin), anthrones and their glycosides (e.g. 10-(1, 5'' anhydroglucosyl)-aloeemodin- 9-anthrone, also known as aloin A and B), chromones, carbohydrates, proteins, glycoproteins, amino acids, organic acids, lipids, sugars, vitamins, minerals. Various studies have revealed that *Aloe vera* leaf possesses many pharmaceutical activities, including antimicrobial, anticancer, antioxidant, antidiabetic, antiulcer, hepatoprotective, immunomodulatory and many more activities. Many of the health benefits associated with *Aloe vera* have been attributed to the polysaccharides contained in the gel of the leaves.

Research on endophytes dates back to over one hundred years. During this period, several aspects of endophyte biology were thoroughly studied, including the diversity, taxonomy, reproduction, host ecology and effects on the host. Because natural selection favors the evolution of beneficial endophytic strains, several endophytes were found to secrete secondary metabolites that protect plants against insect pests, pathogenetic organisms as well as herbivores, thus, endophytes represent a promising source of novel, biologically active metabolites for pharmacological and agricultural applications. Biochemical research revealed that a wide variety of natural products can be obtained from endophytic microbes. Natural products from endophytic fungi were observed to inhibit many pathogenetic organisms including bacteria, fungi, and viruses.

Materials and Methods:-

Sample collection:-

Healthy (showing no visual disease symptom) and mature plants were carefully chosen for sampling. Fresh plant materials (branches, leaves and roots) were collected from ten different sites at Bhoopsandra area, Bangalore, India. The plant material was brought to the laboratory in sterile bags and processed within few hours after sampling. Fresh plant materials were used for the isolation work to reduce the chance of contamination.

Glassware, Chemicals and Media:-

The glassware used were made up of borosilicate glass obtained from M/s Borosil India Limited. The chemicals used were of analytical grade obtained from M/s Himedia Laboratories Pvt. Limited & M/s Sigma-Aldrich Pvt. Limited, Mumbai, India. The Media used for the experiments were obtained from M/s Himedia Laboratories Pvt. Limited, Mumbai, India.

Sample Processing:-

For Tulsi:-

Isolation of endophytic fungi from *Ocimum sanctum* was carried out by using the protocol described by strobel *et*

al., (2003) ^[8] with slight modification. The plant was washed under running tap water for 10 minutes to remove dust and debris. Highly sterile conditions were maintained for the isolation of endophytes. Before surface sterilization, the leaves, branches and roots were cut into small pieces of about 1cm long and sterilized in series with 70% ethanol for 1min, 1% mercuric chloride for 1 min and further cleaned by passing through two sets of sterile distilled water. All of work needs to be performed in the laminar air flow using sterile glassware and mechanical instruments. The sterile samples were placed on plate containing potato dextrose agar (PDA) media with 200mg/L concentration of streptomycin to suppress the bacterial contamination. The parafilm wrapped petridishes were incubated at 25-27°C till the mycelia start growing from the samples. The endophytic fungi was transferred into new agar slant and stored at 4°C for further studies.

For Aloe vera:-

The plant was washed under running tap water for 10 minutes to remove dust and debris. Before surface sterilization plant material was cut into small pieces of about 1cm long and sterilized in a series of 70% ethanol for 1-3 minutes, 4% mercuric chloride for 3-5 minutes, and rinse with 70% ethanol for 2-10 seconds further cleansed by passing through two sets of sterile distilled water. All of work needs to be performed in the laminar air flow using sterile glassware and mechanical instruments. The sterile samples were placed on plate containing potato dextrose agar (PDA) with 200mg/L of streptomycin to suppress the bacterial contamination. The parafilm wrapped petridishes were incubated at 25-27°C till the mycelia start growing from the samples. The endophytic fungi was transferred into new agar slant and stored at 4°C for further studies.

Media preparation:-

Potato Dextrose Agar (PDA) was used for isolation and purification of endophytic fungi. Antibiotic, streptomycin (200mg/L) was added to suppress bacterial growth. The media and antibiotics were purchased from Himedia, India.

Endophyte Fungal identification:-

The identification procedure of endophytic fungi was based on morphology. The five isolated species were described according to their macroscopic features (i.e. the color, shape and growth of cultured colonies) as well as microscopic characteristics (i.e. the structure of hyphae, conidia and conidiophores). The morphology of fungal culture colony or hyphae and the characteristics of the spore were identified by temporary mounts using lacto phenol cotton blue (LPCB) and viewed under the microscope at 40X. Obtained data were then compared with the descriptions of endophytic fungi species from standard identification manuals and matches were recorded. Analysis of the antibacterial activity was carried out on all species identified.

Preliminary antibacterial assay:-

Antibacterial activity of isolated endophytic fungi was tested based on the protocol of Zhang *et al.*, (2009) ^[10] with slight modification. The Petridish containing respective media for growth of bacteria were prepared and the test organism was spreaded on the surface of agar with the help of sterile cotton swab. Nine millimeter diameter of actively growing fungal culture disc from PDA plates were cut using sterile cork borer and placed on the surface of respective agar media seeded with the test bacteria. The plates were sealed with parafilm and kept in refrigerator at 4°C for 6 hours for the complete diffusion of the antimicrobial compounds if any, they were incubated at room temperature for next 12 hours for all bacterial cultures. After incubation the diameter of zone of inhibition was measured in millimeter by using scale.

Secondary metabolite extraction:-

Secondary metabolite extraction was carried out by using the protocol Radji *et al.*, (2011) ^[11]. Positive endophytic fungal isolate was inoculated into 1000 ml conical flask containing 500 ml potato dextrose broth and incubated at room temperature for 21 days under stationary condition with intermittent shaking. The broth culture was filtered to separate the mycelia and filtrate. To the filtrate equal volume of ethyl acetate was added, mixed well for 10 min, keep for 5min, till the two clear immiscible layers are formed. The upper layer of ethyl acetate containing extracted compound was separated using separating funnel. The mycelium was grinded properly in pestle and mortar using ethyl acetate as solvent and then it was filtered using cheese cloth. Both mycelia and culture filtrate were pooled together and evaporated to dryness using water bath. The extracted residue was dissolved in dimethyl sulfoxide (DMSO) and stored at 4°C to be used as stock solution for antibacterial assay.

Preparation of test organism:-

Endophytic fungal extracts isolated from Tulsi and Aloe vera were evaluated for their antibacterial potential against both Gram-positive and Gram-negative bacteria. *Staphylococcus aureus*, *Escherichia coli*, *Streptococcus pyogenes*, *Pseudomonas aeruginosa* were isolated from the clinical samples (like wound, blood and urine) obtained from patients attending KIIMS Hospital, K.R market, Bangalore, India and M. S. Ramaiah Memorial Hospital, New Bell road, Bangalore, India. These bacteria were identified on the basis of biochemical tests, by observation of the morphological structure microscopically and colony characters visually. The isolated organisms were stained with Gram's stain and viewed under microscope at 100X. The organisms were isolated in nutrient agar medium and selectively cultured at 37°C for 24 hrs. These bacterial cultures were maintained in nutrient agar slants at 37°C and were stored at 4°C for further study. Each of the bacteria was reactivated prior to susceptibility testing by transferring them into a separate test tube containing nutrient broth and incubated overnight at 37°C.

Secondary metabolite antibacterial assay:-

Antibacterial activities of secondary metabolites were tested by agar diffusion method. The bacteria used for antimicrobial assay are the pathogenic *Pseudomonas aeruginosa*, *E.coli*, *Staphylococcus aureus*, *Streptococcus pyogenes* which were isolated from urine, blood and wound samples. Muller Hinton agar was used for the antimicrobial assay. The sterile growth media plate specific for test organism were prepared and inoculated with the test organism. Twenty microliters of crude extract was added on to sterile 6mm paper disc using a micropipette and allowed to dry. Disc containing compounds were placed on the surface of the medium. In another plate 6mm diameter wells were made using a sterile cork borer and 20 microlitres of sample was added to each well. The plates were incubated at 24-48 hours at 37°C. The diameter for zone of inhibition was measured.

Determination of Minimum inhibitory concentration [MIC]:-

Minimum inhibitory concentration was carried out using broth dilution as previously reported by Lennette *et al.*, (1974)^[12]. Dilutions of different concentrations of extract and fractions that exhibited sensitivity against the test organisms were prepared using test tubes containing 9 ml of double strength broth. The test tubes were inoculated with (0.2 ml) suspension of the standardized inocula and incubated at 37°C for 24h. MIC's were recorded as the lowest concentration of extract showing no visible growth of the broth.

TLC Screening:-

About 1 mg of each extract was used for chromatography. A solvent system of dichloromethane: ethyl acetate 7:3 was prepared and placed in a tank and the lid was replaced. The fungal extracts were spotted, each one on separate origin on the plate. Chromatographic separations were carried out using silica gel (E. Merck, type 60), pre-coated silica gel GF254. The plates were placed carefully into the tank and covered with the lid. After development, the plate was removed and the solvent front was marked with pencil and allowed to dry. TLC plates were viewed under UV light at 254 nm for fluorescence spots and at 366nm for fluorescent spots. The metabolites from all the bands obtained were extracted by centrifugation of silica containing band in ethyl acetate at 10,000 rpm for 10 minutes. The supernatant was used for the antibacterial activity against all the clinical pathogens.

Results:-

Medicinal plants have been the main source for drugs over many centuries in many countries, in both developed and developing countries. Traditional medicine products are not officially recognized in many countries, and the European Union presently developing regulatory laws for quality traditional medicines. It is estimated that at least 25% of all modern medicines are derived either directly or indirectly from medicinal plants. Traditional medicines play important role in world health treating millions of people. The medicinal property of herbs is due to the presence of different complex chemical substances as secondary metabolites, which are exclusively accumulated in different parts of the plant. The need for new antimicrobial agent, in general, comes from increasing rate of resistance to existing antibiotics. This problem extends beyond the clinical application of antimicrobial drugs, such as agricultural microorganisms are also known to have acquired resistance to commonly used antimicrobial chemicals. In the present study, a total of 5 endophytic fungi (Table 1) were isolated from two medicinal plants (tulsi, aloe vera). All the isolated endophytic fungi showed preliminary antibacterial activity against the clinical pathogen. BCEF-01* showed maximum activity against *E.coli* and *Streptococcus pyogenes* while BCEF-04* and BCEF-05* showed maximum activity against *Streptococcus pyogenes* (Fig.1, Table 2). All the isolated endophytic fungi were cultivated in stationary condition (Fig.2) and the secondary metabolites produced by them are used for the antibacterial activity in different concentrations by agar as well as broth dilution methods. In agar dilution method, the results of antibacterial screening showed that BCEF-01*, BCEF-02*, BCEF-03*, BCEF-04* and

BCEF-05* inhibited all the test organisms. BCEF-01*, showed resistance at concentration of 2.1mg/ml against *P. aeruginosa* while other all test organisms are susceptible at various concentrations of fungal extract. *Staphylococcus aureus* shows maximum zone of inhibition of 15mm at extract concentration 2.6mg/ml (Table 3). For BCEF-02*, all the test organisms were found to be susceptible to the fungal extracts at various concentrations. The maximum zone of inhibition was found to be 17mm for *E. coli* at extract concentration of 2.0 mg/ml (Table 4). For BCEF-03*, *E. coli* was found to be resistant at concentration of 2.1 mg/ml while at rest of the concentrations all the test organisms are susceptible to the fungal extracts (Table 5). For BCEF-04*, *E. coli* is resistant at concentration 1.5mg/ml and 1.6mg/ml and *S. Pyogens* is resistant up to concentration of 1.8mg/ml while *S. aureus* and *P. aeruginosa* are resistant till the concentration of 2.0mg/ml. Rest of the test organisms are susceptible at various concentrations (Table 6). For BCEF-05*, *Pseudomonas aeruginosa* is resistant at concentration 1.5mg/ml while at other concentrations all test organisms are susceptible to the fungal extract (Table 7). The inhibitory effect was promising to all the test organisms showing sufficient zone of inhibition with respect to all the fungal extracts. The summary for which is given in the Table 8. The secondary metabolites of the endophytic fungi were further determined for Minimum Inhibitory concentration (MIC) against all the clinical pathogens by broth dilution method. The MIC of BCEF-02*, extract for all test organisms was found to be 1.42mg/ml (Fig.3).

For BCEF-05*, MIC was found to be 1.5mg/ml for *S. aureus* and *S. pyogens*, while for *E. coli* and *P. aeruginosa* the MIC were found to be 1.54mg/ml and 1.56mg/ml respectively (Fig.4). For BCEF-03*, MIC value obtained for *S. aureus*, *S. pyogens* and *P. aeruginosa* was found to be 2.02mg/ml while for that of *E. coli* MIC value obtained was 2.06mg/ml (Fig.5). For BCEF-01*, MIC value obtained for *E. coli* and *S. pyogens* was found to be 2.04mg/ml while for *S. aureus* and *P. aeruginosa* the MIC values obtained were 2.02mg/ml and 2.06mg/ml respectively (Fig.6). For BCEF-04*, MIC value for *S. aureus* and *S. pyogens* was found to be 2.02mg/ml and for that of *E.coli* and *P. aeruginosa* the MIC value obtained was 2.06mg/ml (Fig.7). Further the secondary metabolites produced by endophytic fungi were subjected to TLC and the bands obtained were used for antibacterial activity (Fig.8).



Fig.1:- Preliminary antimicrobial activity



Fig. 2:- Cultivation of endophytic fungi

Table 1:- List of endophytic fungi isolated from different parts of medicinal plants.

S. No.	Code	Plant	Plant part	Identification
1.	BCEF-01*	Tulsi	Root	<i>Aspergillus</i> sp.
2.	BCEF-02*	Tulsi	Leaf	<i>Penicillium</i> sp.
3.	BCEF-03*	Aloe vera	Root	<i>Cladosporium</i> sp.
4.	BCEF-04*	Aloe vera	Leaf	<i>Nigrospora</i> sp.
5.	BCEF-05*	Tulsi	Stem	<i>Gliocladium roseum</i>

Table 2:- Preliminary antibacterial activity of endophytic fungi.

Name of endophytic fungi	Zone of inhibition (mm)			
	<i>E.coli</i>	<i>Streptococcus pyogens</i>	<i>Staphylococcus aureus</i>	<i>Pseudomonas aeruginosa</i>
<i>Penicillium</i> sp.	10	11	10	12
<i>Gliocladium roseum</i>	15	25	12	15
<i>Cladosporium</i> sp.	10	20	10	10
<i>Aspergillus</i> sp.	25	25	15	10
<i>Nigrospora</i> sp.	10	26	11	15

Table 3:- Zone of inhibition shown by *Aspergillus* extract (BCEF-01*).

Name of endophytic fungal extract.	Concentration of extract (mg/ml)	Zone of inhibition (mm)			
		<i>Staphylococcus aureus</i>	<i>E.coli</i>	<i>Streptococcus pyogens</i>	<i>Pseudomonas aeruginosa</i>
<i>Aspergillus</i> sp.	2.1	10	9	9	-
	2.2	11	10	10	9
	2.3	12	10.1	10.1	9.5
	2.4	13	10.2	10.2	10
	2.5	14	10.3	10.3	10.1
	2.6	15	10.4	10.4	10.2

Table 4:- Zone of inhibition by *Penicillium* extract (BCEF-02*).

Name of endophytic fungal extract.	Concentration of extract (mg/ml)	Zone of inhibition (mm)			
		<i>Staphylococcus aureus</i>	<i>E.coli</i>	<i>Streptococcus pyogens</i>	<i>Pseudomonas aeruginosa</i>
<i>Penicillium</i> sp.	1.5	13	13	12	8
	1.6	13.5	15	13	8
	1.7	14	15.5	13.5	8.5
	1.8	14.5	16	14	9
	1.9	15	16.5	14.5	10
	2.0	16	17	15	11

Table 5:- Zone of inhibition by *Cladosporium* extract (BCEF-03*).

Name of endophytic fungal extract.	Concentration of extract (mg/ml)	Zone of inhibition (mm)			
		<i>Staphylococcus aureus</i>	<i>E.coli</i>	<i>Streptococcus pyogens</i>	<i>Pseudomonas aeruginosa</i>
<i>Cladosporium</i> sp.	2.1	8	-	8	8
	2.2	9	9	8.5	8.5
	2.3	9.5	9.5	9	9
	2.4	9.5	10	9.5	9.5
	2.5	10	10.5	10	10
	2.6	10.5	11	10.5	10.5

Table 6:- Zone of inhibition shown by *Nigrospora* extract (BCEF-04*).

Name of endophytic fungal extract	Concentration of extract (mg/ml)	Zone of inhibition (mm)		Concentration of extract (mg/ml)	Zone of inhibition (mm)	
		<i>E.coli</i>	<i>Streptococcus pyogens</i>		<i>S. aureus</i>	<i>P.aeruginosa</i>
<i>Nigrospora</i> sp.	1.5	-	-	2.1	9	9
	1.6	-	-	2.2	9.5	9.5
	1.7	10	-	2.3	9.6	9.6
	1.8	10.5	-	2.4	9.7	9.7
	1.9	11	9	2.5	9.8	9.8
	2.0	11.5	10	2.6	10	10

Table 7:- Zone of inhibition by *Gliocladium* extract (BCEF-05*).

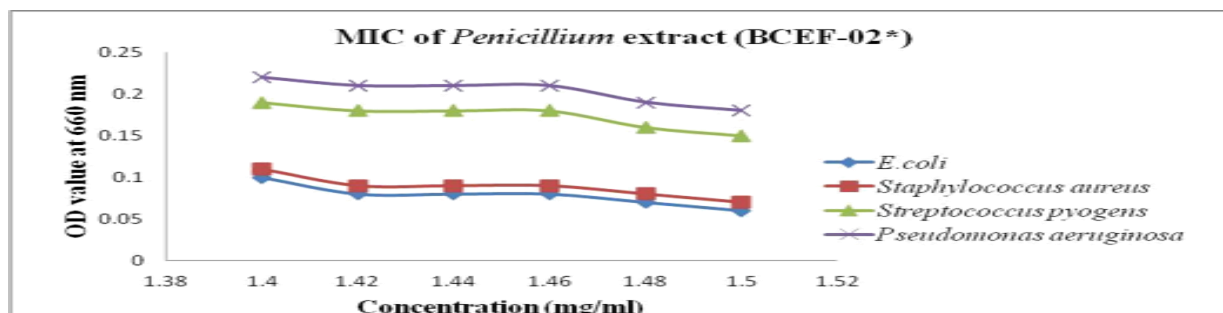
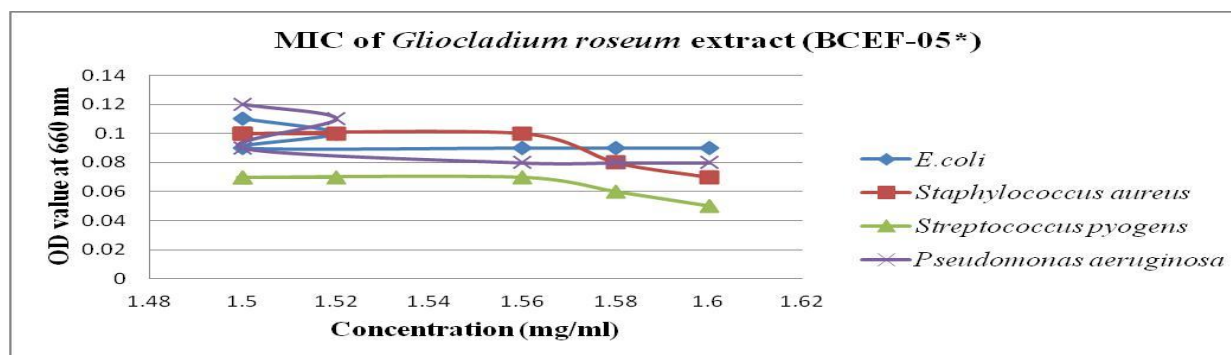
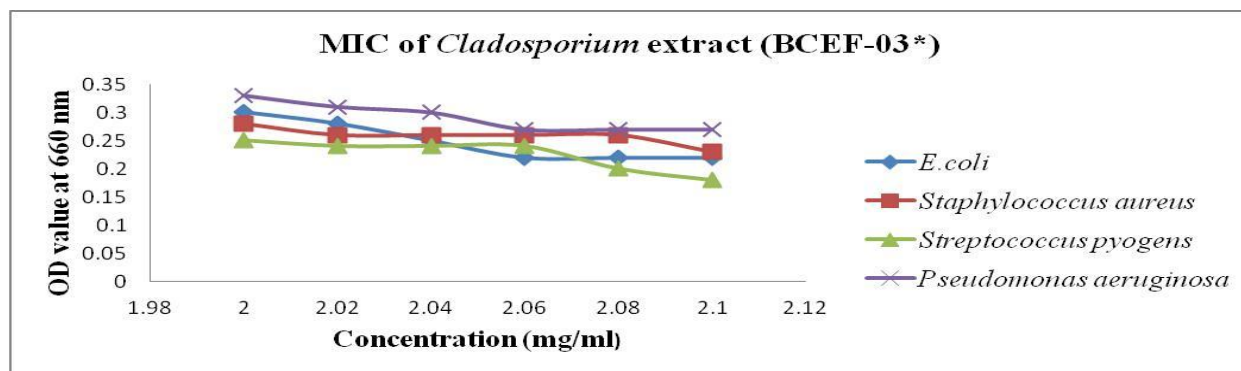
Name of endophytic fungal extract.	Concentration of extract (mg/ml)	Zone of inhibition (mm)			
		<i>Staphylococcus aureus</i>	<i>E.coli</i>	<i>Streptococcus pyogens</i>	<i>Pseudomonas aeruginosa</i>
<i>Gliocladium</i> sp.	1.5	10	9	9	-
	1.6	10.05	10	10	10
	1.7	11	10.5	10.5	10.5
	1.8	11.15	11	11	11
	1.9	12	12	11.5	11.5
	2.0	12.5	12.5	12	14

Table 8:- Summary of antibacterial activity of the fungal extracts.

Endophytic fungi/ code	<i>S.pyogens</i>	<i>S.aureus</i>	<i>E.coli</i>	<i>P.aeruginosa</i>
BCEF-01*	++	+++	++	++
BCEF-02*	+++	+++	+++	++
BCEF-03*	++	++	++	++
BCEF-04*	+	+	++	+
BCEF-05*	++	++	++	++

+: the inhibition zone is up to 10mm ++: the inhibition zone is from 11mm to 14mm

+++: the inhibition zone is 15mm and above

**Fig 3:-** MIC of *Penicillium* extract against test organisms.**Fig. 4:-** MIC of *Gliocladium roseum* extract against test organisms**Fig 5:-** MIC of *Cladosporium* extract against test organisms.

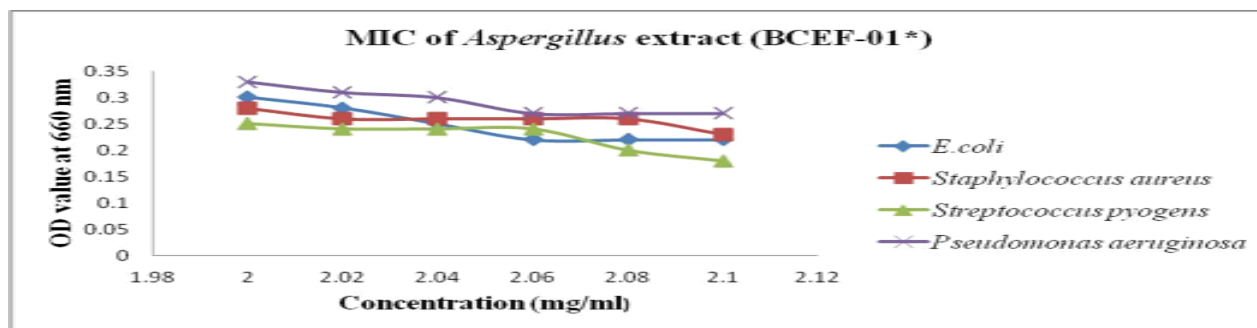


Fig 6:- MIC of *Aspergillus* extract against the test organisms.

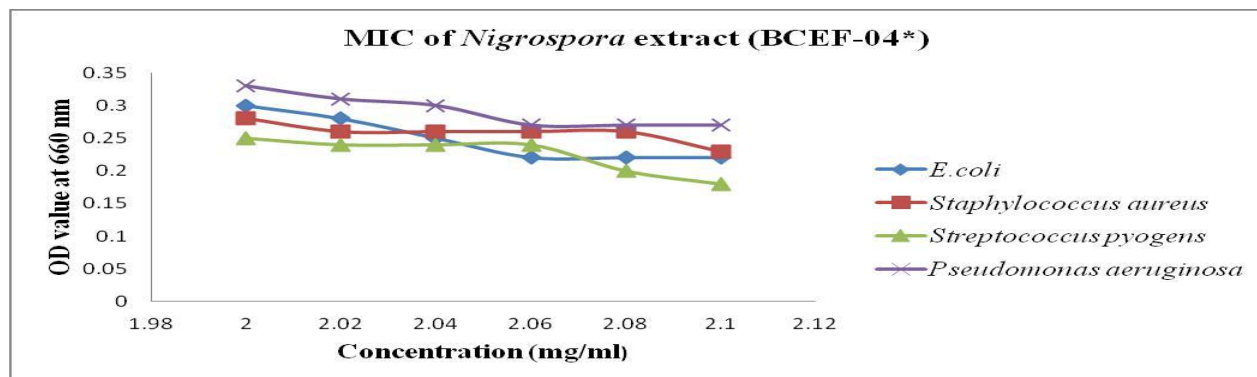


Fig 7:- MIC of *Nigrospora* extract against test organism.

Separation of the metabolites by preparative TLC:-

The fungal extracts were subjected to preparative Thin Layer Chromatography. The spots on the plates were developed by mobile phase (dichloromethane: ethylacetate) in the ratio 7:3 and observed under UV illuminator. Bands obtained were used to check antibacterial activity. And all the fungal extract shows promising antibacterial activity.

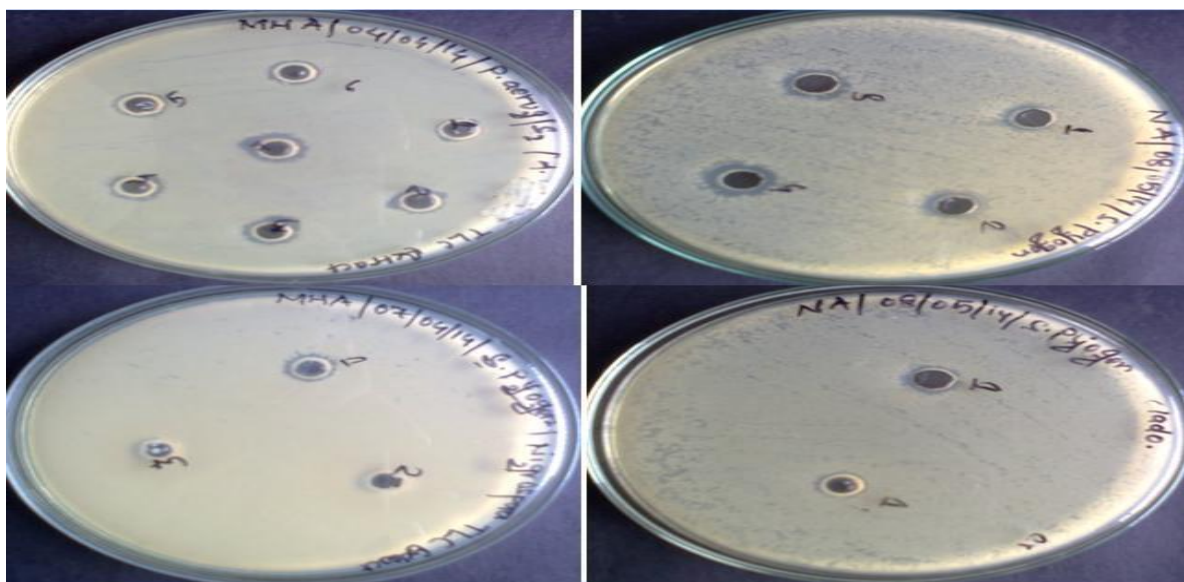


Fig 8:- Antibacterial activity of TLC extracts of different fungal extracts.

Hence the bioactive compounds need to be identified in order to find out which functional group is active against the given pathogens. As for *S. aureus* and *S. pyogenes* the novel compound may be used as a potent bioactive chemical

for topical applications against skin diseases and wounds. For identification of the functional group the study recommends the use of HPLC and GCMS. Still the potential for the same promising novel compound needs to be evaluated before it becomes a potential candidate for treatment. Hence, it may be concluded that these endophytic extracts could be used in the treatment of human diseases which have demonstrated potential values.

Discussion:-

Endophytes colonize inside healthy plant tissues to get nutrition and shelter from the host, and in response produce many functional metabolites, which may enhance the host fitness, and have anti-feedant activity and provide resistance against various biotic and abiotic stresses as well. Endophytic fungi have been recognized as a repository of novel secondary metabolites, some of which have beneficial biological activities (Bills and Polishook, 1991^[13], Strobel and Daisy, 2003^[14]). The results of the present study agree with these earlier findings. The result shows that the fermentation broth of endophytic fungi of *Aspergillus* sp. (BCEF-01*), *Penicillium* sp. (BCEF-02*), *Cladosporium* sp. (BCEF-03*), *Nigrospora* sp. (BCEF-04*) and *Gliocladium roseum* (BCEF-05*) has broad-spectrum of antimicrobial activity against both Gram-positive and Gram-negative pathogenic bacteria like *Staphylococcus aureus*, *Streptococcus pyogenes*, *Pseudomonas aeruginosa*, and *E.coli*.

Siqueira *et al.*, (2011)^[15] reported that 16 out of 203 endophytic isolates showed antimicrobial activity with a wider action spectrum inhibiting Gram-positive and Gram-negative bacteria and fungi. Most of the bioactive metabolite compounds from endophytic fungi reported were more effective against Gram-positive bacteria than Gram-negative bacteria and pathogenic fungi (Chareprasert *et al.*, 2006^[16]). These findings were in agreement with observations by Cos *et al.*, (2006)^[17] that the Gram-negative bacteria are much more resistant than Gram-positive bacteria. It has been stated that the possible mechanism behind the sensitivity of Gram-positive bacteria may lie in the significant differences in the outer layers of Gram-negative and Gram-positive bacteria. Gram-negative bacteria possess an outer membrane and a unique periplasmic space (Duffy and Power, 2001^[18]) whereas the Gram-positive bacteria found to be much more susceptible since they have only an outer peptidoglycan layer which is not an effective permeability barrier (Scherrer and Gerhardt, 1971^[19]).

Since in microbial-plant relationship endophytes contribute with substances that possess various types of bioactivity (Radu and Kqueen 2000^[20]), researchers had been conducting screening for bioactive metabolites and endophytic fungus antimicrobial activity, as was previously revealed (Pela'ez *et al.*, 2000^[21], Radu and Kqueen 2000^[20], Ezra *et al.*, 2004^[22]). Five different endophytic fungi were isolated from the medicinal plant tulsi and aloe vera and all belong to phylum Ascomycota. These results were correlated with the previous findings of Pavithra *et al.*, (2012)^[23]. In the preliminary antibacterial assay the endophytic fungi *Gliocladium roseum* and *Nigrospora* sp. showed the maximum zone of inhibition of 25mm and 26mm against *Streptococcus pyogenes* while *Aspergillus* sp. showed the maximum zone of inhibition of 25mm against *E.coli* and *Streptococcus pyogenes*.

The antibacterial activity of 5 extracts of liquid culture of endophytic fungi were evaluated by diffusion agar assay against Gram-positive and Gram-negative bacteria. The activity of the extracts was estimated from growth inhibition (in mm) as follows: <8mm: Inactive; 8–12 mm: weak activity; 13–15 mm: moderate activity; >15 mm: strong activity (Becerra *et al.* 2002^[24]). Almost all the endophytic fungi showed the antimicrobial activity against the test bacteria but the endophytic fungi *Penicillium* sp. showed the zone of inhibition of 15mm, 16mm, 17mm and 11mm at the concentration of 2mg/ml against *S.aureus*, *E.coli*, *S.pyogens*, and *P.aeruginosa* respectively. Hence the bioactive compound produced by these fungi can be used for the treatment of disease caused by such organism. Although *Cladosporium* sp. showed less antibacterial activity in this experiment, while in a recent study, Zhang *et al.*, (2011)^[25] reported the production of an alkaloid, huperzine A, used in treating the Alzheimer's disease.

The minimal inhibitory concentration (MIC) was defined as the lowest extract concentration resulting in no visible growth after the incubation time. Aligiannis *et al.*, (2001)^[26] proposed a classification for plant materials, based at MIC results as: strong inhibitors: MIC up to 0.5 mg/ml; moderate inhibitors: MIC between 0.6 and 1.5 mg/ml and weak inhibitors: MIC above 1.6 mg/ml. In the present study, we have established MIC from 1.42mg/ml to 2.06mg/ml for all the 5 endophytic fungal extracts. *Penicillium* sp. and *Gliocladium roseum* extracts possess moderate MIC.

Besides the antimicrobial and other bioactivities, endophytic fungi *Gliocladium roseum* reported to produce biodiesel (Strobel *et al.*, 2008^[27]), which may be the future hot cake for endophytic study. Interestingly, the findings of this study provide a strong platform for the isolation and purification of novel natural antimicrobial agents from

endophytic fungi of Tulsi and Aloe vera.

Conclusion and Future Targets:-

Since Stierle *et al.* (1993)^[28] discovered a taxol producing endophyte: *Pestalotiopsis microspore*, there have been an increasing interest in bioactive metabolites isolated from endophytes. Many studies proved that endophytes produce novel secondary metabolites as a resistance mechanism to overcome pathogenic invasion (Tan and Zou, 2001^[29]). Research on endophytes has been carried out and a great amount of antimicrobial natural products were found. So far, a great number of antimicrobial compounds have been found in a handful of the one million different endophyte species (Petrini, 1991^[1]); It is believed that searching for natural products synthesized by endophytes could be a promising way to solve the problem that bacteria are becoming resistant to some commonly used drugs according to WHO and meet the emergency demand of discovering highly effective, low toxicity, and no environmental impacted antibiotics to fight against resistant bacteria species.

The ultimate purpose of endophytes research is to find new antibiotics or pesticides, thus the following aspects should be intensively studied:

1. To find better bioactive antimicrobial substances without any side effect to human, plant and environment.
2. To enhance the antimicrobial activity or decrease the side effects of known metabolites from endophytes by modifying their structures in order to improve the efficacy and specificity to microbes.
3. To optimize conditions of endophyte fermentation that has been found to show bioactivity in order to enhance the yield of active substances synthesized by endophyte.
4. To search for the regulatory gene in synthesis path of antimicrobial compound, and use genetic engineering technology to increase the production of antibacterial substances.

As so many antimicrobial compounds were isolated from endophytes which only occupied a small portion of total endophyte species, it is obviously that there is a great opportunity to find reliable and novel antimicrobial natural products in endophytes, which may be used as clinically effective antibiotics in future.

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RESEARCH ARTICLE

INTRODUCING LITERARY WRITINGS TO DEVELOP COGNIZANCE IN PERSONNEL

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Abstract

Management of organization is the management of its staff in real terms because the work is executed by them and it ultimately runs organization. Every employee has its own customs to solve the problems of life. Literature studies not only enhance the knowledge, but also provide solutions to the life issues. Reading motivational and inspirational stories fills us with a new insight. For this study, employees have been asked that an interesting proverb or saying written on notices can ever encourage them. They admit that fictional writings can be used a new technique to get clarifications of their glitches. The aim of this study is to develop new criteria of literature and management, so that we get to know our culture, traditions and history. The modern management studies can be enriched with literature studies and organization can have better people with more alertness and improved work efficiency.

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Introduction:-

Literature is the base of any culture. One can easily identify the richness of culture by studying it. It demonstrates the traditions, rituals, styles, habits and beliefs of the various present and earliest civilizations. These practices are available in the form of stories, sayings, teachings, poems and proverbs. It constitutes a folk wisdom through creative literature. Some sayings are most popular and become a part of everyday life. 'Folk psychology is a unique kind of narrative practice and that viewing it as such is the best way to account for its ultimate origins and everyday applications. Many of our routine encounters with others take place in situations in which the social roles and rules are well established, so much so that unless we behave in deviant manner we typically have no need to understand one another by means of the belief or desire schema'. 'Folk psychological narratives work to regulate our actions, as such they are instruments of culture, and they summarize not simply how things are but (often implicitly) how they should be (Hutto 2012)'.

Insight Through Narratives:-

"Each narrative text is a singular unit and needs to be seen as such. The characters in the stories are identified by their caste, and when the stories are interpreted collectively, the social fabric of attitudes towards each caste emerges. This adds variety to the folktales. The narration of the oral tales undergoes constant innovation and renewal. For this reason, the narratives of late nineteenth century contain many direct references to contemporary social reality (Naithani 2006)". 'Stories are a way of understanding the underlying operating principles behind organizational communication and learning. Organizations need to be their own unique, customized processes and tools to implement the push - to - pull -to - push strategy. Stories are giving employees a greater sense of alignment between their job responsibilities, the business objectives they need to execute, and the organization's mission'. The majority of management literature on stories places emphasis on characterizing stories as vehicle of communication.

'The communicating power of stories lies in their role as a tool for listening and reflecting. Stories also demystify the way in which our mind synthesizes information in order to produce new insights and learning (Gargiulo 2014)'. The wisdom loving person is the one who searches for the timeless and unchanging truths, never contend with the shifting phenomenon of the material world (Sternberg 1990). "Folk wisdom has been to explore sources of wisdom that have stood the test of time and culture and that might serve to help individuals cope with challenges of life (Lopez 2011)".

Importance Of Vasudhaive Kutumbakam To Entrepreneurs:-

'The most important message that one can extract from the study of Indian literature is *Vasudhaive Kutumbakam*. This word has its own significance and it gives the message of unity being a family. This word has been used by many scholars in their speeches and writings. We are trying to relate this with the practices of management studies in the organization, because the organization is not a single entity. It's a group of people in which people work for their mutual benefits and for the organization itself. So the theory of *Vasudhaive Kutumbakam* can be best applied in the organization which can help the organization to bring the change in the performance of employees by having a feeling of unity among them and can work for each other's benefit rather than having individualistic approach. Everyone has their own worldview which is quite often different from person to person. *Vasudhaive Kutumbakam* means respecting this difference. Contrary to single worldview, *Vasudhaive Kutumbakam* says animals, birds, plants, trees and other organisms in the ecosystem have Atma they are part of our family (Nanuk 2012)'. "*Vasudhaive Kutumbakam* occurs in the *Hitopadesha* or 'Beneficial Instruction' a collection of fables that was intended to be a manual for teaching young princes how to get ahead in the world. One of the fables concern the machinations of hungry jackal. As the larger narrative frames provide the best context for interpreting the verse in question, I shall paraphrase loosely from the *hitopadesha* and find in each case the end in mind is a dastardly one. 'The cat wants to devour the birds, and the jackal has his eyes on the deer. Both make hypocritical appeals to what sounds like accepted moral rules in order to attain their ends. Thus, both play upon the conscience of their prey, turning it to their own evil purpose. The one difference between the verses cited by these two scoundrels is that while the cat's epigram suggests the possible fate that might befall a consistently dutiful host, the jackal cites a verse that has no shadow side'. It seems merely to exhort one to a higher level of humanity (overlooking the character's animal personas) (Hatcher 1994)".

"Far away in Behar there is a forest called Champak-Grove, and in it had long lived in much affection a Deer and a Crow. The Deer, roaming unrestrained, happy and fat of the carcase, was one day described by a Jackal. 'Ho! Ho!' thought the Jackal on observing him, 'if I could but get this soft meat for a meal! It might be—if I can only win his confidence.' Thus reflecting he approached, and saluted him. 'Health be to thee, friend Deer!' 'Who art thou?' said the Deer. 'I'm Small-wit, the Jackal,' replied the other. 'I live in the wood here, as the dead do, without a friend; but now that I have met with such a friend as thou, I feel as if I were beginning life again with plenty of relations. Consider me your faithful servant.' 'Very well,' said the Deer; and then, as the glorious King of Day, whose diadem is the light, had withdrawn himself, the two went together to the residence of the Deer. In that same spot, on a branch of Champak, dwelt the Crow Sharp-sense, an old friend of the Deer. Seeing them approach together, the Crow said, 'Who is this number two, friend Deer?' 'It is a Jackal,' answered the Deer, 'that desires our acquaintance.' 'You should not become friendly to a stranger without reason,' Said Sharp-sense. 'Don't you know?' 'To folks by no one known house-room deny:-

The Vulture housed the Cat, and thence did die." 'No! How was that?' said both.

'In this wise,' answered the Crow. "On the banks of the Ganges there is a cliff called Vulture-Crag, and thereupon grew a great fig-tree. It was hollow, and within its shelter lived an old Vulture, named Grey-pate, whose hard fortune it was to have lost both eyes and talons. The birds that roosted in the tree made subscriptions from their own store, out of sheer pity for the poor fellow, and by that means he managed to live. One day, when the old birds were gone, Longear, the Cat, came there to get a meal of the nestlings; and they, alarmed at perceiving him, set up a chirruping that roused Greypate. 'Who comes there?' croaked Grey-pate.

"Now Long-ear, on spying the Vulture, thought himself undone; but as flight was impossible, he resolved to trust his destiny and approach. 'My lord,' said he, 'I have the honor to salute them.' 'Who is it?' said the Vulture. 'I am a Cat.' 'Be off, Cat, or I shall slay thee,' said the Vulture. 'I am ready to die if I deserve death,' answered the Cat; 'but let what I have to say be heard.' 'Wherefore, then, comest thou?' said the Vulture. 'I live,' began Long-ear, 'on the Ganges, bathing, and eating no flesh, practicing the moon-penance, like a Bramacharya. The birds that resort thither constantly praise your worship to me as one wholly given to the study of morality, and worthy of all trust; and so I

came here to learn law from the, Sir, who art so deep gone into learning and in years. Dost thou, then, so read the law of strangers as to be ready to slay a guest? What say the books about the householder?-

*'Bar thy door not to the stranger, be he friend or be he foe,
For the tree will shade the Woodman while his axe doth lay it low.'*

And if means fail, what there is should be given with kind words, as-'Greeting fair and room to rest in; fire, and water from the well-Simple gifts-are given freely in the house were good. Men dwell,- and without respect of person-'Young, or bent with many winters; rich, or poor, whate'er thy guest, Honor him for thine own honor-better is he than the best,'

Else comes the rebuke-'Pity them that ask thy pity: who art thou to stint thy hoard, When the holy moon shines equal on the leper and the lord!' And that other, too, 'When thy gate is roughly fastened, and the asker turns away, Thence he bears thy good deeds with him, and his sins on thee doth lay.' For verily, 'In the house the husband ruleth, men the Brahmins "master" call; Agni is the Twice-born Master-but the guest is lord of all.' "To these weighty words Grey-pate answered, 'Yes! But cats like meat, and there are young birds here, and therefore I said, go.' 'Sir,' said the Cat (and as he spoke, he touched the ground, and then his two ears, and called on Krishna to witness to his words), 'I that have overcome passion, and practiced the moonpenance, know the Scriptures; and howsoever they contend, in this primal duty of abstaining from injury they are unanimous. Which of them sayeth not-'He who does and thinks no wrong-He who suffers, being strong-He whose harmlessness men know-Unto Swerga such doth go.' "And so, winning the old Vulture's confidence, Long-ear, the Cat, entered the hollow tree and lived there. And day after day he stole away some of the nestlings, and brought them down to the hollow to devour. Meantime the parent birds, whose little ones were being eaten, made an inquiry after them in all quarters; and the Cat, discovering this fact, slipped out from the hollow, and made his escape. Afterwards, when the birds came to look closely, they found the bones of their young ones in the hollow of the tree where Grey-pate lived; and the birds at once concluded that their nestlings had been killed and eaten by the old Vulture, whom they accordingly executed. That is my story, and why I warned you against unknown acquaintances." (Edwin 1861)

Literature Review:-

"Wisdom consists of proverbial sentence or instruction, debate, intellectual reflection; thematically wisdom comprises self-evident intuitions about mastering life for human betterment, groping after life secrets with regard to innocent suffering, grappling with finitude, and quest for truth concealed in the created order and manifested in Dame Wisdom. When a marriage between form and content exists, there is wisdom literature". 'Wisdom literature is not dry-as-dust academic work, but strongly poetic in character (Weeks 2010)'. 'Wisdom is a comprehensive grasp of knowledge that is characterized by both breadth and depth of understanding. It is one of the cardinal virtues and thus transcends the realm of the merely cognitive. The great philosophers of the wisdom are Socrates (469- 399 B.C.), Plato, Aristotle (384-322 B.C.), Lucretius (99-55B.C.), and Zeno of Citium (336- 265 B.C.) (Sternberg 1990)'. "Panchatantra have become part of common Indian folklore transmitted orally from parents to children down the centuries. The literary sources of the Panchatantra therefore, are twofold: the expert tradition of political science and the folk and literary traditions of storytelling (Olivelle 2009)". "Proverbs are the excellent source of wisdom". 'Proverbs have been classified in to three types: synonymous, antithetic, and synthetic. Another classification includes Aussagewort (Statement) and the Mahnwort (warning or admonition) (Whybray 1995)'. 'Ancient authors sought to capture wisdom in collection of proverbs and fables. Language and Literacy are generally regarded as two of the foundation stones of human civilization (Curnow 2015)'. "In Vasudhaive Kutumbakam, the relationship between individuals, families, institutions, communities and nations and between men and nature should be cemented on the basis of equity, mutuality and respect" (Shah 2013). "Folk wisdom also contain a treasure itself of positive psychology- a psychology that was not just about suffering, trauma, depression ,victims, irrationality, madness and crime" (Lopez 2011).

Research Methodology:-

Hypothesis- Proverbs and Literary writings has its significant place in the modern world and ethical and working knowledge can be imparted to employees through these works.

Data analysis:-

We have done survey and interviewed about 200 people professions. Questions were divided on different issues related to our problem. The sample was taken randomly from the Institute, itself. The respondents are divided in to

the different categories. Almost 200 respondents filled the questionnaire very well. Survey was done in very short duration of time of one week due to busy schedule of the respondents. We have asked the respondents about their interest in literature, the concept of Vasudhaiva Kutumbakam and the moral lessons in the proverbs. Two tables are drawn of the response; one table has answers of the questions. And other table is drawn on the basis of the grouping of the respondents according to their profession. The response that we get is described here in the form of charts and tables.

Table 1:- Response From The Employees.

Issue	Reply –Yes	Reply –No
Issue 1	185	15
Issue 2	190	10
Issue 3	160	40
Issue 4	180	20
Issue 5	195	5
Issue 6	170	30
Issue 7	175	25

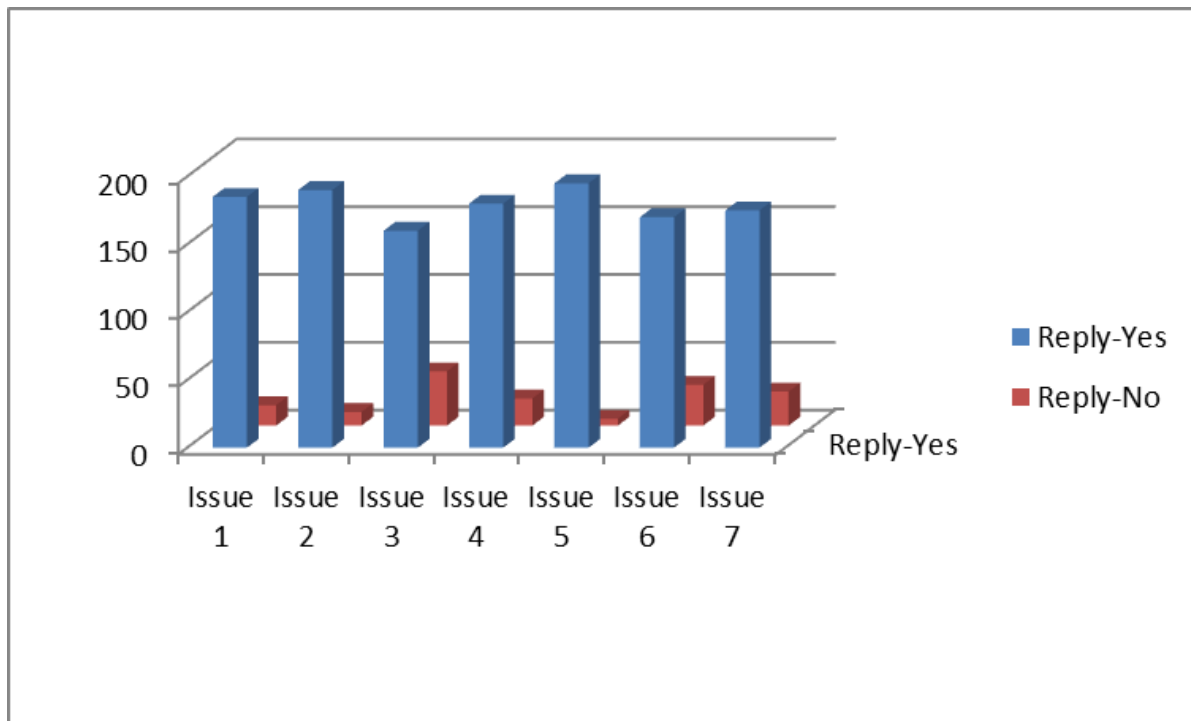


Figure 1:-

Findings:-

After interviewing them, we have found that all the respondents like reading literature. One of the important achievement of the survey is that all the respondents believe that that literature reading definitely helpful to come out of the problematic situations. It will energizes them and helpful in character building. All the respondents were educated persons and they also suggest their colleagues to read literature because it just not entertaining them, but also helps them in building Good moral character. Most of them read literary writings for entertainment but after this survey they read finest literary writing to improve the moral of their employees and set up a good mechanism of management based on literature. Reading literature is not a leisure activity, but it guides and shows the right path. Some entrepreneurs admitted that the business should be run on the basis of the Vasudhaiva kutumbakam. All the respondents gave their wonderful feedback. They agreed that by introducing literature in management studies because one can learn the lessons of good management through their own culture and literary writings. They are very good response about the proverbs because proverbs can be a source of quick learning.

Lessons From The Proverbs:-

'Proverbs expresses what a fundamental truth is apparently; - that is a truism in homely language often adorned, however, with alliteration and rhyme. It is usually short, but need not be; it is usually true, but need not be'. "A proverb must be venerable; it must bear the sign of antiquity, and, since such signs may be counterfeited by a clever literary man, it should be attested in different places at different times". This last requirement we must often waive in dealing with very early literature, where the material at our disposal is incomplete". (Whiting 1932) For example "Do not work under an irrational boss. If it is fate accomplice, try to get rid of him soon" (Dadhich 2014). 'Some proverbs contain a truth of general application which holds good for all time and stands its ground in the phase of social change and political or economic revolution. Such proverbs are based on universal experience and embody the common sense of mankind. Some proverbs have a more limited range. They express a truth rooted in experience, but the experience is that of a particular people, or of a particular nation, and the saying in which it is summed up are colored by the spirit of the time when they were coined and of the nation which produced them. They hold well for their birth place, but not for the entire world' (Risley 1999).

"For example:-

1. A little wit will serve a fortunate man. It means that fortune is nourished by intelligence.
2. A man must plough with such oxen as he hath. It means one should not wait for great efforts and must start with the efforts which he has.
3. Both together do best of all. Appreciation of togetherness.
4. Do not put all your eggs in one basket. One should not risk everything he has in a single venture.
5. Every tub must stand on its own bottom. Hard work is a foundation of mend person.
6. First think and then speak. What will be the effects of speaking? Think it first. First think and then start the work.
7. Go not for every grief to the physician, for every quarrel to the lawyer, nor for every thirst to the pot. Bearness, balance of mind and watchful thirst are the best options of them respectively.
8. Health is wealth. It is true because health earns wealth.
9. Honesty keeps the crown of the cause away. Honesty never is questionable.
10. In to a shut mouth flies fly not. The person who is satisfied never be with ill work. Satisfaction keeps away all the evil things".
11. "Kindness like grain, increase by sowing. Grain increase by sowing so the kindness by doing kind things.
12. Opportunity hardly comes twice. It comes once in a while manner.
13. Punctuality is the soul of business. Most Important merit of business.
14. Religion is the rule of life. It is a chief source of life.
15. Second thoughts are best. Second thoughts are the wise and correct thinking.
16. The greatest wealth is contentment with a little. Gratification is also a great wealth.
17. The morning hour has gold in its mouth. Good hours for effective working as well as good for your health too.
18. There is a good time coming. One should hope for the best.
19. Think before doing. One should think before doing.
20. Truth fears no colours. Truth always is a ferules thing. Whatever the authority is.
21. Union is strength. Untidiness is a sign of power.
22. Virtue has all things in itself. Moral goodness is a perfect thing.
23. We much not lie down and cry, God help us. God helps those who help themselves so it is essential that we should help ourselves first.
24. You may know by a handful the whole sack. Little quantity is sufficient to know the whole sack or if you want to know a person completely then his few specialties are sufficient enough.
25. Zeal without knowledge is fire without light. Enthusiasm without cognition is like a body without soul" (Sharma 2015).

Famous proverbs in the world:-

1. 'By preserving the egg walk on legs. Patience enables one to scucced.to succeeds in any endeavour we have to be patient and preserve till our effort comes to fruition.
2. When the bag tears, the shoulders get a rest. When plans run in to problems, one does not have to become overly frustrated. Frustrating our plans may be god's gracious way of giving.

3. One who bathes willingly with cold water doesn't feel the cold. It expresses the positive benefit of choosing one's fate and accepting circumstances'.
4. 'If you refuse the elder's advice you will walk the whole day.
5. A person who is not disciplined cannot be cautioned
6. Say only a little but say it well.
7. A man a smiling face must not open shop.
8. Experience is a comb which nature gives us when we are bald' (Sawhney 2004).

Conclusion:-

Literature is the medium to learn and explore new things. Ancient literary writings and proverbs are full of wisdom and helpful in transforming personalities of people. People of the modern world always looking for an extra pathway to tackle with the problems of today's demanding life and solution should be quick and easy. In that case proverbs play a better role to show them a true direction and empower them with strength of positivity. We are concluding this paper by significantly remarking that narratives in literature are the best source of wisdom to an ordinary man, and to an entrepreneur.

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RESEARCH ARTICLE

EFFECT OF SYNTHESIS CONDITIONS OF CARBON XEROGELS ON THEIR SURFACE CHEMISTRY, PORE TEXTURE AND METHYLENE BLUE ADSORPTION CAPACITY.

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Pore texture, Methylene blue.

Abstract

Resorcinol-formaldehyde organic xerogels (OXs) were synthesized by the polycondensation of resorcinol (R) and formaldehyde (F) using two different techniques [microwave (MW) - induced synthesis and conventional (C) synthesis methods]. For the MW-induced synthesis of OXs, five samples variable in their R-F solutions pH values [in the range 3.0-7.2] were synthesized. These samples were pyrolyzed at 750 °C under N₂ flow to give five carbon xerogel (CX) samples. To study the effect of the carbonization temperature, the OX sample prepared at R-F solution pH value of 6.5 was selected and carbonized at different temperatures in the range 600 - 800°C, to produce four CX samples. To investigate the effect of the synthesis method on the CX characteristics, another OX sample was prepared by the conventional method at R-F solution pH value of 6.5 and was carbonized at 750°C under N₂ flow to give the last CX sample. The effect of the synthesis conditions on the physicochemical and surface chemistry characteristics of the products was performed using different techniques, such as elemental analysis, determination of the surface acidic and basic active sites by the Boehm method, identification of surface functional groups by FTIR, XRD analysis, SEM and TEM investigations. Pore texture characteristics were evaluated by analysis of the respective N₂/77K adsorption-desorption isotherms. The methylene blue (MB) number (X_m (mg/g)) and surface areas accessible to MB (S_{MB}) were calculated using the Langmuir and Freundlich adsorption isotherm equations. The Langmuir adsorption isotherm model gives better fit as compared to Freundlich model. Ratios of methylene blue surface area to nitrogen surface area (S_{MB} / S_{N₂}) were evaluated and discussed. Removal of MB from aqueous solution is governed by the chemical nature of the CX surface as well as the diffusion through the intricate internal porosity.

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Introduction:-

Carbon gels are a class of porous carbon materials that have received considerable attention due to their potential applications in fuel cells (Alegre et al., 2012), adsorption (Álvarez and Ribeiro, 2015), bio-sensing (Li et al., 2016), catalysis (Moreno-Castilla and Maldonado-Hódar, 2005), energy storage (Ordeñana-Martínez and Rincón, 2016), and supercapacitors (Lufrano et al., 2011; Zapata-Benabith et al., 2016). They can be obtained by sol-gel synthesis

via polymerization reaction between hydroxylated benzenes (such as resorcinol (R)) and formaldehyde (F) (Al-Muhtaseb and Ritter, 2003; Job et al., 2004). After sol-gel polymerization, the solvents used as the reaction media can be removed in three different drying methods; subcritical, supercritical or freezing drying, resulting in xerogels, aerogels and cryogels, respectively (Job et al., 2005). The cheapest and easiest drying method is subcritical drying, which is based on removing the solvent from the gel structure by drying conventionally (at ambient pressure and temperatures of around 100-150 °C) to obtain organic gels with high surface areas and pore volumes (Al-Muhtaseb and Ritter, 2003). Pyrolysis of an organic gel under inert atmosphere allows the removal of labile oxygen and hydrogen surface groups, resulting in thermally stable nano structured porous carbon gel. By varying the synthesis conditions, it is possible to obtain carbon gels with different properties (Zubizarreta et al., 2008). Among the parameters that have a great influence on the final gel properties is the R-F solutions pH values (Zanto et al., 2002a). During the pyrolysis process, the carbonization temperature (CT) is the most important parameter that has a significant influence on the pore structure of carbon gels (Al-Muhtaseb and Ritter, 2003).

Carbon xerogels (CXs) have very interesting features such as three-dimensional nanosized network (Kakunuri et al., 2015), high porosity and surface area, the possibility of designing their porous texture, outstanding electrical, thermal and mechanical properties and simple manufacturing on a laboratory scale (Calvo et al., 2011). However the main problem of CXs synthesis is the too long time in the conventional preparation method. During the last few years, microwave (MW) radiation has become an accepted heating source for chemical synthesis due to numerous advantages it offers. These include a reduction in synthesis time, an easy and safe method, direct heating of molecules and the avoidance of secondary reactions, giving rise to cleaner products with a higher yield (Gedye, 2002). Therefore, MW radiation has been used to produce carbon gels with a considerable saving of time and energy (Kang et al., 2008).

There are a quite large number of studies regarding the production of OXs, as precursors of CXs, using the conventional synthesis method (Naseri I., et al. 2014). In comparison, there is a great lack in the synthesis of OXs using MW radiation as a heating source and the effect of the synthesis conditions on the OXs produced, and as a result on the CXs characteristics. In addition, a large shortage in the studies regarding the surface chemistry and texture characteristics of MW assisted synthesized CXs was observed, although these features determine their behavior towards various applications. A comparative study between the characteristics of CXs produced under the same condition but with these two different techniques (i.e. MW assisted synthesis and conventional synthesis methods) was not given suitable attention (Calvo et al., 2011).

The object of the present investigation is to examine the influence of the R-F solution pH value, as well as the CT on the physicochemical, surface chemistry and pore texture of the MW assisted synthesized CXs, in an attempt to find out the best conditions for preparing high quality CXs by this method. Also, to correlate their surface and textural characteristics with their adsorption capacity for removal of MB (as a standard medium-size organic dyestuff) from aqueous solution. The study was extended, also, to compare the effect of the synthesis method (the MW assisted synthesis and the conventional synthesis methods) on the CX surface chemistry and pore texture characteristics. The results confirmed that there is no need for using the very long conventional synthesis method for CX synthesis, not only because MW assisted method save a lot of time (5 hours compared with 7 days for the conventional method), but also because it produces higher yield, higher carbon content, more surface functional groups, higher surface area, wider pore volume, and more developed mesoporosity.

Experimental:-

Synthesis of Organic and Carbon Xerogels:-

Resorcinol-formaldehyde (R-F) organic xerogels (OXs) were synthesized using two different techniques (microwave-induced synthesis and conventional synthesis methods).

Microwave - induced synthesis Method:-

For the MW-induced synthesis of OXs, the precursor solutions were prepared by the polycondensation of resorcinol and formaldehyde using deionized water as solvent. Resorcinol (Alfa easer, 99%) was first dissolved in deionized water. Then, formaldehyde (Aldrich, 37 wt. % in water, stabilized by 10.7 wt. % methanol) was added under magnetic stirring until obtaining a homogenous solution. The (R/F) molar ratio was (0.5) and the dilution ratio (i.e. total solvent/ reactant molar ratio) was fixed at (5.70). The dilution ratio includes the added deionized water and the water and methanol contained in the formaldehyde in the case of “total solvent”, while “reactant” refers to the resorcinol and formaldehyde. NaOH (1M) and HNO₃ (1M) were added to adjust the pH of the R-F solutions to the

required pH values. The precursor solutions were placed in unsealed glass bottles transparent to MW, and these were then introduced into a multimode microwave oven [MILSTONE START SYNTH 1200 W/ 2450 MHZ]. First, the temperature was kept at 85°C for ~ 3 h for gelation and permitting part of the curing stage to take place. Then, the microwave device was allowed to operate at power (750 Watt) until synthesis had been completed. The total synthesis time of the OX was around 5h. Five OX samples variable in their R-F solutions pH values (between 3.0 and 7.2) were synthesized. These samples were designated as OX3.0(MW), OX5.0(MW), OX6.0(MW), OX6.5(MW) and OX7.2(MW), where the letters OX refer to the organic xerogel and the numbers indicate the R-F solution pH value, whereas (MW) indicates the use of MW-induced synthesis method. Pictures of the OX6.5(MW) sample obtained by the MW induced synthesis method are given in Figure 1(a).

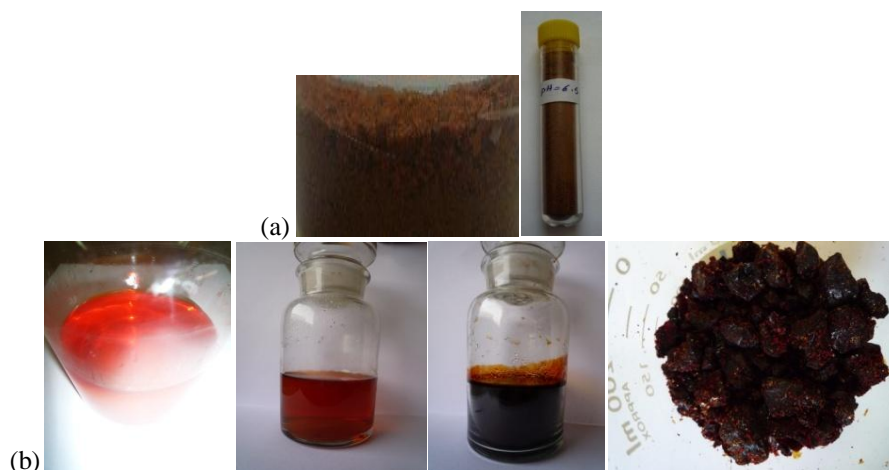


Fig. 1:- Photos of the OX6.5(MW) sample prepared by the MW assisted synthesis method (a), and of the steps of the conventional synthesis method of the OX6.5(C) sample (b).

To study the effect of the R-F solutions pH values on the carbon xerogels characteristics, the OX samples were pyrolyzed at a fixed temperature (750°C) under nitrogen flow in a vertical tubular quartz reactor for 2h. with a heating rate of 50 °C. min⁻¹ to give CXs. These samples were designated as CX3.0(MW)750, CX5.0(MW)750, CX6.0(MW)750, CX6.5(MW)750 and CX7.2(MW)750, where the last number indicates the carbonization temperature. For studying the effect of the carbonization temperature, the OX6.5(MW) sample was selected and pyrolyzed, as described above, at different temperatures in the range 600–800 °C, to produce four CXs variable in their CT. These samples were designated as CX6.5(MW)600, CX6.5(MW)700, CX6.5(MW)750, and CX6.5(MW)800.

Conventional synthesis Method:-

To study the effect of synthesis method on the CX characteristics, another OX sample was prepared by the conventional method at R-F solution pH value of 6.5 and CT of 750 °C to give the last CX sample. These conditions were found to be the best effective ones on the characteristics of the CXs prepared by the MW-induced synthesis method. To synthesize the OX sample by the conventional method, the pH was adjusted to the required value (6.5) using NaOH and HNO₃. The solution was placed in a stopper bottle, which was then introduced in an electrical oven at 85 °C for 72 h. to undergo gelation and curing stages. Afterwards, the bottle was opened and the oven temperature was set at 100 °C for 4 days with the purpose of drying the organic gel via evaporation of the solvent until the process of drying of the organic gel was ended. This sample was designated as OX6.5(C), where (C) indicates the synthesis using the conventional method. Pictures of the steps of the conventional synthesis method of the OX6.5(C) sample are given in Figure 1(b). Finally, the OX6.5(C) sample was pyrolyzed at 750°C under nitrogen flow. This sample was designated as CX6.5(C)750.

Physicochemical and Surface Chemistry Studies:-

Determination of the pH of the Investigated Carbon Xerogels:-

For measuring the pH values of aqueous suspensions of the CX samples, 1.0 gram portions of finely powdered carbon were mixed with 20 cm³ distilled water in 50 cm³ capacity stopper Pyrex bottles. The suspensions were

shaken mechanically for 48 h. at 25°C, following which their pH values were determined using a Jenway 3510 pH meter.

Elemental Analysis:-

The carbon, hydrogen and oxygen contents of the samples were analyzed using Elementary Analyzing-System GmbH, Vario EL V2.3.07 July 1998, CHNS Mode. For the sake of quality assurance and quality control, duplicated analysis was conducted on all samples.

Determination of Active Sites:-

The acidic and basic active sites were determined according to the method of Boehm (Boehm, 2002). 0.2 g of CX sample was placed in 25 ml of the following (0.1N) solutions: sodium hydroxide, sodium carbonate, sodium bicarbonate and hydrochloric acid. The vials were sealed and shaken for 24 h. and then 5 ml of each filtrate was pipetted and the excess of base and acid was titrated with standard HCl and NaOH, respectively. The numbers of acidic sites of various types were calculated under the assumption that NaOH neutralizes carboxyl, phenolic and lactonic groups, while Na_2CO_3 neutralizes carboxyl and lactonic groups, and NaHCO_3 neutralizes only carboxyl groups. The number of surface basic sites was calculated from the amount of HCl which reacted with the CX.

FTIR Analysis:-

The FTIR spectra of the CX samples were recorded using the KBr disc technique in the range $4000\text{--}400\text{ cm}^{-1}$ at the resolution of a Genesis-II Fourier transform infrared spectrophotometer, Shimadzu (Japan), model (Nicolet 6700). About 10 mg of the finally powdered sample was mixed well with 200 mg of KBr powder (spectroscopically pure) in mortar. The mixture was then pressed in a special disc under vacuum at about 8 ton/cm^2 by means of a hydraulic press. The disc produced was 1.2 cm in diameter and about 0.7 mm thickness.

XRD Analysis:-

XRD patterns for the CX samples were recorded using a Philips type Diffractometer (Model PW 2103/00) using Ni-filtered $\text{Cu K}\alpha$ radiation at $\lambda=1.5418\text{ \AA}$. The powders were mounted on silicon monocrystal sample holders. Data acquisition was realized in the range ($4\text{--}80^\circ$) with scan step size of 0.04° . The average particle size of each sample was calculated from Scherrer formula (Khetre et al., 2011) ($t = K \lambda / \beta \cos \theta$, where t is the average size of the particles, assuming particles are spherical, $K=0.9$, λ is the wave length of X-ray radiation, β is the full width at half maximum of the diffracted peak and θ is the angle of diffraction).

Scanning Electron Microscope (SEM) Investigations:-

Scanning electron micrographs of the CX6.5(MW)750 sample were obtained using a JEOL Scanning Microscope, Model JSM-5400 LV (Jeol, Tokyo, Japan). The sample was prepared by the gold sputtering technique (Girgis, 1993), by sprinkling the powder lightly onto a double-sides adhesive tape, which was mounted on a SEM specimen stub. The edge of the double-sided tape was coated with silver paint to minimize charging. Finally, the sample was sputter-coated with gold. Micrographs were obtained in a secondary electron imaging mode using a potential difference of 25 kV.

Transmission Electron Microscope (TEM) Investigations:-

TEM micrographs of the CX6.5(MW)750 sample were recorded using High Resolution Transmission Electron Microscope (HRTEM) TECANI G^2 spirit TWIN. The sample was prepared by mixing the powder with ethanol, and then subjected to sonication for 10 min., and placing a drop onto former-backed carbon coated copper grid. Transmission micrographs were obtained using accelerating voltage 120 kV, conducted by VELETA camera.

Pore Texture Studies:-

To evaluate the textural parameters of the xerogel samples under investigation, the adsorption / desorption isotherms of nitrogen at 77 K were constructed using NOVA 3000 Multi-Station High Speed Gas Sorption Analyzer (Quanta-Chrome Corporation), Version 6.07. Approximately (0.1- 0.2) grams of sample were originally out gassed at 523 K for about 5 h. under a vacuum of about 1.3×10^{-5} KPa. After cooling, the samples were kept in a N_2 atmosphere and later evacuated with a drag pump. The results were expressed in relation to unit mass of out gassed CX. The true adsorption equilibrium checked at regular intervals, following it during long periods. The adsorption isotherms were analyzed to get various porous parameters: i) The BET-equation parameters (Brunauer et al., 1938): S_{BET} (BET-surface area, $\text{m}^2.\text{g}^{-1}$), C_{BET} (the constant C of the BET equation) and $V_{\text{P(total)}}$ (total pore volume, cc.g^{-1}), and ii) The

α_S -plot parameters (Sellés-Pérez and Martín-Martínez, 1991): s_t^α (total surface area, $\text{m}^2.\text{g}^{-1}$), s_n^α (non-microporous surface area, $\text{m}^2.\text{g}^{-1}$), s_{mic}^α (microporous surface area, $\text{m}^2.\text{g}^{-1}$), v_o^α (micropore volume, $\text{cc}.\text{g}^{-1}$), \bar{r} (mean pore radius, Å) and V_{meso} (mesopore volume, $\text{cc}.\text{g}^{-1}$).

Methylene Blue Adsorption Studies:-

A stock aqueous solution of MB (1000 ppm) was prepared. Other solutions of MB were prepared from this solution by dilution to the required concentration. The adsorption of MB was made by placing 50 mg of each of powdered CX, which was dried in an oven at 110°C for 2 h, in a stopper Pyrex bottles containing a 50 ml of dye solution. The bottles with their contents were shaken occasionally for 72 h. at 25°C . Preliminary experiments established that equilibrium times of 48h. were needed to determine solution adsorption isotherms for CXs. The supernatant liquid was removed and analyzed spectrophotometrically using a double beam computerized UV/Visible Scanning Spectrophotometer [type Shimadzu (UV-2101 PC)] and $\lambda_{max} = 662.5 \text{ nm}$. The amount adsorbed was calculated from the difference between the initial and the equilibrium concentrations of MB in the aqueous solution (Girgis and El-Hendawy, 2002). The experimental error was about 5%. Reproducibility was ensured by repeating the experiment under the same conditions.

Results and Discussion:-

Physicochemical and Surface Chemistry Characteristics:-

Yield of Carbon Xerogel:-

Carbonization of the prepared OXs causes removal of the captured solvent, release of volatile matter including residual organic components and non cross linked organic chains, and volatilization of labile oxygen and hydrogen leading to an additional weight loss. This results in the formation of thermally stable nanostructured material formed mainly by carbon. The results of the % yield (expressed as the ratio between the mass of CX and the mass of OX under different conditions) are summarized in Table (1). The results indicate the following: 1) The CX yield depends largely on the R-F solution pH values. It increases continuously with increasing the pH value from 3.0 (46.6 %) up to 6.5 (66.0 %) then declines at pH 7.2 (54.8 %). 2) The CX yield depends also on the CT. As expected, as the CT was increased the yield was reduced. It decreases continuously from 72.0 to 60.0 wt % by increasing the CT from 600 to 800°C , which can be attributed to more and more release of oxygen and hydrogen, as well as a decrease in carbon content, with raising the CT. 3) The synthesis method largely affects the yield. MW induced synthesis method promotes the yield of CX by 10 % compared to the conventional method (66.0 and 56.0 %, respectively). The higher CX yield of the microwave assisted method is a very important factor favors it over the conventional method.

Elemental Analysis:-

The results of elemental analysis of the CXs investigated are presented in Table (1). It can be observed that the content of elemental carbon, hydrogen and oxygen depends on the preparation conditions, as follows: 1) The content of carbon depends on the original R-F solution pH value. It decreases very slightly with the increase of the pH value from 3.0 (94.19 %) to 5.0 (94.11 %), then continuously up to pH 7.2 (87.24 %). 2) The content of carbon, also, depends on the CT. As the CT was increased the content of carbon was decreased. It decreases linearly from 91.45 % to 81.45 % by raising the CT from 600 to 800°C . 3) Carbon content depends, also, on the synthesis method. The MW induced synthesis method shows a higher content of carbon compared to the conventional method (88.71% and 83.37 %, respectively).

Slurry pH of the Investigated CXs Surfaces:-

The pH value of a carbon surface represents the average effect of species of various strength and number, i.e. the average effect of both acidic and basic groups existing on the surface. As an indication of pH of the CX surface, the slurry pH of a CX suspension was measured. All values were obtained at the same conditions. The pH values of the investigated CXs surfaces are given in Table (2). As deduced from the pH values, the chemical nature of the CX surfaces is different. 1) The surface pH value depends largely on of the original R-F solution pH value. It increases linearly with increasing the pH value from 3.0 up to 7.2. 2) The surface pH value is influenced also with the CT. As the CT was increased, the surface pH value was increased. It increases continuously from 6.04 to 7.02 by raising the CT from 600 to 800°C . The acidic functional groups are easily decomposed with raising the CT (Bansal and Goyal, 2005) leading to a decrease of the acidity of the prepared CXs and, as a result, an increase of the pH value. 3) The chemical nature of the CX surfaces depends on the synthesis method. The MW induced synthesis method shows slightly acidic character compared to the conventional method (pH= 6.70 and 7.21, respectively).

Table 1:- Yield (wt.%), elemental analysis and average particle size of the investigated CX samples.

Sample Notation	% Yield	Elemental analysis						Average particle size* (nm)
		wt.% of the element			Atomic content			
		C	H	O	C _{at}	H _{at}	O _{at}	
Effect of Initial pH								
CX3.0(MW)750	46.6	94.19	2.28	3.53	7.85	2.28	0.22	9.0
CX5.0(MW)750	49.6	94.11	1.34	4.55	7.84	1.34	0.28	8.0
CX6.0(MW)750	51.8	92.63	1.43	5.94	7.72	1.43	0.37	9.0
CX6.5(MW)750	66.0	88.71	3.04	8.25	7.39	3.04	0.51	5.9
CX7.2(MW)750	54.8	87.24	2.59	10.16	7.27	2.59	0.63	6.4
Effect of CT								
CX6.5(MW)600	72.0	91.45	2.88	5.67	7.62	2.88	0.35	8.0
CX6.5(MW)700	68.6	87.41	4.34	8.25	7.28	2.67	0.51	9.7
CX6.5(MW)750	66.0	88.71	1.37	9.92	7.39	3.04	0.08	5.9
CX6.5(MW)800	60.0	81.45	2.98	15.29	6.79	2.97	0.96	9.0
Effect of synthesis method								
CX6.5(C)750	56.0	83.37	4.00	12.63	6.95	4.00	0.79	6.2
CX6.5(MW)750	66.0	88.71	1.37	9.92	7.39	3.04	0.09	5.9

*Calculated by Scherrer formula .

Surface acidic and basic active sites:-

The chemical properties of the investigated CXs surfaces were assessed by neutralization with bases of various strengths (NaHCO₃, Na₂CO₃ and NaOH) and HCl. According to Boehm (Boehm, 2002), the bases consumption can be described in the presence of surface acidic groups with various strength while the acid uptake characterizing the carbon surface basicity. Only the strong acidic groups (carboxyl) can be neutralized by NaHCO₃ (pK_a= 6.37), whereas Na₂CO₃ (pK_a= 10.25) titrates carboxyl and lactones. NaOH (pK_a= 15.74) neutralizes carboxyl, lactones and phenolic (hydroxyl) groups. The results of the acid-base titration to determine the surface acidic and basic active sites by the Boehm method are shown in Table (2). Total surface acidic sites included phenolic, lactonic and carboxylic sites, whereas basic sites may include chromene-, pyrone-, and quinone-type structures (Boehm, 2002). The most distinguishing feature of these CXs is the co-existence of the acidic and basic sites; i.e. the representation of all types of surface functional groups. Thus, these CXs are amphoteric in nature. The concentration of the surface acidic and basic active sites depends on the CX synthesis conditions. The analysis of the data given in Table (2) provided the following: 1) The surface acidic and basic active sites depend largely on the original R-F solution pH value. The total surface acidic sites decrease linearly from 2.37 to 1.00 meq.g⁻¹, whereas the total surface basic sites increase continuously from 0.50 to 1.25 meq.g⁻¹ with raising the pH value from 3.0 up to 7.2. Thus, increasing the initial pH from 3.0 to 7.2 (i.e. increasing the amount of basic catalyst) results in a decrease in the acidic function groups and an increase in the basic function groups. 2) The surface acidic and basic active sites depend largely on the CT. The total surface acidic sites decrease from 1.30 to 0.85 meq.g⁻¹, whereas the total surface basic sites increase from 0.60 to 1.22 meq.g⁻¹ by raising the CT from 600 to 800 °C. The decrease in the surface acidic groups by increasing the CT indicates that these groups are thermally unstable, whereas the surface basic groups are stable up to 800°C, as reported by Bansal and Goyal (Bansal and Goyal, 2005). 3) The surface acidic and basic active sites depend on the synthesis method. The MW induced synthesis method shows more total surface acidic and basic sites compared to the conventional method. It is possible that, during the microwave synthesis, some secondary reactions take place producing a higher cross linked organic gel with more stable oxygen functional groups (Caddick, 1995; Le Van and Gourdenne, 1987). These oxygen groups will remain after the carbonization process in a higher extent than in the CXs prepared conventionally. So, to develop more functional groups on the CX surface, it is advisable to use MW assisted synthesis method.

Table 2:- pH values of the investigated CXs surfaces, and number of surface species (meq.g⁻¹) obtained from Boehm titration.

Sample notation	pH	Carboxylic sites	Lactonic sites	Phenolic sites	Total Acidic sites	Basic sites	Total surface coverage
Effect of pH							
CX3.0(MW)750	2.65	0.51	0.44	1.42	2.37	0.50	2.87
CX5.0(MW)750	4.76	0.32	0.30	1.13	1.75	0.80	2.55
CX6.0(MW)750	5.89	0.27	0.25	0.73	1.25	1.20	2.45
CX6.5(MW)750	6.70	0.25	0.24	0.72	1.21	1.20	2.41
CX7.2(MW)750	7.31	0.20	0.16	0.64	1.00	1.25	2.25
Effect of CT							
CX6.5(MW)600	6.04	0.09	0.76	0.45	1.30	0.60	1.90
CX6.5(MW)700	6.20	0.13	0.48	0.66	1.27	1.00	2.27
CX6.5(MW)750	6.70	0.25	0.24	0.72	1.21	1.20	2.41
CX6.5(MW)800	7.02	0.29	0.12	0.75	0.85	1.22	2.07
Effect of Synthesis method							
CX6.5(C)750	7.21	0.09	0.18	0.37	0.64	0.85	1.49
CX6.5(MW)750	6.70	0.25	0.24	0.72	1.21	1.20	2.41

Identification of surface functional groups by FTIR:-

This study provides qualitative information on the chemical structure of the investigated CX surface. Figure (2) displays the FTIR absorption spectra obtained for the CX samples prepared under different conditions. It is clear that these samples have nearly the same absorption peaks, which ensures the fact that most porous carbon materials are characterized by the presence of carboxylic, phenolic hydroxyl, carbonyl and lactone groups (Shafeeyan et al., 2010). However the difference in the relative intensities of the respective bands indicate that some differences are present in the chemical structure of these samples due to the different preparation methods and conditions. Analysis of the absorption bands for the CX6.5(MW)750 sample, for example [c.f. Figure (2 (a))], shows the following: 1) The absorptions displayed in the range 3688-3082 cm⁻¹ as broad band with maximum absorption centered at 3400 cm⁻¹ is ascribed to O-H stretching vibrations [$\nu(\text{O-H})$] due to the existence of surface hydroxyl groups, with participation of adsorbed water. The asymmetry of this band at lower wave number indicates the presence of strong hydrogen bonds (O'reilly and Mosher, 1983). 2) The weak absorptions displayed in the range 2997-2750 cm⁻¹ as broad band centered at 2918 cm⁻¹ are ascribed to C-H symmetric and asymmetric stretching of some aliphatic species on the sample, i.e. methyl and methylene groups (Li et al., 2013). 3) The absorptions displayed in the range 1360-1138 cm⁻¹ with a maximum centered at 1200 cm⁻¹ can be assigned to C-O stretching in alcohols, phenols, ethers and esters (Figueiredo et al., 1999). 4) The absorption bands displayed in the range 1695-1397 cm⁻¹ with maximum absorptions centered at 1600 and 1460 cm⁻¹ can be attributed to the stretching vibrations of the C=O moieties in carboxylic and lactone systems, overlapping the C=C stretching vibrations of aromatic rings of quinone and keto-enol groups (Figueiredo et al., 1999). The results obtained in the present FTIR absorption studies are in agreement with those obtained by the Boehm-titration method.

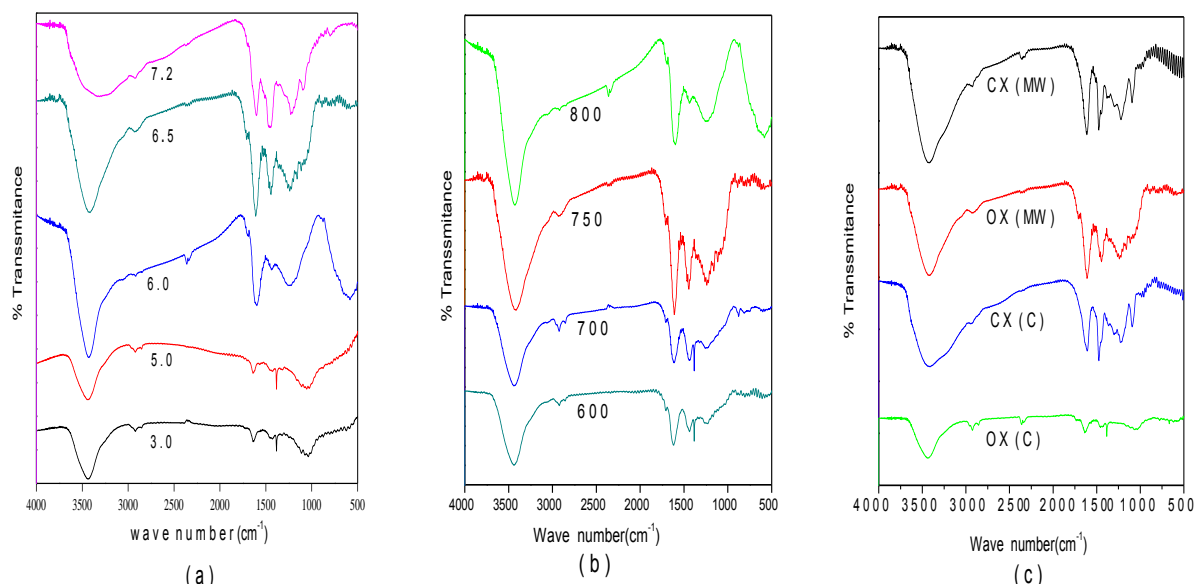
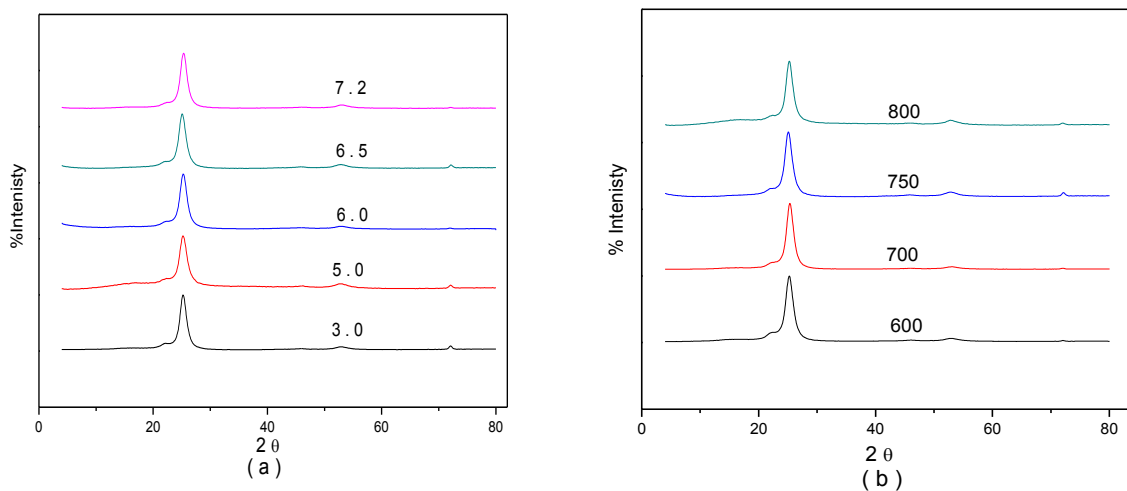


Fig. 2:- FTIR spectra of the CX samples obtained under different conditions: (a) different R-F solutions pH values, (b) different CTs and (c) different synthesis methods.

XRD Analysis:-

The X-ray diffraction patterns of the CX samples obtained under different conditions are given in Fig.3 (a-c), whereas those of the OX samples are represented in Fig.3 (d). The peak located at 25.3 can be assigned to the maximum diffraction of graphitic carbon (002) plane (Yoon et al., 2005). Thus, the XRD results indicate that all CX samples are crystalline in nature and consists of a single phase with a graphite structure, as reported also by Oyedoh et al. (Oyedoh et al., 2013). On the other hand, the OX samples are semi crystalline in nature, as shown in figure 3(d). The average crystallite size of the CX samples obtained under different conditions are given in Table (1). It can be observed that the average crystallite size is in the nanoscale range between 5.9 and 9.7 nm.



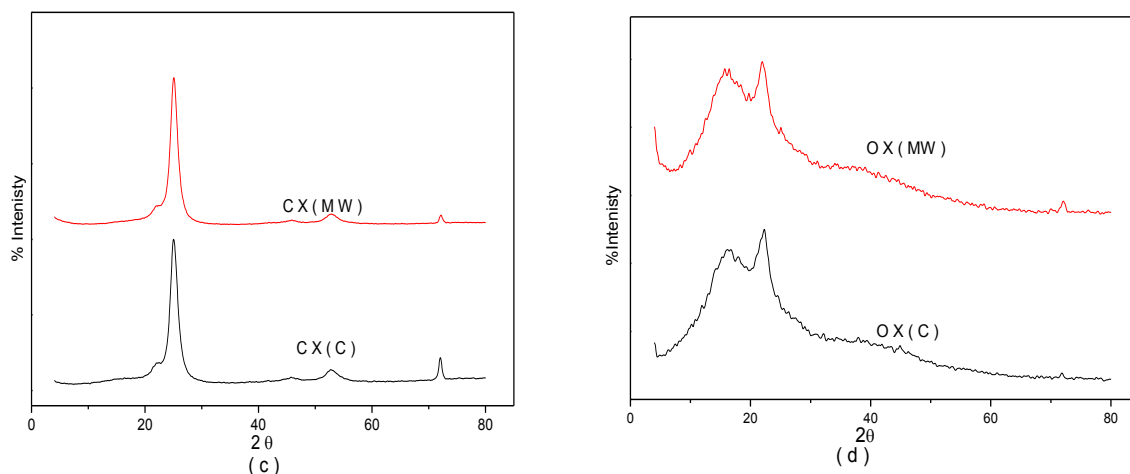


Fig. 3:- XRD patterns recorded for the CX samples obtained under different conditions: (a) different R-F solutions pH values, (b) different CTs and (c) different synthesis methods, also for the OX samples prepared with different methods (d).

Scanning Electron Microscope (SEM) Observations:-

The surface features of a selected sample [CX6.5(MW)750] was examined by SEM for structure, degree of uniformity and the development of porosity. The SEM results at high magnification, Figure (4(b)), confirm the fine and nanostructured nature of the CX under consideration with fine nanosized grains, wide interparticle voids, interconnected particles with irregular shapes. A uniform pore distribution all over the surface was also observed.

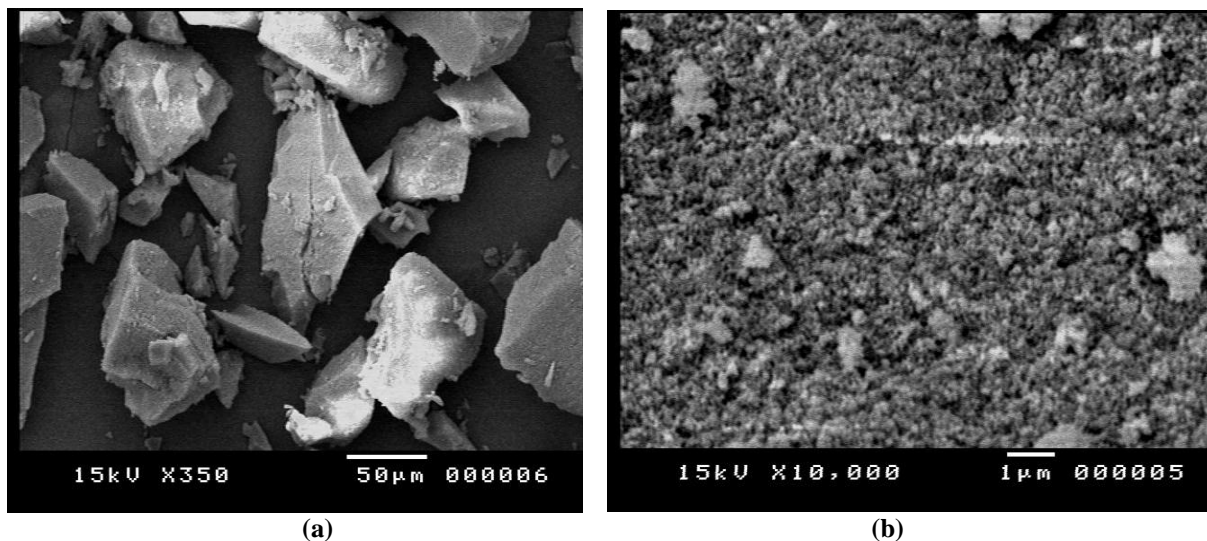


Fig. 4:- SEM micrographs of the CX6.5(MW)750 sample at two different magnifications.

Transmission Electron Microscope (TEM) Investigations:-

The TEM micrographs in Figure (5) at two different magnifications confirms the fine and nanostructured nature of the CX sample. They show distinctly beads of pearl nature of the very fine particles (< 50 nm) and the pores which are originated by the interconnection of CX particles.

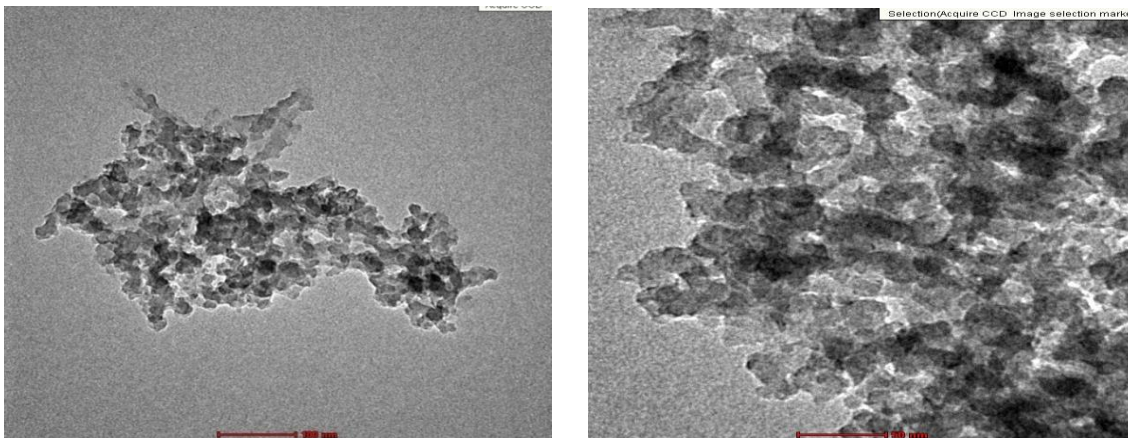
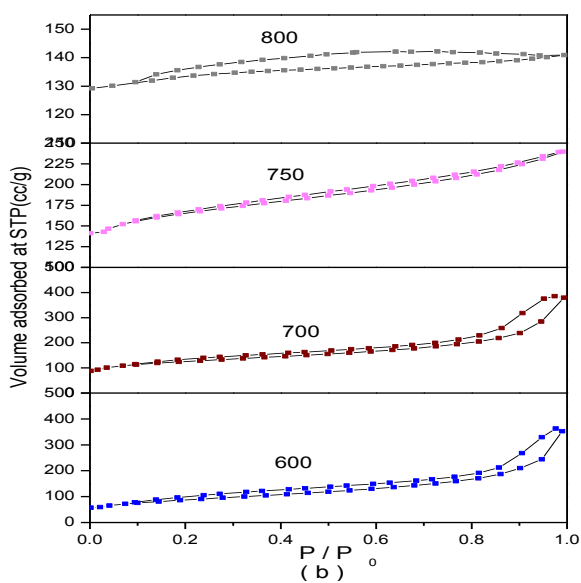
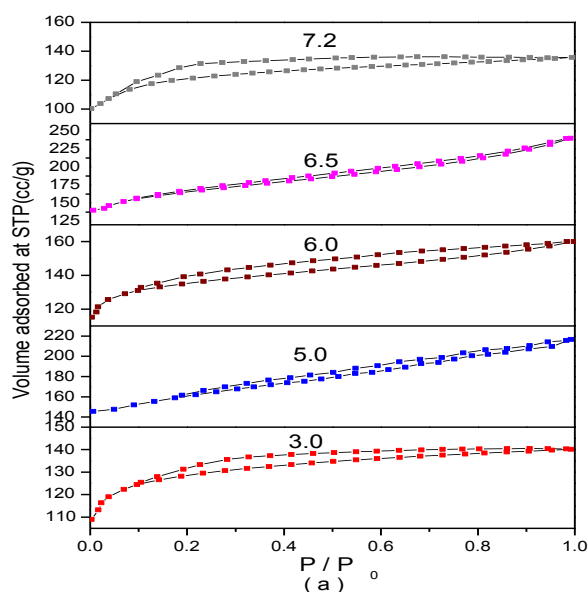


Fig. 5:- TEM micrographs of the CX6.5(MW)750 sample at two different magnifications.

Pore Texture characteristics:-

The sorption isotherms of N_2 at 77 K for the CX samples synthesized at different initial R-F solutions pH values are given in Fig. (6 (a)),



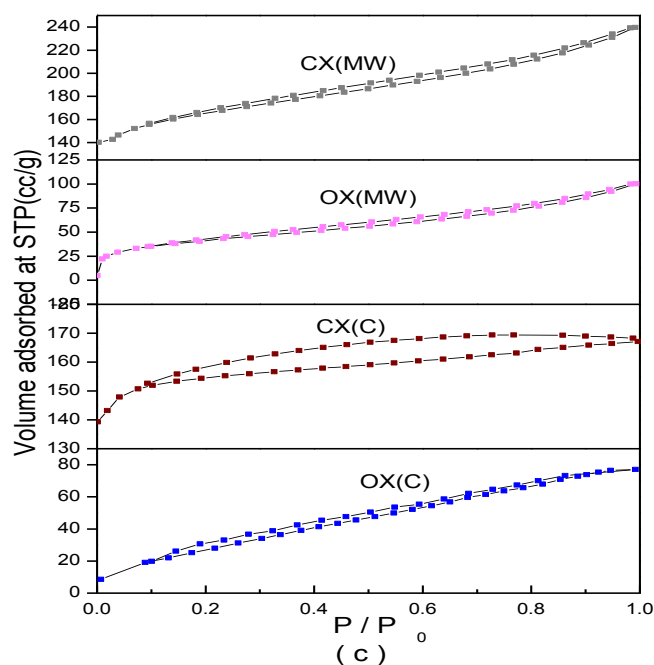
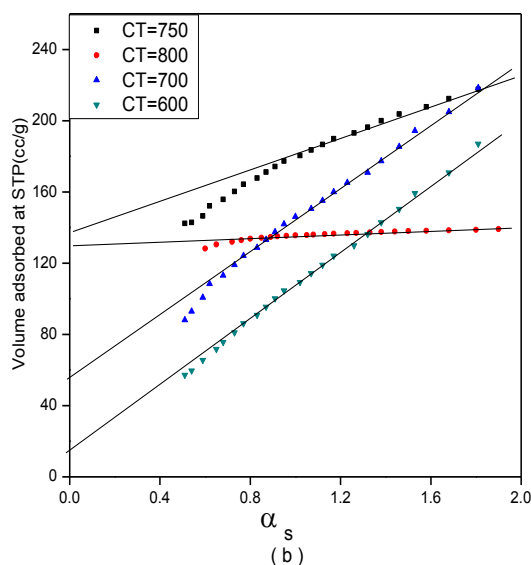
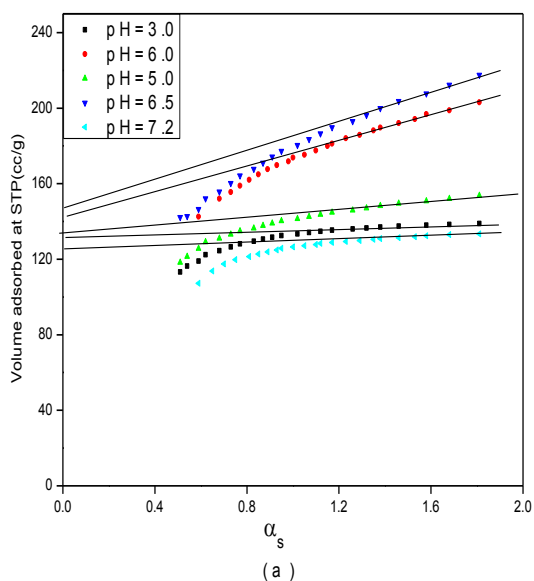


Fig. 6:- Nitrogen sorption isotherms at 77K for the CXs obtained under different conditions: (a) different R-F solutions pH values, (b) different CTs and (c) different synthesis methods.

whereas their α_s -plots are shown in Fig. (7 (a)). As shown in Fig. (6 (a)), the type of the isotherm depends largely on the R-F pH value. The adsorption isotherm of the CX3.0(MW)750 sample exhibits type I feature in the BDDT classification (Brunauer et al., 1940) with very sharp "knee" at the low pressures end ($P/P_0 < 0.02$) and very little of the type II features at relative higher pressures. This indicates a developed microporosity with some mesoporosity and capillary condensation (Pradhan and Sandle, 1999). The type II features increase with raising the R-F pH value. The isotherm of the CX6.5(MW)750 sample exhibits type II feature which indicates the more development of mesoporosity, i.e. the presence of mixed micro- and mesoporosity.



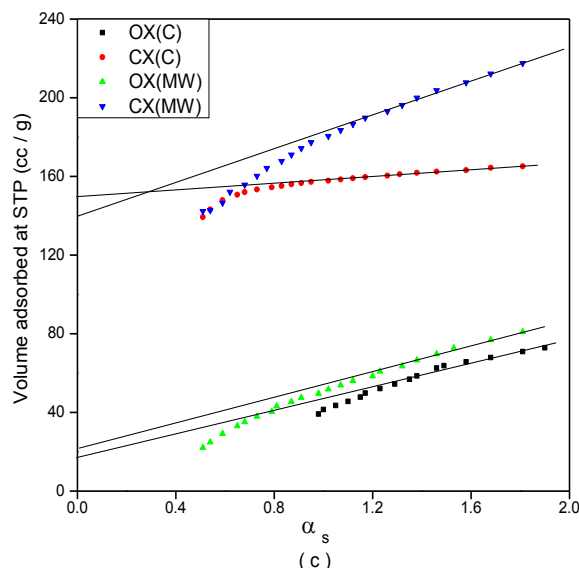


Fig. 7:- α_s -Plots for the CXs obtained under different conditions: (a) different R-F solutions pH values, (b) different CTs and (c) different synthesis methods.

The α_s -plots complement the observations obtained from the sorption isotherms, as these plots exhibit features of the type (f) of the α_s -plots classification (Sellés-Pérez and Martín-Martínez, 1991). The α_s plot in (f) is derived from a Type I isotherm or composite of Type I and II isotherms; here micropore filling is followed by multilayer adsorption on a small external surface (Carrott et al., 1987). The evaluated texture characteristics obtained from the BET-equation and α_s -plots are summarized in Table (3). A comparative representation of the data for total surface area (s_t^a), micropore surface area (s_{mic}^a) and non microporous surface area (s_n^a) obtained from the α_s -plots for these CX samples, prepared at different initial R-F solution pH values, are given in Fig. (8 (a)). The following can be observed: 1) The evaluated texture parameters of the CXs prepared under different R-F pH values indicate that these CXs are in general microporous (s_{mic}^a / s_t^a in the range 72.0- 97.0 %) with some mesoporosity (s_n^a / s_t^a in the range 3.0-28.0 %). 2) As the pH increased from 3.0 to 6.5, the surface area increased from 484 to 622 $\text{m}^2 \cdot \text{g}^{-1}$, then decreased sharply to 412 at pH 7.2. Increasing the pH value from 3.0 to 6.0 leads to a greater cross-linking of the already formed clusters due to lower concentration of protons in the media which resulted in a more porous polymeric structure (Zanto et al., 2002b). On the other hand, increasing the pH up to 7.2 causes the condensation reaction to be hindered leading to less cross-linking of the structures and resulting in a much weaker porous structure than at lower pH value. 3) The CX6.5(MW)750 sample possesses the highest total and micropore surface area parameters. It has mixed micro- and mesoporosity. The mesopore volume for this sample is ~ 43 % of the total pore volume.

The sorption isotherms of N_2 at 77 K for the CX samples synthesized at fixed R-F pH value (pH = 6.5) and different CTs are given in Fig.(6 (b)), whereas their α_s -plots are shown in Fig.(7 (b)). These isotherms exhibit type II features in the BDDT classification with sharp "knee" at the low pressures end, which indicates carbons with mixed micro- and mesoporosity. The evaluated texture characteristics are also summarized in Table (3). A comparative representation of the data for s_t^a , s_{mic}^a and s_n^a obtained from the α_s -plots for these samples are given in Fig. (8 (b)). The following can be observed: 1) The texture parameters of the CX samples prepared at fixed R-F pH value (pH = 6.5) and different CTs reflect that raising the CT is accompanied by a continuous development in the total surface area (s_t^a) with a maximum value at 750°C. 2) The micropore surface area (s_{mic}^a) increases continuously with raising the CT up to 800°C. The s_{mic}^a / s_t^a value increases from 17.7% to 98.2 % by raising the CT from 600°C up to 800°C. Also, the V_{mic} / V_p (total) value increases continuously from 5.1 % to 93.6 %. 3) The non-microporous surface area (s_n^a) decreases sharply with raising the CT up to 800°C. 4) When the pyrolysis temperature increases, the micropore volume first increases (due to the opening of the closed pores and a widening of the narrowest micropores already present), then passes through a maximum ($0.212 \text{ cm}^3 \cdot \text{g}^{-1}$) at 750 °C and finally decreases to reach ($0.203 \text{ cm}^3 \cdot \text{g}^{-1}$) at 800 °C.

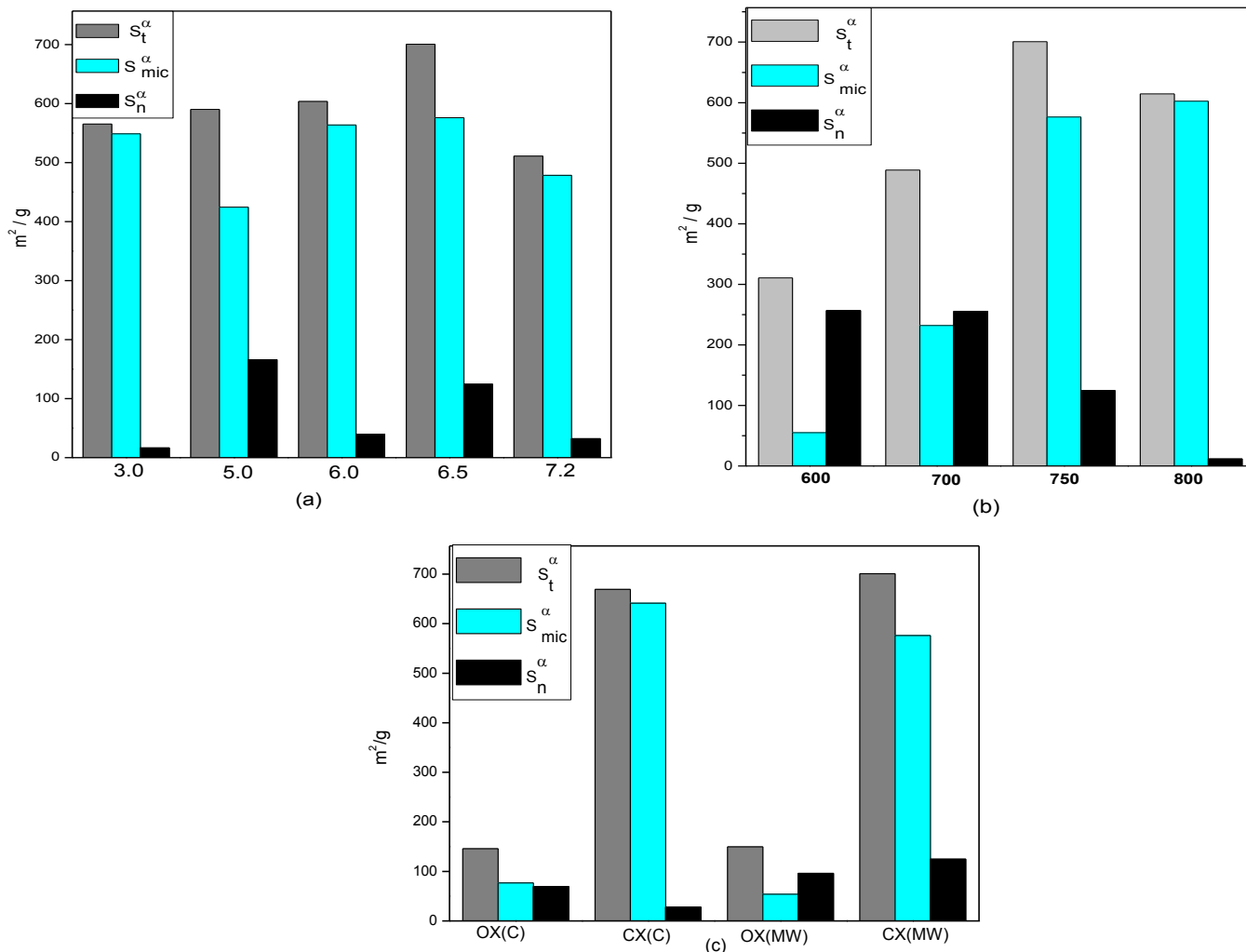


Fig. 8:- A comparative representation of the data for total surface area (S_t^α); microporous surface area (S_{mic}^α); and non-microporous surface area (S_n^α); obtained from the α_s -plots for the CXs obtained under different conditions: (a) different R-F solutions pH values, (b) different CTs and (c) different synthesis methods.

The sorption isotherms of N_2 at 77 K for the OX and CX samples synthesized by the two different methods are given in Fig. (6 (c)), whereas their α_s -plots are shown in Fig. (7 (c)). As shown in Fig. (6 (c)), the type of the isotherm of the organic or carbon xerogel depends on the synthesis methods. The adsorption isotherm of the OX6.5(C) and CX6.5(C)750 samples exhibits type I feature in the BDDT classification with sharp "knee" at the low pressures end and very little of the type II features at relative higher pressures ($> 0.65 P/P^0$). This indicates a developed microporosity with some mesoporosity and capillary condensation. On the other hand, the isotherm of the OX6.5(MW) sample exhibits type II feature which indicates the presence of mixed micro- and mesoporosity. The isotherm of the CX6.5(MW)750 sample preserves the type II feature with sharp "knee" at the low pressures end ($P/P^0 < 0.05$) indicating also a carbon with micro- and mesoporous structure.

Table 3:- Porous characteristics of the organic and carbon xerogels under different synthesis conditions

Sample notation				α_s - plots						$S_{mic}^\alpha / S_t^\alpha$ (%)	S_n^α / S_t^α (%)	$V_{mic} / V_{p(total)}$ (%)	$V_{meso} / V_{p(total)}$ (%)
	S_{BET} (m ² /g)	C_{BET}	$V_{p(total)}$ (cc/g)	S_t^α (m ² /g)	S_n^α (m ² /g)	S_{mic}^α (m ² /g)	V_0^α (cc/g)	\bar{r} (Å)	V_{meso} (cc/g)				
Effect of pH													
CX3.0(MW)750	484	1455	0.217	566	17	549	0.198	7.6	0.019	97.0	3.0	91.2	8.8
CX5.0(MW)750	498	1285	0.374	590	165	425	0.177	12.6	0.197	72.0	28.0	47.3	52.7
CX6.0(MW)750	513	567	0.247	604	40	564	0.198	9.1	0.049	93.4	6.6	80.2	19.8
CX6.5(MW)750	622.	368	0.370	701	125	576	0.212	10.6	0.158	82.2	17.8	57.3	42.7
CX7.0(MW)750	412	155	0.210	511	32	479	0.177	8.2	0.033	93.7	6.3	84.3	15.7
Effect of CT													
CX6.5(MW)600	308	256	0.545	311	256	55	0.028	7.6	0.517	17.7	82.3	5.1	94.9
CX6.5(MW)700	449	410	0.586	489	257	232	0.081	23.9	0.505	47.4	52.6	13.8	86.2
CX6.5(MW)750	622	368	0.370	701	125	576	0.212	10.6	0.158	82.2	17.8	57.3	42.7
CX6.5(MW)800	433	82	0.217	614	11	603	0.203	7.06	0.014	98.2	1.8	93.6	6.4
Effect of synthesis method													
OX6.5(C)	144	1179	0.119	146	69	77	0.041	19.5	0.078	52.7	47.3	34.5	65.5
OX6.5(MW)	144	215	0.155	150	96	54	0.003	2.07	0.152	36.0	64.0	1.9	98.1
CX6.5(C)750	541	449	0.259	669	28	641	0.228	7.73	0.031	95.8	4.2	88.0	12.0
CX6.5(MW)750	622	368	0.370	701	125	576	0.212	10.6	0.158	82.2	17.8	57.3	42.7

The α_s -plots complement the observation obtained from the sorption isotherms. For the OX6.5(C) and CX6.5(C)750 samples, these plots exhibit features of the type (f) of the α_s -plots classification, where the micropore filling is followed by multilayer adsorption on a small external surface. On the other hand, the OX6.5(MW) and CX6.5(MW)750 samples exhibit features of the α -2 type of the α_s plots classification, indicating developed carbon microporosity with some mesoporosity.

The evaluated texture characteristics obtained from the BET-equation and α_s -plots are summarized also in Table (3), which reflects that the organic and carbon xerogels pore texture depends on the synthesis method. A comparative representation of the data for S_t^α , S_{mic}^α and S_n^α obtained from the α_s -plots for the organic and carbon xerogels under conventional and MW synthesis methods are given in Fig.(8 (c)).The following can be observed: 1) Comparing the texture parameters of the OXs [OX6.5(C) and OX6.5(MW) samples] with those of the corresponding CXs [CX6.5(C)750 and CX6.5(MW)750 samples, respectively], reflect that the carbonization process is accompanied by a drastic development in these parameters. 2) The texture parameters of the OX6.5(C) and OX6.5 (MW) samples show that an OX with slightly more developed total surface area and lower microporosity is obtained by the MW induced synthesis method. The OX6.5(MW) sample has predominantly non-microporous character. The non-microporous surface area (S_n^α) is more than 64.0 % of the total surface area (S_t^α) for this sample. This percentage decreases sharply from 64.0 to ~ 17.8 % by carbonization at 750 °C. 3) The texture parameters of the CX6.5(C)750 and CX6.5(MW)750 samples reflect that a CX with more developed porosity (17.8 %) is obtained by the MW induced synthesis method rather than by the conventional synthesis one (4.2 %). 4) The texture parameters of the CX6.5(C)750 sample reflect that the conventional synthesis method produces essentially microporous carbon with wide microporosity (95.8 %) and containing some mesopores (4.2 %). On the other hand, the parameters of the CX6.5(MW)750 sample indicate that the MW induced synthesis method produces, also, microporous carbon (82.2 %) but with slight shift to mesoporosity (17.8 %).

Adsorption of Methylene Blue:-

Fig. (9 (a)) shows the adsorption isotherms of MB on the CX samples synthesized at different R-F pH values and fixed CT of 750°C in nitrogen atmosphere. The graphs are plotted in the form of the amount of MB adsorbed (X) per unit mass of carbon, against the concentration of MB remaining in solution, C_{eq} . The adsorption isotherms show type-H according to the Giles classification (Giles et al., 1960) . They show very steep behavior with various degrees of sharpness or roundness at the low concentration values, tending to a plateau parallel to the C_{eq} -axis. Type-H indicates high affinity isotherm with strong preferential adsorption of the solute (Giles et al., 1960). The results of the Langmuir and Freundlich plots show satisfactory straight lines, which indicate that both of these

adsorption models are applicable. The Langmuir plots are represented in Fig. (10 (a)), whereas the Freundlich plots are illustrated in Fig. (11 (a)).

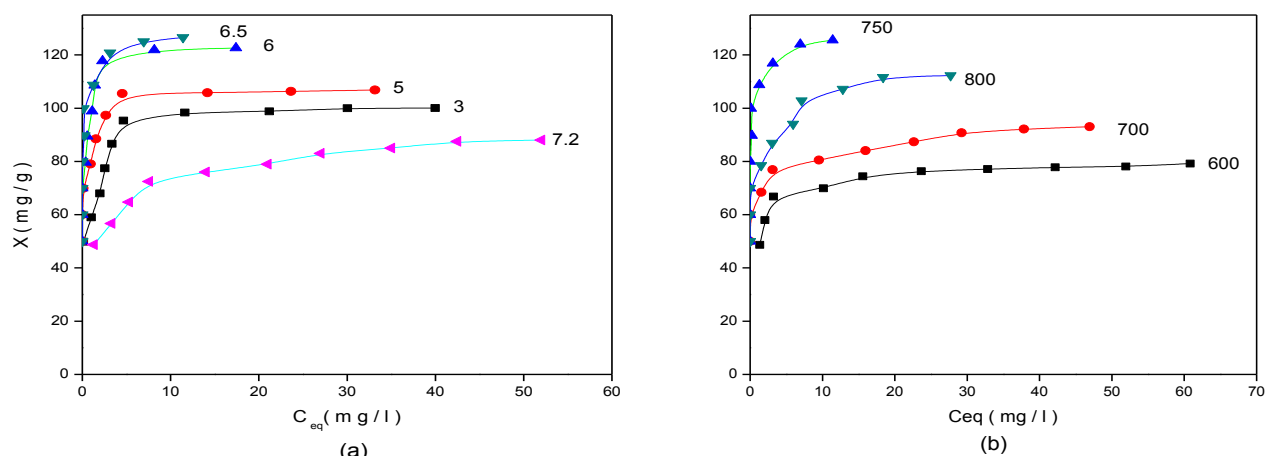


Fig. 9:- The adsorption isotherms of MB on the CXs obtained under different conditions: (a) different R-F solutions pH values and (b) different CTs.

Table (4) summarizes the adsorption parameters (X_m , K_L , n_F and K_F) derived from both adsorption models. The following can be observed: i) Application of the Langmuir equation is more satisfactory as evident from their higher correlation coefficients (R^2) compared to those of the Freundlich equation. ii) The dye adsorption capacity increases with the increase of the R-F solution pH value up to pH=6.5 then declines. The adsorption capacity increases continuously from 102 to 128 mg. g⁻¹ with increasing the initial pH value from 3.0 to 6.5, respectively. This can be ascribed to the increased adsorption space on the CX surface, due to the increase in the surface area and number of pores accessible to MB molecules with raising the R-F solution pH value up to pH=6.5. The sample prepared at pH= 7.2 has a lower surface area, lower pore volume, and, as a result, lower uptake of MB.

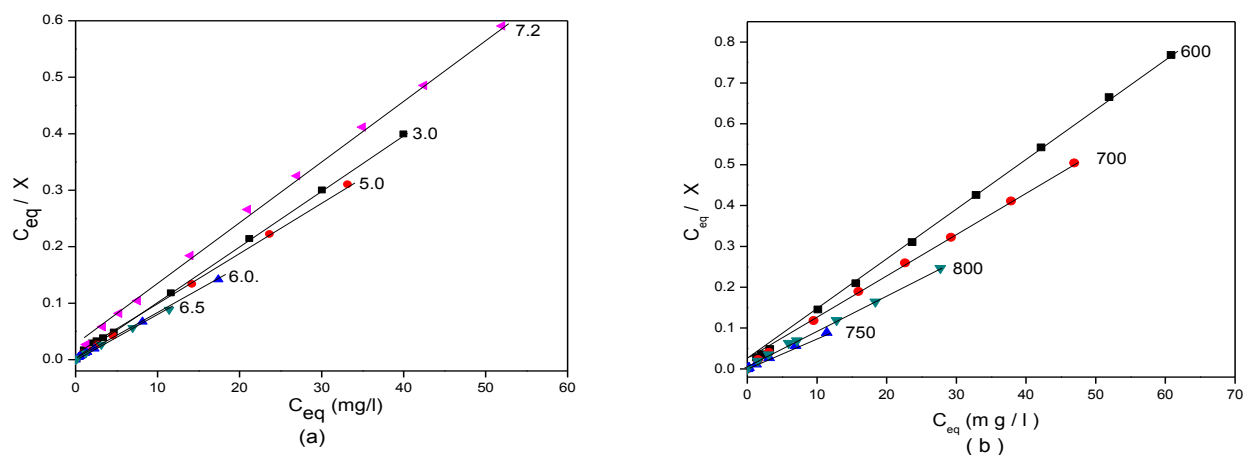


Fig. 10:- Langmuir plots of MB by the CXs obtained under different conditions: (a) different R-F solutions pH values, and (b) different CTs.

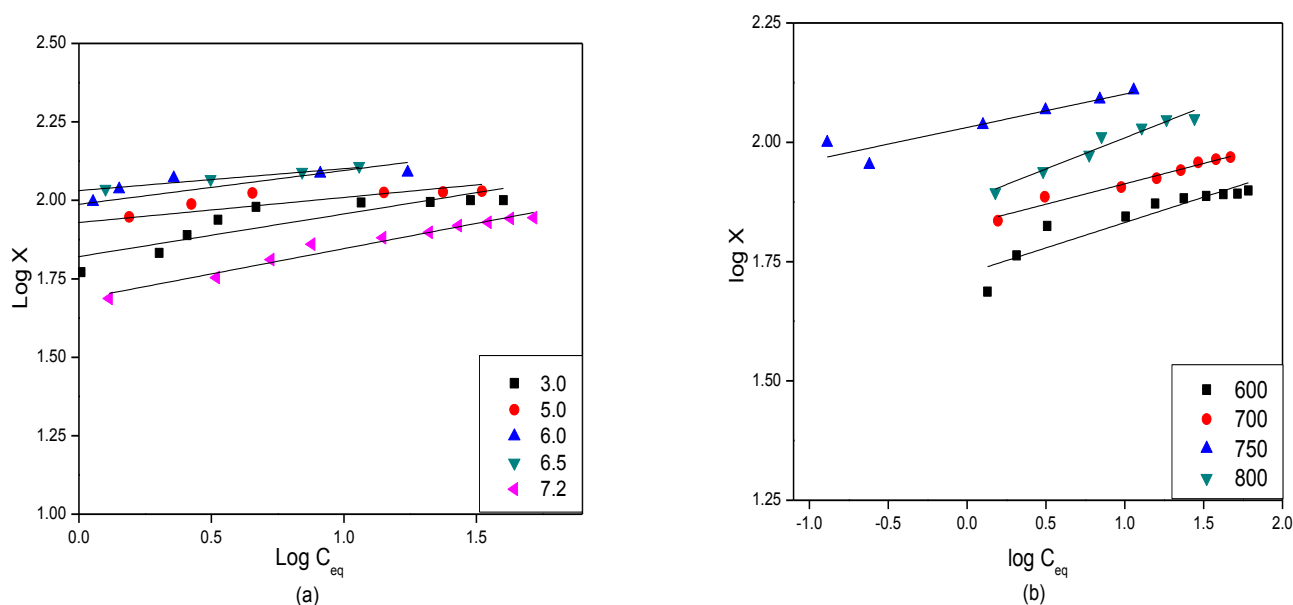


Fig. 11:- Freundlich plots of MB by the CXs obtained under different conditions: (a) different R-F solutions pH values, and (b) different CTs.

Table 4:- Equilibrium values of MB adsorption, surface area accessible to M.B and ratio to nitrogen surface area for the investigated CXs.

Sample notation	Langmuir-plots			Freundlich-plots			S _{MB} (m ² .g ⁻¹)	S ^α _{N₂}	S _{MB} / S ^α _{N₂}
	X _m (mg.g ⁻¹)	K _L (l.mg ⁻¹)	R ²	K _F (mg.g ⁻¹)	n _F	R ²			
Effect of pH									
CX3.0(MW)750	102	1.751	0.9996	70.790	7.407	0.8619	397	566	0.702
CX5.0(MW)750	108	6.643	0.9999	84.950	12.515	0.8660	421	590	0.713
CX6.0(MW)750	123	8.345	0.9996	97.140	9.286	0.7830	483	604	0.799
CX6.5(MW)750	128	12.59	0.9987	107.424	14.308	0.8445	499	701	0.713
CX7.2(MW)750	91	0.473	0.9987	48.421	6.210	0.9641	205	511	0.401
Effect of CT									
CX6.5(MW)600	80	1.0024	0.9998	53.156	9.406	0.8441	313	311	1.007
CX6.5(MW)700	93	1.3827	0.9983	67.234	11.713	0.9649	365	489	0.906
CX6.5(MW)750	128	12.59	0.9987	107.424	14.308	0.8445	499	701	0.713
CX7.2(MW)800	113	2.135	0.9976	75.664	7.666	0.9462	443	614	0.721

The adsorption capacity of CXs depends not only on their textural characteristics, but also on their surface chemistry, i.e. the nature of the CX surface, c.f. Table (5) and Fig. (12 (a, b)). The pH of the dye solutions was not adjusted, so that the pH of the CX surface becomes an important determining factor. The pH determines the surface charge of the carbon material, and therefore the electrostatic interaction (attractive or repulsive) between the dye molecules and the adsorbent (Moreno-Castilla and Rivera-Utrilla, 2001). The maximum adsorptivity belongs to the CX6.5 sample. At lower pH values, excessive protonation of the carbon surface occurs which enhances the electrostatic repulsion between the CX surface and MB cations (positively charged), resulting in lower adsorption capacity. At a higher pH value, although the surface of the adsorbent is negatively charged which could favor the adsorption of positively charged MB, the low porosity and surface area decrease this effect. In this case the net synergistic effect of texture properties and surface chemistry of CX7.2 is to reduce the adsorptivity towards MB.

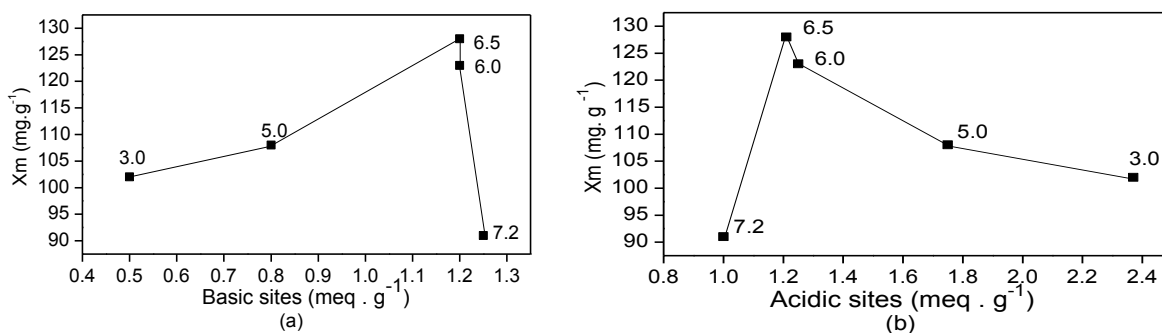


Fig. 12:- Adsorption amounts X_m of MB vs. surface basic (a) and acidic (b) sites obtained from Boehm titration for the CXs obtained at different R-F solutions pH values.

Table 5:- Adsorption amounts X_m ($\mu\text{mol/m}^2$) of MB, total acidic and basic sites (meq/m^2) obtained from Boehm titration for the investigated carbons

Sample notation	X_m (mg.g^{-1})	Total acidic sites (meq.g^{-1})	Basic sites (meq.g^{-1})
Effect of pH			
CX3.0(MW)750	102	2.37	0.5
CX5.0(MW)750	108	1.75	0.8
CX6.0(MW)750	123	1.25	1.20
CX6.5(MW)750	128	1.21	1.20
CX7.2(MW)750	91	1.00	1.25
Effect of CT			
CX6.5(MW)600	80	1.30	0.60
CX6.5(MW)700	93	1.27	1.00
CX6.5(MW)750	128	1.21	1.20
CX7.2(MW)800	113	0.85	1.22

Taking the evaluated X_m and the value of cross-sectional area covered by an adsorbed MB molecule as 120 \AA^2 (Giles, 1969), the surface areas accessible to MB (S_{MB}) were determined. The estimated S_{MB} values are given in Table (4) along with the (S_{MB} / S_t^a) ratios. A comparative representation of the data for total surface area (S_t^a) obtained from N_2 sorption isotherms and surface area accessible to methylene blue (S_{MB}) for the CX samples are given in Fig. (13 (a)). The CX6.5 sample has higher S_{MB} value ($S_{MB} = 499 \text{ m}^2.\text{g}^{-1}$) compared to that for the CX6.0 sample ($S_{MB} = 483 \text{ m}^2.\text{g}^{-1}$), which can be correlated to the less surface acidic nature of this sample (acidic sites = $1.21 \times 10^{-3} \text{ (meq.g}^{-1})$) compared to that of the CX6.0 sample (acidic sites = $1.25 \times 10^{-3} \text{ (meq.g}^{-1})$) [c.f. Table (5)]. However the $S_{MB} / S_{N_2}^a$ value for the CX6.5 sample is lower than that for the CX6.0 sample (i.e. 0.713 and 0.799, respectively) which can be attributed to the development of micro pores inaccessible to MB in the CX6.5 sample. The ability of these CXs for adsorption of MB is reduced by surface acidic groups and enhanced by surface basic sites.

Fig.(9 (b)) shows the adsorption isotherms of MB on the CXs prepared at different CTs in N_2 atmosphere and fixed R-F pH value of 6.5. The isotherms show also type-H according to the Giles classification (Giles et al., 1960). The results of the Langmuir and Freundlich plots show satisfactory straight lines. The Langmuir plots are represented in Fig. (10 (b)), whereas the Freundlich plots are illustrated in Fig.(11 (b)). The Langmuir parameters (X_m and K_L) and Freundlich constants (K_F and n_F) are given also in Table (4). It can be observed that the dye adsorption capacity increases continuously with the increase of the carbonization temperature up to 750°C then declines, the highest value is observed for the CX750 sample. A comparative representation of the data for the CXs obtained at different CTs are given in Table (4) along with the (S_{MB} / S_t^a) ratios, and are represented graphically in Fig. (13 (b)). Table (4) reflects a gradual decrease in the MB accessible surface area ratio (i.e. S_{MB} / S_t^a) with the increase of the CT up to

750°C. The effect of raising the CT was found to increase the porosity up to 750 °C, as indicated previously from the calculated values of S_t^a , S_n^a , and S_{mic}^a , c.f. Fig.(8 (b)). Also, the non-microporous surface area is 82.3 % of the total surface area for the CX600 sample. It decreases sharply to 1.8 % by raising the CT up to 800 °C (i.e. the CX800 sample), [c.f. Table (3)]. It should be remembered here that MB, due to its bulky size, measures only the surface area within pores of diameter $\geq 15 \text{ \AA}$ (Girgis and El-Hendawy, 2002).

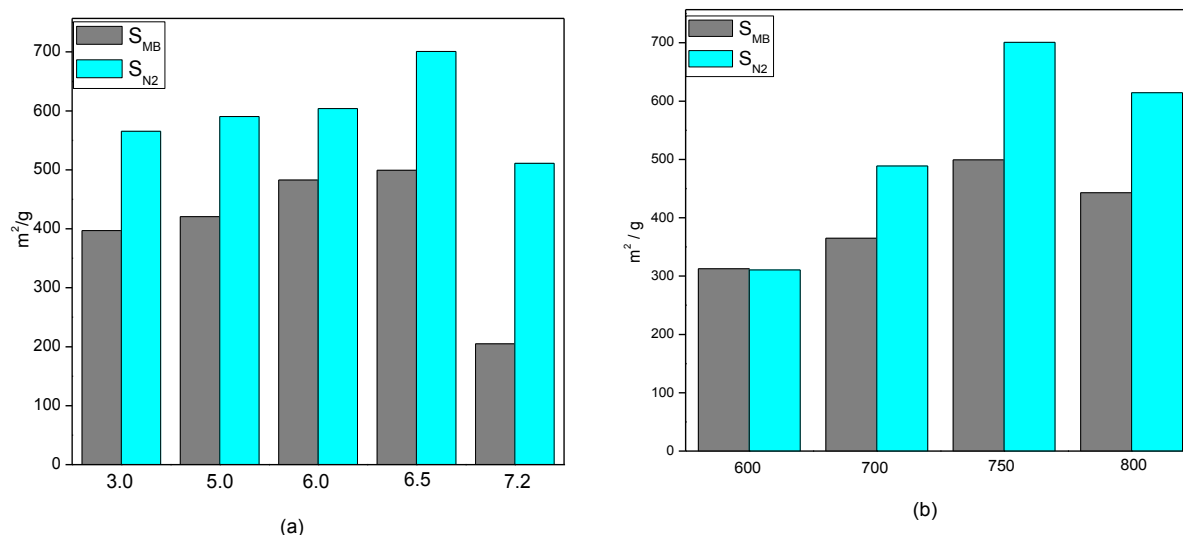


Fig. 13:- A comparative representation of the data for total surface area (S_t^a) obtained from N_2 sorption isotherms and surface area accessible to methylene blue (S_{MB}) for the CXs obtained under different conditions: (a) different R-F solutions pH values, and (b) different CTs.

The results obtained from Fig. (14) are in agreement with those observed in Fig. (12), i.e. the ability of these CXs for adsorption of MB is reduced by surface acidic groups and enhanced by surface basic ones.

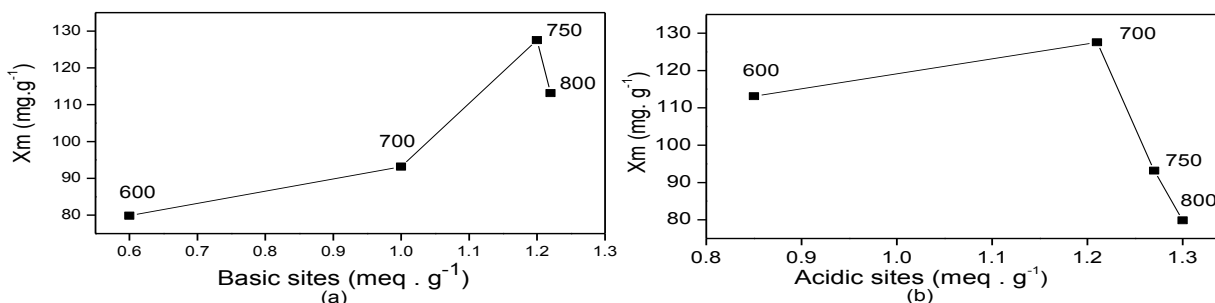


Fig. 14:- Adsorption amounts X_m of MB vs. surface basic (a) and acidic (b) sites obtained from Boehm titration for the CXs obtained at different CTs.

Conclusions:-

1. The MW induced synthesis method of CXs confirmed the following: a) By varying synthesis conditions such as initial solution pH and calcinations temperature, it is possible to obtain nanostructured porous carbons with different physicochemical properties. The chemical nature of the CXs surfaces depends strongly on the synthesis conditions. b) The texture characteristics obtained from the BET-equation and α_s - plots indicate developed carbon microporosity with some mesoporosity, which depends on the preparation conditions. c) The removal capacity of MB is governed by the chemical structure of the CX surface (i.e. the acidic and basic nature) as well as the diffusion through the intricate internal porosity. d) The best conditions for producing CXs by the MW induced synthesis method is at initial solution pH value of 6.5 and CT of 750°C. These conditions

favor higher yield and carbon content, more surface function groups, higher surface area, wider pore volume and more developed mesoporosity.

2. The effect of the synthesis method confirmed that the MW assisted method has a lot of advantages over the conventional method. The main advantage is the saving of time, resulting in a reduction in the energy consumed, also the prepared CXs have higher surface area, more developed porosity, higher carbon content and more yield than those obtained by the conventional synthesis method.

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RESEARCH ARTICLE

THE UNIVERSE IS IN GOOD HANDS.

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Abstract

On 13 June 1936, Paul Kazuo Kuroda noticed confusion about terms used in Aston's lecture on nuclear energy that would later isolate humanity from reality (God). In 1945, Kuroda secretly retained a personal copy of Japan's design for atomic bombs to finesse post-WWII standard scientific models that obscured Aston's 1922 finding of "powers beyond the dreams of scientific fiction" in rest masses (m) of atoms. Precise nuclear and space age measurements later revealed neutron repulsion as the source of energy (E) in cores of heavy elements like uranium, planets like Jupiter, the Sun, and galaxies that maintain the dynamic, cyclic, now-expanding universe through reversible transitions between two forms of one fundamental particle: Neutrons (compacted electron-proton pairs) and hydrogen (expanded electron-proton pairs), with nuclear charge densities of $Z/A = 0$ and $Z/A = 1$ and reversibly, different rest masses, are the alpha and omega of the cosmos and other all atomic matter with nuclear charge densities of $0 < Z/A < 1.0$. Weizsäcker had mistakenly assumed in 1935 that differences in rest masses of free neutrons and hydrogen atoms remained unchanged when these combine to produce atoms of heavier elements, and thus obtained misleading values of "nuclear binding energy" that obscured neutron repulsion as an important force in the origin and evolution of the cosmos and life in the solar system.

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Introduction:-

Kuroda was born on 1 April 1917 and died on 16 April 2001 (Manuel 2001a,b). Kuroda noticed on 13 June 1936, and reported a serious misunderstanding of nuclear energy in his autobiography (Kuroda, 1992, page 6). BBC News (2002) reported that Kuroda had secretly retained a personal copy of Japan's design for atomic bombs, since "just before Japan's surrender in 1945."

The author does not know, but will explain why he believes, Kuroda retained the copy to show others a misunderstanding of Aston's (1922) "packing fraction" might block public acceptance of his conclusion "the beginning of the world may have been just like" Hiroshima's destruction on 6 Aug 1945 (Kuroda, 1982, page 2). Many discoveries subsequently made on the origin and early history of the solar system in fact pointed to the powerful nuclear force in the Sun that was obscured by this 1936 misunderstanding of nuclear energy (Manuel 2016a).

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The author and five students finally discovered neutron repulsion in atomic rest mass data, aka Aston's "nuclear packing fractions," (Manuel et al., 2000) and reported this as the source of solar energy at the 32nd Lunar & Planetary Science Conference (Manuel et al., 2001). A copy of the "Cradle of the Nuclides" from the LPSC abstract was sent to Kuroda, beneath the title of this paper as an encrypted message of hope, indelibly recorded in the rest masses of atoms.

This paper celebrates the centennial of Kuroda's birth on 1 April 1917 and his contributions, as a nuclear, geo-, cosmo-chemist at the University of Tokyo (1936-1949) and later at the University of Arkansas (1952-2001), to dating the geo-chronology of hot springs (e.g., Kuroda, et al., 1954), "fall-out" particles from atmospheric nuclear weapons testing (e.g., Clark et al., 1967), and the solar system's origin and chronology at the "beginning of the world" (Kuroda, 1982, page 2).

Kuroda published about 400 scholarly research papers and supervised the research of ~70 PhD students and several postdoctoral research associates (W.A. Myers, University of Arkansas, personal communication, 2002). Two of his papers were cited as Benchmark Papers in Nuclear Chemistry (Seaborg and Loveland, 1982). Rowe (1988), Braun (2013) and Manuel (2016a) summarized Kuroda's major research contributions, and the author nominated Kuroda for posthumous recognition (Manuel, 2017a). The title and first figure of this paper are from the author's last message to Kuroda in 2001.

An outline of this paper was written to confirm recent reports (Manuel 2016a, 2016b, 2016c) of "An Error in Calculated Nuclear Energy" (Manuel, 2017b), posted for public comments, corrected, expanded and retitled, "The Universe Is In Good Hands," aka "There Is A God," to convey a more appropriate message for the centennial celebration of Kuroda's birth - a message of hope for today's distressed society.

Through the throes, violence and racial nationalism in Japan during WWII, and in the United States after WWII, Kuroda developed a magnificent obsession for truth as the ultimate salvation for his own life and for society as a whole (Kuroda, 1982, 1992). Kuroda's obsession was contagiously passed to his students, who learned that rigorous adherence to basic principles of the scientific method and their own creativity and logic could also provide them meaningful lives of continuous discovery. Since Kuroda encouraged students to think for themselves, the author is therefore solely responsible for any errors in the conclusions of this paper. Kuroda did not endorse them, although many are based on Kuroda's scholarly research publications, especially those cited above and in his book, *The Origin of the Chemical Elements and the Oklo Phenomenon* (Kuroda, 1982), in his autobiography, *My Early Days at the Imperial University of Tokyo* (Kuroda, 1992), and in his summary paper with Myers published in *Proceedings of the American Chemical Society symposium on Origin of Elements in the Solar System*, "Xenology, FUN Anomalies and the Plutonium-244 Story" (Kuroda and Myers, 2000).

The conclusions of this paper are simply the author's own interpretation of Kuroda's actions and published reports, in view of his status as an immigrant from a former enemy nation that secretly retained a personal copy of Japan's successful design for atomic bombs (BBC News, 2002).

1. Kuroda noticed on 13 June 1936 and reported publicly in his autobiography (Kuroda, 1992, p.7) that someone "who appeared to be a physicist in his 30's" did not grasp the meaning of Aston's "packing fraction." The goal of this paper and Kuroda's nomination for posthumous recognition (Manuel, 2017a,b) is public awareness of the misunderstanding of nuclear energy that Kuroda noticed in 1936 (Kuroda, 1992, p. 7) and its long-term impact on modern science and public perception of our place in the universe.
2. While standing in the ruins of Hiroshima one day in August 1945, Kuroda was "overwhelmed by the power of nuclear energy. The sight before my eyes was just like the end of the world, but I also felt that the beginning of the world may have been just like this" (Kuroda, 1982, p. 2).
3. As a research mentor, Kuroda demonstrated and taught his students to appreciate the scientific method as the design for a meaningful life of continuous discovery.
4. In a series of papers with W.A. Myers in the 1990s, Kuroda dated the geo-chronology of the early solar system's formation from a supernova explosion, like fallout particles from a nuclear weapons explosion, using a combination of ^{26}Al , ^{238}U and ^{244}Pu age dating to show that refractory silicon carbide (SiC) and graphite/diamond (C) grains in primitive meteorites formed ~1-10 Ma after a supernova explosion (Kuroda and Myers, 1996) made the solar system's actinide elements ~5.1 Ga ago (Kuroda and Myers, 1994).

In 1961 the author independently noticed the error in Weizsäcker's concept of "nuclear binding energy" (Weizsäcker, 1935) and reported the error at the 1966 Annual Meeting of the American Physical Society (Manuel, 1966). Kuroda did not identify the name of the physicist who misunderstood Aston's talk on "nuclear packing fractions" at the Imperial University of Tokyo on 13 June 1936, except by approximate age. Three possible candidates, whose research achievements also reflected a misunderstanding of Aston's (1922) "nuclear packing fractions," are:

- Hideka Yukawa (born 1907) received the 1949 Nobel Prize in Physics for a model of the nucleus that overlooks neutron repulsion (Yukawa, 1949);
- Hans Bethe (born 1906) received the 1967 Nobel Prize in Physics for a solar model based on hydrogen-fusion, rather than neutron repulsion, as the primary source of energy production in stars (Bethe, 1967).
- Carl von Weizsäcker (born 1912) received the 1957 Max Planck Medal for extra-ordinary achievements in theoretical physics. Noting that modern science "has given us the power of Greek gods" (Drieschner, 2014, page 173), Weizsäcker established the Carl Friedrich von Weizsäcker Society in 1994 to promote "wissen und verant-wirtung," aka "knowledge and responsibility," in the areas of "Physics, philosophy, theology, economics and altered awareness" (Drieschner, 2014, page 173-174).

Publication of Weizsacker's (July 1935) neutron-proton model of the nucleus may have influenced Chadwick's (Dec 1935) Nobel Lecture, or visa versa. Chadwick (1935) assumed the validity of the "scheme of quantum mechanics" as reason to alter his earlier conclusion the neutron consists of "a proton and an electron in close combination" (Chadwick, 1932, pages 697 and 708), as had been suggested earlier in Rutherford's Bakerian Lecture (Rutherford, 1920).

Scientists more qualified than the author to evaluate quantum mechanics (Weinberg, 2017, Weinberg et al, 2017) continue to express doubts about the validity of quantum mechanics, but there is no reasonable doubt calculated values of "nuclear binding energy" (Weizsacker, 1935) consistently disagree with observed values of beta-decay by 0.782 MeV (Manuel, 1966) - the exact rest mass difference between the free neutron and the hydrogen atom - and obscure the powerful force of neutron repulsion in ordinary atoms (Manuel et al., 2000), in the Sun (Manuel et al., 2001), in changing Earth's climate (Manuel, et al., 2002), and in the cosmos (Manuel, 2011, 2012, 2016a,b,c, 2017b, and reference cited therein).

Adherence to theoretical models of reality, over experimental measurements of reality, may also explain current unrest over government-funded science in Washington, DC, with the US NAS supporting ScienceDebate.org (2017), a private 501c3 organization calling for a public "March for Science" in Washington, DC next month in response to President Trump concerns about the validity of scientific evidence for human-induced global warming (AGW).

Publication of this paper – as first outlined and posted for public comment as "An error in calculated nuclear energy" (Manuel, 2017b) – will offer "March for Science" promoters an opportunity to consider first evidence the error in calculated nuclear energy (Manuel, 2017b) has altered awareness of evidence the Sun made our elements (Manuel and Sabu, 1975; Sabu and Manuel, 1976a; Manuel et al., 1977; Toth, 1977; Manuel, 1978; Manuel et al., 1979; Sabu and Manuel, 1980; Manuel and Hwaung, 1983), birthed the solar system in a supernova explosion ~5.1 Ga ago (Kuroda and Myers, 1994), controls Earth's climate (Manuel et al., 2002), now holds Earth in continuous harmonic vibration with the Sun (Thomson et al., 2007) and has sustained the origin and evolution of life on Earth for ~4 Ga ago (Michaelian and Manuel, 2011).

The author first met Kuroda in May 1960, two years after Kuroda returned to the University of Arkansas from a personally challenging and professionally unsuccessful year (1957-1958) at Argonne National Laboratory studying radioactive fallout from a series of "nuclear weapons tests being recklessly conducted in the South Pacific" (Kuroda, 1996, p. 64) - a study that "has remained unpublished in scientific journals up to the present time" (Kuroda, 1996, p. 65).

Four years earlier, Kuroda's suggestion of natural, self-sustaining nuclear fission reactors in the early Earth (Kuroda, 1956) had been criticized as impossible at the 1956 National Meeting of the American Geophysical Union (AGU). The author was nevertheless convinced of Kuroda's genius and intellectual integrity and gratefully accepted Kuroda's PhD research assignment on the "Origin of the solar system and its elements" after discussing these recent findings with him:

- Reynolds' (1960a) discovery that the Richardton meteorite contained excess radiogenic ^{129}Xe from in situ decay

- of extinct, 17-Ma year ^{129}I , synthesized just before the meteorite formed;
- Reynolds' (1960b) report that the overall abundance pattern of all nine stable isotopes of primordial xenon in meteorites is distinct from that in air; and
- Kuroda's (1960) suggestion that spontaneous fission of extinct 82-Ma year ^{244}Pu , made just before the Earth formed, generated excess heavy xenon isotopes (^{131}Xe - ^{136}Xe) in air.

These 1960 discoveries were all later confirmed, the last in 1972 by observing an anomalous mix of xenon isotopes (xenon-X) released from primitive carbonaceous meteorites at $\sim 800^\circ\text{C}$ (Manuel et al., 1972) that would cause the overall isotopic abundance pattern of primordial xenon in meteorites to be distinct from that in air (Reynolds, 1960b).

Xenon-X had properties expected in fresh supernova debris, with "the greatest enrichment in the lighter isotopes, $^{124,126}\text{Xe}$, and the heavier isotopes, $^{134,136}\text{Xe}$, relative to ^{130}Xe . These are the isotopes expected to be produced in supernova explosions," (Burbidge et al., 1957) "the $^{124,126}\text{Xe}$ isotopes by the p-process and the $^{134,136}\text{Xe}$ by the r-process" (Manuel et al., 1972, page 100). It was therefore concluded that "no known nuclear or physical process is capable of producing both anomalies in situ" (Manuel et al., 1972, p. 101), within the meteorite itself.

Scientists at the University of Chicago separated mineral fractions of the Allende carbonaceous meteorite, better isolating the fraction containing "strange xenon" (Lewis et al., 1975), or xenon-X (Manuel et al., 1972), and concluded in situ fission of an unknown super-heavy element made "strange xenon" ("xenon-X") inside the meteorite itself.

However the University of Chicago analysis revealed that essentially all primordial helium and neon in the meteorite was trapped in the mineral fraction that trapped "strange xenon," whereas mineral fractions of the Allende meteorite that trapped "normal xenon" had little or no primordial helium or neon (Manuel et al., 1975).

This observation of primordial He and Ne – trapped only in mineral fractions of the primitive Allende meteorite that trapped "strange xenon" (aka, "xenon-X"), and not in the mineral fractions that trapped xenon of "normal" isotopic composition (Lewis et al., 1975) – matched the prediction by Burbidge et al. (1957) that fusion reactions would have depleted light elements from the interior of a star that evolved to the terminal supernova stage, when r- and p-processes of explosive nucleosynthesis might produce "strange xenon," aka "xenon-X" in outer layers of the exploding star - where light elements like hydrogen, helium, carbon and neon remained.

Being convinced that linked, elemental and isotopic anomalies in the Allende meteorite (Lewis et al., 1975) were nucleogenetic, D.D. Sabu and the author wrote and submitted three papers saying a single, local supernova explosion at the birth of the solar system made strange "xenon-X" (Manuel et al., 1972), primordial helium and neon, ^{244}Pu and perhaps some other isotopic anomalies and decay products of short-lived radioactive isotopes that had been observed in meteorites (Sabu and Manuel, 1976a, Sabu and Manuel, 1976b, Manuel and Sabu, 1975) – before going to the 1976 Annual Spring Meeting of the American Geophysical Union to present the first paper.

A protracted discussion of events that occurred there and in other open discussions of the supernova beginning of the solar system in 1976-77 would only distract from the central purpose of this paper and its outline (Manuel, 2017b):

1. Identification of the logical error in Weizsäcker's (1935) definition of "nuclear binding energy" that isolated humanity from reality after WWII (Manuel, 2017b);
2. Celebration of the centennial of Kuroda's birth on 1 April 1917; and
3. Gratitude that we were anyway able to confirm Aston's report of "powers beyond the dreams of scientific fiction" in atoms (Aston, 1922, page 20): Neutron repulsion - indelibly recorded in rest masses of atoms (Manuel et al., 2000, 2001, 2016a,b,c) – powers the Sun, the cosmos and offers humanity a rational scientific explanation for the simple, logical sequence of events that:
 - Made our elements and birthed the solar system in a supernova explosion (Sabu and Manuel, 1976a,b; Manuel and Sabu, 1975; Manuel et al., 1977; Ballard et al., 1979).
 - Condensed highly refractory "fallout" grains of silicon carbide (SiC) and graphite- diamond (C) within the first 1-10 Ma (Kuroda and Myers, 1996) after the supernova exploded, ~ 5.1 Ga ago (Kuroda and Myers, 1994).
 - Accreted terrestrial planets in layers from heterogeneous supernova debris, beginning with formation of iron

cores from Fe-rich debris near the pulsar (Manuel and Sabu 1981).

- Separated d- and l-forms of amino acids (that would later become precursors to life) in primitive carbonaceous chondrites with CP-circularly polarized light from the pulsar (Cronin and Pizzarello, 1997), before a
- Gravitationally trapped photosphere of waste products, mostly H and He (Manuel, 2012), accumulated over the pulsar and moderated the energy of solar radiation to allow the origin and evolution of life to begin on Earth, ~4 Ga ago, when intense UV radiation from the evolving Sun were first absorbed by primitive molecules of life, like RNA or DNA (Michaelian and Manuel, 2011).
- Sustained the evolution of life into humans, with incredible talents, ~79 year life spans, and inalienable rights to self-governance on the water-covered planet orbiting only 1 AU (one astronomical unit) from the supernova pulsar remnant that birthed the solar system.

Results and Discussion:-

Before considering the figures posted below, readers are encouraged to **read carefully** and **consider thoughtfully** two figures in the outline (Manuel, 2017b) showing

- The consistent error, and
- The way it was obscured

In values of nuclear binding energy Weizsacker (1935) calculated for the neutron-proton model of a nucleus composed of two, always distinguishable forms of nucleons: Neutrons and Protons.

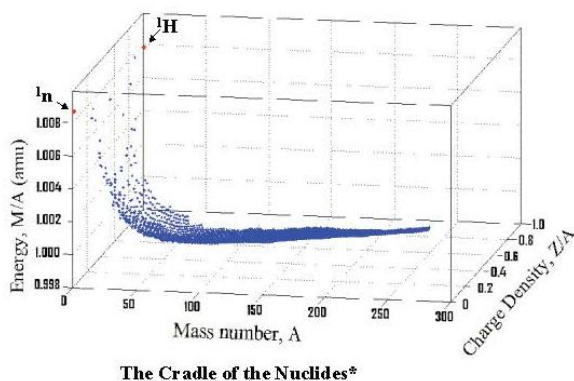
The consistent discrepancy between real nuclear stability observed in the ordinary β -decay of isobars – as measured by Aston’s “nuclear packing fractions” (Aston, 1922) and always calculated incorrectly, by exactly 0.782 MeV (the difference in rest masses of the neutron and the hydrogen atom), in Weizsacker’s values of “nuclear binding energies” – shows neutrons and hydrogen atoms simply become two, reversibly interchangeable forms of one fundamental particle when combined to make heavier atoms with mass number, $A > 1$ (Manuel, 2011).

Widespread adoption of Weizsacker’s (1935) flawed model of the nucleus after WWII blocked serious consideration of Kuroda’s realization, standing in the ruins of Hiroshima in August 1945, “the beginning of the world may have been just like this” nuclear explosion (Kuroda, 1982, p. 2).

Kuroda probably retained a personal copy of Japan’s atomic bomb design after WWII (BBC News, 2002) to show others that the same source of energy that destroyed Hiroshima made our elements, birthed the solar system and began a measurable geochronology for the formation of the solar system, its elements and the beginning of the world (e.g., Kuroda, 1982; Kuroda, 1992; Kuroda and Myers, 1994; Kuroda and Myers, 1996; Manuel et al., 2000; Manuel et al., 2001).

Figure 1 shows the message the author sent shortly before Kuroda died in April 2001, a message of hope, indelibly recorded in rest masses of the ~2,850 distinct types of atoms that comprised all known matter in 2000 (Manuel et al., 2000). The author had just returned from the 32nd Lunar and Planetary Science Conference in March (Manuel et al., 2001) where the “Cradle of the Nuclides” was presented to show that neutron repulsion powered the giant nuclear explosion that triggered the formation of the solar system and sustained the current operation of the Sun:

“The Universe Is In Good Hands”



Each dot represents one of the 2,850 nuclides listed in *Nuclear Wallet Cards*, 6th edition (2000) National Nuclear Data Center, Brookhaven National Laboratory, 74 pp.

Figure 1:- The above message of hope for humanity was sent to Kuroda in April 2001. This “Cradle of the Nuclides” had been published in research papers. It was published on the front cover and inserted in the front flyer pages of the Proceedings of the 1999 ACS Symposium on the Origin of Elements in the Solar System as it was going to press (Manuel, 2000). Combined U/Pb, Pu/Xe and Al/Mg age dating of primitive meteorites and their refractory inclusions by Kuroda and Myers (1994, 1996) showed that a supernova exploded shortly before the oldest meteorites formed, about five billion years (~5.1 Ga or ~5,100 My) ago. Five students enrolled in a graduate course, Special Topics in Nuclear Chemistry, at the University of Missouri-Rolla in the spring semester of 2000 helped the author use computer-assisted 3-D images of atomic rest masses to identify neutron repulsion there (Manuel et al, 2000)

Figure 2 shows how the discovery and confirmation of decay products from 82 My ^{244}Pu in the Earth, in meteorites and in man-made ^{244}Pu (Kuroda, 1960; Rowe and Kuroda, 1965; Alexander et al., 1971) was used to determine and illustrate the geo-chronology and early history of the solar system’s formation.

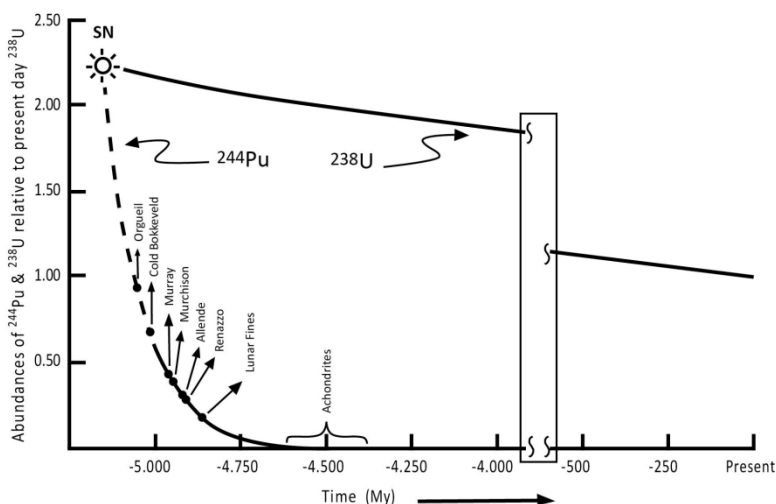


Figure 2:- Combined U/Pb and Pu/Xe age dating of primitive meteorites, dated back from the present by Kuroda and Myers (1994), show the geochronology of primitive meteorites in the early solar system after a supernova explosion birthed the solar system, shortly before the oldest meteorites formed about five billion years (~5.1 Ga or ~5,100 My) ago.

Figure 3 shows how Kuroda and Myers (1996) used the discovery of excess ^{26}Mg from the decay of extinct ^{26}Al in the Allende meteorite (Gray and Compston, 1974) to demonstrate the geo-chronology of formation of silicon carbide and graphite-diamond refractory grains in primitive meteorites with the physical properties of single fallout particles after nuclear detonation (Clark et al., 1967).

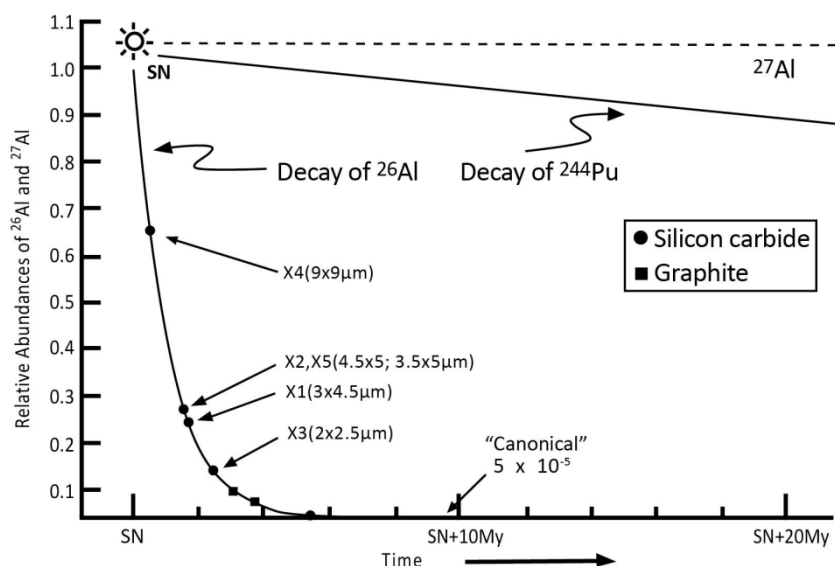


Figure 3:- $^{26}\text{Al}/^{26}\text{Mg}$ age dating of refractory meteorite inclusions of silicon carbide and graphite (diamond) show their formation within the first 1-10 My after the supernova, with physical properties like those of "fall-out" particles from nuclear explosions (Kuroda and Myers, 1996). Grains that formed first grew larger and trapped higher levels of radioactive ^{26}Al ($t_{1/2} = 740,000$ yr or 0.74 My), like the fractionation of fission products in single fallout particles after a nuclear detonation (Clark et al, 1967).

We cannot see the Sun's core, but it may be indistinguishable from this Chandra x-ray image of the pulsar formed in 1054 AD by the supernova explosion that produced the Crab Nebula.

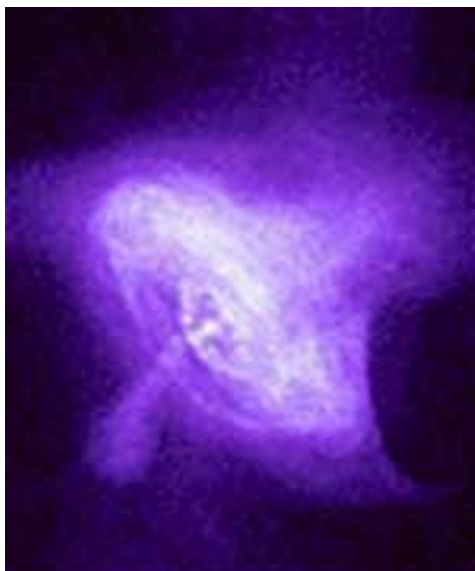


Fig. 4:- The creator, destroyer and sustainer of every atom, life and planet in the solar system is a pulsar like the one above that birthed the Crab Nebula in 1054 AD. Toth (1977) proposed a solar pulsar, Kotov (1996) measured and confirmed the 160 minute solar pulse Toth (1977) had noted earlier as the forth of regular, 40-minute pulses, Manuel et al. (2002) noted the implications for solar eruptions and Earth's climate, Thomson et al. (2007) demonstrated the effects of solar vib-rations on engineering and scientific systems, interplanetary magnetic fields,

atmospheric pressure, induced voltages on ocean cables, and terrestrial seismic data, and Karoff et al. (2016), Clery (2016), Persson (2016) note abrupt solar eruptions may periodically reset civilization.

Conclusion and Acknowledgements:-

The author is honored to write this manuscript to celebrate the centennial of the birth of Paul Kazuo Kuroda on April 1, 1917. Kuroda risked his life and the wrath of united world governments after WWII so humanity might know the simple sequence of geo-, cosmo- chrono-logical events that produced life from highly radioactive debris of a supernova that exploded here five billion years (~5.1 Ga) ago, sustained the origin and evolution of life into humans, and endowed them with remarkable talents and inalienable rights to self-governance on a water-covered planet orbiting ~1 AU (one astronomical unit) from the pulsar remnant that was, and still is, powered mostly by neutron repulsion, the mysterious source of energy Aston discovered and reported as “powers beyond the dreams of scientific fiction” in “nuclear packing fractions” of ordinary atoms (Aston, 1922).

This paper is meant to be suggestive only. We are at the beginning of a new awakening to reality and know only a little. More will be revealed if we selflessly practice the basic principles of science for the benefit of humanity, as exemplified by the life and career of Paul Kazuo Kuroda.

Numerous friends, anonymous scientists and bloggers encouraged publication of this summary of the heroic efforts of the late Professor Paul Kazuo Kuroda (1917-2001) to prevent the misuse of nuclear secrets to isolate humanity from reality, including two well-known geo-ethicists, Drs. Nils-Axel Möerner and VáclavNěmec .

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RESEARCH ARTICLE

RATIO ANALYSIS OF TEXTILE INDUSTRY IN TAMIL NADU: (WITH THE SPECIAL REFERENCE TO CMIE LISTED COMPANY)

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Abstract

The financial performance of the textile industry in Tamil Nadu had been analyzed with the help of the financial ratios. The financial structure of a company can show its capacity to generate the funds needed to undertake the desired expansion. The financial performance assessment together with other efficiency criteria, will give an idea of the total efficiency and industry performance. The success of the company ultimately depends upon its future growth and development. The company's future can never be predicted with accuracy without having precise information related to its present financial position and its past earnings. In addition, the present study analyzed the impact of the performance of liquidity, solvency and efficiency on the profitability of the textile industry using CMIE data. The study found the impact of financial ratios, such as return on capital employed and net profit ratio, on the profitability of textile industry in Tamil Nadu, was meticulously studied. From the Generalized Least Square method it was found that absolute liquidity ratio had the highest impact among the financial ratios on the return on net profit ratio as well as creditors' turnover ratio is highest impact on return on capital employed during the study period with statistical significance.

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Introduction:-

It presents a detailed analysis of the determinants of financial performance of textile industry in Tamil Nadu using financial ratios. The financial structure of a company can show its capacity to generate the funds needed to undertake the desired expansion. The financial performance assessment together with other efficiency criteria, will give an idea of the total efficiency and industry performance. The success of the company ultimately depends upon its future growth and development. The company's future can never be predicted with accuracy without having precise information related to its present financial position and its past earnings. Ratio Analysis is an age-old technique of financial analysis. The financial position of the textile industry in Tamil Nadu is analyzed with the help of the financial facts and ratios such as: liquidity, solvency, efficiency and profitability. In addition, the present study analyzed the impact of the performance of liquidity, solvency and efficiency on the profitability of the textile industry. Liquidity and profitability are the two desired goals of financial management and they are directly affected by the working capital management. With increase in working capital size beyond the adequacy level, liquidity improves and profitability declines, and vice versa. **Ratio Analysis:** This is an age-old technique of financial analysis. It provides the financial statements in an absolute, historical and static form. It is also designed to show

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how one number is related to another and the meaning of such relationships. A ratio is worked out by dividing one number by another number.

Objectives of the Study:-

- To study the financial performance of the textile industry in Tamil Nadu
- To find out the impact of financial ratios (liquidity, solvency and activity) on the textile industry of profitability ratio

Hypothesis of the Study:-

H1 There is a significant impact of Liquidity, Solvency and Activity ratios on ROCE

H1 There is a significant impact of Liquidity, Solvency and Activity ratio's on the Net Profit Ratio

Research Methodology:-

The study sought to measure the financial performance of textile companies, registered under CMIE list. These industries are spread over the state of Tamil Nadu in India. 234 companies were listed under CMIE and 18 companies were listed under NSE/BSE as well as the availability of data for the last ten years (2005 to 2014), which was chosen for the study.

Review of Literature:-

The SINET report (2007) has briefly discussed about the Indian cotton textile industry and its contribution to our country's export, economy, production and employment. It has also analysed the industry's financial indicators of return on capital employed (ROCE), operating and net margins and also earning stability, etc. Anupkumar&Subhash (2011) measured the level of technical efficiency of firms in the Indian textile industry and identified the factors that account for efficiency variation across firms, using the data envelopment analysis (DEA) in non-parametric methodology. Kunal and Lokinder (2009) sought to understand the impact of liberalisation on the Indian textile industry, by comparing the performance of firms incorporated before and after the liberalisation period. Kataria (1996) analysed the financial position of some selected units of the cotton textile industry of Malwa region to judge their profitability and financial strength. Noel, John and Scott (1990) determinants of industry, firm and business financial performance can be used as measures of individual relationships in models linking various hypothesized causal variables of various performance indicators. Mansur (2003) assessed the financial performance of textile industry, using ratio analysis, to determine the financial and operational efficiency. Ram and Mayank (2002) extend the prior work done on the relation of Indian firm characteristics and their financial performance. Alovsat&Abdulmecit (2001) focused on the profitability margin of the export-oriented textile industry of Turkey in the post crisis period. Manasranjan, et. al, (2005) examined the causal relationship between the three variables, the study used granger causality and co-integration test for the period of 1970-2004. Waqas, et.al, (2013) analyzed the factors affecting firm performance in the textile and food sector of Pakistan, using panel/longitudinal data of companies (non-financial) listed in KSE for the years 2005 to 2010.

Analysis and Interpretations:-

Liquidity Ratios: -

Liquidity ratios measure the capacity of the business to meet its short term financial commitments as and when these become due. Investors regularly take a close look at liquidity ratios when performing fundamental analysis on a company. Since a company consistently having trouble meeting its short-term debt, it is at a higher risk of bankruptcy, liquidity ratios are a good measure of whether the firm will be able to comfortably continue as a going concern (Walker, 2011: Ukessays, 2013: Saravanan&Abarina, 2014).

Current Ratio:-

Current Ratio expresses the precise relation between current assets and current liabilities. It indicates the availability of current assets in rupees for every one rupee of current liabilities. A high current ratio indicates high liquidity; while a low current ratio indicates low liquidity. It is sometimes difficult to decide what should be the satisfactory current ratio of any company. Generally, the manufacturing industry ratio of 2:1 is traditionally considered as a benchmark of adequate liquidity, and, in India, as 1.33:1 (Prasanna Chandra, 2004). The current ratio is a very important tool to the outsiders, as well as to the management. It is a measure of the firm's ability to meet its short-term liabilities to the outsiders. From the management side, this ratio discloses the magnitude of the current assets that the firm carries, in relation to its current liabilities. For an outsider, the larger the ratio, the higher is the liquidity

of the company. A very high ratio means that the firm is having idle assets and, hence, there could be some inefficiency in management of its short term funds. Nevertheless, the current ratio is a quick measure of the company's liquidity, as it tests only the quantity - and not the quality. The constraint of this ratio indicator lies due to size, type of the investor and the quality of the receivables of the enterprise.

Table-1 reveals that the average current ratio of the sample companies varied between 0.90:1 and 4.76:1 during the study period. The current ratio was 1.47:1 in 2005, which increased to 1.66:1 in 2006 and then decreased to 1.42:1 in 2008. Thereafter, it marginally declined to 1.39:1 in 2009. Thereafter, it gradually increased to 1.80:1 and reached the level of 1.42:1 in 2014. The analysis shows that the average ratio was lower than the standard, which indicates that sample companies had maintained a sufficient level of liquidity during the study period since the current ratio was above the benchmark of 1.33:1. This indicates that most of the sample companies have maintained the current ratio, as compared to the benchmark of 1.33:1

An in-depth analysis of Table-1 reveals that the companies VTML, BASL and SCL had a very high current ratio. In contrast, companies SSL, KSML and KGDL had a very low current ratio. SSL, KSML & KGDL are likely to have difficulties in meeting their short term obligations, because most of their current assets consist of inventory. VTML, BASL and SCL are likely to meet their current obligations as and when these become due, because a large portion of their current assets consists of cash and accounts receivables. Normally, accounts receivable are highly liquid and can be converted into cash quickly. The standard deviation is universally used to measure the confidence level in statistical conclusions. Usually, the standard deviation close to zero indicates that the data points tend to be very close to the average/mean of the set, although a high standard deviation indicates that the data points are spread over a wider range of values. In this study, when the standard deviation was high, it indicated that the firm has more deviation from the mean/average. It explains that firms (KGDL, KSML SSML and SSL) had not maintained consistent current assets to meet their obligations.

Quick Ratio:-

Table-2 shows the quick ratios of the sample companies. Quick ratio indicates the direct liquidity of current assets that are easily convertible into cash. Recognising that inventory might not be very liquid, this ratio takes into account the quickly realisable assets and measures these against the current liabilities. This is a more refined and conservative estimate of the company's liquidity than the current ratio, since it establishes a relation between liquid assets and current liabilities. Conventionally, a quick ratio of 1:1 is considered to be a more satisfactory measure of the liquidity position of a concern. While this ratio does not entirely supplement the current ratio and, when used in conjunction with it, tends to give a better picture of the company's ability to meet its claims out of the quick assets.

It is evident from Table-2 that the overall average of the quick ratio was 0.62:1 during the period under study. The quick ratio showed a downward trend throughout the period of ten years. It was 0.58:1 in 2005, which gradually decreased and reached lowest level of 0.70:1 in 2013. It is evident from the Table that the overall quick ratio of the sample companies, taken together, was more than unity. The trend clearly reveals that the sample companies had not improved their overall liquidity position over a period of time. In this study, when the standard deviation was found to be high, it indicated that the firm has more deviation from the mean/average. It explains that firms, VTML and NEPCTL, had not maintained consistent liquid assets to repay their creditors. The coefficient of variation of the sample companies was 35.08%, which shows a lesser variation among the units, indicating that they had followed a uniform policy for quick ratio during the period under study. The coefficient of correlation of the quick assets and current liabilities was +0.99. This reveals that there had been a perfect positive correlation between quick assets and current liabilities. This leads to the conclusion that an increase in current liabilities can trigger an increase in quick assets in the same proportion. An in-depth analysis of Table-2 reveals that companies, VTML and NEPCTL, had very high quick ratios, while companies, ACML and KSML, had very low quick ratios.

Absolute Liquidity Ratio: Cash to Current Liabilities:-

This ratio is also known as cash position ratio. Though current ratio and acid-test ratio are important tools to measure the liquidity position of the company of a going concern, this ratio is appropriate to measure the absolute liquidity of the concern. As it indicates the availability of cash to meet the current obligations immediately. If the firm begins with a shortage of absolute cash in meeting its current obligations and if this trend mounts up to heavy burden on the finances of the company, this may even cause cash insolvency of the business. Early detection of this kind of situation by the management is a sine-qua-non for the continuity of the business. Table-3 exhibits the ratio of cash to current liabilities.

Among the companies, the proportion of cash to current liabilities reveals that this was in the range of 0.96% to 0.02%. The average of all the sample companies was 0.12% in 2005, which decreased to 0.11% in 2008 and then marginally increased to 0.20% in 2010. Thereafter, it decreased to 0.06% in 2011 and reached 0.10% in 2014. Most of the companies were not in a position to meet their current liabilities from their cash balances. The study reveals that, during the study period, on an average, the sample companies had 0.16% of cash against their current liabilities. The acceptable specific norm for this ratio is 0.25:1 or 1:4, i.e., Rs.1 worth of cash is considered adequate to pay Rs. 4 worth of current liabilities in time, as all the creditors are not expected to demand at the same time. The company's cash may also be realised from receivables and inventories. As the selected companies had almost equivalent amounts to the specific norm, it indicates their sound cash position. The overall average of total cash to total current liabilities of the selected companies was 0.16% during the study period. The coefficient of variation of the selected companies was very high, i.e., 73.35%. It clearly indicates that they had not followed a uniform policy of maintaining cash during the period under study. In this study, when the standard deviation was high (VTXIL & BASL), it indicated that the firm has more deviation from the mean/average. It explains that firms were not maintaining a sufficient liquid assets position to meet their quick obligations. A deeper analysis of the Table reveals that the companies, VTML, BASL and STL, had exceptionally high cash balance, whereas companies, RML and SSL, had an exceptionally low cash balance of current liabilities. In addition, absolute liquid assets, such as marketable securities and cash in bank, were found to be high, whereas the current liabilities, such as bank overdraft, sundry creditors, bills payable and creditors for outstanding expenses, were found to be low. High ratio reflects that absolute liquid assets worth one half of the value of current liabilities was sufficient for satisfactory liquidity position of the companies.

Solvency Ratios:-

Solvency ratios throw light on the long-term solvency of a firm, while the liquidity ratios, on the short-term solvency. These measure the long term financial viability of a business and its ability to pay off its long-term obligations such as bank loans and bonds payable. It is critical for banks, government, employees, institutional investors, bond holders, owners, etc. (Obaidullah, 2011). Solvency ratios include: debt-to-equity ratio, fixed charge coverage ratio, debt-to capital ratio, times interest earned ratio, and debt-to assets ratio.

Debt-Equity Ratio:-

Table-4 shows the Debt-Equity ratio, the ratio of total liabilities of a business to its shareholders' equity. It is a leverage ratio and measures the degree to which the assets of the business are financed by the debts and the shareholders' equity in a business. Lower values of the debt-equity ratio are favourable and indicate less risk. A higher debt-equity ratio is unfavourable, because it means that the business relies more on external factors. Thus, it is at higher risk, especially at higher interest rates. A debt-equity ratio of 1.00:1 means that half of the assets of a business are financed by debts and the rest by the shareholders' equity. A value higher than 1.00 means that more assets are financed by debt than by the money of shareholders.

It is evident from Table-4 that the overall average of debt-equity ratio was 2.42:1 during the period under study. The debt-equity ratio showed an upward trend throughout the period of ten years. It was 0.12:1 in 2005, which gradually increased and reached a peak level of 0.25:1 in 2006. It is evident from the Table that the overall debt-equity ratio of the sample companies, taken together, was more than unity, suggesting thereby that the companies were investing more funds from outside, compared with shareholders' funds. If it is more than the ideal ratio, it should meet more obligations from outside with high risk. However, less than the ideal ratio means that the company is utilising own funds with its structure, by avoiding the risk.

The standard deviation close to zero indicates that this data points tend to be very close to the average/mean of the companies, although a high standard deviation indicates that the data points are spread out line over a wider range of values. In this study, when the standard deviation was high (CFL & VTML), it indicated that the firms had more deviation from the mean/average. It explains that the firms were borrowing more funds from the outsiders, rather than utilising their own assets effectively. An in-depth analysis of the Table reveals that companies, SSIL, KPRML and VTML, had a very high debt-equity ratio, while companies, LMCL and GTL, had a very low debt-equity ratio.

Interest Coverage Ratio:-

This ratio measures the debt servicing of a firm, in so far as fixed interest on long term loan is concerned. It is determined by dividing the Net Profit before Interest and Tax by the fixed interest charges. It is also known as "time-interest-earned ratio," or debt service ratio, or net income to debt service ratio, or coverage ratio, or fixed

charges over. The lower the interest coverage ratio, the higher would be the company's debt burden, and greater the possibility of bankruptcy, or default. A lower ICR means less earnings are available to meet interest payments and that the business is more vulnerable to an increase in interest rates. When a company's interest coverage ratio is only 1.5, or lower, its ability to meet interest expenses may be questionable. An interest coverage ratio below 1.0:1 indicates that the business is having difficulties in generating the cash necessary to pay its interest obligations, i.e., the interest payments exceed its earnings (EBIT).

It is evident from Table-5 that the overall average of the interest coverage ratio was 2.08 during the period under study. Interest coverage ratio showed an upward trend throughout the period of ten years. It was 2.83 in 2005, which gradually increased to 3.14:1 in 2006. Thereafter, it decreased to 1.56:1 in 2008, and went up to 2.59 in 2011. It reached 1.77 in 2014. There was an impact of the global meltdown on textile companies during 2008 to 2009. The interest coverage ratio indicates the capacity of an organisation to meet its interest obligations. An interest cover ratio of 2 implies that the entity has sufficient profitability to bear twice the amount of its current financial costs. On the whole, the standard deviation close to zero indicates that this data points tend to be very close to the average/mean of the companies, although a high standard deviation indicates that the data points are spread over a wider range of values. In this study, when the standard deviation was high (as in the case of VTXL & CFL), it indicated that the firms had more deviation from mean/average. It explains that the firms were not capitalising on the relatively cheaper source of finance (debt). Also, in such instances, an increase in gearing ratio may actually add value to the company. The company expected the least financial risk (low debt financing) and high interest coverage ratio. It explains that the company preferred equity financing through venture capital institutions, rather than loan financing due to the high level of risk. An in-depth analysis of the Table reveals that the VTML and KPRML companies had a very high ICR, while LMCL and GTL companies had a very low ICR.

Any company tries to control its borrowings, to avoid more interest repayment. Due to the impact of a weak ratio, a company may have to face difficulties in raising funds for its operations. Companies operating in plants that are exposed to a high level of business risk and uncertainty would generally prefer to maintain lower levels of financial risk (lower debt financing) and higher interest coverage ratios. Many start-up companies prefer equity financing through venture capital institutions, rather than loan financing, due to the high level of risk involved and such companies would tend to have very high interest ratios. Some industries tend to have higher interest coverage ratios than others, and cyclical companies, in particular, can experience significant swings in their interest coverage ratios (especially during downturns). Thus, comparison of interest coverage ratios is generally most meaningful among companies within the same industry, and the definition of a "high" or "low" ratio should be made within this context.

Activity Ratios:-

Activity ratios are concerned with measuring the efficiency in asset management. It measures the efficiency with which a business utilises its assets, such as accounts receivable, inventories, and fixed assets. The common operating ratios are: the average collection period ratio, the inventory turnover ratio, days of inventory ratio, the fixed asset turnover ratio, and the total asset turnover ratio. The success or failure of the concern too depends much upon proper and judicious use of the resources. This ratio may be defined as a test of the relationship between sales and the various assets of a firm.

Debtors' Turnover Ratio:-

The debtors' turnover ratio indicates the efficiency achieved by using the funds invested in debtors. A high debtors' turnover ratio indicates quick collections and enables the firm to transact a larger volume of business, without an increase in the investment of receivables. As per Spiller and Gosman, the analysis of the receivables turnover ratio supplements the information regarding the liquidity of the receivables. Table-6 shows the debtors' turnover ratio during 2005 to 2014. The overall average turnover ratio of the sample companies registered a fluctuating trend throughout the period under study and was 12.03 times in 2005, which decreased to 9.26 times in 2006 and then showed an upward trend and fell to 11.67 times in 2010 and finally it marginally increased to 32.23 times in 2014. It is evident from Table-6 that the overall average of the debtor's turnover ratio was 18.28 times during the period under study. This shows that the selected companies had efficient management of receivables, as compared to the overall average ratios. The coefficient of variation of 72.71% of the sample companies indicates that they had followed a uniform policy with regard to debtors during the period under study. Out of the eighteen selected textile companies, eight of them were found to have a turnover rate higher than the overall average of 18.28 times, while 10 had a lower overall average debtor's turnover ratio. An in-depth analysis of the Table reveals that companies,

VTML and GTL, had a very high turnover ratio, while companies, VTXIL, KGDL and CTL, had a low turnover ratio.

Inventory Turnover Ratio:-

Inventory Turnover Ratio is one of the efficiency ratios and measures the number of times, on an average, that the inventory is sold and replaced during the fiscal year. It measures a company's efficiency in turning its inventory into sales. Its purpose is to measure the liquidity of the inventory. Inventory Turnover Ratio is figured as "turnover times". Average inventory should be used for inventory level, to minimise the effect of seasonality. Table-7 shows the inventory turnover ratio during 2005 to 2014. The overall average turnover ratio of the sample companies registered a fluctuating trend throughout the period under study and was 4.46 times in 2005; which increased to 4.54 times in 2009, and then showed a downward trend and fell to 3.71 times in 2011. Finally, it marginally increased to 5.29 times in 2014.

It is evident from Table-7 that the overall average Inventory Turnover Ratio was 4.39 times during the period under study. It reflects that the selected companies had an efficient inventory management system, as compared to the overall average ratios. The coefficient of variation of the sample companies is 34.17%, which indicates that they had followed a uniform policy with regard to inventory during the period under study. Out of the eighteen selected textile companies, seven companies had a turnover rate higher than the overall average of 4.39 times, while 11 had a lower overall average inventory turnover ratio. The high variation of coefficient of ACML and BSSL reflects that these companies had not followed the inventories accounting methods properly, which are indicated by: "last in first out (LIFO)" method, which shows higher costs of goods sold and lower inventories than companies using "first in first out (FIFO)" Method. In addition, high variation shows that special promotions, new product introductions can suddenly and somewhat artificially change the company's inventory ratio. An in-depth analysis of the Table reveals that NEPCTIL and KSML companies had a very high turnover ratio, while ACML, VTXIL and STL companies had a low turnover ratio.

Creditors' Turnover Ratio:-

This ratio is calculated by taking the total purchases made and dividing it by the average accounts payable during the period. It is used to measure the rate at which a firm pays off its suppliers. It's also known as an Accounts Payable Turnover Ratio, or Creditors' Velocity. Accounts payables turnover trends can help a company to assess its cash situation. Just as accounts receivable ratios can be used to judge a company's incoming cash situation, this figure can demonstrate how a business handles its outgoing payments. The higher ratio should indicate that the payments are made promptly. Table-5.8 shows the creditors' turnover ratios during 2005 to 2014. The overall average turnover ratio of the sample companies registered a fluctuating trend throughout the period under study and was 20.10 times in 2005, which increased to 25.14 times in 2007 and then showed a downward trend and fell to 13.44 times in 2009 and finally marginally increased to 39.78 times in 2014. It is evident from Table-8 that the overall average of the creditor's turnover ratio was 25.57 times during the period under study. The average payment period was less than the standard. It shows that the payments are made earlier. This may be due to better liquid resources and working capital. The coefficient of variation of 71.11% of the sample companies indicates that they had followed a uniform policy with regard to the creditors' collections during the period under study. A high ratio (prompt payment) is desirable, but a company should always avail the credit facility allowed by the suppliers.

Out of the eighteen selected textile companies, six were found to have a creditors' turnover ratio higher than the overall average creditors turnover ratio of 19.80, while 12 had a lower than the overall average creditors' turnover ratio. The high standard deviation of CFL, GTL and KGDL shows that data sets of the companies were not close to the mean/average. It explains that suppliers and creditors of the companies were not paying the bills in the right time period. It also suggests that new vendors were being paid back very slow. An in-depth analysis of the table reveals that ACML and NEPCTL companies had a very high creditors' turnover ratio, while VTXIL, SCL and VTML companies had low creditors' turnover ratios.

Profitability Ratios:-

Profits are the measure of the overall efficiency of a business. Profitability is an indication of the efficiency with which the operations of the business are carried on. Poor operational performance indicates poor sales and poor profit. The higher the profits, the more efficient the business is. A lower profitability may arise due to lack of control over the expenses. It must be remembered that profit is an absolute measure of the earning capacity, and profitability is the relative measure of the earning capacity. Important profitability ratios are: return on capital employed ratio,

gross profit margin ratio, net profit margin ratio, operating profit margin ratio, return on assets ratio and return on equity ratio. It highlights how effectively the profitability of a company is being managed.

Return on Capital Employed:-

Return on capital employed is also known as rate of return. It establishes the relationship between profits and the capital employed. It also reveals the earning capacity of the capital invested in the business. It is widely used to measure the overall profitability and efficiency of the business concern. The main objective of making investments in any business is to obtain a satisfactory return on capital invested in the business. It measures the profitability of a company, by expressing its operating profit, as a percentage of its capital employed. Capital employed is the sum of the stockholders' equity and long-term finance. Alternatively, the capital employed can be calculated as the difference between total assets and current liabilities. This ratio is useful to show the efficiency of the business as a whole.

Table-9 shows the return on capital employed during the period under study and also depicts that the overall average return on capital employed was 12.74%. It was 15.92% in 2005, which increased to 16.44% in 2006. Thereafter, for the next three consecutive years, it showed a declining trend, and fell to 3.55% in 2009, but increased to 16.98% in 2014. The ROCE greater than unity indicates, that the higher efficiency of the sample companies in the utilisation of assets to generate sales. The coefficient of variation of 98.88% of the samples companies shows higher variation among the company ratios', which indicates that these companies had followed different policies for the return on capital employed during the period under study. It reveals that there was a lesser perfect positive correlation between profits and capital employed. This leads to the conclusion that increases in capital employed led to an increase in proportion of profits of the companies. An in-depth analysis of the table reveals that companies SSIL and KPRML had a very high return, while companies GTL and CFL had a very low return on capital employed.

Return on Equity:-

This ratio reveals the relationship between profits of a company and its equity shareholders' funds. Net profit after interest, tax and preference dividend is divided by the equity shareholders' funds. It is a measure of the profitability of the stockholders' investments. It shows the net income as a percentage of shareholder equity. Net income is considered for the full fiscal year, after taxes and preferred stock dividends, but before common stock dividends. Shareholders' Equity does not include preferred stocks. Return on Equity varies substantially across different industries. Therefore, it is recommended to compare returns on equity against company's previous values or return of a similar company. The ratio of return on equity of the sample companies moved in a very wide range - varying from -96.60% to 77.09% during the period under study. This ratio was 15.26 in 2005, which increased to 19.60% in 2006. It then maintained an increasing trend and reached 16.61% in 2014. The companies show the negative - 11.92% in 2009 due to the impact of global meltdown and recovered in the year 2011 and reached a peak level of 21.09. The average percentage of return on equity was 9.07% during the period under study. This shows the conservative policy adopted by the managements of the sample companies.

It is evident from Table-10 that the overall average of the return on equity ratio was 9.07% during the period under study. It is assumed that the assets, without corresponding liabilities, are the direct creation of the shareholder capital that helps the company to grow in the first place. The coefficient of variation of 460.83% of the sample companies indicates that they had not followed a uniform policy with regard to equity returns during the period under study. The high standard deviation of CFL and NEPCTL shows that the data set points were not close to the mean and were away from the mean line. An in-depth analysis of the Table reveals that the companies GTL and SSIL had very high returns on equity ratio, while companies NEPCTL and CFL had low returns on equity ratio.

Operating Profit Ratio:-

The operating profit ratio measures whatever proportion of a company's revenue is left over, after deducting direct costs and overheads, before taxes and other indirect costs, such as interest. It measures a company's pricing strategy and operating efficiency. It gives an idea of how much a company makes (before interest and taxes) on each rupee of sales. The operating margin ratio shows whether the fixed costs are too high for the production or sales volume. The operating cost is equal to the cost of goods sold, plus the operating expenses. Non-operating expenses, such as interest charges and taxes, are excluded from the computations. The operating profit ratio of the sample companies moved in a narrow range varying from 7.95% to 22.34 during the period under study. This ratio was 17.51 in 2005, which reached the peak level of 22.34% in 2006. Thereafter, it declined for the three consecutive years to 7.95% in 2009 and increased to 18.12% in 2011 and reached 20.09% in 2014. The average operating profit ratio was 15.84%

during the study period. This shows the conservative policy adopted by the managements of the sample companies to use's sales revenue to cover the cost of goods sold and operating expenses.

It is evident from Table-11 that the overall average ratio of the operating profit was 15.84% during the period under study. The coefficient of variation of 87.21% of the sample companies indicates that they had not followed a uniform policy with regard to operating profit ratio. The high standard deviation of the companies, CFL and GTL, shows that the data sets of these companies were varying too high and not very close to mean/average. It explains that, the companies were not making enough money from their ongoing operations to pay for their variable and fixed costs. An in-depth analysis of the Table reveals that the companies, ACML and RML, had a very high ratio of the operating profit, while companies, CFL, GTL and CTL, had a low ratio of the operating profit.

Net Profit Ratio:-

This ratio reveals the overall profitability of the concern. Net profit ratio establishes the relationship between net profit (after taxes) and sales. It is expressed as a percentage of sales. It indicates the efficiency of the management in manufacturing, selling, administrative and other activities of the firm. This ratio is very useful to the proprietors, because it reveals the overall profitability of the business concern. It is also called "Net Profit to Net Sales ratio". This ratio indicates the result of overall operations of the firm. The net profit ratio of the sample companies moved in a wide range, varying from -7.27 to 9.45 during the period under study. This ratio was 5.52 in 2005, which increased to the high level of 9.45 in 2006. Thereafter, it decreased for the next three consecutive years to touch -7.27 in 2009, due to the impact of the global meltdown, and recovered within a year, to reach 4.51 in 2011. Once again, it fell to -2.74% in 2012 due to low market price for the raw materials and reached 5.12 in 2014. The average percentage of the net profit ratio was 2.83 during the period 2005 to 2014. This shows the conservative policy adopted by the managements of the sample companies.

It is evident from Table-12 that the overall average ratio of the net profit was 2.83 and the coefficient of variation of the sample companies was negatively placed at 124.84%. The high standard deviation of the companies reflects that the companies were not constantly improving their profitability. Also, they were not comparing the ratio with those of the previous years, the industry's average and the budgeted net profit ratio. An in-depth analysis of the table reveals that BASL and ACML companies had a very high ratio of the net profit, while companies, GTL and CFL, had a low ratio of the net profit.

Impact of Liquidity, Solvency and Activity ratios on Return on Capital Employed using Generalised Least Square Method:-

(OLS) techniques were used due to high correlation. Before going to use OLS, one should consider the impact of independent variables on the ROCE of the profitability ratio. In order to examine the impact of both independent and dependent variables of the textile companies, ordinary least squares tests the stationarity on the variables, if it is a time series data. The study applied the unit root tests as per the augmented Dickey Fuller and Phillips-Perron (PP, 1988) method to avoid spurious regression results. If the first and second difference does not satisfy the stationarity, it would reflect that multi-collinearity, or auto correlation.

$$Y = \alpha + \beta X_i + e.$$

It has been observed that the correlation is given the multi-collinearity, to avoid more than two variable of multi-collinearity, used in the generalised linear square method.

$$\text{Generalised Least Square } Y^* = \alpha + \beta X_i^* + U_i$$

Here, Y^* = Transformed Dependent Variable (Y/σ), X_i^* = Transformed Independent Variables (X_i/σ), σ = Standard Error

The generalised Linear Square method indicated that the profitability was high, as depicted by its independent variables. It is also evident from the value of R^2 that 0.967 percent of variation in ROCE was accounted for by the joint variation in the independent variables of financial performance. The Adjusted 'R' Square (R^2) signifies that 0.935 percent of the positive variation in the ROCE is explained by the independent variables. The high standard error of the regression coefficients demonstrates that there is really a line of estimates among the variables.

H1 There is a significant impact of Liquidity, Solvency and Activity ratios on ROCE

Generalized Least Square Statistics and Hypotheses Results

	Coefficients	Standard Error	t Stat	P-value	H
Intercept	1.88	20.09	0.09	0.93	
Current Ratio	96.85	46.23	2.09	0.07	Not Accepted
Quick Ratio	-41.52**	13.20	-2.52	0.03	Accepted
Absolute Liquidity Ratio	23.99	28.09	1.02	0.33	Not Accepted
Debt Equity Ratio	-0.68	5.49	-0.12	0.90	Not Accepted
Interest Coverage Ratio	5.12**	0.81	6.33	0.00	Accepted
Debtors Turnover Ratio	13.37	6.98	1.92	0.09	Not Accepted
Creditors Turnover Ratio	-152.42	72.88	-2.09	0.07	Not Accepted
Inventory Turnover Ratio	1.29	1.87	0.69	0.51	Not Accepted

** = statistically significant at 5% level

It can be observed from the above Table that current ratio is having the high positive impact on the return on capital employed and was statistically insignificant at 5% level. It reflects that the capital employed was the sum of stockholders' equity and long term finance. The capital employed is the difference between total assets and current liabilities. Here, the assets are impacting more on the return on capital employed. On the other hand, the estimated coefficient of quick ratio shows the negative impact on the capital employed, which is statistically significant. The estimates coefficients of absolute liquidity ratio were found to be having a high negative impact on the ROCE, which was statistically insignificant. DER and ICR of the coefficients shows the positive impact on the ROCE, which was statistically insignificant by DER and significant by ICR at the level of 5%. It reflects that debt into a company's total capital provides a more comprehensive evaluation, as well as management's usage of debt and equity disposal. DTR, CTR and ITR were found to be having a positive and negative impact on the ROCE, which was statistically insignificant. During the years 2005 and 2008 to 2010, the stock turnover and the average collection period had shown adverse trends, which indicate the deteriorating level of inventory management and stock collection policy. In the year 2012 all those ratios showed high and low fluctuations. The main reason for that was the availability of raw materials at low cost and the less duty on textile products by the government. An insignificant variation in financial ratios could be the result of the composite effect adopted in the analysis, as well as many other financial ratios-related unexplained variables. Hence, we can agree with the alternative hypothesis *there is no significant impact of Liquidity, Solvency and Activity ratios on ROCE of the textile industry in Tamil Nadu*.

Impact of Liquidity, Solvency and Activity ratios on Net Profit Ratio, using the Generalised Least Squares Method:-

The strength of the relationship between the dependent variable, net profit ratio (NPR) and all the independent variables are taken together to look into the impact of independent variables on the NPR of the profitability ratio. In order to examine the impact of both independent and dependent variables of textile companies, ordinary least squares (OLS) techniques were used, due to the high degree of correlation. Before going to use OLS, one should test the stationarity on the variables, if it is time series data. The study applied the unit root tests as per the augmented Dickey Fuller and Phillips-Perron (PP, 1988) method to avoid spurious regression results. The first and second differences were not satisfying the stationarity, which reflect the multi-collinearity, or auto correlation.

$$Y = \alpha + \beta X_i + e.$$

It is observed that the correlation given the multi-collinearity, to avoid more than two variable of multi-collinearity, used in the generalized linear square method.

$$\text{Generalised Least Square } Y^* = \alpha + \beta X_i^* + U_i$$

Here, Y^* = Transformed Dependent Variable (Y/σ), X_i^* = Transformed Independent Variables (X_i/σ), σ = Standard Error

The Generalised Least Square method indicates that the profitability was high impacted by its independent variables. It is also evident from the value of R^2 that 0.99 percent of variation in NPR was accounted for by the joint variation in independent variables of financial performance. The Adjusted 'R' square (R^2) signifies that 9.97 percent of the positive variation in the NPR is explained by the independent variables. The low standard error of the regression coefficients demonstrates that there is really a line of estimates among the variables. Note that the p-values for all the coefficients, with the exception of the coefficient for profitability, are lower than 0.05. This means that we

cannot reject the hypothesis that they are zero or below 0.05 (so it can be eliminated from the model). This is also confirmed from the fact that the 0 lies in the interval between the confidence level of the lower 95% and the upper 95% for each of these coefficients.

H1 There is a significant impact of Liquidity, Solvency and Activity ratio's on the Net Profit Ratio

Generalized Least Square Statistics and Hypotheses Result.

	Coefficients	Standard Error	t Stat	P-value	Hypotheses
Intercept	0.69	1.40	0.49	0.64	
Current Ratio	0.98	0.91	1.08	0.31	Not Accepted
Quick Ratio	11.71**	3.30	3.55	0.01	Accepted
Absolute Liquidity Ratio	-22.96	33.66	-0.68	0.51	Not Accepted
Debt Equity Ratio	-0.26	0.65	-0.40	0.70	Not Accepted
Interest Coverage Ratio	0.03	0.03	1.32	0.22	Not Accepted
Debtors Turnover Ratio	-0.98**	0.26	-3.71	0.00	Accepted
Creditors Turnover Ratio	22.48**	9.66	2.33	0.04	Accepted
Inventory Turnover Ratio	0.00	0.29	-0.01	0.99	Not Accepted

** = statistically significant at 5% level.

It was observed from the above Table that current ratio having the high positive impact on the net profit ratio was insignificant at 5% level. It replicates that an increase or decrease in current assets will impact the net profit ratio of the companies. The estimated coefficients of the QR and ALR were found to be having a positive and high negative impact on the net profit ratio of the textile companies, which was statistically significant at 5% level and insignificant by ALR. On the other hand, the estimated coefficient of ICR and CTR were found to be having a positive impact on the net profit ratio, which is statistically insignificant by ICR. The collection system was found to be faulty, because the debtors were enjoying a credit facility beyond the normal period. The performance of debt collection was found to be poor. It reflects that company were neglecting the increase of the credit position, as also the decreasing debt burden, while making repayment at high interest rates. Two other important indicators of efficiency, DTR and ITR, were having a low negative impact on the net profit ratio of the companies, which are statistically significant at 5% level by DTR and insignificant by ITR. Hence, we can agree with the alternative hypothesis *there is no significant impact of Liquidity, Solvency and Activity ratio's on Net Profit Ratio of the textile industry in Tamil Nadu.*

Findings, Suggestions and Conclusion:-

The financial performance of the textile industry in Tamil Nadu had been analysed with the help of the financial ratios. VTML Company performed better than the other companies throughout the period of the study in the areas of: current ratio, quick ratio and absolute liquidity ratio. As regards debt-equity ratio, CTL and RML Company scored better than the other companies. ACML Company was the best performer in the area of interest-coverage ratio. A higher debt-equity ratio was noticed in SSML. VTML Company better performed in less time to collect the receivables compared with other companies in the debtor's turnover ratio. The inventory turnover ratio of textile industry in Tamil Nadu fluctuated significantly during the period under study. NEPCTL also occupied the first place in inventory turnover ratio. The inventory management was better in KGDL. However, the age of inventory gradually declined during the period of the study. The high accounts payable turnover ratio is not always good enough in the interest of a company. LMCL Company performed better than the other companies during the study period in the area of medium creditor's turnover. The contribution made by shareholders to the capital structure was found to have gradually increased in textile industry of Tamil Nadu. ROCE is a more comprehensive test of profitability and SSIL Company performed with high return among the companies. At the same time, CFL Company's average return on equity fluctuated very badly during the period of the study. The higher operating profit ratio reveals the less net profit ratio. RML Company was found to have a high operating profit among the textile companies. However, BASL Company was earning the highest net profit among the select textile companies during the study period. The impact of financial ratios, such as return on capital employed and net profit ratio, on the profitability of textile industry in Tamil Nadu, was meticulously studied. From the Generalised Least Square method it was found that absolute liquidity ratio had the highest impact among the financial ratios on the return on net profit ratio as well as creditors' turnover ratio is highest impact on return on capital employed during the study period with statistical significance. During the course of the study, it was found that some companies were giving more attention to only liquidity and solvency aspects of the performance and taking more conservative decisions, leading to the

decline in their profitability. There is an urgent requirement to bring about changes in the approach of the management. Each company should give suitable weightage to the performance aspects of liquidity, solvency, and activity and profitability ratios. STL showed good performance in a very short span, as it was able to maintain its aggressive approach towards the working capital. Other companies also need to adopt more aggressiveness in maintaining their current ratio at 2:1 and improving their profitability. Both VTXIL and STL Company were found to be having very huge funds blocked in inventories and receivables. This could be due to the company's size of operation, and also to obviate issues like diseconomies of scale. The selected companies are advised to review their working capital frequently with tools like funds flow and cash flow statement and cash control reports. All the companies were found to have enough resources to meet their obligations over the next business cycle. At the same time, the current ratio values were varying from company to company, due to their respective demand and supply conditions and their own position in the market. However, it is suggested that all the sample companies should maintain the ratios at least equal to the benchmark level of 2:1. Meanwhile, the companies should avoid the idle assets, rather than keeping these as cash/liquidity in the business. Companies should maintain the ideal Absolute liquidity ratio in the business, which is productive and earning, and also taking care of liquidity. If the firm begins with a shortage of absolute cash in meeting its current obligations and if this trend imposes a heavy burden on the finances of the company, this may even cause cash insolvency of the business. Further it is suggested that companies should avoid the sine-qua-non for the continuity of the business. Overall debt-equity ratio of the sample companies, taken together, was more than unity, suggesting thereby that the companies were investing more funds from outside, compared with shareholders' funds. If it is more than the ideal ratio, it should meet more obligations from outside with a risk. However, less than the ideal ratio means that the company is utilizing its own funds within its structure, by avoiding the risk. Companies should maintain the average inventory level, instead of the ending the inventory level, to minimize the adverse impact of seasonal factors. Further, the companies should also solve the negative coefficients of absolute liquidity ratio, debtor's turnover ratio, as well as debt-equity ratio.

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Appendix:-

S. No	CMIE Listed Companies in Tamil Nadu	Code
1	Ambika Cotton Mills Limited	ACML
2	Bannari Amman Spinning Mills Limited	BASL
3	Celebrity Fashions Limited	CFL
4	Cheslind Textiles Limited	CTL
5	Gangotri Textiles Limited	GTL
6	K G Denim Limited	KGDL
7	K P R Mill Limited	KPRML
8	Kandagiri Spinning Mills Limited	KSML
9	Lakshmi Mills Company Limited	LMCL
10	N E P C Textiles Limited	NEPCTL
11	Rajapalayam Mills Limited	RML
12	SalonaCotspin Limited	SCL
13	Sambandam Spinning Mills Limited	SSML
14	Shiva Texyarn Limited	STL
15	Super Sales India Limited	SSIL
16	Super Spinning Mills Limited	SSL
17	V T M Limited	VTML
18	V T X Industries Limited	VTXIL
	CMIE = Centre for Monitoring and Indian Economy	

Annexure:-**Table 1:-** Current Ratio Of The Tamil Nadu Textile Companies During The Period 2005 To 2014.

CR	Mar -05	Mar -06	Mar -07	Mar -08	Mar -09	Mar -10	Mar -11	Mar -12	Mar -13	Mar -14	M in	Ma x	A vg	S D	CV
ACML	1.3	1.6	1.55	1.48	1.38	1.32	1.28	1.25	1.41	1.56	1.25	1.6	1.41	0.13	9.03
BASL	2.83	4.51	2.42	1.61	1.86	1.65	1.4	1.23	1.58	1.27	1.23	4.51	2.04	1.01	49.41
CFL	1.27	2.76	1.25	1.04	0.99	1.25	0.97	0.9	0.91	0.9	0.9	2.76	1.22	0.56	45.77
CTL	1.3	1.26	1.1	1.14	0.76	1.61	1.41	1.31	1.27	1.25	0.76	1.61	1.24	0.22	17.73
GTL	0.79	1.66	1	1.2	0.84	1.87	1.72	0.82	0.69	0.6	0.6	1.87	1.12	0.47	41.78
KGDL	1.11	1.37	1.25	1.14	1.03	1.02	1.02	1.02	0.97	0.9	0.9	1.37	1.08	0.14	12.88
KPRML	1.08	1.41	1.66	1.41	1.68	1.36	1.6	0.8	0.86	1.41	0.8	1.68	1.33	0.31	23.68
KSML	1.15	1.04	1.06	1.01	0.91	1.06	1.24	0.69	1.07	0.97	0.69	1.24	1.02	0.15	14.43
LMCL	0.79	0.73	0.81	1	0.83	3.02	2.76	2.41	2.44	2.18	0.73	3.02	1.70	0.94	55.40
NEPCTL	2.58	1.11	1.67	2.34	2.05	0.61	0.6	0.72	0.93	0.93	0.6	2.58	1.35	0.75	55.06
RML	1.48	1.68	1.68	1.57	0.96	1.19	1.38	1	1.06	0.95	0.95	1.68	1.30	0.30	22.99
SCL	2.01	1.7	1.59	1.47	1.59	1.46	1.56	1.79	2.3	1.72	1.46	2.3	1.72	0.26	15.21
SSML	1.09	1.18	1	1.39	0.95	1.12	1.21	0.83	1.17	1.05	0.83	1.39	1.10	0.15	14.09
STL	2.39	1.95	1.9	1.68	1.37	1.27	1.58	1.13	1.12	1.23	1.12	2.39	1.56	0.42	26.82
SSIL	1.17	1.29	1.39	1.38	1.53	1.64	1.23	0.81	0.95	1.12	0.81	1.64	1.25	0.25	20.22
SSL	1.1	1.47	1.02	1.1	0.81	0.82	0.95	0.48	0.55	0.68	0.48	1.47	0.90	0.30	32.86
VTML	2.11	2.17	3	1.96	4.18	4.32	3.73	7.81	12.01	6.33	1.96	12.01	4.76	3.17	66.62
VTXIL	0.93	1.04	1.73	1.67	1.28	1.3	1.35	1.17	1.19	0.51	0.51	1.73	1.22	0.35	28.90
Min	0.79	0.73	0.81	1.00	0.76	0.61	0.60	0.48	0.55	0.51					
Max	2.83	4.51	3.00	2.34	4.18	4.32	3.73	7.81	12.01	6.33					
MEAN	1.47	1.66	1.50	1.42	1.39	1.55	1.50	1.45	1.80	1.42			1.52		30.72
SD	0.63	0.85	0.55	0.35	0.80	0.86	0.71	1.65	2.59	1.29					
CV	0.43	0.51	0.36	0.25	0.58	0.55	0.47	1.13	1.44	0.91					

Table 5:-Quick Ratio Of The Tamil Nadu Textile Companies During The Period 2005 To 2014

QR	Mar -05	Mar -06	Mar -07	Mar -08	Mar -09	Mar -10	Mar -11	Mar -12	Mar -13	Mar -14	M in	M ax	Me an	S D	CV
ACM L	0.17	0.19	0.19	0.4	0.21	0.29	0.24	0.24	0.32	0.28	0. 17	0.4	0.2 5	0. 07	28. 01
BAS L	0.43	1.72	1	0.56	0.63	0.66	0.38	0.5	0.76	0.47	0. 38	1.7 2	0.7 1	0. 40	56. 01
CFL	0.55	1.52	0.32	0.29	0.45	0.67	0.5	0.53	0.72	0.61	0. 29	1.5 2	0.6 2	0. 35	56. 17
CTL	0.51	0.41	0.37	0.54	0.39	0.42	0.31	0.71	0.66	0.64	0. 31	0.7 1	0.5 0	0. 14	27. 74
GTL	0.39	1.01	0.4	0.61	0.45	0.84	0.75	0.38	0.47	0.29	0. 29	1.0 1	0.5 6	0. 24	42. 14
KGD L	0.31	0.45	0.38	0.39	0.41	0.41	0.39	0.29	0.34	0.34	0. 29	0.4 5	0.3 7	0. 05	13. 41
KPR ML	0.45	0.61	0.84	0.62	1.01	0.56	0.7	0.44	0.52	0.74	0. 44	1.0 1	0.6 5	0. 18	27. 56
KSM L	0.34	0.2	0.33	0.31	0.5	0.27	0.31	0.34	0.4	0.25	0. 2	0.5	0.3 3	0. 08	25. 42
LMC L	0.23	0.25	0.18	0.45	0.39	0.44	0.46	0.45	0.49	0.41	0. 18	0.4 9	0.3 8	0. 11	29. 75
NEP CTL	2.22	0.75	1.16	1.68	1.46	0.73	0.53	0.2	0.26	0.26	0. 2	2.2 2	0.9 3	0. 69	74. 06
RML	0.64	0.56	0.78	0.79	0.64	0.71	0.75	0.46	0.42	0.3	0. 3	0.7 9	0.6 1	0. 17	27. 53
SCL	0.85	0.56	0.69	0.57	0.73	0.46	0.28	0.57	0.89	0.74	0. 28	0.8 9	0.6 3	0. 18	29. 04
SSM L	0.26	0.31	0.27	0.55	0.53	0.32	0.33	0.57	0.56	0.42	0. 26	0.5 7	0.4 1	0. 13	31. 21
STL	0.8	0.53	0.43	0.68	0.65	0.53	0.55	0.46	0.66	0.43	0. 43	0.8	0.5 7	0. 12	21. 36
SSIL	0.33	0.49	0.58	0.55	0.78	0.77	0.48	0.49	0.61	0.67	0. 33	0.7 8	0.5 8	0. 14	24. 21
SSL	0.5	0.53	0.38	0.45	0.42	0.49	0.41	0.26	0.28	0.33	0. 26	0.5 3	0.4 1	0. 09	22. 85
VTM L	1.01	1.03	1.23	1.32	2.39	2.57	2.15	2.28	3.75	4.2	1. 01	4.2	2.1 9	1. 11	50. 60
VTXI L	0.37	0.5	1.02	0.62	0.45	0.38	0.36	0.39	0.48	0.25	0. 25	1.0 2	0.4 8	0. 21	44. 28
Min	0.17	0.19	0.18	0.29	0.21	0.27	0.24	0.20	0.26	0.25					
Max	2.22	1.72	1.23	1.68	2.39	2.57	2.15	2.28	3.75	4.20					
MEA N	0.58	0.65	0.59	0.63	0.69	0.64	0.55	0.53	0.70	0.65			0.6 2		35. 08
STDE V	0.47	0.42	0.34	0.35	0.51	0.51	0.43	0.46	0.78	0.90					
CV	0.81	0.66	0.58	0.55	0.73	0.80	0.78	0.86	1.12	1.40					

Table3:-Absolute Liquidity Ratio Of The Tamil Nadu Textile Companies During The Period 2005 To 2014.

ALR	Mar-05	Mar-06	Mar-07	Mar-08	Mar-09	Mar-10	Mar-11	Mar-12	Mar-13	Mar-14	M in	M ax	Me an	STD EV	CV
ACML	0.1	0.06	0.06	0.13	0.02	0.04	0.02	0.02	0.04	0.02	0.02	0.13	0.05	0.04	74.21
BASL	0.06	1.39	0.62	0.19	0.05	0.14	0.02	0.05	0.14	0.02	0.02	1.39	0.27	0.43	161.35
CFL	0.14	1.04	0.05	0.02	0.12	0.18	0.11	0.13	0.15	0.11	0.02	1.04	0.21	0.30	144.91
CTL	0.07	0.06	0.11	0.09	0.04	0.07	0.04	0.04	0.07	0.04	0.04	0.11	0.06	0.02	38.19
GTL	0.06	0.13	0.03	0.02	0.02	0.14	0.03	0.02	0.13	0.03	0.02	0.14	0.06	0.05	84.11
KGDL	0.1	0.1	0.07	0.06	0.07	0.06	0.05	0.07	0.06	0.05	0.05	0.1	0.07	0.02	25.97
KPRML	0.18	0.11	0.17	0.27	0.17	0.26	0.1	0.17	0.26	0.16	0.1	0.27	0.19	0.06	32.56
KSML	0.11	0.07	0.03	0.01	0.05	0.03	0.02	0.05	0.03	0.12	0.01	0.12	0.05	0.04	71.84
LMCL	0.16	0.2	0.15	0.31	0.12	0.16	0.12	0.12	0.16	0.12	0.12	0.31	0.16	0.06	35.97
NEPCTL	0.04	0.02	0.01	0.01	0.06	0.09	0.1	0.12	0.08	0.17	0.01	0.17	0.07	0.05	74.69
RML	0.06	0.1	0.03	0.04	0.01	0.03	0.03	0.04	0.03	0.03	0.01	0.1	0.04	0.02	61.24
SCL	0.07	0.05	0.18	0.06	0.12	0.1	0.06	0.12	0.13	0.26	0.05	0.26	0.12	0.07	56.54
SSML	0.13	0.2	0.03	0.05	0.06	0.06	0.02	0.06	0.06	0.02	0.02	0.2	0.07	0.06	80.68
STL	0.18	0.13	0.13	0.34	0.45	0.24	0.16	0.25	0.24	0.26	0.13	0.45	0.24	0.10	41.73
SSIL	0.11	0.08	0.06	0.05	0.06	0.14	0.06	0.06	0.14	0.16	0.05	0.16	0.09	0.04	45.20
SSL	0.01	0.01	0.03	0.02	0.02	0.01	0.02	0.02	0.03	0.04	0.01	0.04	0.02	0.01	47.35
VTML	0.59	0.7	1.15	0.26	2.05	1.73	0.15	1.05	1.73	0.15	0.15	2.05	0.96	0.70	73.34
VTXIL	0.03	0.01	0.6	0.08	0.03	0.1	0.03	0.03	0.1	0.03	0.01	0.6	0.10	0.18	170.46
Min	0.01	0.01	0.01	0.01	0.01	0.01	0.02	0.02	0.03	0.02					
Max	0.59	1.39	1.15	0.34	2.05	1.73	0.16	1.05	1.73	0.26					
MEAN	0.12	0.25	0.20	0.11	0.20	0.20	0.06	0.13	0.20	0.10			0.16		73.35
STD EV	0.13	0.39	0.30	0.11	0.47	0.39	0.05	0.24	0.39	0.08					
CV	1.04	1.57	1.53	1.00	2.42	1.95	0.75	1.76	1.95	0.81					

Table4:-Debt-Equity Ratio Of The Tamil Nadu Textile Companies During The Period 2005 To 2014.

DER	Mar -05	Mar -06	Mar -07	Mar -08	Mar -09	Mar -10	Mar -11	Mar -12	Mar -13	Mar -14	M in	M ax	Me an	S D	CV
ACM L	4.18	4.22	3.51	2.44	1.74	2.44	4.88	1.48	0.78	0.43	0. 43	4.8 8	2.6 1	1. 54	58. 82
BAS L	3.66	1.11	4.24	3.15	1.67	2.15	4.26	2.17	1.79	1.85	1. 11	4.2 6	2.6 1	1. 13	43. 51
CFL	3.94	3.84	0.51	0.16	5.48	0.2	1.04	1.21	1.67	1.13	0. 16	5.4 8	1.9 2	1. 84	95. 90
CTL	1.13	2.17	0.75	0.4	3.08	0.7	2.22	3.6	2.77	2.6	0. 4	3.6	1.9 4	1. 12	57. 68
GTL	2.04	1.78	1.55	0.26	0.9	0.35	0.15	0.81	0.76	0.43	0. 15	2.0 4	0.9 0	0. 67	74. 01
KGD L	2.45	4.35	0.43	0.39	0.04	1.01	1.47	2.24	1.91	1.89	0. 04	4.3 5	1.6 2	1. 27	78. 46
KPR ML	8.93	5.73	6.19	1.51	3.69	4.07	3.87	1.21	0.92	0.9	0. 9	8.9 3	3.7 0	2. 68	72. 26
KSM L	2.22	2.44	2.36	1.69	0.59	1.46	2.58	6.01	4.13	3.23	0. 59	6.0 1	2.6 7	1. 52	56. 79
LMC L	0.47	1.59	0.16	1.5	0.14	0.31	1.2	2.57	2.93	2.05	0. 14	2.9 3	1.2 9	1. 02	78. 59
NEP CTL	4	1.33	3	2.25	0.65	3.84	1.81	0.14	0.14	0.14	0. 14	4	1.7 3	1. 51	87. 04
RML	2.64	3.26	2.94	1.92	1.15	1.5	2.35	1.89	1.62	1.79	1. 15	3.2 6	2.1 1	0. 67	31. 93
SCL	1.99	2.29	3.72	1.37	1.44	1.99	2.35	3.67	2.6	1.65	1. 37	3.7 2	2.3 1	0. 83	35. 99
SSM L	2.96	3.44	2.68	1.45	0.59	1.51	2.42	5	3.05	2.33	0. 59	5	2.5 4	1. 22	48. 03
STL	1.8	1.51	3.6	2.82	2.1	1.73	3.07	2.7	1.8	2	1. 51	3.6	2.3 1	0. 69	29. 85
SSIL	2.94	5.97	4.72	3.34	1.16	6.41	5.94	1.76	1.28	0.89	0. 89	6.4 1	3.4 4	2. 17	63. 20
SSL	2.05	3.54	2.33	1.13	0.31	0.87	1.43	2.28	1.89	1.9	0. 31	3.5 4	1.7 7	0. 90	50. 74
VTM L	2.46	5.6	2.43	0.71	7.78	4.09	4.18	0.11	0.23	1.02	0. 11	7.7 8	2.8 6	2. 53	88. 58
VTXI L	1.06	2.29	3.14	1.65	0.33	1.36	1.43	2.49	2.61	2.34	0. 33	3.1 4	1.8 7	0. 85	45. 35
Min	0.47	1.11	0.16	0.16	0.04	0.20	0.15	0.11	0.14	0.14					
Max	8.93	5.97	6.19	3.34	7.78	6.41	5.94	6.01	4.13	3.23					
MEA N	2.83	3.14	2.68	1.56	1.82	2.00	2.59	2.30	1.83	1.59			2.2 3		61. 93
STDE V	1.84	1.56	1.60	0.97	2.04	1.64	1.51	1.54	1.06	0.83					
CV	0.65	0.50	0.60	0.62	1.12	0.82	0.58	0.67	0.58	0.52					

Table 5:-Interest Coverage Ratio Of The Tamil Nadu Textile Companies During The Period 2005 To 2014.

ICR	Mar -05	Mar -06	Mar -07	Mar -08	Mar -09	Mar -10	Mar -11	Mar -12	Mar -13	Mar -14	Min	Max	Mean	STDEV	CV
ACML	4.18	4.22	3.51	2.44	1.74	2.44	4.88	4.66	3.02	3.39	1.74	4.88	3.45	1.04	30.25
BASL	3.66	1.11	4.24	3.15	1.67	2.15	4.26	-0.08	2.07	2.22	-0.08	4.26	2.45	1.40	57.07
CFL	3.94	3.84	0.51	0.16	5.48	0.2	1.04	0	-0.14	0.72	-0.14	5.48	1.58	2.04	129.50
CTL	1.13	2.17	0.75	0.4	3.08	0.7	2.22	-0.65	1.75	1.21	-0.65	3.08	1.28	1.07	83.76
GTL	2.04	1.78	1.55	0.26	0.9	0.35	0.15	-0.67	-0.73	-0.75	-0.75	2.04	0.49	1.05	215.11
KGDL	2.45	4.35	0.43	0.39	0.04	1.01	1.47	1.47	1.91	1.36	0.04	4.35	1.49	1.25	83.70
KPRML	8.93	5.73	6.19	1.51	3.69	4.07	3.87	1.7	3.46	3.93	1.51	8.93	4.31	2.19	50.77
KSML	2.22	2.44	2.36	1.69	0.59	1.46	2.58	-0.44	1.67	1.51	-0.44	2.58	1.61	0.93	58.08
LMCL	0.47	1.59	0.16	1.5	0.14	0.31	1.2	-0.76	1.22	2.37	-0.76	2.37	0.82	0.92	111.62
NEPCTL	4	1.33	3	2.25	0.65	3.84	1.81	2.01	1.98	1.67	0.65	4	2.25	1.07	47.28
RML	2.64	3.26	2.94	1.92	1.15	1.5	2.35	0.85	2.43	2.44	0.85	3.26	2.15	0.78	36.27
SCL	1.99	2.29	3.72	1.37	1.44	1.99	2.35	-0.58	1.89	3.39	-0.58	3.72	1.99	1.18	59.31
SSML	2.96	3.44	2.68	1.45	0.59	1.51	2.42	-0.17	1.96	2	-0.17	3.44	1.88	1.09	57.98
STL	1.8	1.51	3.6	2.82	2.1	1.73	3.07	-0.11	2.25	2.5	-0.11	3.6	2.13	1.02	47.90
SSIL	2.94	5.97	4.72	3.34	1.16	6.41	5.94	-0.78	2.52	2.91	-0.78	6.41	3.51	2.30	65.36
SSL	2.05	3.54	2.33	1.13	0.31	0.87	1.43	-1.51	1.11	1.05	-1.51	3.54	1.23	1.32	107.63
VTML	2.46	5.6	2.43	0.71	7.78	4.09	4.18	4.75	2.63	4.04	0.71	7.78	3.87	1.97	50.86
VTXIL	1.06	2.29	3.14	1.65	0.33	1.36	1.43	0.68	1.06	-4.06	-4.06	3.14	0.89	1.92	214.29
Min	0.47	1.11	0.16	0.16	0.04	0.20	0.15	-1.51	-0.73	-4.06					
Max	8.93	5.97	6.19	3.34	7.78	6.41	5.94	4.75	3.46	4.04					
MEA	2.83	3.14	2.68	1.56	1.82	2.00	2.59	0.58	1.78	1.77			2.0		83.

N													8		71
STD EV	1.84	1.56	1.60	0.97	2.04	1.64	1.51	1.77	1.02	1.89					
CV	0.65	0.50	0.60	0.62	1.12	0.82	0.58	3.08	0.57	1.06					

Table6:-Debtors' Turnover Ratio Of The Tamil Nadu Textile Companies During The Period 2005 To 2014.

DTR	Mar-05	Mar-06	Mar-07	Mar-08	Mar-09	Mar-10	Mar-11	Mar-12	Mar-13	Mar-14	Min	Max	Mean	STD EV	CV
ACML	4.75	2.78	6.34	37.69	35.81	37.31	43.91	25.45	14.78	7.77	2.78	43.91	21.66	16.09	74.27
BASL	54.24	19.93	19.9	22.94	11.04	11.38	18.5	27.62	25.19	29.18	11.04	54.24	23.99	12.24	51.01
CFL	6.95	7.24	10.3	5.81	6.82	7.97	6.04	44.71	34.42	52.83	5.81	52.83	18.31	18.29	99.88
CTL	5.86	5.62	6.34	7.13	7.5	7.57	14.04	12.31	32.35	20.24	5.62	32.35	11.90	8.56	72.00
GTL	35.1	25.72	18.57	9.57	6.02	7.48	7.33	43.61	85.87	79.94	6.02	85.87	31.92	29.73	93.12
KGDL	8.51	4.39	4.94	4.34	3.89	3.34	4.13	22.97	21.74	24.66	3.34	24.66	10.29	8.99	87.37
KPRML	4.69	6.61	6.25	14.22	8.13	7.02	15.23	33	30.84	31.06	4.69	33	15.71	11.51	73.30
KSML	8.51	8.21	8.67	5.73	7.61	9.71	9.01	40.11	25.72	21.75	5.73	40.11	14.50	11.16	76.96
LMCL	7.14	12.14	10.56	9.4	13.35	13.47	12.27	32.87	39.84	40.12	7.14	40.12	19.12	13.04	68.24
NEPCTL	9.99	8.72	7.92	27.13	17.1	7.07	7.09	39.45	12.69	17.69	7.07	39.45	15.49	10.54	68.06
RML	6.05	3.15	4.51	6.65	7.13	7.96	10.27	44.45	36.79	32.57	3.15	44.45	15.95	15.55	97.47
SCL	16.42	15.28	29.87	38.17	27.65	16.88	13.23	27.4	31.77	38.02	13.23	38.02	25.17	9.40	36.92
SSML	4.72	6.09	6.9	7.08	9.08	8.4	5.88	25.77	37.62	34.53	4.72	37.62	14.61	12.84	87.87
STL	7	8.44	15.06	26.95	17.08	14.9	20.51	38.04	27.19	28.63	7	38.04	20.38	9.78	47.97
SSIL	3.95	4.18	5.23	5.71	9.53	16.15	8.65	33.53	41.01	47.73	3.95	47.73	17.57	16.73	95.21
SSL	4.69	5.36	6.87	5.93	4.91	4.73	4.78	34.13	31.83	29.41	4.69	34.13	13.26	12.84	96.86
VTML	24.61	19.88	21.8	33.09	37.74	25.59	34	58.42	44.86	39.52	19.88	58.42	33.95	11.86	34.94
VTXIL	3.43	2.97	4.1	3.66	3.83	3.16	5.99	9.57	8.76	4.43	2.97	9.57	4.99	2.36	47.36
Min	3.43	2.78	4.10	3.66	3.83	3.16	4.13	9.57	8.76	4.43					
Max	54.24	25.72	29.87	38.17	37.74	37.31	43.91	58.42	85.87	79.94					
MEAN	12.03	9.26	10.79	15.07	13.01	11.67	13.38	32.97	32.40	32.23			18.28		72.71
STD EV	13.33	6.70	7.28	12.29	10.43	8.48	10.55	11.87	16.61	17.18					
CV	1.11	0.72	0.67	0.82	0.80	0.73	0.79	0.36	0.51	0.53					

Table7:-Inventory Turnover Ratios Of The Tamil Nadu Textile Companies During The Period 2005 To 2014.

ITR	Mar -05	Mar -06	Mar -07	Mar -08	Mar -09	Mar -10	Mar -11	Mar -12	Mar -13	Mar -14	M in	M ax	Me an	STD EV	C V
ACM L	1.24	1.12	1	1.36	1.5	1.4	1.96	2.13	3.27	4.59	1	4.5 9	1.9 6	1.14	58. 24
BAS L	2.03	1.55	1.62	1.8	2.59	3.42	2.56	3.49	5.62	6.28	1. 55	6.2 8	3.1 0	1.66	53. 51
CFL	3.25	2.18	2.99	2.38	5.36	5.79	7.48	9.26	8.19	6.02	2. 18	9.2 6	5.2 9	2.52	47. 72
CTL	4.79	3.5	4.18	3.28	4.12	4.34	3.6	4.39	5.12	4.08	3. 28	5.1 2	4.1 4	0.57	13. 79
GTL	5.68	5.15	4.04	2.7	5.2	4.57	4.98	6.31	4.28	3.89	2. 7	6.3 1	4.6 8	1.02	21. 80
KGD L	8.88	2.94	2.97	3.48	3.41	4.02	3.94	4.27	5.28	7.19	2. 94	8.8 8	4.6 4	1.96	42. 18
KPR ML	0.9	4.87	3.74	2.45	3.13	6.14	3.49	2.78	4.87	5.07	0. 9	6.1 4	3.7 4	1.53	40. 96
KSM L	3.59	3.38	4.89	3.43	3.66	9.73	4.44	8.73	6.82	5.98	3. 38	9.7 3	5.4 7	2.30	42. 08
LMC L	6.06	6.38	5.26	4.74	6.2	0.99	1.14	2.89	5.23	6.72	0. 99	6.7 2	4.5 6	2.14	46. 86
NEP CTL	13.2 5	13.0 7	10.7	11.0 6	12.6 7	11.8 3	10.8 8	14.2 7	16.2 3	13.6 4	10 .7	16. 23	12. 76	1.73	13. 57
RML	4.6	2.73	3.29	3.27	3.61	3.1	2.58	3.34	3.68	5.23	2. 58	5.2 3	3.5 4	0.81	22. 95
SCL	4.58	3.46	3.88	4.31	4.73	2.68	1.8	2.45	3.87	4.82	1. 8	4.8 2	3.6 6	1.04	28. 49
SSM L	2.52	3	3.09	3.41	5.91	2.76	1.95	3.12	2.89	3.18	1. 95	5.9 1	3.1 8	1.04	32. 71
STL	1.8	1.63	1.97	2.62	3.33	2.84	2.83	3.14	3.72	2.37	1. 63	3.7 2	2.6 3	0.68	26. 07
SSIL	3.1	3.12	3.66	2.13	5.3	3.8	2.72	4.83	4.13	3.23	2. 13	5.3	3.6 0	0.96	26. 63
SSL	4.25	2.89	4.32	3.35	4.81	6.31	3.19	7.17	6.23	5.28	2. 89	7.1 7	4.7 8	1.45	30. 43
VTM L	7.42	4.85	5.06	3.12	4.91	3.74	5.9	6.27	5.28	4.59	3. 12	7.4 2	5.1 1	1.23	23. 99
VTXI L	2.26	1.93	2.01	1.41	1.24	1.11	1.28	3.76	3.06	2.98	1. 11	3.7 6	2.1 0	0.91	43. 05
Min	0.90	1.12	1.00	1.36	1.24	0.99	1.14	2.13	2.89	2.37					
Max	13.2 5	13.0 7	10.7 0	11.0 6	12.6 7	11.8 3	10.8 8	14.2 7	16.2 3	13.6 4					
MEA N	4.46	3.76	3.82	3.35	4.54	4.37	3.71	5.14	5.43	5.29			4.3 9		34. 17
STD EV	3.05	2.70	2.09	2.12	2.46	2.82	2.42	3.11	3.02	2.48					
CV	0.69	0.72	0.55	0.63	0.54	0.65	0.65	0.60	0.56	0.47					

Table 8:-Creditors' Turnover Ratios Of The Tamil Nadu Textile Companies During The Period 2005 To 2014.

CTR	Mar -05	Mar -06	Mar -07	Mar -08	Mar -09	Mar -10	Mar -11	Mar -12	Mar -13	Mar -14	Mi n	Ma x	Me an	STD EV	CV
ACML	69.9	53.3 2	38.6 8	34.9 9	31.3 3	14.0 2	14.3 4	88.6 4	55.8 8	55.5 1	14. 02	88. 64	45. 66	23.6 8	51. 85
BASL	30.8 7	30.7 1	29.3 1	22.1 9	17.3 1	14.7 9	16.8 1	35.9 6	34.2 1	25.3 7	14. 79	35. 96	25. 75	7.63	29. 61
CFL	5.94	6.18	8.78	5.82	4.77	5.65	4.44	62.4 5	68.2	54.4 3	4.4 4	68. 2	22. 67	27.1 5	119 .79
CTL	15.7 2	14.1 3	14.7 6	8.44	6.42	11.0 9	30.9	12.2	60.3 3	40.3 5	6.4 2	60. 33	21. 43	17.2 5	80. 47
GTL	8.15	7.78	9.2	7.37	6.97	10.9 8	10.3 1	79.9 1	27.4 6	43.0 3	6.9 7	79. 91	21. 12	23.7 4	112 .44
KGDL	12.5	11.2 3	9.79	7.84	8.3	9.77	11.7 7	81.4 4	76.6 9	79.4 8	7.8 4	81. 44	30. 88	33.4 0	108 .14
KPRML	11.0 5	13.4	9.64	7.68	6.97	7.37	8.58	61.9 7	69.2 8	43.6 5	6.9 7	69. 28	23. 96	24.5 7	102 .57
KSML	12.5	12.8 2	12.9 5	8.63	7.37	8.85	10.8 3	33.1 5	12.1 2	19.8	7.3 7	33. 15	13. 90	7.59	54. 57
LMCL	18.1	23.0 1	22.8 5	12.4	9.6	11.8 5	14.4 3	50.6	52.4 5	53.0 5	9.6	53. 05	26. 83	17.9 5	66. 88
NEPCTL	43.8	47.1 9	186. 43	168. 63	76.6 9	79.4 8	98.3 3	42.3 1	74.1 7	76.0 4	42. 31	186 .43	89. 31	50.0 2	56. 01
RML	29.3 4	22.3 2	20.3 2	12.9 4	11.0 8	11.7 4	10.0 5	35.1 4	30.5 7	31.9 9	10. 05	35. 14	21. 55	9.72	45. 10
SCL	8.03	6.58	7.31	7.58	8.13	8.26	12.0 4	21.8 8	12.9 7	12.8 7	6.5 8	21. 88	10. 57	4.65	43. 99
SSML	15.9 9	17.9 2	14.5 4	8.44	5.83	7.6	8.53	62.4 1	37.9 1	47.2 4	5.8 3	62. 41	22. 64	19.6 1	86. 62
STL	12.1	11.5 4	14.4 7	11.9 1	6.52	7.83	11.2 9	30.2 5	40.4 1	42.8 9	6.5 2	42. 89	18. 92	13.6 2	71. 99
SSIL	12.9 1	13.4 8	12.1 4	9.11	8.97	9.52	10.1 6	61.3 4	36.0 3	28.8 4	8.9 7	61. 34	20. 25	17.1 4	84. 66
SSL	21.7 2	19.2 6	18.9 5	14.7 1	13.6 8	17.6	22.9 4	50.3 3	49.4 4	44.7 6	13. 68	50. 33	27. 34	14.7 1	53. 81
VTML	29.8	20.5 8	18.8 1	12.5 3	9.54	7.22	7.73	8.7	8.05	7.54	7.2 2	29. 8	13. 05	7.60	58. 22
VTXIL	3.29	2.92	3.51	2.68	2.44	2.91	4.12	4.73	7.91	9.21	2.4 4	9.2 1	4.3 7	2.33	53. 30
Min	3.29	2.92	3.51	2.68	2.44	2.91	4.12	4.73	7.91	7.54					
Max	69.9 0	53.3 2	186. 43	168. 63	76.6 9	79.4 8	98.3 3	88.6 4	76.6 9	79.4 8					
MEAN	20.1 0	18.5 8	25.1 4	20.2 2	13.4 4	13.7 0	17.0 9	45.7 5	41.8 9	39.7 8			25. 57		71. 11
STD EV	16.2 1	13.4 6	41.1 3	37.7 3	17.0 1	16.7 8	21.2 4	25.1 6	22.8 4	20.4 8					
CV	0.81	0.72	1.64	1.87	1.27	1.23	1.24	0.55	0.55	0.51					

Table9:-Return On Capital Employed Of The Tamil Nadu Textile Companies During The Period 2005 To 2014

ROC E	Mar -05	Mar -06	Mar -07	Mar -08	Mar -09	Mar -10	Mar -11	Mar -12	Mar -13	Mar -14	Min	Max	Mean	STD EV	CV
ACML	13.25	14.58	9.73	11.72	11.93	14.85	20.52	20.57	22.78	25.48	9.73	25.48	16.54	5.37	32.45
BASL	30.18	9.07	6.44	6.54	7.22	9.41	15.15	5.27	16.97	15.45	5.27	30.18	12.17	7.61	62.52
CFL	19.47	7.9	4.88	2.54	-45.92	7.49	1.74	25.36	14.62	26.22	-45.92	26.22	6.43	20.47	318.32
CTL	9.97	13.45	11.69	5.08	-12.04	12.27	23.29	0.53	13.82	21.65	-12.04	23.29	9.97	10.25	102.85
GTL	13.37	11.86	5.9	3.75	-3.22	1.84	6.25	0.07	-19.95	-1.89	-19.95	13.37	1.80	9.37	521.50
KGDL	14.23	19.07	8.21	9.02	7.07	16.94	20.69	27.68	31.16	24.97	7.07	31.16	17.90	8.40	46.92
KPRML	23.56	17.17	13.18	10.2	15.68	18.14	15.81	13.98	27.26	25.42	10.20	27.26	18.04	5.61	31.08
KSML	17.26	16.53	17.49	13.05	7.24	12.54	15.35	1.18	17.86	16.38	1.18	17.86	13.49	5.40	40.01
LMCL	8.87	10.34	9.2	1.35	4.1	6.92	9.46	0.53	21.79	14.30	0.53	21.79	8.69	6.25	71.92
NEPCTL	6.65	35.06	8.1	6.76	-2.13	0.57	-0.49	1.70	3.97	3.97	-2.13	35.06	6.42	10.61	165.30
RML	15.59	14.17	16.56	12.7	11.2	13.86	16.42	12.22	17.75	16.92	11.20	17.75	14.74	2.23	15.14
SCL	13.42	15.64	19.07	13.83	15.8	14.53	19.67	4.61	23.14	28.96	4.61	28.96	16.87	6.48	38.41
SSML	22.06	19.74	18.53	12.23	7.59	11.87	15.04	3.34	18.56	18.54	3.34	22.06	14.75	5.94	40.28
STL	10.21	14.02	14.44	11.42	5.49	7.32	14.54	4.21	19.77	15.72	4.21	19.77	11.71	4.93	42.05
SSIL	22.64	26.51	25.04	20.76	15.28	27.65	26.53	4.62	26.69	26.21	4.62	27.65	22.19	7.23	32.58
SSL	15.71	18.48	14.99	11.75	4.88	14.8	17.61	-7.18	22.92	20.03	-7.18	22.92	13.40	8.73	65.19
VTML	21.14	11.83	15.91	10.09	11.97	13.44	25.57	15.43	21.42	21.09	10.09	25.57	16.79	5.19	30.92
VTXIL	9.03	20.43	10.87	10.25	1.7	6.44	9.72	4.84	14.62	-13.83	-13.83	20.43	7.41	9.07	122.49
Min	6.65	7.90	4.88	1.35	-45.92	0.57	-0.49	-7.18	-19.95	-13.83					
Max	30.18	35.06	25.04	20.76	15.80	27.65	26.53	27.68	31.16	28.96					
MEAN	15.92	16.44	12.79	9.61	3.55	11.72	15.16	7.72	17.51	16.98			12.74		12.74
STD EV	6.23	6.50	5.41	4.69	14.28	6.29	7.51	9.46	11.14	11.00					
CV	0.39	0.40	0.42	0.49	4.03	0.54	0.50	1.23	0.64	0.65					

Table10:-Return On Equity Of The Tamil Nadu Textile Companies During The Period 2005 To 2014.

ROE	Mar -05	Mar -06	Mar -07	Mar -08	Mar -09	Mar -10	Mar -11	Mar -12	Mar -13	Mar -14	Min	Max	Mean	STD EV	CV
ACML	19.64	19.17	2.57	12.88	9.23	12.73	19.24	17.34	12.19	14.06	2.57	19.64	13.90	5.34	38.40
BASL	34.65	10.96	10.92	7.17	5.02	8.72	18.68	-8.56	12.71	13.00	-8.56	34.65	11.33	10.86	95.92
CFL	18.95	6.03	-3.66	-14.99	-96.6	-35.41	64.67	31.18	10.40	35.70	-96.60	64.67	1.63	44.38	272.815
CTL	-1.84	5.68	-2.33	-7.56	-16.89	-14.24	37.24	34.98	16.87	10.82	-16.89	37.24	6.27	18.87	300.83
GTL	10.25	9.31	4.2	-11.72	-16.12	12.55	17.76	77.09	64.58	24.56	-16.12	77.09	19.25	29.99	155.80
KGDL	12.3	26.92	-13.43	-12.53	-28.61	0.4	9.54	14.10	24.54	9.74	-28.61	26.92	4.30	17.75	412.98
KPRML	33.63	17.56	15.6	1.98	9.36	12.28	12.8	5.39	15.21	17.18	1.98	33.63	14.10	8.54	60.59
KSML	18.72	24.67	24.7	19.93	-12.95	13.78	29.2	-69.15	23.86	13.46	-69.15	29.2	8.62	29.75	345.07
LMCL	38.07	37.37	23.24	-28.63	-15.55	6.27	19.35	-91.46	45.12	28.78	-91.46	45.12	6.26	41.77	667.64
NEPCTL	-1.94	29.24	2.41	-8.58	-14.48	-12.97	-0.57	3.95	4.51	4.51	-14.48	29.24	0.61	12.33	202.757
RML	9.74	11.73	15.65	9.75	5.74	9.23	18.13	0.84	13.54	14.23	0.84	18.13	10.86	5.02	46.24
SCL	8.17	19.1	24.91	9.48	1.12	16.98	25.94	-39.66	20.00	27.56	-39.66	27.56	11.36	19.85	174.75
SSML	30.6	30.87	40.35	18.17	-13.09	13.74	28.14	-47.93	23.32	19.72	-47.93	40.35	14.39	26.16	181.79
STL	6.51	16.93	15.06	6.26	4.44	4.97	20.95	-18.05	17.58	14.42	-18.05	20.95	8.91	11.19	125.69
SSIL	16.96	28.98	30.79	19.75	1.86	29.31	26.96	-21.64	14.04	18.56	-21.64	30.79	16.56	16.07	97.05
SSL	11.7	19.69	11.39	1.42	-30.82	0.35	13.18	-8.66	10.73	4.53	-30.82	19.69	3.35	14.45	431.23
VTML	9.57	1.06	7.03	0.48	3.99	4.72	12.97	6.54	11.74	10.01	0.48	12.97	6.81	4.29	63.00
VTXIL	-0.92	37.51	11.3	6.4	-10.14	2.92	5.49	-23.82	0.75	18.07	-23.82	37.51	4.76	16.28	342.32
Min	-1.94	1.06	-13.4	-28.6	-96.6	-35.4	-0.57	-91.4	0.75	4.51					

			3	3	0	1		6							
Max	38.0 7	37.5 1	40.3 5	19.9 3	9.36	29.3 1	64.6 7	77.0 9	64.5 8	35.7 0					
MEAN	15.2 6	19.6 0	12.2 6	1.65	- 11.9 2	4.80	21.0 9	- 7.64	18.9 8	16.6 1			9.0 7		460. 83
STDEV	12.3 7	10.8 5	13.3 1	13.3 4	24.5 6	14.2 7	14.1 6	39.2 6	14.8 2	8.29					
CV	0.81	0.55	1.09	8.10	- 2.06	2.98	0.67	- 5.14	0.78	0.50					

Table11:-Operating Profit Margins Of The Tamil Nadu Textile Companies During The Period 2005 To 2014.

OPM	Mar -05	Mar -06	Mar -07	Mar -08	Mar -09	Mar -10	Mar -11	Mar -12	Mar -13	Mar -14	Min	Max	Mean	STDEV	CV
ACML	26.1 7	33.0 8	22.0 3	29.2 6	26.8 4	27.6 8	30.8 4	30.9 1	23.0 3	22.8 1	22. 03	33. 08	27. 26	3.81	13. 97
BASL	44.0 9	26.7 7	26.9	20.6 6	15.9 9	19	22.0 4	7.85	20.4 8	17.0 0	7.8 5	44. 09	22. 08	9.48	42. 95
CFL	11.8 6	13.8 9	4.89	2.53	- 34.8 7	5.04	1.13	12.8 2	8.36	11.1 7	- 34. 87	13. 89	3.6 8	14.2 6	387. 23
CTL	9.61	15.2 9	12.8 8	8.14	- 12.9 5	11.0 5	16.4	0.22	13.8 1	9.62	- 12. 95	16. 40	8.4 1	8.78	104. 41
GTL	10.9 7	14.4 5	15.4 5	12.3 3	- 12.8 6	6.08	13.8 9	0.15	- 8.05	- 6.20	- 12. 86	15. 45	4.6 2	10.5 7	228. 60
KGDL	9.03	18.2 3	7.94	8.44	5.76	11	10.4 3	12.0 3	11.8 9	8.76	5.7 6	18. 23	10. 35	3.38	32. 61
KPRML	29.7	26.9 8	25.7	14.8 6	20.0 8	23.0 3	22.3 1	15.8 0	24.3 2	19.3 1	14. 86	29. 70	22. 21	4.76	21. 45
KSML	18.5 3	21.7 6	22.8 9	21.8 7	13.5 6	19.6 8	19.0 7	1.53	17.1 7	12.8 7	1.5 3	22. 89	16. 89	6.35	37. 60
LMCL	9.58	11.1 6	11.7 5	2.93	8.49	11.6 3	12.1 9	0.78	26.3 7	13.1 4	0.7 8	26. 37	10. 80	6.84	63. 30
NEPCTL	10.0 6	36.3 1	10.3 4	7.71	- 2.12	0.46	- 0.41	18.5 8	54.8 4	54.8 4	- 2.1 2	54. 84	19. 06	21.9 0	114. 91
RML	22.9 5	27.6 5	29.4 5	24.6 6	25.2 2	28.2 6	30.6 4	20.4 9	26.4 3	22.5 7	20. 49	30. 64	25. 83	3.25	12. 58
SCL	12.8 3	20.3 9	21.7 5	15.8 2	15.9 4	17.4 8	20.1 9	3.72	16.4 3	15.8 2	3.7 2	21. 75	16. 04	5.09	31. 74
SSML	22.4 5	21.9	24.4 1	19.5 1	15.3 5	20.6 9	20.6 7	4.27	18.2 8	15.7 7	4.2 7	24. 41	18. 33	5.70	31. 09
STL	23.4 2	33.2 6	29.5 7	24.5 4	17.8 5	16.9 7	19.3 1	5.57	18.6 9	14.8 0	5.5 7	33. 26	20. 40	7.81	38. 30
SSIL	15.1 7	24.9 8	30.7 2	31.8	19.1	32.0 8	29.9	6.30	26.5 2	22.0 3	6.3 0	32. 08	23. 86	8.38	35. 13
SSL	11.6 4	18.0 9	15.2 7	12.1 8	4.74	11.6	11.9 1	- 4.14	11.3 1	8.57	- 4.1 4	18. 09	10. 12	6.13	60. 59
VTML	17.2 1	12.2 7	15.1 5	10.7 5	11.9 5	15.1	20.7	13.7 8	16.9 1	14.9 7	10. 75	20. 70	14. 88	2.93	19. 69
VTXIL	9.85	25.6	19.7 8	22.8 9	5.02	22.4 3	25.0 3	10.5 3	34.8 5	- 72.6	- 72.	34. 85	10. 34	30.4 6	294. 70

										1	61				
Min	9.03	11.16	4.89	2.53	-34.87	0.46	-0.41	-4.14	-8.05	-72.61					
Max	44.09	36.31	30.72	31.80	26.84	32.08	30.84	30.91	54.84	54.84					
MEAN	17.51	22.34	19.27	16.16	7.95	16.63	18.12	8.96	20.09	11.40			15.84		87.27
STDEV	9.29	7.54	7.87	8.70	15.50	8.49	8.89	8.82	12.66	24.01					
CV	0.53	0.34	0.41	0.54	1.95	0.51	0.49	0.98	0.63	2.11					

Table12:-net profit margins of the tamilnadu textile companies during the period 2005 to 2014.

NPM	Mar-05	Mar-06	Mar-07	Mar-08	Mar-09	Mar-10	Mar-11	Mar-12	Mar-13	Mar-14	Min	Max	Mean	STDEV	CV
ACML	14.56	17.61	1.77	8.55	6.21	8.02	13.23	13.23	6.10	7.73	1.77	17.61	9.70	4.80	49.46
BASL	21.61	18.84	16.38	5.64	2.81	4.24	7.07	-3.56	4.88	4.51	-3.56	21.61	8.24	8.00	97.02
CFL	4.31	6.05	-1.68	-6.1	-45.58	-4.14	-10.08	-6.47	-2.35	3.43	-45.58	6.05	-6.26	14.75	-235.64
CTL	-0.81	2.67	-1.14	-4.25	-27.9	-2.32	6.27	-12.38	4.47	1.33	-27.90	6.27	-3.41	10.05	-295.18
GTL	1.81	1.94	1.95	-5.87	-41.45	-23.06	-7.55	-8.30	-12.19	-13.70	-41.45	1.94	-10.64	13.44	-126.26
KGDL	2.63	7.22	-3.21	-3.12	-5.21	0.06	1.3	1.75	3.00	1.11	-5.21	7.22	0.55	3.62	655.03
KPRML	17.15	11.76	13.26	1.36	6.1	6.9	7.98	2.63	6.75	6.58	1.36	17.15	8.05	4.79	59.56
KSML	3.84	5.39	4.92	5.11	-2.84	2.9	5.7	-9.75	3.41	1.88	-9.75	5.70	2.06	4.84	235.55
LMCL	2.74	4.12	3.81	-4.81	-1.74	1.07	3.03	-8.78	6.70	4.26	-8.78	6.70	1.04	4.77	458.25
NEPCTL	-1.32	26.94	2.76	-8.75	-12.7	-9.01	-0.21	8.58	34.84	24.84	-12.70	34.84	6.60	16.74	253.81
RML	5.49	6.82	8.16	4.87	2.82	4.52	8.28	0.44	6.92	6.13	0.44	8.28	5.44	2.43	44.61
SCL	1.91	5.04	6.41	2.3	0.26	4.06	5.54	-6.22	3.63	5.32	-6.22	6.41	2.83	3.70	130.81
SSML	5.47	5.56	8.13	4.37	-3.19	3.23	5.92	-7.73	4.19	3.66	-7.73	8.13	2.96	4.77	161.09
STL	5.77	17.62	16.01	6.63	4.6	2.67	6.7	-5.85	5.31	4.13	-5.85	17.62	6.36	6.60	103.82
SSIL	5.41	10.0	11.6	9.51	0.84	14.0	11.9	-	5.60	7.68	-	14.0	6.7	6.94	103.82

		3	6			8	7	9.71			9.7 1	08	1		47
SSL	2.98	6.05	3.53	0.44	- 7.48	0.08	2.91	11.2 7	1.73	0.64	- 7.4 8	11. 27	2.2 1	4.76	214. 95
VTM L	6.15	0.78	5.1	0.39	3.39	4.77	9.01	5.38	8.60	6.69	0.3 9	9.0 1	5.0 3	2.89	57.4 7
VTX IL	- 0.31	15.6 5	10.3 4	6.13	- 9.83	3.07	4.24	- 13.8 8	3.47	15.9 4	- 13. 88	15. 94	3.4 8	9.72	279. 25
Min	- 1.32	0.78	- 3.21	- 8.75	- 45.5 8	- 23.0 6	- 10.0 8	- 13.8 8	- 12.1 9	- 13.7 0					
Max	21.6 1	26.9 4	16.3 8	9.51	6.21	14.0 8	13.2 3	13.2 3	34.8 4	24.8 4					
MEA N	5.52	9.45	6.01	1.24	- 7.27	1.17	4.52	- 2.74	5.28	5.12			2.8 3		124. 84
STD EV	6.19	7.13	5.81	5.55	15.5 5	7.80	5.91	8.22	8.69	7.46					
CV	1.12	0.75	0.97	4.46	- 2.14	6.64	1.31	- 3.00	1.65	1.46					



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RESEARCH ARTICLE

COMPETING IDEAS OF REGULATIONISM – WHAT CAN BE LEARNT FROM THE GERMAN MOVE TO A MORE COMPREHENSIVE REGULATION OF PROSTITUTION.

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Abstract

In 2002, Germany legalised prostitution through the enactment of the Prostitution Act, thereby following one of the most liberal regulationist approaches to date. The following year, New Zealand enacted the Prostitution Reform Act 2003, which has become an often-referenced example of decriminalisation. Despite both jurisdictions following a regulationist approach, New Zealand's model has attracted much positive international attention, whereas the German approach is used as an example of the shortcomings within regulationism. Drawing on the findings of the governmental evaluations of both New Zealand's and Germany's prostitution legislation, this article argues that the significant differences between the New Zealand and German models of regulating prostitution are not to be found within the legislation, but rather in the evaluation thereof. This finding calls into question the evidence presented within the evaluation of approaches to prostitution, and suggests that there may be other societal factors which decide how the success of legislation is determined. In Germany, the evaluation of the 2002 Prostitution Act (Prostitutionsgesetz) resulted in a move towards stricter regulation of prostitution. This article examines the changes to the law that will enter into force in July 2017, which are aimed at ensuring that the rights of sex workers in Germany will not just be protected in theory, but also in practice.

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Introduction:-

As a subject matter that divides people in their opinions regarding morality, best practice, society and laws, prostitution and the way it should be regulated in individual jurisdictions is a contemporary topic that regularly finds its way into governmental debates regarding the best way to regulate the issue. As will be demonstrated in the following article, there are a number of different approaches to regulating prostitution, and much debate regarding whether these approaches are appropriate to effectively deal with prostitution in relation to the desired objectives sought to be achieved by a particular government. In contemporary law reform discussions, a general practice can be found in examining other jurisdictions and the approaches taken in order to evaluate whether a particular approach would be similarly suitable in one's own jurisdiction. As will be demonstrated in the following, a country often referred to in relation to decriminalisation, is New Zealand, with their regulationist approach to prostitution. However, Germany, as one of the countries with the most liberal regulationist approach to prostitution, is rarely used as a similar role model in this sense. In order to examine why this may be the case, the following will examine the German model of regulationism in light of the upcoming changes to their legislation on prostitution, which will be

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entering into force in July 2017. In particular, the focus will be on the German experience of regulationism in legal practice, what has been learnt, and how this has affected the upcoming German law reforms. In light of the German legal terminology, the words prostitutes and sex workers will be used interchangeably.

The Theoretical Approaches to Prostitution:-

There are a number of terms used in relation to the regulation of prostitution in law and policy. Many are commonly used interchangeably to describe different legal approaches, such as “criminalisation” and “prohibitionism,” in some cases “legalisation” and “decriminalisation” as well as in other cases “decriminalisation” and “abolitionism.” However, when investigating the terms, it becomes clear, that what is often used to describe a particular regulatory system in the media or certain policy documents, or even in certain journal articles, is not actually in line with the theoretical meaning of the terms, which leads to confusion. In this sense, it has to be explained that these terms fall into two fundamentally separate categories. In this sense, the Council of Europe Resolution 1579 (2007) is correct when explaining that the three predominant approaches adopted in the 47 member states of the Council of Europe are prohibitionist, regulationist and abolitionist.¹ This is because these three terms actually address and describe regulatory approaches. However, it is often found that decriminalisation is mentioned as a fourth option, often with reference made to the system in New Zealand.² However, “decriminalisation” is not an approach in itself. Instead, it is a function, or a tool used within the development of approaches, which merely describes the absence of criminalisation. In this sense, the terms “criminalisation” and “legalisation” fall into the same category of legal terminology. Accordingly, “criminalisation” describes the process of making an activity prohibited under criminal law provisions³ and “legalisation” describes the process of reform in which something, which was considered illicit, ceases to be viewed as such under the law, and instead becomes legitimate.⁴ In this sense, although these latter three terms may constitute key attributes within the regulatory approaches to prostitution, they are not the same and cannot be used interchangeably.

In terms of the different theoretical regulatory approaches to prostitution, prohibitionist approaches generally target both the sale and the purchase of these services by utilising criminalisation. Prohibitionism can also involve a categorical prohibition of certain behaviours, which are closely related to selling or purchasing sexual services, including activities like street solicitation, or running agencies or sex businesses.⁵

Abolitionist approaches involve policies, which do not unconditionally criminalize acts of prostitution or activities closely related. The defining attribute of an abolitionist approach is that it will target only one side of an interaction in an attempt to make the entire interaction obsolete. Usually the side targeted is the procurement side, which is then categorically criminalized, as it is thought that the elimination of demand will eliminate the entire transaction.⁶

¹Council of Europe, Parliamentary Assembly, 'Resolution 1579 (2007) Prostitution – Which Stance to Take?' Assembly debate on 4 October 2007 (35th Sitting) s.4 et seq.

² See for example: Jane Scoular, *The Subject Of Prostitution. Sex/Work, Law And Social Theory* (1st edn, Taylor & Francis Ltd 2016) 116; Victor Minichiello, *Male sex work and society* (Harrington Park Press, 2014) 191; Joanna Phoenix, *Regulating sex for sale: prostitution policy reform and the UK* (Policy Press, 2009) 19; For an example of the issues of decriminalisation as a term to classify the model taken in New Zealand, see: Kelly, Liz, Maddy Coy, and Rebecca Davenport. 'Shifting sands: a comparison of prostitution regimes across nine countries.' (2009) London: London Metropolitan University.

³ See for example: M J Allen, *Textbook on criminal law* (Oxford University Press, 2015) 12 et seq.; Nina Peršak, *Criminalising harmful conduct: the harm principle, its limits and continental counterparts* (Springer, 2007) 5; Caron Beaton-Wells; Ariel Ezrachi, *Criminalising Cartels: Critical Studies of an International Regulatory Movement* (Hart Publishing, 2011) Chapter 13.

⁴ See for example: Philip Bean, *Legalising drugs: debates and dilemmas* (Bristol : Policy Press, 2010) iv; Janina Dill, *Legitimate targets?: social construction, international law and US bombing* (Cambridge University Press, 2015) 145.

⁵Peter De Marneffe, *Liberalism And Prostitution* (Oxford University Press 2010) 16; Phil Hubbard, Roger Matthews and Jane Scoular, 'Regulating Sex Work In The EU: Prostitute Women And The New Spaces Of Exclusion' (2008) 15 Gender, Place & Culture.

⁶Peter De Marneffe, *Liberalism And Prostitution* (Oxford University Press 2010) 12, 29; Joyce Outshoorn, *The Politics Of Prostitution* (Cambridge University Press 2004) 8, see also: GeetanjaliGangoli and Nicole Westmarland, *International Approaches To Prostitution* (Policy Press 2006) 175; K. Thorlby, 'May-Len Skilbrei And CharlottaHolmstrom, Prostitution Policy In The Nordic Region: Ambiguous Sympathies.' (2015) 18 Sexualities.

Regulationism refers to a set of policies that neither criminalise the sale or purchase of prostitution services nor any other activities that are closely related. Accordingly, regulationist approaches will legalise the activity itself, however, while imposing certain restrictions, which differentiate prostitution from other transactions or businesses. Restrictions in this sense can involve, for instance, imposing age restrictions or introducing targeted health regulations.⁷ This attribute of regulationism differentiates it from full decriminalisation. Full decriminalisation may be what is meant by “decriminalisation” when used to describe a regulatory approach to prostitution. Full decriminalisation refers to a set of policies that incorporate normalisation or laissez-faire.⁸

Calls for the Decriminalisation of Prostitution:-

Over the past decades, there have been numerous calls for decriminalisation of prostitution. These calls become particularly public, when governments open discussions for potential law reforms in the area of prostitution.⁹ The ideas that are put forward in this situation usually relate to the general feminist ideas of decriminalization, which involve the objective of decriminalizing “prostitution in those places where it is still a crime to be or to visit a whore.”¹⁰ However, as the term “decriminalization” refers to the complete abolition of any laws, which seek to criminalize prostitution as well as any offences that are related to prostitution,¹¹ it is implied that prostitution activities are legitimised as a form of work.¹² Thus, decriminalisation would involve removing any penal sanctions, while avoiding to introduce any legal measures to regulate prostitution.

When looking through a number of bills and reports from governments reviewing potential changes to the laws on prostitution, New Zealand is often referred to and discussed as an example of decriminalisation in the false use of the term.¹³ New Zealand sought to decriminalise prostitution in 2003 with the implementation of the Prostitution Reform Act (PRA).¹⁴ The objectives of this piece of legislation were claimed to be to decriminalise prostitution,¹⁵ construct a legal framework to protect sex workers’ human rights and prevent them from being exploited, as well as to promote sex workers’ welfare and occupational health and safety and to “contribute to public health; and prohibit the use of prostitution of persons under 18 years of age.”¹⁶ These objectives already indicate that what is being referred to as “decriminalisation” is not what the theoretical concept of decriminalisation dictates. Instead, it appears that New Zealand’s decriminalisation approach is a form of regulationist approach. This becomes even more apparent when looking more closely at the various provisions that were implemented through the PRA, such as, for instance the provisions listed in part 2 of the Act which regulate prostitution as commercial sexual services,

⁷Peter De Marneffe, *Liberalism And Prostitution* (Oxford University Press 2010) 29; Brigit C. A Toebe, *The Right To Health. A Multi-Country Study Of Law, Policy And Practice*; Melissa Hope Ditmore, *Encyclopedia Of Prostitution And Sex Work* (Greenwood Press 2006) 372.

⁸ Ibid.

⁹Nadia Khomami, 'Decriminalisation Of Sex Workers In England And Wales Backed By Mps' *The guardian* (2017) <<https://www.theguardian.com/society/2016/jul/01/decriminalisation-of-sex-workers-in-england-and-wales-backed-by-mps>> accessed 28 March 2017; Al Jazeera, 'French Parliament Passes Prostitution Bill' (2017) <<http://www.aljazeera.com/news/europe/2013/12/french-parliament-passes-prostitution-bill-2013124182412605944.html>> accessed 28 March 2017; Joyce Outshoorn, *The Politics Of Prostitution* (1st edn, Cambridge University Press 2004).

¹⁰ L O Ericsson 'Charges Against Prostitution: An Attempt at a Philosophical Assessment' in D Kelly Weisberg (ed) *Application of Feminist Legal Theories to Women's Lives: Sex, Violence, Work and Reproduction* (Temple University Press, 1996) 208-216.

¹¹Judith Kilvington, Sophie Day and Helen Ward, 'Prostitution Policy In Europe: A Time Of Change?' (2001) 67 *Feminist Review*.

¹² South African Law Commission (SALC) Issue Paper 19 (Project 107) 'Sexual Offences: Adult Prostitution' (2002).

¹³UK Parliament, 'Prostitution - Point 5: Other Legislative Models' <<https://www.publicationsparliamentuk/pa/cm201617/cmselect/cmhaff/26/2608htm>> 2016 accessed 31 March 2017.

¹⁴ Prostitution Reform Act 2003 (New Zealand), Public Act, 2003 No 28, 27 June 2003.

¹⁵ Ibid. Part 1, s. 3; while remaining neutral and avoiding any endorsement or comment in relation to morally sanctioning prostitution or the use thereof.

¹⁶ New Zealand Government, Ministry of Justice, 'Report of the Prostitution Law Review Committee on the Operation of the Prostitution Reform Act 2003' (New Zealand Ministry of Justice 2008) <<https://maggiecnell.files.wordpress.com/2012/04/report-of-the-nz-prostitution-law-committee-2008.pdf>> accessed 31 March 2017.

including regulating an age restriction of 18 years of age or a certification regime for owners or managers of brothels.¹⁷

A point which will be interesting when looking at the German regulatory approach is that the first purpose named listed in the PRA 2003 is to “[safeguard] the human rights of sex workers and protects them from exploitation”¹⁸ and that the first regulatory provision of commercial sex in part 2 is that “No contract for the provision of, or arranging the provision of, commercial sexual services is illegal or void on public policy or other similar grounds.”¹⁹ As it is already clear from the theoretical concept of the term “decriminalisation” that the approach taken in New Zealand is regulationist rather than fully decriminalised, the question arises what makes this approach unique in relation to other regulatory approaches to prostitution.

The main reason for this can be the reported success of the approach, as promoted by the New Zealand Government. In this sense, the PRA underwent governmental evaluation in 2008 by a governmentally arranged Law Review Committee.²⁰ The main findings of this report were that the PRA had not had any significant impact on the amount of sex workers working in the sex industry; that the PRA protected the rights of children, in the sense of people under the age of 18 years; that the PRA succeeded in protecting adults’ right to refuse engagement in sex work, which included being able to refuse particular clients or sexual services, and that the PRA offered sufficient protection from exploitative and degrading employment practices.²¹ The report suggests that this protection of the sex workers’ rights was only achieved through decriminalisation, in the sense of having removed the illegality of this kind of work. This is not disputed here. The use here of decriminalisation is clearly in line with the meaning of the term as explained above. In this sense, it can be argued that New Zealand decriminalised prostitution. However, it should be emphasised that this was only one part of the overall regulatory approach taken.²²

Although the report noted that the Act had not managed to eliminate all exploitative practices in practice, there was clear evidence from their qualitative study that more sex workers felt they were likely to report any incidents of violence they had experienced to the police. Street work was still claimed to be dangerous for sex workers, who were urged to either leave the industry entirely or move their work to safer, indoor settings.²³ Tim Barnett, a New Zealand Member of Parliament, who had sponsored the Prostitution Reform Bill while it was passing through Parliament, stated that the decriminalisation of prostitution had resulted in a “massive improvement in police-sex worker cooperation” that had facilitated the solving of murders of sex workers, the exposure and conviction of corrupt and law breaking brothel owners and police, as well as the detection of traffickers of sex workers.²⁴ According to the New Zealand Prostitutes Collective, the approach to prostitution in New Zealand ensures the protection of prostitutes’ rights, health and welfare.²⁵ Furthermore, they emphasise that this not only protects sex

¹⁷ Prostitution Reform Act 2003 (New Zealand) – Part 2, ss. 20-23.

¹⁸ Prostitution Reform Act 2003 (New Zealand) – Part 1, s. 3 Purpose.

¹⁹ Prostitution Reform Act 2003 (New Zealand) –Part 2, s. 1, Commercial sexual services, Contracts for commercial sexual services not void, (7): “No contract for the provision of, or arranging the provision of, commercial sexual services is illegal or void on public policy or other similar grounds.”

²⁰ New Zealand Government, Ministry of Justice (n 16).

²¹ Ibid.

²² See: Prostitution Reform Act 2003 (New Zealand).

²³ New Zealand Government, Ministry of Justice, (n 16).

²⁴ Tim Barnett (PRO0127) cited in House of Commons Home Affairs Committee (UK), ‘Prostitution, Third Report of Session 2016–17’, 15 June 2016, Committee reports are published on the Committee’s website at <www.parliament.uk/homeaffairscom> accessed 28 March, 2017; see also New Zealand Parliament Hansard, ‘Prostitution Reform Bill’ — Procedure, Third Reading, 25 June 2003, Volume: 609, Page: 6585, <https://www.parliament.nz/en/pb/hansard-debates/rhr/document/47HansD_20030625_00001319/prostitution-reform-bill-procedure-third-reading>accessed 28 March, 2017.

²⁵ UK Parliament, Home Affairs Committee, Committee Evidence, ‘Written evidence submitted by the New Zealand Prostitutes’ Collective’ 2016, <http://data.parliament.uk/writtenevidence/committeeevidence.svc/evidencedocument/home-affairs-committee/prostitution/written/29308.pdf>accessed 28 March 2017.

workers from exploitation, but also provides them with the legal opportunity to report any violence they encounter to the relevant authorities without having to fear any action being taken against them or their clients by the police.²⁶

The New Zealand model has been emphasised as an example in various governmental reports looking at the issue of the regulation of prostitution. In this sense, the UK House of Commons, for instance, examined the New Zealand model in their Third Report of Session 2016–17.²⁷

The German Approach to Regulating Prostitution:-

The German approach to prostitution is openly regulationist by nature, with many counting it not only as the most liberal approach to prostitution in Europe, but also as one of the most liberal approaches in the world.²⁸ Nevertheless, the discussions about prostitution reform tend to focus on the New Zealand model, or even the Dutch model, rather than the German approach.²⁹ One of the reasons for this could be the stark criticism Germany has received in light of the size of its sex industry.³⁰ In this sense, Germany is often referred to as the “bordello of Europe.”³¹ However, the following will look at the German model in closer detail, in order to demonstrate that this system may not be as different as the New Zealand model, which is often referred to as “decriminalisation.” In particular, the following will also address how the German criticalness of their own system may be one of the root causes of its international disrepute, rather than the disadvantages within the system itself. In order to understand the German viewpoint in the laws, a brief overview will also be provided on the legal system prior to the introduction of the Prostitutionsgesetz (Prostitution Act) (ProstG) in 2002.³²

One of the initial critiques of the former prostitution laws in Germany, as explained by Philipp S. Fischinger,³³ is that prostitution as the “world’s oldest profession” was essentially, a millennia-old set of double standards which involved a form of legal discrimination. On the one hand, there were the clients, who lived a respectable life as first

²⁶House of Commons Home Affairs Committee (UK), Prostitution, Third Report of Session 2016–17, 15 June 2016, Committee reports are published on the Committee’s website at <www.parliament.uk/homeaffairscom> accessed 28 March, 2017.

²⁷Ibid.

²⁸See: Germany 2002 Prostitution Act (ProstG), Gesetz zur Regelung der Rechtsverhältnisse der Prostituierten, Prostitutionsgesetz of 20th December 2001 (BGBl. I S. 3983); Danna, Daniela. “Trafficking and prostitution of foreigners in the context of the EU countries’ policy about prostitution.” In *NEWR Workshop on Trafficking, Amsterdam*, vol. 26. 2003; Milivojević, Sanja, and Sharon Pickering. “Football and sex: the 2006 FIFA World Cup and sex trafficking.” *Temida* 11, no. 2 (2008): 21-47; Louise Osborne, ‘Why Germany Is Now ‘Europe’s Biggest Brothel’ (the Guardian, 2013) <<https://www.theguardian.com/world/shortcuts/2013/jun/12/germany-now-europes-biggest-brothel>> accessed 28 March 2017.

²⁹Mossman, Elaine. “International approaches to decriminalising or legalising prostitution.” *New Zealand: Ministry of Justice* (2007); Brown, Emma, Imy McCaw, and Raquel Bosó Pérez. “Prostitution Law Reform (Scotland) Evidence.”; Unit, Abuse Studies. “A critical examination of responses to prostitution in four countries: Victoria, Australia; Ireland; the Netherlands; and Sweden.” (2003); Geist, Darren. “Amnesty International’s Empty Promises: Decriminalization, Prostituted Women, and Sex Trafficking.” *Dignity: A Journal on Sexual Exploitation and Violence* 1, no. 1 (2016): 6.

³⁰See for example: Agustín, Laura María. “New research directions: the cultural study of commercial sex.” *Sexualities* 8, no. 5 (2005): 618-631; Jacoby, Wade. “Managing globalization by managing Central and Eastern Europe: the EU’s backyard as threat and opportunity.” *Journal of European Public Policy* 17, no. 3 (2010): 416-432; Macrae, Heather. “The EU as a gender equal polity: Myths and realities.” *JCMS: Journal of Common Market Studies* 48, no. 1 (2010): 155-174; Schneider, Friedrich, and A. T. Kearney. “The shadow economy in Europe, 2013.” *Johannes Kepler Universität, Linz* (2013); Isgro, Kirsten L., Maria Stehle, and Beverly M. Weber. “From sex shacks to mega-brothels: the politics of anti-trafficking and the 2006 soccer World Cup.” *European Journal of Cultural Studies* 16, no. 2 (2013): 171-193.

³¹Kulick, Don. “Sex in the new Europe: The criminalization of clients and Swedish fear of penetration.” *Anthropological theory* 3, no. 2 (2003): 199-218; Hirsh-Ratzkovsky, Roni. “City, Alter City: German Intellectuals writing on Paris, 1900–1933.” *Unpublished PhD thesis. Tel Aviv: Tel-Aviv University* (2011); Katona, Noémi. “Sex work” and “prostitution.” *Solidarity in Struggle* (2016): 89.

³²Germany 2002 Prostitution Act (ProstG).

³³Philipp S Fischinger and Norbert Habermann, §§ 134-138; Anh. Zu § 138: Prostg (1st edn, Sellier - de Gruyter 2011), §1 ProstG.

class citizens, whereas on the other hand, there was the sexual services providing prostitute, who was mostly regarded as a second class citizen and was exposed to social contempt. In Germany, prior to the introduction of the ProstG, this societal double standard was also implemented through the laws of the state: On the one hand, prostitutes were required to pay income and sales tax.³⁴ On the other hand, they were denied access to social security and were categorized as acting in violation of moral standards (*Sittenwidrigkeit*).³⁵

This was also found to be reflected in the German jurisprudence over several decades, starting with the equation of prostitution with ("Ausübung von Gewerbsunzucht") professional criminals in the Bundesverwaltungsgericht (BVerwG - The German Federal Administrative Court).³⁶ This had the effect that prostitution could not fall under the protection of Article 12 of the German Constitution (das Grundgesetz (GG)).³⁷ The Bundesverwaltungsgericht maintained this conception for several decades due to the perception of prostitution being a violation of moral standards and as such *contra bonos mores* and in many aspects socially repugnant.³⁸ This position was also put forward in 1981, when it was held that prostitutes could not be granted free movement rights in accordance with European Economic Community (EEC) law.³⁹ In the area of civil law, the jurisprudence of the Bundesgerichtshof (BGH – The German Federal Court) not only considered prostitution contracts as *contra bonos mores*, but also contracts such as for telephone sex⁴⁰ or for loan agreements for the purpose of financing brothels.⁴¹ In 1973, in the area of criminal law, the BGH even differentiated between whether the victim was an "unblemished woman" or a prostitute, in the determination of the culpability of a rapist.⁴² Around the turn of the millennium, a significant change became apparent in the courts' jurisprudence to a softer attitude towards prostitution. The BVerwG changed its decision regarding the free movement status by revoking its previous decision, which stated that prostitutes were not part of economic life.⁴³ Moreover, the Bundesfinanzhof (BFH – The German Federal Fiscal Court) started to think it possible to reconsider its previous decisions on the taxation of prostitution based on changes in public opinion,⁴⁴ and the BGH distanced itself from its previous decision on telephone sex.⁴⁵ In particular, a decision from the Verwaltungsgericht Berlin (Berlin administrative court) marked a significant change in the attitudes towards prostitution, as it clearly rejected the *contra bonos mores* of sex services as well as the idea of them being a violation of human dignity.⁴⁶

³⁴Ibid.

³⁵See: Wesel, Uwe: Frauen schaffen an, das Patriarchat kassiert ab, NJW (Neue juristische Wochenschrift) 1998, 120-121; Wesel, Uwe: Prostitution als Beruf; in: NJW (Neue juristische Wochenschrift) 1999, S. 2865–2866; Rautenberg, Erardo Cristoforo: Prostitution. Das Ende der Heuchelei ist gekommen! NJW (Neue juristische Wochenschrift) 2002, S. 650 et seq.

³⁶BVerwGE 22, 286 (Decisions of the Federal Administrative Court, vol 22), pp. 286, 289; Monatsschrift für Deutsches Recht (*M.D.R.*) 1996, p. 260; BGH, 30.06.1987 - 4 StR 267/87, NJW (Neue juristische Wochenschrift) 1987, 3209, MDR 1987, 948; BGH (Bundesgerichtshof – German Federal Supreme Court), 20.05.1981 - 2 StR 784/80, NJW (Neue juristische Wochenschrift) 1981, 2071, MDR 1981, 863, NSStZ 1981, 395; BVerwG (Bundesverwaltungsgericht - Federal Administrative Court), 28.06.1995 - 4 B 137.95, NVwZ-RR 1996, 84, BauR 1996, 78, ZfBR 1995, 331 only exception: BVerwG (Bundesverwaltungsgericht - Federal Administrative Court) GewArch (GewerbeArchiv) 1973, 192, 193].

³⁷German Grundgesetz (Basic Law) art 12, Grundgesetz für die Bundesrepublik Deutschland, 23.05.1949 (BGBl. p. 1) last amended 23.12.2014 (BGBl. I S. 2438) m.W.v. 01.01.2015.

³⁸Bénédict Winiger, Essential cases on damage, Digest of European tort law, v. 2 (Berlin: De Gruyter, 2011) 381; Silke Ruth Laskowski, 'New German Prostitution Act-An Important Step to a More Rational View of Prostitution as an Ordinary Profession in Accordance with European Community Law' (2002) *Int'l J. Comp. Lab. L. & Indus. Rel.* 18, 479; Anja Schmidt, 'Die Reform Des Rechts Der Freiwilligen Prostitution' (2015) 48 *Kritische Justiz*.

³⁹BVerwG (Bundesverwaltungsgericht - Federal Administrative Court) 15.07.1980, Az. 1 C 45/77 (Lueneburg) NJW (Neue juristische Wochenschrift) 1981, 1169;

⁴⁰BGH NJW (Neue juristische Wochenschrift) 1998, 2895, 2896.

⁴¹BGHZ (German Supreme Court Reporter for Civil Matters) 67, 119; NJW (Neue juristische Wochenschrift)-RR 1990, 750.

⁴²BGH at Dallinger, MDR 1973, 555.

⁴³BVerwG (German Federal Administrative Court) 18.9.2001, Az. 1 C 17/00 (Mannheim), NVwZ 2002, 339, 340.

⁴⁴BFH (German Federal Fiscal Court) 23.2.2000, Az. X R 142/95 (Schleswig-Holstein), NJW 2000, 2919, 2920.

⁴⁵BGH NJW (Neue juristische Wochenschrift) 2000, 361.

⁴⁶VG Berlin, Urteil v. 01. 12.2000, NJW 2001, 983 (986) - „Widerruf der Gaststätten erlaubnis"

A significant indicator for the change of public opinion in Germany was the result of a representative study used in the rationale of this case. This study looked into the public opinion in Germany as to whether prostitution should be recognised as employment. The study revealed that almost two thirds of the questioned participants thought that prostitution was to be considered to be legal employment.⁴⁷

The significance of this decision was that prior to the ProstG entering into force, the central legal issue surrounding prostitution was the characterisation of prostitution contracts as being *contra bonos mores*.⁴⁸ This meant that even after a prostitute had fulfilled the service, he or she did not have any legally enforceable demands.⁴⁹

The Creation of the Law for the Improvement of the Legal and Social Status of Prostitutes:-

The creation of the law for the improvement of the legal and social status of prostitutes is only comprehensible on the basis of the previously described legal situation. Following calls for reform for many years,⁵⁰ the German legislature started drafting a prostitution code.⁵¹ Despite diverse ideas on how this was to be conducted, all parties of the Bundestag (the German House of Representatives) agreed that the legal status of prostitutes had to be improved.⁵² Apart from the purely legal changes, a primary goal was to send a clear signal that the social double standards were to be removed.⁵³

The German prostitution code is essentially based on three pillars: Under civil law, the prostitute should have an entitlement to the agreed remuneration after the service has been performed.⁵⁴ Secondly, the access of prostitutes to social security should be enabled or facilitated. Finally, the expungement of § 180a Abs. 1 No. 2 StGB (Strafgesetzbuch – The German criminal code) was to give prostitutes the possibility to be legally safeguarded and to voluntarily act as persons in dependent employment in brothels. The objective was to achieve an improvement of working conditions in these types of establishments.⁵⁵

The most significant part of the ProstG (Prostitution Code) is the first of three paragraphs, which states:

⁴⁷Urteil des VG Berlin (Verwaltungsgericht Berlin) (Judgement of the Berlin Administrative Court) NJW (Neue juristische Wochenschrift) 2001, 893, 988; this is in contrast to: Deutscher Bundestag, BT-Drucksache 16/4146, Deutscher Bundestag Drucksache 16/4146 16. Wahlperiode, Bericht der Bundesregierung zu den Auswirkungen des Gesetzes zur Regelung der Rechtsverhältnisse der Prostituierten (Prostitutionsgesetz – ProstG) (Report of the Federal Government on the effects of the law regulating the legal situation of prostitutes) (2007) English translation available online at https://ec.europa.eu/anti-trafficking/sites/antitrafficking/files/federal_government_report_of_the_impact_of_the_act_regulating_the_legal_situation_of_prostitutes_2007_en_1.pdf accessed: 31st March 2017.

⁴⁸See: BGH NJW (Neue juristische Wochenschrift) 1976, 1883, 1884; Hefermehl, in Hans Theodor Soergel, Wolfgang Siebert and Jürgen F Baur, Bürgerliches Gesetzbuch (2nd edn, Kohlhammer 1997).

⁴⁹See: J. von Staudinger and others (n 70) § 138 Rn 453 mwNw; For most current version see: J. von Staudinger and Volker Emmerich, J. Von Staudingers Kommentar zum Bürgerlichen Gesetzbuch, Mit Einführungsgesetz und Nebengesetzen (1st edn, Sellier-de Gruyter 2016).

⁵⁰Deutscher Bundestag, BT-Drucks 14/4456 (German Parliament Print No. 14/4456) – Entwurf eines Gesetzes zur beruflichen Gleichstellung von Prostituierten und anderer sexuell Dienstleistender (Draft law on the professional equality of prostitutes and other sexually employed persons).

⁵¹Deutscher Bundestag, BT-Drucks 14/5958 (German Parliament Print No. 5958) – Entwurf eines Gesetzes zur Verbesserung der rechtlichen und sozialen Situation der Prostituierten (Draft law to improve the legal and social situation of prostitutes).

⁵²See in particular: the critique from the CDU/CSU-fraction: Deutscher Bundestag, BT-Drucks 14/6781, – Entschließungsantrag, Urheber: Fraktion der CDU/CSU (Motion for a resolution, author: Group of the CDU / CSU) 1 et seq; Maria Eichhorn [CDU], Stenographischer Bericht der 196. Sitzung des Deutschen Bundestages (Stenographic report of the 196th meeting of the German Bundestag) 14/196, 19195 et seq.

⁵³See: Remarks of the fraction Bündnis 90/Die Grünen BT-Drucks 14/19171 German Bundestag, 14th election period, 196th meeting, Berlin, Friday, den 19. Oktober 2001, at 8.

⁵⁴Germany 2002 Prostitution Act (ProstG) §§ 1 & 2.

⁵⁵Deutscher Bundestag, (n 51) 6.

§1 When sexual acts have been undertaken for a previously agreed fee, the agreement shall constitute a legally valid claim. The same applies if a person, especially in the context of an employment relationship, keeps the provision of such acts ready against an agreed fee for a certain period of time.⁵⁶

The *ratio legis* of the 2002 code, as evidenced by the draft justification, is solely the legal improvement of the prostitutes' status as opposed to brothel managers.⁵⁷ The two key priorities in the introduction of this provision, were to remove any barriers prostitutes may have accessing social security⁵⁸ and to create a civil claim for prostitutes against clients or brothel and club managers, when sexual acts have been performed, or have been kept ready.⁵⁹ The intention was to help prostitutes in two respects: Legally, by granting prostitutes an enforceable claim due to the repeal of the contractual finding of *contra bonos mores*, as well as factually, by reducing the dependence of prostitutes on procurers or pimps, as prostitutes no longer required the help of so-called "fist law."⁶⁰ This point is crucial in looking back at the provisions in New Zealand, as it becomes apparent that both models base their legalisation on the contractual notion of eliminating contracts for sexual services becoming void on the basis of *contra bonos mores* provisions.⁶¹

Another interesting point to be made is one of the key motivations for the German legislators to argue for the legalisation of sex services. Due to the history of human rights violations during the Third Reich period in relation to the holocaust and other atrocities which took place during the time of the Second World War, the highest legal provision on the basis of which all other laws need to be measured can be found in the first article of the German Basic Law, which essentially is the German constitution, states that human dignity shall be inviolable.⁶² The strong sense that human dignity needs to be ensured and protected at all costs is typical of a German legal philosophy. Accordingly, it is interpreted widely. As a basic human right, the protection of human dignity is a defensive tool to protect citizens from the state. In this sense, the wide understanding of the inviolability of human dignity includes preventing the state from dictating what an individual's dignity entails. Accordingly, part of the protection of dignity entails being able to decide for oneself what is and what is not in accordance with one's own dignity.⁶³ However, although the state understands that it is not able to decide whether providing sexual services is in accordance with one's own understanding of one's dignity, the German government accepts the responsibility to protect its citizens regardless of the way they perceive their own human dignity, and the life choices they make on the basis of this.⁶⁴ Thus, the German model is based mostly on ensuring the human rights' protection of prostitutes, including providing them legal protection from harm, by ensuring they are able to press charges under criminal law, as well as to make civil action claims,⁶⁵ as well as ensuring access to social security provisions. In this sense, prostitutes in Germany are entitled to the same rights as any other worker, including pension rights, holidays, sick leave, maternity

⁵⁶Translated from German: § 1 ProstG: "Sind sexuelle Handlungen gegen ein vorher vereinbartes Entgelt vorgenommen worden, begründet diese Vereinbarung eine rechtswirksame Forderung. Das Gleiche gilt, wenn sich eine Person, insbesondere im Rahmen eines Beschäftigungsverhältnisses, für die Erbringung derartiger Handlungen gegen ein vorher vereinbartes Entgelt für eine bestimmte Zeitdauer bereithält."

⁵⁷Deutscher Bundestag, (n 51) 4, 6.

⁵⁸Bruckert, Chris, and Stacey Hannem, 'Rethinking the prostitution debates: Transcending structural stigma in systemic responses to sex work.' (2013) Canadian Journal of Law and Society 28, no. 1, 43-63.

⁵⁹Germany 2002 Prostitution Act (ProstG) § 1; Bernhard Pichler, *Sex als Arbeit: Prostitution als Tätigkeit im Sinne des Arbeitsrechts* (Hamburg: Disserta Verlag, 2013) 104.

⁶⁰Philipp S Fischinger and Norbert Habermann (n 33) §1 ProstG.

⁶¹See: Germany 2002 Prostitution Act (ProstG) § 12; Part 2. (7), Prostitution Reform Act 2003 (New Zealand).

⁶²German Grundgesetz (Basic Law) Article 1, para. 1: Die Würde des Menschen ist unantastbar (English translation: The human dignity is inviolable).

⁶³BVerfGE (Decision of the German Federal Constitutional Court) 45, 187, 227f. – Lebenslange Freiheitsstrafe; Robert Esser; Hans-Heiner Kühne, *Festschrift für Hans -Heiner Kühne* (Müller, 2013) 97; Barbara Sandfuchs, *Privatheit wider Willen? Verhinderung informationeller Preisgabe im Internet nach deutschem und US-amerikanischem Verfassungsrecht* (Mohr Siebeck, 2015) 126 – 129.

⁶⁴Rahel Gugel, *Das Spannungsverhältnis zwischen Prostitutionsgesetz und Art 3 II Grundgesetz in einer rechtspolitischen Untersuchung* (Münster Lit 2010) 115 et seq.

⁶⁵Raymond, Janice G. 'Prostitution on demand: Legalizing the buyers as sexual consumers.' *Violence against women* 10, no. 10 (2004): 1156-1186; Philipp S Fischinger and Norbert Habermann, (n 33) §1 ProstG.

or parental leave and healthcare.⁶⁶ Again, this human rights emphasis mirrors the same objectives found in the New Zealand model.

The Scope of the German Prostitution Act:-

Due to the fact that the ProstG only consists of three paragraphs, in which the working has deliberately been kept as general as possible, the scope of the code and its effects in practice are far reaching. The way this works in practice will be addressed in the following, in particular, by laying out the scope of application.

In personal terms, the ProstG applies to all genders of prostitutes, including male, female, transsexual and hermaphrodites.⁶⁷ This has been the case since the date the code came into force, in other words since the 1st January 2002.⁶⁸ Due to the short and general nature of the ProstG, it is not surprising that the material scope was not clearly defined within the wording of the act, and, thus, needed to be established by legal interpretation.

According to von Galen, the material scope of the ProstG is to be interpreted narrowly. Hence, “sexual acts” in the sense of the first sentence of the first paragraph are to be understood the same way as the term would be understood in society, meaning the performance of sexual acts with or in front of a client with direct contact.⁶⁹ This interpretation would exclude practices such as telephone sex, the performance of sexual intercourse on stage in front of a public audience, peep-shows, striptease performances or webcam transmissions of a sexual nature.

However, the prevailing opinion appears to distance itself from this narrow interpretation by concentrating on the term “sexual act” in the sense of § 1 S 1 ProstG instead of “prostitution.”⁷⁰ The only counterargument to this is title of the act referring to the “legal relationship of prostitutes” as well as the fact that the legal materials and § 3 of the act consistently refer to the term “prostitution.” However, it is apparent that the drafters did not understand the term prostitution in the same narrow sense as stated above. Instead, in accordance with the draft act, prostitution is understood to mean “the commercial performance of sexual acts.”⁷¹ Furthermore, in accordance with the intentions of the drafters, the reference to “sexual act” will not only apply to sexual acts in the traditional sense between two people, commonly of different genders. It is also argued that the initial objective to protect the status of prostitutes justifies a wide interpretation. Moreover, the fluctuations⁷² in practices, pose difficulties in determining what is included within the definition or not. This difficulty in setting boundaries within the definitions is seen as a further supporting argument for a wide interpretation.⁷³

It appears that the entire scope of the ProstG relies on the term “sexual act.” This commonly refers to “every human act or acquiescence, which is directed at sexually arousing and satisfying another.”⁷⁴ This may include all forms of sexual acts, regardless whether a form of penetration has taken place. The key indicator is intention. Hypothetically, this could even include neutral acts such as gynaecological examinations or gymnastic exercises as long as the sexual intent can be subjectively established. This widens the scope of the definition of “sexual act” in the ProstG beyond the definition of the same term used in German criminal law.⁷⁵ In this sense, even acts such as telephone sex or cyber transmission could be included.⁷⁶ It is assumed, that a wide interpretation is accepted here, due to the fact

⁶⁶ Ibid.

⁶⁷ Götz Schulze, *Die Naturalobligation :Rechtsfigur und Instrument des Rechtsverkehrs einst und heute - zugleich Grundlegung einer zivilrechtlichen Forderungslehre* (Mohr Siebeck, 2008) 548, 549.

⁶⁸ Germany 2002 Prostitution Act (ProstG) § 2.

⁶⁹ Margarete von Galen, *Rechtsfragen Der Prostitution* (1st edn, Beck 2004) n.43.

⁷⁰ J. von Staudinger and others, *J. Von Staudingers Kommentar Zum Bürgerlichen Gesetzbuch, Mit Einführungsgesetz Und Nebengesetzen* (1st edn, Sellier-de Gruyter 2003) § 138 Rn 452; Franz Jürgen Sacker, *Münchener Kommentar Zum Bürgerlichen Gesetzbuch* (1st edn, CH Beck 2015) Rn 3 et seq.

⁷¹ Deutscher Bundestag, (n 51) 1.

⁷² For example: Someone may book a private striptease which could include physical touching, ranging from light to actual sexual intercourse.

⁷³ Philipp S Fischinger and Norbert Habermann, (n 33) §1 ProstG, Rn 5.

⁷⁴ Ibid.

⁷⁵ See: Perron and Eisele in Adolf Schönke and others, *Strafgesetzbuch* (1st edn, Beck 2010) § 184 Rn 6.

⁷⁶ Christian Armbrüster, Kurt Rebmann and Franz Jürgen Sacker, *Allgemeiner Teil (Auszug)* (1st edn, Beck 2003) Rn 4 in Franz Jürgen Sacker, Christian Armbrüster and Kurt Rebmann, *Münchener Kommentar Zum Bürgerlichen Gesetzbuch* (1st edn, Beck 2003).

that this is a matter of civil law, and is, thus, not connected to punitive evidence.⁷⁷ This also explains the fact that the mere acquiescence is sufficient without any requirement to have actively performed something.⁷⁸ Thus, the passive participation in sexual acts is also included.

Despite these differences between the civil and criminal legal realms, it is debated whether the definition of a "sexual act" in civil law requires an element of significance, as is the case in § 184g of the German Criminal Code (hereinafter referred to as StGB).⁷⁹

It is the German view, that transferring criminal law concepts and principles in such a manner would mix two things that are unrelated.⁸⁰ On the one hand, § 184g no. 1 StGB deals with the question whether actions against a protected interest, good or right are insignificant.⁸¹ On the other hand, it is merely relevant for the scope of the ProstG to establish whether the sexual activity in question was of "some weight."⁸² Furthermore, the fact that § 184 no 1 StGB is limited to, in the words of the paragraph, "this law," strengthens this argument. The limitation of culpability can be explained by the fact that the criminal law is only *ultima ratio*, whereas the objective of the ProstG is to establish the greatest possible protection for the sexual services providers, regardless of the nature of the service provided.⁸³ Finally, as the intention of the action is to satisfy the clients' sexual desire or arousal is a highly personal and subjective matter, it is argued that this cannot be determined on the basis of objective criteria, which ultimately means that the application of the ProstG is not dependent on establishing significance of the performed service.⁸⁴

Prostitutes as Employees:-

A point that is quite different between the German and the New Zealand models of regulating prostitution relate to the employee status of prostitutes. In this sense, although New Zealand's provisions may refer to prostitutes as being employed, this refers merely to the situations in which prostitutes work within brothels, or out of brothels, in the sense, that the brothels distribute work to them. However, as in accordance to Section 16 of the PRA, brothel operators are unable to sanction workers for lateness, unprofessional conduct or other misdemeanours,⁸⁵ as well as the fact that prostitutes are required to pay a "shift fee" to the brothels, regardless of how much business comes in,⁸⁶ and apart from this are able to keep their proceeds, the working relationship would be classified as self-employed according to EU standards.⁸⁷ These practices are often criticised in the New Zealand model, as it often means that prostitutes will potentially lose money on slow nights.⁸⁸ In contrast, in Germany it is possible for prostitutes to work as employees in brothels, which means that they receive a fixed salary. In these circumstances, there is no

⁷⁷ Ibid.

⁷⁸ Dirk Looschelders, Dirk Olzen and Gottfried Schiemann, J. Von Staudingers Kommentar zum Bürgerlichen Gesetzbuch (1st edn, 2015) Anhang zu § 138 Rn 6.

⁷⁹ Christian Armbrüster, Kurt Rebmann and Franz Jürgen Sacker (n 76); A contrasting approach can be found in: Margarete von Galen, (n 69) 40 et seq.

⁸⁰ Philipp S Fischinger and Norbert Habermann, (n 33) §1 ProstG.

⁸¹ See: Perron and Eisele in Adolf Schönke and others (n 75) § 184, 16.

⁸² Philipp S Fischinger and Norbert Habermann, (n 33) §1 ProstG.

⁸³ Dirk Looschelders, Dirk Olzen and Gottfried Schiemann, (n 78) Rn 5.

⁸⁴ Philipp S Fischinger and Norbert Habermann, (n 33) §1 ProstG, Rn. 6.

⁸⁵ See Section 16 Prostitution Reform Act 2003 (New Zealand).

⁸⁶ Jan Jordan, *The Sex Industry in New Zealand: A Literature Review* (Ministry of Justice (New Zealand), 2005) 29; Mossman, Elaine (n 29); Barrington, Jane. 'Shapeshifting: prostitution and the problem of harm: a discourse analysis of media reportage of prostitution law reform in New Zealand in 2003.' (2008) PhD diss., Auckland University of Technology; Cheryl Brunton; Lisa Fitzgerald; Catherine Healy; Gillian Abel, *Taking the crime out of sex work: New Zealand sex workers' fight for decriminalisation* (Policy Press, 2010) 119 et seq.

⁸⁷ Werner, Arndt, Johanna Gast, and Sascha Kraus. "The effect of working time preferences and fair wage perceptions on entrepreneurial intentions among employees." (2014) *Small Business Economics* 43, no. 1, 137-160; Cruz, Katie, Kate Hardy, and Teela Sanders. 'False Self- Employment, Autonomy and Regulating for Decent Work: Improving Working Conditions in the UK Stripping Industry.' (2016) *British Journal of Industrial Relations*; Nyombi, Chrispas. 'A Response to the Challenges Posed by the Binary Divide between Employee and Self-employed.' *International Journal of Law and Management* 57, no. 1 (2015): 3-16; Nigel G Foster, Foster on EU law (Oxford University Press, 2015) 315 et seq.

⁸⁸ Charles B Fields and Richter H Moore, *Comparative And International Criminal Justice* (1st edn, Waveland Press 2005) 73.

contractual relationship between the prostitute and the clients, but rather between the clients and the brothel owners.⁸⁹

The contract between the prostitute and the brothel will herein be referred to as a brothel contract. In accordance with § 1 S2 of the ProstG, the prostitute is considered to be keeping his or her services ready to provide these to clients for a particular amount of time. In return the prostitute will receive his or her “previously agreed salary.” However, it is clear from § 3 ProstG that the legislator aimed at only granting brothel owners a limited right of direction.⁹⁰ This, however, only applies to the place and time of the acts to be undertaken and not to the selection of clients and the nature of the services provided. In this sense, the prostitutes are not employed for the specific services, but rather for keeping his or her services ready for a particular time and at a particular place.⁹¹ This allows for employed prostitutes to also have the right to reject any clients without losing any claim towards the brothel owner in relation to salary.⁹² Similarly to the classification of the prostitution contract, the brothel contract will also be classified as a services contract. The only difference is that this contract is not unilateral and falls within the scope of a regular services contract. This is based on the fact that the prostitute is only contracted to keep the services ready for provision, and thus does not directly apply to the actual provision of the sexual act provisions per se.⁹³

However, the brothel owners’ right of direction, even if it is only limited, can only be explained constructively if the contractual partners have certain obligations. These obligations are seen in the keeping ready of the services. This obligation would not fall under the human dignity protection of the German constitution, as it cannot be equated to actually providing the sexual act itself.⁹⁴ This legal obligation can, however, not be enforced by the brothel owner, as a prostitute is able to terminate the contract at any point without notice.⁹⁵ Nevertheless, in order for the contractual relationship to be categorised as an employment contract, which brings with it the entitlement to social security in accordance with § 7 Abs. 1 S 1 SGB IV (Sozialgesetzbuch IV – the German Social Code: book IV), obligations need to be assumed, despite the fact that a brothel owner cannot enforce these.⁹⁶

If a brothel contract constitutes a contract for employment, the normal individual and collective employment laws apply. This includes that the contract can only be limited by § 14 TzBfG,⁹⁷ that the prostitute has holiday entitlements, an entitlement to continued payment in case of illness (sick leave), a protection against dismissal as well as the right to form a union.⁹⁸

In principle, although the brothel contract is not specifically referred to in the ProstG, it is widely accepted that it will not be considered *contra bonos mores*, similarly to prostitution contracts. However, this will only be applicable within reason, meaning that brothel contracts may, nevertheless, be considered *contra bonos mores* in exceptional circumstances, for instance, if the contract were to result in the exploitation of the prostitute.⁹⁹

A significant advantage in contrast to the New Zealand model is that under a brothel contract, prostitutes are merely contractually obliged to keep themselves ready to provide sexual acts.¹⁰⁰ This means that there is no due

⁸⁹ Philipp S Fischinger and Norbert Habermann, (n 33) §1 ProstG, Rn. 48-53.

⁹⁰ Deutscher Bundestag, (n 51) 6.

⁹¹ Philipp S Fischinger and Norbert Habermann, (n 33) §1 ProstG, rn. 52; Margarete von Galen(n 69) Rn. 132 et seq.

⁹² Ibid § 2 ProstG, Rn. 15.

⁹³ Margarete von Galen (n 69) Rn. 132 et seq.

⁹⁴ Ibid.

⁹⁵ Deutscher Bundestag, (n 51).

⁹⁶ J. von Staudinger, George Annub and Dieter Reuter, J. Von Staudingers Kommentar Zum Bürgerlichen Gesetzbuch Mit Einfuhrungsgesetz Und Nebengesetzen (1st edn, Sellier 2011) §§ 138, 586, 50.

⁹⁷ Gesetz über Teilzeitarbeit und befristete Arbeitsverträge (Teilzeit- und Befristungsgesetz -TzBfG) (Act on part-time work and temporary contracts) 21st December 2000 (BGBl. I S. 1966)

⁹⁸ Christian Armbrüster, Kurt Rebmann and Franz Jürgen Sacker, (n 76).

⁹⁹ Ibid.

¹⁰⁰ Margarete von Galen, (n 69) Rn 147 et seq.

performance of the actual provision of sexual acts and prostitutes will be owed remuneration, even when sexual acts have not been performed.¹⁰¹

The question that naturally follows is how brothel owners' interests can be protected from prostitutes declining to perform sexual acts persistently or at least in an unreasonably large number of cases. The answer to this question can be found in the understanding of the term "keeping ready." It is understood that to keep something ready involves an internal willingness to serve suitable clients who are willing to pay for the service.¹⁰² According to von Galen,¹⁰³ this requirement of willingness extends to the prostitutes' appearance as well, meaning that it requires prostitutes to present themselves in an externally sufficiently attractive manner. The employment nature of the work relationship between prostitutes and brothel owners also assumes instruction rights of owners or managers in relation to prostitutes' appearances and clothing.¹⁰⁴ However, this does not mean that a prostitute can be forced against her will to follow these requirements. Instead, it merely means that non-compliance will be understood as an absence of internal willingness,¹⁰⁵ which in the worst case will result in a termination of the employment contract.¹⁰⁶

Despite being regulated under German employment law, the termination of brothel contracts involves a number of exceptions due to the nature of the work. In this sense, brothel contracts may be terminated mutually by agreement, or in the event of the death of the prostitute.¹⁰⁷ Furthermore, it has been emphasised that prostitutes, as they cannot be forced into providing sexual services against their will, have the right to terminate brothel contracts at any time without giving previous notice.¹⁰⁸ However, there is no corresponding termination right for brothel owners. Thus, without a qualifying reason for an extraordinary termination,¹⁰⁹ it can be assumed that the ordinary termination provisions apply in accordance with § 620 BGB. Accordingly, a brothel owner would have to give a prostitute between 2 weeks and 7 months' notice, depending on the length of the brothel contract.¹¹⁰

Finally, a point worth mentioning in relation to the current effects of the ProstG in Germany involve the way damages are regulated. Accordingly, claims for damages in prostitution usually follow the same *culpa in contrahendo* rules as in any other contractual relationship.¹¹¹ However, an area which deserves particular attention in the issue of damages related to parties to the contract being infected with a disease by the other party. The resulting claim in such a scenario would be derived from § 280 s. 1 in conjunction with § 241 s. 2 BGB, due to a breach of the duty of care towards the other party. In the event that the prostitute infected the client, this protected interest is the client's integrity interest. As there are no specific provisions within the ProstG, the majority opinion is that prostitutes do not fall under any general limitation of liability. The reason for this is that the non-use of barrier protection, such as condoms, is regarded as negligence, unless the surrounding circumstances of the risk of infection was known to the client and he or she nevertheless requested the omission of said protective measures.¹¹²

Understanding the German move to stricter Regulationism:-

In recent years, the German government has worked towards drafting new legislation targeted as implementing further regulation of prostitution, which will enter into force on 1st July, 2017.¹¹³

¹⁰¹ Dirk Looschelders, Dirk Olzen and Gottfried Schiemann, (n 78) Rn 9.

¹⁰² Philipp S Fischinger and Norbert Habermann, (n 33) §1 ProstG, rn. 52.

¹⁰³ Margarete von Galen (n 69) Rn 148.

¹⁰⁴ Ibid.

¹⁰⁵ Philipp S Fischinger and Norbert Habermann, (n 33) §2 ProstG, Rn 56.

¹⁰⁶ Ibid.

¹⁰⁷ However, not in the event of the death of the brothel owner, in accordance with § 672 BGB.

¹⁰⁸ Deutscher Bundestag, (n 51) 4 et seq.

¹⁰⁹ § 626 BGB.

¹¹⁰ See: § 622 BGB - Kündigungsfristen bei Arbeitsverhältnissen (Section 622 Notice periods in the case of employment relationships).

¹¹¹ Philipp S Fischinger and Norbert Habermann, (n 33) §1 ProstG, Rn. 45; Bergmann 2003 JR, 2003, 270, 275.

¹¹² Ibid. Rn 47.

¹¹³ Committee on the Elimination of Discrimination against Women (CEDAW) Consideration of reports submitted by States parties under article 18 of the Convention on the Elimination of All Forms of Discrimination against Women, List of issues and questions in relation to the combined seventh and eighth periodic reports of Germany, Sixty-sixth session, 13 February-3 March 2017, CEDAW/C/DEU/Q/7-8/Add.1, available online at: <http://www.institut-fuer-menschenrechte.de/fileadmin/user_upload/PDF-

In order to understand the planned changes to the current laws, it is important to understand the issues and gaps revealed from the initial Prostitution Code (ProstG).

When the ProstG was drafted, a key philosophical stance towards prostitution was that it was a given, which realistically had to be dealt with regardless of moral attitudes.¹¹⁴ Accordingly, the Act sought neither to end prostitution, nor to augment its status.¹¹⁵ Instead, a particular focus was directed towards the improvement of prostitutes' working conditions, in order to support people voluntarily earning their living through the provision of sex services.¹¹⁶

Thus, the Act sought to enhance prostitutes' legal as well as social situations, by removing the negative effects of prostitution having been classified as immoral, and as such invalid, legal transactions,¹¹⁷ as well as by facilitating access to social insurance.¹¹⁸ The idea was for the ProstG to assist in reducing prostitution-related crimes as well as making it easier for people working in prostitution to exit the industry.¹¹⁹ A crucial point, however, was that prostitution was not "job like any other," in particular, due to strict legal protection provisions related to the rights of prostitutes to sexual self-determination.¹²⁰

A key rationale here is the notion that the state is not responsible for the protection of people from their own life-choices, especially when these have been a product of free self-determination. In this context, freedom as part of the right to sexual self-determination proscribes that people should be allowed to freely choose whether, how and when to engage in sexual acts.¹²¹

The basis for this is found, in particular in the fact that, today, prostitution, as an occupation aimed at sustaining one's livelihood, is protected by Article 12 s. 1 GG.¹²² As an occupation, people working in the area need to be protected from harm in the same way as anyone else. The German government recognises the vulnerability of people working in prostitution. Thus, it has been emphasised that the state needs to combat any prostitution-related

Dateien/Pakte_Konventionen/CEDAW/cedaw_state_report_germany_7_8_2015_loi_reply_en.pdf> accessed 31 March, 2017.

¹¹⁴Deutscher Bundestag, (n 7) 9, English translation available online at: <https://ec.europa.eu/anti-trafficking/sites/antitrafficking/files/federal_government_report_of_the_impact_of_the_act_regulating_the_legal_situation_of_prostitutes_2007_en_1.pdf> accessed: 31 March 2017.

¹¹⁵ Ibid.

¹¹⁶Pajnik, Mojca. 'Reconciling paradigms of prostitution through narration.' (2013) *Društvena istraživanja* 22, no. 2, 257-276; Bruckert, Chris, and Stacey Hannem (n 58) 43-63.

¹¹⁷Venla Roth, Defining human trafficking and identifying its victims : a study on the impact and future challenges of international, European and Finnish legal responses to prostitution-related trafficking in human beings (MartinusNijhoff Publishers, 2012) 36, 37; Kavemann, Barbara, and Heike Rabe. 'The Act Regulating the Legal Situation of Prostitutes—Implementation, Impact, Current Developments: Findings of a Study on the Impact of the German Prostitution Act.' (2007) *Berlin: Sozialwissenschaftliches FrauenForschungsInstitut eV*, <www.cahrv.uni-osnabrueck.de/reddot/BroschuereProstGenglisch.pdf> accessed: 31 March, 2017.

¹¹⁸Laskowski, Silke Ruth. 'The New German Prostitution Act-An Important Step to a More Rational View of Prostitution as an Ordinary Profession in Accordance with European Community Law' (2002) *Int'l J. Comp. Lab. L. & Indus. Rel.* 18, 479; Dodillet, Susanne. 'Cultural clash on prostitution: Debates on prostitution in Germany and Sweden in the 1990s.' (2004) In *First Global Conference: Critical Issues in Sexuality, Salzburg, Austria, October*.

¹¹⁹Elfriede Steffan, Prof.Dr. Barbara Kavemann, Tzvetina Arsova Netzelmann & Prof.Dr. Cornelia Helfferich, 'Abschlussbericht der wissenschaftlichen Begleitung zum Bundesmodellprojekt Unterstützung des Ausstiegs aus der Prostitution' (translation into English: Final report of the scientific accompaniment to the federal model project Supporting the withdrawal from prostitution) (2015) Federal Ministry of Family Affairs, Senior Citizens, Women and Youth [Germany] <<https://www.bmfsfj.de/blob/95446/b1f0b6af91ed2ddf0545d1cf0e68bd5e/unterstuetzung-des-ausstiegs-aus-der-prostitution-langfassung-data.pdf>> accessed: 31st March, 2017.

¹²⁰Greggor Mattson, *The cultural politics of European prostitution reform: governing loose women* (Palgrave Macmillan, 2016) 101.

¹²¹Sick, Brigitte, and Joachim Renzikowski. 'Lücken beim Schutz der sexuellen Selbstbestimmung aus menschenrechtlicher Sicht.' (2015) In *Über allem: Menschlichkeit* 928-942. Nomos Verlagsgesellschaft mbH & Co. KG.

¹²²Deutscher Bundestag (n 114).

crimes, in particular socially damaging human rights violations, such as forced prostitution, trafficking in human beings, child prostitution and other human rights violations by all available means provided by law.¹²³ These means include criminal prosecution, monitoring, preventive and repressive measures, and victim protection, including the provision of assistance to victims.¹²⁴

In line with the German gender equality framework, the Federal Government has recognised a need to take countermeasures against the gender equality issues found within prostitution.¹²⁵ This includes the high proportion of women working in the industry in contrast to men, as well as considerations of the nature of the industry, which can be “physically and psychologically demanding, risky and dangerous.”¹²⁶ Thus, while accepting the free choice of adults to work in this industry, it is understood that any threats, including poor hygiene conditions, as well as any other threats to prostitutes’ health, safety and overall wellbeing need to be addressed. In particular, it is important to ensure the voluntariness of the decision to work in prostitution. Accordingly, it needs to be ensured that the social reality of many prostitutes finding themselves in social and psychological situations, in which the ability to freely and autonomously choose to practice this occupation can be called into question, are counteracted. The social responsibility taken up by the Federal Government in this respect include providing possibilities for prostitutes to “earn their living by other means and to prevent them drifting into dependencies which make prostitution appear to be the lesser evil or an acceptable way out.”¹²⁷ These objectives are sought to be achieved through state-funded assistance programmes as well as labour market and education policy in order to offer viable alternatives.¹²⁸

Although the legal measures taken by Germany in 2002 strongly resemble the legislation introduced in New Zealand, the German Federal Government appears to have been more critical in their 5-year review of their prostitution laws. The New Zealand government undertook a four-stage research study, which, despite conducting a quantitative element by means of a survey, which was evaluated on the basis of statistical estimations, sought to avoid significant evaluation based on these statistics within their report, and instead focussed on the predominantly qualitative stages of the study. This consisted of interviews conducted with 58 voluntary participants who worked in prostitution.¹²⁹ This evaluation can be criticised on the basis, that any dark figures would not have been revealed, as it can be assumed that vulnerable prostitutes who had not been positively affected by the PRA, would not necessarily be able to take part in such studies. Moreover, New Zealand’s evaluation of the PRA, focussed more on the black letter law achievements. This means that the success of the PRA was predominantly based on the human rights protection the law provided in theory rather than in practice. In this sense, the report concludes that

“[t]he PRA has been in force for five years. During that time, the sex industry has not increased in size, and many of the social evils predicted by some who opposed the decriminalisation of the sex industry have not been experienced. On the whole, the PRA has been effective in achieving its purpose, and the Committee is confident that the vast majority of people involved in the sex industry are better off under the PRA than they were previously. However, progress in some areas has been slower than may have been hoped. Many sex workers are still vulnerable to exploitative employment conditions, and there are still reports of sex workers being forced to take clients against their will.”¹³⁰

Despite the negative findings that the PRA’s success had been slower than intended and that there still were many exploitative practices, it is significant that New Zealand found the PRA to be a success. In this sense, they justified this finding by stating that “[i]t is a truism that traditions and attitudes developed over many years cannot be

¹²³ Ibid.

¹²⁴ European Parliament, Directorate General For Internal Policies Policy Department C: Citizens' Rights And Constitutional Affairs, 'Sexual Exploitation And Prostitution And Its Impact On Gender Equality' (2014) 40-43, <[http://www.europarl.europa.eu/RegData/etudes/etudes/join/2014/493040/IPOL-FEMM_ET\(2014\)493040_EN.pdf](http://www.europarl.europa.eu/RegData/etudes/etudes/join/2014/493040/IPOL-FEMM_ET(2014)493040_EN.pdf)> accessed 31 March 2017.

¹²⁵ Deutscher Bundestag (n 114).

¹²⁶ Ibid.

¹²⁷ Ibid 12.

¹²⁸ Ibid.

¹²⁹ New Zealand Government, Ministry of Justice, (n 16).

¹³⁰ Ibid at 168

changed overnight”¹³¹ and concluded that the committee did not consider it to be necessary to undertake further reviews of the operation of the PRA at that stage.”¹³²

The optimism in the theoretical, legal application of the legislation in contrast to some of the more negative findings in practice may explain why the evaluation of the ProstG by the German government was less optimistic in its findings. In this sense, the German Federal government took a more pragmatic stance in its evaluation. It explained that despite the unreliability of statistical data, the legislators had to assume that the ProstG had only achieved its objectives to a limited extent. As previously explained, the objectives of the ProstG included to eliminate the legal immorality of prostitution, to enable prostitutes to take legal action, not only under criminal law, but also to ensure they could take civil measures, such as to enforce payment, to ensure access to social insurance, to combat prostitution-related crime, to facilitate prostitutes to exit prostitution, and to better working conditions.¹³³

Accordingly, the German government stressed that the legislation itself ensured that all the objectives were met legally, the situation in practice revealed that not all people working in prostitution were making use of these legal provisions. In particular, it was noted that there were no indications of the legislation having worsened the situation in prostitution, nor of human trafficking for the purpose of sexual exploitation as well as other prostitution related crimes having been enhanced or made more difficult to prosecute through the legalisation of prostitution, as initially feared by critics of the ProstG. However, the improvements through the legislation were still tentative, and despite some indications of improvements, these suggested that the ProstG proved rather a long-term rather than a short-term solution to the situation of prostitution in practice.¹³⁴ These final points essentially demonstrate the same kinds of findings as the report from the New Zealand Government. It appears that the most significant difference is that the New Zealand Government interpreted these findings as a success, whereas the German government interpreted their findings as an indication to take further action.

It was recognised by the Government, that the particularly liberal approach taken, which based on the mere three paragraphs, was in fact less regulated than the New Zealand model, only represented a limited approach, and as such could only be taken as a first step towards achieving the government’s objectives regarding the regulation of prostitution.¹³⁵

The Introduction of the Act on the Protection of Persons engaged in Prostitution:-

Following the evaluation of the ProstG in 2007, the Act on the Protection of Persons engaged in Prostitution (short: Prostituiertenschutzgesetz - ProstSchG) was issued as the first Article of the Law on the regulation of Prostitution and the Protection of Persons engaged in Prostitution (Gesetz zur Regulierung des Prostitutions sowie zum Schutz von in der Prostitution tätigen Personen - ProstSchGEG) on the 21st October 2016 and will enter into force on 1st July 2017.¹³⁶

The aim of the ProstSchG is to improve the situation for prostitutes by strengthening their right to self-determination as well as ensuring the protection from exploitation, coercion, violence and trafficking in human beings.¹³⁷ The legal basis also serves to improve the legal instruments relating to the monitoring of the prostitution industry in order to tackle any threats therein.¹³⁸

¹³¹ Ibid.

¹³² Ibid.

¹³³ Deutscher Bundestag (n 114).

¹³⁴ Ibid.

¹³⁵ Ibid.

¹³⁶ Article 7, Gesetz zur Regulierung des Prostitutionsgewerbes sowie zum Schutz von in der Prostitution tätigen Personen.

¹³⁷ Deutscher Bundestag, Drucksache 18/8556, Entwurf eines Gesetzes zur Regulierung des Prostitutionsgewerbes sowie zum Schutz von in der Prostitution tätigen Personen (Draft Law On The Regulation Of Prostitution And The Protection Of Persons Engaged In Prostitution)' (Bundesanzeiger Verlag GmbH 2016).

¹³⁸ Ibid.

The act is based on two key pillars, namely, the regulation of the prostitution industry, as well as the protection of persons engaged in prostitution.¹³⁹

The first pillar primarily involves introducing an authorization requirement within the prostitution industry for brothels and brothel-like establishments, as well as all other forms of commercial prostitution.¹⁴⁰ Accordingly, operators will need to undergo a personal reliability test. This will also apply to any persons acting as a legal agent or has been entrusted with any managerial or security related duties.¹⁴¹ In order to obtain authorisation, operators will be required to draw up an operating concept and comply with the minimum requirements for the equipment within the business premises.¹⁴² A significant milestone, according to the German Federal Ministry of Family Affairs, Senior Citizens, Women and Youth, is the newly created obligations to ensure acceptable working conditions.¹⁴³

The obligatory checks introduced by the new regulation will ensure, for instance, that people who have previously been convicted of human trafficking offences will no longer be permitted to operate brothels.¹⁴⁴ It will also ensure that there is an evaluation of reasonableness of operating concepts and business models prior to the start-up of prostitution businesses. This can be interpreted as a direct response to the consequences of capitalist market forces in Germany following the particularly liberal legalisation of prostitution, which resulted in new business models, such as “flat rate brothels” being introduced, as well as an increasing demand for harmful or dangerous sex acts, such as group sex” or “gangbangs.”¹⁴⁵ Any breaches would result in the operators being subject to sanctions, such as the loss of authorisation as well as penalties.¹⁴⁶

The second pillar, covering the protection of persons engaged in prostitution, includes the implementation of a personal notification obligation as well as health advice provisions for prostitutes, which are to be repeated at regular intervals.¹⁴⁷ The idea is to provide prostitutes with additional support and to ensure that prostitutes are actually able to exercise their rights and to work freely in a self-determined manner.¹⁴⁸

¹³⁹BundesministeriumfürFamilie, Senioren, Frauen und Jugend (BMFSFJ) Federal Ministry of Family Affairs, Senior Citizens, Women and Youth [Germany], 'RahmenbedingungenFür Die Legale Prostitution Schaffen (Framework For Legal Prostitution)' (2016) <<https://www.bmfsfj.de/bmfsfj/aktuelles/alle-meldungen/rahmenbedingungen-fuer-die-legale-prostitution-schaffen/83928>> accessed 31 March 2017.

¹⁴⁰Ibid.

¹⁴¹§ 15 Gesetz zur Regulierung des Prostitutionsgewerbes sowie zum Schutz von in der Prostitution tätigen Personen.

¹⁴²§ 12 Gesetz zur Regulierung des Prostitutionsgewerbes sowie zum Schutz von in der Prostitution tätigen Personen.

¹⁴³BundesministeriumfürFamilie, Senioren, Frauen und Jugend (BMFSFJ) Federal Ministry of Family Affairs, Senior Citizens, Women and Youth [Germany], 'RahmenbedingungenFür Die Legale Prostitution Schaffen (Framework For Legal Prostitution)' (2016) <<https://www.bmfsfj.de/bmfsfj/aktuelles/alle-meldungen/rahmenbedingungen-fuer-die-legale-prostitution-schaffen/83928>> accessed 31 March 2017, commenting on § 18 Gesetz zur Regulierung des Prostitutionsgewerbes sowie zum Schutz von in der Prostitution tätigen Personen.

¹⁴⁴BundesministeriumfürFamilie, Senioren, Frauen und Jugend (BMFSFJ) Federal Ministry of Family Affairs, Senior Citizens, Women and Youth [Germany], 'RahmenbedingungenFür Die Legale Prostitution Schaffen (Framework For Legal Prostitution)' (2016) <<https://www.bmfsfj.de/bmfsfj/aktuelles/alle-meldungen/rahmenbedingungen-fuer-die-legale-prostitution-schaffen/83928>> accessed 31 March 2017.

¹⁴⁵Mathieson, Ane, Easton Braman, and Anya Noble. "Prostitution Policy: Legalization, Decriminalization and the Nordic Model." (2016) *Seattle Journal for Social Justice* 14, no. 2, 10, 395; Stephens, Monica. "Gender and the GeoWeb: divisions in the production of user-generated cartographic information." (2013) *GeoJournal* 78, no. 6, 981-996.

¹⁴⁶Section 6, § 33 Gesetz zur Regulierung des Prostitutionsgewerbes sowie zum Schutz von in der Prostitution tätigen Personen.

¹⁴⁷BundesministeriumfürFamilie, Senioren, Frauen und Jugend (BMFSFJ) Federal Ministry of Family Affairs, Senior Citizens, Women and Youth [Germany], 'RahmenbedingungenFür Die Legale Prostitution Schaffen (Framework For Legal Prostitution)' (2016) <<https://www.bmfsfj.de/bmfsfj/aktuelles/alle-meldungen/rahmenbedingungen-fuer-die-legale-prostitution-schaffen/83928>> accessed 31 March 2017.

¹⁴⁸Deutscher Bundestag, Drucksache 18/8556, Entwurf eines Gesetzes zur Regulierung des Prostitutionsgewerbes sowie zum Schutz von in der Prostitution

Thus, from July 2017, prostitutes will receive a personal information and consultation session at registration, during which they will be informed about their rights and receive important information on available support,¹⁴⁹ such as the Germany-wide "Violence against women support Hotline."¹⁵⁰ The provided services and information are available in currently 17 languages,¹⁵¹ however, § 7 (3) ProstSchG indicates that the provided information should be provided in a language understood by the prostitute, which may require further languages to be made available. Moreover, prostitutes are to be given the opportunity to discuss any health-related aspects of their work in confidentially in one-to-ones.¹⁵²

It has been argued that the new stricter regulation is a step backward from the ideal decriminalised regulation of prostitution, with merely few regulatory measures, such as age-restrictions, in particular from a number of advocacy groups for prostitution, such as Hydra,¹⁵³ BesD,¹⁵⁴ BSD¹⁵⁵ and the Doña Carmen eV.¹⁵⁶ The main criticism is based on the argument that any restrictions, such as imposed by the obligatory registration and authorisation for businesses, will unreasonably interfere with and potentially limit prostitutes' right to self-determination and other basic rights protected in the German constitution. However, the German Federal government has argued that these provisions are necessary in order to ensure the self-determination of vulnerable prostitutes and the protection of victims. Accordingly, it can be argued that a balancing of these competing interests, namely the limitation of the general right to self-determination and the rights of vulnerable prostitutes and exploitation victims to be protected from harm, justifies the implementation of such measures.¹⁵⁷

Conclusion:-

It has been revealed that the supposed decriminalisation approach in New Zealand is in fact a regulationist approach much like the German one. In fact, in terms of the amount of provisions, it became apparent that the German approach was less regulated than the New Zealand model. Both approaches targeted the elimination of invalidity of contracts due to prostitution being *contra bonos mores*, as their primary provision. Moreover, both systems appear to

TätigenPersonen (Draft Law On The Regulation Of Prostitution And The Protection Of Persons Engaged In Prostitution)' (Bundesanzeiger Verlag GmbH 2016) 1.

¹⁴⁹ §§7-9, Gesetz zur Regulierung des Prostitutionsgewerbes sowie zum Schutz von in der Prostitution tätigen Personen.

¹⁵⁰ 'The Violence Against Women Support Hotline – Support And Counselling For Women' (Hilfetelefon.de, 2017) <<https://www.hilfetelefon.de/en.html>> accessed 31 March 2017.

¹⁵¹ 'Beratung In 17 Sprachen: Hilfetelefon' (Hilfetelefon.de, 2017) <<https://www.hilfetelefon.de/das-hilfetelefon/beratung/beratung-in-17-sprachen.html>> accessed 31 March 2017.

¹⁵² §8 Gesetz zur Regulierung des Prostitutionsgewerbes sowie zum Schutz von in der Prostitution tätigen Personen.

¹⁵³ HYDRA e.V., 'Information About The 'Prostitution Protection Law', That Will Go Into Effect Nationwide Starting 01.07.2017' (2017) <http://www.hydra-berlin.de/sexarbeit_von_a_bis_z/infos_zum_neuen_gesetz/information_about_the_new_law/> accessed 31 March 2017.

¹⁵⁴ Berufsverband erotische und sexuelle Dienstleistungen e.V. (BesDe.V.) - (English Translation: Professional association erotic And sexual services e.V.), 'BesD Unterstützt Verfassungsbeschwerde Gegen Das Prostschg (English Translation: BesD Supports Constitutional Complaints Against The Prostschg)' (2017) <<http://berufsverband-sexarbeit.de/besd-unterstuetzt-verfassungsbeschwerde-gegen-das-prostschg/>> accessed 31 March 2017.

¹⁵⁵ Bundesverband Sexuelle Dienstleistungen e.V. (English Translation: Federal Association for Sexual Services e.V.), 'Stellungnahme Zum Referentenentwurf Des Bundesministeriums Für Familie, Senioren, Frauen Und Jugend: „Entwurf Eines Gesetzes Zur Regulierung Des Prostitutionsgewerbes Sowie Zum Schutz Von In Der Prostitution Tätigen Personen,, (Statement To The Draft Paper By The Federal Ministry For Family Affairs, Senior Citizens, Women And Youth: "Draft Law For The Regulation Of The Prostitution Industry As Well As For The Protection Of Prostitution Persons")' (2015) <<http://www.bsd-ev.info/publikationen/index.php>> accessed 31 March 2017.

¹⁵⁶ Doña Carmen e.V., '25 Gute Gründe Für Ein Klares NEIN Zur Geplanten „Erlaubnispflicht Für Prostitutionsgewerbe' (English Translation: For A Clear NO To The Planned , Permission For Prostitutes')' (2016) <<http://www.donacarmen.de/wp-content/uploads/25-gute-Gr%C3%BCnde-gegen-Erlaubnispflicht.pdf>> accessed 31 March 2017.

¹⁵⁷ Dunn, Michael C., Isabel CH Clare, and Anthony J. Holland. "To empower or to protect? Constructing the 'vulnerable adult' in English law and public policy." *Legal studies* 28, no. 2 (2008): 234-253; Klein, Alana. "Criminal Law and the Counter-Hegemonic Potential of Harm Reduction." (2015) *Dalhousie LJ* 38 447.

have prioritised the right to sexual self-determination as well as the protection of other human rights affected in the commercial sale of sex services in their choices to decriminalise the sale and procurement of sex services. Interestingly, the governmental evaluations of each model, which were conducted five years after the implementation of each approach revealed similar findings as well. However, these findings appear to have resulted in opposite conclusions. The evaluation of the New Zealand model was seen as a success, in particular due to the theoretical legal application and the way this was able to protect the rights of sex workers. It was understood that in the long-term, when society adapted, the approach would achieve the initially set objectives. The German evaluation, however, has focused on the short term application of the model, and sees a slow adaptation as something that needs addressing in order to ensure the rights of vulnerable prostitutes are protected. Thus, the German government has sought to create new legislation, which will tighten the regulation of prostitution. In particular, this move to stricter regulation will involve regulating and monitoring the industry as well as enhancing the protection of the individual sex workers by implementing mandatory registration and consultancy, as well as extending the provision of support and education, in order to ensure all voluntary sex workers are adequately protected, and any victims of exploitation or forced prostitution are helped. Consequently, what can be learnt from the German regulationism is that although decriminalisation, from a philosophical human rights perspective is desirable, in practice it appears that there are a number of other factors apart from the laws, which currently interfere with the realisation of the theoretical objectives of such a regulatory approach. Thus, in the short-term, negative practices cannot be ignored and require addressing to ensure the wellbeing of all sex workers. Only time will show whether the new German regulationist approach will be able to close any current deficiencies in the decriminalisation of prostitution.



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RESEARCH ARTICLE

EFFECTIVENESS OF SELF DEVELOPED INFORMATIONAL BOOKLET REGARDING BIRTH PREPAREDNESS AND COMPLICATION READINESS ON KNOWLEDGE OF ANTENATAL WOMEN AND ASSESSMENT OF COMPETENCIES OF ASHAS REGARDING BIRTH PREPAREDNESS AND COMPLICATION READINESS - A LITERATURE REVIEW.

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Key words:-

Birth preparedness and complication readiness (BPCR), knowledge, antenatal women, competency, health care provider.

Abstract

Birth preparedness and complication readiness (BPCR) is an approach that focuses on raising awareness and creating a stronger demand for quality healthcare services. Childbirth is a crucial biological event which is socially constructed as it takes place within a cultural context and gets shaped by the perception and practices. According to the Integrated Management of Pregnancy and Childbirth (IMPAC) manual of WHO, birth preparedness would significantly increase the capacities of women, their partners and their families to remain healthy, to take appropriate steps to ensure a safe birth and to seek timely skilled care in emergencies. BPCR also reduces delays in receiving appropriate care as it requires the health care providers like nurses, doctors, community health workers to be prepared to attend births and be ready to manage or prevent complications through appropriate measures. On reviewing literature, the researcher found various studies which have shown that not only that antenatal women lack in BPCR, even the health care providers lack in knowledge and/or skills in delivering services related to BPCR.

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Introduction:-

A positive childbirth experience is very important not only to the woman but also to the newborn's health and well-being. Childbirth is a normal physiological process for the majority of women and a process that, like all other life events, is looked upon with a mixture of anticipation and happy expectation. Studies in developed countries have shown a positive impact on pregnancy and birth outcomes when the woman feels in control of the process of pregnancy and birth; childbirth preparation has been shown to facilitate this feeling of self-control and autonomy.

In the Standards for Maternal and Neonatal Care developed by the Department of Making Pregnancy Safer, World Health Organization, a birth plan/emergency preparedness plan includes identification of the following elements: the desired place of birth; the preferred birth attendant; the location of the closest appropriate care facility; funds for birth-related and emergency expenses; a birth companion; support in looking after the home and children while the woman is away; transport to a health facility for the birth; transport in the case of an obstetric emergency; and identification of compatible blood donors in case of emergency.¹

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Since pregnancy is perceived as an ordinary event, most families do not effectively prepare for an emergency. When a pregnancy complication arises, the family is unprepared and while in a hasty attempt to handle the situation precious time is lost in most of the cases. Even to have BPCR at the provider level-health care facility, the doctors, midwives, and other health care workers like ASHAs must have the knowledge and skills necessary to treat or stabilize and refer women with complications.

Hence, the researcher strongly felt the need of assessing the knowledge of antenatal women regarding BPCR after developing and administering an informational booklet and also assessing the competencies of ASHAs regarding the same. In the same context, the researcher reviewed many literatures obtained through various databases like CINAHL, MEDLINE, PubMed, Science Direct, ProQuest and Google Scholar and found out that most of the studies showed lack of BPCR among both the antenatal women and the health care providers.

Materials, Methods And Findings:-

This study is mainly headed in two areas based on the target population and they are knowledge of antenatal women regarding BPCR and competencies of ASHAs regarding the same.

A cross-sectional study was conducted to assess the Birth Preparedness and Complication Readiness of ASHAs at Koppal, Karnataka. (2013). A structured pre-tested interview schedule was used to collect data on socio-demographic and BPCR from 225 randomly selected ASHAs. The study findings showed that the proportion of ASHAs who knew key danger signs were just 1% for labor and child birth, 4.8% for postpartum period and 7.2% for pregnancy. Composite score out of 10 showed a poor score of 0-3 in 55.1%, 4-5 in 37.7%, 6-7 in 7.2%, with none scoring above 7. Score for BP/CR out of 8 showed a maximum score of 8 in 1.4%, 4-7 in 71% and 1-3 in 27.5%. However, knowledge of antenatal care components was good with $\geq 90\%$ in 50.2% of ASHAs. Thus the study concluded that the ASHAs were poorly equipped to identify obstetric complications. They had considerable potential but needed sustained support from the health system to be able to make a difference to maternal mortality and morbidity.²

A quasi-experimental study was conducted to evaluate the effectiveness of Home Based Life Saving Skills training on knowledge of danger signs, birth preparedness, complication readiness and facility delivery, among women in Rural Tanzania. (2016). A two-stage cluster sampling strategy was employed to select 700 samples each from two districts. An intervention district received training with routine care whereas the comparison district continued to receive routine antenatal care. The main findings of the study showed significant improvement of knowledge of three or more danger signs during pregnancy (15.2 % vs. 48.1 %) with a net intervention effect of 29.0 % (95 % CI: 12.8–36.2; $p < .0001$) compared to the comparison district. Birth preparedness practice improved for those who made more than three actions (20.8 vs. 35.3 %) with a net intervention effect of 10.3 % (95 % CI: 10.3–20.3; $p < .0001$) between the intervention and control district at pre-intervention and post intervention. Use of facility delivery improved in the intervention area as compared to comparison district. Thus the study concluded that a community-based intervention in delivering Home Based Life Saving Skills program improved the knowledge of mothers & families regarding preparedness for childbirth and increased deliveries at health facilities.³

A community-based study was done to assess perception and practices regarding BPCR and to identify the related factors among women in a rural community of Bankura district, West Bengal, India. (2011). Cluster sampling technique was used to select 240 women who had delivered in last 12 months with the help from ASHA & Anganwadi workers. Data collection was done using semi structured questionnaire and a set of ten indicators constructing the BPCR index. The study result showed that the proportion of women who had identified a blood donor was low 12.9% (+3.3). Around only one-sixth respondents were aware of key danger signs of pregnancy, childbirth, postpartum and neonatal period, which indicated low awareness at the individual level. BP/ CR index of the study population was 49.4 (+1.7). The study noted that women who were more vulnerable to obstetric complications showed reluctance in following BPCR practices. Thus the study concluded to emphasize the need of motivating the health workers to utilize every contact with the beneficiaries and making them equipped with the concept of 'focused antenatal care' in order to empower women and their family to take decision, plan and implement BPCR practices.⁴

A cross-sectional study was conducted by NIHFV and Department of Community Medicine, S.S. Medical College, Rewa to assess the status of BP/CR at individual, institute and community levels in Rewa district, Madhya Pradesh. (2008-09). 2022 mothers who had delivered before one year of the study and pregnant women who were in 2nd & 3rd

trimester of pregnancy were selected using cluster sampling technique and among the 9 CHCs of Rewa district, 9 medical officers and 11 peripheral health staffs like staff nurses and ANMs were included. The study findings noted that all the CHCs severely lacked manpower like an anesthetist and Ob -Gynae specialist. None of the CHCs had blood storage facilities and were not providing Comprehensive Emergency Obstetric Care like CS facility, blood transfusion, and anesthetic kit. Among the knowledge and skills of health care providers less than 50% of them were competent in diagnosing and management of complications. BP/CR index was only 47.5%. The Birth preparedness indicators like knowledge of danger signs, knowledge of transportation services, 1st trimester registration and population saved money was found to be very low like 18.6%, 18.6%, 24.1% and 44.2% respectively. Thus the study concluded the need to enhance the facilities and manpower at the CHCs through effective resource management and adequate staff training. It also stressed to encourage the mothers and community to effectively involve in BPCR activities.⁵

A descriptive study was done to assess the proportion of women with birth preparedness plans, and the socio-demographic, maternal and institutional determinants of birth preparedness among 345 systematically selected women who had delivered within two years prior to the study attending maternal and child health clinics at Tharaka, Kenya. (2014). Data collection was done using interviewer administered questionnaires, focus group discussion guide and key informant interview guide. The study noted that only 20% of the women were prepared for birth. The aspect of birth preparedness that was identified and planned for by the highest number of women was finances for delivery expenses (74%) followed by place of delivery (68%) while the least was the mode of transport to the facility (35%). The maternal determinants of birth preparedness were history of still birth and attendance of 4 or more antenatal visits. There were no institutional determinants in this study. Thus the study concluded that the level of birth preparedness among the women of Tharaka was low and thus recommended to their ministry of health to sensitize women of reproductive age on birth preparedness.⁶

A cross sectional study was conducted to assess the status of BPCR and socio-demographic factors affecting BPCR among 417 antenatal mothers attending a primary health center, Palam, New Delhi. (2012). The study revealed that the BPCR index was very low 41%. Only 42.9% women were aware about early registration of pregnancy. Only one-third 33.1% of women knew about four or more antenatal visits during pregnancy. Overall, only 27.8% women knew about any one danger sign of pregnancy. Thus the study concluded that efforts should be targeted to increase the awareness regarding components of BPCR among pregnant women and their families at the PHC as well as at the community level.⁷

A cross-sectional survey was done to assess the knowledge and practice towards BPCR among women in Northern Ghana. (2015). Systematic random sampling technique was used to select 211 pregnant women and 211 women who had given birth in the 12 months preceding the study who attended antenatal or postnatal care in health facilities within the 5 zones of the study area. A structured survey questionnaire adapted from the safe motherhood research tools by the Maternal Neonatal Programme of JHPIEGO was used for data collection. The study findings related to knowledge noted that only 3.79% respondents mentioned of knowing about arrangement for someone to cater for house and kids, 3.08% knew about making transport arrangements, 4.5% mentioned arrangement for delivery place ahead of time, 0.24% mentioned the identification of blood donor, 0.71% mentioned identification of skilled attendants and 12.09% did not know anything about BPCR. The study findings related to practices noted that only 18.01% of respondents planned ahead for a place of delivery, only 10.18% had prior arrangement for transport, only 0.95% & 0.47% had identified a potential blood donor and a skilled provider respectively. Thus the study concluded that antenatal care education should be emphasized on BPCR to improve access to skilled and emergency obstetric care.⁸

Qualitative study using focus group discussions (FGD) were conducted to explore the perception of community on BPCR at Rufiji district, Eastern Tanzania (2011). Purposively selected 65 young women and men, as well as 67 older men and women, participated in the FGDs. A pretested topic guide that included cultural and social issues surrounding birth preparedness, pregnancy, and childbirth, steps taken to prepare for pregnancy and potential complications during childbirth in the community, recognition of maternal danger signs, and the use of traditional medicine during pregnancy and childbirth was used to conduct the FGDs. The study finding noted that the community members expressed a perceived need to prepare for childbirth. They were aware of some aspects of BPCR however, certain factors like stigma on unmarried women and transportation were identified as hindrance to birth preparedness and hence utilization of skilled care. Challenges were related to the consequences of poverty they perceived difficulties due to informal user fees. Thus the study concluded to stress upon community reinforcement

for BPCR, by using insurance schemes, community health funds, and by providing information on other birth preparedness messages via community health workers.⁹

A cross-sectional study was done to assess BPCR among slum women in Indore, India. (2004-06). A pretested interview schedule was used to collect data from purposively selected 312 mothers of infants aged 2-4 months in 11 slums of Indore. The study finding noted that only 47.8% respondents were well-prepared. Factors associated with well-preparedness were maternal literacy and availing of antenatal services. Deliveries in the slum-home were high 56.4%. Among these, skilled attendance was very low 7.4%. Skilled attendance during delivery was three times higher in well-prepared mothers compared to less-prepared mothers. Thus the study concluded that antenatal outreach sessions should be used for promoting BPCR as it will be important to increase the competency of slum-based traditional birth attendants, along with promoting institutional deliveries.¹⁰

A Facility-based cross-sectional study was conducted to assess the determinants of BPCR among pregnant women attending antenatal care at Dilchora Referral Hospital, Dire Dawa City, East Ethiopia. (2015). A pre-tested structured questionnaire was used to collect data from 405 systematically selected participants. The study findings noted that the proportion of women who were well prepared for birth and ready for complications were 54.7%. Attending tertiary level education and being knowledgeable on obstetric danger signs were found to be significantly associated with BPCR. The study concluded that the proportion of women who were well prepared for birth was still low and thus improving their awareness and reinforcing counseling on obstetric danger signs was recommended to increase level of BPCR.¹¹

A community-based mixed method study was conducted to find out awareness and practices regarding BPCR among pregnant and recently delivered women in Uttar Dinajpur, West Bengal. (2011). Two-stage, 40 cluster sampling technique was used to select 360 participants. Information on socio-demographic variables as well as awareness and practices regarding BPCR were collected through semi-structured interview. In-depth interviews with one respondent per cluster were also conducted. The study findings indicated that overall BPCR index of the study population was 34.5. Proportion of women aware of at least one key danger sign each of pregnancy, labor, postpartum, and newborn ranged from 12.1% to 37.2%. Proportions of women with first ANC within 12 weeks, four or more ANCs, institutional delivery, saving money, identifying transport, and blood donor were low. Thus the study concluded that preparedness in health system, ensuring competence, and motivation of workers were needed for promotion of BPCR among the study population.¹²

Conclusion:-

Various studies have been done to assess BPCR among the antenatal women and even the health care providers. These studies showed that there is a varied range of factors that affect BPCR. In spite of various health programs for antenatal women, and training of health care providers regarding BPCR, they are found to be not effectively prepared for birth and emergencies. As knowledge of antenatal women and competence of health care providers play substantial role to reduce maternal mortality and morbidity, the researcher feels the need to assess both.

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RESEARCH ARTICLE

PREDICTION OF RESPONSE TO PEGYLATED INTERFERON PLUS RIBAVIRIN IN HEPATITIS C PATIENTS BY IL28B VARIATION AND ITS RELATION TO OTHER DIFFERENT PREDICTORS.

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HCV, Ribavirin, Pegylated- Interferon, IL28B

Abstract

Worldwide, hepatitis C Virus (HCV) is recognized as a major threat to global public health. Accurately predicting the patients who will respond to therapy of HCV by interferon and ribavirin is still important, for patient care and also with respect to the cost as new DAAs are not always available. Studies have shown that IL28B is strongly associated with the response to treatment of hepatitis C by interferon and ribavirin. Our patients are 200 divided into two equal groups, responders and non- responders to Interferon ribavirin treatment. They are submitted to detailed history and clinical examination including body mass index calculation, laboratory examination: CBC, PT, PC, serum Albumin, AST, ALT, Bilirubin total, viral load by PCR, abdominal ultrasound, liver biopsy, IL28B gene. We proved that IL28B is the strongest predictor of response of treatment by Peg/interferon ribavirin and CC allele has the best chance to respond to treatment. Also, BMI, ALT and ALP could be used as predictors for response.

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Introduction:-

Hepatitis C virus (HCV) infection is a major cause of chronic liver disease (Berg T et al., 2003). About 2.35% of the world's population is infected with HCV (Lavanchy D, 2011).

In Egypt, hepatitis C virus (HCV) has the largest epidemic in the world. The released Egyptian Demographic Health Survey [EDHS] declared that the overall prevalence of HCV is 14.7% (El-Zanaty F and Way A, 2009). The blood borne virus, which is highly infectious, kills an estimated 40 000 Egyptians a year and at least 1 in 10 of the population aged 15 to 59 is infected (Henk B, 2014).

Studies have shown that the prevalence of antibodies to HCV (anti-HCV) specially, to a single subtype, 4a in Egypt is approximately 10-fold greater than in the United States and Europe (Strickland GT et al., 2002). Clinical trials demonstrated that type 4 response to treatment by Pegylated- Interferon (PEG- INT) and ribavirin is better than type 1 response but it is worse than type 2 and 3 response (Martinot-Peignoux M et al., 2010).

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Accurately predicting the patients who will respond to therapy is becoming increasingly important, both from the point of patient care and also with respect to the healthcare cost as clinicians need to continue treatment in patients who will respond and stop treatment in patients who are unlikely to respond and use new protease inhibitors instead of Peg- interferon and ribavirin (Zeuzem S et al., 2004). Also in era of new direct antivirals, as they are not always available.

Older age, advanced stage of fibrosis, African-American ethnicity and HCV- related factors, including HCV genotype 1 and high viral load at baseline, predict poor response to anti-viral therapy. Furthermore, metabolic factors, such as high body mass index (BMI), presence and severity of liver steatosis and increasing homeostasis model assessment of insulin resistance (HOMA-IR) score have been reported as negative predictors of response (Bressler BL et al., 2003).

On the other hand, early on-treatment kinetics of HCV RNA, e.g. undetectable HCV RNA at week 4, has a high positive predictive value of SVR (Yu ML et al., 2008). Several independent studies have consistently shown that single nucleotide polymorphisms (SNPs) near IL28B, which encodes the type III interferon are strongly associated with the response to treatment of chronic hepatitis C (Ge D et al., 2009).

However, very few studies are available where all these novel pre- treatment variables are analyzed together in multivariate models.

The aim of this study was to find if there is a relation between IL28B gene single nucleotide polymorphism and the response to treatment of HCV by combination of PEG- Interferon and ribavirin. Also, to find if IL28B was a surrogate predictor of the response to HCV treatment compared to other predicting factors like age, sex, liver enzymes, liver biopsy and degree of steatosis, body mass index (BMI) and viral load.

Material and Methods:-

Study population:-

A prospective study was conducted on 200 patients, including 124 males and 76 females & the mean age was (41.93 ± 9.686) years taken from 657 patients after they took pegylated-interferon/ ribavirin. They were recruited from the outpatient clinics of the Internal Medicine department, Kasr Al Aini hospital, Cairo University, Beni-Suef Public Hospital and National liver institute from 2012 to 2014, while patients with Co- infection with HBV and were receiving immunosuppressant drugs, chemotherapy or corticosteroids concomitantly with the study were excluded. Following the response to treatment with PEG interferon and ribavirin after 48 weeks, the patients were classified into two groups, responders and non- responders each group contained 100 patients. The study protocol conformed to the ethical guidelines of 1975 the Helsinki declaration and was approved by the ethical committee of internal medicine, faculty of medicine, Cairo University. Written informed consents were obtained from participants in this study.

Blood collection and sample preparation:-

Blood (10 ml) was withdrawn by venipuncture; After analyzing cell blood counts (CBC) on aliquots of whole blood in EDTA tube (2 ml) using the (sysmex corp, Kope, Japan) automated hematology analyzer. Liver function tests (bilirubin, ALT, AST, ALP, albumin, PT, PC, & INR), urea and creatinine were measured. Viral load by PCR DNA extraction. DNA was extracted from mononuclear cell layer of whole blood sample using QIAamp kit supplied by Qiagen (USA) according to manufacturer's specifications. IL28B genotyping (rs12979860) was carried out by means of custom TaqMan Assay (Applied Biosystem, USA) designed to detect the following SNP: rs12979860. Primers and probes sequence as following; rs12979860 (5'-GCTTATCGCATACGGCTAGG-3', sense) and rs12979860 (5'-AGGCTCAGGGTCAATCACAG-3', antisense). The TaqMan minor groove binder probe sequences were VIC 5'- ACCTGCCTACCCCTT G-3', and FAM 5'- CTGCCCCACC CTTG-3'. The detection was carried out by qPCR in a StepOne thermocycler by using fluorescent probes. The amplification conditions were as follows: After a denaturation time of 10 min at 96 °C, 45 cycles of 92°C 15 sec 60°C 90 sec were carried out and fluorescence was measured at the end of every cycle and at endpoint. All samples were determined by duplicate and genotypes were assigned both, by the gene identification software (StepOne 2.0, Applied Biosystem) and by analysis of the reference cycle number for each fluorescence curve, calculated by the use of algorithm (Applied Biosystem).

Statistical analysis:-

Pre-coded data was entered into the computer program SPSS (Statistical Package for the Social Science; SPSS Inc., Chicago, IL, USA) release 15 for Microsoft Windows (2006) to be statistically analyzed. Data was summarized using mean and standard deviation for quantitative variables and frequency and percentage for qualitative ones. Comparison of numerical variables between the study groups was done using Student *t* test for independent samples while comparing categorical data was done using Chi square (χ^2) test. Exact test was used instead when the expected frequency is less than 5. Univariate and multivariate analysis models were used to test for the preferential effect of the independent variable(s) on response to treatment. *p* values less than 0.05 was considered statistically significant.

Results:-

Two hundred patients were included in our study. The demographic characteristics and laboratory results are shown in (table 1).

Table 1:- Univariate analysis. HS: highly significant S: significant N S: non- significant

Variables		Non responder	Responder	P value
Number		100	100	
Age(yrs.) (Mean \pm SD)		44.42 \pm 9.111	39.44 \pm 9.647	0.000(HS)
Sex	Male (N, %)	52(53.1%)	30(30%)	0.14(NS)
	Female (N, %)	48 (46.9%)	70(70%)	
BMI (%) (Mean \pm SD)		29.5260 \pm 5.15565	26.3260 \pm 3.39585	0.000(HS)
AST(IU/L)		64.99 \pm 41.528	27.44 \pm 16.469	0.000(HS)
ALT (IU/L)		69.71 \pm 40.743	25.46 \pm 14.206	0.000(HS)
ALP(IU/L)		120.41 \pm 62.545	156.17 \pm 52.245	0.000(HS)
Viral load (before treatment)		1,752,561.87 \pm 8,147,842.224	742,197.40 \pm 1,641,246.876	0.226(NS)
IL28b SNP	CC (N, %)	6(6%)	47(47%)	0.000(HS)
	CT (N, %)	60(60%)	47(47%)	
	TT (N, %)	34(34%)	6(6%)	
Fibrosis	0 grade (N, %)	10(10%)	4(4%)	0.29(NS)
	1 grade (N, %)	59(.59%)	76(76%)	
	2 grade (N, %)	21(21%)	14(14%)	
	3 grade (N, %)	7(7%)	6(6%)	
	4 grade (N, %)	3(3%)	0(0%)	

By using univariate analysis, the results demonstrate a highly significant relation between age, sex of the patient and the response to treatment of HCV by PEG interferon and ribavirin. The *p* value was respectively (0.00 & 0.014). Also the study shows significant relation between treatment response and elevated ALT & ALP of the patients. The study reported that a highly significant relation between IL28B subtype and the response to treatment of HCV by PEG interferon and ribavirin. CC showed more incidence to respond to treatment and TT less incidence to respond. In group of responders (CC/CT/TT) represented (47%/47%/6%) respectively. In non- responders group (CC/CT/TT) represented (6%/60%/34%) respectively. The *P* value was (0.000). However, regarding both viral load & fibrosis grades, no significant relation between those & the response to treatment of HCV by PEG interferon and ribavirin (*p* value= 0.226 & 0.092) respectively.

By multivariate analysis: The present study illustrated a significant relation between BMI and the response to treatment, the lesser the BMI is, the better response to treatment (*p*= 0.005). The *p* value for the ALT, ALP and its relation to response to treatment were (0.000, 0.028) respectively. As well as the *p* value for the IL28B subtype and its relation to response to treatment was (0.000) in multivariate analysis. These result showed highly significant relation between IL28B subtype and the response to treatment. CC subtype has more incidence to respond to treatment by PEG interferon and ribavirin. TT subtype has less incidence to respond to treatment by PEG interferon and ribavirin. The odd ratio for BMI, ALT, ALP and IL28B 0.76, 0.92, 0.01 and 8.79 respectively. On the other hand, no significant relation between the response to treatment of HCV by PEG interferon and ribavirin and each of these factors including age, sex, AST, viral load & fibrosis (*p* value = 0.405, 0.851, 0.051, 0.531 & 0.405 respectively) in multivariate analysis.

Discussion:-

Many studies were done to find predictors of treatment response to INF and ribavirin in HCV. Finding those patients who will mostly respond to treatment will help in decreasing the cost and side effects of medications. Moreover, this will help in choosing patients to use the triple therapy with direct acting antiviral drugs (Butti M, 2002). An important factor influencing treatment outcome in chronic hepatitis C has been discovered IL28B gene polymorphism that is also distributed according to ethnic and geographical areas. The CC allele polymorphism occurs in 33% of Americans of European ancestry, 14% of African Americans and 29% of Hispanic Americans (Thomas DL et al., 2009). Individuals carrying IL28B polymorphism with CC alleles have the best prognosis, with two to three times the rate of sustained virological response (SVR) with IFN-based anti-HCV therapy, while those with TT alleles have the worst SVR rates (Thompson AJ et al., 2010). Most of the initial studies of this polymorphism were conducted on genotype 1 patients. However, a study by Asselah T, et al., (2012) has shown that, in patients with HCV genotype 4, patients with CC, CT and TT genotypes have 82%, 47% and 29% SVR rates respectively.

IL28B genotype is the most important pretreatment predictor of response to peg-IFN and RBV therapy for HCV-1. Patients who carry the good response genotype (e.g. C/C at rs12979860) have reported SVR rates $\geq 70\%$, a two- to threefold increase over patients who carry one of the poor response genotypes (e.g. C/T, T/T at rs12979860) (Chen Y et al., 2012). While in genotype 2 and 3 infected patients, this association seems to be less pronounced (Scherzer TM et al., 2011).

The primary goal of the present study was to evaluate the variable predictors of treatment response (PEG interferon and ribavirin) in Egyptian patients infected with HCV subtypes 4a, mainly age, sex, BMI, fibrosis on liver biopsy, viral load, AST, ALT, ALP and IL28B SNP.

For HCV genotype 4 (HCV-4), three studies on HCV mono-infected (Stättermayer AF et al., 2011; De Nicola S et al., 2012; Asselah T et al., 2012) and two in patients coinfecting with HCV and the human immunodeficiency virus (HIV) (Pineda JA et al., 2010; Rallón NI et al., 2011) suggest that single nucleotide polymorphisms (SNP) near IL28B are strong predictors of treatment outcome.

The current study was conducted on 200 Egyptian patients infected by HCV; 124 males and 76 females. The patients were classified into two groups; responders and non-responders according to response to treatment with PEG interferon and ribavirin following 48 weeks. Univariate and multivariate analysis were used to detect the relation between different predictors and the response to treatment by c. We studied the age, sex, BMI, fibrosis on liver biopsy, viral load, AST, ALT, ALP and IL28B SNP as predictors of response. Our results showed a highly significant relation between IL28B subtype and the response to treatment of HCV by PEG interferon and ribavirin in univariate analysis. CC subtype showed more incidence to respond to treatment and TT less incidence to respond. In group of responders (CC/CT/TT) represented (47%/47%/6%) respectively. In non-responders group (CC/CT/TT) represented (6%/60%/34%) respectively. The P value was (0.000). The p value for the IL28B subtype and its relation to response to treatment was (0.000) in multivariate analysis. These results showed highly significant relation between IL28B subtype and the response to treatment. CC subtype has more incidence to respond to treatment by PEG interferon and ribavirin. TT subtype has fewer incidences to respond to treatment by PEG interferon and ribavirin. These results are in accordance with Stättermayer AF, et al., (2011); De Nicola S, et al., (2012); Asselah T et al., (2012).

In our study, we found that the age has a highly significant relation to the response to treatment of HCV by PEG interferon and ribavirin in univariate analysis. The responders mean age mean is 39.44 and the lesser the age, the better the response with a p value (0.000). However, this relation was not detected in multivariate analysis. The p value for the age and its relation to response to treatment in multivariate analysis was (0.567). These results showed no significant relation between age and the response to treatment of HCV by PEG interferon and ribavirin. The SVR was found significantly higher in patients below 40 years of age compared to those above 40 years ($P = 0.022$) in a study done by Muhammad Idrees and Sheikh Riazuddin, (2009) and this was disagreed by Gamal El Din E, et al., (2010) found no relation between age and response to treatment by Peg/ribavirin.

Strangely, in our study, the fibrosis stage did not show significant relation to SVR neither in univariate or multivariate analysis (p value =0.050 and =0.851, respectively). Only in one study by Thomas Berg et al., (2003), the stage of fibrosis was not significantly associated with SVR.

We found a better response to treatment with PEG and ribavirin in males compared to females. The p value of the sex and its relation to response to treatment was (0.014) in favor of the male sex. However, in multivariate analysis this effect disappeared. The p value of the sex and its relation to response to treatment was (0.405). These results showed no significant relation between sex and the response to treatment of HCV by PEG interferon and ribavirin. While, Hadziyannis et al., (2004) and Conjeevaram et al., (2006) found that males have a lower SVR than do females, no statistically significant correlation was found between sex and SVR by Kau A et al., (2008).

In our study, a highly significant relation was found between BMI of the patient and the response to treatment of HCV by PEG interferon and ribavirin. The P value was (0.000) in univariate analysis. The p value was (0.005) in multivariate analysis in our study. These results showed highly significant relation between BMI and the response to treatment. Bressler BL et al., (2003), reported that body fat mass, as estimated by body mass index (BMI), influenced viral eradication in response to antiviral treatment for HCV and found that obesity defined as a BMI greater than 30 was a negative predictor of response to HCV treatment. Body weight was not significantly associated with SVR in a study done by Thomas Berg et al., (2003).

Our results univariate analysis showed a highly significant relation between AST of the patient and the response to treatment of HCV by PEG interferon and ribavirin as the P value was (0.000). While the p value for the AST and its relation to response to treatment was (0.051) in multivariate analysis, which showed no relation between AST and the response to treatment of HCV by PEG interferon and ribavirin. Other studies showed mixed results. In a study done by AS Hgar H et al., (2009), they found the lesser AST was, the better the response to Peg/ribavirin. Gamal El Din E et al., (2010) found no relation between AST level and SVR.

Our results showed a highly significant relation between ALT of the patient and the response to treatment of HCV by PEG interferon and ribavirin in univariate analysis. The P value was (0.000). The p value for the ALT and its relation to response to treatment was (0.000) in multivariate analysis. These result showed highly significant relation between ALT and the response to treatment. The lesser the ALT is, the better the response to treatment. The mean for responders was 25.46. The studied baseline ALT levels, which were an indicator of liver damage, were significantly higher in responder patients than in non-responders in treatment ($p = 0.002$) (Berg T et al., 2003). Pre-treatment ALT levels had no predictive role in treatment response rate in study done by Muhammad Idrees and Sheikh Riazuddin, (2009).

Our results showed a highly significant relation between ALP of the patient and the response to treatment of HCV by PEG interferon and ribavirin in univariate analysis. The P value was (0.000). The mean for responders was 156.17units/litter. The higher the ALP is, the better the response. The p value for the ALP and its relation to response to treatment was (0.028) in multivariate analysis in our study. These results showed significant relation between ALP and the response to treatment. ALP was of no significant impact in a study done by Gamal El Din E et al., (2010). In a study done by Ekram W et al., (2015); they reported a significantly higher in alkaline phosphatase, in non-responder group than in responder group at baseline assessment ($p=0.037$).

In our study the p value for the viral load and its relation to response to treatment was (0.226) in univariate analysis. These result showed no significant relation between viral load and the response to treatment of HCV by PEG interferon and ribavirin. The p value for the viral load and its relation to response to treatment was (0.531) in multivariate analysis in our study. These result showed no significant relation between viral load and the response to treatment of HCV by PEG interferon and ribavirin. A low baseline viral load ($P= 0.04$) was independent predictor for sustained virologic response in a study done by Thomas Berg et al., (2003). The predictive effect of baseline viral load was confirmed by several studies, even in the studies of patients co-infected with HCV and HIV (Zeuzem S et al., 2012).

Conclusion:-

We concluded that IL28B is the strongest independent predictor of response of treatment by Peg/ ribavirin by the highest OR 8.79. Also, BMI, ALT and ALP could be used as predictors for response. AST, age, sex, viral load and fibrosis are not predictors for response according to our study.

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RESEARCH ARTICLE

“AWARENESS AND ATTITUDE OF MEDICAL FRATERNITY TOWARDS WHOLE BODY AND ORGAN DONATION”.

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Abstract

This study is undertaken to evaluate the awareness and attitude of medical fraternity towards whole body and organ donation. A cross sectional study was conducted among undergraduates, graduates and post graduates. Total 500 participants answered the multiple choice questionnaire containing information about demographic features, awareness and attitude towards whole body and organ donation. Out of 500 participants, 44% were willing to donate organs, 47.8% were willing to donate body only for organ transplantation, while only 5.8% for dissection purpose. 16.2% and 36.4% were not willing to donate organs and body respectively. 55.4% were self motivated followed by media (27.7%). Mistrust or misconception was the major reason for unwillingness to donate followed by premature organ removal. Although there is high level of awareness about whole body and organ donation, most of the medical students did not filled the pledge form. It is suggested that there is a need for reviewing medical school curricula and focussing on organ and whole body donation so, that there will be an altruistic change in attitude of future medical professionals.

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Introduction:-

Best method to deliver anatomy training to medical students is by cadaveric dissection. Voluntary whole body donation and unclaimed bodies obtained by the police is the principal means by which universities acquire bodies for dissection. In India, the rate of increase in number of medical students is not correlating with the number of whole body donors. As a result, there is a diminished opportunity for an individual student to carry out dissection of the cadaver. In Japan, cadavers for dissection were totally supplied by body donation.⁽²⁾

The idea of donating bodies to medical science was decreased significantly from 31.5% to 19.6% among medical students after exposure to dissection.⁽¹¹⁾

Replacing the organ was the only treatment for patients with end stage organ disease. There are two kinds of donors: live and cadaveric (brain dead persons). Highest rate of cadaveric donation in world occurs in Spain being 32 per

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million population, in USA and UK, it is 22 and 14 per million population, where as in India, it is just 0.16 per million population ⁽¹³⁾. Only 23% knew about the concept of cadaver as brain death and 93% were able to distinguish between brain death and persistent vegetative state ⁽¹³⁾.

The medical personnel are the one who derive maximal benefit from dissection and they are also the one who will have maximum knowledge on organ and whole body donation. So, this study was undertaken to examine the awareness and attitude of medical students and doctors towards both whole body and organ donation.

Materials and Methods:-

A cross sectional study was conducted in GITAM Dental College and Hospital, Andhra Pradesh, among 500 medical students from Nov 2015 to Jan 2016 to evaluate the awareness and attitude towards whole body and organ donation. Medical students were categorized as Undergraduates, Graduates and Post graduates. Undergraduates were again categorized as third and fourth year students. For this, a multiple choice questionnaire had been given to the participants which includes information about donation.

Attitude and Awareness of Medical Fraternity towards Organ / Whole Body Donation

A.) Age (year):

B.) Sex:

C.) Religion:

- | | |
|----------------|-------------------|
| • Hinduism | • Islam |
| • Christianity | • Others(specify) |

D.) Educational Qualifications

- | | |
|--------------------------|------------|
| • Inter/Higher secondary | • Graduate |
| • Post graduate | • Other |

OPINION REGARDING ORGAN / BODY DONATION

E.) What is your idea about organ donation:

- | | |
|---|---|
| • Organ donation from living donor only | • Organ donation from living & cadaver both |
| • Organ donation from cadaver only | • No idea |

F.) What is your idea about Body Donation:

- | | |
|---|-----------|
| • For dissection purpose of medical students only | • Both |
| • For purpose of organ transplantation only | • No idea |

J.) Will your family agrees for body donation:

- Yes without any hesitation
- No absolutely not
- Yes but need persuasion
- No idea

G.) Are you willing to donate your organ after death:

- | | |
|--------------------------------|-------------|
| • Yes, with out any hesitation | • No idea |
| • Yes, but need persuasion | • For money |
| • No absolutely not | |

H.) Do you want to donate your body after death:

- | | |
|---------------------------------------|-------------|
| • Yes, only for organ transplantation | • Unwilling |
| • Yes, only for dissection purpose | • For money |
| • For both purpose | |

I.) Will your family agrees for organ donation:

- | | |
|------------------------------|----------------------|
| • Yes without any hesitation | • No, absolutely not |
| • Yes but need persuasion | • No idea |

J.) For those who are willing to donate Body or Organ only continue this; And those who are not willing to donate continue page no.4

FOR THOSE WHO ARE WILLING ONLY

K.) From where you are motivated for organ / body donation:

- By medical person / Doctor
 - By media (TV, Radio, News paper, Internet etc)
 - By family members
- L.) Have you already filled up donor card pledge form:
- Yes
 - No
- M.) Reason behind willingness to donate body / organ but not filled up pledge form till now:
- Not decided yet
 - Will do later
 - Want to know much more
 - No idea about procedure
 - Religious Barrier
 - Family not agreed
 - No benefit
- N.) What is your opinion regarding positive attitude of Body / Organ donation:
- To be live by other peoples life
 - To avoid unnecessary wastage of body by cremation
 - To save other needy people's live
 - To facilitated medical teaching process more
 - To avoid environmental pollution
 - All of above
 - Others
- O.) If willing which organ would you like to donate:
- Kidney
 - Heart
 - Liver
 - Brain
 - Whole body
- FOR THOSE WHO ARE NOT WILLING**
- P.) Reason behind willingness regarding body / organ donation:
- Organ could be wasted
 - Don't want to cut body in to pieces
 - Organ / Body could be misused / abused
 - Religious barrier
 - Prevented by family members
 - Psychological anxiety
 - No reason
 - No knowledge about this
 - Any other reason
 - No monetary benefit for family
- Q.) Do you have any Religious beliefs for not donating organ:
- Yes
 - No
- R.) Do you have any fear of premature organ removal:
- Yes
 - No
- S.) Do you have any mistrust / misconception about how the medical community treats registered organs:
- Yes
 - No
- T.) If you have monetary benefit, will you accept for donation of Body / Organ?
- Yes
 - No

[Table/Fig-1]: Questionnaire

Results:-

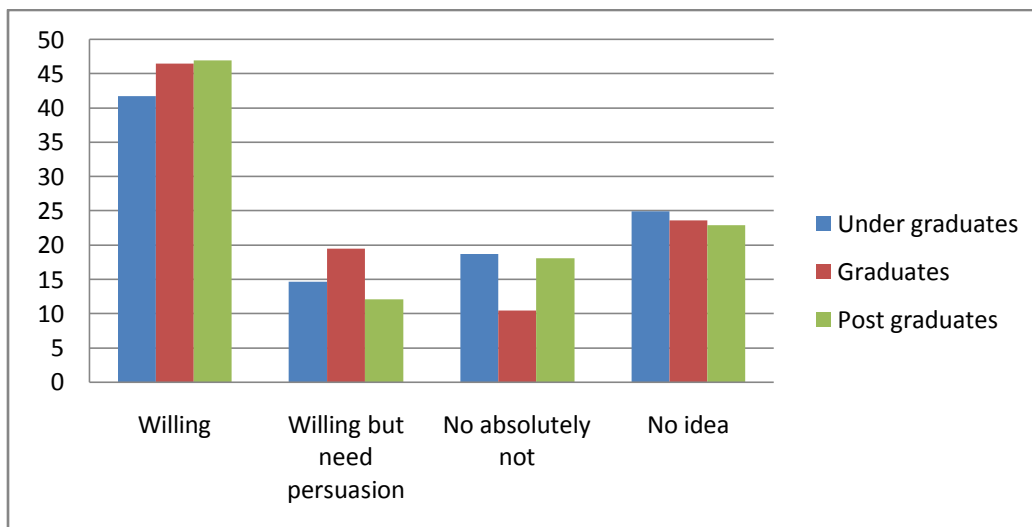
Questionnaires were answered by all 500 participants, out of which 273 were undergraduates, 144 were graduates and 83 were post graduates.

Majority of participants were willing to donate organ and 24% had no idea about organ donation. Responses from organ donation was close among undergraduates, graduates and post graduates with a little higher willingness among graduates and post graduates. Regarding cadaveric donation, maximum (48%) of participants were willing to donate only for organ transplantation while for dissection purpose was very poor (6%).

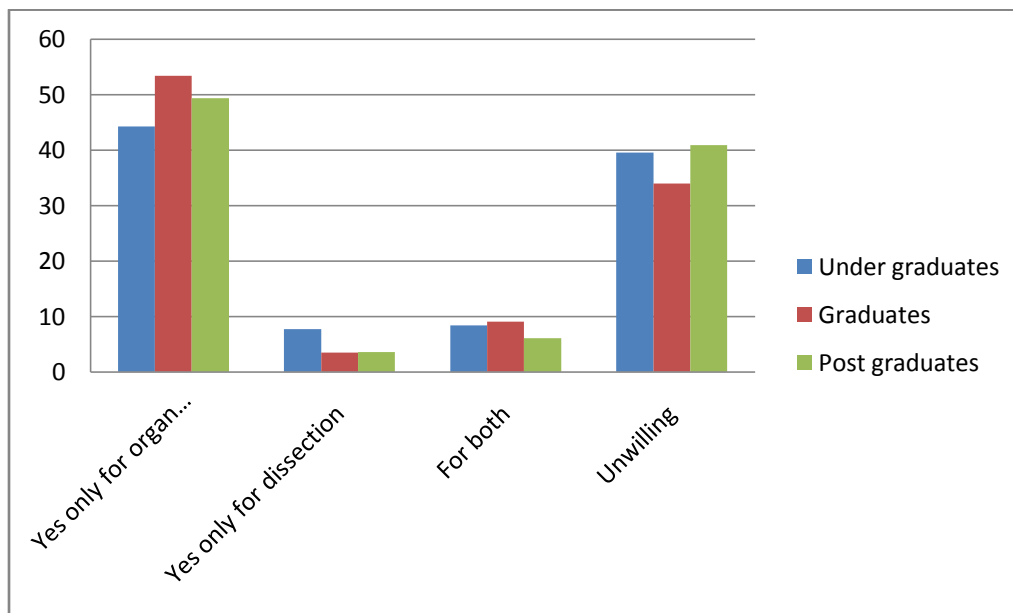
Majority of participants were self motivated followed by media. Very less percentage of participants were motivated by medical person / doctor. Even though, majority of participants were willing to donate, only one had filled donor card. 33% of participants had no idea about donation. Mistrust or misconception was the main reason behind unwillingness to donate organs. Reasons behind unwillingness to donate organs is shown in Table/Fig 7.

Groups		Male	Female	Both	Percent
Undergraduates	Third year	31	113	144	28.8
	Fourth year	32	97	129	25.6
	Total	63	210	273	56.4
Graduates		24	120	144	28.8
Post graduates		31	52	83	16.6

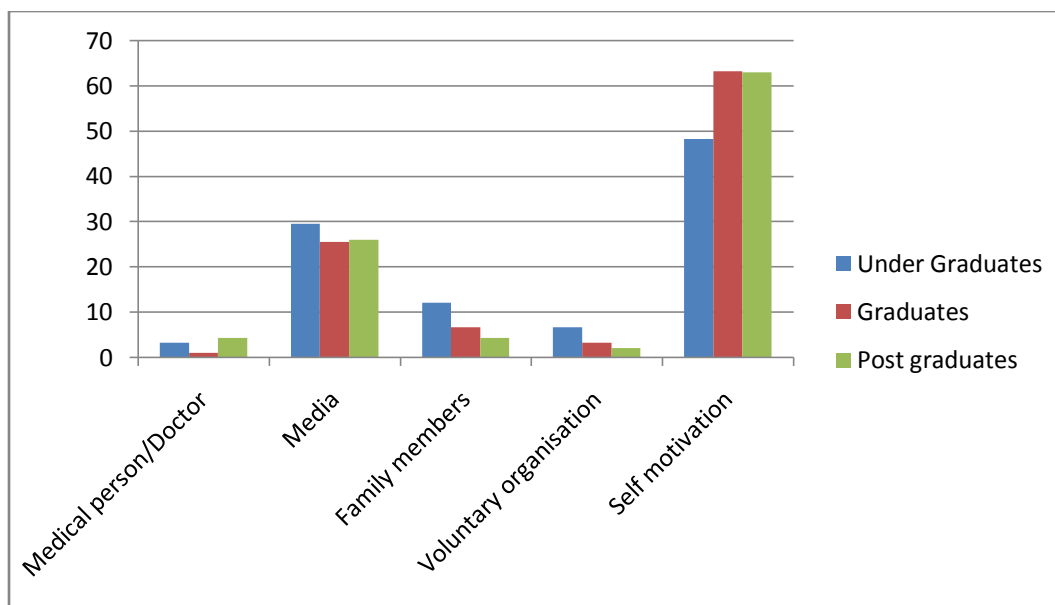
Table/Fig-2: Distribution of demographic parameters



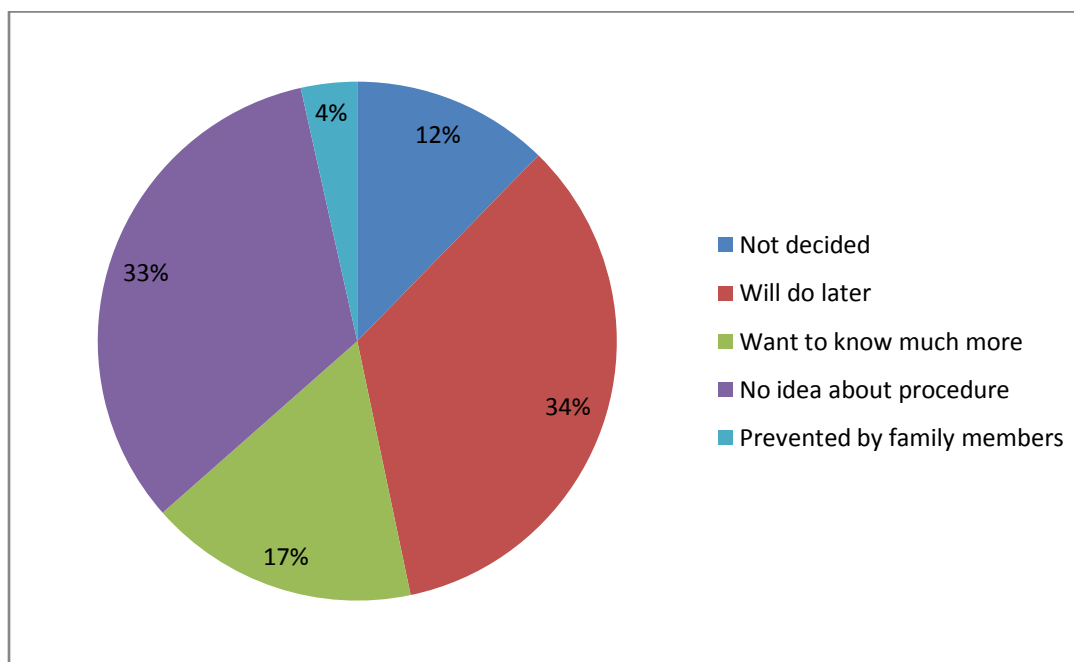
Table/Fig-3: Attitude towards organ donation



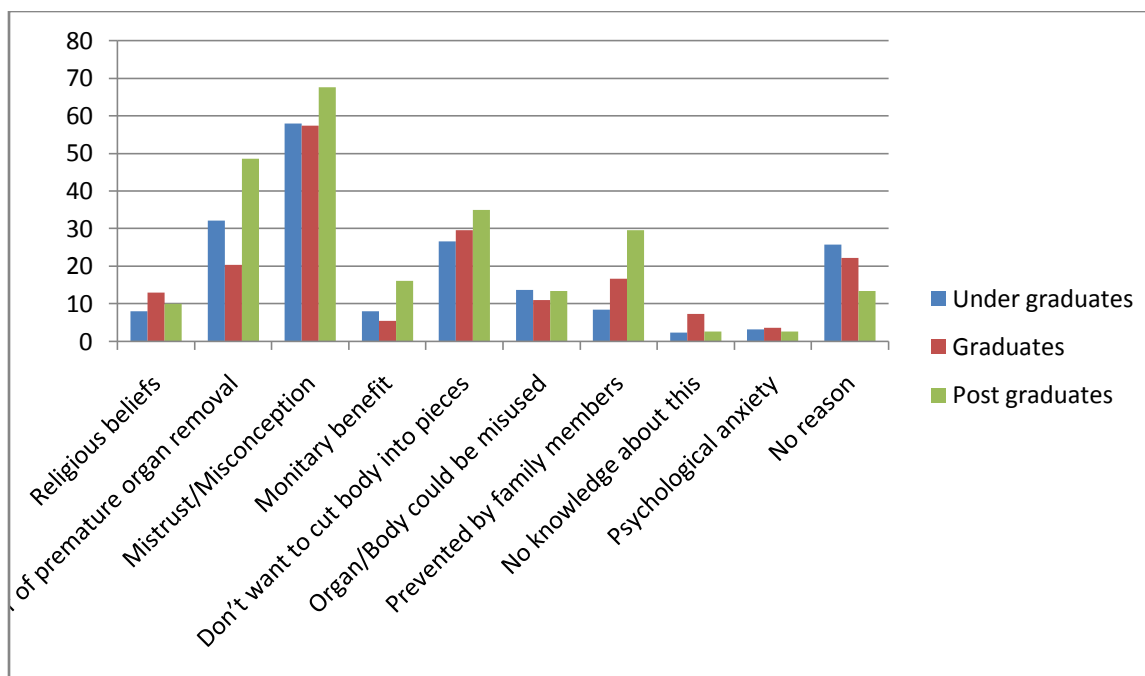
Table/Fig-4: Attitude towards body donation.



Table/Fig-5: Motivating factors for donation



Table/Fig-6: Reasons for not filled up donor card



Table/Fig-7: Causes of unwillingness to donate organs

Discussion:-

Body donation to science was first occurred in 1832, when British utilitarian philosopher, Jurist and social reformer Jeremy Bentham's body donated their body according to their will ⁽¹⁰⁾.

In India, it was first experienced in year 1956, when the body of pandurang Sridhar Apte was donated to BJ Medical college, Pune ⁽¹⁰⁾. Even though, body donation was started in 1956 there was no significant improvement in percentage of body donation. The rate of increase in medical students was not correlating with the body donors for cadaveric dissection and research purposes. And the demand for organs was also increasing as the patients with end stage organ diseases were increasing. As physicians will have maximum knowledge on organ donation process, they can play a key role in solving this problem of shortage of organs and body by eliminating the barriers for organ and body donation by creating awareness, educating and motivating others but they may fail because of lack of knowledge about donation⁽⁹⁾. So, this study is undertaken, to know the awareness and attitude among 500 medical personnel which includes undergraduates, graduates and post graduates.

In this study, we found that 44% of respondents were willing to donate organs, 47.8% were willing to donate body only for organ transplantation while for dissection purpose was very poor and females were more willing to donate than males. Very negligible members were willing to donate for money where as in Turkey, 58.4% were willing to donate their organs and the acceptance of live organ donation was higher than cadaveric donation ⁽³⁾.

Majority of students were self motivated followed by media and only 8 members were motivated by medical person / doctor and 59.3% of students said that they want to save other needy people's live. In other studies they found that print / electronic media as their major and primary source of knowledge ^(2,3,4,8).

Although, more than half of our students were willing to donate, only one student has filled up donor card and only 8 of them asked for donor cards which were similar to a study done in turkey, where only 1.2% had a organ donor card ⁽³⁾. It states that the participants who are willing to donate are not whole heartedly willing to donate and some participants did not know about the procedure. So, proper guidance and counselling of and also incorporating organ donation topics in medical curriculum thereby creating awareness about it is very much essential.

Reasons behind unwillingness to donate organ or body was not wanting cut their body into pieces because of their previous experience with dissection hall followed by mistrust or misconception, fear of premature organ removal, religious barriers and monetary benefit Very less percentage of people were prevented by their family members.

Idea of donating bodies to medical science among medical students decreases significantly from 31.5% to 19.6% but not levels reported in general population⁽¹⁰⁾.

In a study done among medical and dental members, showed that 80% believed in mistrust and are not ready to donate their bodies⁽⁶⁾ which was similar to a study done in Malaysia⁽⁷⁾.

So, in India, Anatomy act was implemented in 1949, a state act published in state government gazette, which regulates the use of unclaimed bodies for medical research and education. It states that any death occurring in the state hospital or in a public place within the prescribed zone of medical institution and there are no claimants for body provided the police declared a lapse of 48 hours, they can be used for medical education⁽¹⁰⁾.

In Pakistan, Transplantation of human tissues and organ bill was passed by Pakistan parliament in 2007 and it includes restriction of organ donation only to a closed blood relative. It also created the "Human organs transplantation authority" which established transplant registry and recognized centers for transplantation⁽²⁾⁽⁶⁾.

A similar act was established in 2004 in Libya known as Libyan organ transplantation program to initiate deceased organ transplantation activity and helps in obtaining legal approval for organ donation and also organizes educational campaigns and meetings with the religious leaders to overcome the religious beliefs⁽¹²⁾.

Awareness of brain death and its importance for organ transplantation was extremely low in India especially among villagers⁽¹⁴⁾. Brain stem death was now legal in India under the passage of legislation by the Indian parliament act 1994 and it applies only when organ transplantation was intended⁽¹⁴⁾.

Most important reason for nobody donation in Indians is lack of awareness followed by religious beliefs⁽¹⁰⁾. In Libya, lack of adequate knowledge and religious implications were the major reasons for refusal⁽⁵⁾.

So, shortage of organs was not only due to mistrust or misconception but also because of lack of awareness, thought of being dissected and religious implication.

Conclusion:-

It is emphasized that there is a need for reviewing medical school curricula and focusing on organ and whole body donation so, that there will be an altruistic change in attitude of future medical professionals towards organ and whole body donation.

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RESEARCH ARTICLE

ASSESSMENT OF KNOWLEDGE, ATTITUDE AND PRACTICES TOWARD FIRST AID OF HYPOGLYCEMIA IN SAUDI ARABIA, RIYADH AND JEDDAH CITIES 2016.

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Ahmad Abdullah Alghamdi, zainalaabdeen sayyar alyami, Khalid ali shbeeli, matar Mahmoud yousuf
alsomali, Faisal aliwi qidhi alenezi, Mahdi mana abdullah alabbas and Mohammed Nasser Makeen.

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Abstract

Background: Hypoglycemia is one of the most common complications of diabetes management. The over enthusiastic approach in maintaining tight blood sugar control so as to reduce the long-term complications of diabetes have resulted in increase in frequency of this complication. This situation seems to get worsened with use of combination of anti-diabetic drugs.

Objectives: To assess the level of awareness toward First Aid Related To Hypoglycemia among Population In Riyadh And Jeddah cities to identify barriers.

Methods: A cross sectional analytical questionnaire based study among the general population of Riyadh And Jeddah cities.

Results: A total of 360 subjects answered the questionnaires. The mean age were 36 years, ranged from 16 to 70 and 45.2% females and 43.7% male respondents .Of these, 67% had attended college, 5% had postgraduate degree, 23.5% had completed high school, and 3.1% had basic school. The majority of participants had high level of monthly income (38%). The majority of subjects included in the research were (59.6%) married and (51.3%) were unemployed.

Most subjects (338) had poor knowledge about risk of hypoglycemia and there was no association between the knowledge and demographics of participants except for education as the higher the levels of education, the more significant association with good knowledge.

Conclusion: The Results of the present study reveal that the knowledge about the risks of hypoglycemia and first aid of hypoglycemia was poor among the studied population. Also, education significantly impacts the knowledge of hypoglycemia thus there is a need for providing the population and patients with necessary information to improve their hypoglycemia awareness.

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Background:-

Hypoglycemia is one of the most common complications of diabetes management. The over enthusiastic approach in maintaining tight blood sugar control so as to reduce the long-term complications of diabetes have resulted in increase in frequency of this complication. This situation seems to get worsened with use of combination of anti-diabetic drugs.

The American Diabetes Association defines the hypoglycemia as “any abnormally low plasma glucose concentration that exposes the subject to potential harm”, and proposes a threshold of $<70 \text{ mg\%}$ ⁽¹⁾. In subjects with type 1 diabetes, autoimmune destruction of pancreatic β -cells leads eventually to an absolute requirement for insulin replacement therapy. Insulin delivered exogenously is not subject to normal physiological feedback regulation, so it may induce hypoglycemia even in the presence of an intact counter regulatory response. The average individual with type 1 diabetes experiences about two episodes of symptomatic hypoglycemia per week, a figure that has not changed substantially in the last 20 years ⁽²⁾. This risk is increased markedly with the increasing duration of the disease and strict glycemic control. In subjects with type 2 diabetes, the increasing duration of the disease and the more widespread use of insulin therapy also increase the risk of severe hypoglycemia. This was reflected in a recent survey in Tayside, Scotland, which found the proportion of severe hypoglycemic episodes needing emergency medical assistance was similar between type 1 and insulin-treated type 2 diabetic patients ⁽³⁾. Our Study Was Designed To Examine Health Beliefs And Assessment Level OF Awareness Toward Early First Aid Related To Hypoglycemia among Population In Saudi Arabia, Riyadh And Jeddah And Applies The Health Belief Model To Determine Barriers.

Rationale:-

Diabetes mellitus is a chronic disease that constitutes a major public health problem. The worldwide prevalence of DM has risen tremendously over the past two decades. Hypoglycemia is one of the most common complications of diabetes management. The over enthusiastic approach in maintaining tight blood sugar control so as to reduce the long-term complications of diabetes have resulted in increase in frequency of this complication. This situation seems to get worsened with use of combination of anti-diabetic drugs.

Objectives:-

General objectives:-

To assess the level of awareness toward First Aid Related To Hypoglycemia among Population In Riyadh And Jeddah cities to identify barriers.

Specific objective:-

The goal of this study was to examine community health beliefs regarding hypoglycemia and their perceptions related to it and evaluate the role of demographic factors in shaping beliefs about first aid related to hypoglycemia and assess possible associations between demographic characteristics with the preventive behavior of interest.

Methods:-

Study design: cross sectional study.

Setting and data collection:-

This survey analysis was conducted among community population in Riyadh and Jeddah cities. A preformed self-administered questionnaire was distributed among the community population.

Sample:-

Subjects were chosen according to geographical and sex distribution. Sample size was calculated based on web-site calculator ⁽⁴⁾ taking the total size of Riyadh and Jeddah population (4,087,000+2,800,000) ⁽⁵⁾, confidence level (95%) and margin error (5%) to be 285. Additional 20 % was added to cover the missing data. The total sample obtained was 360.

Study population:-

The study population included were both male and female in Riyadh and Jeddah cities.

Study tool:-

Preformed Self-administered questionnaire that requires information about:

- 1- Demographic characteristics: age, gender, education level, monthly income, marital status, and employment.
- 2- Knowledge assessment including 7 questions about risk factor of hypoglycemia and first aid of hypoglycemia. A score of 1 was given to yes and 0 otherwise. For each subject, a maximum score of 7 was calculated. A scoring system was applied to measure the respondents' knowledge towards hypoglycemia impacts. The hypoglycemia knowledge score was calculated as a continuous variable by summing the participant's number

of yes answers to the questions. One point was awarded for each yes, and zero for each no or don't know, with a maximum obtainable correct score of 7 for each respondent. The knowledge score was categorized into two levels indicated by poor (0–4.5), and good (5–7).

Ethical considerations:-

An informed consent was obtained from the participants included in this research before filling the questionnaire.

Statistical analysis:-

Data were entered into the Statistical Package for Social Sciences (SPSS, version 24, SPSS, Chicago, IL, U.S.A.) and descriptive analysis conducted. The results were reported as percentage (95% confidence interval).

The internal consistency was assessed using Cronbach's α test. The test results were for the 7 statements of knowledge about hypoglycemia first aid was 0.422.

Association of respondents' characteristics with about hypoglycemia first aid, was evaluated using univariate logistic regression. Results were reported showing odds ratio (OR) and 95% confidence interval. Statistical significance was accepted at $p < 0.05$. The dependent variables: knowledge of hypoglycemia risk (1 = Poor knowledge and 0 = good knowledge). The following independent variables were included: (1) age: ≤ 20 years, [21–30 years], [31–40 years], [41–50 years], > 50 years; (2) gender: males and females; (3) level of education: low, for those who completed secondary school or less, intermediate for those who finished college degree or have bachelor degree and high for those who had postgraduate degree; (4) monthly income: low [< 3000 Saudi Riyal (SR)], middle [3000–10000 SR] and high [> 10000 SR]; (5) marital status: single and married; (6) employment: unemployed and employed.

Results:-

Demographics of the studied subjects:

The socio-demographic characteristics were shown in Table. 1.

Table 1:- Socio-Demographic Characteristics of Respondents (n = 360)

	Frequency	Percentage (%)
Age (Year)		
≤ 20.00	36	8.60%
21.00 - 30.00	125	29.70%
31.00 - 40.00	56	13.30%
41.00 - 50.00	47	11.20%
51.00+	63	15.0%
Missing	94	22.30%
Mean\pmSD (Min.-Max.)	36 \pm 14 (16 – 70)	
Gender		
Female	228	54.20%
Male	184	43.70%
Missing	9	2.10%
Education level		
Basic school	13	3.10%
High School	99	23.50%
Collage degree	282	67.00%
Post-graduate	21	5.00%
Missing	6	1.40%
Monthly Income		
< 3000	90	21.40%
3000-5000	46	10.90%
5000-7000	34	8.10%
7000-10000	80	19.00%
> 10000	160	38.00%

Missing	11	2.60%
Marital Status		
Married	251	59.60%
Un Married	169	40.10%
Missing	1	0.20%
Employment		
Employed	194	46.10%
Un Employed	216	51.30%
Missing	11	2.60%

A total of 360 subjects were included in the study and answered the questionnaire. The age ranged from 16 to 70 years. The mean was 36 and about 8.6% of subjects were less than 20 years old, 29.7% of participants were from 21-30 years old, 13.3% were from 31-40 years old, 11.2% ranged from 41-50 years old, 15% were more than 51 years old and 22.3% of participants had missing data about age.

The gender distribution showed that 54.2% of participants were females, 43.7% were males and 2.1% had missing data about gender.

The study population represents a highly educated group of people, with 67% having attended university or college, 23.5% having completed high school, 5% having post-graduate degree and 3.1% completed basic school and 1.4% had missing data about education level.

The majority of participants had high level of monthly income (38%) more than 10.000 SR, followed by 21.4% of patients had monthly income less than 3000 SR then 19% had income ranged from 7000-10.000 SR, 10.95% had an income that ranged from 3000-5000 SR and 8.1% had an income ranged from 5000-7000 SR.

The marital status showed that the majority of subjects included in the research were (59.6%) married and 40.1% were un-married.

The most of subjects were unemployed (51.3%), and 46.1% were employed, however only 2.6% had missing data.

Responses to questions of knowledge assessment questionnaire (Table. 2):-

The response of participants to question 1 showed that 76.5% of patients answered that they had good knowledge about risks of hypoglycemia and hypoglycemia first aid, 58.4% of subjects answered yes to question 2 as they have knowledge about the importance of hypoglycemia assessment by doctors.

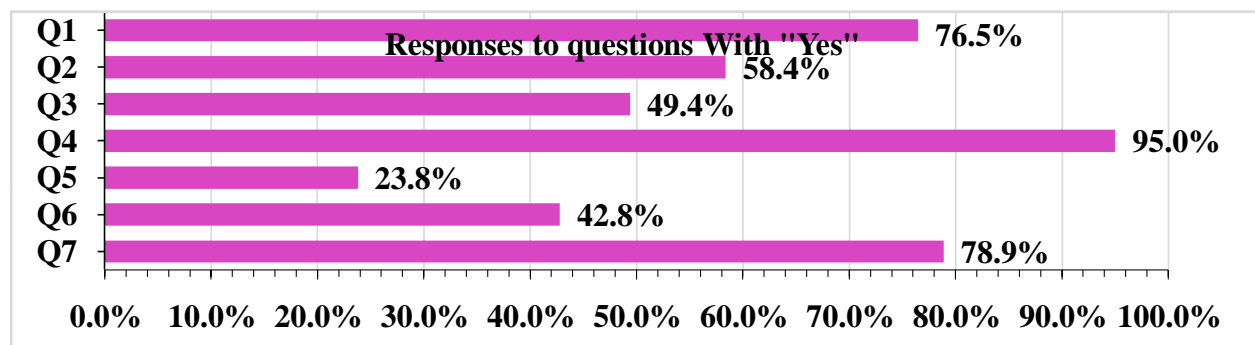
As for question 3, 50.6% of participants had no knowledge about the first aid hypoglycemia and 49.4% answered yes to this question.

Regarding to Q 4, 95% of subjects said yes to their knowledge about the treatment of diabetes have hypoglycemic effect on blood glucose. But 76.2% of participants said that the majority of doctors underestimate informing patients about the risks of diabetic therapy in Q 5.

57.2% of subjects said that doctors doesn't give them enough information about hypoglycemia and 42.8% had been given information about hypoglycemia in question 6. In question 78.9% of patients had knowledge about the complication of hypoglycemia (Figure. 1).

Table 2:- Responses to questions on assessment level of awareness toward hypoglycemia risk

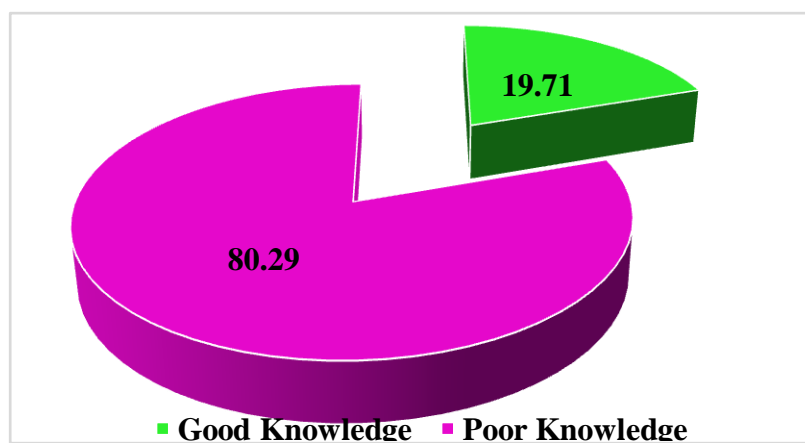
	No	Yes	Don't Know
Q1: Do you think that the hypoglycemia has risks?	99(23.50%)	322(76.50 %)	0 (0.0%)
Q2: Do you think that hypoglycemia assessment by doctors are important?	129 (30.60%)	246 (58.40%)	46(10.90 %)
Q3: Do you have knowledge about the first aid of hypoglycemia?	213 (50.60%)	208 (49.40%)	0 (0.0%)
Q4: Do you think treatment of diabetes have hypoglycemic effect on blood glucose?	21 (5.00%)	400 (95.00%)	0 (0.0%)
Q5: Do doctors clarify the impacts of diabetes therapy?	321 (76.20%)	100 (23.80%)	0 (0.0%)
Q6: Do doctors provide adequate information for people to avoid hypoglycemia ?	241 (57.20%)	180 (42.80%)	0 (0.0%)
Q7: Do you think hypoglycemia leads to sever complication can cause death?	89 (21.10%)	332 (78.90%)	0 (0.0%)



hypoglycemia risks (Figure. 2).

Table 3:- Knowledge of awareness toward first aid of hypoglycemia

	Knowledge Score
Mean± SD	4.25±1.44
Min.- Max.	0-7
Good Knowledge (≥ 5.69)	83 (19.71%)
Poor knowledge (< 5.69)	338 (80.29%)

**Figure 2:-** Respondent's Knowledge about first aid of hypoglycemia

Association between knowledge and demographics of included participants:-

Univariate logistic regression to study the association between knowledge and participant's demographics showed that, neither age nor other demographic variables showed significant association with being aware of hypoglycemic risks ($P > 0.05$). However, education level achieved statistical significance ($p < 0.0001$) association with knowledge as higher education resulted in good levels of hypoglycemia first aid (Table. 4). Interestingly, it was found that respondents who have completed their college degree have had a higher likelihood to have poor knowledge about hypoglycemia risks with an OR (95% CI) of 8.07 (3.11 - 20.94) when compared with those who get higher post-graduate degree (Master of PhD). Nearly the same difference was found in respondents, who have completed either high school or lower, with % of poor knowledge about (80.4%) when compared with individuals with high post graduate degrees (Figure. 3).

Table. 4:- Univariate logistic regression model for association between hypoglycemia knowledge and socio-demographic variables:

	Good Knowledge (n=83)	Poor Knowledge (n=338)	OR (95% CI)	P-value
Age				
<= 20.00	5(13.9%)	31(86.1%)	1	0.434
21.00 - 30.00	30(24.0%)	95(76.0%)	0.51(0.18-1.43)	0.201
31.00 - 40.00	8(14.3%)	48(85.7%)	0.97 (0.29-3.23)	0.957
41.00 - 50.00	9(19.1%)	38(80.9%)	0.68 (0.21-2.24)	0.527
51.00+	10(15.9%)	53(84.1%)	0.86 (0.27-2.73)	0.791
Gender				
Female	39(17.1%)	189 (82.9%)	1	0.115
Male	43(23.4%)	141(76.6%)	0.68 (0.42-1.1)	
Education Level				
High	14 (66.7%)	7 (33.3%)	1	< 0.0001
Intermediate	56 (19.9%)	226 (80.1%)	8.07 (3.11 - 20.94)	< 0.0001
Low	22 (19.6%)	90 (80.4%)	8.18 (2.95 -22.69)	< 0.0001
Monthly Income (SR)				
> 10,000 SR	31(19.4%)	129(80.6%)	1	0.68
3000-10000 SR	34(21.3%)	126(78.8%)	0.89 (0.52-1.54)	0.677
< 3000 SR	15(16.7%)	75(83.3%)	1.2 (0.61-2.37)	0.596
Marital Status				
Married	51(20.3%)	200(79.7%)	1	0.727
Un Married	32(18.9%)	137(81.1%)	1.09(0.67-1.79)	
Employment				
Employed	42(21.6%)	152(78.4%)	1	0.429
Un Employed	40(18.5%)	176(81.5%)	1.22(0.75-1.97)	

OR: Odds ratio, CI: Confidence Interval

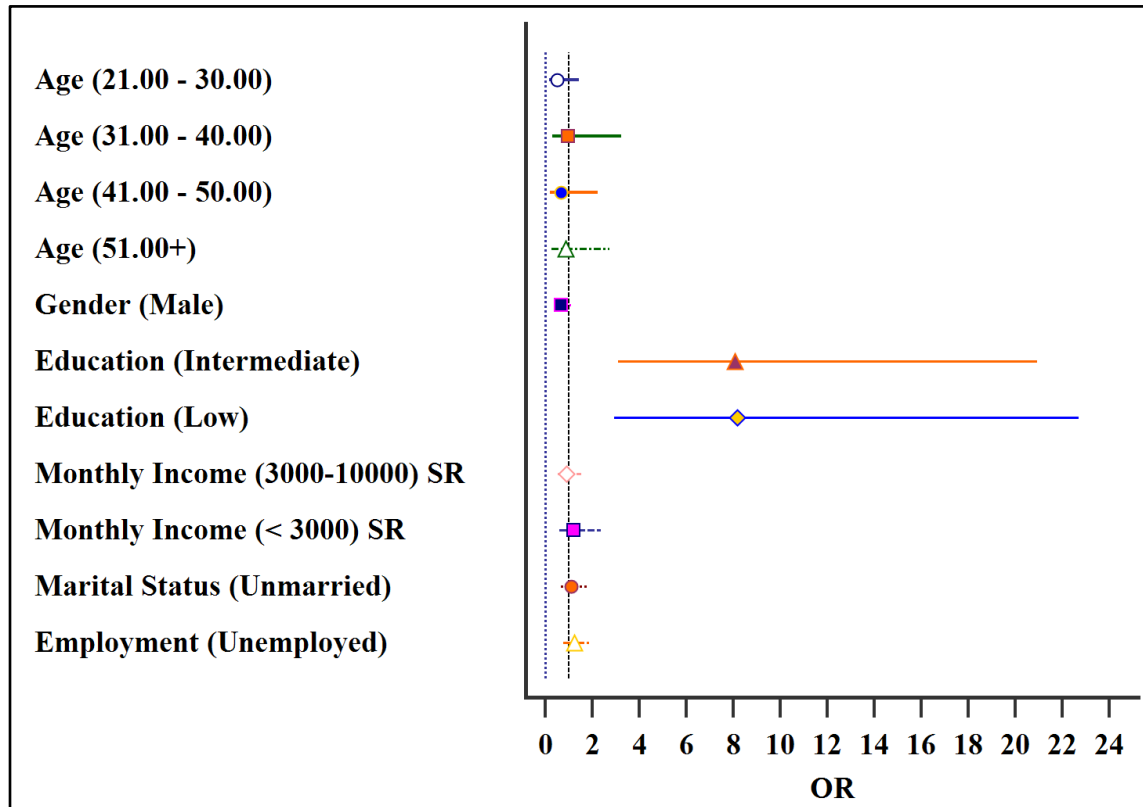


Figure 3:- Forest plot illustrating the odd ratios with 95% confidence intervals of the different socio-demographic predictors for Respondent's Knowledge about hypoglycemia first aid

Discussion and conclusion:-

This study has some limitations including that sample size in this research were educated group, and doesn't represent the whole population of the Riyadh And Jeddah cities thus the results can't be generalized. Also, Regardless of the sample size limitations, this study showed that there is a poor knowledge in the general population awareness about risks and first aid of hypoglycemia .

The response of participants to the questions showed that the majority had good knowledge about risks and first aid of hypoglycemia .

On the other hand the majority of doctors don't give the patients adequate information for risks nor first aid of hypoglycemia.

Thus doctors should provide patients with the necessary information to increase their awareness toward risks and first aid of hypoglycemia as it is a part of the responsibility of healthcare providers. Also, hypoglycemia significantly affected the general knowledge of the included participants.

In conclusion, the knowledge about risks of hypoglycemia and first aid of hypoglycemia was poor , thus the awareness about hypoglycemia and first aid of hypoglycemia must be increased. This study showed a poor awareness about hypoglycemia and first aid of hypoglycemia in the general population. Thus many studies should be conducted to provide the necessary information in order to increase of hypoglycemia and first aid of hypoglycemia awareness of population .

Budget:-

Item	Price
Transportations	700 SR
Paper work	800 SR
Software programs	2000 SR
Books	1000SR
Stationaries	1000SR

Work plan:-

Tasks in the work plan	Time period
Literature review	2 Months
Preparation for data collection	1 Months
Data collection	3 Months
Statistical analysis	1 Months
Discussion of results	2 months
Writing an abstract	1 months

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RESEARCH ARTICLE

PREVALENCE AND ANTIMICROBIAL RESISTANCE PATTERN OF METHICILLIN RESISTANT STAPHYLOCOCCUS AUREUS ISOLATED FROM DIFFERENT CLINICAL SAMPLES.

Dr. Nida Khan¹, Dr. Vineeta Khare, Dr. Rahmat Farid and Dr. Shadma Yaqoob.

Manuscript Info

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Abstract

Background: Methicillin Resistant *Staphylococcus aureus* (MRSA) since its emergence, has been a challenge for the community owing to its potential to cause life threatening events like sepsis, endocarditis and osteomyelitis.

Aims and objectives: To investigate the prevalence of MRSA in various clinical samples and to find the antibiotic resistance pattern of the MRSA isolates.

Materials and Methods: Oxacillin resistance screening agar with 5.5% NaCl and Kirby–Bauer disk diffusion method (cefoxitin 30µg disk) were used for MRSA confirmation and antibiotic resistance testing was done as per CLSI 2016 guidelines. E-test was done for testing sensitivity to Vancomycin.

Result: *Staphylococcus aureus* was isolated from 350 specimens. Out of these 350 isolates, 220 (62.86%) were methicillin sensitive and rest 130 (37.14%) isolates were MRSA. Most of the MRSA isolates were from urine samples (43.71%) followed by pus (24.0%), and putum (11.14%). MRSA isolates showed high resistance to ciprofloxacin (53.38%), and clindamycin (42.31%). linezolid resistance was seen in only 6.15% and all isolates were sensitive to vancomycin.

Conclusion: A high prevalence of MRSA (37.14%) in our institution warrants the judicious use of antibiotics in treating infections caused by *Staphylococcus aureus*. Vancomycin and linezolid are good treatment options in infections caused by MRSA isolates.

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Introduction:-

The genus *Staphylococcus* is one of the most commonly encountered group of pathogens in both community-acquired as well as hospital-associated infections. Within genus *Staphylococcus*, *Staphylococcus aureus* is the most important and prevalent organism isolated in clinical specimen¹. The organism also exists as a commensal, colonizing the anterior nares of about one third of the healthy human population. Asymptomatic nasal carriers are at a high risk of subsequent *S. aureus* infection and are presumed to be an important source of strains that spread and cause infection in contacts^{2,3,4}.

This centuries-old pathogen is found in almost all the varieties of clinical specimen ranging from minor skin infections to fatal necrotizing pneumonia⁵ and still continues to be the cause of significant morbidity and mortality despite huge advances in medical care *S. aureus* resistance to methicillin was reported for the first time in 1961 in

England, and since then it continues to be the most dreadful strains of *S. aureus*. It is one of the most common pathogens that cause nosocomial infections^{6,7}

In India, a multicentric trial conducted between January 2008 to December 2009 at several healthcare centres⁸ found MRSA prevalence among specimen collected from outpatients, ward inpatients and ICU to be 28, 42 and 43 per cent, respectively in 2008 and 27, 49 and 47 per cent, respectively in 2009, thus indicating that MRSA has assumed a concerning proportion. Moreover, they stressed on the pattern of changing antibiotic susceptibility and also recommended robust antimicrobial stewardship and strengthened infection control measures to prevent spread and reduce emergence of resistance.

It must be kept in mind that current therapeutic options for MRSA are limited few expensive drugs like vancomycin, linezolid, teicoplanin, daptomycin and streptogramins. Another alarming sign is that emergence of resistance to Vancomycin, although at a low level has been reported in literature⁹, thus underlining the observations made by the multicentric study cited above.

Hence, the present study was planned with an aim to assess the prevalence and antibiotic susceptibility of Methicillin resistant *Staphylococcus aureus* at Era's Lucknow Medical College and Hospital (ELMCH)

Aims and Objectives:-

The present study was carried out with an aim to evaluate the prevalence, antibiotic susceptibility of MRSA in a tertiary care centre. This aim was fulfilled with the help of following objectives:

1. To investigate the prevalence of MRSA in various samples obtained for culture.
2. To find the antibiotic resistant pattern of MRSA isolates

Material And Methods:-

Place Of Study Study was conducted at Department of Microbiology, Era's Lucknow Medical College.

Duration:

12 months from Nov 2014 to Oct 2015

Study Population:

Part I: All patients visiting Era's Lucknow Medical College and Hospital, Lucknow.

Sampling Frame:-

Sample size: 350

Inclusion criteria:-

All patients including outpatients and admitted patients

Exclusion criteria:-

Patients who did not show cooperative attitude or refuse to provide necessary information were excluded.

Sample Collection:-

Part I: The sample collection was done at different clinical Department of Microbiology, Era's Lucknow Medical College, Lucknow using standardized sampling techniques. The specimen obtained from different sources were labeled and tagged with information

Lab Diagnosis:-

Microscopy:-

Gram staining

Culture was done on following media:

Blood agar

Mac Conkey agar

MRSA confirmation by:-**Oxacillin resistance screening agar with 5.5% NaCl:-**

Suspend 51.75 grams in 500 ml distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool at 45-50°C and aseptically add rehydrated contents of 1 vial of Oxacillin Resistance Selective Supplement (FD191). Mix well and pour into sterile Petri plates.

Disc diffusion method [Kirby bauer (cefoxitin 30 mg disk)]¹⁰

Procedure (As per manufacturer's instructions):-

Bacterial Suspension was prepared according to 0.5 McFarland standard and results were interpreted as Resistant, Intermediate or Sensitive for each antimicrobial using CLSI guidelines.

E-TEST: Vancomycin EzyMIC strips (HiMedia) were used to test for vancomycin sensitivity over a range of MIC from 0.016-256 mcg/ml. The E Test was done and interpreted as per manufacturer's instructions. Apply the Etest strip to the agar surface with the MIC scale facing upwards. MICs were read where the edge of inhibition ellipse intersected the strips.

Statistical Tools Employed:-

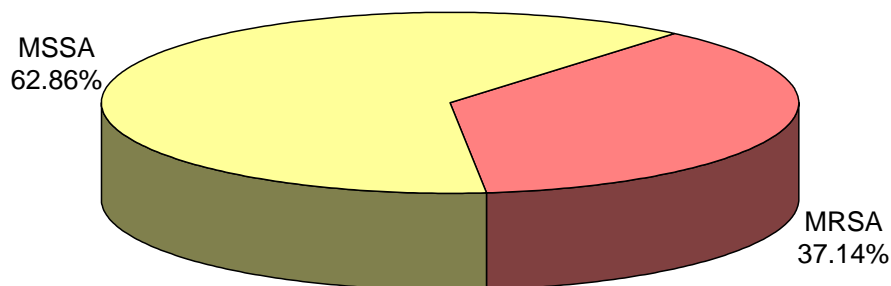
The statistical analysis was done using SPSS (Statistical Package for Social Sciences) Version 15.0 statistical Analysis Software. The values of mean, standard deviation and p value were calculated and a p value of $p < 0.05$ was considered significant. Chi square test was done to differentiate between MRSA and MSSA.

Results:-

A total of 350 specimens from the patients were collected which were subjected to MRSA sensitivity.

Table 1:- Distribution of Cases according to MRSA status (n=350)

	Number of cases	Percentage
MRSA	130	37.14
MSSA	220	62.86



Out of 350 specimens, 220 (62.86%) were found to be MSSA and only 130 (37.14%) specimens were found to be MRSA. Prevalence of MRSA in our tertiary care centre was found to be 37.14.

Table 2:- Comparison of Type of specimen between MRSA and MSSA cases

	Total	MRSA (n=130)		MSSA (n=220)		Statistical significance	
		No.	%	No.	%	χ^2	p
BC	5	5	3.85	0	0.00	47.289	<0.001
Blood	15	2	1.54	13	5.91		
Body fluid	30	2	1.54	28	12.73		
Catheter tip	24	10	7.69	14	6.36		
Pus	84	51	39.23	33	15.00		
Sputum	39	11	8.46	28	12.73		
Urine	153	49	37.69	104	47.27		

Out of 350 specimens, most common specimen was Urine (n=153; 43.71%), followed by Pus (n=84; 24.0%), Sputum (n=39; 11.14%), Body fluid (n=30; 8.57%). 15 (4.29%) were blood specimens and 5 (1.43%) were from blood culture (BC).

Proportion of MRSA specimens was higher than MSSA for Blood components – Blood culture (3.85% vs. 0.0%), Catheter tip (7.69% vs. 6.36%), Pus (39.23% vs. 15.00%).

Table 3: Distribution of MRSA positive specimen according to Method of Identification (n=130)

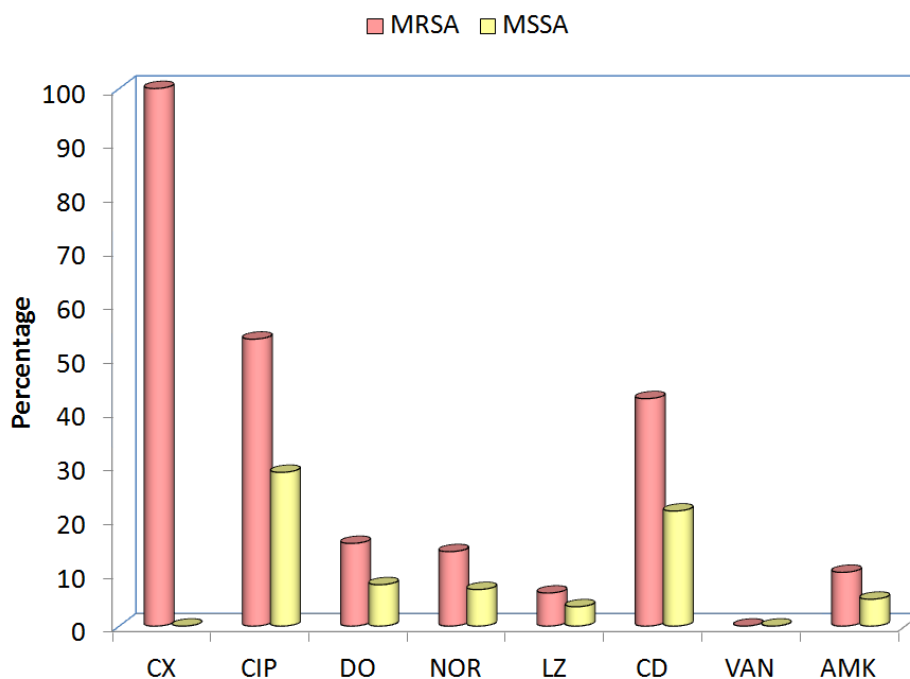
SN	Method	No. of specimen identified	Percentage
1.	Disc diffusion method : Kirby Bauer (cefoxitin 30mg disk)	130	100
2.	Oxacillin resistance screening agar with 5.5% NaCl	90	69.2
3.	Both	90	69.2

All the MRSA samples were identified by Kirby-Baur disc diffusion method (cefoxitin 30 mg). Using Oxacillin resistance screening agar with 5.5% NaCl helped in identification of 90 (69.2%) specimen. Both the methods identified MRSA in 90 (69.2%) specimens

Difference in type of specimens between MRSA and MSSA was found to be statistically significant ($p < 0.001$).

Table 9:- Comparison of Resistance of MRSA and MSSA cases

Resistant	MRSA (n=130)		MSSA (n=220)		Statistical significance	
	No.	%	No.	%	χ^2	P
CEFOXITIN	130	100.00	0	0.00	350.00	<0.001
CIPROFLOXACIN	70	53.38	63	28.6	22.04	<0.001
DOXYCYCLINE	20	15.38	17	7.7	5.068	0.024
NORFLOXACIN	18	13.85	15	6.8	4.726	0.030
LINEZOLID	8	6.15	8	3.6	1.187	0.276
CLINDAMYCIN	55	42.31	47	21.4	17.37	<0.001
VANCOMYCIN	0	0.00	0	0	-	-
AMIKACIN	13	10.00	11	5.0	3.198	0.074



All the MRSA specimen were resistant against Cefoxitin. The resistance rate in decreasing order was Ciprofloxacin (53.38%), Clindamycin (42.31%), Doxycycline (15.38%), Norfloxacin (13.85%), Amikacin (10%) and Linezolid (6.15%). Vancomycin was sensitive against 100% of MRSA isolates.

All MSSA specimen were sensitive against Cefoxitin. The resistance rate in decreasing order was Ciprofloxacin (28.6%), Clindamycin (21.4%), Doxycycline (7.7%), Norfloxacin (6.8%), Amikacin (5%) and Linezolid (3.6%). Vancomycin was sensitive against 100% of MRSA isolates.

Discussion:-

Methicillin resistant *Staphylococcus aureus* since its emergence has been a challenge for the healthcare workers owing to its potential to cause life-threatening events like sepsis, endocarditis, and osteomyelitis. Major outbreaks of MRSA and its different phage types have also been recorded and reported in healthcare facilities¹¹. Since resistance to multiple antibiotics among MRSA isolates is very common, there is a possibility of extensive outbreaks, which may be difficult to control. MRSA is now one of the commonest nosocomial pathogens, and asymptotically colonized healthcare workers are the major sources of MRSA in the hospital environment. Early detection of MRSA and formulation of effective antibiotic policy in tertiary care hospitals is of paramount importance from the epidemiological point.

In our study a total of 350 clinical specimen positive for *Staphylococcus aureus* were obtained and assessed for methicillin resistance. Out of these 130 (37.14%) were found to be Methicillin resistant. Thus prevalence of Methicillin resistant *S. aureus* was found to be 37.14% in present study. Prevalence of MRSA has been shown to vary substantially in some contemporary clinical series from the region. Table D1 shows the prevalence of MRSA in some contemporary clinical series:

Table D1:- Prevalence of MRSA in some contemporary clinical series

SN	Author (Year)	Sample size and characteristic	MRSA prevalence rate
1.	Saikia <i>et al.</i> (2009) ¹² Dibrugarh, Assam	276 Clinical specimen	34.78
2.	Ahmad <i>et al.</i> (2009) ¹³ , Armed Forces Hospital, Saudi Arabia	106 Specimen collected from different hospital	22.3%
3.	Tiwari <i>et al.</i> (2011) ¹⁴ , Bhubaneswar, India	204 Clinical samples	55.8%
4.	Sharma <i>et al.</i> (2013) ¹⁵ , Mangalore, India	685	23.25%

SAAn overview of Table D1 above shows a wide variability in clinical prevalence rate of MRSA in different studies.

In present study, Kirby Bauer disc diffusion method (Cefoxitin 30 mg) proved to be more sensitive than Oxacillin resistance agar method for detection of MRSA. This finding is in accordance with the observations made by Datta *et al.* who also showed that Cefoxitin disc diffusion is more sensitive than Oxacillin resistance agar method in the detection of MRSA¹⁶.

In present study, pus was the most common source of MRSA (39.23%) followed by urine (37.69%), sputum (8.46%) and catheter tip (7.69%) respectively. Saikia *et al.* (2009)¹² also showed maximum isolation of MRSA from pus/wound swabs (46.67%) followed by sputum/throat swab (42.86%) while Ahmad *et al.*¹³ found source of MRSA to be 22.2% from pus, 23.8% from wound swabs, 33.3% from aspirates and 13.3% from sputum. The findings of present study are in accordance with the observations of Tiwari *et al.*¹⁴ (2011) who reported pus (45%) followed by urine (20.5%) to be the major source of MRSA in their study. With slight difference in proportions, these two sources comprise the major source of MRSA in our study too. In present study, MRSA positivity rate in pus samples was significantly higher (51/84; 60.7%) as compared to other specimen (79/266; 29.7%). This finding is in accordance with some other studies too that have also reported pus to have a higher MRSA positivity rate as compared to other specimen^{12,14,15}.

MRSA specimens were most sensitive against Vancomycin (100.0%) and Linezolid (93.85%). Cefoxitin, Ciprofloxacin and Clindamycin showed maximum resistance (100%, 53.38% and 42.31% respectively). The antibiotic susceptibility has been reported to vary substantially in different studies. Goyal *et al.* (2013)¹⁷ showed from a study conducted in a teaching hospital at Agra that MRSA specimens were most sensitive against

Vancomycin, Teicoplanin and Linezolid (100%), Ampicillin, Erythromycin and Chloramphenicol showed maximum resistance (100%, 88%, 66.7% respectively). Another study conducted by Sharma et al (2013)¹⁸ from Mangalore, India showed most of the MRSA strains were sensitive against Linezolid (98.27%) and Ciprofloxacin (64.7%), Tetracycline (58.38%) showed maximum resistance. The findings of the present study are in accordance with Tiwari et al (2011)¹⁹ Bhubaneswar, India who showed all the isolated MRSA strains were sensitive against Vancomycin and Linezolid (100%). Vancomycin and Linezolid are most sensitive drugs with most of the studies reporting their sensitivity rates between 90% to 100%. The findings of present study also emulated the same and showed vancomycin and linezolid to be 100% and 93.85% sensitive. For other conventional antibiotics different studies have shown a high resistance pattern. A number of studies have also reported multidrug resistance. In present study too for conventional antibiotics the resistance rates were ranged from 42.31% to 100%. A number of specimens were polypersistent too.

These findings in turn indicate the need to develop an antibiotic protocol dependent on MRSA profile of the pathogen in order to reduce the unnecessary burden of antibiotics.

Conclusions:-

The present study was carried out in the Department of Microbiology, Era's Lucknow Medical College & Hospitals to evaluate the prevalence, antibiotic susceptibility and carriage rate of MRSA in a tertiary care centre

1. Out of 350 specimens during the study period, 220 (62.86%) specimens were found to be MSSA and 130 (37.14%) were MRSA. Prevalence of MRSA in our institution was 37.14%.

2- Proportion of MRSA specimens was higher than MSSA for Blood culture (3.85% vs. 0.0%), Catheter tip (7.69% vs. 6.36%), Pus (39.23% vs. 15.00%) while proportion of MSSA was higher than MRSA for body fluid (12.73% vs. 1.54%), Sputum (12.73% vs. 8.46%) and Urine (47.27% vs. 37.69%).

3-MRSA specimens were most sensitive against Vancomycin (100.0%) and Linezolid (93.85%) while least sensitive for Cefoxitin (0.0%).

4- 90 isolates were detected by oxacillin resistance screening agar and 130 isolates were detected by Kirby Bauer disk diffusion method. Thus Kirby Bauer method provided an addition of 30.8% in overall MRSA detection

The findings of present study, thus showed that MRSA was prevalent in our hospital. There is a progressive increase in MRSA positivity and multi-drug resistance in strains of *Staphylococci*. Vancomycin and linezolid were found to have absolute sensitivity.

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RESEARCH ARTICLE

ASSOCIATION BETWEEN GRAIN YIELD AND YIELD COMPONENTS OF MAIZE DURING INBREEDING SYSTEM.

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Manuscript Info

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Abstract

Evaluation of cobs-to-row progenies raised from previously selected 800 progenies for their early vigour at 30 DAS and grain yield and its components at maturity indicated the presence of high degree of variation for leaf area and leaf width compared to seedling girth and seedling height. Further median and mode values of traits related to early vigour exhibited negative skewed distribution with a platykurtic pattern indicating greater scope of selection among the progenies for improving early vigour. Among the traits studied at the time of maturity cob weight, pith weight, number of kernels per cob and grain yield per plant showed high coefficient of variability. Plant height and shelling percentage exhibited very low coefficient of variation.

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Introduction:-

Maize (*Zea mays* L.; $2n = 20$) is an important cereal crop with high yield potential. The total production at world level has surpassed both sorghum and pearl millet gaining a third place after wheat and rice. The demand for maize grain is increasing every year due to its diversified use in poultry, piggery and industry. In India, about 50-55 per cent of maize production is consumed directly as food, 30-35 per cent for poultry, piggery and fishmeal and 10 to 12 per cent in wet milling industry viz., in starch and oil and about 3 per cent in dry milling for traditional requirement like *Dalia* and *Sattu*. A number of genotypes viz., single cross, three way cross, double top cross, double cross hybrids, composites, synthetic population are being cultivated by farmers.

The efficiency and success of hybrid maize breeding depends on the contribution of heterotic effects from the superior inbred lines. The superior cross combination may be exploited as commercial single cross. Usually, the hybrid development programme in maize involves development of inbred lines, evaluation of inbred lines, crossing of selected inbreds and production of hybrids. In this context, a programme on development of inbred lines was initiated by using inbreeding system utilizing yellow pool as the base population. Yellow pool is a blend of 50 elite lines received from Directorate of Maize Research (DMR), New Delhi and inter mated for four cycles at Agricultural Research Station, Arabhavi. The S_2 lines derived from this population were evaluated by line \times tester analysis involving four diverse testers in to know their performance, combining ability and nature of inheritance of different quantitative traits. The line \times tester ($L \times T$) design is basically an extension of top cross analysis involving more than one tester (Kempthorne, 1957). Thus, keeping these views in background, the proposed work was undertaken to develop hybrids suitable for *kharif* season.

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***Part of the Ph.D thesis submitted to the University of Agricultural Sciences, Dharwad**

Material and methods:-

Fifty elite breeding lines (Table 1) were received from Directorate of Maize Research (DMR), IARI, New Delhi for development of full season heterotic yellow pool. These lines were raised and allowed to open pollinate for four generations (intermating) without exercising any selection. In each generation, the plants were raised in half-sib family method and at the end of fourth generation, the seeds were collected and grown as base population at College of Agriculture, Dharwad in isolation and allowed to open pollinate. Sufficient care was taken to maintain more than 10 thousand plant population. Out of this population, 150 plants were tagged in each grid by dividing entire population into 10 grids of 150 sq. mt. at the time of tassel emergence, based on vigour, standing ability, silk and tassel traits and free from disease incidence, 800 cobs were selected based on grain yield (*per se*) performance and this material formed basis for further inbreeding and isolation of inbred lines.

The experiments were laid out at Agricultural Research Station, Arabhavi and the experimental details are mentioned below under.

After thorough land preparation, hand dibbling was done with two seeds per hill and thinned out to maintain single plant per hill during second week after sowing (10-15 days after sowing). The crop was applied with recommended dose of fertilizers as per the package of practices.

Observations On Quantitative Traits:-

In these investigations, data were collected in respect of quantitative traits. Observations on quantitatively inherited traits were recorded on five competitive randomly selected plants. The average was taken as the mean of the treatment. The way in which observations were recorded is described below.

Plant height at 60 days after sowing (cm) :-

Plant height was expressed in centimetres by measuring the plant stalk from the ground level to the base of the last leaf sheath of the plant.

Ear height at 60 days after sowing (cm) :-

This observation was recorded at the same time as plant height (at harvest), measured as the height of the plant from the ground level to the uppermost ear bearing node and expressed in centimetre.

Leaf area at 60 days after sowing (cm²) :-

The average of five leaves from five different plants at 60 days after sowing was calculated by using formula $L \times B \times 0.75$ and expressed in square centimetre (Montgomery, 1911).

Ear weight (g) :-

Five ears were sun dried and weight in grams was recorded at stable moisture content.

Ear length (cm) :-

Length of the ear was measured and recorded in centimetres at the time of harvest as its total length (from the base to the tip of the ear).

Ear circumference (cm) :-

Ear circumference at one-third length from the base of the ear was measured and recorded in centimetre.

Number of kernel rows per cob:-

Number of kernel rows in each ear was counted for five cobs and mean was worked out.

Number of kernels per row:-

The number of kernels in any one of the row in each ear was counted and mean was worked out.

Number of kernels per ear:-

Total number of kernels in each ear of the selected plants was recorded.

Hundred grain weight (g) :-

One hundred randomly selected grains per cob were weighed and weight was recorded in grams.

Shelling percentage:-

The ratio of grain weight to the ear weight was expressed in percentage.

$$\text{Shelling percentage} = \frac{\text{Grain weight}}{\text{Cob weight}}$$

Pith / shank / heart weight (g) :-

The total pith / shank / heart weight from each ear was recorded and the average heart weight was expressed as heart weight per plant in grams.

Kernel yield per plant (g) :-

At the time of harvesting, fresh ear weight was recorded in grams per plant. Moisture determinations were made from shelled samples from five random ears of each plot with the help of electronic moisture meter. The fresh ear weight data was used to work out the dry weight at 15 per cent moisture grain yield in grams per plant.

Kernel yield per plot (kg/plot) :-

At the time of harvest, fresh ear weight was recorded in kilograms for each plot. This fresh ear weight was adjusted to zero per cent grain moisture and recorded as grain yield per plot. Moisture percentage was determined on universal moisture tested in a grain sample obtained from two rows of each of five ears selected at random at the time of harvest were used for genetical analysis following Combstock (1948) and Robinson (1968).

$$\text{Yield per plot (kg/plot)} = \frac{\text{Fresh ear weight} \times (100 - \text{AVM})}{100}$$

Where,

$$K = \frac{\text{Average stand/plot of the trial} \times \text{plot area} \times 0.9412}{\text{Final stand}}$$

AVM = Average moisture

100 × plot area

Kernel / grain yield per hectare (q/ha):-

Fresh dehusked ear weight per plot was recorded at the time of harvest. Moisture content of 100 grams of fresh kernels was recorded by moisture meter of each entry and the moisture percentage was calculated by using moisture conversion charts. Kernel yield per hectare was calculated using the formula given below:-

$$\text{Yield (q/ha)} = \frac{\text{Fresh ear weight} \times 100 - \text{AVM}}{100} \times K$$

Where,

$$\text{K} = \frac{\text{Final stand harvest}}{\text{Average stand of the trial} \times \text{Area (ha)} \times 0.9412}$$

Statistical Analysis:-**Analysis of variance:-**

The data were subjected to statistical analysis for randomised block design (RBD) as described by Panse and Sukhatme (1962). The mean values of the entries in each replication were used for analysis of variance (ANOVA). The significant difference among genotypes was tested by 'F' test at one per cent and five per cent levels of probability.

Genotypic and phenotypic correlation coefficients:-

Genotypic and phenotypic correlation coefficients were worked out as per the methods suggested by Johnson *et al.* (1955) to find out the relationship between yield and its contributing characters. Analysis of covariance was done similar to that of analysis of variance taking two characters at a time. The genotypic and phenotypic co-variances were derived as detailed for genotypic variance. The genotypic and phenotypic variance and co-variance components were utilized to calculate genotypic and phenotypic correlation coefficients (r) by applying the formula.

$$r = \sqrt{\frac{\text{Cov. g. 1,2}}{(\sigma^2_{g1}) \times (\sigma^2_{g2})}} \text{ and } \sqrt{\frac{\text{Cov. p. 1,2}}{(\sigma^2_{p1}) \times (\sigma^2_{p2})}}$$

Where,

Cov.g. 1:2 : Genotypic covariance between two traits (1 and 2)

Cov.p. 1:2 : Phenotypic covariance between two traits (1 and 2)

$\sigma^2_{g_1}$: Genotypic covariance for first trait

$\sigma^2_{p_1}$: Phenotypic covariance for first trait

$\sigma^2_{g_2}$: Genotypic covariance for second trait

$\sigma^2_{p_2}$: Phenotypic covariance for second trait

Referring to correlation table of Snedecor and Cochran (1967), the significance of phenotypic correlation coefficient was tested.

Results:-

Association studies in S_1 progenies:-

Among all traits, selection for seed yield is a complex phenomenon that involved interaction among different growth and yield contributing character of a genotype. In the present investigation an attempt was made to understand association of different traits at population level contributing towards yield improvement.

The phenotypic and genotypic correlation of grain yield with 16 other yield components among S_1 progenies were worked out and the same is presented in Table 2 and 3.

A general observation of the data across different characters under study revealed that seed yield exhibited significant positive correlation with few yield traits, but also exhibited significant negative association with few other traits. The results obtained on the basis of association of individual trait with grain yield are presented below.

Plant height at 60 days after sowing (cm):-

Plant height at 60 DAS showed highly significant association in positive direction with other traits under study except for shelling percentage which had non-significant association with this trait (0.086) only at genotypic level (Table 2). Whereas highly significant phenotypic association was recorded in desirable direction, except for number of kernels per rows (0.112) which expressed only significant association in positive direction (Table 3).

Ear height (cm):-

All the traits exhibited highly significant positive association with the ear height, except shelling percentage (0.072) which was non-significant at genotypic level (Table 2). A highly significant positive phenotypic association was recorded for all the traits studied, except for plant girth at 30 DAS (0.108), leaf area at 30 DAS (0.121), number of kernel rows per cob (0.110) and number of kernels per row (0.123), at five per cent level of probability (Table 3).

Leaf area at 60 days after sowing (cm²):-

The data revealed significant positive genotypic association with all traits except for number of kernel rows per cob (0.028), shelling percentage (0.082) and number of grains per cob (0.184) which recorded non-significant association (Table 2).

A highly significant positive phenotypic association was recorded for all the traits studied, except for number of kernel rows per cob (0.036) and shelling percentage (0.070) (Table 3).

Cob weight (cm) :-

All the traits exhibited highly significant association with cob weight at both genotypic and as well as phenotypic level in the desirable direction (Table 2 and 3).

Cob length (cm) :-

The trait cob length exhibited highly significant positive genotypic association with all the characters, except for number of kernel rows per cob (0.062) and shelling percentage (0.061) (Table 2). A similar association of cob length with all the characters at phenotypic level was exhibited except for plant height at 30 DAS (0.122), number of kernel rows per cob (0.068) and shelling percentage (0.053) (Table 3).

Cob girth (cm) :-

It is interesting to note that all the traits under study exhibited highly significant association in the desirable direction in both genotypic as well as phenotypic level with cob weight (Table 2 and 3).

Number of kernel rows per cob:-

Majority of the characters exhibited highly significant positive genotypic association with number of kernel rows per cob, except for the traits like, number of kernels per row (0.101), leaf area at 60 DAS (0.028) and cob length (0.062) (Table 2). Similarly, a highly significant positive phenotypic association was recorded for all the traits studied, except the characters like plant girth at 30 DAS (0.117), leaf area at 30 DAS (0.127), ear height (0.110), number of kernels per row (0.103) and shelling percentage (0.125), which recorded significance only at five per cent level of probability (Table 3). But, the traits such as leaf area at 60 DAS (0.036) and cob length (0.068) exhibited non-significant association.

Number of kernels per row:-

A highly significant positive genotypic association with number of kernels per row was observed for all the characters under study, except for number of kernel rows per cob which exhibited significance at five per cent level of probability in positive direction (0.101) (Table 2). Similarly, with phenotypic correlation, trait under study had highly significant positive association with all the characters except for plant girth at 30 DAS (0.102), plant height at 60 DAS (0.112), ear height (0.123) and number of kernel rows per cob (0.103) which exhibited only a positive significant association at five per cent level of probability (Table 3).

Pith weight (cm):-

The study revealed that the trait pith weight exhibited highly significant association at one per cent and five per cent level of probability with all the traits under study at genotypic and phenotypic levels, except with shelling percentage which had significance in negative direction (Table 2 and 3).

Hundred seed weight (g):-

From the results of present study, it is clear that all the characters exhibited highly significant association with 100-seed weight at both genotypic and phenotypic levels, except for number of kernel rows per cob (0.121) which recorded positive association with phenotypic level at five per cent level of probability (Table 2 and 3).

Shelling percentage (%):-

The character shelling percentage exhibited highly significant positive genotypic association with all the traits under study, except for plant height at 30 DAS (0.115) which recorded only significant positive association (Table 2). The characters plant girth at 30 DAS (0.039, 0.009), plant height at 60 DAS (0.086, 0.075), ear height (0.072, 0.056), leaf area at 60 DAS (0.082, 0.070) and cob length (0.061, 0.053) exhibited non-significant association, whereas pith weight (-0.450, -0.450) recorded highly significant negative association at both genotypic and phenotypic levels (Table 2 and 3). Similarly at phenotypic level, a highly significant positive association was recorded for all the traits, except for plant height at 30 DAS (0.103), leaf area at 30 DAS (0.102) and number of kernel rows per cob (0.125) at five per cent level of probability (Table 2 and 3).

Number of kernels per cob:-

Number of kernels per cob recorded highly significant positive association at both genotypic and phenotypic levels, except for leaf area at 60 DAS (0.184) which showed non-significant association at genotypic level (Table 2 and 3).

Grain yield per plant (g):-

In maize, increase in grain yield is mainly achieved by manifestation of many characters like number of kernel rows per cob, number of kernels per row, cob length, cob girth, 100-seed weight, *etc.* Therefore it is necessary to understand the nature of association existing between all characters and grain yield, particularly in early generation (S_1).

Grain yield per plant had highly significant positive association with all the characters at both genotypic and phenotypic levels, except for plant girth at 30 DAS (0.393) which showed significance only at five per cent level of probability at genotypic level (Table 2 and 3).

Discussion:-

In general genotypic correlation was higher in magnitude than that of phenotypic correlation. Similar trend of higher values of genotypic correlation coefficients than phenotypic coefficient correlation was also reported in earlier studies. Muhammad Rafique *et al.* (2004) reported higher GCV in case of grain yield per plant followed by plant height, ear height, kernels per row, 1000-kernel weight and ear length. Number of kernels per row had significant

correlation with seed yield followed by plant height, ear height, cob diameter and length of cob. Similar results were also reported by earlier worker, Khatum *et al.* (1999) and Devi *et al.* (2001).

Grain yield per plant exhibited positive association with all growth and yield parameters in the present investigation. Most of the growth and yield parameters are inter dependent as evidenced by significant and positive correlation among themselves. However, number of kernel rows per cob was not associated significantly with leaf area and cob length indicating these as independent traits. Likewise shelling percentage was also found to be independent of plant girth at 30 DAS, plant height at 30 DAS, ear height at 30 DAS, leaf area at 60 DAS and cob length as it has recorded non-significant correlation with these traits. As expected pith weight exhibited significant and negative association with shelling percentage, which are interdependent among the productivity traits. Cob weight, cob length, cob girth, grain number per cob, number of kernels per cob, 100-seed weight and shelling percentage recorded high correlation coefficient more than 0.50 indicating these are important yield contributing traits. Similar reports were also made by Kumar (1974), Bhalla and Khehra (1977) and Ramamurthy (1980).

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RESEARCH ARTICLE

DRY NEEDLING TECHNIQUE IN MYOFASCIAL PAIN - CLINICAL CASE REPORT.

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Abstract

Myofascial pain characterized by the presence of myofascial trigger points (TP) that can cause local pain and referred pain, that is, perceived in a different location. TP is a palpable nodule and a painful point of compression, which, in addition to local pain, promotes pain. It is called referred pain when pain caused by TP also affects other regions, such as: other muscles, teeth, gums, TMJ and ears. It is of fundamental importance the anatomical knowledge of the muscles, since it facilitates the localization of the TP and palpation, as well as the functional knowledge of the muscles, since the TP can be activated when the patient makes a certain movement / gesture, this facilitates the correct diagnosis. When the procedures performed reach the TP areas, muscle pain is rapidly eliminated, restoring function and normal muscle length. One of the most widely used treatments has been dry pressing, the theory is that the needle through mechanical trauma stimulates the release of intracellular potassium, which blocks the transmission of nerve impulses temporarily. The needle also breaks the adhesions of the fibers that may exist. The objective of this work is to report two cases of myofascial pain treatment using the dry needling technique. Where, Ms D.S.R and Ms O.H.S.R. attended the UNIFEB clinic, with symptoms of muscular pain and difficulty opening the mouth and chewing, dry needling was performed and there was instant improvement of the pain picture, and improvement in movements that were previously limited.

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Introduction:-

Temporomandibular Disorders (TMDs) involve a series of changes, painful or not, in the Temporomandibular Joint (TMJ) region, chewing muscles, or both [1]. TMD pains, especially those in the temporalis muscle (temples) or in the joint itself, can produce a sensation of pain in the head due to the location of these structures. It should be clear that such manifestation is not a primary headache, that is, those of neurological origin. In the case of TMD pain, a classic manifestation is the worsening of pain with the use of the jaw to speak, to feed, among others [2].

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One of the most debated topics today is chronic orofacial pain that according to the World Health Organization (WHO) [1] it is estimated that this type of pain reaches 30% of the world population. According to the Brazilian Society for the Study of Pain, women feel more pain and are at greater risk of experiencing numerous painful conditions. In this way, pains in the neck and shoulders, abdomen, tension-type headaches, migraine after puberty, temporomandibular joint (TMJ) disorders are more common in women.

In men, the risk of painful manifestations of this type is lower and one of the great novelties in the research is the signaling that testosterone protects males in relation to TMJ pain, where a difference in the biological mechanisms of TMJ pain is also observed with respect to other parts of the body [3]. In Brazil, it is estimated that 50 million people feel this type of pain, and that at some point 15 to 25% of people will suffer from chronic pain, and in individuals over 65 this rate can increase to 50% .

Myofascial pain syndrome is the most common musculoskeletal dysfunction (6), this is common in clinical practice where about 10% of the population has one or more disorders of the musculoskeletal system [4,5,6,7]. This syndrome is characterized by localized or referred muscle pain that is originated in a myofascial trigger point [9]. The term trigger point (TP) describes a nodule in the musculature located mainly at the end of the muscle insertion, where a hypersensitive and painful area is formed [8,10]. However, the mechanisms of TP formation are hardly evident. It is known that local muscle pain is related to the activation of muscle nociceptors for a variety of endogenous substances including neuropeptides, arachidonic acid derivatives, inflammatory mediators and others.

The therapeutic approach of myofascial pain syndrome is extremely difficult due to the difficulty of diagnosis, which occurs in only 15% of cases [12]. The treatment for myofascial pain syndrome is broad and has many techniques, most commonly used: ischemic pressure (IP), dry needling (DN), wet needling, passive stretching, stretching and spraying, TENS, massage [12], ultrasound and laser [8, 14, 15]. In these techniques DN and IP appear to be very effective in the treatment of myofascial pain syndrome [15,16,17].

The DN, a technique similar to acupuncture, is an effective treatment for pain relief. This technique may increase pain threshold and range of motion on return of pain. Hsieh et al. [16] provided evidence that acute dry-needle inactivation of a primary TPs inhibits activity in satellite TPs located in their pain reference zone.

Thus, this study compares the efficacy of dry needling in the treatment of myofascial pain in addition to reducing pain and analyzing the influence on quality of life. The present study is justified aiming at the effectiveness of dry needling therapy when applied in cases of myofascial pain. Therefore, the present study aimed to report clinical cases of myofascial pain treatment with the dry needling technique, to demonstrate the importance of the dental surgeon in the diagnosis of temporomandibular disorders.

Case Reports:-

The present work has a descriptive character as object of study the clinical cases and the bibliographic survey, through a systematized review of scientific materials as articles found in databases available in electronic medium, such as: Scielo, Bireme and Google academic.

Cases:-

1. Ms. O. H. S. R., melanoderma, attended the UNIFEB clinic, with symptoms of muscular pain and difficulties to open the mouth and chew, performing dry needling with instant improvement of the condition. Patient reported constant headache and limited movement (Figure 1).
2. Ms. D.S.R., leucoderma, attended the clinic of UNIFEB, with symptoms of muscle pain and difficulties to open the mouth and chew, performing dry needling with instant improvement of pain. Patient still reported going through emotional problems and making use of medication for treatment (Figure 2).

The materials used were: fine monofilament sterile needles; Pen for marking trigger points; 70% alcohol and sterile gauze for site asepsis;

Ethics:-

The current resolutions of the National Health Council - Ministry of Health (CNS / MS), which regulate human research standards (Resolution CNS 196/96), were followed. Before the beginning of the study, the protocol will be submitted to the Research Ethics Committee of the University Center of the Teaching Foundation of Barretos.

Discussion:-

According to Schmidt et al. [1], temporomandibular dysfunction (TMD) is a type of orofacial pathology arising from disorders of the masticatory system as well as in its underlying structures, which may present with muscular or articular characteristics. This, in turn, is located near the ear and is responsible for the connection between the jaw and the skull. It presents functions such as chewing movements, facial expression and speech.

The etiology of TMD is complex and multifactorial in relation to its predispositions, involving emotions, traumas, postures and muscular hyperactivity. It presents an evolutionary course varying in days, months and years, aggravating itself with parafunctional habits, characterized by not presenting normal functions of the masticatory system [3].

The main symptoms of TMD are related to otalgia, headache, dizziness, pain in the face and / or TMJ region, tinnitus, TMJ noises, cracking and crackling, and movement-related disorders. Patients may still present inappetence, fatigue, sleep disturbance, labor difficulties and loss of quality of life [3,4].

Pain is an unfeasible experience and its importance is recognized by exerting biological function, manifesting protection to the organism. Patients who present chronic pain, because they report pain in long periods and complain of therapeutic failures, result in the presence of signs of depression and behavioral changes. They cease to be understood as warning signs and are observed as a biopsychosocial process that affects daily life in general [3,5].

Dry needling is a relatively new method in the arsenal of painkillers. Its widespread use began after Lewit's publication more than 30 years ago [6-9]. Since the beginning of the 21st century, serious scientific attention has been given to this method [10,11]. The dry needling technique emerged empirically, in different schools and conceptual models, were developed including the radiculopathy model which is a form of deep dry needling and the myofascial trigger point model is a form of superficial needling [12-14]. Patients with cardiovascular problems and bleeding or those who are taking drugs for the blood thinner will not be allowed to use the dry needling technique [15-17].

Conclusion:-

Needle treatment has been shown to be very effective for immediate pain relief, requiring follow-up for TMD treatment.

Conflict of Interest:-

The authors declare no conflict of interest.



Figure 1:- Image of Case 1 showing the act of dry needling.



Figure 2:- Image of Case 2 showing the act of dry needling.

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RESEARCH ARTICLE

FLUORIDE RELEASING AND COLOURCHANGING PIT AND FISSURE SEALANTS – a review.

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Abstract

Aim- to review about the colour changing and fluoride releasing pit and fissure sealants.

Background- dental caries is the most important and the most common intra oral disease that is seen both in children and adults. This being the era of preventive dentistry, the main aim of the dentist is to prevent the teeth from any carious involvement. Earlier, two main methods prophylactic restoration and prophylactic odontotomy were used in the management of pit and fissure caries. It is even possible to establish a caries free dentition in childhood with the increasing advancements in pit and fissure sealants. This article mainly aims to review the recent advancements in pit and fissure sealants like color changing sealants, fluoride releasing sealants and resin based sealants.

Reason- to understand more about the colour changing and fluoride releasing pit and fissure sealants.

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Introduction:-

Dental sealants are preventive dental treatment where pit and fissure of primary or permanent molar and premolar are filled with material thereby preventing caries [1]. Aim of pit and fissure sealant is to prevent developing caries which is achieved by blocking the surface and prevents bacteria from getting attached to it [2]. Deep pits and fissures favor food retention and are difficult to clean by routine brushing. It provides a favorable environment for the oral microorganisms to thrive and convert the carbohydrates to acids, leading to demineralization of the enamel. [3]

Previously, during the early 1920s two main procedures were done for the management of pit and fissure caries, they are prophylactic restoration and prophylactic odontotomy. Which was later followed by Kline and Knutson who discovered treatment of pit and fissure caries with ammoniacal silver nitrate and Bowen RL et al who discovered Bis-GMA (Bisphenol A - Glycidyl Methacrylate) a resin sealant material. Thus, resin sealant methods were born [4].

Sealants are the most effective clinical technique to prevent pit and fissure caries. The cost effectiveness of sealants, naturally, is based upon sealant retention. While the rates of sealant retention on occlusal surfaces are relatively high at five years, [5-8] sealant retention for buccal and lingual pits and fissures of molars is considerably lower [9].

A recent, significant development with resin based sealants is the development of moisture-tolerant chemistry. In the past, isolation and exclusion of moisture with resin-based sealants was required. A moisture tolerant, resin-based sealant does not require an additional bonding agent [10-12].

Discussion:-

Indications and Contraindications of Pit and Fissure sealants

The main indications of application of pit and fissure sealants are deep retentive pit and fissures and stained pit and fissures. Patients which have a very poor plaque control and patients undergoing orthodontic treatment. Other indications of application are no evidence of proximal caries[13].

The contraindications are mainly partially erupted teeth without any adequate moisture control and in teeth where isolation is not possible[14].

Generations of pit and fissure sealants

FIRST GENERATION	UV cured pit and fissure sealants
SECOND GENERATION	Chemically cured pit and fissure sealants
THIRD GENERATION	Visible light cured pit and fissure sealants
FOURTH GENERATION	Fluoride releasing sealants

Fluoride releasing pit and fissure sealants:-

The primary approach in prevention of dental caries comprises of use of pit and fissure sealants. Fluoride from the sealants reduces demineralization and enhances remineralization, thus helping to the prevent initiation and progression of caries[15]. Fluoride releasing sealants are commonly used in dental treatment as a primary preventive measure against caries[16]. The focus of most sealant studies is the occlusal surface of permanent teeth. Permanent molars have been selected as teeth most at risk for occlusal caries and thus, the teeth that most benefit from sealants. This perspective comes from population data. It reflects the realities of “normal” tooth anatomy and thus average susceptibility to caries. It does not account for individual differences among patients and among teeth[17].

Fluoride is incorporated into resins by two ways-

the first utilizes a soluble fluoride salt which, after application, dissolves releasing fluoride ions, which may compromise the integrity of the resin[18]. This method has been questioned, because fluoride release resulting from the dissolution of a soluble salt might weaken the sealant and thereby might reduce its usefulness as a preventive agent[19]. The other system uses an organic fluoride that is subsequently released by an exchange with other ions in the system[20]. In this method (anion exchange systems), fluoride constitutes only a small amount of the total structure, and is replaced rather than lost. Thus, there should not be any significant decrease in the strength of the sealant[19]. The addition of fluoride to pit and fissure sealants was considered more than 25 years ago[20,21] but did not prove that they reduce caries as they were poorly retained on the tooth surface.

The properties of a fluoride containing sealant that the conventional sealants did not have are better retention rates when compared to the conventional, constant fluoride release for a prolonged period of time and the ability to function as a reservoir of fluoride ion for enamel and to promote fluorapatite formation in enamel[22].

Colour Changing Sealants:-

In March of 1977, the first colored sealant (3M's Concise White Sealant) was introduced to the market. The advantages to a color as long as it is esthetically acceptable. It is easier to see the sealant during application, and it is much retention. Faster and easier to assess with a white sealant than with a clear sealant at later time intervals.[23] While a Helioclear material, which changes color from clear to green when exposed to a visible light has some clinical utility, particularly on subsequent follow-up examinations when clear sealant is very hard to see, it is hard to understand any benefit to dentist or patient of the Clinpro material that changes color from pink to white on polymerization. The combined identification error rate for opaque resin was only 1%, while for clear resin it was 23%. Significant differences were also found in the accuracy with which the 3 dentists identified each type of resin. The most common error was to identify the presence of clear resin on an untreated tooth.[24] This study raises questions about the accuracy of studies done with clear resin.

Conclusion:-

Of all the surfaces of the tooth pit and fissures are very much susceptible to caries and hence pit and fissure sealants help mainly to prevent caries and also are cost effective. Fluorides in the pit and fissure sealants reduce enamel solubility and stimulate remineralization thereby reversing the course of caries in the early stages itself.

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RESEARCH ARTICLE

STUDY ON POPULATION BUILDS UP OF RICE LEAF FOLDER *Cnaphalocrocis medinalis* (Guenee) IN RELATION TO WEATHER PARAMETERS.

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Abstract

Study on population build up of rice leaf folder *Cnaphalocrocis medinalis* (Guenee) was carried out at NARP Farm, Navsari Agricultural University, Navsari during Kharif 2012 and 2013. The observation on per cent damaged leaves was recorded immediately after transplanting up to the harvest of the crop. Results shows that maximum 21.9 per cent damaged leaves were observed during fourth week of July (30th SMW) and that of minimum (0.2 per cent) were in fourth week of October (43rd SMW). In the present investigation, per cent damage leaves showed significant and positive correlation with morning relative humidity, evening relative humidity, average relative humidity and rainfall but significant negative correlation with maximum temperature, sunshine hours and evaporation rate.

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Introduction:-

Rice (*Oryza sativa*, Linnaeus.) is a staple food of Southeast Asian population. (FAO, 2004; Kumar, et al, 2009). Global rice production must increase by 36% by 2025 to feed estimated 4 billion rice consumers (Anderson et al., 1997). But the rice production is limited by both biotic and abiotic stresses of which insect pests alone caused about 25 % losses amounting to Rs.240,138 million (Dhaliwal et al., 2007). Bautista et al., (1984) have clearly shown that loss in yield, due to the rice leaf folder, is positively related to the percentage of damaged leaves. Weather factors are the major regulating causes for the insect pest populations under field circumstances. Certain limits support and other disfavour their multiplication and movement; therefore, it results in serious outbreaks of different insect pests (Hyslops, 1941). Insects are mostly dependent on the environmental temperature for the maintenance of their activity.

Materials and Methods:-

In order to study the influence of weather parameters on population build up of rice leaf folder *Cnaphalocrocis medinalis* Guenee experiment was carried out at N.A.R.P. Farm, N.A.U., Navsari during Kharif 2012 and 2013 in 1000 m² area in GR 11 rice variety was transplanted with 20 x 15 cm² spacing. All the post sowing recommended agronomical practices were followed. The experimental area was kept free from insecticide spray throughout the crop season in order to record incidence of rice leaf folder *C. medinalis*. The observations on per cent damaged leaves caused by rice leaf folder *C. medinalis* were recorded at weekly interval in each standard meteorological week immediately after transplanting up to the harvest of the crop. The leaf folder incidence was assessed by calculating per cent damaged leaves per 10 hills.

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Results and Discussion:-

The data on incidence of rice leaf folder in terms of per cent damaged leaves (DL) per ten hills during kharif-2012, 2013 and pooled were found significant. During Kharif 2012, data recorded on per cent damaged leaves indicated that infestation was ranged from 1.04 to 20.72 per cent. the average infestation started appearing just after establishment of transplanted paddy seedlings i.e. from third week of July [30th SMW] and continued up to the second week of October (41th SMW). The maximum infestation 20.72 per cent DL was found in first week of September (36th SMW).

During Kharif 2013, infestation commences from fourth week of July (31st SMW) and continued up to the fourth week of October (43rd SMW). The maximum infestation (35.96 per cent DL) was found in second week of July. In 4th week of September 39th STW sudden increase in per cent leaf folder damage thereafter started declining up to 4th week of November (42nd SMW) which noticed minimum 1.44 per cent (DL) and no incidence in next SMW.

Results on pooled data of (Kharif 2012-2013), indicated that in both the years (Table 1). Maximum infestation 21.9 per cent (DL) was observed in fourth week of July (30th SMW). Minimum infestation of leaf folder 0.2.0 per cent (DL) was noticed during fourth week of October (43rd SMW).

The results on correlation of per cent leaf damaged of rice leaf folder, *C. medinalis* during Kharif-2012 -2013 and pooled were worked out with weather parameters indicated that during first year morning relative humidity (X4) ($r = 0.5749$), evening relative humidity (X5) ($r = 0.5409$) and average relative humidity (X6) ($r = 0.5651$) had highly significant positive correlation and rain fall (X9) ($r = 0.04847$) had significant positive relationship with per cent damage leaves (Y1). The maximum temperature (X1) ($r = -0.7230$), sunshine hours (X8) ($r = -0.5495$) and evaporation rate (X10) ($r = -0.7986$) had highly significant negative correlation with per cent damage leaves. The minimum temperature (X2) ($r = 0.3426$) showed non-significant positive correlation but wind speed (X7) ($r = -0.0571$) showed non-significant negatively correlation with per cent damage leaves (Table 2).

Likewise, in the subsequent year (Kharif 2013), per cent damage leaves indicated highly significant positive correlation with wind speed (X7) ($r = 0.5758$) and rain fall (X9) ($r = 0.5455$). However, the maximum temperature (X1) ($r = -0.6317$) and evaporation rate (X10) ($r = -0.6416$) exhibited highly significant negative correlation with damage of leaves. The evening relative humidity (X5) ($r = 0.4266$) and average relative humidity (X6) ($r = 0.4234$) showed significant positive correlation but Average temperature (X3) ($r = -0.4265$) and sunshine (X8) ($r = -0.4525$) showed non-significant negative correlation while minimum temperature (X3) ($r = 0.2928$) and morning relative humidity (X4) ($r = 0.3832$) had non-significant positive correlation with per cent damage leaves (Table 6.1).

In pooled analysis, overall per cent damaged leaves (Y1) showed significant positive correlation with morning relative humidity (X4) ($r = 0.5170$), evening relative humidity (X5) ($r = 0.5083$), average relative humidity (X6) ($r = 0.5114$) and rainfall (X9) ($r = 0.4554$). The maximum temperature (X1) ($r = -0.7130$), sunshine hours (X8) ($r = -0.5626$) and evaporation rate (X10) ($r = -0.6416$) exhibited highly significant negative correlation, whereas morning relative humidity (X4) ($r = 0.5170$), evening relative humidity (X5) (5083), average relative humidity (X6) (0.5154) and rain fall (X9) ($r = 0.454$) had significant positive correlation. But, average temperature (X3) ($r = -0.2294$) had non-significant negative correlation with per cent damage leaves. Whereas minimum temperature (X2) ($r = 0.3934$) and wind speed had non-significant positive correlation (Table 2).

It is revealed from the forgoing discussion that, per cent damage leaves showed significant and positive correlation with morning relative humidity, evening relative humidity, average relative humidity and rainfall but significant negative correlation was observed with maximum temperature, sunshine hours and evaporation rate. Earlier, Balasubramanian et al. (2005) depicted that a significant positive correlation was found between pest incidence and morning relative humidity, evening relative humidity, total rainfall and total number of rainy days. A significant negative correlation was observed between pest incidence and hours of sunshine and maximum temperature.

Table: 1 Population build up of rice leaf folder in relation to weather parameters during Kharif -2012 and 2013.

Sr. No	Standard Meteorological Week (SMW)	Period	Per cent leaves damaged by <i>C. medinalis</i>		
			Kharif 2012	Kharif 2013	Pooled
1	33	13.08.14 to 19.08.14	0	0	0.0
2	34	20.08.14 to 26.08.14	0	0	0.0
3	35	27.08.14 to 02.09.14	0	0	0.0
4	36	03.09.14 to 09.09.14	0	0	0.0
5	37	10.09.14 to 16.09.14	0	0	0.0
6	38	17.09.14 to 23.09.14	7.86	35.97	21.9
7	39	24.09.14 to 30.09.14	12.81	28.73	20.8
8	40	01.10.14 to 07.10.14	14.95	12.71	13.8
9	41	08.10.14 to 14.10.14	17.25	14.71	16.0
10	42	15.10.14 to 21.10.14	16.45	12.32	14.4
11	43	22.10.14 to 28.10.14	14.87	0.00	7.4
12	44	29.10.14 to 04.11.14	20.39	5.86	13.1
13	45	05.11.14 to 11.11.14	20.72	0.00	10.4
14	46	12.11.14 to 18.11.14	16.75	7.54	12.1
15	47	19.11.14 to 25.11.14	12.17	25.47	18.8
16	48	26.11.14 to 02.12.14	5.58	5.53	5.6
17	49	03.12.14 to 09.12.14	4.00	3.86	3.9
18	50	10.12.14 to 16.12.14	1.04	0.85	0.9
19	51	17.12.14 to 23.12.14	2.63	1.44	2.0
20	52	24.12.14 to 31.12.14	0	0	0.0
21	1	01.01.15 to 07.01.15	0	0	0.0
22	2	08.01.15 to 14.01.15	0	0	0.0
23	3	15.01.15 to 21.01.15	0	0	0.0
24	4	22.01.15 to 28.01.15	0	0	0.0

Table: 2 Correlation analysis of rice leaf folder *C. medinalis*, infestation with meteorological parameters

Correlation coefficient										
Kharif 2012										
	1	2	3	4	5	6	7	8	9	10
	Max. T	Min. T	Ave. T	Mor. RH	Eve. RH	Ave. RH	WS	SS	RF	Eva.
% DL 12 (Y1)	- 0.7230*	0.342 6	- 0.2370	0.5749*	0.5409*	0.5651*	-0.0571	- 0.5495*	0.4847*	- 0.7986**
NL 12 (Y2)	- 0.6516*	0.241 2	- 0.3040	0.4820*	0.4635*	0.4808*	-0.0995	- 0.4350*	0.4936*	- 0.8033**
Kharif 2013										
% DL 13 (Y1)	- 0.6317*	0.292 8	- 0.4265*	0.3832	0.4266*	0.4234*	0.5758*	- 0.4525*	0.5455*	- 0.64162*
NL 13 (Y2)	- 0.5380*	0.269 3	- 0.3349	0.3995	0.3895	0.3989	0.5505*	- 0.4360*	0.5820*	- 0.61800*
Pooled 2012-2013										
	Max. T	Min. T	Ave. T	Mor. RH	Eve. RH	Ave. RH	WS	SS	RF	Eva.
% DL poole	- 0.7130*	0.393 4	- 0.2294	0.5170*	0.5083*	0.5154*	0.2364	- 0.5626*	0.4554*	- 0.6481**

d (Y1)	*							*		
NL poole d (Y2)	- 0.6282* *	0.345 8	- 0.2034	0.5148*	0.4592*	0.4791*	0.0243	- 0.4630*	0.4838*	- 0.6293**
*Significant at 5 % level 0.4227					** Significant at 1 % level 0.5368					

DL =Damaged leaves, NL= Number of Larva, T=Temperature, Mor. =Morning, Eve. = Evening, RH= Relative Humidity, WS =Wind speed, SS= Sunshine, RF=Rain Fall, Eva=Evaporation rate

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RESEARCH ARTICLE

MAIN PREDICTORS OF BONE CREST PRESERVATION WITH THE USE OF THE SWITCHING PLATFORM.

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Abstract

This study examined the effects of the use of Platform Switching in rehabilitation implantossuportadas. It is a technique that consists in fitting a prosthetic component of smaller diameter the platform of implant with larger diameter, so that there is no combination in the size of both, as in conventional manner. The results showed that the benefits and the indication of the use of this technique is due to reason that the same initial reduces the reabsorption of bone Christian peri-deploy and maintain this level bone in the course of time, as well as reduces the forces on the bone tissue adjacent to the cervical region of the implant. Although there are possible disadvantages in the use of Platform Switching, which may cause an increase of tensions in the region of the screw and prosthetic component, its adaptation in rehabilitation implantossuportadas has been very attractive.

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Introduction:-

Dentistry was favored by the discovery of osseointegration, which became of its importance in the treatment of partially and totally edentulous patients. Initially, osseointegration was only used to treat fully edentulous patients in the maxilla and mandible, through a single type of implant, prosthetic abutment and rehabilitation protocol [1,2]. Surgeons, as soon as they began to use osseointegration, sought only to obtain and maintain the anchorage of the implants, without caring much about aesthetics. However, this has been gradually changed and is now established as a reality, thanks to the longevity of the treatments and to the functional and aesthetic results obtained [2].

Over time, the basic principle of osseointegration has been proven, so as to allow its application in the most varied clinical situations of tooth failure, both unitary and partial. It is necessary that new concepts, principles and technologies be discovered and improved [3]. However, as the applications of osseointegration have evolved, complications have also increased, which have become more numerous and are no longer the result of surgical and mechanical problems of the components of a single care protocol, requiring more research, studies and Care [3].

Among the most varied concerns that prosthodontists experience, peri-implant bone loss is found because a treatment with implants will only have satisfactory results if the soft and hard tissues are maintained in the long term around the implant [2,3] . This led to a reassessment and questioning of the traditional protocols, resulting in new approaches to implantology, among which, the concept of Switching Platform, considered as the use of a prosthetic

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component smaller than the implant platform , In order to reduce reabsorption of vertical bone crest levels around the implants over time [2,3].

Once the prosthesis is definitively installed on the implants, an expected peri-implant bone loss occurs, which is called saucerization [3,4]. This is due to the reestablishment of biological distances. Thus, the application of prosthetic components smaller than the implant platform creates a difference in horizontal adaptation circumference size, limiting bone crest resorption and preserving bone levels around implants [3].

Therefore, Platform Switching consists of a technique adopted to preserve the bone, through the prosthetic connection of a smaller diameter than that of the implant, in order to transfer the existing slit to an innermost position horizontally. Its concept has presented satisfactory results, by reducing peri-implant bone loss through the internalization of the junction between the implant and the prosthetic connection [1-3].

The objective of the present study was to evaluate the effects of the use of the Switching Platform in implant - supported rehabilitations. It is a technique that consists of connecting a smaller diameter prosthetic component to the larger diameter implant platform, so that there is no combination in size of both, as in the conventional form.

Methods:-

For the identification of studies in this review study, carried out a detailed search strategy for Medline, Pubmed, in the years 1986 - 2016. They were used as descriptors: Switching Platform; Regular Platform; Implantology; Implant Supports Rehabilitations. Analyzed studies systematic review, meta-analysis, randomized controlled cases, nonrandomized clinical cases and opinion articles that addressed the term atraumatic surgery. The data are analyzed, correlated to the discussion of the results highlighted in the literature, as shown in figure 1.

Continuous Predictors:-

The continuous predictors were Switching Platform Implant.

Response Predictor:-

The response predictor were increased tensions in the region of the prosthetic component and retention screw, which could lead to loosening and maladjustment and even fracture.

Literature revision:-

Implant concept and Osseointegration:-

The implant is performed when a titanium screw, which can be of various shapes, is inserted into the bone, by means of various surgical techniques. This screw will support a prosthetic crown, which can be cemented or screwed, which will restore the functions of the natural tooth crown [40].

When analyzing the blood microcirculation in rabbit tibia in the 1960s, Branemark verified that there was a perfect integration of the bone with the titanium used in the research, which led to the appearance of the term osseointegration, that is, structural and functional junction that occurs Between the living, ordered bone and the surface of the implant submitted to the functional load [6].

In this context, it is not only the osseointegration that Branemark discovered that it is enough for an implant to be successful, also needing other factors, among them, a correct occlusion, that is, a harmonic relationship between the upper and lower arches without nothing interfering. If there is no such occlusal harmony, occlusal trauma or premature contact may occur, which may lead to changes in the functioning of the masticatory apparatus, significantly altering the position of the teeth, gingival recessions, bone resorption, as well as interfering with the implants installed, generating undesired results. Patient and the surgeon [43].

Bone Loss:-

In implants where the heights of the structures are greater than 180 μm , even if there was no inflammation in the peri-implant tissue, an increase in bone resorption may occur due to occlusal overload [35]. This is because bone resorption is generated where there is an inflammatory reaction or a prolonged excessive stimulus [6,16].

However, in the first implants performed, there was correspondence between the prosthetic components, observing the occurrence of an apical remodeling of the bone crest of about 1.5 to 2.0 mm of the implant implant pillar or at the level of the first thread, the Which altered the successful results of implant maintenance [5,27].

This bone loss was defined as saucerization, which consists of a process that affects all types of osseointegrated implants, regardless of even the general conditions of the patient. It is a mechanism of adaptation of the organism in the establishment of the physiological dimensions of the periodontium, that is, of the periodontal biological space. This process of saucerization occurs mainly around external hexagon type implants [1,2].

The Peri-implant junctional epithelium is shown to have more layers of cells, assuming a conformation very similar to the junctional epithelium of the natural teeth, which approximates the Peri-implant junctional epithelium of the osseointegrated surface, increasing the concentration of Gingival connective tissue at the site, represented by the arrows and, consequently, accelerating bone resorption, beginning the saucerization [2].

In conventional implants, the apical remodeling of the bone crest occurred when exposed to the buccal medium after a healing was performed or the second stage surgery performed. This is due to the fact that an inflammatory infiltrate arises in the implant interface pillar implant or the attempt of the soft tissue to create a barrier, in order to seal the environment of the implant platform [11,16]. However, bone resorption can also be induced by stress concentrations in the coronal region of the implant [16,19].

Many factors may lead to loss of bone crest around conventional implants, among which the most common are: surgical trauma, recovery of biological space, occlusal overload, peri-implantite, gaps [23], as well as manipulations Tissues [11]. Some consequences can be caused by the resorption of the bone crest around dental implants, especially in relation to aesthetics, because the soft tissues change in the proximal faces, which can be seen in the papillae and, on the vestibular face, through the Presence of recessions [16,23].

Keeping the level of the bone crest in the ridge of implants was only possible when, occasionally, a radiographic study was able to make this observation. This is due to the fact that the Implantodontics company, called Innovations 3i, has introduced wide-diameter platform implants (5.0 mm and 6.0 mm), which should be used in cases of implants failure Standard and poor bone region [2,3]. Above all, because there were no prosthetic components corresponding to these implants, they were restored with 4.1 mm abutments, so that a step of 0.45 mm occurred and, radiographically, a variation of 0.45 mm at 0.95 mm [32].

After 13 years of follow-up, by means of radiographs, these restored restorations with smaller diameter pillars than the platform, it was possible to verify that the levels of bone crest were maintained with minimal marginal bone loss, thus raising the concept Of Platform Switching [16].

Platform Switching:-

The concept of Switching Platform used in rehabilitation with osseointegrated implants has been increasingly used, both in the literature and in implant clinics, because it presents satisfactory results, both aesthetic and functional. This is a concept created by Lazzara and Porter (2006) [32], which consists of the use of a smaller diameter prosthetic component connected to the platform of a larger diameter implant, in order to create a "step" of 90 Between the implant and the prosthetic component.

Although a number of factors influence bone resorption, many studies show that implants with a Switching Platform system are more advantageous in relation to implants in which there is a combination of the standard prosthetic component with the type of implant, to maintain the crest level Because it improves the results of peri-implant soft and hard tissues, maintenance of the position of the interdental papilla and marginal gingiva, in addition, when using a prosthetic component of smaller size, the concentration of force area on the implant occurs of the marginal bone crest area. [8,37].

The Switching Platform has been considered a very significant form of treatment to maintain soft and hard peri-implant tissues, not only in two-stage implants, but also in immediate-loading immediate implant protocols. This system removes the concentration of stresses from the peri-implant bone margin and reduces its effect on marginal bone resorption [30]. There is a chain of theorists who affirm that the occurrence of change in the prosthetic-implant

component connection contributes to the maintenance of the biological space, thus reducing bone loss of the marginal ridge [32,37].

Based on Maeda et al. (2007) [34] verified the biomechanical advantages of the Switching Platform, analyzing three-dimensional finite elements, through the use of an external hexagon-type implant model (4.0 mm x 15 mm), on which two types of implants were performed: One with a prosthetic component of 4.0 mm in diameter and the other with a prosthetic component of 3.25 mm, using the Switching Platform configuration.

According to Canullo et al. (2010) [18] developed a study to evaluate changes in bone level in implants with Switching Platform system, making use of various combinations between implant / prosthetic component. After 90 days, they verified that the connection of the prosthetic components of 3.8 mm to the implants occurred, and the restorations were definitively installed. The bone was measured by radiographs performed at implant placement (control), and after 9, 15, 21 and 33 months. After 21 months, all implants were clinically osseointegrated in all 31 patients studied. However, according to the radiographs, the authors verified the occurrence of bone loss of 0.99 mm (± 0.42 mm) for Second experimental group; 0.82 mm (± 0.36 mm) for the third experimental group and 0.56 mm (± 0.31 mm) for the fourth experimental group. However, these values were lower in comparison with the control group (1.49 mm ± 0.54 mm). After 33 months, no difference was found compared to the 21-month results, with the exception of the third experimental group (0.87 mm) and the fourth experimental group (0.64 mm), leading to the conclusion that the existing relationship is Inverse relationship between size / extent of implant-prosthetic component decompensation and amount of bone loss, with the maintenance of better bone margins levels using the Switching Platform configuration.

The study by Calvo-Guirado et al. (2009) [17] evaluated the survival rate of implants made in the maxilla, anterior and premolar areas and restored with single crowns after one year and also loss of bone crest. In this study, implants were fixed in the alveoli, after tooth extraction, with an immediate temporary restoration, and the definitive implant was placed after 15 days, through the Switching Platform concept.

Still in the same year of 2009, Schrotenboer et al. (2009) [42], studying the interaction of the platform in the transcortical section of the adjacent bone of an osseointegrated dental implant, developed a two-dimensional model to analyze finite elements in order to observe the bone-implant interactions under mastication forces. They simulated, together with a 5.0 mm diameter implant, two prosthetic components, one of 4.5 mm in diameter, to represent the switching platform and the other 5.0 mm, to represent a standard diameter platform, Applying static forces of 100 N, vertically (90°) and obliquely (15°) on the pillars. The results revealed that with the standard platform model, the maximum von Mises stress pressure on the bone crest was 28 MPa under oblique load and 6,977 MPa under vertical load.

Hsu et al. (2009) [29] when checking the bone tension and micro-movement at the bone-implant interface for switching platform, simulated finite elements and analysis by extensometry with different diameters of a single implant with immediate loading. They created four models, 5.0 mm diameter implants with prosthetic components 5.0 mm and 4.0 mm in diameter, cemented (late loading) and screwed (immediate loading), and a model with implant of 3.75 Mm diameter, in which vertical and lateral loads of 130 N were applied, which resulted in the following: bone forces were reduced by up to 10% with the use of the switching platform.

Another evaluation of bone crest alterations was made by Vigolo et al. (2009) [47], who did so for a long period of five years, after implanting external hexagon type 5.0 mm diameter restoration with components of the same diameter or switching platform. One group had mean bone resorption values of 0.9 mm (± 0.3 mm), while the other group had 0.6 mm (± 0.2 mm). In consecutive years no marginal bone resorption was observed, leading to the conclusion that restored implants with prosthetic components of standard diameters had greater bone loss than those restored with a switching platform.

Cocchetto et al. (2010) [22] sought to verify that the change of the junction to a more interior region of the implant platform may result in less resorption of the bone crest, with an increase in the discrepancy between the implant platform and the prosthetic component diameter. This study was performed with 10 patients who needed restoration with mandibular or maxillary implants. Fifteen 5.0 mm diameter implants with a switching platform with a diameter of 5.8 mm in the collar were used, and 5.0 mm prosthetic surfaces were attached, which were connected to healing wounds of 4.1 Mm for eight weeks, following single-stage protocol. The results showed a peri-implant bone loss averaging 0.30 mm.

Bilhan et al. (2010) [13] performed a study with 51 patients to compare bone preservation around regular implants and switching platform that supported overdentures. A total of 126 implants were performed, with follow-up for 6, 12, 24 and 36 months after the installation of the prostheses. Through a series of radiographs they were able to show that, with the use of the switching platform, there were lower bone and distal bone losses in 36 months, although there were differences in mesial and distal surface loss in both groups. They verified, therefore, that the smaller bone losses in implants that support overdentures occur with the use of platform switching.

Chang et al. (2010) [21] performed an analysis and comparison of tensions at the bone-implant interface with protein elements of standard diameters and diameters according to the concept of switching platform in the posterior maxilla region, using finite elements. They created two finite element models, one of a first maxillary molar section, with an osseointegrated implant of 4.1 mm x 10.0 mm, with the simulation of a prosthetic component of 4.1 mm in diameter and another model with a little narrower component, that is, with 3.4 mm, simulating the switching platform.

Fickl et al. (2010) [24] studied the influence of the use of the switching platform at the height of the bone crest around dental implants in 89 dental implants and 36 patients. For this, these authors observed implants made in healthy bone, which did not require an increase of ridge. In this way, they created the following: Group 1, with the placement of wide diameter implants in the region below the bony crest and the connection of regular diameter scars; Group 2, placement of regular diameter implants on the ridge line and connection of regular diameter scars. The results showed that, with the use of the switching platform system (n = 75), bone loss was significantly lower at the moment of definitive insertion of the prosthesis (0.30 ± 0.07 mm, versus 0.68 ± 0.17 mm; $p < 0.05$), and in one year (0.39 ± 0.07 mm versus 1.00 ± 0.22 mm, $p < 0.01$), compared with conventional implants (n = 14), leading them to conclude that implants With switching platform limit bone crest remodeling.

Discussion:-

For Lazzara and Porter (2006) [32] the term Switching Platform has been extensively studied in Implantology today, which means that the base of the restorative abutment is smaller than the head of the implant where it is embedded. An implant performed according to the Switching Platform concept presents, around the cervical region, a greater quantity of soft tissues and maintenance of more bone, because of the reduction of the remodeling generated by the establishment of the biological space.

The ideal for the success of an implant would be minimal or no bone loss, however, there is always some bone loss, especially after one year of masticatory function [28]. Several biological and mechanical factors influence this resorption around the implant, such as bacterial microleakage, localization of inflammatory connective tissue area, concentration of stresses in the cervical region of the implant, location of the implant / abutment junction and micro-movements [38].

The maintenance of the bone / implant interface, whether biomechanically, in the induction of stresses, or biological, depends on the type of connection that can allow bacterial infiltration at the interface, which directly influences the success of the rehabilitation. [14,20,46]. This fact is evidenced, especially, in the alteration of the height of the bone crest, as a consequence of the concentration of tension in the cervical region of the implant [21]. Thus, the implant system must be designed to better dissipate the load in the peri-implant bone, so that it can support restoration in function and maintaining osseointegration [48]. In view of this, some studies have shown that resorption of bone crest can be reduced by the use of the Switching Platform technique [32,38].

The rehabilitations that make use of the concept of Switching Platform have been much studied, aiming to verify their real advantages and disadvantages in the short and long term. Among the advantages, the literature points to the reduction of reabsorption of the peri-implant bone crest, as verified by the following authors: Calvo-Guirado et al. (2009) [17], which evidenced a reduction in bone loss in osseointegrated implants with the use of the Platform Switching concept; Canullo et al. (2010) [18] who, in an evaluation of sixty implants, observed the occurrence of an inverse relationship between the extent of implant-prosthetic component decompensation with amount of bone loss, so that the greater the discrepancy, the lower the loss Bone at the peri-implant crest, still acting on the longitudinal maintenance of this bone level. Also Cocchetto et al. (2010) [22] and Bilhan et al. (2010) [13], have concluded that increased discrepancy between implant and prosthetic component reduces subsequent bone loss.

In relation to the distribution of forces under occlusal loads, Schrotenboer et al. (2009) [42] observed that the Switching Platform system for the Von Mises test, on cortical bone crest, had a measurable but minimal effect. Hsu et al. (2009) [29], when using finite elements in their studies, concluded that there is a 10% reduction in forces exerted on bone when there is the use of the Switching Platform concept in relation to conventional implants and that the increase of implant diameter (Extended platform) led to an expressive reduction of the stress in the surrounding bone to the implant, leading to understand that not only the Switching Platform system, but the increased implant diameter reduces stress in the bone tissue. Still in the same study, they stated that none of the settings used. However, regarding micromovements to improve the stability of the implant, no differences were identified when comparing with those presenting conventional implants.

In relation to stress concentration, Chang et al. (2010) [21], found that the Switching Platform system led to a reduction of stress concentration in areas of compact bone but transferred this concentration to areas of spongy bone. Also Tabata et al. (2011) [45] have shown that implants with Switching Platform are advantageous in terms of improving biomechanical stress distributions in peri-implant bone tissue.

However, Maeda et al. (2007) [34] concluded that although the Switching Platform system shifts to more distal from the cervical stress area concentration, which has been considered as important for maintaining the peri-implant bone crest, it generates an increase in stress on the component Prosthetic and retaining screw, which may lead to maladjustment, fracture or loosening of the screw. In addition, according to Vigolo and Givani (2009) [47], the 85 implants from his work in which wide platform components were used showed greater bone loss than the 97 implants in which the switching platform was used in the components.

Through statistical analysis of 850 implants with switching platform and the evaluation of radiographic changes of marginal bone levels and their amount of loss compared to implants without switching platform. The results showed that the implants with switching platform lost less marginal bone in relation to the rehabilitated ones without platform switching, except for the study of Kielbassa et al. [50] where decreasing the size of the component relative to the platform resulted in a loss of 0.8mm. This bone loss was greater than the conventional rehabilitated implants, where a loss of 0.63 mm was detected.

Figure 1:-

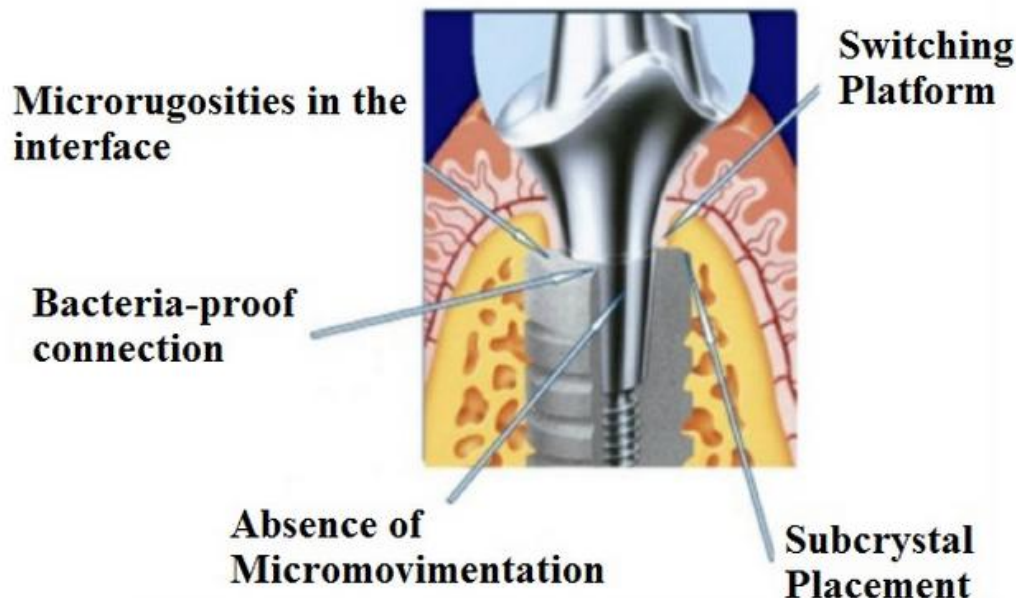
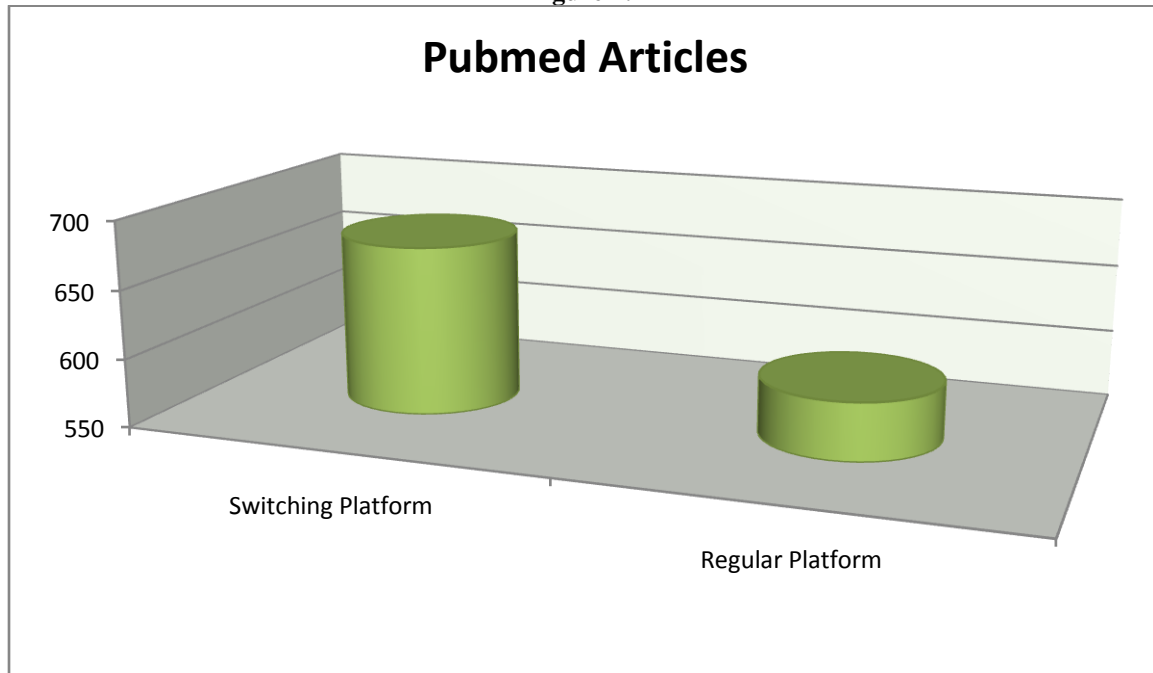


Figure 2:-



Conclusion:-

The biological advantages and the clinical performance of the Switching Platform technique in the implant-supported rehabilitations were evaluated, reducing the bone resorption at the marginal ridge adjacent to the implant with the maintenance of the implant, as well as a reduction of about 10% in the forces on the bone tissue. However, the use of this technique can increase tensions in the region of the prosthetic component and retention screw, which can lead to loosening and maladjustment or even fracture.

Competing Interests:-

The authors declare no competing interests.

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RESEARCH ARTICLE

STUDY OF SOME SELECTED AQUATIC AND SEMI-AQUATIC FAUNA OF BAANGANGA WETLANDS, HARIDWAR (UTTARAKHAND, INDIA).

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Abstract

Baanganga, the riverine wetland lies between Hastinapur Wildlife Sanctuary in Uttar Pradesh and Jhilmil Jheel Conservation Reserve, in Uttarakhand is considered a major habitat for wintering waterfowl. The habitat, known for swamp deer and hog deer and also for aquatic and semi aquatic fauna, is deteriorating as revealed by the analysis of physico-chemical properties of water. The present study recorded the occurrence of four species of freshwater turtles, marsh crocodile, three amphibians and 18 species of fishes in the Baanganga wetland. Freshwater turtle, *Morenia petersi* recorded from the area is a new record for the State. We suggest measures for conservation of these wetlands.

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Introduction:-

Wetlands harbor variety of fauna that inhabit terrestrial-semi-aquatic and aquatic environments. Baanganga wetland, Indrishpur village in Haridwar district lies between Hastinapur Wildlife Sanctuary in Uttar Pradesh and Jhilmil Jheel Conservation Reserve in Uttarakhand. It represents riverine wetland ecosystem (Fig.1). The construction of a dam across river Ganga, near Baanganga and other anthropogenic activities have led to the conversion of channel into isolated water bodies. These wetlands are still considered as major habitat for wintering waterfowl, though they are surrounded by large agricultural fields and human settlements (Adhikari 2008). The Indo-Gangetic plain and the Terai region have diverse Chelonian fauna with 20 species of freshwater turtles (Das 2002, Rao 1990). The present study describes the occurrence freshwater turtles, amphibians and fishes based on collection and sightings in and around Baanganga. Water quality of the area and suitability for aquatic fauna is also described.

Material And Methods:-

Study area: Baanganga wetland (78°2'43"E to 78°2'15" E and 29°44'5" N to 29°36'01"N) is located 60 km. south-west of Haridwar on Laksar-Purkaji road, and it has about 10 km² water spread area. Land use and land cover include; water bodies (55 %), marsh (4 %), islands (15 %), moist shores (8 %) and upland (18 %) (Adhikari, 2008, Fig 2-4).

Monthly surveys were conducted along the Baanganga River covering seven localities (Table 1) during May –Sep 2008 to 2010. The wetland was divided into five land use categories: water bodies, marshy area, islands, moist

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shores and upland (Adhikari 2008) and surveyed randomly for freshwater turtles, amphibian and fishes. Enquiries were also made on the occurrence of taxa studied.

A minimum of 10 water samples were collected from each (eight) water bodies for analysis (Table 6, 7). The ambient temperature of the atmosphere and of surface water was taken by using digital thermometer. Dissolved oxygen of water was calculated following/ NIH (1987-1988) and electrical conductivity was determined following method given by Jackson (1962). The levels of calcium, magnesium, chloride and bicarbonate were assessed titrimetrically (Jackson 1962a, b, c, d).

Mollusca were collected qualitatively together with other fauna using a plastic hand-net of 1 mm mesh size and hand picking method directly from the habitat. Larger specimens of the samples were pre-identified in the field and preserved in 15% ethyl alcohol to avoid hardening and contraction, later they were transferred into 70% ethyl alcohol. The material is deposited in the Zoological Survey of India, Northern Regional Centre, Dehradun. Identification was done with the help of Preston (1915), Subba Rao (1989) and Ramakrishna & Dey (2007).

Results and Discussion:-

Fauna: Molluscs, fishes and amphibia are known to be the major food for many freshwater turtles (Tikader and Sharma 1985). The aquatic Mollusca fauna associated with the investigated habitats comprise 8 snails (Class Gastropoda) and 6 shells (Class Pelecypoda or Bivalvia). All records are relatively large-sized species of riverine and wetland or lake habitats. These are the most frequently found and common mollusks of the Gangetic Plains including the Himalayan foothills and the Kumaon Lakes (Prashad, 1922; Neemann *et al.*, 2003-2007) (Table 2).

The family Viviparidae is represented with two species. The most common snail in different types of running and stagnant water bodies was *Bellamya bengalensis* with fifteen localities during the present field studies. The second much smaller Viviparidae *Idiopoma dissimilis* was found in three localities and this species was limited on stagnant water bodies. Among the air-breathing snails, the two species *Lymnaea acuminata* with seven localities and *Indoplanorbis exustus* with five localities were distributed in all types of water bodies with shallow zones and presence of submerged aquatic macrophytes including paddy fields as well as the shorelines of lakes and reservoirs in the Himalayan middle mountains (Table 2).

The freshwater mussels were found in two rivers with six widely distributed species of the Indian subcontinent, e.g. *Parreysia favidens*, *Radiatula* and the clam shell *Corbicula striatella*. The Unionidae were identified with results of Neemann *et. al* (2003) and the updated species catalogue of Haas (1969), Graf and Cummings (2007). The presence of a number of large bivalves species always indicates stable ecological conditions with fairly good sediment quality. The larvae or glochidia need different host fishes to complete metamorphosis into a young freshwater mussel.

The pollution tolerant Physidae *Haitia mexicana* is a true invasive species or neozoan that reached the northern Indian subcontinent by introduction to the capitals as the main centers of aquarium trade with plants and substrate. *Haitia mexicana* started rapid spreading from Delhi after 1994 (Surya Rao *et al.* 1997), reaching Allahabad in the year 1998 and Patna in the year 2000 (Sinha *et al.* 2003). It simultaneously colonized the Bagmati Kosi River system starting from Kathmandu after 1994. Nowadays *Haitia mexicana* is a widespread species of various polluted and degraded water bodies. The species remains absent from most natural habitats which have developed a rich diversified native fauna. Thus *H. mexicana* is always an indicator for direct anthropogenic impact. However in the present study there was no record of this species in any of the water body studied indicating that the area is still suitable for aquatic and semi aquatic fauna but small measures can be taken to improve the quality of the habitat.

Eighteen species of fishes belonging to 14 genera, 9 families and 3 orders were observed during this study (Table3). The heterogeneity of the wetlands could be one of the reasons for the observed diversity (Kumar, 2007).

Three species of anurans, *Fejervarya sahydrensis* (Annandale); Syhadra frog, *Euphlyctis cyanophlyctis* (Schneider) and Indian bull frog, *Hoplobatrachu stigerinus* (Daudin) were collected during this study (Table 4). The cricket frog has been reported to be available in small hill streams and water logged areas near paddy fields. The population of the species is denser in eastern parts of the district Dehradun (Ray 1998). *Zakerana* were split off from *Fejervarya* in 2011. This was the culmination of recognizing that *Fejervarya* (as known before) was paraphyletic and consisted of two different groups; this recognition had already developing some years before. The sister taxon of *Zakerana* is

Sphaerotheca (Frost 2014, Howalder 2011). *Fejervarya*, as currently defined, has a more eastern distribution than the southern Asiatic *Zakerana*, from eastern India eastwards. (Frost 2014). They are sometimes known as the South Asian cricket frogs.

Euphlyctis cyanophlyctis was the most common anuran recorded from the area. It can tolerate considerable amount of contamination and organic pollution in temporary water pools (Ray1998), (Table 4).

Four species of freshwater turtles; *Lissemys punctata andersonii* (Webb), *Pangshura tecta* (Gray), *Pangshura tentoria circumdata* (Gray) and *Morenia petersi* (Anderson) were observed (Table 4, Fig. 8, 9). *Lissemys punctata andersonii* and *Pangshura tecta* are listed under Schedule- I of the Wildlife (Protection) Act, 1972, and *Pangshura tentorian circumdata* and *Morenia petersi* are new distributional records for Uttarakhand state (Bahuguna 2009, Bahuguna 2010) (Figs 6-9). Rawat and Pandav (2006) also reported three species; *Geoclemys hamiltonii*, *Kachuga kachuga* and *Lissemys punctata* from these wetlands and all are listed in Scheduled I of the IWPA (Table 5).

Marsh crocodile (*Crocodylus palustris* Lesson) was observed during this study and also reported in the past (Rawat and Pandav 2006, Rashtriya Sahara 2009). The crocodiles were reported to often enter the human settlements near Laksar in Ajitpur area. The distribution of crocodiles in Uttarakhand is restricted to Corbett Tiger Reserve and Baanganga wetlands.

Physico- Chemical Parameters:-

Temperature: Mean surface temperature of the various water bodies ranged from 25° to 30° C during May - September 2008 and 2009 (at 1200hrs- 1230 hrs). The highest ambient temperature (39° C) was recorded during May and June.

The increase in temperature in water leads to speeding up of chemical reactions and reduced solubility of gases (Hem 1989, NIH 1987-1988). Continuous depletion of ground water reservoirs and deterioration of quality in Haridwar is reported by Rao & Kumar (2002). The increase in temperature in the area must be due to recent spate of urbanization, increased vehicular pollution, regular incoming of pilgrims to Haridwar and ever increasing population in Haridwar. This increase in temperature can deteriorate the conditions of some of the water bodies (2-3 meters depth, Nainpur) harboring the important aquatic, semi aquatic fauna and water birds (Figs 6, 7).

pH and electrical conductivity : The pH recorded in the present study (Table 6) varied from 7.02 to 8.14 (in Lalpur near to Stone crusher unit). The range of pH for freshwater was within the permissible limit of BIS 10500 (1991). Electrical conductivity of water corresponds to the total concentration of dissolved compounds (electrolyte). It was noted to be varying from 0.191 to 0.588 ds/m. Highest has been noted to be in water bodies near to human settlements

Dissolved oxygen: Dissolved oxygen varied from 3 to 4 ppm in the present study. DO for healthy water has been recommended to be >6 ppm by BIS 1991 (Table 5).

Calcium: In the present study, Calcium concentration from all localities ranged from 24-76 ppm, highest Nainpur i.e. 64 ppm, whereas for Lalpur it was 48 ppm (Table 7). Calcium concentrations in natural waters are typically less than 15 mg l⁻¹ (NIH 1987-88). For waters associated with carbonate rich rocks, level may reach 30-100mg l⁻¹. Cremation sites near Nainpur (Fig 4) and stone crusher facility in Lalpur area (Fig 2) and intense human activities near Shahpur might have led to increased level of calcium in the water sample. It has been reported that acidic rainwater can increase the leaching of calcium from soil (Hem 1989, NIH. 1987-1988).

Magnesium: As noted, the magnesium concentration ranged from 12.0 ppm to 48 ppm, which is within the range of natural conditions. Natural levels of magnesium in freshwater may range from 1 to 100 mg l⁻¹. Magnesium occurs in many organometallic compounds and in organic matter since it is an essential element for living organisms. Although magnesium is used in many industrial processes these contribute relatively little to the total magnesium in surface waters (EIFAC 1964-90, ISO 1984, Hem 1989, WHO 1984a, 1984b).

Carbonate: Carbonate concentration in water ranged from 6 to 18 mg l⁻¹, it was noted to be nil at Nainpur and Tada Bhagmal localities (Table 6). Carbonate is uncommon in natural surface waters because they rarely exceed pH 9, whereas the groundwater can be more alkaline and may have concentrations of carbonate up to 10 mg per liter.

Bicarbonate concentration in surface water is usually less than 500 mg l⁻¹ (WHO 1984a, 1984b). In the present study, it was noted to be within the range and was the dominating anion present in surface water (Table 7).

Chloride: In the present observations it was noted to be very high i.e. of the 56.8 to 78.1 ppm. In freshwater, chloride concentrations are usually lower than 10 mg l⁻¹ and sometimes less than 2 mg l⁻¹ (APHA 1989, Hem 1989). Higher concentrations can occur near sewage and other waste outlets, irrigation drains, salt water intrusions, in arid areas and in wet coastal lines. In the present study, it seemed to be mainly through irrigation drains and waste outlets. As chloride is frequently associated with sewage, it is often incorporated into assessments as an indication of possible faecal contamination or as a measure of the extent of the dispersion of sewage discharge in water bodies (APHA 1989; Bestemyanov and Krotov, 1985; CEC 1976).

The physicochemical parameters recorded depicted that for the fishes pH was optimum. The value of dissolved oxygen in the present study varied from 3 to 4 mg /l. Oxygen plays a vital role to regulate the metabolic processes and is indicator of aquatic productivity. For average, good productivity dissolved oxygen should have concentration >5 mg/l (Banerjee 1967) and thus the D.O. recorded is less for the survival of the fish. The chloride level (56.8 to 78.1 mg/l) was noted to be too high depicting that the water might be leading towards eutrophication (CEC 1976, Jairam 1999).

Human activities around and in the area like cleaning cattle's, taking bath and discharge of remains of human cremation in small water bodies were affecting the health of the water as revealed by physical and chemical analysis of water (Tables 6,7). Agriculture practices around the area were also affecting the water level in remnant habitats as the drainage through water pumps were done and still going on to increase the land area and for irrigation purpose. The upland and moist shores of the study area were noted to be utilized for agriculture. The area was also exploited to collect stone for stone crusher unit present in the area. The local enquiries revealed the presence of channel full of water till 1980 in the same locality meeting Ganga, thus called Baanganga, but the construction of barrages/ dam on Ganga led to decrease in the water level in the channel, thus produced different land categories (Adhikari, 2008). Rao (1998) extensively surveyed from Rishikesh to Kanpur and reported 12 species of freshwater turtles. Localities near Baanganga (Haridwar) were surveyed by Adhikari (2008) and Kumar (2007) for the study of vegetation types and fish fauna. They reported that the study area harbored many islands, varying in shape and size, which remained underwater during the rainy season and provided a good habitat to various plant taxa, birds (resident as well as migratory) and animals mainly Swamp deer, *Cervus duvauceli duvauceli* Cuvier, a critically endangered species and Hog deer, *Axis porcinus* (Zimmermann).

Activities like sand mining, agriculture, reclamation of wetlands and riparian areas, alternation of rivers for irrigation and generation of hydroelectric power, pollution, siltation, eutrophication and intensive fishing have been reported to cause the killing of turtles, affecting their nesting (Bhupathy and Chaudhury 1995, Sharma 1998, Rao 1998). Surveys to the present study area since May 2008-September 2010 unfolded the deteriorating situation of the habitat for aquatic and semi-aquatic fauna. Main factors, which have affected the habitat, are: barrage/dam construction nearest to habitat, increase in human activities including agriculture, use of open water bodies for cattle and human bath, cremation ground including weed proliferation in many water bodies. The increase in ambient atmospheric temperature that resulted in the low level of water in many small water bodies causing death of fishes and other aquatic fauna as observed during the surveys.

Rao (1998) undertook extensive survey in Ganga River (645 km) at a stretch between Rishikesh to Kanpur to know the status of freshwater turtles and reported 12 species of freshwater turtles under seven genera. He observed that all the way from Rishikesh to Kanpur most of the Ganga Ghats have religious importance. Large numbers of the pilgrims take holy bath, do cremation and post cremation activities and thus become a major source of pollution to the river; large number of factories like IDPL, BHEL, sugar, chemicals, fertilizers, engineering, cotton and tanneries are situated on the bank of the river. The natural flow of the Ganga River has been checked due to construction of hydro-power barrages up-stream at Rishikesh, Haridwar, Bijnor and Narora. Two barrages have been constructed at Haridwar and Bijnor for irrigation purpose through Upper Ganga canal and Madhya Ganga Canal, respectively (Rao 1985). The present study although limited to a small area of 8-10 km has significant importance as the area still has freshwater turtle species in various water bodies and they need conservation and protection, as lowering the water levels and scarcity of the food might increase the mortality rate besides exposing them to the danger of illegal trade. Local people were though aware of the legal protection of the freshwater turtles were not

able to differentiate Scheduled and unscheduled species. Local enquiry revealed that the people from outside were coming to the area and asking for particular species of freshwater turtles for medicine.

Illegal trade of freshwater turtles is a global problem, involving many of the developing countries. There is a demand of turtles in China, Singapore and Hong Kong markets. In India, the illegal trade of turtles is going on and the most common routes are from Kanpur to Kolkata market. Uttarakhand State earlier rich in biodiversity of turtles but recently the population of turtles has now been found to be restricted (Husain and Ray 1995), thus indicating threats to the population of turtles and tortoises in the State. There is an urgent need for the management to conserve the habitat and the species.

Conclusion:-

Water bodies in Baanganga wetlands of Haridwar, Uttarakhand are not in healthy conditions. This river is also passing through the conservation area Jhilmil Jheel, thus needs special attention for habitat management. Available oxygen, adequate nutrients and absence of toxic chemicals are essential factors for growth and reproduction for aquatic and semi-aquatic fauna. In the present study increasing atmospheric temperature, increased pH in some water bodies, low oxygen in water, high level of chloride ions were noted to be harmful for the habitat of aquatic and semi aquatic fauna still thriving in the area. Many scheduled (under IWPA) species were found to be present in the area including turtles, crocodile and mammals, indicating the importance of the area. Low level of water during summer and increased pressure due to anthropogenic activities were noted to be increasing the threat to their survival and make them easy target of poaching.

Management plan for the area:

1. The water body in Nainpur locality can be a good habitat for aquatic and semi aquatic fauna after implementation of some management measures like: removal of weeds, pumping air, shifting cremation ground and involvement of local people for management of habitat, reintroduction of aquatic-semi aquatic fauna from deteriorating habitat to habitat in Nainpur.
2. Afforestation programmes around the area should be initiated to improve the habitat and to control the rising temperature.
3. Strong actions are required to conserve the threatened and endangered species as some of them may reach the threshold of critical endangerment.
4. Involvement of local people: generate awareness among local community about the importance of fauna residing in the area; involve them in reintroduction programmes, afforestation programme. Local people and turtle trappers should be involved in various surveys to get information of illegal trade.

Table 1:- Land types present

S.No.	Localities	Land types
1	Shahpur	Small water bodies 20%, upland, agriculture area covering 60 % of the area, human settlements 20 %
2	TandaBhagmal	Few water bodies 10%, agriculture area 55 % and human settlement 20 %
3	Lalpur	Open water bodies 30 %, Agriculture 50 %, rest other types
4	Nainpur	Open water bodies 50%, marshy area 20%,upland10%,agriculture 10%,human settlements 10%
5	Indreshpur	Open water bodies 10%, agriculture 55%, rest human settlements
6	Ranjitpur	Open water body 75% (near to Ganga bank), agriculture 25%, rest human settlement
7	Bhikampur	Open water body 25%,agriculture area 50%, human settlements 25%

Table 2:- Mollusca diversity in Baanganga area

Sl. No.	Name of Place	Date of Collection	Species	No. of specimen	Identified by
1.	Near Solani Bridge Dist.Haridwar	9.11.2009	Gastropoda		Dr. G. Sharma/ Dr.H.Nesemann
			<i>Bellamyia bengalensis</i>	19	-do-
2.	Solani River, Haridwar	25.06.2010	Gastropoda	2	-do-
			<i>Lymnaea accuminata</i>		
			<i>Bellamyia bengalensis</i>	3	-do-
			Pelecypoda		
			<i>Parreysia (favidens)viridula</i>	1	-do-
			<i>Radiatula occata</i>	9	-do-
			<i>Radiatula caerulia</i>	1	-do-
			<i>Lamellidens marginalis</i>	1	-do-
3	Nainpur(From large open water body) Haridwar	11.10.2008	Gastropoda		
			<i>Indoplanorbis exustus</i>	2	-do-
			<i>Bellamyia bengalensis</i>	2	-do-
			<i>Idiopoma dissimilis</i>	20	-do-
4	Haridwar and around(then away from Solani River)	25.02.2010	Gastropoda		
			<i>Lymnaea accuminata</i>	3	-do-
			<i>Radix luteola</i>	1	-do-
			<i>Bellamyia bengalensis</i>	17	-do-
4	Kanpur Ganga (U.P.)	19.04.2011	Gastropoda		
5	Nainpur(From large open water body) Haridwar	11.10.2008	Gastropoda		
			<i>Indoplanorbis exustus</i>	2	-do-
			<i>Bellamyia bengalensis</i>	10	-do-
7.	Baan Ganga, Nainpur, Haridwar	18.06.2009	Gastropoda		
			<i>Bellamyia bengalensis</i>	16	-do-
			<i>Lymnaea accuminata</i>	1	-do-
8	Khatoli (Pond) Haridwar	29.07.2009	Gastropoda		
			<i>Bellamyia bengalensis</i>	79	-do-
9.	Baan Ganga near village Tanda, Haridwar	14.05.2008	Gastropoda		
			<i>Bellamyia bengalensis</i>	10	-do-
			<i>Indoplanorbis exustus</i>	2	-do-
			<i>Lymnaea accuminata</i>	1	-do-
10.	Ranjitpur (Haridwar)	17.10.2008			
			<i>Idiopoma dissimilis</i>	6	-do-
			<i>Radix luteola</i>	1	-do-
11	Nainpur near Luksar (Dist Haridwar).	30.12.2008	Gastropoda		
			<i>Bellamyia bengalensis</i>	6	-do-
			<i>Indoplanorbis exustus</i>	1	-do-
			<i>Lymnaea accuminata</i>	2	-do-
12.	Lalpur (Near Haridwar)	14.11.2008	Gastropoda		
			<i>Bellamyia bengalensis</i>	5	-do-
			<i>Idiopoma dissimilis</i>	3	-do-
			<i>Indoplanorbis exustus</i>	1	-do-
			<i>Lymnaea accuminata</i>	1	-do-

13.	Solani River Fc no.3	15.09.2009	Pelecypoda		
			<i>Radiatula occata</i>	1	-do-
14.	Nainpur : Haridwar & around	18.07.2008	Gastropoda		
			<i>Bellamya bengalensis</i>	1	-do-
			<i>Pila globosa</i>	1	-do-
			<i>Lymnaea accuminata</i>	1	-do-
			<i>Indoplanorbis exustus</i>	6	-do-

Table 5:- List of species of freshwater turtles from various localities.

S.No	Species	Localities	Common Name	Category under IWPA 1972
1	<i>Pangshura tecta</i> (Gray)	Shahpur, TandaBhagmal, Lalpur, Nainpur, Indreshpur, Ranjitpur,	Indian roofed turtle	Schedule- I
2	<i>Pangshura tentoria circumdata</i> (Gray)	Ranjitpur, Shahpur	Circled Indian tent turtle	Not listed
3	<i>Lissemys punctata andersonii</i> (Webb)	TandaBhagmal, Lalpur, Nainpur, Ranjitpur	Indian flap shelled turtle	Schedule- I
4	<i>Morenia petersi</i> (Anderson)	Bhikampur	Indian eyed turtle	Not listed

Table 3:- List of fishes

1 Order Cypriniformes				
Family 1 Cyprinidae		Subfamily Rasborinae		
S.No..	Species	Locality	Common Name	Local Name
1	<i>Daniorerio</i> (Hamilton.1822)	Ranjitpur village, Ganga River-19exs,Bann Ganga, Near Village Tanda Bhagma-01ex, Lalpur, Luxar, dist. Haridwar-01ex	Zebrafish	Chahal
2	<i>Esomus danricus</i> (Hamilton.1822)	Nainpur, Luxar-06exs	Flying barb	Chiddullu
3	<i>Rasboradaniconius</i> (Hamilton.1822)	Ranjitpur village, Ganga River-12exs, Bann Ganga, Near Village Tanda Bhagma, Nainpur, Luxar-02exs, Nainpur, Luxar-02exs	Blacklinerasbora	Chinoodolah
Subfamily Cyprininae				
S.No..	Species	Locality	Common Name	Local Name
1	<i>Puntiuschola</i> (Hamilton..1822)	Ranjitpur village, Ganga River-01ex, Nainpur, Luxar-12exs	Chola barb	Pothi
2	<i>Puntiussophore</i> (Hamilton. 1822)	Bann Ganga near Village Shahpur, Sitakhera-03exs, Nainpur, Luxar-01ex	Spotfin swamp barb	Punti
3	<i>Puntiusticto</i> (Hamilton.1822)	Bann Ganga near Village Shahpur, Sitakhera-05exs,Nainpur-02exs	Ticto barb	Pothia
Subfamily Cultrinae				
1	<i>Laubucalaubuca</i> (Hamilton. 1822)	Ranjitpur village, Ganga River - 07exs	Indian glass-barb	Dennahrah
Family 2 Homalopteridae				

Subfamily Noemachlinae				
S.No..	Species	Locality	Common Name	Local Name
1	<i>Acanthocobitis.botia</i> (Hamilton. 1822)	Nainpur, Luxar-01ex	Loach	
Family 3 Cobitidae				
Subfamily: Cobitinae				
1	<i>Lepidocephalichthysguntea</i> (Hamilton. 1822)	Bann Ganga, Near Village Tanda Bhagma-01ex, Nainpur, Luxar-16exs	Loach	Jiwal
2. Order Siluriformes				
Family 1 Siluridae				
S.No..	Species	Locality	Common Name	Local Name
1	<i>Ompokpabda</i> (Hamilton. 1822)	Shahpur (Haridwar and Around)-01ex	Pabada Catfish	Pabdaha
Family 2 Siluridae				
S.No..	Species	Locality	Common Name	Local Name
1	<i>Hara jerdoni</i> Day.1870	Nainpur, Luxar, dist. Haridwar- 02exs	Sylhethara	
3. Order Perciformes				
Family 1 Nandidae				
1	<i>Nandusnandus</i> (Hamilton. 1822)	Ranjitpur village, Ganga River- 01ex,Bann Ganga, Near Village Tanda Bhagma- 03exs, Nainpur, Luxar- 03exs,	Mottled Nandus	Chambal
2	<i>Badisbadis</i> (Hamilton. 1822)	Nainpur, Luxar-01ex	Dwarf chameleon fish	Sumha
Family 2 Belontiinae				
Subfamily Trichogasterinae				
S.No..	Species	Locality	Common Name	Local Name
1	<i>Trichogasterfasciata</i> Bloch& Schneider,1801	Bann Ganga near Village Shahpur, Sitakhera- 09exs,Bann Ganga, Near Village Tanda Bhagma- 21ex,Nainpur- 02exs,Shahpur(Haridwar and Around)-01ex, Nainpur, Luxar-01ex	Striped Gourami	Kanghee
2	<i>Trichogasterchuna</i> (Hamilton 1822)	Ranjitpur village, Ganga River-01ex,Bann Ganga, Near Village Tanda Bhagma, Dist. Haridwar- 08exs, Nainpur (Haridwar and around)- 01ex	Sunset Gourami	Kanghee
3	<i>Trichogaster latius</i> (Hamilton. 1822)	Nainpur, Luxar-01ex, Lalpur, Luxar, dist. Haridwar-03exs	Dwarf Gourami	Khosti
Family 3. Channidae				
1	<i>Channa</i>	Ranjitpur villae, Ganga	Spotted	Phool dhok

	<i>punctata</i> (Bloch.1793)	River (Haridwar)-01ex, Bann Ganga near Village Shahpur, Sitakhera, dist. Haridwar-01ex, Nainpur (Haridwar and around)-02exs, Nainpur, Luxar-21exs	Snakehead	
Family 4. Mastacembelidae				
1	<i>Macrognathus pancalus</i> Hamilton.1822	Ranjitpur village, Ganga River (Haridwar)-01ex, Bann Ganga near Village Shahpur, Sitakhera, dist. Haridwar-01ex, Nainpur, Luxar-01ex	Striped Spiny eel	Bamimachi

Table 4:- Species of anurans, Amphibia from the study area

S.No.	Species	Examples	Locality	Common Names
1	<i>Fejervarya</i> species	5 exs	Nainpur	Alpine cricket frog
2	<i>Euphlyctis cyanophlyctis</i> (Schneider)	15 exs	Nainpur, Ranjitpur, Lalpur	Common Skittering frog
3	<i>Hoplobatrachus tigerinus</i> (Daudin)	3 exs	Nainpur, Bhikampur	Indian bull frog

Table 6:- Water analysis of different localities covered during survey (April 2008- Sep.2009)

S.No.	Localities	Mean Atmospheric temp (in °C) (12 am to 12.30 pm)	Mean Water surface temperature (in °C) (12 am to 12.30 pm)	pH Mean \pm SD	EC(in ds/m) Mean \pm SD	Dissolved oxygen(DO) in ppm Mean \pm SD
		Sd: not recommended	Sd: not recommended	Sd: pH 6.5 to 8.5	Sd: EC not recommended	Sd: DO >6 ppm
1	Shahpur (in May)	39	31	7.02 \pm 0.0	0.588 \pm 0.0	4 \pm 0.01
2	Tanda Bhagmal (in May)	39	30	7.65 \pm 0.0	0.528 \pm 0.0	3 \pm 0.01
3	Lalpur (near to Stone crusher)(in June)	39	31	8.14 \pm 0.1	0.465 \pm 0.1	3 \pm 0.1
7	Lalpur (in June)	39	30	7.31 \pm 0.0	0.203 \pm 0.0	4 \pm 0.02
4	Nainpur (large open water body) (in July)	38	32	7.26 \pm 0.0	0.257 \pm 0.01	4 \pm 0.01
5	Nainpur (large open water body) (in July)	38	30	7.35 \pm 0.2	0.288 \pm 0.0	4 \pm 0.01
6	Shahpur (in Aug)	34	30	7.75 \pm 0.0	0.324 \pm 0.01	3 \pm 0.02

8	Ranjitpur (near to Ganga) Se(in p)	32	25	7.03±0.0	0.191±0.0	4±0.02
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Table 7:- Chemical parameters of water analyzed SR Standard Range**SD: Standard value as recommended by BIS 10500 (Bureau of Indian Standards) 1991:**

S.No	Samples	Calcium (ppm) Mean±SD	Magnesium (ppm) Mean ± SD	Chloride (ppm) Mean ± SD	Carbonate (ppm) Mean ± SD	Bicarbonate (ppm) Mean ± SD
		SR 15ppm (Ref NIH 1987-88)	SR 1-100ppm (Ref Hem 1989, WHO 1984a, 1984b)	SR <10ppm (Ref APHA 1989; CEC 1976)	SR <10ppm (Ref. WHO 1984a, 1984b)	SR < 500ppm (Ref WHO 1984a, 1984b)
1	Shahpur	76± 0.01	40.8± 0.0	63.9± 0.02	18± 0.1	335.5± 0.2
2	TandaBhagmal	68± 0.0	12.0± 0.0	71.0± 0.1	nil	164.7± 0.1
3	Lalpur (near to Sone crusher)(in June)	48± 0.01	33.6± 0.0	63.9± 0.0	15± 0.0	329.4± 0.2
4	Nainpur (large open water body) (in July)	24± 0.0	31.2± 0.1	63.9± 0.2	nil	134.2± 0.0
5	Nainpur (large open water body) (in July)	64± 0.1	12.0± 0.2	56.81± 0.1	18± 0.1	231.8± 0.01
6	Shahpur (in Aug)	44± 0.0	48.0± 0.01	78.1± 0.02	15± 0.01	311.1± 0.01
7	Lalpur (in June)	40± 0.01	26.4± 0.01	78.1± 0.01	12± 0.01	231.8± 0.01
8	Ranjitpur (near to Ganga) (in Sep)	28± 0.1	24.0± 0.2	78.1± 0.02	6.0± 0.01	164.7± 0.01

Source: www.bis.org.in



Fig 1:- Map of the study area: Banganga, Near Lakshar road. District Haridwar , scale 1:500,000, source : Survey of India, Dehradun.

Localities of study area.



Fig 2:- Lalpur: small water body with stone crusher machine on roadside.



Fig 3:- Ranjitpur: river Ganga; the bank of which are covered with sugarcane plantation.



Fig 5:- Bhikampur: infested with weeds, Is a habitat of crocodile.



Fig 6:- *Pangshura tecta* (Gray) Indian roofed turtle.



Fig 7:- *Leissymus punctataandersoni* Flap Shelled turtle.



Fig 8:- *Pangshura tentoria circumdata*, South Indian roofed turtle.



Fig 9:- *Morenia petersi*), Indian eyed turtle.



Fig 10:- *Hoplobatrachus tigerinus* (Daudin), Indian bull frog.

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RESEARCH ARTICLE

SURVEY ON CONTRIBUTIONS TOWARDS LIVE FACE RECOGNITION IN VIDEO STREAMING.

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Image attributes, Face recognition, Templates, Compression, Content analysis, Video content extraction, Image processing, Temporal segmentation, Scene segmentation.

Abstract

Image detection to identify the targeted image and extract its contents in any of the live videos which are being streamed. Every particular image of a layman contains several unique attributes for Example, the space between the eyebrows or a different type of nose. The proposed project looks into recognizing this special features and detecting them through the videos being processed in of the environment. This was attributed to the ability of artificial intelligence management, as a computer technology, to generate, process, and analyze large volume of data at a time, compared to when humans are used, to help managers in quick decision making because results of the analysis are available in time.

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Introduction:-

With the advancement in technology and more and more invention being done in the field of computer technology, the risk of theft and destruction has been increased to vast levels. A security system for the protection of all kinds of destructive things round the world has become one of the major necessity. This kind of a system would be creating help to not only the field of technology but also in vehicle monitoring, theft and illegal activity investigation. The approach can also be used in detecting murders and crimes happening around.

A self-recognizing system that detects image in live video streaming is being studied. Every human face is different, every face is gifted with certain attributes that is one's own identity and this makes each face different from other. This personal identity can be used as a tool to detect and verify different people round the ground. The basic requirement for the work are: Images, for representing the frame data, a numerical matrix whose values can be colours, colour map entries, etc. Colour Maps, which map entries into a colour space, allowing an additional indexation level. Image Display Converters and Image IO Handlers, that convert images in the specific formats of the platforms and vice-versa

The approach seeks to store the unique feature of the image in its database. Further, the extraction of this feature is verified through Eigen- Face algorithm. Here whenever the system detects an image, it crops the unique feature from the face and creates a similar possible and identical image of the face in its image dataset. So the next time the same face occurs in front of the face it detects the image and extracts the contents about the image from the video streaming. This indirectly detects and monitors

Related work:-

The idea for extracting the content of information is the Accessibility Problem. A problem that is even more relevant for dynamic multimedia data, which also have to be searched and retrieved. While content extraction techniques are

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reasonably developed for text, video data still is essentially opaque. Its richness and complexity suggests that there is a long way to go in extracting video features, and the implementation of more suitable and Effective processing procedures are an important goal to be achieved [1]. The increase in the diversity and availability of electronic information led to additional processing requirements, in order to retrieve relevant and useful data: the accessibility problem. This problem is even more relevant for audio visual information, where huge amounts of data have to be searched, indexed and processed. Most of the solutions for this type of problems point towards a common need: to extract relevant information features for a given content domain [2]. A process which underlies two difficult tasks: deciding what is relevant and extracting it. In fact, while content extraction techniques are reasonably developed for text, video data still is essentially opaque. Despite its obvious advantages as a communication medium, the lack of suitable processing and communication supporting platforms has delayed its introduction in a generalized way. This situation is changing and new video based applications are being developed. In our research group, we are tools for indexing video archives for later reuse, a system for content analysis of TV news, and hyper video systems where hyperlinks are established based on content identification in different video streams. These applications greatly rely on efficient computational support, combining powerful image analysis and processing [3]. Each of these concepts is represented by a (C++) class and integrated in a systematic hierarchy. Tools for data processing are applied to the described data modelling classes, and also modelled as a hierarchy of classes [4]: The Image OPs, these operators represent functions which are applied to image regions and extract "single-image" or sequential content features. A shot object is a discrete sequence of images with a set of temporal attributes such as frame rate and duration and represents a video segment [5]. A shot sequence object groups several shots using some semantic criteria. Views, are used to visualize and browse shots and shot sequences.

The approach in this study is to construct dynamic templates in a bottom-up fashion using information on problem types and associated discourse structure [6]. The classification algorithm uses different cropping styles to detect the 3D image in the video streaming.

Face recognition has been a tough problem of biometrics and it has a variety of applications in modern life. The problems of face recognition attract researchers working in biometrics, pattern- recognition and computer vision. Several face recognition algorithms are also used in many different applications apart from biometrics, such as video compressions, indexing's etc. [7]. They can also be used to classify multimedia content, to allow fast and efficient searching for material that is of interest to the user. An efficient face recognition system can be of great help in forensic sciences, identification for law enforcement, surveillance authentication for banking and security system, and giving preferential access to authorised users i.e. access control for secured areas etc. [8]. The problem of face recognition has gained even more importance after the recent increase in the terrorism related incidents.

Background: Image Detection:-

This section introduces image detection and presents a list of common performance objectives and the techniques used to achieve them. Moreover, a brief overview of the evolution of image processing until the appearance of the PCE-based architecture is included. Finally, the limitations of current solutions are described, in order to identify how system can be future improved.

A. In this paper, it proposes to improve MPW with the aim of sampling a smaller number of PWs without any loss in detection rate and false positive rate. In addition, we solve the problem of determining the number of PWs in each stage. We call the proposed algorithm iPW does not need to generate a large number of PWs at the initial iteration (stage), which greatly reduces the detection time. Even if the initial PWs do not contain any object, iPW can detect the objects at next stages. 2) MPW unnecessarily draws too many PWs around the positive windows whereas iPW avoids generating these redundant PWs by using the information of both rejected and accepted PWs. 3) To use MPW, one has to empirically set the number of PWs in each stage whereas this is not a problem because iPW generates a single PW in each iteration (stage). 4) IPW fully makes use of the information of rejected negative PWs while MPW almost completely depends on the accepted positive PWs. Rejected negative PWs are used to directly suppress the PD around these windows and at the same time indirectly enhance the PD beyond the windows. In this sense, iPW is rejection-oriented while MPW is acceptance-oriented. 5) In MPW, the uniform distribution is used in the initialization and plays an unimportant role in the later stages so that it can be omitted. In iPW, a dense uniform distribution is used to play an important role for sampling useful PWs by rejecting background. IPW fully makes use of the information of rejected negative PWs while MPW almost completely depends on the accepted positive PWs. Rejected negative PWs are used to directly suppress the PD around these windows and at the same time indirectly enhance the PD beyond the windows. In this sense, iPW is rejection-oriented while MPW is acceptance-oriented. 5)

In MPW, the uniform distribution is used in the initialization stage and play unimportant role in theater stages so that it can be omitted. In iPW, a dented uniform distribution is used to play an important role for sampling useful PWs by rejecting background(i.e., no object) regions.6) iPW utilizes dented Gaussian distribution while MPWutilizes full Gaussian distribution for sampling PWs. Byusing dented Gaussian distribution, iPW avoids drawingmany unnecessary PWs around the object and object-likeregions.7)

B. It provides a comprehensive review on the recent advances in face anti-spoofing. While most previous works on face spoofing detection are based on analyzing only the luminance (i.e. gray-scale) information of the face images, we introduce a novel and appealing approach using color texture analysis and demonstrate that the Chroma component can be very useful in discriminating fake faces from genuine ones. We exploit the joint color-texture information from theluminance and the chrominance channels by computinglow-level features using different descriptors extractedfrom different colorspace. We investigate how welldifferentcolor spaces and descriptors can be used for describing the intrinsic disparities in the color texture between genuine faces andfake ones. We also perform afusion study to analyze the complementarity of the different descriptors and different colorspace. We conduct extensive experimental analysis on the three latest and most challenging spoofing databases using their pre-defined publicly well-defined experimental evaluation protocols ensuring the reproducibility of the results and a fair comparison against the state of the art. We provide an extensive comparative analysis againstthe state-of-the-art face spoofing detection techniques andshow that our proposed approach outperforms all existingmethods on two databases and achieves very competitiveperformance on the third databaseUnlike most of the methods proposed in the literature, our proposed approach is able to achieve stable performance across all the three benchmark datasets. Furthermore, in our inter-database evaluation, the facial color texture representation showed promising generalization capabilities,thus suggesting that color texture can be more stable in unknown conditions compared to its gray-scale counterparts.

Research onnon-intrusive software-based face spoofingdetection has mainly been focusing on analyzing gray-scaleimages and hence discarding the color information whichcan be a vital visual cue for discriminating fake faces fromgenuine ones. In a very recent work, Wen et al. [6] proposedcolor quality based features that describe the chromaticdegradation and the lack of color diversity of recaptured faceimages. However, the actual local variations in color textureinformation was not exploited for face spoofing detection

There are many ways to perform face scrambling. Forexample, scrambling can be done simply by masking orcartooning. However, this kind of scrambling will simplylose the facial information, and hence subsequent facerecognition or verification becomes unsuccessful in this case.Especially for security reasons, it is obviously not a goodchoice to really erase human faces from surveillance videos.In comparison, the Arnold transform as a basic step in many encryption algorithms, is a kind of recoverable scrambling method. Scrambled faces can be unscrambledby several manual tries.

C.Moving object detection plays an important rolein many video processing applications, such asobject tracking, categorization, identification, and videocondensation. It often serves as pre-processing for higher levelvideo analyses and its performance directly affects the performance of the subsequent applications. For object tracking, if moving object is detected as two or two moving objects are detected as one, the tracking result may be incorrect. For object categorization, incomplete or adhesive detection of moving objects may lead to wrong categorization, and it is the same case for object identification. For video condensation, object tracking is also an indispensable part. It's not the desired result if the head and legs of one person appear at different time in the condensed video. Ideally, a detection method should detect each moving object separately without breaking. Backgroundmodelling is indispensable for moving object detection in many cases and many works have been done in this research area. In early works, the background model was constructed for each pixel independently. In , a single Gaussian model was used to model the value variations of each pixel, and the parameters of the Gaussian model were updated recursively with an adaptive filter. It is robust in modelling the static background, but is sensitive to dynamic background variations. To address these problems, the mixture of Gaussian models (Moggs) was proposed in . In addition, a series of variants were proposed to improve the performance of background modelling. However, the Gaussian model-based methods are all based on the assumption that the pixel intensity follows a Gaussian distribution, which is not always correct. In contrast, a nonparametric method was proposed in , where each pixel is directly modelled with a probability density function without any assumption on the distribution of the pixel values. Since all the possible pixel values are modelled into the probability density function, dynamic background modelling problem can be handled and illumination change can be adapted progressively. In [6], a Bayesian decision rule for

background and foreground classification was constructed based on different feature selections for static and moving backgrounds, and is able to deal with dynamic background problems better than MoG. In [7] and [8], background pixel values are quantized into a set of codebooks to form the background model which is efficient in memory and speed.

D. Given a training dataset, faces are forwarded to the training procedure. The offline procedure then learns its semantic salience map. Following this, the database is scrambled and the feature space is reconstructed by multiplying salient feature according to their semantic salience weights. Random sampling is then applied to select features sparsely to construct as many kernels as is allowed, and discriminant analysis is used to learn a kernel subspace for each kernel. After a scrambled facial image is input as a test, the input is projected into each kernel subspace, and the distance to each training sample is computed. The decision procedure is based on the combination of all kernel subspaces. It is noted that we can have unscrambled images (mainly for statistic salience learning) in the offline training because offline training is carried out centrally with authorities'/business supervisors' permission and will not undermine users' privacy. Privacy protection is mainly an issue with distribution over the internet. In this scheme, the training procedure can be carried out offline. The online verification then becomes purely a data-driven process. In the test procedure, all test images, we have identified a new challenge in scrambled face recognition originated from the need for biometric verification in emerging IoT applications, and developed a salience-aware face recognition scheme that can work with chaotic patterns in the scrambled domain. In our method, we conjectured that scrambled facial recognition could generate a new problem in which "many manifolds" need to be discovered for discriminating these chaotic signals, and we proposed a new ensemble approach – Many-Kernel Random Discriminant Analysis (MK-RDA) for scrambled face recognition. We also incorporated a salience-aware strategy into the proposed ensemble method to handle chaotic facial patterns in the scrambled domain, where random selection of features is biased towards semantic components via salience modelling. In our experiments, the proposed MK-RDA was tested rigorously on three standard human face datasets. The experimental results successfully validated that the proposed scheme can effectively handle chaotic signals and drastically improve the recognition accuracy, making our method a promising candidate for emerging IoT applications. One of the most important tasks for video analysis is to specify a unit set, in which the video temporal sequence may be organized.

The different video transitions are important for video content identification and for the definition of the semantics of the video language, making their detection one of the primary goals to be achieved. The basic assumption of the transition detection procedures is that the video segments are spatially and temporally continuous, and thus the boundary images must suffer significant content changes. Changes, which depend on the transition type and can be measured. The original problem is reduced to the search of suitable difference quantification metrics, whose maximums identify, with great probability, the transition temporal locations

Goals:-

Once installed, test whether or not it works by firing up a python session. If there are no any errors then, move further. Next step is the cascading the image (the cascade is just a Xml file that contains the data to detect faces. Read the image and convert it to greyscale. Many operations in Open Cv are done in grayscale. This is detection of the face that comes in front of camera. Once the face detection is done, next part to be done is its recognition in such a way that it stores the value of each faces, so that the next time the face appears on the screen it is able to name and retrieve data by its own. For this Eigen-face Algorithm is used, the Eigen values of each face is stored under the database. Prepare a training set of face images. The pictures constituting the training set should have been taken under the same lighting conditions, and must be normalized to have the eyes and mouths aligned across all images. Subtract the mean. The average image \bar{a} has to be calculated and then subtracted from each original image. Calculate the eigenvectors and eigenvalues of the covariance matrix S . Choose the principal components. Sort the eigenvalues in descending order and arrange eigenvectors accordingly. The number of principal components k is determined arbitrarily by setting a threshold ϵ on the total variance. These Eigen faces can now be used to represent both existing and new faces.

Identified Challenges:-

Extensive experiments on the three latest and most challenging spoofing databases (the CASIA FASD, the Replay-Attack Database and MSU MFSD) showed excellent results. On the CASIA FASD and the MSU MFSD, the proposed facial colour texture representation based on the combination CoLBP and LPQ features computed over HSV and YCbCr colour spaces outperformed the state of the art, while very competitive results were achieved on the Replay-Attack Database. More importantly, the proposed approach was able to achieve stable performance

across all three benchmark datasets unlike most of the methods proposed in the literature. Furthermore, in our inter-database evaluation, the facial colour texture representation showed promising generalization capabilities, thus suggesting that colour texture seems to be more stable in unknown conditions compared to its gray-scale counterparts. To be consistent with many previous studies, it is worth noting that face normalization or the limits of the face bounding box were not optimized in our experiments.

Identified Analysis:-

The system contains four modules:

Image detection, Image recognition, Image classification and content extraction. Image detection is the basic step towards identification of the image here the face in front of the screen is identified and a cropping structure (dialog box) appears over the detected area. The next step is to recognize the unique feature of the face the system recognized the basic pixels and creates the base feature of the face. Further a similar possible image of the face is created using this base feature and the image is classified under various folders. The live streaming data of the previously stored is compared and the contents from the image is detected and conclusions are made.

Experimental Setup:-

Following explain the basic experimental setup of the system:

Accept the image from the live video streams

Classify it and crop appropriate features

Identify function using Eigen-face algorithm

Generate equations (with the help of Eigen values)

Solve Equations

Display Content

Summary:-

present a multi-stage investigation system that can be used in various fields of cyber security systems. The crimes and various threats can be easily solved through recognizing images in the pictures as well as live video streams ensuring a safety towards a destructive world.

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RESEARCH ARTICLE

PULMONARY ALVEOLAR MICROLITHIASIS: CLINICAL AND RADIOPATHOLOGICAL REVIEW.

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Abstract

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Introduction:-

Pulmonary alveolar microlithiasis (PAM) is a rare and slowly progressive diffuse chronic lung disease characterized by deposition of small calcium phosphate spherules within the alveolar cavity.(1) The etiology of the disease is still unknown and is supposed to be due to defect in the inherited local enzyme that is responsible for calcium deposition.(2)The disease is usually seen from birth up to 40 years of age.(1,3) Clinically, the disease course is different. Most of the patients are asymptomatic or having very mild symptoms and majority of patients either having normal or restrictive pulmonary function tests. (1,3)

Case:-

We report a case of 44-year-old male, tailor by occupation who presented with breathlessness on exertion for 3 years. Initially, grade I MMRC dyspnea progressed to grade II MMRC on presentation, over 3 years. Associated with mild cough and scanty expectoration which was gradually progressive, non-foul smelling, non-blood tinged. No history of fever/chest pain.

Personal history-Smoker for 10 years,1-2 cigarettes/day, stopped 5years back. Also, had a history of cement dust exposure from a nearby cement factory for 25-30 years. No significant family history.

On general examination vitals, stable, clubbing grade 2 observed. No pallor, icterus, cyanosis, lymphadenopathy or edema. Systemic examination: Upper respiratory tract normal.

On inspection, chest bilaterally symmetrical, trachea sign negative. No scars, sinuses or dilated veins seen, movements bilaterally equal. Palpation: Inspectory findings confirmed, no local rise of temperature or tenderness, tactile vocal fremitus equal on both sides. Percussion: Resonant note all over lung fields. On auscultation: Late fine inspiratory crepitations heard over interscapular, infrascapular and infraaxillary areas. Vocal fremitus: equal on both sides. Rest of the systems are normal.

On chest x-ray (Fig-1) diffuse, dense, large calcifications noted in bilateral lung fields. HRCT chest (Fig-2,3) revealed dense, large parenchymal calcifications in apico-posterior and anterior segments of bilateral upper lobes, thick septal calcifications in rest of the lung fields, predominantly in the basal segments of the bilateral lower lobes. Spirometry showed restrictive pattern. Bronchoscopy showed fibrosis in the secondary bronchi Bronchoscopic guided lung biopsy was done.

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Histopathological examination (Fig-4) of the lung section revealed calcified microlith in the intra-alveolar spaces. Special stain -Von kossa stain (Fig-5) positive confirmatory of pulmonary alveolar microlithiasis.

Treatment:-

Treatment with corticosteroids and symptomatic treatment was given and on follow up visits, patient improved symptomatically.

Discussion:-

PAM is a rare chronic parenchymal lung disease often diagnosed incidentally in asymptomatic patients or become symptomatic between 3rd and 4th decades(1,3,4,5). Basically characterized by numerous small calculi (denominated calciferites, calcospherites or microlites) within air spaces.(5) Calculi are round or ovoid in shape and range from 0.01 to 2.8mm.(2) Studies confirmed that the disease is caused by autosomal recessive inheritance of a mutation in the gene (SCL34A2 gene) on chromosome 4, that produces the protein that is the only known sodium-dependent phosphate transporter in the lungs.(4,6) As a result of this mutation, the protein is unable to transport phosphorus ions from the alveolar space into type II pneumocytes, and this failure leads to the development of calcospherites in the alveolar space.(6) At clinical presentation patients usually demonstrate a lung disorder with restrictive pattern as seen in our case. Adult patients commonly show progressive deterioration of the pulmonary function and death usually occurs in mid-life because of respiratory failure associated with cor pulmonale.(5)

Plain chest x-ray usually shows diffuse bilateral symmetric micronodular calcification (sandstorm) mainly in middle and lower zones; apical bullae and black pleural lines. HRCT thorax remains the imaging technique of choice to diagnose PAM. Findings include ground glass opacity (most common), sub-pleural calcification, confluent and diffuse calcified nodules and dense consolidation. Septal thickening, 'black pleura' sign due to sub-pleural cyst and crazy paving pattern have also been described. (4,5) Although the imaging can help in diagnosing the disease but the lung biopsy is the best for definitive diagnosis.(4) Measurement of surfactant protein A (SP-A) and surfactant protein D (SP-D) can be considered as an alternative in monitoring disease activity.(4,5) Differential considerations include sarcoidosis, pneumoconiosis, pulmonary hemosiderosis, amyloidosis, miliary tuberculosis, and metastatic pulmonary calcifications associated with chronic renal failure.(6) There is no effective treatment for PAM. Use of disodium etidronate in the treatment of PAM is associated with mixed results. Lung transplantation is the only hope for end-stage PAM. (4)

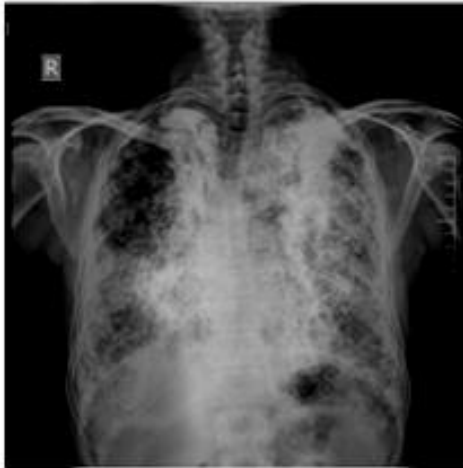


Fig-1

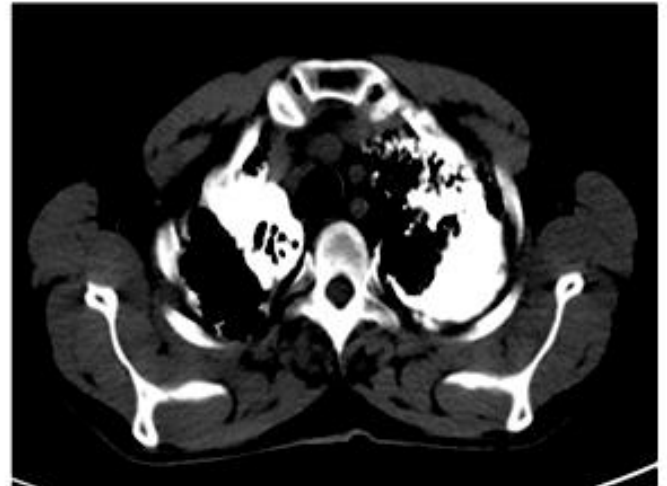


Fig-2

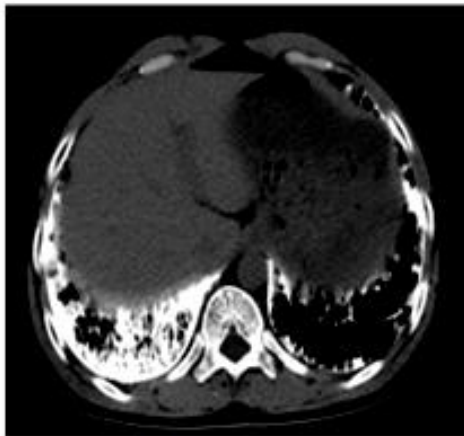


Fig-3



Fig-4



Fig-5

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RESEARCH ARTICLE

Stock market prediction using Neural Networks and sentiment analysis of News Articles.

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Key words:-

Stock Market, Text Mining, Sentiment Analysis, Artificial neural network, Stock Market Prediction

Abstract

Stock market prediction is considered as one of the classic problems of time series prediction due to high volatility of the financial market. Stock prices tend to be dependent on various factors like historical values of the company stock, P/E ratio of the company, news articles related to the company and public opinion which includes faith in the company and rumours related to the activities of the company. The system tries to predict the opening price of stock market for few particular companies based on the company's past performance and information available from various news sources.

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Introduction:-

A stock is a type of security that represents a claim on part of the corporation's assets and earning and thus signifies ownership [1]. Companies raise funds by allowing customers to buy the company's shares at an agreed price. Stock Market thus, in turn, affects the economy of a country. Buying and selling (the right) stocks lead to profits. Act of trying to determine the future value of company stock traded on an exchange is Stock Market Prediction. Stock markets have gained speed since the use of internet increased. Trading market analysis aims to predict the market fluctuations and the rise or fall of market in future.

It involves an assumption that the fundamental information that is publicly available in the past is somehow predictively related to the future stock returns. The objective of this study is to extract fundamental information from relevant news sources and use them in analysing or forecasting the stock market from any common investor's viewpoint. Researchers have developed a lot of interest in this area due to its dynamic and unpredictable nature. Since stock prices are dependent on various parameters there exists no clean equation to determine the movement of the stock price.

Predicting stock market is difficult, since relationship between stock prices and the financial economic variables is not linear. Therefore, nonlinear models could produce more reliable and correct predictions of share prices. Machine learning techniques like Artificial neural networks (ANN) has ability to map nonlinear nature and hence can be used much effectively. This paper investigates systematic usage of neural network to predict stock market prices in future.

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Literature survey:-

A lot of work had been done previously in the field of stock market prediction. Stock market prices are mostly dependent on the principle of demand-and-supply. Demand of a particular company share varies depending on various factors mentioned below:

1. Historical Prices of the company share which includes the opening prices, closing prices, highest prices and lowest prices for a time span varying from a month to an year.[3]
2. Financial status of the company, Dividends and balance sheets, Volatility of the stock[5]
3. Overseas markets that close around the same time when the concerned stock market opens up.[2]
4. Prices of oil, interest rates, inflation, GDP[4]
5. P/E ratio, general index, Volume traded[5]
6. Tweets made by people regarding the company
7. News articles related to the company.[8]

Various methodologies were used to predict the values of the stock. Decision trees were used in-order to determine whether the market will rise or fall. Linear regression approach was used to predict the trading volume of the stocks. Use of Support vector machines has also been done for predicting the rates of NASDAQ market.[2] But, a lot of previous work has proved that use of Artificial neural network is the best choice for predicting the stock markets because:

1. Ability to implicitly detect complex nonlinear relationships between dependent and independent variables.[4][5]
2. ANN is nonlinear model that is easy to use and understand compared to statistical methods.

Background: stock market prediction:-

There are few factors that help to analyse and predict its movement. These forces come under three categories:

1. Fundamental Factors
2. Technical Factors
3. Market Sentiments

Fundamental Factors:-

Stock prices would be determined primarily in an efficient market by these fundamentals, which, at some point, refer to a combination of two things:[5]

1. Earnings base (earnings per share (EPS))
2. Valuation multiple (a P/E ratio).

The level of earning base, the expected growth in the earning base, the discount rate, which is itself a function of inflation and the perceived risk of the stock are the key fundamental factors. EPS means the profit that a particular company has made per share and that too on the last quarter.

An owner of a common stock has a claim on earnings, and earnings per share (EPS) are the owner's return on his or her investment. Whenever a stock is been purchased, a proportional share of an entire future stream of earnings is purchased. That's the reason for the valuation multiple: it is the price one is willing to pay for future earnings stream. Part of these earnings might be distributed as dividends, while the remainder would be retained by the company for purpose of reinvestment.

Technical Factors:-

There are certain technical factors that helps to predict the bumpy ride of stock market, some them are Expected moving average (EMA), Rate of change indicator, Moving average (MA), Relative strength index (RSI), On Balance Volume etc.

Market Sentiment:-

It refers to the psychology of market participants, individually and collectively. Market sentiment is often subjective, biased and obstinate that helps to make a judgement about a stock's future growth prospects.

The psychology of market participants is hugely dependent on the news and hence, news is another factor that affects the share price. When there is good news about a particular stock or a company which might result in a good future of the company and increase in profits, people try to invest all their money in that particular company. This leads to increase in the interest of buying the stock whereas there exists a contrary situation too, where news could bring a negative effect where it could ruin the prospect of the particular stock.

Thus, investors depend on various different factors. Short term traders usually prioritize the technical factors of trading and hence incorporate them. While the long-term investors recognize that technical factors play a major role they also prioritize the fundamentals of trading. At the most fundamental level, supply and demand in the market determines stock price. Price times the number of shares outstanding (market capitalization) is the value of a company. It's meaningless to compare just the share values of two companies. Theoretically what affects the investor's valuation of a company is Earning, but along with this there are certain other indicators also that can be used to predict the stock hence it is investor's sentiments, attitude and expectations play an equally important role in determining the prices of the stock market

Trading analysis can be of following types-

1. Fundamental analysis:-

It mainly focuses on statistical data of the company. The goal of fundamental analysis is to find a stock's intrinsic value. It basically means what a stock is really worth as opposed to the value at which it is being traded in the market. Fundamental analysis may involve examination of financial data, management, business concept and competition. It also includes quarterly balance sheets, daily news and rumours related to the company, return on investments etc. It finds intrinsic value of a stock and if the current value of stock is below intrinsic value, it generates a buy signal and is helpful for long term trading.

2. Technical analysis:-

The Fundamental analysts examine earnings, dividends, assets, quality, ratio, new products, research and the like[11]. Technicians employ many methods, tools and techniques as well, one of which is the use of charts. Using charts, technical analysts seek to identify price patterns and market trends in financial markets and attempt to exploit those patterns.

Technicians using charts search for archetypal price chart patterns, such as the well-known head and shoulders or double top/bottom reversal patterns, study technical indicators, moving averages, and look for forms such as lines of support, resistance, channels, and more obscure formations such as flags, pennants, balance days and cup and handle patterns.[9]

Technical analysis is done considering various market indicators, some of which include price transformations, up and down volume, all time high and low etc. This factor helps to know whether the asset is trending and also its direction of growth in near days. Technical analysis also considers relationships between price/volume indices and market indicators such as moving average, relative strength, and price/volume index.

Goals:-

The system first analyses the past prices of the stock market and news related to the company to predict the opening value of that particular stock. The information from news is scraped from a variety of web sources, the open source selenium web driver is a good choice for accomplishing this task. The system scrapes the financial information like news articles, prices in history, volume of stocks sold etc. The scraped data is then processed to obtain only the data that is useful to us. The system then tries to combine sentiment analysis on this processed data along with fundamental and technical analysis.. This data is then passed through "neural network" in order to forecast the opening value of a particular stock. Output will be the approximate Prediction of highest value or the lowest value for a particular share for the given day.

The system's input consists of the news, public opinion (for rumours and people's faith in the company), volume traded, and public shareholding, stability of the share and various technical parameters of the stock market trading system. Stability of the stock's price of a particular company plays a major role in predicting the future value of a share. It depicts the performance of the share over past time and how the share has recovered from market falls. Thus it provides a measure of how good the stock is likely to be in its present position.

Identified analysis:-

Following are the modules in the system:

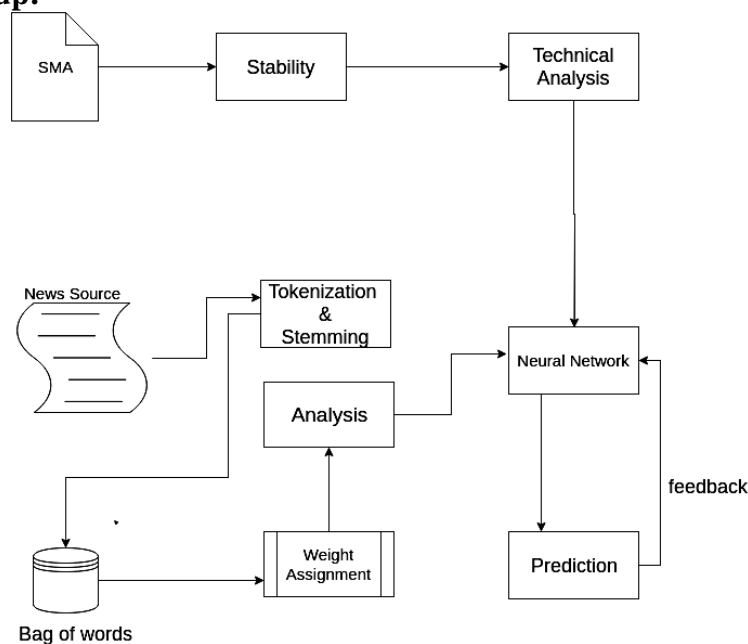
1. Scraper:
 - a. There are three scrapers in the system as follows:
 - i. First, the scraper that scrapes the values of the company stock like:
 1. Highest price
 2. Lowest price
 3. Opening price
 4. Closing price.

The prices scraped are over an interval of one-month
 - ii. Second, the scraper that scrapes the news articles related to a particular company.
 - iii. Third, the scraper that extracts tweets from the twitter related to a particular company.
2. Sentiment analyser
 - a. News related to a particular company are passed through the analyser which vectorizes the news articles and also the tweets.
 - b. The overall sentiment of the news articles and tweets is analysed using bag of words.
 - c. This is done using 'gensim' library of python which help in the Doc2Vec conversion.
3. Neural Network:
 - a. The results from the scraper and the sentiment analyser are passed as input the neural network.
 - b. Expected results are also fed to the network so as to train it.
 - c. Error values are back-propagated to update the weights.
 - d. 'Tensorflow' and 'keras' modules of python are used to implement the neural network

Information gathering:-

Scraper extracts the news articles regarding the recent company activities and the analyser shows how it will affect its share value. It extracts highest value and the lowest value of a share from the given company so as to get its stability that will help in predicting its future value. The news information will be scrapped from a collection of financial websites. BeautifulSoup[10] is a Python library which is used for scraping the live data and it converts incoming documents to Unicode and outgoing documents to UTF-8. Along with beautiful soup the open source Selenium web driver is used so as to continuously fetch the information as it is visible to a visitor on the webpage. The scrapping would be constrained only to gather information about a particular company or a set of companies.

The process of sentimental analysis comes into picture that will show how the collected news affects the sentiments of people which in turn affects the trading of the stock. We also consider Sentiment analysis on twitter feeds which is performed on the tweets extracted from the Twitter feed. This is done using 'tweepy' library. The system then uses a set of technical analysis methods, variables such as public shareholding which means equity shares of the company held by public. Such Parameters aid in the technical analysis of the trading market so as to minimize the market risk. This parameter along with various other parameters like all-time high, all time low, average returns in the form of numerical value will be input for an artificial neural network.

Experimental setup:-

The basic experimental setup of the system is as follows:

1. Scrape the news information, tweets related to the company and other stock related information from the web.
 - a. The news articles are scraped from reliable news sources like reuterindia.com
 - b. Historical prices of the company shares along with its volatility is scraped from nseindia.com
 - c. Tweets related to the company are extracted from twitter using 'tweepy' library
2. The news articles are passed through the sentiment analyser which returns the overall sentiment of the news. The same process is performed on the tweets to quantify the public opinion about the company
3. The values are passed to the neural network where number of nodes, activation function and number of hidden nodes are decided based on the accuracy of the system. After a right amount of training, the neural network is passed test values to check for its accuracy.

Summary:-

The system tries to predict the opening price of the share of a particular company by analysing news related to the company for the next trading day. The system also considers the stability of the stock price of the particular company. Sentimental analysis is used for vectorizing the news, along with technical and fundamental analysis. Artificial neural networks are used as they hold good for pattern recognition, classification and forecasting of the market so as to model nonlinear process without any prior assumptions. The nature of this process helps in obtaining accurate results.

Various stakeholders attempt to have such a system as a strategy that aims towards solving the financial situations as well as improving the efficiency and efficacy of financial risk management. Such proposed model can be a helpful tool for the investors for taking the right decision in regards to their stocks in order to extract any predictive information from that given data. The system will also display a graph of "Stock price" vs. "Date" in which the predicted values and real values in the past are plotted.

Hence it can be concluded that hybridized parameters like combination of technical and fundamental variable gives better prediction accuracy over application of standalone parameters and news being a vital factor that affects investor sentiments can be used to predict the drive of the market.

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RESEARCH ARTICLE

Exploitation of Rib Cartilage in Septorhinoplasty in Primary and Revision Cases at king Abdullah Medical City (KAMC).

Alhareth Adnan Baarimah, Mohammed Saad Aly, Omar Abdullah Qudos and Redwan Anwar Khan.

Manuscript Info

Manuscript History

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Abstract

Background: Rhinoplasty is one of the most popular cosmetic procedures in the developed countries with Plethora of techniques available to successfully complete the Rhinoplasty. Rib cartilage graft is used as an alternative to septum cartilage, with the advantage of providing sufficient amount of cartilage even for large or revision Rhinoplasty.

In this study, we will use rib cartilage graft in primary and revision Rhinoplasty cases. The rib cartilage graft will be obtained from different anatomical sites, depending on patient gender while using different techniques. The patient nasal angles and lines will be measured before and after the operation with recording the patient satisfaction in functional and cosmetic manners.

Methodology: Reconstruction Rhinoplasty cases performed from 2012 to 2016 were chosen from the King Abdullah Medical Center KAMC in Saudi Arabia. A total of 18 patients (10 females and 8 male) were selected from ENT clinic, with the age above 18 years and all of which must presented with nasal deformity and nasal obstruction. Both primary and revision Rhinoplasty cases were constituted. Patients presented with normal nose shape had been excluded from research along with the cosmetic cases. Rib cartilages were used in all patients as the graft of choice for the Rhinoplasty procedure and different technique were used as well.

Results: A total of 18 cases; 8 (44.4%) male patients, 10 (55.6%) female patients, most of them are Saudis (88.9%). They are presented with different nose presentation, majority of them had dorsal irregularity (77.8%) and deviated nose (66.7%), and they are complaining of poor breathing and Aesthetic look.

Regarding the nasal angles (Nasofrontal, Nasolipial angle, and Tip deviation) have been measured pre and post-operative as well. And for all the angles, their P Values are significant statically.

Pre-op Nasofrontal angle, Mean \pm SD (144.4 \pm 7.9). Post-op Nasofrontal angle, Mean \pm SD (135 \pm 7.8). P value is < 0.001 which is significant. Pre-op Nasolipial angle, Mean \pm SD (86.4 \pm 9.5). Post-op Nasolipial angle, Mean \pm SD (97.9 \pm 7.7). P value is < 0.001 which is significant. Pre-op tip deviation, Mean \pm SD (1.9 \pm 2.3). Post-op tip deviation, Mean \pm SD (0.47 \pm 0.73).

Conclusion: We conclude that autogenous rib cartilage grafts are an outstanding material in septorhinoplasty when structural, functional, and aesthetic problems are present.

Autogenous rib cartilage grafts are the gold standard for nasal reconstruction in patients with cartilage depletion and when large amount of tissue are needed.

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Introduction:-

Rhinoplasty is one of the most popular cosmetic procedures in the developed countries with Plethora of techniques available to successfully complete the Rhinoplasty. However, nowadays autografts is preferred, mainly due to less complication when compared to other types of grafts.

Cartilage grafts usually obtained from conceal, septum and costal cartilage. The size of the required graft and the type of the procedure (either primary or revision Rhinoplasty) usually determine the site of where the graft is taken. In small graft, the septum cartilage is considered the graft of choice. But in large reconstruction or revision procedures, the use of septum cartilage is limited because it is either too small or it because it damaged (1,2,6).

Rib cartilage graft is used as an alternative to septum cartilage, with the advantage of providing sufficient amount of cartilage even for large or revision Rhinoplasty. complains about the rib graft has been introduced and different techniques have been suggested to improve rib graft(2-4).

Autologous or homologous tissues grafts can be used in Septorhinoplasty. Comparing different types of graft, autologous tissue graft has the most advantages than the other types. Autologous graft can be obtained from septum, rib, or auricular concha [2].

The 5th or 9th rib cartilage is the graft of choice in female or male respectively in dorsal augmentation and reconstructive support in general. Specially in secondary Rhinoplasty because there is not sufficient septal cartilage [1,2].

Numbers of Factors make Secondary Rhinoplasty more difficult than the primary operation; different surgeons, sufficient septal cartilage, thickening of the skin, etc. Rhinoplasty considered one of the most challenging surgery, and it has a revision rate of 21% [3].

In this study, we will use rib cartilage graft in primary and revision Rhinoplasty cases. The rib cartilage graft will be obtained from different anatomical sites, depending on patient gender while using different techniques. The patient nasal angles and lines will be measured before and after the operation with recording the patient satisfaction in functional and cosmetic manners.

Methodology:-

The cases presented from January 2011 to December 2016 were chosen from the ENT department at King Abdullah Medical City, Makkah, Saudi Arabia. A total of 18 patients fro 250 septorhinoplasty cases (10 females and 8 male) were selected from ENT clinic retrospectively with consent obtained before starting ,and IRB approval was tekened .

Inclusion criteria:-

The age above 18 years, and presented with nasal deformity and obstruction. Both primary and revision Septo-Rhinoplasty cases were included.

All patients have been examined before and after the operation, and all the complains were documented as well. The measurements of the Nasal lines and angles were obtained from patients pictures. The patient's satisfactions from breathing and aesthetic point of view pre and post operative estimated by using a scale from 1 to 5 (1=not satisfied) and (5 =completely satisfied).

Post operatively the patients observed for complication and recorded if existed, then the follow up continued until one year at least after the procedure and all the measurements and data were repeated again.

Costal cartilage is the graft of choice when there is no preserved septal or conchal cartilage in the Septo-Rhinoplasty in both revision and primary severely depressed and saddled nose. Costal cartilage harvested from the 6th rib in male patients while in female patients was taken from the 5th rib just above the inframammary fold to obscure the appearance of the scar.

Two techniques have been applied to the harvested costal cartilage:

1st is the core of the rib was used as a spreader and strong strut graft and the outer part of the cartilage is diced and wrapped with either rectus fascia or temporalis fascia (16 cases),

2nd technique slanting incision was done to form straight slices which used as spreader and strut graft and the remnant was diced and wrapped in rectus fascia (2 cases).

Diced Rib Cartilage Graft Wrapped in Rectus Fascia:-

After the cartilage extracted from the rib, the surgeon isolate and separate the graft from any remnant fat or muscles, then the outer part of the rib cartilage was taken and diced into small cubes (0.7mm-1mm). In the same time, the rectus muscle fascia is taken and compressed to be more thinner, Then the diced cartilage was wrapped by the rectus fascia. This combination of the diced cartilage and fascia gives the graft its unique characteristics; Strong, smooth, camouflage effect, long standing, and the natural look.

Remnants of the sliced rib cartilage graft wrapped in rectus fascia:-

This technique is similar to the diced cartilage in rectus fascia

Dr. Mohamed Saad- notice that by slicing the cartilage, it will lose its strength, and it is weaker than the core of the cartilage when used as spreader and strut graft.

Results:-

In this study 18 cases have been included, 8 (44.4%) male patients and 10 (55.6%) female patients. Most of them are Saudi (88.9%).

They are presented with different nose presentation, majority of them had dorsal irregularity (77.8%) and deviated nose (66.7%). Other presentation includes saddle nose, radix depression.

Pre and post-operative nasal lines have been measured, Intra pupillary line has been fixed at 30mm, and then all other nasal lines (Basal, Dorsal, and Interdomal line) have been measured pre and post-operative, and they all show a significant P values.

Pre-operative basal line, Mean \pm SD (12.6 \pm 2.7). And post-operative measurements are Mean \pm SD (11.4 \pm 2.7), the p value is 0.004 which is statically significant. Pre-op Dorsal line, Mean \pm SD (5.8 \pm 1.4). And Post-op Dorsal line, Mean \pm SD (5 \pm 1.2), p value is 0.001 which is statically significant. Pre-op interdomal line, Mean \pm SD (7.8 \pm 1.4). Post-op interdomal line, Mean \pm SD (7.1 \pm 1.3), the P value is 0.009 which is statically significant. table 4.

Regarding the nasal angles (Nasofrontal, Nasolipial angle, and Tip deviation) have been measured pre and post-operative as well. And for all the angles, their P Values are significant statically.

Pre-op Nasofrontal angle, Mean \pm SD (144.4 \pm 7.9). Post-op Nasofrontal angle, Mean \pm SD (135 \pm 7.8). P value is < 0.001 which is significant. Pre-op Nasolipial angle, Mean \pm SD (86.4 \pm 9.5). Post-op Nasolipial angle, Mean \pm SD (97.9 \pm 7.7). P value is < 0.001 which is significant. Pre-op tip deviation, Mean \pm SD (1.9 \pm 2.3). Post-op tip deviation, Mean \pm SD (0.47 \pm 0.73).

table 6.

The patients satisfactions regarding breathing and Aesthetic look have been included in the study. There were dramatically improvement in both breathing and cosmetic appearance. Breathing and Aesthetic P values are less than 0.001 which are statically significant.

Table 5

None of the patients had Diabetes, Hypertension, or other co-morbidities.

All patient removed their sutures after 7 days of surgery, and they all received oral antibiotics and analgesics after surgery. The follow-up of all 18 cases have been done 12 months post-operative.

The rib cartilage used as a graft of choice. Two techniques were used; cucumber and take core techniques. 16 (88.9%) cases have been done with take core technique, 2 (11.1%) cases with cucumber technique.

Table 1:- Demographic and pre-operative presentation

	Number	Percentage
Nationality :		
Saudi	16	88.9
Non-Saudi	2	11.1
Total	18	100
Gender :		
Male	8	44.4
Female	10	55.6
Total	18	100
Smokers	1	5.6
From Makkah	17	94.4
Hyposomnia	10	55.6
Headache	9	50
Sinusitis	4	22.2

Table 2:- Nose presentation

	Number	Percentage
Radix depression	5	27.8
Saddle nose	9	50
Dorsal irregularity	14	77.8
Deviated nose	12	66.7

Table 3:- post-operative complications

	Numbers	Percentage
Dorsal irregularity	2	11.1
Seroma	0	16.7
Scars	4	22.2
Septal perforation	1	5.6

Non of the 18 cases had other complications; infection, bleeding, nasal obstruction, ...etc

Table 4:- pre and post operative nasal lines measurements

	Basal line		Dorsal line		Interdomal line	
	Pre	Post	Pre	Post	Pre	Post
Mean \pm	12.6 \pm	11.4 \pm 2.7	5.8 \pm 1.4	5 \pm 1.2	7.8 \pm 1.4	7.1 \pm 1.3
P value	0.004		0.001		0.009	

Table 5:- satisfaction

	Breathing		Aesthetic	
	Pre	Post	Pre	Post
Mean \pm	2.3 \pm 0.8	4.9 \pm 0.5	1.8 \pm 0.7	4.7 \pm 0.5
P value	< 0.001		<0.001	

Table 6:- pre and post operative angles measurements

	Nasofrontal angel		Nasolipial angel		Tip deviation	
	Pre	Post	Pre	Post	Pre	Post
Mean \pm SD	144.4 \pm 7.9	135 \pm 7.8	86.4 \pm 9.5	97.9 \pm 7.7	1.9 \pm 2.3	0.47 \pm 0.73
P value	<0.001		<0.001			

Discussion:-

The main goal of septorhinoplasty is to achieve the optimal functional and aesthetic results, regarding the patient and surgeon prevalence. Autogenous costal cartilage graft is the graft of choice in revision septorhinoplasty [1,2]. costal cartilage graft provide the adequate amount of tissue needed to reconstruct the defected cartilages, and has low rate of infection[2].Some complications have been noted, and to avoid those complications multiple techniques are used[5]. Knowing which method is better is mandatory as the use of costal cartilage graft become the standard of care in revision septorhinoplasty [2,5].The costal cartilage graft has been used in deferent techniques. 16 out of 18 cases have been done by taking the core of the cartilage graft, then diced and wrapped in rectus (8 cases), and temporalis (7 cases) fascia. The other two cases have been done by taking the costal cartilage in a slices technique,this used for spread and strut graft, and then the remnants diced and wrapped in rectus fascia. One case has been done using the core of the outer part of the cartilage alone, and this has the lowest results of them all. The best technique was the diced core in rectus fascia, it is the strongest,has smooth contour, and doesn't develop the wrapping effect. In the other hand, slices technique is much weaker.

the result depends on three main factors, patient satisfaction, nasal measurements (angels and lines), and post operative complication.

Patient satisfactions in function (breathing) and aesthetic look were taken pre and one year post operative. All the patient are satisfied with the result.

Regarding the nasal measurements, the goal was to reduce theNasofrontal angel to the range between 115-135 degree. And to increase the nasolapial angel to the range between 90-95 in males, and 95-105 in females. Both angels have significant P value.

Non of the patients developed significant post operative complication, except slices technique, we did for him augmentation of the lower lateral cartilage with chonchal cartilage graft.

Advantages of our research:-

18 cases were obtained from jan2011 to 2016 from KAMC center with an inclusion criteria involved only revision septorhinoplasty. All patients complain of function and structural defect, Cosmetic cases were excluded from the research. Adult patient were only selected with 10 female and 8 male patients. Rib cartilage graft used in all patients with different techniques involved: Core cartilage, slices and diced cartilage wrapped in either temporal or rectus abdominal fascia. Objective and subjective measurement were done for the patients before and after operation with follow up duration post operatively for one year and complication documented if present.

The difficulties we faced in our research which include cases were obtained from KAMC only and that reflected the low number of cases. Some of the techniques were added lately in the research, which caused unequal distribution of cases per technique. In future research we will include multiple centers to increase the number of data involved to get more accurate results.

Conclusion:-

We conclude that autogenous rib cartilage grafts are an outstanding material in septorhinoplasty when structural, functional, and aesthetic problems are present.

Autogenous rib cartilage grafts are the gold standard for nasal reconstruction in patients with cartilage depletion and when large amount of tissue are needed.

Autogenous costal cartilage grafting offers strength for nasal support to replace or augment missing tissue.



RESEARCH ARTICLE

Maternal Intake of Folic Acid Supplementation and Non- syndromic Orofacial Clefts Risk: Systematic Review and Meta Analysis.

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Abstract

Background: Orofacial clefts are common congenital birth defects that are influenced by genetic and environmental factors. Affected babies and their families suffer from burdensome consequences emotionally, socially and economically. The association of orofacial clefts with maternal intake of folic acid still unclear.

Objective: This study was conducted to collectively examine evidences released regarding the possible protective effect of maternal use of folic acid against orofacial clefts of any type.

Methods: We conducted a meta analysis and searched bibliographic databases for potential prospective and retrospective studies that report the association between folic acid and orofacial clefts. Our search was limited to human studies published in English in the last five years. Results revealed by included studies were combined using the random effect model. Q test and I^2 statistics were used to examine heterogeneity and further investigation made by subgroup analysis and meta regression. The publication bias assessed visually by funnel plot and statistically by Begg's test and Egger's test.

Results: The combined odds ratio revealed by analyzing 6 case-control studies was 0.387 (95% CI: 0.263, 0.567; $p < 0.001$). Significant heterogeneity between studies was noted ($Q = 12.4$, $p = .003$, $I^2 = 59.7$).

Conclusions: Our meta analysis supported the hypothesis of the preventive role of folic acid to orofacial clefts but more attention has to be paid for examining this association using prospective and interventional study designs.

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Introduction:-

Orofacial clefts are considered as common congenital birth defects with a prevalence of 1.47 per 1000 live births globally [1]. Structurally, they can be categorized as either isolated, in forms of isolated cleft lip (CL) and isolated cleft palate (CP), or combined cleft lip and plate (CLP) [2]. As it needs a multidisciplinary approach with several steps, the treatment process could be intolerable and burdensome [3].

Folic acid, also known as vitamin B₉, is an essential amino acid that must be supplemented exogenously to humans since they can't synthesize it de novo [4]. It has a critical role in cell division, DNA synthesis, methylation and repair, particularly in infancy and pregnancy periods that has rapid growth and multiple cell divisions [5].

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It is thought that non-syndromic orofacial clefts (NSCLP) are developed under the influence of genetic and environmental factors' interactions [6]. In the light of the fact that the neural tube is the embryological origin of facial and oral tissues via its neural crest cells in which folate receptors are expressed extremely high [7] and since maternal intake of folic acid supplementation during the preconception period had been demonstrated by experimental studies' findings to reduce the risk of neural tube birth defects (NTDs) [8], many studies were conducted hypothesizing that similar effect of folic acid might exist for reduction of orofacial clefts risk. In addition, some evidences showed that epileptic mothers who are taking anti-epileptic drugs that interfere with folate metabolism are more prone to have babies with orofacial defects [9].

Conflicting results came out from the observational studies examining the potential protective role of folic acid's intake against orofacial clefts in human. These mixed evidences might be explained by the difficulty in investigating the folic acid role in reducing the risk of orofacial clefts independent from the effects of other multivitamins supplementations taken by mothers in the same time [10]. Other factors that might contribute to this contradiction include variations in study designs, sample sizes, populations and different analytical approaches for potential confounders [8].

In 2006, the World Health Organization recommended Mandatory wheat flour fortification with folic acid programs to be implemented as a national law [11]. Reduction of orofacial clefts risk following mandatory fortification of food was observed by several studies. This inverse association was statistically significant in some of these researches [12,13,14] and insignificant in others [15,16,17].

Since the evidences still controversial and not conclusive, we conducted this meta analysis to combine the results of different studies on the possible protective role of maternal use of folic acid supplementation during the period of preconception and the first trimester against the risk of orofacial clefts of any type.

Methodology:-

Search Strategy:-

We conducted our systematic review with the guidance of the Preferred Reporting Items for Systematic Reviews and Meta-Analyses Statement [18] as relevant and required. Our search started in 15th of December 2015 using bibliographic databases including the PUBMED interface to MEDLINE, SCOPUS and COCHRANE databases to identify potentially eligible citations by using the mesh terms for [folic acid] and [clefts]. The search was limited to human studies published in English in the last 5 years. We communicated with experts in the field of Obstetrics and Gynecology as well as the field of Pediatrics Surgery for further identifications of potential eligible studies. Our search was repeated every two weeks and the electronic automated PUBMED alert system was used monthly to identify any newly published studies up to the end of February 2016. The reference lists of all included studies as well as those of relevant systematic review articles were scanned to enhance our search deductibility for further potentially eligible articles.

Inclusion and Exclusion Criteria:-

Studies included in our systematic review had to meet the following inclusion criteria: 1- is designed as a randomized controlled trial, cohort, case-control, or cross-sectional study. 2- Provide sufficient data for estimations on the association between maternal folic acid intake in preconception period and first trimester and non syndromic orofacial clefts of any type. Studies that reported the folic acid intake to general population and studies that included syndromic orofacial clefts were excluded.

Data extraction:-

The following elements were extracted from each study: citation, study design, study place, study time, the amount of folic acid supplemented, folic acid supplementation timing in relation to conception and antenatal period, sample size, crude and adjusted estimations with 95% CI, p-values and standard errors. After the four investigators extracted data from each study independently, they cross-checked the extracted data and met to resolve any discrepancies.

Statistical methods:-

We used Comprehensive Meta-Analysis Software version 3 with were *p* values of 0.05 or less and a confidence intervals of 95 %, considered statistically significant for all statistical analyses performed in our review. All studies included in Meta analysis were of case control study design, since there was only one eligible cohort study met our inclusion criteria. As the included studies brought out results from different populations, we used the random effect

model for combined effect estimation. The heterogeneity was tested using Q statistics calculations in which statistically significant values different from 0 indicates presence of heterogeneity. Further evaluation of heterogeneity made by I^2 statistics which evaluated the percentage of variation between studies that cannot be attributed to within-study variation. The value of $I^2 > 50\%$ was considered significant for heterogeneity. In order to explore the source of heterogeneity, we used the subgroup analysis and further investigation of heterogeneity made using meta regression for selected variables and study characteristics.

The National Heart, Lung, and Blood Institute (NHLBI) quality assessment tool was used to assess the studies qualities. This tool rates the study quality as good, fair or poor. The good quality study is considered to be of a least risk of bias and valid results, while the fair and poor studies are considered to be susceptible to some risk of bias and indicate a significant risk of bias, respectively[19]. All investigators assessed the quality of each eligible study and they met to make a consensus on their study quality assessment ratings. Only studies rated as of good quality were included in our review and meta analysis.

Graphical assessment of publication bias was made by funnel plot that assessed asymmetry visually, while Begg's test and Egger's test were chosen for statistical assessment.

Results:-

Data search results:-

Our search in electronic databases initially revealed 112 citations; in which 43 articles were duplicated. The titles and abstracts of the remaining 69 articles were scanned and the full texts of 20 of them were retrieved since they were identified as potential articles to be included in our review. After in detail review of their full texts, 13 articles were excluded for not fitting with the inclusion criteria, leaving 7 articles for final inclusion in the systematic review and meta analysis.

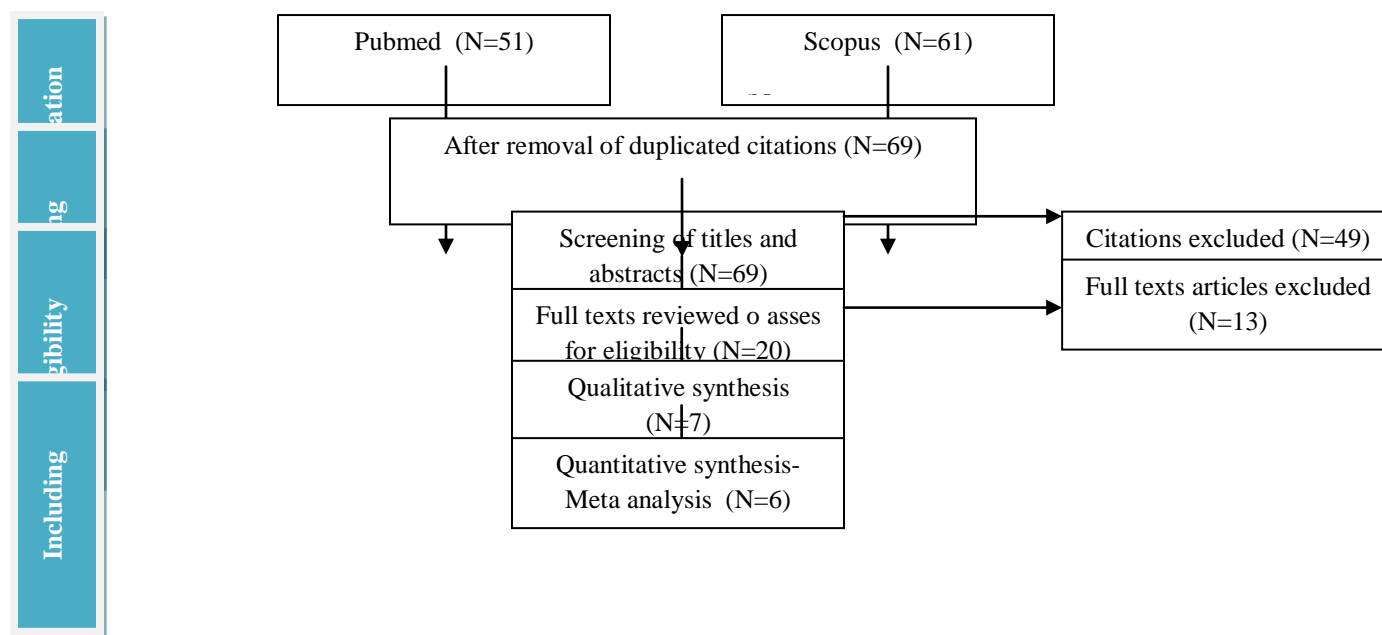


Figure1. Flow Chart for Studies Selection

Characteristics of studies included in the systematic review:-

One prospective cohort (Li Song,2012) and six retrospective case-control (Li Fang,2015; Lin,Yu, 2014; McKinney,2013; Hao,2015; Jia,2011; Figueiredo,2015) studies met the inclusion criteria. The cohort study showed a statistically significant association between maternal folic acid supplementation and orofacial clefts (aRR= 0.69; 95% confidence interval = 0.55– 0.87) suggesting that daily maternal intake of 0.4mg of folic acid, started during the period of preconception and in early pregnancy , was associated with a reduced risk of orofacial clefts in offspring born in a highly prevalent region of China [20].

Five retrospective case control studies demonstrated a statistically significant association between maternal folic acid supplementation and orofacial clefts of any types. Four of these studies (Li Fang, Xu, 2015; Lin, Yu, 2014; Hao, 2015; Jia, 2011) were conducted in China. Xu Li Fang showed that folic acid intake in first trimester confers a reduced risk of having babies with orofacial clefts (aOR= 0.23; 95% confidence interval = 0.10–0.55) [21]. Similarly, a significant relationship between folic acid supplementations during pregnancy and having offspring with orofacial clefts was established using multiple logistic regression analyses of different environmental factors in Yu Lin's study, indicating a possible preventive role of folic acid to orofacial clefts (aOR= 0.36; 95% confidence interval = 0.274–0.473) [22].

Plausible associations between preconceptional and 1st trimester use of recommended dose of folic acid and risk of orofacial clefts was evident among Chinese population as revealed by Yanru Hao (aOR= 0.43; 95% confidence interval = 0.210–0.880) [23]. Furthermore, the protective role of folic acid against orofacial clefts was emphasized in Z-L Jia's assessment of the risk of orofacial clefts and its relation to exposures to different environmental factors and nutritional supplements intake as demonstrated by multiple logistic regression analyses (aOR= 0.67; 95% confidence interval = 0.44–1.00) [24].

According to RF Figueiredo, women in Brazil who took a daily dose of 0.40 mg of folic acid in 1st trimester were significantly 0.14 times as likely to have an affected child with orofacial clefts than those who did not [25]. The relationship between micronutrients and orofacial clefts was examined by C.M. McKinney among the Thai population and women who reported taking a folic acid supplement in the preconception period were 0.51 times as likely to have an affected child than those who did not, although this finding was not statistically significant (P= 0.12) [26]. All included studies characteristics are listed in table 1.

Table 1. Studies Characteristics.

Folic Acid Use and Nonsyndromic Orofacial Clefts in China	Li, Song 2012	240,244	China	Prospective Cohort Study	RR= 0.75 95% CI= 0.61 - 0.91	RR=0.69; 95% CI = 0.55–0.87	Age.	Preconception and first trimester
Maternal malnutrition, environmental exposure during pregnancy and the risk of non-syndromic orofacial clefts	Jia, Z. L., 2011	934	China	case-control study	OR= 0.52 95% CI= 0.33–0.81	OR= 0.67, 95% CI = 0.44–1.00	Gender, vitamins intake, maternal age, maternal weight addition, passive smoking during pregnancy, calcium intake.	First trimester.
Association of Parental Environmental Exposures and Supplementations Intake with Risk of Nonsyndromic Orofacial Clefts: A Case-Control Study	Hao, Yanru 2015	979	China	case-control study	OR=0.43 95% CI= 0.21-0.88	OR=0.69 95% CI= 0.28, 1.69	Maternal illness, Cold without fever, History of skin disease, medication use, Smoking.	Preconception and first trimester

in Heilongjiang Province, China								
case-control study of environmental exposures for nonsyndromic cleft of the lip and/or palate in eastern Guangdong, China	Lin, Yu 2014	858	China	case-control study	OR=0.334 95% CI= 0.25–0.43	OR=0.36; 95% CI = 0.27–0.47	Gender, childbearing age, parental education level, family history, abortion history, common cold & drug use during pregnancy	First trimester of pregnancy.
A Case-control Study of Environmental Risk Factors for Nonsyndromic Cleft of the Lip and/or Palate in Xuzhou, China*	Li Fang, 2015	527	China	case-control study	OR = 0.18; 95% CI= 0.19-0.26	OR= 0.23; 95% CI= 0.10–0.55	family history, pesticide exposure, indoor decoration pollution, multivitamins.	First trimester of pregnancy.
The role of the folic acid to the prevention of orofacial cleft: an epidemiological study	Figueiredo, Rf, 2015	80	Brazil	case-control study	OR= 0.14 95% CI= 0.049 – 0.401	Not adjusted	No adjustment	Preconception and first trimester.
Micronutrients and oral clefts: a case-control study.	McKinney, C M, 2013	342	Thailand	case-control study	OR= 0.68 95% CI= 0.32, 1.45	OR= 0.51 95% CI= 0.22, 1.19	Diabetes, history of cold, vitamin A intake, Liver intake, Menstrual cycle regulation supplements	Preconception and first trimester

Using the random effect model, the combined data of six case-control studies yielded a combined odds ratio of 0.387 (95% CI: 0.263, 0.567; $p < 0.001$) suggesting a statically significant role of maternal intake of folic acid supplementation in reducing the risk of orofacial clefts in offspring (Figure 2).

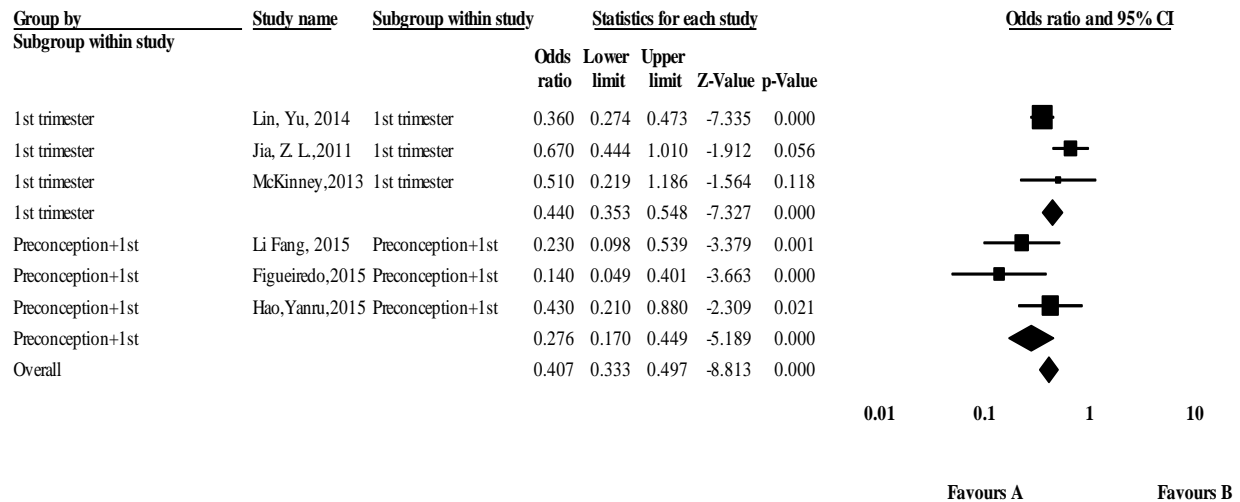


Figure 2. Meta Analysis of Case-Control Studies

The Q test and I^2 statistics showed a significant moderate heterogeneity between the analyzed studies ($Q=12.4, p=.003, I^2=59.7$). In order to estimate the effect of folic acid supplementation maternal intake timing, a subgroup analysis was performed based on the folic acid intake timing relative to conception and antenatal period. Half of the included case-control studies confined the time of maternal intake of folic acid to the first trimester (Lin, Yu, 2014; Jia, Z. L., 2011 and McKinney, 2013). Combining the odd ratios of these studies revealed a combined OR of 0.440 (95% CI: 0.353, 0.548; $p<0.001$) and the tests for heterogeneity were significant ($Q=12, I^2=59.7, P=0.03$) while the combined odd ratios of the remaining three studies that include the preconceptional intake to their analyses (Li Fang, 2015, Figueiredo, 2015 and Hao, Yanru, 2015) was 0.276 (95% CI: 0.170, 0.449; $p<0.001$) and the tests for heterogeneity were not significant ($Q=3.01, I^2=26.7, P=0.9$). Thus, the subgroup analysis showed a folic acid intake role in reducing the risk of orofacial clefts to be statistically significant in both subgroups and adding the analysis of preconceptional use of folic acid reduced the dispersion to insignificant level statistically (figure 3).

All studies (Random Effect Model)

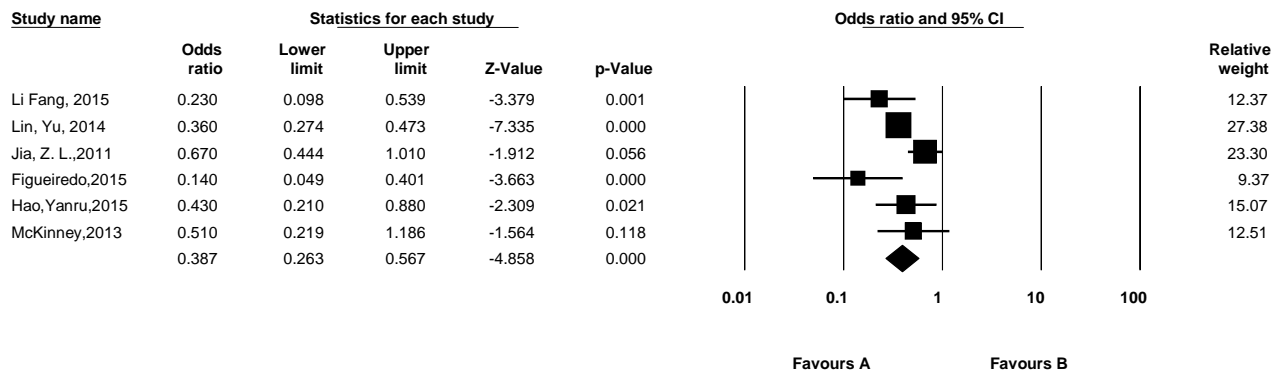


Figure 3. Subgroup Analysis Results.

Further investigation of possible sources of heterogeneity was made using the meta regression. Neither the sample size ($Q=2.1, P=0.146$) nor the study location ($Q=3.05, P=0.217$) were found to show statistically significant relation to heterogeneity (figure 4)

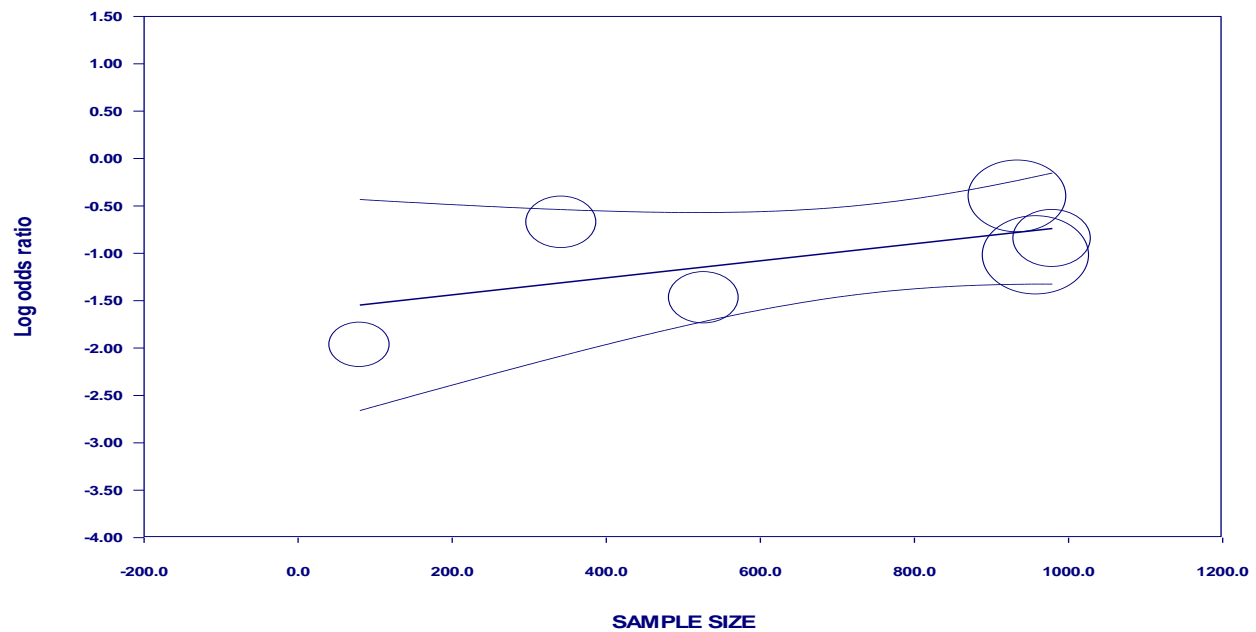


Figure 4. Regression of Log Odds Ratios on Sample SIZE

By using the funnel plots, the visual check for publication bias did not appear to be asymmetrical, and the Begg's test for publication bias was not significant ($p=0.18$). Moreover, Egger's Test, that has the better statistical power to discover publication bias, was also not significant ($p=0.6$).

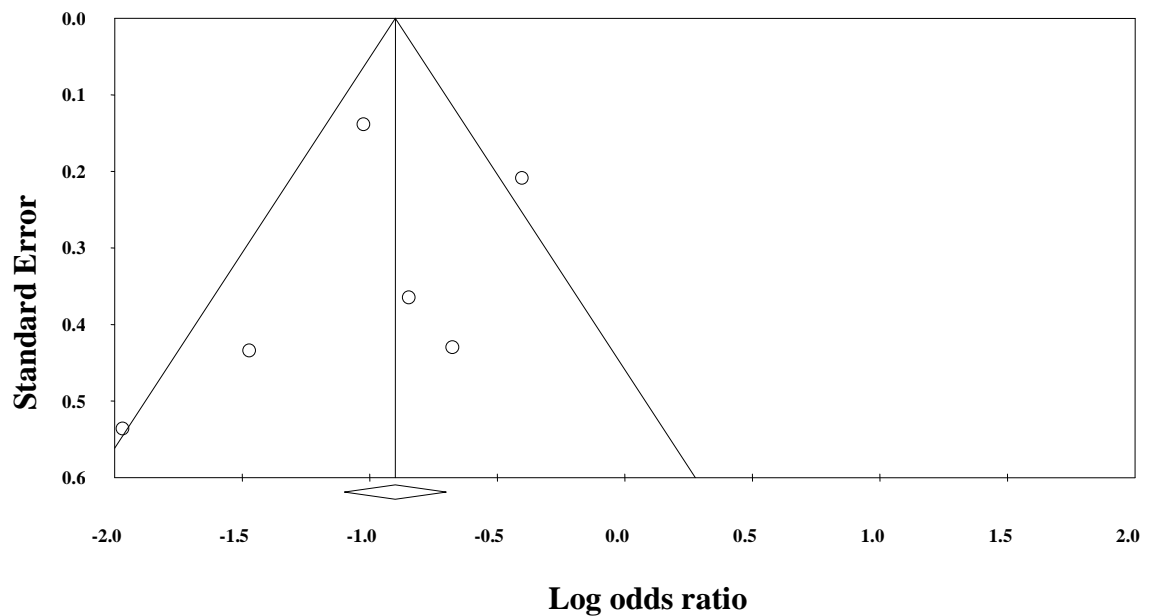


Figure 5. Funnel Plot of standard Error by Log Odds Ratios

Discussion:-**Outcome:-**

Our systematic review and meta analysis advocated the hypothesis of the protective role of maternal intake of folic acid's supplements against orofacial clefts in offspring. The point estimates yielded from the meta analysis of included case-control studies revealed that folic acid intake by mothers during the preconception period and first trimester reduced the risk of orofacial clefts in their offspring by 38,7 %.

A Similar significant effect was reflected by combining estimations using a subgroup analysis of the studies restricted the time of maternal intake to the first trimester only and those studies that included the use in the preconception period. Mothers who took the folic acid supplements in 1st trimester only were 44% less likely to have babies with orofacial clefts than mothers who did not. Similar effect was shown in the preconception period and 1st trimester subgroup, but with less effect size.

A previous meta analysis showed results that indicate an inverse relationship between folic acid and orofacial clefts of any type, but yielded a positive relationship for cleft palate among the prospective studies [27]. Another meta analysis among the populations in the United States and Canada concluded that the evidence of folic acid association with orofacial clefts is not strong, however, it suggested a possible beneficial effect of the use of multivitamins supplements in early pregnancy in preventing the occurrence of orofacial clefts and have shown that a reduction of about 7% of cleft lip and palate [28].

The results of a recent individual participant data (IPD) meta-analysis for the studies that examined the interaction between genetic and environmental etiological factors, focusing mainly on the folic acid and MTHFR C677T genotype interaction, showed reduced risk of cleft lip and palate with maternal folic acid use ($p=0.008$; OR=0.70, 95% CI: 0.65–0.94) and with supplements containing folic acid ($p=0.028$, OR=0.80, 95% CI: 0.65–0.94) suggesting a protective influence of folic acid against orofacial clefts[29]. A meta-analysis of studies that examined the association of fortification with orofacial clefts among the populations in United States and Canada have shown a reduction of about 7% of cleft lip and palate[30].

A randomized control trial found no difference in the recurrence rates of orofacial clefts among two groups of women assigned to high dose (4mg) and low dose (0.4mg) of folic acid intake, suggesting that potential ineffectiveness of the higher dose was possibly ineffective in further reducing recurrence of orofacial clefts relative to the lower dose [30].

Limitations:-

Our results were limited by relatively few number of studies included in the meta analysis (6 studies) which might affect the statistical power for estimating the combined effect.

All included studies in our meta analysis were of retrospective case-control design making our results vulnerable to recall bias. Maternal consumptions of different vitamins and trace elements supplements that have potential contribution to the risk of orofacial clefts were difficult to be examined independently among included studies, creating a bias of non random selection of supplements use.

Another main limitation in our review was the lack of randomized clinical trials which experimentally test the folic acid role in protecting offspring against orofacial clefts. Since the protective role of folic acid supplements against neural tube defect has been well established, conducting a randomized control trial with low dose folic acid against the control of placebo will interfere with the concept of nonmaleficence, thereby, considered unethical.

The potential confounders, such as intake of other supplements, family history, age, gender, were analyzed differently among the included studies and one study did not show the results of analytical manipulation to control for potential confounding factors.

Significant moderate heterogeneity observed between included studies is another source of limitation to our findings. Aside from narrowing the time window of exposure in the first trimester, no factor was identified to explain the significant heterogeneity.

The concern of publication bias is considered in any meta analysis. The results of our analysis did not show this bias to be evident.

Conclusion:-

Our results supported the hypothesis that taking folic acid-containing supplements during pregnancy may protect against oral clefts. However, more prospective studies conduction in different populations with different ethnicities would be beneficial in expressing a causal association. Moreover, it is essential for future retrospective observational studies consider careful analysis of suspected confounders and improve the statistical power by enhancing the representativeness of the sampling techniques and sample sizes. Enhancing the Implementation of Mandatory fortification programs with close monitoring, might be beneficial in reducing the risk of orofacial clefts in offspring.

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RESEARCH ARTICLE

TO EVALUATE THE ANTIMICROBIAL EFFICACY OF SIX MOUTHWASHES AGAINST STAPHYLOCOCCUS AUREUS AND STREPTOCOCCUS MUTANS – IN INVITRO STUDY

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Key words:-

Mouthwashes, Staphylococcus aureus, Streptococcus mutans, Agar well diffusion method.

Abstract

In-vitro comparative study to evaluate the efficacy of antimicrobial activity of Thymol(A), Chlorhexidine(B), Chlorhexidine plus triclosan (C), Cetylpyridinium chloride (D), Belleric myrobalan (E), Povidone iodine (F) mouthwashes against Staphylococcus aureus and Streptococcus mutans with distilled water as a control group was undertaken. The mouthwashes were tested at four different concentrations 1:4(25%), 1:1(50%), 3:4(75%) and full strength (100%). The antimicrobial activity was determined by using agar well diffusion method. The plates were allowed to incubated at 37°C for 24hr, 48hr and 72hr. Chlorhexidine plus triclosan emerged as the most effective mouthwash against Staphylococcus aureus (30mm to 35.33mm) and Streptococcus mutans (19mm to 27.5mm) followed by Cetylpyridinium chloride, Chlorhexidine, Thymol, Belleric myrobalan. Povidone iodine and distilled water had no antimicrobial activity. It was concluded that the Chlorhexidine plus triclosan shows excellent antimicrobial activity but povidone iodine has no antimicrobial activity.

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Introduction:-

Prevention of oral diseases is easier than cure¹. Oral hygiene is an important aspect in our daily life for maintaining a healthy life style. The oral cavity contains more than 500 types of pathogenic and non-pathogenic bacteria, out of which 22 types are dominant. Improvement of oral health can influence the quality of life. Scientific research has shown that the use of mouthwashes (antimicrobial solutions) plays a vital role in the reduction of oral microbial load in the oral cavity.

Antimicrobial solutions are used for cleaning food remnants, decreasing oral malodour, prevention of plaque accumulation on tooth and gingival surfaces, used prior to and after oral surgical procedures in order to prevent oral pathogenic infections. Within the oral cavity, the most common microorganisms are staphylococcus aureus

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(formerly also called staphylococcus pyogenes) and Streptococcus mutans. Staphylococcus aureus is a gram positive, non motile, non spore forming, catalase positive cocci that occur in grape like clusters, it is a major human pathogen, responsible for a number of hospital-acquired infections, initially colonizes several locations in the human body, but the oral cavity and hands are the main reservoirs for propagation of this pathogen in the hospital environment¹. Streptococcus mutans is a gram positive, non motile, non spore forming, catalase negative, facultative anaerobic cocci arranged in chains or pairs and commonly found in the oral cavity and has a major role in tooth decay formation and sore throat. S.mutans metabolise sucrose to lactic acid causes tooth decay.

There are 25 species of oral streptococci in human oral cavity². Each of them develops specialised properties for colonisation in oral sites and constantly changes conditions to fight competing bacteria. Oral diseases can be initiated by imbalances in the microbial flora. In specific conditions, streptococci can change to opportunistic pathogen that can initiate the disease and damage the host. Oral streptococci is mentioned both as harmless and harmful bacteria.

To decrease infections in the Oral cavity different antimicrobial mouthwashes are used. So an in vitro study was under taken to know the antibacterial effectiveness of various mouthwashes such as A – Thymol, B – Chlorhexidine, C – Chlorhexidine gluconate with triclosan, D - Cetylpyridinium chloride, E - Belleric myrobalan, F - Povidone iodine on Staphylococcus aureus and Streptococcus mutans.

Aims and objectives of the study:-

The aim of this invitro study is to evaluate efficiency of antimicrobial activity of different mouth washes such as Thymol, Chlorhexidine, Chlorhexidine plus triclosan, Cetylpyridinium chloride, Belleric myrobalan, Povidone iodine mouthwashes in different concentrations against staphylococcus aureus and Streptococcus mutans, by using culture media such as Nutrient agar and Blood agar respectively, compared with distilled water as a control group.

Materials and Methods:-

Six commonly used mouthwash products (Table 1) were evaluated for their antimicrobial activity compared with a control group (distilled water) in Department of Oral and Maxillofacial Surgery, GITAM Dental College and Hospital, Visakhapatnam, Andhra Pradesh.

Microorganisms:-

The test microorganisms Staphylococcus aureus, Streptococcus mutans were subcultured on specific media such as Nutrient Agar broth and Brain Heart Infusion Agar respectively incubated aerobically at 37°C. The identification of all the microbes was confirmed by standard staining and biochemical methods.

Identification of pathogen:-

The oral specimens are plated on BHI agar, blood agar, and BHI broth for the isolation of the pathogenic strains in both aerobic and anaerobic conditions. The isolated strains were identified to the genus level by staining, microscopy, and biochemical tests. The agar slants of brain heart infusion agar are used for maintaining the identified culture. The culture slants were stored at 4°C in the refrigerator.

Staining Characteristics:-

The isolated colonies were stained by Grams staining procedure. The prepared smear was air-dried and heat fixed. Crystal violet was flooded over the smear for 1 min and drained. After washing, Gram's iodine was added and left for 1 min. The smear was then washed in 95% (v/v) ethanol for 30 sec. It was counterstained with Safranin for 2 min. After air drying, the smear was examined under oil immersion objective in a light microscope.

Biochemical Characteristics:-

The isolated strains were subcultured on BHI, blood agar and BHI broth. These Plates were incubated at 37°C overnight and cultural characteristics of the colonies were studied after incubation. Biochemical tests such as indole production test, methyl red test, citrate utilization test, catalase, indole, nitrate reduction, carbohydrate fermentation test, starch hydrolysis, urease test, hydrogen sulfide test, gelatin hydrolysis were performed.

Methods:-

Antimicrobial effectiveness of various mouthwashes was assessed by using Agar well diffusion method:

Agar well Diffusion Method:-

The mouthwashes were tested at four different concentrations: 1:4(25%), 1:1(50%), 3:4(75%) and full strength (100%), taking sterile distilled water as the diluent, using agar well diffusion method or cup plate method. In this method, pure isolate of each microbe was subcultured on the recommended specific media for each microorganism at 37°C for 24hrs. From each inoculated agar plate, a minimum of four colonies were touched with a sterile loop and transferred into a tube containing normal saline (0.85%). A 100µl volume of each mouthwash concentration (full strength, 3:4, 1:1, 1:4) and the control was propelled directly into the wells of the inoculated specific medias nutrient agar petri plates for *Staphylococcus aureus* and blood agar petri plates for *Streptococcus mutans*. The petri plates were allowed to stand for ten minutes for diffusion of the mouthwash to take place and incubated at 37°C for 24hr, 48hr and 72hr. The antimicrobial activity, indicated by an inhibition zone surrounding the well containing the mouthwash measures in mm. The inhibition zone for different mouthwashes are measured and noted.

Observations and Results:

In the present study the antimicrobial activity of the six mouthwashes was evaluated against *Staphylococcus aureus* and *Streptococcus mutans*. It was determined by agar well diffusion method, was measuring growth of inhibition zone in millimeters (mm) including the diameter of well (6mm). The inhibition zone diameters were measured accordingly with dilutions of 1:4, 1:2, 3:4 and full strength, during the respective time intervals of 24hrs, 48hrs, 72hrs

Based on the mean diameter of the zones of microbial inhibition produced by the six mouth washes in agar well diffusion method, against *Staphylococcus aureus* and *Streptococcus mutans*, the mouthwash 'C' emerged as the most effective antimicrobial activity followed by mouthwash 'D' that showed excellent level of antimicrobial activity. Following which mouthwash 'B' and 'A' showed good antimicrobial activity and finally, displaying very little antimicrobial activity was mouthwash 'E'. No antimicrobial activity against *Staphylococcus aureus* and *Streptococcus mutans* was elicited when tested with mouthwash F and distilled water.

Table 1:- Ingredients of six mouthwashes tested for antimicrobial potential.

Mouthwash	Ingredients	Active ingredients
A	Purified water, sorbitol, alcohol, poloxamer 407, benzoic acid, sodium saccharin, eucalyptol, flavour, methyl salicylate, thymol, sodium benzoate, menthol	Thymol Eucalyptol Methyl salicylate Menthol
B	Chlorhexidine gluconate 0.2% w/v in a pleasantly flavoured aqueous base.	Chlorhexidine gluconate
C	Chlorhexidine gluconate 0.2% w/v, triclosan USP 0.05% w/v, sodium monoflourophosphate USP 0.07% w/v in a pleasantly flavoured aqueous base.	Chlorhexidine gluconate Triclosan
D	Water, glycerine, propylene glycol, sorbitol, poloxamer 407, cetylpyridinium chloride, potassium sorbate, sodium fluoride, sodium saccharin, menthal, citric acid	Cetylpyridinium chloride, Sodium fluoride
E	Belleric myrobalan (Bibhitaki) 10mg, Betel (Nagavalli) 10mg, Pilu 5mg powders, Peppermint satva 0.4mg oils, Gandhayva taila 1.2mg, Ela , other ingredients Sodium benzoate IP, Bronopol IP, Potassium sorbate	Belleric myrobalan (Bibhitaki)
F	Povidone iodine IP 2% w/v, absolute alcohol content 8.38% v/v in a mint flavour aqueous base.	Povidone iodine

Table 2:- Antimicrobial activity of mouthwashes against *Staphylococcus aureus* and *Streptococcus mutans* determined by agar well diffusion method by measuring diameter of growth of inhibition zones (mm)

Dilutions Mouthwash	Staphylococcus aureus				Streptococcus mutans			
	24 hrs	48hrs	72 hrs	Average	24 hrs	48hrs	72 hrs	Average
A								
1:4(25%)	16	16	16	16	16.5	16.5	16.5	16.5
1:2 (50%)	17	17.5	17.5	17.33	17	17	17	17
3:4 (75%)	19	20	20	19.66	18.5	18.5	18.5	18.5
(100%)	21	21.5	21.5	21.33	18.5	19	19	18.83
B								
1:4(25%)	18	18.5	18.5	18.33	19	19.5	19.5	19.33
1:2 (50%)	19	19.5	19.5	19.33	21	21.5	21.5	21.33
3:4 (75%)	21	21.5	21.5	21.33	22	22.5	22.5	22.33
(100%)	24	25	25	24.66	23.5	25	25	24.5
C								
1:4(25%)	30	30.5	30.5	30.33	23	23.5	23.5	23.33
1:2 (50%)	31	31.5	31.5	31.33	25	25	25	25
3:4 (75%)	33	33.5	33.5	33.33	26	27	27.5	26.83
(100%)	35	35.5	35.5	35.33	28	29	29.5	28.33
D								
1:4(25%)	22	22.5	22.5	22.33	20	21	21	20.66
1:2 (50%)	24	24.5	24.5	24.33	23	23.5	23.5	23.33
3:4 (75%)	27	27.5	28	27.50	25	25.5	26	25.5
(100%)	29	29.5	29.5	29.33	26	26.5	27	26.5
E								
1:4(25%)	0	0	0	0	0	0	0	0
1:2 (50%)	0	0	0	0	0	0	0	0
3:4 (75%)	16	16	16	16	14	14	14	14
(100%)	18	18.5	18.5	18.33	16	16	16.5	16.17
F								
1:4(25%)	0	0	0	0	0	0	0	0
1:2 (50%)	0	0	0	0	0	0	0	0
3:4 (75%)	0	0	0	0	0	0	0	0
(100%)	0	0	0	0	0	0	0	0
Distilled water	0	0	0	0	0	0	0	0

Discussion:-

P.K. Sreenivasan .et.al³ (2013) stated that the human oral cavity and its various ecological niches (i.e. dorsal tongue, buccal mucosa, subgingival spaces, etc.) harbour substantial numbers of Gram-positive and Gram-negative bacteria. Dental plaque is one of the most thoroughly investigated types of microbial biofilms with clinical studies clearly. The oral cavity is a major gateway to the human body. The microorganisms colonizing one area of the oral cavity have a significant probability of spreading on contiguous epithelial surfaces to neighbouring sites. Microorganisms from the oral cavity have been shown to cause a number of oral infectious diseases, including caries (tooth decay), periodontitis (gum disease), endodontic (root canal) infections, alveolar osteitis (dry socket), and tonsillitis. Evidence is accumulating which links oral bacteria to a number of systemic diseases, including cardiovascular disease, stroke, and pneumonia.

In the present study determination of the antimicrobial activity of six mouthwashes, and its effectiveness of active ingredients was done. The antimicrobial activity of the mouthwashes were assessed where statistical ranking procedures were used to place the six different mouthwashes following the order of antimicrobial effectiveness. The results revealed variations in their effectiveness against the two tested microorganisms. Based on the mean diameter of the zones of microbial inhibition produced by the mouthwashes in agar well diffusion method, tested against *staphylococcus aureus* and *streptococcus mutans*, chlorhexidine gluconate with triclosan showed best results where as Cetylpyridinium chloride also emerged as a mouthwash with the excellent antimicrobial activity. Chlorhexidine

gluconate and Thymol (essential oils) showed good antimicrobial activity and finally, displaying very little antimicrobial activity was Belleric myrobalan while povidone iodine had no antimicrobial activity.

The mouthwash 'A' contains active ingredients such as four phenol-related essential oils (thymol, eucalyptol, menthol and methyl salicylate) which claim to penetrate the plaque biofilm and thus kill micro-organisms that causes gingivitis. Furgang D .et.al⁴ (2005) stated that the mouthwashes display broad spectrum antimicrobial activity, prevent bacterial aggregation, slow bacterial multiplication, retard plaque maturation and decrease plaque mass and pathogenicity.

The mouthwashes 'B' and 'C' contains active ingredients. They are Chlorhexidine gluconate is a cationic biguanide with broad spectrum antimicrobial activity. It is most effective mouthwash for reducing plaque and gingivitis. Arweiler NB .et.al⁵ (2001) stated that the Chlorhexidine has an important feature of substantivity, that helps in prolonged adherence of the antiseptic to the oral surfaces (mucosa & teeth) & its slow release at effective doses that guarantees the persistence of its microbial activity. Kamal Raj Current recommendations are for twice-daily i.e. chlorhexidine to be used only as a short-term adjunct, or as an aid in disinfection of surgical sites, to improve wound healing, or as a short-term treatment of halitosis. It is not recommended for long-term use due to its numerous adverse effects. These include tooth and restoration staining, soft tissue staining, increased calculus deposition, unpleasant taste, taste alteration, burning sensation, desquamation and mucosal irritation. Chlorhexidine may also potentiate oral discomfort in patients with chemotherapy-induced mucositis, xerostomia or ulcerative oral mucosal conditions.

The mouthwash 'C' contains active ingredients, Chlorhexidine gluconate and Triclosan. Triclosan is synthetic non-ionic chlorinated phenolic agent with antiseptic qualities. Witt J.et.al^{6,7} (2005) stated that broad-spectrum efficacy on Gram-positive and most Gram-negative bacteria. It is also effective against mycobacterium, anaerobic bacteria, the spores and fungi of the Candida species. The mechanism of its antiseptic action is by acting on the microbial cytoplasmic membrane, inducing leakage of cellular constituents and thereby causing lysis of the micro-organisms.

The mouthwash 'D' contains active ingredients, they are Cetylpyridinium chloride is a quaternary ammonium compound with antiseptic and antimicrobial properties. Narinder Pahwa .et.al⁸ (2011) stated that the cationic and binds to bacterial surfaces causing disruption of the cell membrane, leakage of intracellular components and disruption of metabolism. Mouthwashes containing cetylpyridinium chloride inhibit and reduce plaque build-up. Hadi Darvishikhezri et al³ (2013), investigated that the mouthwashes containing cetylpyridinium chloride with alcohol and cetylpyridinium chloride without alcohol were most effective antimicrobial activity against supragingival plaque microorganism.

The mouthwash 'E' contains active ingredients such as Belleric myrobalan. Rupali Mahajan .et.al⁹ (2016) stated that the antimicrobial properties, analgesic and antioxidant properties.

The mouthwash 'F' contains active ingredients such as Povidone iodine, an iodophore in which iodine is linked to povidone that displays an affinity for the cell membrane there by delivering free iodine directly to the bacterial cell surface. Adamietz I.A.et.al¹⁰ (1998) stated that the broad spectrum of activity against bacteria, fungi, protozoa and viruses. The mouthwash has been shown to be effective in reducing plaque and gingivitis and may be a useful adjunct to routine oral hygiene. It also reduces the incidence, severity and duration of radiation mucositis. Absorption of excess iodine has been postulated to result in metabolic complications, however this is not of concern in patients without pre-existing thyroid disease and if the patient spits out the solution.

Chlorhexidine gluconate with triclosan has shown most effective antimicrobial activity at full strength concentration against Staphylococcus aureus and Streptococcus mutans, which causes dental caries and oral infections respectively in comparison with the other five mouthwashes (i.e Thymol, Chlorhexidine, Cetylpyridinium chloride, Belleric myrobalan and Povidone iodine).

Conclusion:-

According to this study, Chlorhexidine gluconate with triclosan has shown most effective antimicrobial activity at full strength concentration against Staphylococcus aureus and Streptococcus mutans which causes dental caries and oral infections respectively in comparison with the other five mouthwashes (i.e Thymol, Chlorhexidine, Cetylpyridinium chloride, Belleric myrobalan and Povidone iodine).

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RESEARCH ARTICLE

SLEEP QUALITY, DAYTIME SLEEPINESS, AND DEPRESSION AMONG ER PHYSICIANS IN RIYADH'S REFERRAL HOSPITALS..

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Key words:-

ER physicians, sleep quality, daytime sleepiness, depression.

Abstract

Objectives: To assess the prevalence of daytime sleepiness, poor sleep quality and depression among (emergency room) ER physicians in comparison to family medicine physicians.

Methods: We compared 68 ER physicians with 69 family medicine physicians in an observational case control study. A self-administered questionnaire composed of the Epworth Sleepiness Scale (ESS), the Pittsburgh Sleep Quality Index (PSQI) and the Beck Depression Inventory Scale (BDI).

Results: The mean total ESS score of ER physicians was significantly higher than the score of family medicine physicians as 41.2% of ER physicians scored ≥ 10 , while only 14.5% of family medicine physicians scored ≥ 10 . Moreover, ER physicians had a higher on the PSQI (indicating poorer sleep quality), and 83.8% of them had poor sleep quality (≥ 5) in comparison to 50.7% of family medicine physicians. The BDI scores of the ER and family medicine physicians indicated that 41% and 22% suffered from some level of mood disturbance, respectively. Pearson correlation showed a positive and significant relationship between sleep quality and depression among ER physicians ($P < 0.001$, $r = 0.437$).

Conclusion: Our study demonstrated that daytime sleepiness, poor sleep quality and depression are prevalent among ER physicians. Moreover, there was a strong correlation between poor sleep quality and depression.

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Introduction:-

Night shift work is a well-known risk factor for disturbed circadian rhythm¹, leading to significant drawbacks in quality of sleep and sleep efficiency²⁻⁴. Therefore, night shift workers, such as emergency room physicians, have a significant liability to develop poor sleep quality. A fair number of studies have focused on the effects of shift work on the general health, and have found a link between shift work and cardiovascular diseases⁶⁻¹², diabetes mellitus type 2¹³⁻¹⁷, breast cancer²¹, metabolic syndrome²²⁻²³, memory loss²⁴ and obesity²⁵⁻²⁶. In contrast, fewer studies have addressed the relation between shift work and its effect on sleep quality and its negative consequences on sleep patterns and mood. Shift work schedule has numerous effects on the quality of sleep and wakefulness of workers^{25, 27}. Some studies mentioned that the irregular work hours seem to exert acute effects on sleep and alertness of individuals similar to those witnessed in clinical insomnia, which could be responsible for major negative human

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and economic impacts due to fatigue related accidents and diminished productivity²⁵. There is a definite association between low sleep quality and shift work^{27, 28}. A previous study done in Kingdom of Saudi Arabia revealed that the quality of sleep of shift-workers was significantly affected in comparison to those without shift work²⁹. Sleepiness and stress are among the risk factors for worsening sleep quality among shift workers³⁰. Poor sleep quality was associated with increased work injury³⁰. Low sleep quality was reported to correlate with a poor quality of life among nurses working rotating shifts²⁸. Insomnia among shift workers plays a role in experiencing sleep difficulties³¹. Regarding poor sleep quality and depression, one study was done on multiple sclerosis patient had found a link between poor sleep quality and depression³². One studies suggested that the increase in depressive symptoms across adolescence is partially mediated by sleep-related developmental changes³³. Increasing depression among sleep apnea patients was associated with poor sleep quality, as well³⁴.

No study has assessed the effect of shift work on ER physicians on sleep and mood in Saudi Arabia; therefore, we conducted this study to estimate the prevalence of poor sleep quality, daytime sleepiness, and depression among ER physicians.

Methods:-

This is an observational case control study that was conducted between November 2015 and April 2016. The cases were randomly chosen and comprised of 68 emergency medicine physicians from 8 major Riyadh hospitals (King Khalid University Hospital, King Faisal Specialist Hospital and Research Center, King Fahad Medical City, National Guard Hospital, Prince Sultan Military Medical City, King Saud Medical Complex, Imam Abdulrahman bin Faisal Hospital, King Salman Hospital) who have worked regular shifts for at least 3 months. The controls comprised of 69 family physicians randomly chosen from 5 major Riyadh Hospitals (King Khalid University Hospital, King Fahad Medical City, National Guard Hospital, Prince Mohammed bin Abdulaziz Hospital, King Saud Medical Complex). The project was approved by the Ethics Committee and informed consent was obtained from participants.

Data collection and Questionnaire:-

As medicine is taught in English in Saudi Arabia, a self-administered questionnaire in the English language was distributed to all participating physicians. The questionnaire used in this study was composed of four sections: Demographic information, position, previous diagnosis of either depression or a sleeping disorder, and medications.

The Epworth Sleepiness Scale (ESS): This is a validated questionnaire that is used to assess daytime sleepiness. It contains 8 items that assess the likelihood of dozing in a variety of daily living situations. The highest score is 24, and a score ≥ 10 indicates sleepiness³⁶.

The Pittsburgh Sleep Quality Index questionnaire: This is an instrument used to measure the quality and patterns of sleep in adults. It differentiates “poor” from “good” sleep quality by measuring seven areas (components): subjective sleep quality, sleep latency, sleep duration, habitual sleep efficiency, sleep disturbances, use of sleeping medications, and daytime dysfunction over the last month. A total score of 5 or higher is indicative of poor sleep quality³⁵.

The Beck Depression Inventory: This is a validated questionnaire to assess depression. It is composed of items relating to symptoms of depression. It contains 21 questions; each answer being scored on a scale value of 0 to 3. Higher total scores indicate more severe depressive symptoms. A score of 11-16 indicates mild mood disturbance, 17-20 indicates borderline clinical depression, 21-30 indicates moderate depression, 31-40 indicates severe depression, and a score of over 40 indicates extreme depression³⁷.

Data Analysis:-

Data were expressed as mean \pm SD for continuous data. Categorical data were expressed as n (%). Student t-test or Mann-Whitney U-test (depending on normality) and chi-square test were used to compare numerical or categorical data, respectively. One way ANOVA or Kruskal Wallis test (if normality assumption violated) were used to compare BDI groups. The results were considered statistically significant if $P \leq 0.05$. Pearson correlation or Spearman rank-order correlation (if normality assumption violated) were used to assess correlation between ESS, PSQI and BDI scores. Statistical Package for Social Sciences (SPSS), IBM version 22 (SPSS Inc., Chicago, IL, USA) software was used for data analysis and management.

Results:-

The study included 68 ER physicians and 69 family medicine physicians. Table 1 shows that the two groups were similar in all parameters except for age; the mean age for ER physicians was 35.2 years (SD: 8.6), and mean age of 40.4 (SD:10.5) in family medicine physicians. There was a difference in the position parameter, as well. There were a higher proportion of male physicians in both groups; 70.6% of ER physicians were males, and 58% were males in family medicine physicians group. One subject of ER physicians reported being diagnosed with a sleeping disorder, and two from the control group, one of them specified being diagnosed with obstructive sleep apnea. In addition, 8.8% of ER physicians are diagnosed with depression; almost the same percentages, 8.7%, of family medicine physicians are diagnosed with depression as well. Three subjects of the ER physicians reported being on Selective Serotonin Reuptake Inhibitors, and one reported being on beta-adrenergic blocking agents. Only one subject of family medicine physicians is on beta-adrenergic blocking agents.

The mean total ESS score of ER physicians was significantly higher than the score of family medicine physicians. Furthermore, 41.2% of ER physicians scored ≥ 10 , while only 14.5% of family medicine physicians scored ≥ 10 [Table 2]. ER physicians also scored higher on the PSQI, 83.8% of them were having poor sleep quality (≥ 5) in comparison to 50.7% of family medicine physicians [Table 2]. Finally, a strong correlation was observed between sleep quality and depression among ER physicians, Pearson correlation shows a positive and significant relation ($P < 0.001$, $r = 0.437$) [Figure, Table 3]. The results of Pearson correlation between ESS score and BDI score represents that daytime sleepiness has a weak correlation with depression in ER physicians ($r = 0.172$) [Figure, Table 3]. Pearson correlation between sleep quality and daytime sleepiness has a weak correlation ($r = 0.178$), as well. [figure, Table 3].

Table I:- Characteristics; comparison between ER physicians and family medicine physicians.

Variable	Mean \pm SD		P-Value
	ER physicians n=68	Family medicine n=69	
Age	35.2 \pm 8.6	40.4 \pm 10.5	0.003
Gender			
Female	20 (29.4)	29 (42)	0.123
Male	48 (70.6)	40 (58)	
Marital Status			
Single	17 (25)	12 (17.4)	0.276
Married/Divorced/Widower/Widow	51 (75)	57 (82.6)	
Position			
Consultant	13 (19.1)	26 (38.2)	0.014
Assistant professor/Registrar/Resident	55 (80.9)	42 (61.8)	
Diagnosed with a sleeping disorder			
No	67 (98.5)	67 (97.1)	1.000
Yes	1 (1.5)	2 (2.9)	
Sleep Disorders Diagnoses			
OSA	0 (0)	1 (1.4)	1.000
Diagnosed with depression			
No	62 (91.2)	63 (91.3)	0.979
Yes	6 (8.8)	6 (8.7)	
Medication			
Not on medication	64 (94.1)	68 (98.6)	0.208
On Medication	4 (5.9)	1 (1.4)	

Table II:- ESS, PSQI and BDI results, comparison between ER physicians and family medicine physicians.

Variable	Mean \pm SD		P-Value
	ER physicians n=68	Family medicine n=69	
ESS Score	9.1 \pm 4.6	6.1 \pm 3.8	<0.001
ESS Diagnostic	40 (58.8)	59 (85.5)	<0.001
< 10	28 (41.2)	10 (14.5)	
≥ 10			
Global PSQI Score	8.3 \pm 4	5.3 \pm 3.2	<0.001
PSQI Diagnostic	57 (83.8)	35 (50.7)	<0.001
≥ 5	11 (16.2)	34 (49.3)	
< 5			
BDI Score	11.5 \pm 10.1	6 \pm 7.1	< 0.001

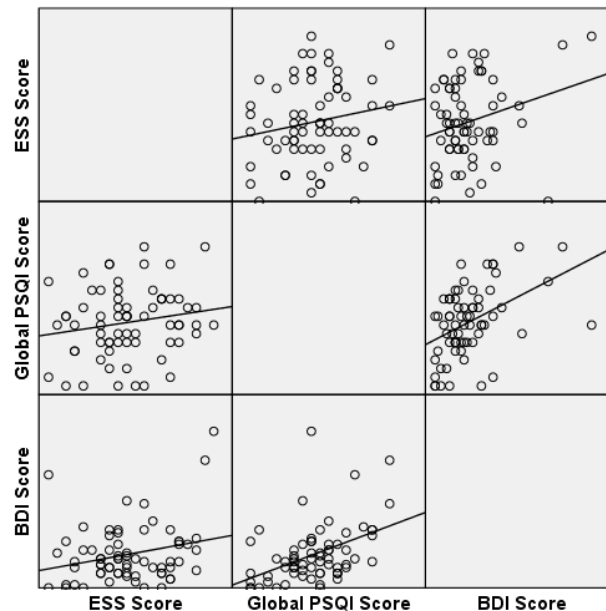
ESS: Epworth Sleepiness Scale

PSQI: Pittsburgh Sleep Quality Index

BDI: The Beck Depression Inventory

Table III:- Correlations between ESS, PSQI and BDI scores in ER physicians.

Variable	n=68	r	p-Value
ESS Score Vs Global PSQI Score		0.178	0.146
ESS Score Vs BDI Score		0.172	0.161
BDI Score Vs Global PSQI Score		0.437	< 0.001

**Figure:-** Scatter plot between ESS, PSQI and BDI scores in ER physicians.**Discussion:-**

This study was conducted to assess the prevalence of daytime sleepiness, poor sleep quality and depression among emergency room physicians with a control group of family physicians in Riyadh referral hospitals.

The results of this study showed by using the PSQI³⁵ that poor sleep quality was prevalent among emergency room physicians with a prevalence of 84% compared to 51% in family medicine physicians. Moreover, we found that falling asleep during daily activities is more prevalent among ER physicians with a prevalence of 41% compared to 14% reported in family medicine physicians. In addition, by using Becks Depression Inventory (BDI)³⁷ it was found to be statically significant and indicating that 41% of emergency room physicians room physicians were depressed

compared to 22% of family medicine physicians who were found to be depressed. Moreover, we found that poor sleep quality in emergency room physicians is strongly associated with depression.

A previous study conducted on male factory workers in Aseer, Saudi Arabia²⁸ showed that poor sleep quality among shift workers was reported to be around 67.6% compared to only 20.3% in non-shift workers. Furthermore, another study was conducted on ER physicians in Pittsburgh³⁸, reported that a percentage of almost 31% suffered from poor sleep quality. In comparison, the percentage of shift workers and ER physicians suffering from poor sleep quality are much less than what was reported in ER physicians in our recent study. Both studies focused on the effect of shift work on the sleep quality of the shift workers. It is possible that doctors in our study do more shifts than ER physicians in the American study.

Limitations:-

A limitation of this study was the relatively small sample size. However, the number of emergency room physicians in Riyadh is relatively small. Another limitations were that a higher proportion of the participants in this study were males.

Conclusion:-

Our study demonstrated that daytime sleepiness, poor sleep quality and depression are prevalent among emergency room physicians. Moreover, there was a strong correlation between poor sleep quality and depression. Efforts are needed to investigate the causes of these problems among emergency room physicians and to propose practical solutions.

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RESEARCH ARTICLE

STUDY THE ROLE AND EFFECT OF DISTANCE EDUCATION ON REGULAR COURSES.

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Abstract

This research addressed the effect of distance learning courses on regular courses. Distance Learning Courses are very popular now days and persons are much interested in distance learning courses because this is cheaper than the regular courses but we found that some they do not believe in this system because they think this will not help in grooming the personality of the executive. So that some people were satisfied with distance Learning System but still some people believe in regular studies. Regular class attendance keeps students on track with their course work in classroom courses. Distance learning courses do not have regular class meetings, so students must be self-disciplined enough to keep up with their studies. Distance learning has opened doors for many students, especially for those who cannot afford to pay for the high fees of full-time courses. Although online distance learning courses have many advantages over on-campus degree courses, many factors come into play in order to make this a success for students.

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Introduction:-

Mukhopadhyay (1998) described distance education as “Distance education is a mode of delivery and not an educational system; open education is a system-distance education is often the delivery mode of open education”. The first three Open Learning Institutes were set up in Australia, Canada and New Zealand in 1916, 1919 and 1922, respectively. The success of Delhi University’s endeavor in distance education motivated other universities to introduce several kinds of programmes through correspondence courses.

As distance learning has evolved, various terms have been used to describe the particular technique being utilized. Moreover, distance learning techniques have become more widely diffuse across educational and organizational settings, spawning even more unique terms and definitions. Distance learning is training that takes place largely synchronously; that is, the material is delivered to all participants at the same time even though participants are separated by geographical distance. The US Distance Learning Association defines distance learning as “the delivery of education or training through electronically mediated instruction including satellite, video, audio, audiographic computer, multimedia technology etc.

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Literature Review:-

Mr. Wei Yuan Zhang at all from the open University of Hong Kong SAR, given their views on “A Preliminary study of current states of distance Education research in India”. They suggested that, In India, higher education through the distance mode was first offered by the School of Correspondence and Continuing Education, the University of Delhi, in 1962. At present there are 10 open universities and 62 conventional universities offering correspondence courses, little literature is available on the current state of distance education research in India. This study surveys 29 experts in 14 distance education institutions in India and examines their foci in tertiary distance education research. The data for each expert include gender, administrative positions, electronic mail address and homepage, research areas, recent research projects, and proposed future studies.

Mr. M.M Ansari from *Distance Education Council, IGNOU, New Delhi, India* wrote on “Best Practices in Open and Distance Learning Systems in India: An Assessment” The success of Open Distance Learning institutions has contributed to increased credibility and acceptance of distance learning systems as an effective mode of teaching and learning. This genre of education has made significant contributions to human resource development. At least 40 per cent of incremental students population at post-secondary level, of the total of about 0.5 million students per year, prefer to study through distance mode. They opt for different types and levels of degree/diploma programmes that are offered by the QOL institutions.

Mr. Satya Sundar SETHY from Staff Training and Research Institute of Distance Education Indira Gandhi National Open University Maidangarhi, New Delhi, India On “Distance Education In The Age Of Globalization: An Overwhelming Desire towards Blended Learning The aim of this paper is to discuss the nature and status of distance education in the age of globalization, i.e. how best it fits for the present educational scenario.

Dr. Ashok Kumar Gaba, Senior Research Officer, Staff Training and Research Institute of Distance Education Indira Gandhi National Open University given his view on “The Role of Distance Education in the Human Resource Development of India. The objective of this paper is to analyze the role of distance education in the human resource development (HRD) of India. The paper has been divided into three parts. First, part discusses the relationship between distance education and human resource development in general and conceptual context in particular. Indian rank in HRD corresponding to other countries has also been highlighted in this part. Second part highlighted how distance education helped to learners to bring changes economically in their personal life after getting degree from distance education institution. The third and last part discusses the futuristic model of distance training for on the job learners.

Research Objectives:-

The main aim of this study is to analyze effect of Distance education on regular courses. This research is an endeavor towards to analyze the attitude of the students towards distance education. Today number of distance institutions has crossed a number of 850 which offer more than 16,000 courses to over 15 million learners all around the world. During this research we conducted a survey among the youth of Uttar Pradesh in order to verify the roll and impact of distance education on regular courses.

Area of Research:-

For our convenience we have selected many area of *district of Uttar Pradesh (East)* as a research area that includes Faizabad, Lucknow, Gonda, Ambedkarnagar, Barabanki, Sultanpur for the research project.

Sample Unit:-

My sampling units were students of different *district of Uttar Pradesh (East)*. I have taken 150 respondents for my study.

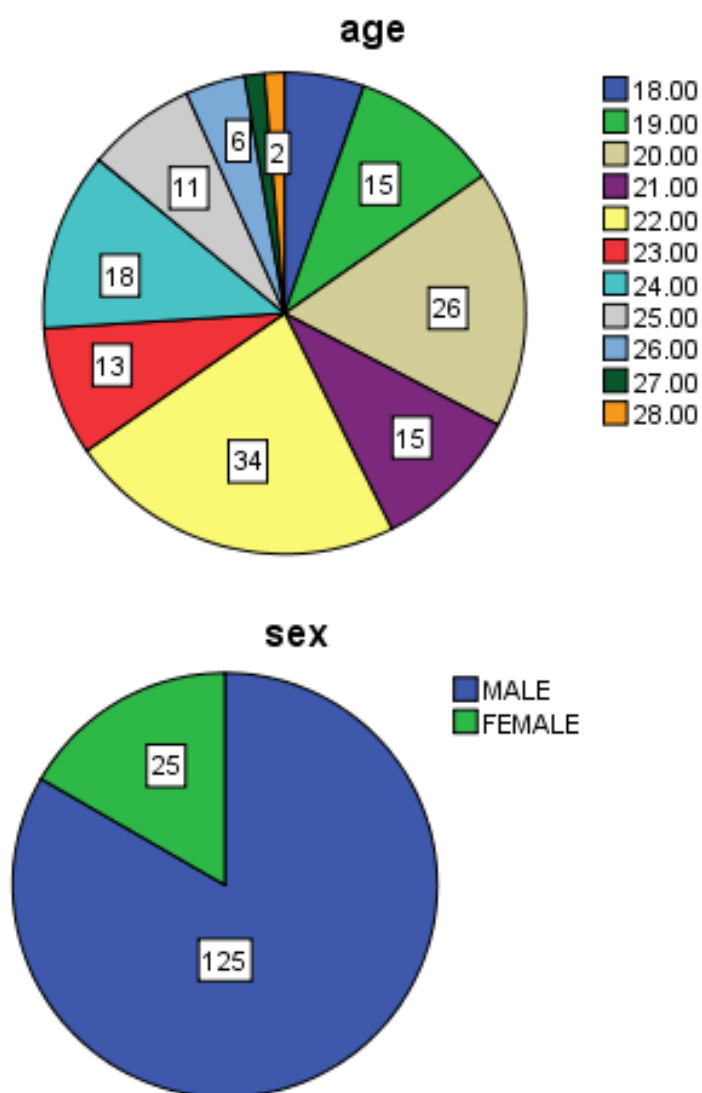
Hypothesis-Let us take the hypothesis that Distance education having no effect on regular courses.

I have used the conclusive Research Method in this study. As our project deals with the study of distance learning education so we need to meet the student. Our primary aim is to find the psychology of student for what they think about the distance learning education. We had interacted with students of different areas and came out with some conclusions, for that we had framed questions.

Statistical Analysis
Frequencies

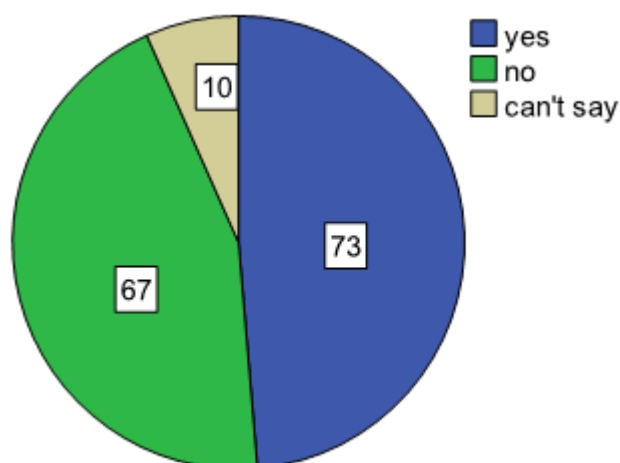
Statistics					
		age		Sex	
N	Valid	150		150	
	Missing	0		0	
Age					
Age		Frequency	Percent	Valid Percent	Cumulative Percent
Valid	18.00	8	5.3	5.3	5.3
	19.00	15	10.0	10.0	15.3
	20.00	26	17.3	17.3	32.7
	21.00	15	10.0	10.0	42.7
	22.00	34	22.7	22.7	65.3
	23.00	13	8.7	8.7	74.0
	24.00	18	12.0	12.0	86.0
	25.00	11	7.3	7.3	93.3
	26.00	6	4.0	4.0	97.3
	27.00	2	1.3	1.3	98.7
	28.00	2	1.3	1.3	100.0
	Total	150	100.0	100.0	

Sex					
		Frequency	Percent	Valid Percent	Cumulative Percent
Valid	MALE	125	83.3	83.3	83.3
	FEMALE	25	16.7	16.7	100.0
	Total	150	100.0	100.0	



Statistics					
Response Table-					
N	Valid		150		
	Missing		0		

		Frequency	Percent	Valid Percent	Cumulative Percent
Valid	Yes	73	48.7	48.7	48.7
	No	67	44.7	44.7	93.3
	can't say	10	6.7	6.7	100.0
	Total	150	100.0	100.0	

**Chi-Square Test:-**

Hypothesis-Let us take the hypothesis that Distance education having no effect on regular courses.

Frequencies

Table			
	Observed N	Expected N	Residual
yes	73	50.0	23.0
no	67	50.0	17.0
can't say	10	50.0	-40.0
Total	150		

Test Statistics

Test Statistics	
	QUESTION13
Chi-Square	48.360 ^a
Df	2
Asymp. Sig.	.000
a. 0 cells (.0%) have expected frequencies less than 5. The minimum expected cell frequency is 50.0.	

Finding/Interpretation:-

For two degree of freedom at 5% level of significance, the table value of $\chi^2=5.991$. The calculated of χ^2 is much greater than the table value and hence our hypothesis stands rejected. We, therefore conclude that Distance education having effect on regular courses.

The above diagram shows that exactly half of the percentage in yes and half in no that means students think that distance learning has effect on regular courses are equal in number as thinkers who think that it has no effect on regular courses. It happens generally as government give priority to regular courses in comparison of distance learning. Distance students can save costs because people do not have to travel to campus to attend classes on regular scheduled basis, nor live on campus, nor leave their current professional commitments, although they will have to pay for tuition fees and expenses for the purchase of learning materials. This relatively much lower cost of distance education on the parts of the students compared to the face-to-face mode of education should improve participation and expand greater opportunities for those socially and economically less advantaged groups of the society, particularly women to have access to quality education higher education at affordable costs. Those who thought that distance education was passé are in for a surprise as close to one-fourth of the country's students are getting educated through the distance mode. The Open Distance Learning (ODL) system, also known as Distance Education (DE) system, has evolved as one of the effective modes of education and training as the overall annual growth. In fact, the enrollment in distance education has been increasing approximately at the rate of more than 10% in last two decades. Enrollment in Open Universities (OUs) and Distance Education Institutions (DEI) has increased steadily at a higher pace than in conventional programmes. According to the report of a seven-member committee headed by NR Madhava Menon, the share of distance education increased

from 2.6% in 1975-76 to 8.9% in 1985-86 and further improved to 10.7% in 1990-91 and to 20.56% in 2008-09. "At present, close to 24% of all enrollments are in the ODL system and growing fast because of the reach of this mode and the opportunities it gives to those who are already employed and seek to enhance their qualifications.

Conclusion:-

Distance Education plays a vital role in the development and access to education. It is less expensive than the conventional system though the cost per student differs among the distance education institutions. The cost variations are due to the expenditure pattern and the enrolment trend. The fixed cost for the Open University is high because it has to create physical infrastructure and appoint teaching and administrative staff. But the CCIs utilize the physical infrastructure of the concerned university and hire the services of the resource persons from the formal system. Hence, the fixed cost is comparatively low in the correspondence course institutions. As far as the variable cost is concerned, where the enrolment is more, the per capita cost tends to reduce. This is due to large scale production of learning materials, and other expenditures like remuneration to lesson writers, payment to resource persons, rent and other expenditures to conduct PCPs. Hence, the per head cost is low in the institutions where the enrolment is high. To determine the cost in any distance education system following four factors need to be considered:

- ❖ Compare the cost per student in distance education system with the cost per student in the formal system.
- ❖ Quality of equipment/material used.
- ❖ Number of students enrolled in each system.
- ❖ Students' private costs.

Distance education has opened a new window of opportunities for those who craved for further studies. In a way it has helped numerous professionals, dropouts (school & college), housewives, etc to accomplish their unfinished dreams.

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RESEARCH ARTICLE

A COMPARATIVE ANALYSIS OF FINANCIAL RATIOS WITH SPECIAL REFERENCE TO TATA STEEL (2014 AND 2015).

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TATA, Ratio, Liquidity, Profitability, leverage

Abstract

In 2014, the world crude steel production reached 1665 million tonnes (mt) and showed a growth of 1% over 2013. China remained the world's largest crude steel producer in 2014 (823 mt) followed by Japan (110.7 mt), the USA (88.2 mt) and India (86.5 mt) at the 4th position. WSA has projected Indian steel demand to grow by 6.2% in 2015 and by 7.3% in 2016 as compared to global steel use growth of 0.5% and 1.4% respectively (Ministry of Steel, Government of India 2016).

Established in 1907 as Asia's first integrated private sector steel company, Tata Steel Group is among the top-ten global steel companies with an annual crude steel capacity of over 29 million tonnes per annum. It is now the world's second-most geographically-diversified steel producer, with operations in 26 countries and a commercial presence in over 50 countries. The Tata Steel Group, with a turnover of Rs. 1, 48,614 crores in FY 14, has over 80,000 employees across five continents and is a Fortune 500 company (Tata Steel 2016).

All research used for this paper will be secondary information. Secondary information was taken from Tatasteel.com. The method of analysis is through ratios. The study concentrates on profitability ratios and concentrates on company's two-year growth.

Liquidity ratios affect profitability as they have a positive direct relationship. If liquidity is low, profitability will be affected negatively. If Liquidity is high, profitability will be affected positively. Tata Steel, one of the largest players in the Indian despite this, their economic activity had decreased in 2015 compared to 2014.

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Introduction:-

Tata Steel Limited is an Indian multinational steel-making company headquartered in Mumbai, Maharashtra, India. A subsidiary of the Tata Group, it has a total number of 79,657 employees (2015) and incurs revenues of US\$ 20 billion or 135,278 Crore rupees. Tata Steel's larger production facilities comprise those in India, the UK, the Netherlands, Thailand, Singapore, China and Australia. Operating companies within the Group include Tata Steel Limited (India), Tata Steel Europe Limited (formerly Corus), Tata Steel Singapore and Tata Steel Thailand. Tata Steel's competitors consist of JSW steel, SAIL and Jindal industries, all major contributors in the steel industry.

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The Tata Steel Group's vision is to be the world's steel industry benchmarks in "Value Creation" and "Corporate Citizenship" through the excellence of its people, its innovative approach and overall conduct. Underpinning this vision is a performance culture committed to aspiration targets, safety and social responsibility, continuous improvement, openness and transparency. In 2008, Tata Steel India became the first integrated steel plant in the world, outside Japan, to be awarded the Deming Application Prize 2008 for excellence in Total Quality Management. In 2012, Tata Steel became the first integrated steel company in the world, outside Japan, to win the Deming Grand Prize 2012 instituted by the Japanese Union of Scientists and Engineers (Tata Steel 2016).

Literature review

The ratio data has been taken from Moneycontrol.com, a reputed and sought after source of information related to finance and business in India. However, this paper is not being analytical about the steel industry in India and is only considering one big player (Tata Steel). Our study of the company is also limited as we only consider data from the most recent 3 years of the company's activities. All definitions of ratio's as well as their interpretation have been taken from the book Introduction to Managerial Accounting (Brewer, Garrison and Noreen 2016).

As said by Florenz Tugas, most financial statement analyses focus on firms belonging to industries that either contributes significantly to economic figures or post it in a highly competitive business environment. Whatever the motivation may be, financial statement analysis should be made available to all industries for reasons of comparability and benchmarking (Tugas 2012). In addition, empirical results of a financial statement can be used to adequately signal business failure in the context of normal economic circumstances (Tuvadaratragool 2013). According to Savchuk, liquidity is one of the most essential factors for assessing the company's position (Savchuk 2014).

Limitations of research:-

- The paper analyzes only one company from the steel industry.
- Restricted study from year 2014 to year 2015.
- Secondary information from online resources.

Research methodology:-

All research used for this paper will be secondary information. A trusted Indian online source, Moneycontrol.com (Money Control 2016), used by seasoned professionals as well as stock brokers is the source of the secondary research and information employed in this paper. Secondary information was also used from Tatasteel.com. The method of analysis is through ratios. The study concentrates on profitability ratios and concentrates on company's two-year growth (Kothari and Garg 2015).

Data Analysis & Findings:-

Liquidity Ratios:-

H₍₀₎: There is no significant difference in the liquidity ratios in year 2014 and 2015

H₍₁₎: There is significant difference in the liquidity ratios in year 2014 and 2015

Table (1):- Descriptive Statistics

Liquidity Ratios	Mar'15	Mar'14	Industry Standard
Current Ratio	0.52	0.62	2:1
Quick Ratio	0.32	0.27	1:1

Current Ratio:-

H₍₀₎ a: There is no significant difference in the liquidity ratios in year 2014 and 2015

H₍₁₎ a: There is significant difference in the liquidity ratios in year 2014 and 2015

In the table above, the industry standard is mentioned. However when we look at Tata Steel's ratios, we see that their current ratio is lower than the standard. A relatively low current ratio represents that the liquidity position of the firm is not good and the firm shall not be able to pay its current liabilities in time without facing difficulties.

As we can see, the current ratio should be 2, where as Tata's is 0.62 in March 2014 and decreases to 0.52 in March 2015. The decrease will affect Tata negatively. This could be due to many reasons, some being insufficient funds to pay off liabilities or the business trading beyond its capacity. In the case of Tata's current ratio, hypotheses H₍₁₎ a, proves to be applicable as there is significant difference in the liquidity ratios in year 2014 and 2015.

B) Quick Ratio

H₍₀₎ b: There is no significant difference in the liquidity ratios in year 2014 and 2015

H₍₁₎ b: There is significant difference in the liquidity ratios in year 2014 and 2015

On the other hand, Quick ratio should be 1. Tata's quick ratio is low at a 0.27 in March 2014 and 0.32 in March 2015, however there is an increase when year 2014 is compared to year 2015. This increase will have a positive affect on the overall function and business activity of Tata. This could be because Tata is meeting its short-term obligations in time because of its very efficient inventory management. In the case of Tata's quick ratio, hypotheses H₍₁₎ b proves to be applicable as there is significant difference in the liquidity ratios in year 2014 and 2015.

After looking at the balance sheets of Tata we can make some assumptions for why this effect might have occurred. Cash and bank Balances reduced from 961.16 crore rupees in 2014 to 478.59 crore rupees in 2015. Their cash flow has been affected and reduced; this might have an effect on the liquidity ratio as 'liquidity' of the company has reduced. A step toward improving the ratios could be that of increase liquidity or the inflow and outflow of cash. In the case of Tata's Liquidity ratios, hypotheses H₍₁₎ proves to be applicable as there is significant difference in the liquidity ratios in year 2014 and 2015.

Profitability Ratios:-

H₍₀₎ : There is no significant difference in the liquidity ratios in year 2014 and 2015

H₍₁₎ : There is significant difference in the liquidity ratios in year 2014 and 2015

Table (2):- Descriptive Statistics

Profitability Ratios	Mar'15	Mar'14
Operating Profit Margin (%)	18.87	23.95
Gross Profit Margin (%)	13.81	19.17
Net Profit Margin (%)	12.82	15.41
Return On Capital Employed (%)	9.03	9.25

Gross Profit Ratio:-

H₍₀₎ a: There is no significant difference in the liquidity ratios in year 2014 and 2015

H₍₁₎ a: There is significant difference in the liquidity ratios in year 2014 and 2015

Gross profit is very important for any business. It should be sufficient to cover all expenses and provide for profit. There is no norm or standard to interpret gross profit ratio. Generally, a higher ratio is considered better. To judge whether the ratio is satisfactory or not, it should be compared with its own past ratios.

A consistent improvement in gross profit ratio over the past years is the indication of continuous improvement. However as we see above, Tata's gross profit has fallen from 19.17% in 2014 to 13.81% in 2015. This could have occurred due to a decrease in overall sales volume caused by a decrease in units sold or decreased selling prices or due to an increase in purchasing costs that is not offset by an increase in selling prices. For Tata's gross profit, hypotheses H₍₁₎ b is applicable as there is a negative change.

Operating Ratio:-

H₍₀₎ b: There is no significant difference in the liquidity ratios in year 2014 and 2015

H₍₁₎ b: There is significant difference in the liquidity ratios in year 2014 and 2015

This ratio is used to measure the operational efficiency of the management. It shows whether the cost component in the sales figure is within normal range. A low operating ratio means high net profit ratio i.e., more operating profit. To judge whether the ratio is satisfactory or not, it should be compared with its own past ratios.

As shown above, Tata's operating ratio has fallen from 23.95% in 2014 to 18.87% in 2015. This reduction could be due to either a decrease in sales or an increase in expenses.

Net Profit Ratio:-

H₍₀₎ c: There is no significant difference in the Net Profit Ratio in year 2014 and 2015

H₍₁₎ c: There is significant difference in the Net Profit Ratio in year 2014 and 2015

For the purpose of this ratio, net profit is equal to gross profit minus operating expenses and income tax. All non-operating revenues and expenses are not taken into account because the purpose of this ratio is to evaluate the profitability of the business from its primary operations. To judge whether the ratio is satisfactory or not, it should be compared with its own past ratios.

As shown above, Tata's operating ratio has fallen from 23.95% in 2014 to 18.87% in 2015. This reduction could be due to either a decrease in sales or an increase in expenses.

Return on Capital Employed/ Return on Investment:-

H₍₀₎d: There is no significant difference in the Return on Capital Employed in year 2014 and 2015

H₍₁₎d: There is significant difference in the Return on Capital Employed in year 2014 and 2015

Return on capital employed ratio measures the efficiency with which the investment made by shareholders and creditors is used in the business. Managers use this ratio for various financial decisions. It is a ratio of overall profitability and a higher ratio is, therefore, better.

As shown above, Tata's Return On Capital Employed has fallen from 9.25% in 2014 to 9.03% in 2015. For this case, hypotheses H₍₀₎d is applicable as there is no significant difference in the Return On Capital Employed. This is a positive sign as their return on investment has not changed much. This keeps all stakeholders feeling secure and confident.

Conclusion:-

As shown in the analysis above, Tata Steel, one of the largest players in the Indian steel market has suffered some fallbacks. However, the company has made an effort to maintain their stance in the market and have been successful in doing so. Despite this, their economic activity had decreased in 2015 compared to 2014. This could be due to a few reasons, as mentioned above. Tata needs to take some steps in moving forward as their growth slows down and is hampered. Tata Steel exports 67% of their revenue and should work on the untapped domestic market by being more competitive. Though Tata Steel's revenues from the domestic market are 32%, this figure can be increased, leading to higher sales revenue and eventually higher profits. Tata also needs to concentrate on reducing their expenses and seen above and in their other financial statements, as expenses eat away at the company's profits quite largely.

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RESEARCH ARTICLE

RELEVANCE OF BIBLICAL VALUES TO THE MODERN EDUCATIONAL SYSTEM.

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loyalty

Abstract

The present study has great scope in the modern Educational system. We are living in a world which never considers values. Values can change the behavior and attitudes of every one. Value education is important for our students. And Bible always remains us about values. We can see the different teaching methods and techniques used by Jesus Christ and we can also understand that many of them have great relevance today. There are also guiding principles to teachers, parents and students. Bible strongly advocates the need for discipline and it is also helpful for our educational system.

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Introduction:-

“The Bible is no Mere a book, but a living creature, with a power that conquers all that opposite it” “NAPOLEON BONAPARTE(1769-1821)

Bible is like a software, it enters in your life, scan your problems, edit your tension, download solutions, delete your worries and saves you.

A child gets his first education from his family (proverbs 17:6)” Children are the crown of old men and the glory of children in their father”. Little children are Humble, dependent, harmless, godly and deserve to be cherished by parents. Each family believes in certain religion. So there is an interconnection between education and religion believes imparted from the family. Each religion has its own norms and principle and these are conveying through religious books. The thoughts and ideas of bible great related to values.

Bible and values:-

Philippians 23:4, “don’t be Selfish; don’t live to make a good impression on others. Be humble, thinking of others as better than yourself. Don’t think only about your own affairs, but be interested in others too, and what they are doing .The implies the morality and good character that showed be promoted by a family and educational system on their children. Bible does not teach what method to use, but it does teach what character to build. One of the main aims of Bible is to inculcate virtues among individuals. If the teacher in any setting loves children and promotes Christ like conduct by example, the school conforms to the bible. This does not mean that the child will only learn to love the lord, but that he will also learn to diligent at work, competent in labour, kind in relationships, forgiving in conflict and gracious in judgement.

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Compassion:-

The definition of "Compassion" simply says that " it is the sympathetic consciousness of others distress along with a desire to alleviate it"

The parable of the good Samaritan (Luke 10: 25-37) is a best example for compassion. There are more than to specific references to compassion in the bible. Most old testament version refer to the lord's compassion toward his people and most new testament version refer to Jesus compassion in need.

"Lord will always have compassion on us" (2 kings 13:23, Psalm 103:13, Isaiah 54:8)

"Jesus felt compassion on those in need" (Matt 9:36, 14:14, Mark 6:34, 8:2, 15:20)

"Compassion fulfill the law of Christ" (Galatians 6:3)

Loyalty:-

The world loyalty means unswerving in allegiance as faithful to one's sovereign or government, faithful to a private person to whom fidelity in due faithful to a cause, ideal or custom. Bible has to say about loyalty and being loyal.

"Loyalty is unwavering in good times and bad" (Proverb 17:7)

"Loyalty is what you do, not what you say (Matt 26:33-35)

Loyalty is in your heart. It is willing and not Reluctant (Psalm 78:8)

Forgiveness:-

Bible says you should forgive seventy seven times. The benefits of forgiveness are

It is necessary for our own forgiveness:-

1. Jesus said. "But you are Praying, first forgives any one are holding a grudge against, so that your father in heaven will forgive your sins too" (Mark 11:25, 26)
2. Spiritual cleaning
3. James says that the elders of the church should pray over those who are and lord will make them well, adding "and *Anyone who has committed sins will be forgiven*" (James 5:15-16)

Integrity:-

Integrity means firm adherence to a code of moral values, ie, incorruptible, honor;

Bible says about integrity are "Integrity means treating people fairly and honest"(Deut 25:15, Proverbs 16:11-13)"Integrity is giving your word and keeping it.(Exodus 8:28-32)

"Lord hates lies and lack of integrity (Zachariah 8:16-17)

Moral values:-

Moral values refer to those values that are related to an individual's character and personality conforming to what is right and virtuous. They reveal a person's self control. Some of the moral values envisaged in bible are as follows.

Leadership value and Bible:-

Maintaining one's values can cost a leader dearly. So how do we decide what matters most when we are weighing the bottom line costs against our bottom line conviction. The first step in effective leadership is defining core values. Until that is done, the ship the leader is trying to steer has no rudder. Vision, mission, strategy and outcomes are difficult-if not impossible to define until values are clear. Jesus knew that. Early in the process of developing his team of disciples, he forced them to confront this foundational issue.

Jesus told his disciples that the core value, the driving value, the internal value is this "Does what I am doing please God" Every other value is second to that one. When that value is in place all other values line up. Matthew 6 is among the most definitive chapters in the bible for shaping a leader's philosophy of life and leadership. Jesus urged his disciples to focus their values on things that would bear an eternal return. But how, while making a living on earth, while responsibly leading an enterprise on earth, while providing jobs, product, service and profit on earth, do we build treasure in heaven.

Matthew records Jesus' primer on values in Matthew 6:1. Jesus focused his lesson in verses 19-21. Do not store up for yourselves treasures on earth, where moth and rust destroy, and where thieves break in and steal. But store up for

yourselves treasures in heaven, where moth and rust do not destroy, and where thieves do not break in and steal. For where your treasure is there your heart will be also.

Bibles quotes about family values:-

Psalms 127:35 "Children are a heritage from the lord the fruit of womb is a reward, like arrows in the land of a warrior, so are the children of one's youth. Happy is the man who has quiver full of them". **Proverbs 17:6** "Children are the crown of old man and the glory of children is their father"

Ephesians 6:14:-

'Children obey your parents in the lord, for there is the right, honor your father and mother, which is the first commandment with promise. That it may be well with you and you may live long on the earth. And you father's, do not provoke your children to wrath, but bring them up in the training and dominations of the lord', Titus 2:4 "they admonish the young women to love their husbands, to their children.

Bible And Discipline:-

Discipline is essential and important for the development of children. As a matter of fact, it is discipline which makes an individual a dynamic, co-operative, sympathetic and tolerant citizen include with social ideas of service and sacrifice at the cost of his own self-interest. Only this sense of discipline creates such conditions in a school which promotes desirable development of a child. In ancient times, too much importance was given to discipline that it was regarded as an end in itself instead of a means only.

For a well adjured life, development of human and moral qualities are essential, because this qualities make a child cultured and civilized for the purpose of leading one's life happily and also contributing to the welfare of society. To achieve this great aim, discipline is very essential. According to Nunn, "discipline is involved in the control of emotions and powers and it in through that we get the ability to work. The aim of discipline is the creation of virtues and development of personality".

Bible also gave importance to discipline. God uses discipline as a last resort. If all god needed to justify mankind to himself was a bigger cattle-prod, then Christ died for nothing. No, god's plan for the earth is love first, discipline last. To get us to listen and repent, he will use deep conviction, the rebuke of friends, coincident sermons, the quickening of scripture, and every other means before discipline us. But if we resist God's spirit of light and mercy. We are promised the rod of correction. When god disciplines, the punishment fits the crime.

Quotes from Bible about discipline:-

Prov. 3: 11-12 :- "May son, do not despise the Lord's discipline and do not resent his rebuke, because the Lord disciplines those he loves as a father the son he delights in".

Prov: 15: 5:-" A fool spurns his father's discipline but who ever heads correction shows prudence".

Job 5: 17-18:-"Blessed is the man whom God corrects so do not despise the discipline of almighty for he wounds, but he also binds up, he injured but his hands also heal"

1 Cor 11:32:-"But when God does judge us, the disciplines us as his own sons, that we may not be involved in the general condemnation of the world".

Prov: 20 : 30:-"Blows and wounds cleanse away evil and beatings purge the innermost being".

Rev: 3 : 19:-"Those whom I love I rebuke and discipline. So be earnest and repent".

Child discipline :-"He that loveth his son, frequently chastiseth him, that he may rejoice in his latter end, and not grope after the doors of his neighbours" (**Ecc 30:1**)

"A horse not broken becometh stubborn and a child left to himself will become head strong" (**Ecc. 30:8**)

"Give him not liberty in his fourth, and wink not at his devices bow down his neck while he is young, and beat his sides while he is a child, lest he grow stubborn, and regard thee not and so be a sorrow of heart to thee" (**Ecc. 30: 11-12**)

"He who spareth the rod hateth his son; but he that loveth him correcteth him betimes" (**Proverbs 13:24**)

"With hold not correction from a child: for is thou striketh him with a rod he shall not die. Thou shall beat him with a rod, and deliver his soul hell".(Prov. 23: 13-14)

1 Pet 4: 17-19:- "The time has evidently arrived for God's judgment to begin, and it is beginning at his own household. And if it starts with us, what is it going to mean for those who refuse to obey the gospel of God? If even the good man is only just saved, what will be the fate of the wicked and the sinner?... Therefore those who suffer according to god's will can safely commit their souls to their wonderful creator, and go on bound up in the heart of a child, but the rod of discipline will drive it far from him".

Prov. 22: 15:- "Foolishness is bound up in the heart of a child, but the rod of discipline will drive it far from him".

Proverbs 13:24 -- "He who spares the rod hates his son, but he who loves him disciplines him promptly."

Proverbs 19:18 - "Discipline your children while they are young enough to learn. If you don't you are helping them destroy themselves".

Proverbs 21:15 - "Children will naturally do silly, careless things, but a good spanking will teach them how to behave"

Proverbs 23:13 - "Don't hesitate to discipline children. A good spanking won't kill them. As a matter of fact it may save their lives"

Proverbs 23:22 - "Listen to your father. Without him you would not exist. When your mother is old, show her your appreciation"

Proverbs 29:15 - "Correction and discipline are good for children" "Foolishness is bound in the heart of a child; but the rod of correction shall drive it far from him" (**Proverbs 22:15**)

Withhold not correction from the child: for if thou beatest him with the rod, he shall not die. Thou shalt beat him with the rod, and shalt deliver his soul from hell (**Proverbs 23:13-14**).

"Correct thy son and he shall give thee rest; yea, he shall give delight unto thy soul "(**Proverbs 29:17**).

Physical discipline in Bible:-

The bible is replete with exhortations about discipline and examples of stiff disciplines allowed or brought by the Lord, in both Old and New Testaments. Some examples;

"Folly is bound up in the heart of a boy, but the rod of discipline drives it far away" (Prov. 22:12)

"Do not withhold discipline from your children, if you beat them with a rod, they will not die. If beat them with a rod, you will save their lives from sheol" (Prov. 23: 14-15)

"The rod and reproof give wisdom" (Prov. 29:15) Discipline your children and they will give you rest, they will give delight to your heart" (Prov. 29 : 17)

"On the lips of one who has understanding wisdom is found, but the rod is for the back of one who lacks sense" (Prov. 10: 27)

"Those who spare the rod hate their children, but those who love them are diligent to discipline them" (Prov. 13: 24).

"The Lord disciplines those whom he loves, and scourges every child whom he accepts" (Hebrews 12:6)

Discipline believes that good behaviour is a teachable skill, not unlike math or reading. Because of this, it makes use of the tools that a good teacher would use. Tools like: good relationship/ rapport building, teaching stories following through with logical consequences, real life examples, personal sharing, redirecting, practice, and giving information in respectful, repeated and varied ways. People who use discipline correctly, do not necessary differ in the number of limits they establish.

Conclusion:-

Values can change the behavior and attitude of every one. Value education is important for our children. And Bible always remains us about values. We can see the different teaching methods and techniques used by Jesus Christ and we can also understand that many of them have great relevance today. There are also some guiding principles to teachers, parents & students. Bible strongly advocates the need for discipline and it is also helpful for our educational system.

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RESEARCH ARTICLE

THE MAIN RESOURCES OF DIVERSITY AND ITS EFFECTS ON THE QUALITY OF WORK LIFE OF HUMAN RESOURCES.

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Abstract

This study aimed at Identifying the impact of the case of functional diversity on the quality of work in the surveyed organization system, and the impact of diversity by gender, diversity by age, Diversity by specialization, and diversity, according to the functional level the stuff on the quality of work in surveyed organization. The study concluded that there is high impact for these variables on the quality of work. It recommends that the bank strategy should take in consideration the unity and balance of the organization, and carry out the requirements of its integration.

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Introduction:-

All organizations, of all kinds, whether governmental organizations or business organizations or civil society organizations, in different sectors of work, if it is agricultural organizations, industrial or service ...it is working hard to be a unified and balanced organizations, so that it meets all components of each organization, and interact together to form a (system)to be integrated and balanced, so it looked like a human body that if one part was suffering all body will be suffering and felt the pain too . The human element is the base for this Organization unity, it is the only vital element in the organization, which gives it's a vitality and governs it's movement, in it the organization life cycle begins to take shape, and finishes when it's finish. Thus, it should develop an integrated strategy for the management of human resources, to effectively manage the organization to achieve balance and united desired. Multiple components that play in any integrated strategy, but the demand to manage the diversity of human resources and the understanding of its effects on the unity of the organization and the elements of its balance, is one of the key components of this strategy. It demands to build a system for the quality of working life and is a major part in it. And that this study will revolve around these two variables, with a focus on the impact of the case of functional diversity on the quality of working life in the Jordan Islamic Bank.

He problem of the study:-

Contemporary organizations concerned with the requirement to build an advanced system to improve the quality of life work, has also become interested in the phenomenon of functional diversity as one of the main variables that affect the lives of organizations, and that the problem of this study centered on the search this matter to answer the following question:

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Is the case of functional diversity affect the quality of working life of the system in the Jordan Islamic Bank.

The importance of the study:-

The importance of the study from a scientific point that it's working on a research influential relationship between two variables and they are considered the most important variables that are keen various of the organizations on their management to manage it effectively, due to their uncertain role in preserving the unity, integrity and balance of organizations. They also have practical importance where the results of it will help the specialized departments, in particular, the human resources management in the surveyed organization to develop strategies in a more rational and effective.

Objectives of the study:-

The main objective in the study is the impact of the case of functional diversity on the quality of work in the surveyed organization system. The ramifications of the following sub-goals:

- Knowledge of the impact of diversity by gender on the quality of work in surveyed organization.
- Knowledge of the impact of diversity by age on the quality of work in the surveyed organization.
- Knowledge of the effect of diversity, according to specialization on the quality of work in surveyed organization.
- Knowledge the effect of diversity, according to the functional level the stuff on the quality of work in surveyed organization.

Study model: based on two variables:-

The independent variable: the diversity of human resources, and the ramifications of several sub-variants, have been some of the sub variables is appropriate exception to the case of the Islamic bank because there is no diversity of this kind, and that as a variable of race or nationality or values, emphasis was placed on four variables are: (diversity by gender, by age, according to the categories of staff (administrators and executives) and according to the functional level (senior management, central and lower). The dependent variable: the quality of working life of the system, and the ramifications sub-variables (working conditions, labor problems, and the system of security and public safety). Source: prepared by the researcher, based on (Sail & Alavi, 2010), (Sing, 2011), (Dessler, 2004) (Assaf, 2012).

Hypotheses:-

The main hypothesis (H01):-

No effect is statistically significant at a moral level ($P \leq 0.05$) for the cases of diversity in human resources in terms of its scopes (gender, age and categories of employees, career level), the quality of work life of the system in terms of its scopes (the conditions of the physical work and the problems of work and the system of security and safety) in the Jordan Islamic Bank.

Sub-hypotheses:-

- The first hypothesis (H01-1): No effect is statistically significant at a moral level ($P \leq 0.05$) for diversity by gender, the quality of work life of the system in terms of its scopes (physical working conditions and labor problems and the system of security and safety) in the Jordan Islamic Bank.
- The second hypothesis (H01-2): No effect is statistically significant at a moral level ($P \leq 0.05$) for diversity by age, the quality of work life of the system in terms of its scopes (physical working conditions and labor problems and the system of security and safety) in the Jordan Islamic Bank.
- The third hypothesis (H01-3): No effect is statistically significant at moral level ($P \leq 0.05$) diversity, according to specialization, the quality of work life of the system in terms of its scopes (physical working conditions and labor problems and the system of security and safety) in the Jordan Islamic Bank.
- hypothesis fourth (H01-4): No effect is statistically significant at a moral level ($P \leq 0.05$) diversity, according to the functional level, the quality of work life of the system in terms of its scopes (the conditions of the physical work and the problems of work and the system of security and safety) in the Jordan Islamic Bank.

Methodology of the study:-

- The study relies on two approaches descriptive and analytical, a case study of the Jordan Islamic Bank. The study includes managers of the bank's branches in Amman community, which is numbering 30 branches, and the study included a survey of all levels of society.
- Questionnaire was designed for the purposes of this study, it has been distributed to all members of the sample, and 24 forms were obtained from the numbers, which was 80%. Also Likert scale has been used; it was an appropriate statistical analysis to test the hypotheses of the study.

Literature:-

Managing the diversity in human Resources:-

All organizations, of whatever type, is working hard to be a unified and balanced organizations, to integrate all the components of each organization, and interact together to form both the (system) integrated and balanced. (Kehinde,2012) .The human element in any organization considered the single vital element which gives it vitality and governs its movement. As the human resources by nature is characterized by diversity and difference, its presence within any organization will ask an important question about the impact of this diversity or differences on the unity of the organization and the elements of balance, and the performance and effectiveness levels, and this is what forces the organizations to take the necessary degree of attention, and to include it within their strategies. (Noori,2013). Have multiple definitions offered by scholars of the concept of diversity despite the clarity of the concept which is inherently in it, it does not provoke any controversy around the researchers to try to clarify the truth, diversity is the variety and differences in the types of human resources, and its degree, according to each organization and what are the sources that they get their human resources, and we do not have to define the axioms. The distinct and more voluminous organizations and geographical spread, especially cross-border and multi-national organizations, the more diversity is more apparent, it has helped the phenomenon of globalization, and the phenomenon of complexity and entanglement in international economic and political relations, the proliferation of such organizations. (Richard,2008) (Nasyira,Othman,&Gazali,H ,2014).

Cases of the diversity of human Resources:-

First, the diversity of gender ; this has become a source of the most common causes of diversity in various organizations, without exception, even the military and security ones, as a result of the expansion of the entry of women into the work field, and markedly and growing, due mostly to: (Polk,2011,167),(Suhasini&, Naresh, 2013).

- The growth of public awareness of the heads of families in different communities, especially in Arab societies, as a result of the spread of education among them, the need to give girls opportunities in education, in response to the imperatives of social life on the one hand, and the evolution of consciousness and religious understanding, which focuses on the importance of girls' education.
- The general sense of the growth of the necessity of equality between men and women, especially in front of education and employment opportunities, has become the general legislation in most states which imposed it. International pressure operated global trend organizations which they have supported it.
- The pressure of economic life, and the weight of the economic burden on many families, prompting the woman to go and female heads of households and their daughters to enter the practical life to help men to provide the growing needs of the family members.

There is no doubt that the differences are organic and differences between males and females are differences (Congenital), are present at the origin of the creation of all of them and that it has direct effects and remark on their behavior in public life and working life, and that is necessary for each organization to put these differences into account, and what is most important can be referred to as: (Zikmund,2000,54),(Ichi, , &Arthur, 1984,68-74)

- distinguish between jobs that require physical efforts and capabilities, and guide the male workers to it because they are characterized as having high capabilities more than females in this area.
- recognize the high degree of emotional transparency among females than males, and by taking into account the situation of women and not to put her in a decision-making positions, which are constantly under pressure from the centers of power in the organization or community. If it is necessary to put women in certain positions of leadership, it is important to have a number of male advisers who are able to support in any situation like this.
- Note some of the characteristics that can be characterized by the female often, that they do things in details, and have ordinal and protocol capabilities, and so on, so you can take advantage of the functions or activities that they do.

Second, the diversity of culture; where is the culture in the organization is the most important factors that affect the unity and balance of the organization, if the organization has a clear and unified culture, and workers with different functional levels stick to it, and to be organized at their best, then it will be cohesion and balanced. But if there were cultural differences among workers, and is rooted in their consciousness, and visible in practice, this is one of the most dangerous things that can be faced by the organization, and inevitably leads to disagreements and conflicts that threaten the unity of the organization and perhaps its existence as well. (Chuai, 2008) .The concept of the culture is important, which needs to clarify and be determined, when we follow up on the most important definitions provided,

we can offer an integrated definition which defines culture as the outcome of three basic systems of interaction : (Assaf, 2012,265):

- System prevailing values
- System prevailing public knowledge
- The skills and expertise which include customs, traditions and tools, rituals and symbols and techniques and means. that prevail in the life of the community or organization system.

Cultural diversity challenge in most international organizations has become a reality case, where these organizations are spread and have hundreds of branches around the world, and attracts thousands upon thousands of workers from different cultures, and this is what is in fact the most important factors that should be on the public authorities of any such organization, be taken into account, and that the design is suitable to make a bigger deal of cultural integration, and the development of the spirit of citizenship functional strategies, and without it threatening the unity of the organization, and more likely failures. (Drucker, 1998,165-172).

Third, the diversity of nationalities; due to global openness among many regional organizations or cross-border, as well as the many local organizations in some countries that rely on imported labor from abroad, as in the case of the Gulf countries, and some other oil states like Libya and Algeria. As it happens for most major industrial countries, which amounted to the degree of expansion of their organizations to import manpower from abroad. The diversity of nationalities represents one of the main sources of cultural diversity in many cases; in addition to that it may represent a source of some of the conflicts that may arise as a result of the existence of political sensitivities, economic or ... between some of the countries to which it belongs. The organizations that are monitoring these cases and practices, and put suitable strategies to manage this diversity and overcome the disadvantages, and take advantage of it to greater vitality to its life and operations.

Fourth:- The diversity of races and colors; in turn its reflected in the cultural diversity on the one hand, it also represents one of the major factors of difference and disagreement, and perhaps conflict within organizations, especially if it has been associated with the multiplicity of nationalities, or social and political sensitivities. That the organizations be aware of the truth of this matter, and take him into consideration which put its strategies to build a united and balance.

Fifth:- The religious and sectarian diversity; and it is considered the most dangerous sources and the most important events in the cultural diversity, and it set on the flame of the rivalry between the workers and to the degree it may sometimes be devastating. It is true that organizations can paint a functional policy which abolish the status of religions and sects, but it is important to give it a constant attention, and concern for the lack of full openness to diverse sources of religiously and ideologically as much as possible, because in the case where the religious and sectarian rivalry, if happened it will become out of control, and difficult to be contained, and it threatens the life of organizations actually.

Sixth:- The diversity of ages; the wide gap between the ages of workers in any organization lead undoubtedly to the disparity in scores of experience, and functional and professional expertise, as reflected in the form of variations in the level of general wisdom, and in the degree of flexibility and personal capabilities to bring about mutual understanding and interaction between workers of all levels horizontally and vertically, may lead to a kind of generational conflict within organizations. The organizations that took this issue a keen interest and designed work strategies, any organization is undoubtedly in desperate need for the elderly and their experience and wisdom, as it is in definite need for new generations and their knowledge, skills and enthusiasm, and thus become a required to paint scenarios needed to bring about integration and knowledge among all workers which have different ages.

Seventh:- Functional diversity itself; any organization has had the category of administrative or white collar stuff, also has executives and normal class workers or blue collar workers, there is no doubt that there is a significant gap or legal and psychological between these two categories, and should not be ignored, and the development of appropriate formalities, and appropriate guidance, to reduce its size and to avoid effects. (Wilson, 2014,68-74). Also up in the functional diversity among the category of managerial and professional category within organizations, and it's important to the organizations authorities realize the truth of that and its importance, and put all the formalities needed to bring interactive and complementary relationships between these groups and guidance. Administrators should realize the importance of technicians for the life of the organization, and they're part of an integral part, and they are the most important elements of the organization's success in its operations and to achieve its objectives.

Technicians also must understand the role stature and importance of administrators who do all administrative arrangements that help technicians on their success, and that they (professionals). And do not aspire to administrative positions that if they have succeeded access to it, leading to disruption of artistic energies on the one hand, and they are not expected to succeed in their administrative roles because of the elements of personal and technical skills that they have differ from the administrative personal elements. (Worrel & Cary, 2012.76-77) .As well as functional diversity among the category of managers and the category of experts and advisers, and the possibility of escalation of the conflict between them sometimes, this is reflected on the organization negatively. The organization responsible authority required to put the necessary regulatory scenarios for the development of each category in the proper context, so as to convince the advisers and experts that the work of administrative differs in terms of success on their work, and therefore, on these experts and consultants to focus on the tasks required of them in all honesty (consultant is - intimate), and does not aspire to management positions that may fail miserably if it were transferred to them. Well it is necessary to convince managers that they need the expertise of experts and advisers who are without their advice, and knowledge and experience; they may not be able to make decisions effectively.

Finally, functional diversity and differences between the Central upper levels of management and administrative levels and lower levels, this diversity may result in many organizations to the outbreak of the manifestations of hatred and personal intrigues, and manifestations of espionage and absent ... etc. Especially if there was a large and spacious gaps between the levels of compensation that workers get in each level of them.

The power authority in the organization should take care of this matter, and to estimated the severity of it, and put administrative and financial formalities necessary to minimize the negative effects and containment.

8-2 the quality of work life:-

It is not considered modern in the quality of work life or new in the management thought, and administrative practice, as some researchers may think, but it is due in fact to the model liberal-west in the mid-thirties of the last century with the surfacing of human relations school , and the escalation of this attention with the rise of behaviorism school in the fifties of the last century, which grows until it reached the limits of its peak following the emergence of Total quality management School at the beginning of the eighties of the last century, until now.

The concept of quality of work life:-

Regardless of any definitions may be presented by some scholars here or there, it could be argued that this concept of sufficient clarity that does not require to make hardship in order to identify the vocabulary of it, and if it was necessary to enter into this subject, it should be recognized that determining this concept needs to consider the following: (Wilson, 2014.86-87).

- overview of all work life so that it is taking into account all the components of life, whether these components related to the internal environment of the organization, or affecting the external environment.
- focus on quality necessary to improve all environmental components related to the life of the organization requirements, as a prerequisite to improve human resources in the organization, and make it truly the real capital in it.

Thus, the quality of life can be defined as the work situation which reflects the improvement or development of all elements of the organization and the foundations of all environments to ensure the provision to adapt the necessary atmosphere for human resources and resettlement in the organization, and to enable them to be the true capital of it.

Core areas of working life:-

Varied and multiple attention should be paid out to improve the lives of work in different organizations elements of the environment, and the most important thing that should be taken care of and focused on:

Improve basic working conditions:-

The basis for the improvement of the various working conditions on the demand aims at adapting the material elements of work with the needs of workers and material and moral circumstances and not vice versa. Dealing with the working conditions and development is supposed not to rely on the fact that workers are characterized by flexibility and speed of adaptation. But we must make the other elements are flexible and compatible with the requirements and expectations of employees and not inconsistent with them, and the most important conditions that must be observed.

- a. light intensity; It has been proved experimentally that the increase of light does not lead to increased productivity, and that there is a critical lighting degree of increase then stop point, this with the need to take into account individual differences among workers, some of whom needs severity of high illumination to works effectively, and some of them needs the intensity of lighting to be moderate, and so on ..., but the normal situation is that there are a critical lighting one minimum and the other is high point, and that productivity negatively affected if the degree of intensity of illumination less than the minimum, and it is fixed if above the maximum, it has been adversely affected in some exceptional cases. (Assaf, 2012,265-272)
- b. Days and hours of work; (William Martar) conducting an experiment on the number of hours of work and its impact on productivity he found that the reduction of working hours has led to increased productivity and gave workers important opportunities for reflection and self-development and development of other work items. But reducing the number of weekly working days effect was present, but less degree of reduced working hours. (Assaf, 2012,267-274)
- c. periods of daily comfort; experiments on the effects of rest periods has shown the importance of human relation during work, and the necessity of having breaks during work, it reactivates workers, whether their work that they carry out heavy and stressful, or boring and stressful psychologically. The breaks should not be multiple which leads to cutting business continuity and preparations for its implementation, and long breaks leads to inaction and lack of enthusiasm to return to work. (Morshal, 2012,56-65)
- d. Ventilation, temperature, humidity and cold conditions noise and dust ... etc. All the factors that affect the impact, stressing the sense of working comfort and satisfaction, and it is a must for any organization that is keen to ensure the safety of its facilities and arrangements to have all this, in an effort to provide a good working environment as a requirement on one hand, and the demand for motivating employees and raise their performance levels on the other hand.

Overcoming the problems:-

Studies suggest that problems of work have a prominent role in the increase of accidents at work, says one study, 95% of work-related accidents were caused by human error, and 5% were by machines, as was proven by the study that there is a relationship between the state of mental and physical of the worker and accidents before they happen. (Assaf, 2013), and the most important problems that can be referred to:

- a. The problem of fatigue or stress; an experience shows that the patterns of social and psychological characteristics that could lead to stress and fatigue as the physical fatigue, studies have indicated that the cases of physical exhaustion less common in modern organizations than the psychological exhaustion. Some scholars say that the solution to fatigue problems do not have to remove fatigue completely by rest periods or other procedures within the organization, but to take the necessary steps to keep the fatigue at minimum before it happens, and this requires accurate studies, and the creation of appropriate working conditions.
- b. the problem of boredom; some scholars believed that boredom is not as a result of physical causes, but rather is the result of psychological reasons, and occur as a result of fragmentation detailed work, which makes the worker feels futility and irrelevance, and leads him to boredom and then the physical and psychological stress.
- c. To address the cases of boredom can change the work type of the employee from time to time, and develop a sense of the importance to the workers who do simple jobs and repetitive it is their roles that they do that make the organization to continue and elevate (Great job), as well as the development of the case of job stability, and granting appropriate wages, and to provide appropriate break periods, etc.
- d. The problem of absenteeism and turnover work; highlights the absences as a problem when repeated markedly, and when top managers have doubts that absences did not have compelling excuse, but rather the negative feeling towards the business or organization.
- e. One study has shown that rates of 25-33% of absences due to neurotic diseases may be already exist among workers, and appeared while working out in unsuitable conditions, or as a result of the organization's policies concerning them. (Assaf, 2012,266-274)
- f. The problem of incompatibility or functional adaptation; adaptation intended to find a balanced and acceptable relationship between workers and their work environment with all its components. The consensus is that each of the organization and employee convergence between each other to achieve a balance between their needs and objectives. It was the technological developments and its accelerated entry into the life of organizations, especially the computer revolution and related technical developments and administrative, which has a big impact and made a wide gap in the relations between workers and organizations, and vice versa, thereby exacerbating adjustment and functional compatibility issues needed and it is the most important thing to be

recommended to provide an adequate amount of the balance of working and supporting the case of functional adaptation: (Parakash, 2015), (Wilson, 2014,112-116)

- The development of the organization's preparations for compatibility with the interests of workers and their goals, and to highlight that in the rules and regulations prescribed by the management of their employees, and to develop policies and practices that actually confirms it.
- Promote the spirit of social and collective solidarity with the workers through cultural and educational programs, and through some of the practices that are programmed for this purpose.
- The design of technical programs for the use of computer systems and related support systems, skills, and educational and behavioral programs to promote new systems of relations between the workers themselves, and between them and the organization, in line with the imposed technological developments of relations.
- Pursue advanced career and objective methods for selecting and directing careers, for the person who is chosen to work to be psychologically and professionally appropriate to the requirements of the work that he will exercise when his appointed.
- Take into account individual differences among workers, and try to deal with all of them in accordance with the nature of his personality, and the system needs.

Security management and occupational safety:-

There is no disagreement about the concept of occupational safety and security, and it is known as expressing some most important areas which are looking at the ways to protect workers in the organizations in which they work for, from the effects resulting from all existing environment elements of each organization of risk factors mechanisms, and lead to health effects, directly or indirectly on the safety of workers and their health. This is in addition to the protection of the property of the various organizations, facilities, reputation and position in the market and society. (George & Weimerskirch, 1998). It was the industrial revolution which caused the emergence of this issue to take its place and its importance, especially in the industrial organizations, which have many risks in the workplace, mechanical, electrical, chemical and physical, etc., which cannot be ignored by any organization or neglected, and it must manage it to ensure the provision of safe and healthy working environment. It is sufficient demonstration of the importance of this subject to the annual Statistics of the International Labor Organization (ILO) show that there are about 115 million workers in the world are exposed annually to accidents and injuries, of which about a variety of work (180000) injury leading to death. (AlAkailah, 2011,17-24). It was the first established law by the nations to protect the employees from the dangers of the environment of work is in Britain in (1802). The interest increased with the founding of the International Labor Organization at the end of the First World War, which became to take care of this matter. Then the formation of the Arab Labor Organization as one of the Arab League specialized organizations, issued Convention No. (7) of the occupational safety and health, and put obligations on the Arab countries in order to achieve protection for workers from exposure to any injuries of work or occupational diseases, as stipulated by the Convention No. 13; the need to improve the working conditions of physical, chemical and biological, technical and social (AlAkailah, 2011,16-29). Security management and safety functions: The main objective of this department is the protection of persons, individuals or groups, of any risks, which revolves around the functions of the department concerned, in addition to: (Sheraz, Salid, Quresh, & Rizwan, 2014)

- Protection of buildings, facilities and equipment and various equipment from the risk of damage or theft or other threats and risks.
- Conservation and protection of raw materials during storage or use during the production process, as well as the conservation and protection of materials when processed and stored to avoid any potential risks.
- Measuring the effects of all chemical and biological, electrical and lay the foundations for preventive basics, and build a foundation for complete information about all the environment of the organization, and to benefit from all of this in a safe and disciplined work environment, according to the required standards of prevention.
- take care on the highest levels of mental health and physical and social, this requires the activation of legislation, regulations and procedures and technical and administrative arrangements in forced by the organization. And design training programs that build the skills of workers in the organization.

Organization environment risks: each organization is keen to be environment-friendly and all occupational safety and health conditions are secure, in order to avoid the most important risks: (Kakkos, 2011) (AlAkaileh, 2011,26-54)

First, the physical hazards, including noise, humidity, ventilation, heat, cold, luminescent radiation and atmospheric pressure, and others.

Second: chemical hazards, including the risk of toxic effects, and the effects of irritants or allergic or that which lead to distortions on birth or mind or otherwise.

Third: mechanical risks that come as a result of the wrong operation of machinery and equipment and various tools. Or as a result of error in carrying the loads and the method of downloading and others.

Fourth: the risks of construction, which relates to the fall of the materials or falling from scaffolding or cranes, or as a result of falling into the excavation or the result of some fires .. and so on.

Fifth: human risks, that stems from the weakness in some senses like smell or sight, or produced as a result of the psychological conditions of workers and their feelings of stress physical or moral, or the result of carelessness or negligence, and so on.

Sixth: the risks of mismatch between the employee and the work for any reason, which leads to the situations of stress and increase job errors and risks.

Seventh: biological risks: which result from the spread of harmful viruses and infectious bacteria, parasites and fungi.

Eighth: electrical hazards: caused by any errors in the use of machinery, equipment, fixtures, that uses electrical power.

The responsibility to improve security and safety conditions: the human resources management, in particular units responsible for the subject of security management and occupational safety, required to manage a system for the basic relations between all parties of the production processes in the organization in order to accomplish a number of important responsibilities, namely: (Ibrahim, 2014) (Kakkos, 2011)

1. The responsibility of the organization to develop an integrated and clear system of security and safety.
2. The responsibility of continuous coordination with the higher authorities in the organization, so as to develop the rules and regulations and issue decisions and instructions appropriate and necessary for their implementation.
3. Responsibility for coordination with suppliers, in order to determine where the source of the danger in all this, and what are the possibilities of the risks, and the preparation of evidence to show these things, and the definition of hazardous substances and their characteristics, the design of catalogs showing ways to use and contribute to the training of personnel on the use and operation. And determine what contingency plans in the event of a particular incident
4. Responsibility for coordination with the workers, so as to identify training needs of all equipment, tools and techniques used, and identify training plans aimed to explain to the workers the operating and maintenance details, and the development of operational skills, and familiarize them with the tools of prevention, systems and procedures. It is responsible for coordination with the various trade unions in order to approve the necessary protection systems for workers with all the details of that technical and procedural.
5. The responsibility of continuous coordination with the administrators of safety and security so as to ensure they are doing their duties in the best way possible, and to develop plans from their knowledge and skills.

Security and safety of workers system: There is no doubt that the protection of employees and providing good conditions for their security and safety, is one of the most important arrangements of any organization, and makes them at the head of the organizational and administrative priorities, and that their the absolute goal is to be zero accidents, because if any breaches of the cases of security or integrity happened, the results could be disastrous, notably: (Sing, 2011) (Parakash, 2015).

1. The humanitarian consequences that come as a result of injury to any of the workers with some work-related injuries that may cause him bodily harm leading to death or partial disability or permanent, which will reflect the occurrence of psychological and social effects on him or his family and relatives.
2. Social consequences for the society as a whole from some incidents of disabling some of the forces operating in it, and what consequences on their families from its effects.
3. The economic consequences of the organization as a result of any incidents that may be damaging to its assets or property or workers or the general population and the environment surrounding it.

4. Legal consequences that may result if the argument broke out between the organization and employees affected or their families about their compensation for damages for their losses as a result of exposure to accidents at work, which is reflected on the reputation of the organization and its place in society.

That the organization must design an integrated system of security and safety which is one of the most important prerequisites that must be provided, and that the human resources management in the organization must working to verify the construction of this system, and discuss the elements and the specificity with the makers of the design mode. Security and safety system may differ from one organization to another depending on the nature of the work of each organization, its size, and its technologies. There is no doubt that the greater the technical nature of the Organization, and the Organization of substantial size, advanced technology, the degree of risk in it is high, security and safety system would grow more complex and important. Add to it that whenever there was high competition between the organization and some other organizations, the more job security considerations are needed, especially those related to information security and the protection of confidentiality. Ranging safety and security system components, according to the degree of complexity, and the areas it covers, and the most important thing we must focus on:

(A): The system vocabulary and the fields, the system should cover all aspects that could be breached more or less on the state of security and safety within the organization or outside, in particular the state of safety and security for their workers, or the people in the surrounding community, and the most important thing should be covered by the system namely: (AlAkaileh, 2011,55-155) (Siddique, Fatima & Khan, 2013)

- A comprehensive statement identifies all sources of risk, ranging from the highest sources of risk down to the least dangerous.
- Build expectations of environmental risk sources, both with regard to the occurrence of earthquakes or floods, or other things that can happen and has an impact on the organization and its staff.
- Determine the degree of risk entailed in each of these sources, and determine the risk involved in this, and affected stuff.
- Identify the times that have higher degree of risk than others, and the system should identify a comprehensive disclosure list of all sources of danger and times of occurrence of each risk, and the degree of risk, according to each case, all the time.
- Identify areas of risk within the organization, and draw a clear and comprehensive map showing all that, with the identification of risks for each place, and what is the degree of the risk.
- Identify sources of external threats that are related to the organization, and determine the degree of seriousness of each of them, particularly as regards to opposition organizations, or the culture of the surrounding community and the likelihood of burglary, robbery or revenge and sabotage ... etc.

The system processes and procedures, which are carried out by the risk management within the organization and outside, and consists of two-fields: (Moshal, 2011,65-78) (ILO, 2014).

- The first part: a precaution; includes all policies and regulations, tools and procedures that must be adhered to by the employees in the organization at all levels, and the most important as follows:
- Build a comprehensive system of security and safety records include all incidents involving the organization, and what are the effects and results, to represent documentary reference which can be referred to it when necessary.
- The design of criteria that include everything related to the general structure of the organization; electrical and mechanical systems, sewage networks, computer networks and systems. It also includes all the work of the organization and operations of every detail necessary to ensure the security and integrity of the organization and its staff and procedures.
- The provision of supplies for the security and safety system, taking into account that they are consistent with the safety and security specific standards, with the need for continuous update.
- Identify all the actions that are working out in case of any emergency, and circulated to all relevant personnel, and train them to do so in an orderly and accurate way, according to specified criteria.
- Build a system for monitoring and inspection, which guarantees and ensures verification of compliance with all policies, standards and requirements and the specific arrangements and procedures.
- Review of the items of the system as necessary to keep pace with the various developments.

The second part: Therapeutic; in the event of danger for any reason, the security and safety system should include a comprehensive design and actions that must be taken to counter the threat and treatment and reduce its effects, and the most important thing must be contained in the system:

- build your own risk occurrence of expectations, and its causes and degrees of damage that result from each of them, and a system of priorities for dealing with it.
- Identify the arrangements and requirements and procedures that must be applied in every case of danger, and make sure that it is ready constantly.
- Monitor a special budget (risk budget), so the organization will not find itself unable to finance any danger that it may be exposed to.
- Conduct audits of vocabulary of the system and arrangements from time to time and again, and develop what needs to be developed, and add or delete what needs to be added or deleted.

The priorities of safety and security system; it includes priorities of following models; (Parakash, 2015).

First priority: The imminent risk, which confirms the high expectations on the probability of occurrence soon, if happened it will cause great harm to the organization or employees. And you must determine what these risks, and classifying them according to the degree of impact and the expected times of occurrence.

Second priority: large risks related to disasters and accidents that result in high-risk and negative results, determine what these risks are and classify them according to the degree of impact and the expected times of occurrence.

Third priority: the risks of non-compliance with the security standards and safety, or inputs, systems, regulations and technical and administrative specified procedures.

Fourth Priority: small risks relating to certain natural faults that are bound to occur in some of the equipment and techniques or electricity networks and various utilities.

Fourth Priority: The requirements for the system to succeed: it must provide a set of requirements that lead to the success of the system, at the corporate level or the staff level, or the surrounding environment and the surrounding community level. The most important of these requirements (Worral & Cary, 2012,67-69):

- That includes a comprehensive list of the most important reasons that could lead to an accidents or certain risk of the system, and work to develop solutions.
- All levels of the organization's management should be committed to the system and conditions.
- The training of all employees on the implementation of the system and all its standards, arrangements and procedures, and work to make the process of how to arrange of the applications in the event of any accident.
- Identify and provide all the necessary requirements in the event of any accident, and to ensure availability and made available upon request at the time, and work to check it regularly.
- To provide job security and safety nets, health insurance or social security, or the policies of job stability, and make sure they take into account the working conditions, according to each organization, and take into account the workers professional and living standards.
- Design a code of honor to work, focusing on forbidding the abuse of drugs and alcohol and similar substances, and not to violate ethical norms accepted.
- Comply with the regulations imposed by the trade unions technical and administrative.

The results of the study:-

First, it was a multiple linear correlation tests to ensure suitability of the study model, and the results were as follows:

Table 1:- multiple linear correlation test.

Sequence	variation	VIF inflation rate variables
1	diversity by gender	2.767
2	diversity by age	3.336
3	diversity, according to functional category (administrators and executives)	2.155
4	diversity, according to the functional level (senior management, middle management and lower)	1.988

As noted, the results of each variation inflation is higher than one and less than ten, underscoring according to (Gujarati, 2004,352) there is no problem between the variables of the study.

Second:- A test of autocorrelation was conducted (Deran- Watson test) to ensure the absence of data from the autocorrelation problem, and the results were as follows:

Table 2:- autocorrelation test

Hypothesis	value (D-W) d calculated	(dL) minimum	upper limits (du)	result
Ho1-1	1.803	1.758	1.779	No self link
Ho1-2	1.812	1.758	1.779	No self link
Ho1-3	1.852	1.758	1.779	No self link
Ho1-4	1.905	1.758	1.779	No self link
Ho1	1.889	1.728	1.809	No self link

As can be seen, all the values of (d) outside the limits specified in the standard values (dL), In (du), which confirms the absence of evidence of autocorrelation and validity problem in the use of the regression model.

Third:- hypotheses test; was conducted by analyzing the simple linear regression and gradual, and the results were as follows:

(1) test the first sub-hypothesis (H01-1), and the result was as follows in Table (3)

Table 3:- impact test results by age diversity on the quality of working life

Coefficient					ANOVA			Model Summery		The dependant variable
Sig t level of significance	T calculate d	standar d error	B	statement	Sig F	Df degree of freedom	F calculate d	r ²	R correlation coefficient	
0.000	14.763	0.038	0.567	polarization	0.000	1	217.945	0.479	0.692	quality of work life

As can be seen, the value of the correlation coefficient (r) between the diversity variable by gender and variable quality of work life is positive, high-value, according to the standard (Zikmund, 2000, 513) as can be seen that, the coefficient of determination (r²) has interpreted the role of the state of diversity by gender in the level of variation (47.9%), with all other variables remain constant. Also, the value of (F) has confirmed a significant gradient at the level of less than 5%, at a confidence level (Sig = 0.000), as well as the rest of the results confirm a significant coefficient at least 5%, and this leads to the rejection of the hypothesis Sub-first, and to accept the hypothesis (that there is a statistically significant effect in the abstract level of equal to or less than 5% of the state of diversity by gender on the quality of working life of the system in the Jordan Islamic Bank).

(2) The second sub-test the hypothesis (H01-2), and the result was as follows in Table (4)

Table 4:- impact test case diversity results by age on the quality of working life of the system.

Coefficient					ANOVA			Model Summery		The dependant variable
Sig t level of significance	T calculate d	standar d error	B	statement	Sig F	Df degree of freedom	F calculate d	r ²	r correlation coefficient	
0.000	16.496	0.036	0.595	resettlement	0.000	1	272.123	0.534	0.731	Quality of work life of the system

As can be seen, the value of the correlation coefficient (r) between the state of diversity variable by age and variable quality of work life is (73.1%), a positive, high-value, according to the standard (Zikmund, 2000, 513) as can be seen that the coefficient of determination (r^2) has interpreted the role of the state of diversity by age in the quality of work life standard amount of variation of 53.4%, with all other variables remain constant. Also, the value of (F) has confirmed a significant gradient at the level of less than 5%, at a confidence level (Sig = 0.000), as well as the rest of the results a significant coefficient at least 5%, and this leads to the rejection of the hypothesis sub-second, and to accept the hypothesis (that there is a statistically significant effect in the abstract level of equal to or less than 5% of the state of diversity, according to a variable-age on the quality of working life of the system in the Jordan Islamic Bank).

(3) Test the hypothesis Sub-third (H01-3), and the result was as follows in Table (5)..

Table 5:- the impact of the test case of diversity, according to the results of the functional groups on the quality of working life of the system:

Coefficient					ANOVA			Model Summery		The dependant variable
Sig t level of significance	T calculate d	standar d error	B	statement	Sig F	Df degree of freedom	F calculate d	r^2	r correlation coefficient	
0.000	15.604	0.035	0.547	Investment	0.000	1	243.485	0.507	0.712	Quality of work life system

As can be seen, the value of the correlation coefficient (r) between the state of diversity variable depending on the functional groups on the quality of work life is a system (72.1%), which is a positive, high-value, according to the standard (Zikmund, 2000,513) as can be seen that the coefficient of determination (r^2) may be interpreted the role of the state of diversity, according to categories of staff in the quality of working life system, standard amount of variation (50.7%), with all other variables remain constant. Also, the value of (F) has confirmed a significant gradient at the level of less than 5%, at a confidence level (Sig = 0.000), as well as the rest of the results confirm a significant coefficient at least 5%, and this leads to the rejection of the hypothesis Sub-third, and to accept the hypothesis (that there is a statistically significant effect in the abstract level of equal to or less than 5% of the state of diversity, according to the functional groups on the quality of working life of the system in the Jordan Islamic Bank).

(4) Test the hypothesis Sub-fourth (H01-4), and the result was as follows in Table (6).

Table 6:- the impact of the test case of diversity, according to the results of the functional level on the quality of working life of the system.

Coefficient					ANOVA			Model Summery		The dependant variable
Sig t level of significance	T calculate d	standar d error	B	statement	Sig F	Df degree of freedom	F calculated	r^2	r correlation coefficient	
0.000	17.032	0.037	0.639	preservation	0.000	1	290.087	0.550	0.742	Quality of work life system

As can be seen, the value of the correlation coefficient (r) between the state of diversity variable depending on the functional level and the variable quality of work life in a system (74.2%), which is a positive, high-value, according to the standard (Zikmund, 2000,513) as can be seen that the coefficient of determination (r^2) may be interpreted the

role of functional diversity, according to the functional level in the quality of work life of the level of \$ 55.0% variation, with all other variables remain constant. Also, the value of (F) has confirmed a significant gradient at the level of less than 5%, at a confidence level (Sig = 0.000), as well as the rest of the results confirm a significant coefficient at least 5%, and this leads to the rejection of the hypothesis Sub-fourth, and to accept the hypothesis to become (that there is a statistically significant effect in the abstract level of equal to or less than 5% of diversity, according to the functional level on the quality of working life of the system in the Jordan Islamic Bank).

(5) test the hypothesis President, and to identify any of the sub-variables most impact in the quality of life of work system, and has been so for a multi-tiered regression testing, the following table illustrates this:

Table 7:- a summary of the model to test the main hypothesis.

Model summary form				Model
coefficient of determination	standard error	The coefficient of determination	R correlation coefficient	
0.504		0.550	0.742	1
0.435		0.667	0.817	2
0.416		0.697	0.835	3
0.409		0.707	0.841	4

Table 8:- analysis of variance to test the main hypothesis.

Sig F level of significance	F calculated	D f degree of freedom	sum of squares	model	
0.000	290.087	1	73.682	Regression	1
		237	60.198	Residual	
		238	133.880	Total	
0.000	236.138	2	89.270	Regression	2
		236	44.609	Residual	
		238	133.880	Total	
0.000	179.769	3	93.247	Regression	3
		235	40.632	Residual	
		238	133.880	Total	
0.000	141.181	4	94.657	Regression	4
		234	39.222	Residual	
		238	133.880	Total	

Note that the data in Table 7 shows that the first model for gradual regression analysis shows that diversity variable is depending on the functional level and may be interpreted accounted for 55% of the total variation in the quality of work life, and become ratio 66.7 when you add diversity variable by age to diversity variable depending on the functional level, the ratio becomes 69.7 when you add diversity to a business class, for up to 70.7 by adding diversity by gender. While noting that the value of the levels (F) in all the different models within the abstract level (Sig = 0.000) as shown in Table (9) as follows:

Table 9:- the results of regression coefficient is a summary to test the main hypothesis

Sig significance level	t calculated value	Std.Error	B	quality work life	Model
0.000	17.032	0.037	0.639	diversity, according to the functional	First
0.000	9.680	0.042	0.402	diversity, according to the functional	Second
0.000	9.081	0.039	0.357	Diversity by age	
0.000	7.795	0.042	0.331	diversity, according to the	Third

				functional	Fourth
0.000	6.013	0.043	0.258	Diversity by age	
0.000	4.796	0.041	0.194	Diversity, according to the functional level	
0.000	7.020	0.043	0.302	diversity, according to the functional	
0.002	3.167	0.053	0.167	Diversity by age	
0.000	4.851	0.040	0.194	Diversity, according to the functional level	
0.004	2.900	0.048	0.140	Diversity by gender	

By observing these results, we find that the values of (B) in the two models at levels (t) is different at the significance level (Sig = 0.004) or less, and this confirms a significant regression co-efficients, which confirms that the impact of these variables in the models statistically significant. Accordingly, we reject the premise of it and accept the alternative hypothesis that (There is no impact statistically significant at a moral level $P \leq 0.05$ cases diversity of different dimensions (gender, age and job category, career level) on the quality of working life of the system in terms of its various dimensions (working conditions and labor problems and the system of Security and safety) in the Jordan Islamic Bank.

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RESEARCH ARTICLE

HUMAN RESOURCES INFORMATION SYSTEMS AND THEIR IMPACT ON THE EFFECTIVENESS OF ORGANIZATIONAL CHANGE MANAGEMENT-APPLIED STUDY ON THE JORDAN INSURANCE COMPANIES.

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Abstract

The aim of this study is to investigate the effect of human resources information systems on the effectiveness of organizational change management, applied study on Jordanian insurance companies. The study concluded with the following results: there is a correlation relationship between the training on system and the effectiveness of organizational change management, the training on system explained 19.9% of the variation in the effectiveness of organizational change management, the availability of technical support explained 37.8% of the variation in the effectiveness of organizational change, the availability of technical support explained 55.1% of the variation in the effectiveness of organizational change, the availability of technical support explained 57.8% of the variation in the effectiveness of organizational change. The multiple regression results showed that training on the system has no significant effect on the effectiveness of organizational change, while there was a significant effect of (availability of technical support, senior management support, availability of infrastructure) on the effectiveness of organizational change. The study recommended with the following: develop the capacity of workers and trained them on human resources information systems requirements, put a fair system of incentives to encourage innovation and creativity process, develop infrastructure and technological development, and there is a need to be updated with the developments existing in the world, update software and deal with the reliable one, benefit from the experiences of other insurance companies regarding human resources information systems, management of change resistance and use scientific means.

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Introduction:-

Organizational success and management of the knowledge economy is heavily dependent today on the performance of Human Resources Management (HRM) (Lippert and Swiercz, 2005; Troshani et al, 2011). The human resources management (HRM) recently changed to focus on the exchange of knowledge and strategic analysis of the workforce, and its contribution significantly developed widely in the strategic management of the organization and adaptation with the surrounding forces of change that have high impact on the management of its resources and

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maximize the return of them (Rodriguez and Ventura, 2003; Troshani et al., 2011). Therefore, the organizations with various forms seek today to take advantage of all what possesses of resources, potential and abilities to face the developments and challenges that determine their relationship with its customers and affect its position in the market, so there is a new direction to increase the effectiveness of the management of human resources by relying on human resources information systems (Ball, 2001; Lippert and Swiercz, 2005; Troshani, et al., 2011). To do so, the Jordanian insurance companies must use human resources information systems in order to adapt with the surrounding forces of change, and to achieve and build competitive advantage, and this can provide quick access to the customer and meet his/her expectations.

Problem Statement:-

The change management in Jordanian Insurance companies is one of the most important research areas, as it is a tool that affects the reasons for its survival and development of its ability to adapt with the surrounding environmental forces that have effective impact on their ability in leading markets. The human resources information systems interest is one of the strategies that must be taken into account in this aspect, the researcher thinks that there is a lack of awareness in the role of human resources information systems in the organizational change management in the Jordanian insurance companies, so they must develop their human resources systems in line with the size of change that occurred in their environment.

Study Objectives:-

- Know the reality of human resources information systems in the Jordanian insurance companies.
- Know the reality of change management in the Jordanian insurance companies.
- Know the areas of use of human resources information systems in the Jordanian insurance companies.
- Know the effect of use of human resources information systems on the effectiveness of change management in the Jordanian insurance companies.

Study Importance:-

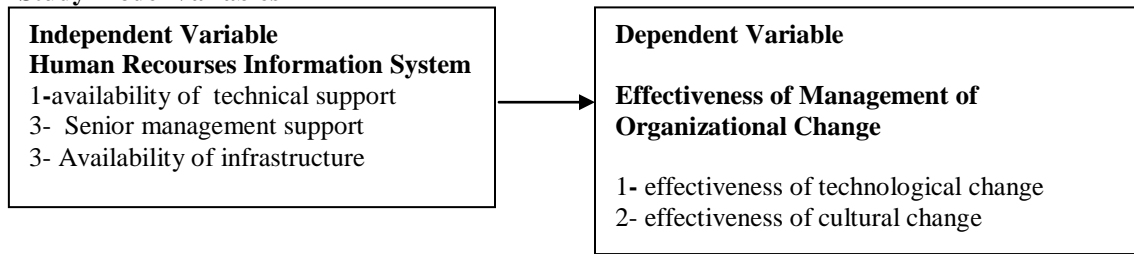
The importance of the study stems out from the importance of human resources information systems and their role in the development of the capacity of workers and raise their readiness for the best use of the resources, and this requires good change management and adapting with that change to achieve the goals and raise the organization ability to deal with the forces of change in the internal and external environment, and this assures that organizations must respond to rational demanding which is based on the principle of rational view to achieve the desired benefits while achieving stakeholder satisfaction. Also, the applied importance of this study stems out of the importance of the researched sector, the Jordanian Insurance companies, which employs a large number of staff and they are reliable sector to make partnership with government agencies in the development and employment and in running the economic resources sectors. The activation of the use of human resources information systems in the Jordanian insurance companies will raise their efficiency, so that the right choice for workers will raise their abilities and their knowledge and develop attitudes, and provide data and knowledge by the required ways.

Hypotheses:-

first hypothesis: There is no statistically significant effect of human resources information systems represented by (training on system, availability of technical support, senior management support, availability of infrastructure) on the effectiveness of change management (technological change, cultural change, change in the organizational structure) in Jordanian insurance companies.

The first hypothesis is divided into the following sub-hypotheses:

- First sub hypothesis: There is no statistically significant effect of (the training on system) on the effectiveness of organizational change in the Jordanian insurance companies.
- Second sub hypothesis: There is no statistically significant effect of viability of technical support on the effectiveness of organizational change in the Jordanian insurance companies.
- Third sub hypothesis: there is no statistically significant effect of senior management support on the effectiveness of organizational change in the Jordanian insurance companies.
- Fourth sub hypothesis: There is no statistically significant effect of the availability of infrastructure on the effectiveness of organizational change in the Jordanian insurance companies.

Study Model Variables

Sub-variables for human resources information systems have been identified by study of (Abu Obeilh ,2010) (Alkdah and Alkarasni, 2008), the variables of the effectiveness of change management were determined by study of (Anzi 2013).

Previous Literature:-**Human resources information systems:-**

The use of technology in the field of human resources has significantly and continuously expanded and therefore the human resources management activities changed at the senior, middle or lower management level (Mathis and Jackson, 2010).human resources information systems have evolved since 1960 (Lederer, 1984), the surveys carried out by the study of (Ball , 2001) indicated that 60% of Fortune Companies used human resources information system to support their daily operations of human resources management (Kovach et al., 2002).With the beginning of the nineties of the past century, human resource management practices have become more complex and there is great pressure to achieve corporate objectives related to improving general performance, and therefore human resources information systems (HRIS) have evolved to use expert information, which includes analytical tools to support the decision-making process in managing human capital (Ostermann, Staudinger & Staudinger, 2009).The human resource planning process makes the process of follow-up easier regarding gaps in the labor force, either quantitative or qualitative or planning to determine the needs of the labor force in the future and all that with the help of knowledge systems used by human resources (Dessler, 2005).The human resources information systems accelerate the formulation and implementation of effective human resources management activities, which include personnel management , payroll administration, and monitoring daily work attendance , skills development, medical history, performance appraisal, training and development, human resources planning, recruitment and career planning, negotiations and so on (Singh et al., 2011).Human resources information system defined as these systems which are based on the technology used for the acquisition, storage, processing, analysis, retrieval and distribution of relevant information on the human resources of the organization (Tannenbaum, 1990). Also it is defined as the integrated system used to collect, store and analyze information related to human resources in the organization by using the necessary data and computer applications and the hardware and software (Hendrickson, 2003).The Human Resources Information System (HRIS) is one of the most important management information systems that contributes to the goals and strategies of the organization related to the management of human resources and increasing their capabilities (Kavangah et al, 2002). (Pyburn, 1983) indicated that (HRIS) is becoming increasingly important for the successful implementation of the company's strategy, whilst (Tanriverdi, 2006) indicated that ((HRIS) is a strategic organizational resource.The human resources information systems have an essential and effective role in achieving the organization's objectives, and therefore they must provide a number of basic requirements interconnected and integrated with each other, and those requirements are as follows : (Almagribi, 2009, p. 352).

Administrative requirements:-

All organizations seek access to its goal and achieve the desired goals of performance, so the organization management is primarily responsible for identifying those goals and the role to be played by human resources information systems, so the organizations has to train workers on the system and to provide all the requirements relating to infrastructure. The system design requires cooperation between different organization and human resources management departments, and provision of all requirements of the multiple resources, and vigorous follow up for all its elements and its components to ensure efficiency and effectiveness in its performance, and that system must consider to the need of beneficiaries regarding information and statements.

Technological requirements:-

The success of the human resources system in the implementation of the tasks entrusted to it depends on what is owned by the organization of devices and machinery and infrastructure within the limits of their capabilities, and the ability of employees to manage and operate those machines by what is available of expertise and technical skills. The data and information provided by the system are extremely important which, therefore, must be handled with absolute secrecy, and the organization management should design an integrated system that considers to maintenance and safety, as they must be characterized by interdependence and integration to utilize them more effectively.

Economic requirements:-

The human element is the most important requirements in the success of the human resources information system, as the optimum utilization of the personnel, and training them on the system skills will contribute in achieving the desired goals, and will reduce the costs, as the time factor will effect on the preparation and design that is based on clear and sound principles.

Social requirements:-

The fact that the human resources information system depends on the amount of joint cooperation between human resources management and beneficiaries of the system on one hand, and between workers in the organization and the various departments on the other hand, as this cooperation results will provide the necessary information, and to help them to extract data necessary, and to encourage positive behavior for employees and beneficiaries of the system which makes it easy to take advantage of its results and outputs.

The effectiveness of organizational change:-

The application of the regulatory change is necessary for organizations in order to compete in the constantly changing business environment. The rapid development of information and communication technology has made many organizations to seek creating ways and ideas for new and innovative solutions to improve existing products and strategy of its operations and its technological system and all this called organizational change, which requires acceptance of its workers in the framing positive behavior, which will inevitably lead to change in the organization (Tan and Nasurdin, 2010). Change management: is a means of planning, implementation and follow-up in the processes of change at the institutional and personal level, and includes a revolutionary change in the management of new projects and developmental changes in the organization structure (Frans, 2010). Change management defined as the effective management of change that practice by executive, managers and employees by using technology and organizational resources. (Korir, Mukotive, 2012).

The areas of change in organizations:-

It is the most influential external power in the organization than the internal power because of its broad scope and difficulty of predicting their dimensions, and also because of rapid changes in the environment in which the organizations work. It turned out that there is a significant increase in competition between companies, especially after the emergence of the current globalization and the accession of many countries to the World Trade Organization (WTO)

These global changes have impacted heavily on the methods and patterns of management in the organizations, and thus they made them to move from the static central and bureaucratic systems to a more flexible system which fit with the nature of the global economic transformations system, and there is a need to change as a strategic choice (Sawalhah, et al 2015). The legal environment has also played a big role, represented by state intervention in economic and social activity and legal environment that lies in the change of laws or amended legislation as these impose restrictions or create opportunities, such as changes in fiscal and monetary government policy (Sawalhah et al, 2015). The technological environment is the most important source for change, where the latest technical development in industrialized countries made parallel change in the labor force structures, and the organizational structures moved from the traditional pyramid shape to the knowledge organizations and not routine business organizations, and consequently there is a change in training curricula and change in performance standards. The most important technological change that drives to change is the advancement in transportation and communication, which overcomes the border that was between the countries, and the aspects of accelerated technological development in digital equipment and machinery. The information revolution appears through information and communication technology (Sawalhah et al, 2015). Organizations also witnessed a dramatic change in values, attitudes and behaviors of individuals, customers and suppliers, culture surrounding environment do not only

affects the behavior of employees and their way of dealing, but this culture is reflected in the existing structure and pattern of the prevailing management and communication systems and information, and people's ways in problem-solving and decision-making, also it effects on customers' trends particularly in demand ,product design ,marketing mix and methods of dealing with customer (Sawalhah, et al 2015).

Hypotheses Testing:-

first hypothesis: There is no statistically significant effect of human resources information systems represented by (training on system, availability of technical support , senior management support, availability of infrastructure) on the effectiveness of change management (technological change, cultural change, change in the organizational structure) in Jordanian insurance companies.

The first hypothesis is divided into the following sub-hypotheses:-

First sub hypothesis: There is no statistically significant effect of (the training on system) on the effectiveness of organizational change in the Jordanian insurance companies.

Table 1:- Simple regression of the first hypothesis.

Dependent variable Effectiveness of organization change	Model Summary		ANOVA			Coefficients				
	R	R square	Adjusted R Square	DF	SIG	VAR Training on system	β	Std. Error	T	SIG
	0.478	0.199	105.756	1	0.000		0.478	0.033	11.096	0.000

The table above shows that there is a strong correlation between the training on system and the effectiveness of management of organizational change, the training on system explained 19.9% of the variation in the effectiveness of organizational change, as (F) value is statistically significant on the test.

Based on the results, we accept the alternative hypothesis:-

There is statistically significant effect of (the training on system) on the effectiveness of organizational change in the Jordanian insurance companies

Second sub hypothesis: There is no statistically significant effect of availability of technical support on the effectiveness of organizational change in the Jordanian insurance companies.

Table 2:- Simple regression of the second hypothesis.

Dependent variable Effectiveness of organization change	Model Summary		ANOVA			Coefficients				
	R	R square	Adjusted R Square	DF	SIG	VAR availability of technical support	β	Std. Error	T	SIG
	0.625	0.378	351.258	1	0.000		0.625	0.033	18.152	0.000

The table above shows that there is a strong correlation between the availability of technical support and effectiveness of organizational change management , the availability of technical support explained 37.8% of the variation in the effectiveness of organizational change, as (F) value is statistically significant on the test. Based on the results, we accept the alternative hypothesis: There is statistically significant effect of (availability of technical support) on the effectiveness of organizational change in the Jordanian insurance companies.

Third sub hypothesis: there is no statistically significant effect of senior management support on the effectiveness of organizational change in the Jordanian insurance companies.

Table 3:- Simple regression of the third hypothesis.

Dependent variable Effectiveness of organization change	Model Summary		ANOVA			Coefficients				
	R	R square	Adjusted R Square	DF	SIG	VAR Senior management support	β	Std. Error	T	SIG
	0.732	0.551	525.237	1	0.000		0.732	0.031	21.103	0.000

The table above that there is a strong correlation between the senior management support and the effectiveness of organizational change management, the technical support explained 55.1% of the variation in the effectiveness of organizational change, as (F) value is statistically significant on the test. Based on the results, we accept the alternative hypothesis: There is statistically significant effect of senior management support on the effectiveness of organizational change in the Jordanian insurance companies.

Fourth sub hypothesis: There is no statistically significant effect of the availability of infrastructure on the effectiveness of organizational change in the Jordanian insurance companies.

Table 4:- Simple regression of the fourth hypothesis

Dependent variable Effectiveness of organization change	Model Summary		ANOVA			Coefficients				
	R	R square	Adjusted R Square	DF	SIG	VAR Availability of infrastructure	β	Std. Error	T	SIG
	0.775	0.578	525.237	1	0.775		0.775	0.025	23.144	0.000

The table above shows that there is a strong correlation between the availability of infrastructure and effective management of organizational change, the technical support explained 57.8% of the variation in the effectiveness of organizational change, as (F) value is statistically significant on the test.

Based on the results, we accept the alternative hypothesis says:

There is statistically significant effect of the availability of infrastructure on the effectiveness of organizational change in the Jordanian insurance companies. To test the main first hypothesis, we used multiple regressions as follows:

Table 5:- Multiple regressions for fourth hypothesis

Dependent variable Effectiveness of organization change	Model Summary		ANOVA			Coefficients				
	R	R square	Adjusted R Square	DF	SIG	VAR	β	Std. Error	T	SIG
	0.805	0.725	175.232	4	0.000	Training on system	-0.075	0.025	0.725-	0.441
						Availability of technical support	0.169	0.047	4.425	0.000
						Senior management support	0.118	0.041	3.045	0.000
						Availability of infrastructure	0.335	0.036	6.321	0.000

The table above show that the correlation coefficient (0.805), indicating a strong correlation, and that the impact of dimensions of human resources information systems on the dependent variable (effectiveness of organizational change) is a statistically significant, according to F value, and R square, it is indicated that 72.5% of change in the effectiveness of organizational change was due to the human resources information systems. The training on the system has no statistically significant impact, while the other variables have statistically significant impact.

Results and Recommendations:-

Results:-

Based on the field study and analysis of survey data and the variables the study the following results was drawn:

1. There is a strong correlation between the training on system and the effectiveness of organizational change management the training on the system explained 19.9% of the variation in the effectiveness of organizational change, as (F) value is statistically significant on the test.
2. There is a strong correlation between the availability of technical support and effectiveness of management of organizational change, the availability of technical support explained 37.8% of the variation in the effectiveness of organizational change, as (F) value is statistically significant on the test.
3. There is a strong correlation between the support of senior management and the effectiveness of management of organizational change, the availability of technical support explained 55.1% of the variation in the effectiveness of organizational change, as (F) value is statistically significant on the test.
4. There is a strong correlation between the availability of infrastructure and effectiveness of organizational change, the availability of infrastructure explained 57.8% of the variation in the effectiveness of organizational change, as (F) value is statistically significant on the test.
5. Multiple Regression Results showed that training on the system has a statistically significant on the effectiveness of organizational change while there was a statistically significant effect of (availability of technical support, senior management support availability of infrastructure) on the effectiveness of organizational change.

Recommendations:-

1. Develop the capacity of workers and training them on human resources information systems requirements.
2. Put a fair system of incentives to encourage innovation and creativity process.
3. Develop Technological infrastructure and the to keep up with the developments in the world
4. Update software and deal with them reliably.
5. Take advantage from other insurance company's experiences with respect to human resources information systems.

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RESEARCH ARTICLE

GROUNDWATER MODELING OF SHALLOW AQUIFER IN LAYLAN SUB-BASIN / NE IRAQ.

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Ground water, Modeling, Laylan sub-basin, Iraq.

Abstract

A Mathematical model has been done for the study area (Laylan sub-basin) in Kirkuk / NE of Iraq by Modflow pro software. To make a simulation for the shallow aquifer to predict a clear future imagination about the hydraulic head. The aquifer receives 23.29 mm in January, 49.44 mm February, 25.17mm in March. According to such conditions with no sinks, the model was run, after converting the water surplus to the used unit in the model m/day. The model output in the monthly flow map depends on the variation of the above package (groundwater recharge). The calculated head values for each step are noticed showing slight differences in head values. Another simulation was run with sinks for one year divided also to 12 stress period. The calculated hydraulic head after 1-year simulation with the withdrawal of groundwater estimated by -432 m³/day of 70 wells, the small difference in hydraulic heads was noticed when comparing the readings of both simulations (with or without pumping). The model was run after reloading the mentioned stresses, and with increasing 10 wells yearly (as an assumption), the result shows that there will be a decrease in the southern and northern parts toward the center of the modeled area.

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Introduction:-

Groundwater Modeling is an efficient tool for groundwater management (Harbaugh AW, et al., 2000). Models are a simplification of reality to investigate certain phenomena or to predict future behavior of groundwater. Despite their efficiency, models can be complicated and produce wrong results if they are not properly designed and interpreted. Regardless of the type of model being used, similar sequences should be followed in modeling. If the conceptual model is not properly designed, all modeling processes will be a waste of time and effort. To build a proper conceptual model, hydrogeological data should be sufficient and reliable. Calibration and verification are the last steps in modeling before writing the final model report (Todd DK, and Mays LW., 2005). Groundwater management and policy decisions must be based on knowledge of the past and present behavior of the groundwater system, the likely response to future changes and the understanding of the uncertainty in those responses (Sinclair Knight Merz, 2012). The purpose of modeling can vary widely, and the approach used may depend on site-specific needs, current understanding of the hydrogeologic system, availability of input data, and expectation and use of the model results. Models are typically used to evaluate ground water movement, flow direction, velocity, and discharge rates beside:

- Evaluate the interaction between hydrogeologic systems.
- Interpolate between known measurement points.

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-Identify data gaps during site characterization.

-Aid in the development and management of groundwater supply systems.

Models are not a substitute for field investigations but should be used as supplementary tools. Results depend on the quality and quantity of the field data available to define input parameters and boundary conditions (Wang, H.F., and Anderson, M.P., 1982). The results should always be evaluated in context with the fundamental assumptions of the model and the adequacy of the input data. A general rule, it is prudent to continually question the results of modeling and the potential consequences of decisions based on misleading results, and consider what can be done to verify the results (U.S. EPA., 1996c).

Location of Study area:-

The study area (Laylan) located in the northeastern part of Iraq, it is a sub-basin belongs to Kirkuk province. Laylan is approximately 26 Km to the southeast of the governorate capital, Kirkuk. Geographically, the area of study lies in between latitudes ($35^{\circ} 26' 30''$ - $35^{\circ} 10' 00''$), and longitudes ($44^{\circ} 43' 30''$ - $44^{\circ} 22' 30''$), the area of study is about 256 km². These places belong to Kirkuk province. Geologically, structural boundaries for the study area from east and northeast by Kirkuk anticline, while in the west and southwest by Jambur Mountains (Anticline), and it is bounded by hydrological natural boundaries from north and northwest by ephemeral stream (intermittent stream named Shireen valley), while in the south and southwest by ephemeral stream named (Qarqacha valley) (Figure 1).

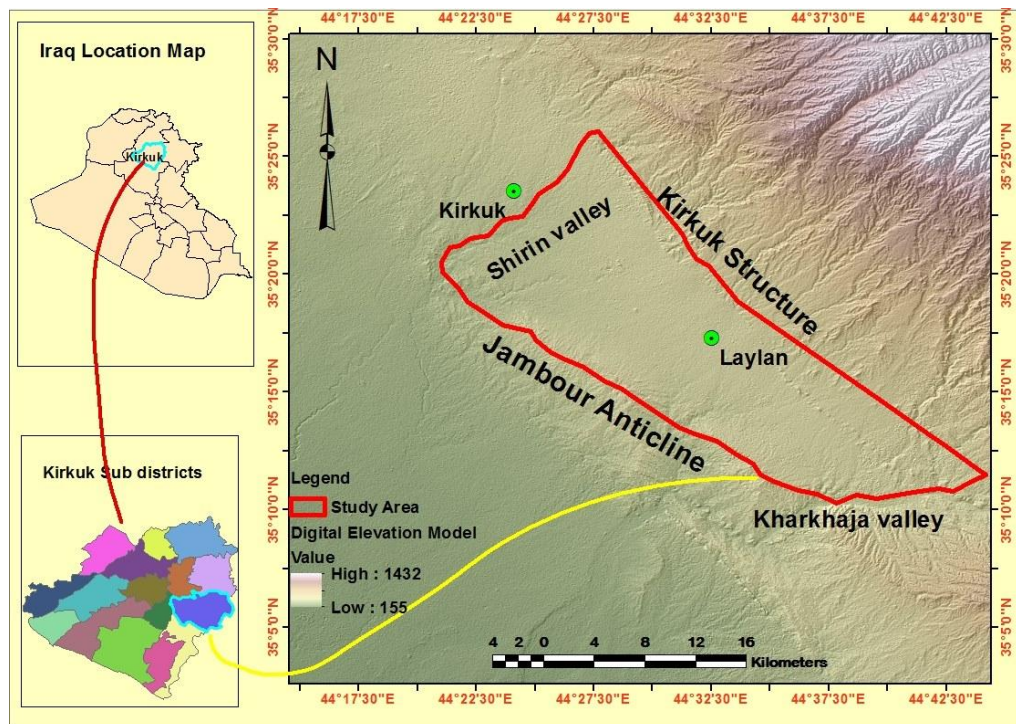


Figure (1):- Location of the study area.

General Setting of Tectonic, Geology, and Structure of the Study Area:-

Lailan sub-basin contains Mukdadyia (L. Pliocene) and Bai-Hassan (U. Pliocene) Formations with a cover of Pleistocene and Holocene Quaternary deposits. Groundwater exists abundantly in sand and gravel layers of Quaternary and Bai Hassan Formation. Lailan located within unstable shelf in the deepest part of the Foothill Zone, which called Hemrin-Makhul Subzone. The subzone was the depocentre of the Neogene molasses but has been a subsiding unit throughout the Mesozoic and Tertiary (Jassim and Goff, 2006), AL-Mamuri (2005).

Methodology:-

The research is done by using the hydrogeological field data to build a proper model for the study area (Lailan sub-basin) through the following steps:

Grid Design:-

The dimensions of the model should be selected during the formulation of the conceptual model. To minimize a variety of sources of numerical errors the model grid should be designed using the finest mesh spacing and time steps. In designing the grid, the length to width ratio (or aspect ratio) of cells or elements should be kept as close to one as possible. Long linear cells or elements can lead to numerical instabilities or errors and should be avoided, particularly if the aspect ratio is greater than about (Jacques W. Delleur, 2006). The modeled area was 256 km², this area was divided into 355 cells, the dimension of each cell is (750 m*1000 m), all these cells are active.

Boundary & Initial Conditions:-

To obtain a unique solution of a partial differential equation corresponding to a given physical process, additional information about the physical state of the process is required. This information is supplied by boundary and initial conditions. The boundary conditions include the geometry of the boundary, for groundwater model applications, the initial conditions are simply the values of the dependent variable specified everywhere inside the boundary at the start of the simulation. Normally, the initial conditions are specified to be a steady-state solution. It should be recognized that heads will change during the transient simulation, not only in response to the new pumping stress but also due to the initial conditions. Boundary conditions have great influence on the computation of flow velocities and heads within the modeled area. The boundary conditions which are used in the modeled area, specified head positive values (+1) in the I Bound array defines active cells when expressing the domain inside.

Initial Hydraulic heads:-

MODFLOW requires initial hydraulic heads at the beginning of a flow simulation. For steady state simulation, the initial heads are used as starting values. Actual, confidential head values were derived from the hydrogeologic data bank of groundwater commission. GIS, the 3D spatial analyst is used to create initial isopotential lines. The isopotential lines of the aquifer, the same values were loaded into the model as initial hydraulic heads.

Aquifer Material Properties:-

The model needs more additional properties such as top and bottom of the aquifer beside transmissivity... Etc, the values were loaded into the model. Flow packages represented by recharge (L/T) which means length unit /time unit, positive values, and wells (L³/T), negative values, will add to the model during simulation of the Transient model. Recharge is defined by assigning the data to each vertical column of cells. The model assumes that a well penetrates the full thickness of the cell. To calculate heads in each cell in finite difference grid, GMS prepares one finite difference equation for each cell. SIP (strongly implicit procedure) package is used to solve the system of a finite difference equation.

Results and Discussion:-**Steady State Simulation:-**

Flow in such simulation means the volume passing a given point per unit of time remains constant. An initial steady-state condition is required for time-dependent modeling of groundwater flow. After loading data and after running the model, hydraulic heads are the primary results of the model as a steady state simulation will appear as in (Figure 2).

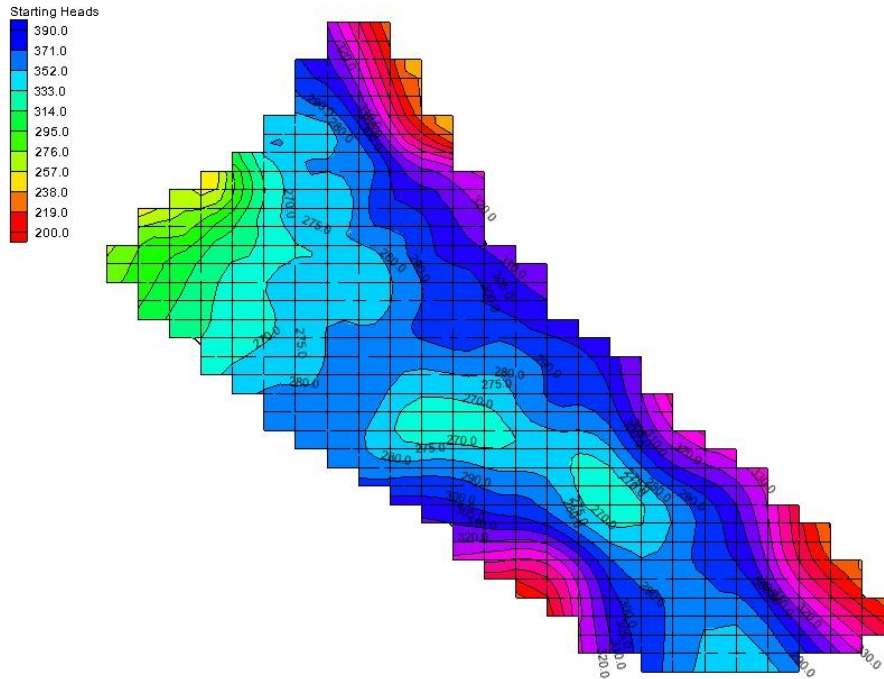


Figure (2):-Steady State Simulation

Model Calibrations:-

Calibration is the process of adjusting model inputs to achieve the desired degree of correspondence between the model simulations and the natural groundwater flow system(Christoph Wels,2012). In other words, calibration methods are used until the solution matches the known (usually hydraulic heads).For this model, calibration is made for inputs to reach the matching between inputs and model simulation. Model calibration should include comparisons between model-simulated conditions and field conditions for the following data.

Transient Simulation:-

Transient simulation involves the change in hydraulic head with time. These simulations are needed to narrow the range of variability in model input data since there are numerous choices of model input data values which may result in similar steady-state simulations.

The first attempt in the transient simulation is made with an assumption without withdrawal water from the aquifer. The flow recharge package is the most important factor included in this simulation due to the difference in climatological factors, groundwater recharge as a source is an influenced factor during three months in a year according to the calculations of water surplus. The aquifer receives 23.29mm in January, 49.44 mm February, 25.17mm in March. According to such conditions with no sinks, the model was run, after converting the water surplus to the used unit in the model m/day.

Time stepping in transient simulation:-

The time for one-year simulation without sinks (wells) is divided to 12 stress periods, each stress period represents a month, while the period length of each stress period are divided into days, so the total time steps equal to 12 months, while the total simulation time equals to 3.65E+2 days. The model output in the monthly flow map depends on the variation of the above package (groundwater recharge). The calculated head values for each step are noticed showing slight differences in head values.

Another simulation was run with sinks for one year divided also to 12 stress period. The calculated hydraulic head after 1 year simulation with withdrawal of groundwater estimated by $-432 \text{ m}^3/\text{day}$ of 70 wells, Small difference in hydraulic heads was noticed when comparing the readings of both simulations (with or without pumping) (Figure 3).

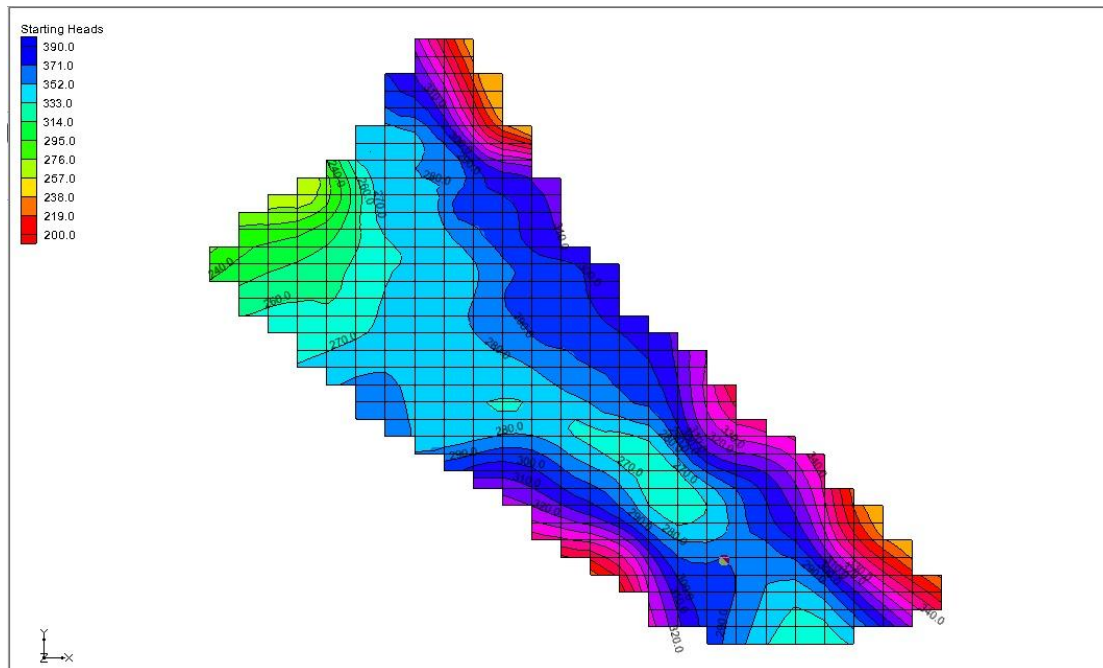


Figure (3):-Transient Simulation

Predictive simulation:-

A model can be used to predict future groundwater flow conditions, such simulation estimates the hydraulic response of an aquifer, and also it can predict the pumping rate needed to monitor the hydraulic heads.

A pumping strategy (groundwater management) is a set of spatially and possibly temporary distributed rates of extracting water from the aquifer.

To predict the simulation for 5 years, the stress period were divided into 20 stress periods, each one-year simulation divides into 2-period lengths, 1 for the wet period represented by 90 days while the another one for the dry period represented by 270 days.

For utilization of 70 wells for agricultural purpose, wells flow package represented by estimated withdrawal of $(-432) \text{ m}^3/\text{day}$ during the dry period and $(-216) \text{ m}^3/\text{day}$ during wet season, recharge flow package represented by $(0.00108) \text{ m/day}$ during the wet season and (0) during the dry season. The model was run after reloading the mentioned stresses, and with increasing 10 wells yearly (as an assumption), the result shows that there will be a decrease in the southern and northern parts toward the center of the modeled area (figure 4)

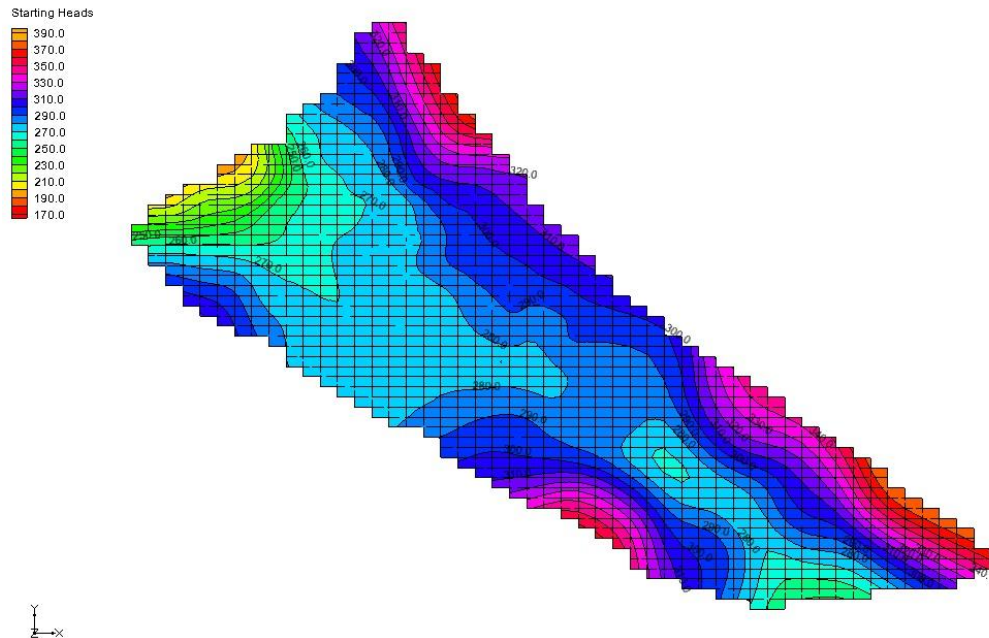


figure (4):- 5 Years later Simulation

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RESEARCH ARTICLE

EFFECT OF TRIDEX EXTRACT ON SEED GERMINATION BIOASSAY.

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Abstract

The fully mature fresh leaves of *Tridax procumbens* L. were collected and prepared extract. *Tridax* weed, used for germination. Make different concentration of extract. This was carried on Cowpea *vigna unguiculata* (L.) Walp. The studies on the influence of extract on seed germination are highly essential and of paramount importance, considering this view, the impact of *Tridax* leaf extract has been studied by using seed germination bioassay of Cowpea in the present investigation.

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Introduction:-

Cowpeas are one of the most important food legume crops in the semiarid tropics covering Asia, Africa, southern Europe, and Central and South America. A drought-tolerant and warm-weather crop, cowpeas are well-adapted to the drier regions of the tropics, where other food legumes do not perform well. It also has the useful ability to fix atmospheric nitrogen through its root nodules, and it grows well in poor soils with more than 85% sand and with less than 0.2% organic matter and low levels of phosphorus. In addition, it is shade tolerant, so is compatible as an intercrop with maize, millet, sorghum, sugarcane, and cotton. This makes cowpeas an important component of traditional intercropping systems, dried stalks of cowpea is a valuable by-product, used as animal feed.

Seed germination is one of the most basic aspects. During seed germination sequential series of physiological and biochemical event takes place in quiescent seed, resulting into seedling. It is transition period between resting and growth phases of plants and considered to be completed at the time of emergence of radical. The stimulatory or inhibitory impact of extract can be initially analysed through seed germination bioassay studies. *Tridax* leaf extract was used to study seed germination bioassay on Cowpea.

Material And Method:-

Collection of material:-

The fully mature fresh leaves were collected in polythene bags from the fixed study area and the material was brought to the laboratory in ice bags. Leaves were washed thoroughly under tap water and spread over the blotting papers under shade for drying.

Preparation of Extracts:-

Freshly collected mature leaves were cleaned and shade dried. The leaves were uniformly homogenized in 100 ml 80% ethanol and condensed in water bath to 10 ml below 60°C to avoid evaporation of volatile compounds and denaturation of active principle in it. This was filtered and filtrate was made to 100 ml final volume with sterile distilled water & stored in sterile coloured bottle in deep freezer at 0°C for further use.

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Seed germination bioassay:-

This is carried out with Cowpea. Uniform seeds were washed thoroughly in water & surface sterilized with 0.5% HgCl₂ solution & again washed with distilled water. Surface sterilized seeds were germinated in different concentration of *Tridax* leaf extract observation on

- (1) Germination %
- (2) Length of plumule & radicle.
- (3) Root/shoot ratio
- (4) Fresh & dry weight of seedling
- (5) Vigor index were recorded. All the sets were triplicated under laboratory condition. Observation on the following parameter were recorded.

Germination percentage:-

Development of seedling from seeds is known germination. It is a process in which the seed sprouts for growing & developing into a plant. Germination percentage is a calculated approximately of the viability of a population at seeds. The germination rate provides a measure of the time course of seed germination.

Record for seeds germinated in each treatment from 2nd day to 9th days after sowing was noted carefully. The seeds with normal length of radicle & plumule were considered as germinated seeds. The percent germination was calculated as

$$\text{Germination percentage} = \frac{\text{Number of seed germinated}}{\text{Total number of seeds kept for germination}} \times 100$$

Length of Plumule & radicle in germinated seeds.

From each replication & each replication and each treatment ten germination seeds were randomly selected & the length of selected Plumule & radicle of these germinating seeds was recorded at 6th & 9th days.

Root/shoot ratio –

The root /shoot ratio for each germinating seeds at each treatment was calculated as follows

$$\text{Root /Shoot Ratio} = \frac{\text{Root length (cm)}}{\text{Shoot length (cm)}}$$

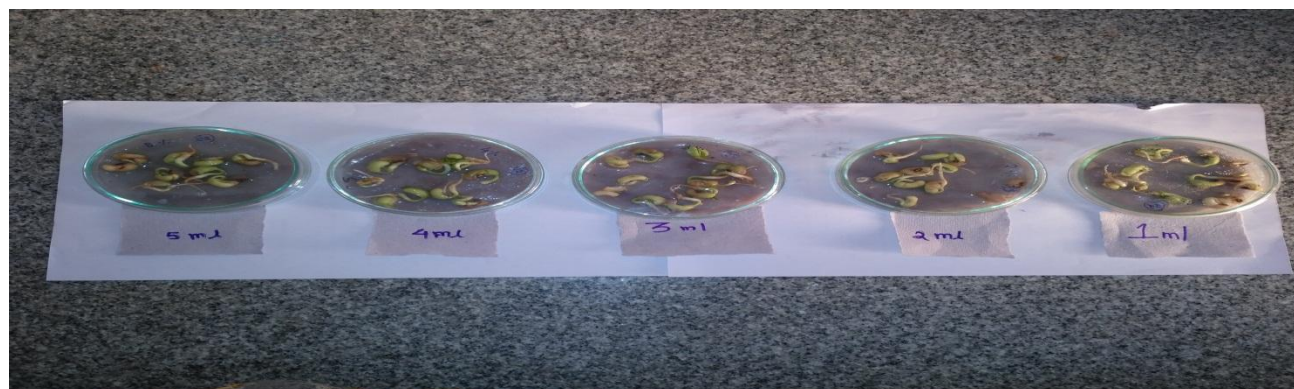
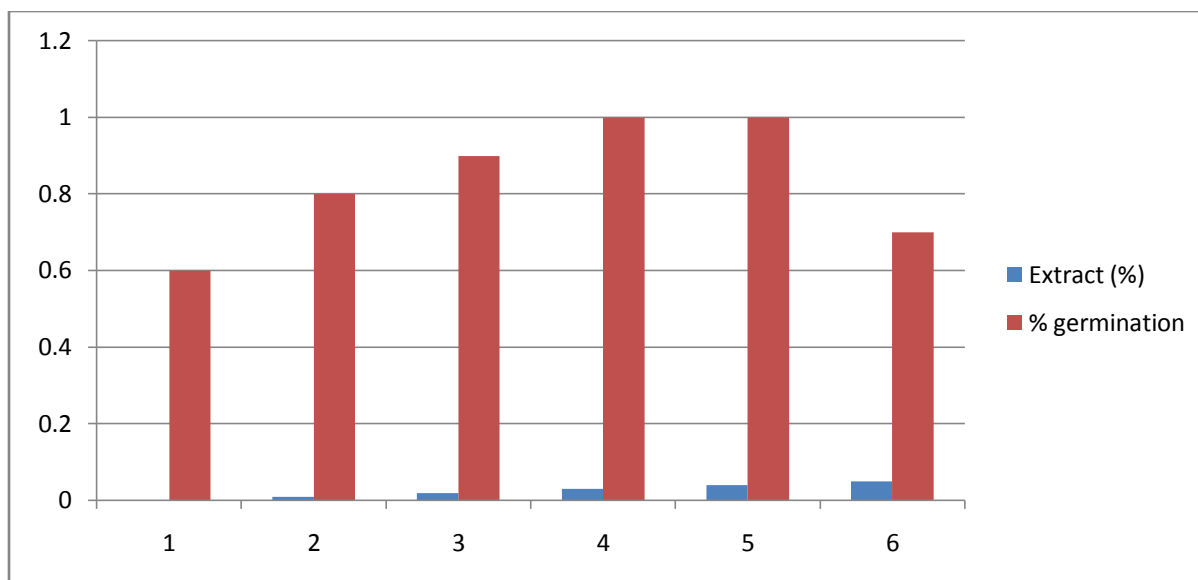
Seed Vigor index (VI):-

Seedling vigor index were determined by germination percentage and seedling length of the same seed lot. Seed vigor index is calculated by multiplying germination % & Seedling length.

Seed vigor index = germination % X seedling length (mm)

Table 1:-

Sr.no	Extract (%)	% germination	Shoot length (cm)	Root length (cm)	Total length (cm)	Root/shoot Ratio (cm)	Vigor Index (VI)
1	Control	60%	2.3	11.5	13.8	5.0	828
2	1%	80%	9.9	10.5	21.4	0.15	1712
3	2%	90%	10.5	12.1	21.0	1.15	1890
4	3%	100%	12.00	14.0	24.1	1.16	2410
5	4%	100%	12.8	9.9	26.8	0.77	2680
6	5%	70%	8.00	11.5	17.9	1.43	1432



Result And Discussion:-

Effects of extracts on seeds germination:-

The result recorded in table 1 indicate that the extracts had positively stimulated the seed germination percentage over control in higher concentration. While the lower conc. (1%,2% 3%) have caused low influence on seed germination. The length of root & shoot was significantly increased at 4% concentration, however at lower conc. it was reduced over control.

Root / shoot length:-

In present investigation the impact of the extracts on root & shoot length in Cowpea seedling was studied in details. For this extracts of (*Tridax*) were used. From the result it is seen that extract of the species was stimulatory enhancing the seed germination at higher conc. (4%) however lower concentration adversely affected the seed germination, root length & shoot length.

Seed vigor index (VI)-

4% *Tridax* extract seed lot showing the higher seed vigor index is considered to be more vigorous.

Conclusion:-

The early seedling growth governs its establishment through root & shoot elongation. The uptake of water & nutrients depends on the early root development & its spreading. Inhibition or stimulation of root & shoot length at seedling level will have adverse or favorable effects on the future growth of the plant.

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RESEARCH ARTICLE

ESTABLISHMENT OF PLAQUE ASSAY METHOD FOR TITRATING YELLOW FEVER VACCINE

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Abstract

The present study was undertaken with a view to look for the use of Vero cell line for plaque assay of Yellow fever vaccine, over Mouse LD₅₀ method, for vaccine potency test. To establish the plaque assay the growth conditions for Vero cell line were standardized. Different parameters like cell count, dilution, inoculum size, adsorption time for virus, concentration for Methyl Cellulose (MC) in the overlay media and incubation period were standardized. And titration of Yellow fever vaccine was performed by Plaque Forming Unit (PFU) method. A cell concentration of $3-4 \times 10^5$ cells per ml, in tissue culture plate gave a complete monolayer after overnight incubation. Virus dilution of 10^{-2} was optimum for desired pfu/0.5ml. An inoculum size of 0.2 ml per well was found suitable. Maximum plaques were obtained after an adsorption time of 4 hours, at $36 \pm 1^\circ\text{C}$, by 7 days of incubation. Different concentrations of methyl cellulose, overlay media were used. 1% methyl cellulose in Minimum Essential Media (MEM) with 2% FBS gave good results. The different samples of Yellow fever 17D vaccines were titrated by Plaque Assay method along with the IRS and titre results were compared with the titre values of manufacturer's potency assay. The vaccine pass the limit of minimum requirement of potency i.e., $4.24 \log_{10}$ PFU/0.5ml dose, equivalent to titre 1000 MouseLD₅₀.

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Introduction:-

Yellow fever was recognized as a clinical entity as early as in the seventeenth century and was familiar to pirates as the 'YELLOW JACK'. It is a native of Africa and was transported along the trade routes to Europe, America. The disease has been largely confined to certain areas of Africa and South and Central America. Yellow fever is a non-contagious acute infectious disease caused by the Yellow fever virus^{[1][2]}. Yellow Fever is member of family *Flaviviridae*, genus *Flavivirus*, and the virus after which the family and genus were named (*flavus*, Latin- Yellow). The virus has a small positive-sense, single stranded (ss) RNA genome. It is a spherical enveloped virus, bearing surface projections. Nucleocapsid has an icosahedral symmetry, 40-50 nm in diameter.

It is a serious health problem with clinical symptoms that may vary from mild to severe disease characterized by sudden onset of fever and jaundice. The most serious forms also include hepatorenal syndrome, bleeding, shock and eventually evolve to death^[3]. It is both Viscerotrophic, causing liver disease and Neurotrophic causing encephalitis^[4].

Among various preventive measures such as vector control, improved hygiene and sanitation and immunization; immunization currently represents the least expensive and most effective means to prevent infection and to avoid subsequent disease ^[5] ^[6]. Yellow fever is a vaccine preventable disease and can be controlled by an effective vaccination program ^[7].

Yellow fever was one of the most feared lethal diseases before the development of an effective vaccine ^[3]. The live attenuated vaccine of Yellow fever was developed in 1936 by Thieler & Smith by attenuating Yellow fever virus Asibi strain through serial passages in tissue culture and animals, originating the 17D attenuated vaccines. Another French neurotropic vaccine was developed by Sellard and Laigret in 1932; it was popular as Dakar vaccine. The serious drawbacks of this vaccine were high incidences of encephalitic reactions in children. Due to which it was stopped being recommended for children under 14 years in 1961 and manufacturing of this vaccine strain was discontinued in 1980 ^[8].

Nowadays, there are two licensed vaccines available in international market, the first vaccine uses the 17D-214 strain and the second uses the 17DD strain (in Brazil) ^[9]. Yellow fever 17D vaccine has an outstanding feature that the vaccine provide protection against other Flaviviruses.

Vaccines are essential tool in the prevention of disease, as they protect the vaccinated individual from developing a potentially serious disease and helps in the reduction of spreading of infectious diseases. Thus the manufacturers are primarily responsible for the safety and efficacy of the vaccines. However, it is country's National Control Authority (NCA) that is responsible for establishing procedures for assuring that vaccines intended for use in that country are of adequate safety and efficacy. The activities required in this concept are incorporated under the responsibility of Quality Control (QC). The NCA grants approval for marketing vaccines and certify manufacturer by monitoring production, testing procedures and processes, performing QC testing of vaccines through National Control Laboratory (NCL) ^[10].

QC tests for a vaccine are; sterility, identity, stability, toxicity, potency test etc. Potency is one of the most important parameters for the QC of vaccines. WHO requirements for potency testing of 17D vaccine are that the titre of the vaccine shall be not less than 1,000 mouse LD₅₀/dose (0.5 ml) or its equivalent PFU ^[11] ^[12]. Potency of Yellow fever is tested by two methods; LD₅₀, PFU ^[11] ^[12] ^[13].

Mouse LD₅₀ is a sensitive method but involves use of animals, with a long observation period *i.e.*, 21 days and involves large number of animals (8 for each dilution). On the other hand plaque technique is quite sensitive, accurate, less time consuming method for titration. The PFU method gives result in 7 days, and has no use of animals. Many problems of taking care of mice, their food, and breeding etc., and problems faced due to premature death, variation in animal immune status, sensitivity to particular immunological products and variability in results can be avoided. Keeping in view the guidelines of RRR (Replacement, Refinement and Reduction of animals) for use of laboratory animals, an attempt has been made to perform the potency assay *in vitro*, *i.e.*, PFU method. PFU method is equally sensitive method as mouse LD₅₀ (*in vivo* method) ^[13].

Material and Method:-

Vaccine:- The samples of the Live-attenuated, 17-D (204) Yellow fever vaccine, with diluent for suspension.

Cell lines:- Vero Cell line was revived at passage level- 126 at CDL, Kasauli.

Reference Standard:- Yellow fever International Reference Standard (IRS); 99/616 IRS

Media, Reagents And Solutions:-

Buffers and solutions:-

- Sterile water for injection (W.F.I.).
- Distilled water: - Solutions used in all procedures were dissolved in distilled water.
- Washing Buffer: Phosphate buffer saline (pH7.4), Sodium chloride (NaCl)-0.8gm, Potassium chloride (KCl)-0.020gm, Sodium Hydrogen Phosphate (Na₂HPO₄)-0.115gm, Potassium Dihydrogen Phosphate (KH₂PO₄)-0.020gm.

The above contents were mixed and solution was sterilized by autoclaving at 121°C, 15 lbs, for 20min. and stored at 2-8°C.

- 0.25% Trypsin-EDTA (1X) (Dissociation Reagent);

Staining and fixing reagents:-

- Trypan Blue (0.4% solutions):- The solution was used for cell counting.
- 0.5% Crystal Violet Solution, Crystal Violet-2.5gm, Absolute ethanol-100ml, Formaldehyde (37-41%) - 135ml, Distilled water-265ml. The above contents were mixed and filtered through filter paper. Solution is stored at room temperature. It was used for staining the plaques.
- Cell fixative, Formal saline (10%):- Normal Saline (NaCl), NaCl-0.85gm, Distilled water-100ml.
- The above contents were mixed and solution was sterilized by autoclaving at 121°C, 15 lbs, for 20min.
- Formal Saline, Formaldehyde (37-41%)-10ml, Normal Saline-90ml. The solution is stored at room temperature. The solution was used as fixative *i.e.*, for fixation of cells.

Media:-

- Foetal Bovine Serum (FBS), Sterile, gamma-irradiated, Mycoplasma tested; It was used at different concentrations in MEM. Stored at 2°C-8°C.
- Minimum Essential Medium (MEM) 1X, MEM Alpha (1X) + GlutaMAX-I [-] Ribonucleosides, [-] Deoxyribonucleosides, Stored at 2°C to 8°C.
- Penicillin-Streptomycin solution, Penicillin-[+] 5,000 units/ml, Streptomycin-[+] 5,000 µg/ml, Stored at 2°C to 8°C.
- Growth medium- MEM= 470ml, FBS-25ml, Pen/Strep (1%)-5ml.
- Maintenance medium; MEM-483ml, FBS-12ml, Pen/Strep (1%)-5ml.
- Overlay medium: Methyl Cellulose (1% Stock solution):-

Solution A:- MEM-97ml, Methyl cellulose-1gm, Vigorous shaking of solution to mix methyl cellulose properly. Autoclave the solution at 121°C, for 15 min. Solution is cooled to room temperature.

Solution B:- FBS-2ml, Penicillin/Streptomycin-1ml.

Solution B was added to solution A; Stored at 2-8°C.

Materials And Equipments:-

Tissue culture plates (Disposable, flat bottomed 6-Well-tissue-culture plates), Tissue culture bottles (Roux bottles) (25cm² and 75cm² sterile disposable cell culture bottles), Autopipettes, Disposable graduated pipette (5ml, 25ml, 100ml), Ependorff, falcon tubes, Disposable syringe and needle (5ml). Glass vials, Measuring cylinders, Glass bottles, Beakers, flasks. Disposable gloves, masks, tissue paper, vaccine vial opener, glass vial stand, vortex mixer, haemocytometer, inverted microscope, biosafety cabinet, autoclave, deep freezer and refrigerator, carbon dioxide incubator.

Method:-

Revival and Propagation of Vero cell line:-

Vero cell line was obtained from CDL, Kasauli at passage level-126 (P-126). The ampoules containing Vero cell line, were stored at -196°C in cryopreservation system (in liquid Nitrogen), were removed and transferred immediately to water bath at 36°C ± 1°C till its content was completely thawed. Then outside of the ampoule was wiped with disinfectant to reduce contamination and the ampoules were opened in Bio-safety Cabinet. The cell suspension was transferred to culture bottles containing growth media, MEM with 5%FBS. The bottles were incubated at 36°C ± 1°C for 4 hrs. The media was decanted carefully to get rid of DMSO present and fresh growth medium was added for the production of cell monolayer and incubated at 36°C ± 1°C until the cell monolayer is complete.

Maintenance of cell lines:-

Cell lines were maintained at 32°C temperature with maintenance medium MEM+2%FBS.

Tissue Culture:-

Trypsinization of Cells:-

Once the monolayer was complete, the cells were harvested using the following procedure-

- A freshly thawed trypsin was warmed to $36^{\circ}\text{C} \pm 1^{\circ}\text{C}$ in incubator.
- The tissue culture bottle was observed under the inverted microscope, for following characteristics: - Confluent monolayer, healthy and viable cells, no or little granularity, no C.P.E., or other changes in the cell growth present.
- Using a sterile pipette, whole spent medium was aspirated from tissue culture bottle.
- The cell layer was rinsed gently with 5 ml of 1X MEM.
- 0.25% trypsin solution was added to the cell monolayer, *i.e.*, 1ml to 25cm^2 tissue culture bottles and 2-3 ml to 75cm^2 tissue culture bottles.
- The tissue culture bottle was incubated in the incubator at $36^{\circ}\text{C} \pm 1^{\circ}\text{C}$ until the cells starts detaching from the surface.
- Discard the trypsin before the cell monolayer detach entirely from the surface of the tissue culture bottle.
- The growth medium was added to cell culture bottle, 10 ml to 25cm^2 culture bottles, 20 ml to 75cm^2 culture bottles. The serum in the growth medium stops the action of trypsin still present.
- The media was aspirated gently to loosen the cells from the surface of the bottles and pooled the cell suspension from each bottle into new tissue culture bottle.
- Re-suspended the cells in growth medium to desired concentration, based on cell counting. Seed fresh culture bottles or 6-well tissue culture plates.

Cell counting:-

Cell counting was done for enumeration of viable cells per ml. Accurate number of cells in a cell suspension was calculated by using haemocytometer (Neubauer chamber). Before cell counting the cells were dispersed thoroughly by pipetting in and out.

- Prepared 1:1 dilution of original cell suspension (obtained by trypsinisation) using 0.4% trypan blue as follows: 0.5ml of diluted cell suspension, 0.5ml of 0.4% trypan blue.
- The cell suspension was mixed well with pipette and immediately aspirated a sufficient volume to fill the haemocytometer.
- All viable (unstained) cells were counted in the 8 squares (1.0mm) constituting the 4 corners of each of the two haemocytometer chamber, omitting cells lying on the upper line and left line of each square. The volume of each corner square = 0.1mm^3 ($1.0 \times 1.0 \times 0.1$).
- Calculated the mean count of the cells per 0.1mm^3 .
- The number of cells in the suspension were calculated using the following formula:

$$N = m \times tb \times V \times 10^4$$

Where,

N- Total number of cells in the suspension

m- Mean of cell count in eight corners

tb- Trypan blue correction factor

V- volume of original cell suspension in ml

10^4 - conversion factor for haemocytometer

PLAQUE ASSAY ^[14]

Preparation of the Vero Cell monolayer:-

After counting cells, the cell concentration was adjusted to $3-4 \times 10^5$ Vero cells/ml of cell suspension, in MEM with 5%FBS.

- 2ml of this cell suspension was added to each well of 6-well tissue culture plate.
- Tissue culture plates were incubated at $36^{\circ}\text{C} \pm 1^{\circ}\text{C}$, at 5%CO₂, overnight for complete monolayer formation.

Reconstitution of Yellow fever vaccine:-

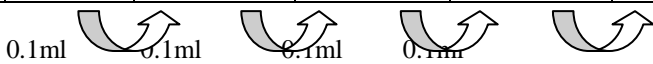
Lyophilized

vaccine (0.5ml dose) was diluted in the diluent provided with the vaccine. It was placed on ice bath till use.

Preparation of vaccine dilution:-

- The sterile glass vials were labelled as 10^{-1} , 10^{-2} , 10^{-3} , 10^{-4} and 10^{-5} .
- The reconstituted vaccine was serially diluted in chilled dilution medium (MEM with 2%FBS), in a range of 10^{-1} to 10^{-5} .
- 0.9ml of chilled dilution medium was dispensed in all 5-vials.

- The 10-fold dilutions were prepared by adding 0.1ml of reconstituted vaccine into first vial (10^{-1}), and mixed on vortex for 15-20 seconds and then transferring 0.1ml of 10^{-1} dilution to next vial making it 10^{-2} and vortex it, and similarly upto 10^{-5} dilution.

Range of dilutions used in plaque assay of Yellow fever vaccine.					
Dilution	10^{-1}	10^{-2}	10^{-3}	10^{-4}	10^{-5}
Diluent (in ml)	0.9	0.9	0.9	0.9	0.9
Vaccine (in ml)	0.1	—	—	—	—
					

Virus infection and Adsorption:-

- The media was aspirated, from the wells of tissue culture plates with confluent monolayer of Vero cell line.
- 0.2ml of each viral dilution from 10^{-1} to 10^{-5} was added to different wells, minimum 2 wells for each dilution.
- Two wells will remain as the cell-control wells, and 0.2 ml of medium (MEM+2% FBS) was added to it.
- The plates were incubated for 4 hours, for adsorption of virus to the cell-monolayer. The plates were gently shaken after every 30 minutes, for uniform distribution of the inoculum.

Addition of Overlay Medium:-

- After 4hrs of adsorption, 3ml of methyl cellulose was added to all 6 wells of the plate, to stop the viral diffusion.
- The plates were incubated for 7 days, at $36^{\circ}\text{C} \pm 1^{\circ}\text{C}$, at 5% CO_2 for the development of plaques.

Plaque Fixation and Staining:-

- On 7th day, the overlay medium was aspirated from the wells, and the plates were washed 2-times with PBS 2ml per well.
- The cells were fixed with 2ml per well 10% formal saline solution for 30 minutes.
- After fixation aspirated the fixative and the plates were washed with PBS.
- The plates were stained with 0.5% crystal violet stain solution for 30 minutes at room temperature.
- The plates were washed with PBS 2-3 times and air dried.
- After proper drying of tissue culture plates, the plates were examined to count the number of plaques, comparing with the cell control.

Calculation of results (PFU/0.5ml dose):-

The number of Plaque Forming Units (PFU), were titred per 0.5ml dose.

Formula: - $\text{PFU} = \text{P} \times \text{D} \times 2.5$

Where: - P - Average No. of plaques

D- Reciprocal of Dilution factor

Results:-

Cell count:-

The cell count of $3-4 \times 10^5$ cells/ml gave a complete monolayer when 2ml of this suspension was used per well of the 6-well tissue culture plate. (Table-1)

Table 1:- Standardization of the Cell Count.

Cell count	Incubation time	Result
2×10^5	Overnight Incubation	Incomplete Monolayer Formation
3×10^5	Overnight Incubation	Complete Monolayer Formation
4×10^5	Overnight Incubation	Complete Monolayer Formation
5×10^5	Overnight Incubation	Complete Monolayer, with overgrowth of cells at centre.

Working Dilution and Inoculum Size:-

It was observed that the dilution of 10^{-2} at the inoculum size of 0.2ml was suitable for the assay (Table-2). Therefore this dilution and inoculum size were used in the entire study.

At 10^{-1} dilution with 0.2ml of inoculum, plaque formation occurs but due to their large number, their boundaries were seen overlapping and hence plaques were difficult to count. With higher inoculum sizes 0.3ml and 0.4ml, the plaques were absent due to excessive degeneration of cells.

At 10^{-3} dilution, very less number of plaques was obtained. Even with highest inoculum size of 0.4ml, the plaque ranged from 10-20, hence was not suitable for the study. With 10^{-2} dilution of the virus, the inoculum size of 0.2ml produced a uniform distribution of plaques (Table-3). With higher inoculum sizes 0.3ml and 0.4ml, the plaques formation occurred yet counting could not be made due to excessive degeneration of cells. So, the inoculum size of 0.2 ml was selected as it gave a discrete plaque distribution with round appearance, sharp boundaries and clear margins.

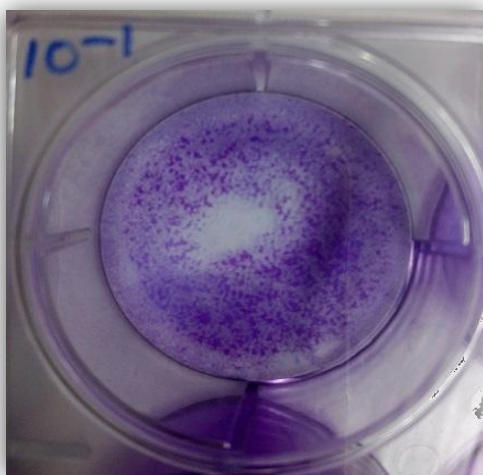
Table 2:- Effect of Different Dilution on the Formation of Plaques.

Virus Dilution	Results
10^{-1}	Very high no. of plaques. Boundaries overlapping. Unable to count.
10^{-2}	Plaques were seen as uniformly distributed, discrete, with round shape.
10^{-3}	Plaque no. very less.
Cell control	Healthy.

Table 3:- Effect of Different Inoculums on the Formation of Plaques.

Inoculum size (10^{-2} dilution)	Results
0.2 ml	Discrete plaques were observed.
0.3 ml	No. of plaques was high, unable to count.
0.4ml	No plaques were seen, rounding of cells and detachment.

Image 1: - Shows 10^{-1} dilution with 0.2ml of inoculum. The plaque formation occurs but due to their large number, their boundaries were seen overlapping and hence plaques were difficult to count.



Adsorption time:-

Maximum plaques were obtained after an adsorption time of 4 hours at $36^{\circ}\text{C} \pm 1^{\circ}\text{C}$. (Table-4) At adsorption time 1 hour there was no plaque development. Therefore the adsorption time of 4 hrs was selected.

Table 4:- Effect of Different Adsorption Times on Plaque Formation.

Adsorption Time	Results
1 hour	No Plaque development.
4 hours	Good plaques development; maximum no. of plaques seen.

Image 2:-

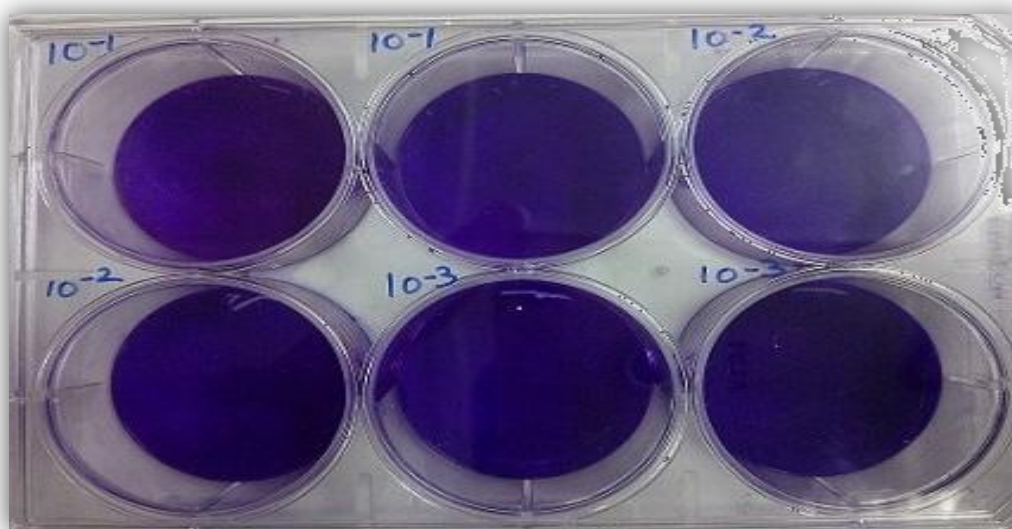


Image 3:-

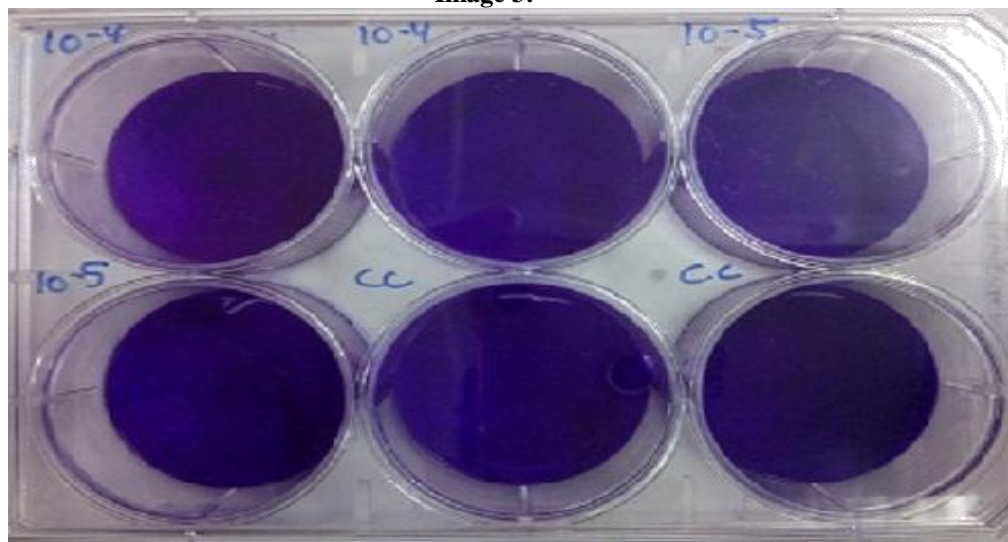


Image-2 and 3:- No plaque development due to incomplete adsorption time period of 1 hr.

Methyl cellulose Concentration:-

It was observed that maximum final concentration of 1% of Methyl cellulose (MC) in medium (MEM with 2% FBS) suited well for Vero cells as well as for the formation of plaques. (Table-5)

Low concentration of MC (0.8%) could not prevent the spreading of the virus on entire plate. The plaques were seen with diffused boundaries and were not countable.

Table 5:- Effect of Different Concentration of MC on Vero Cells and Plaque Formation.

MC Concentration	Results	
	Test	Control
0.8%	Plaques were seen but were not countable	Cells were normal.
1%	Plaque formation occurred.	Cells were normal.

Image 4:-

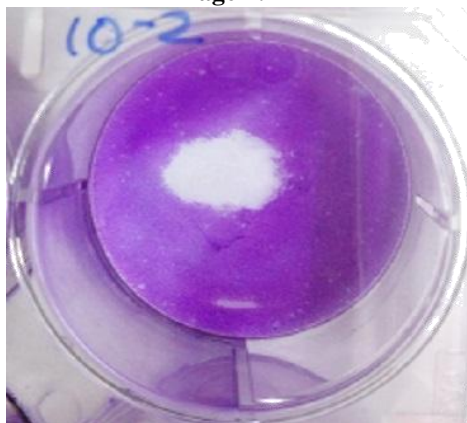


image 5:-



Image 4 and 5:- represents the appearance of plaques of Yellow fever virus in a 6-well tissue culture plates. The first well is inoculated with 10^{-1} dilution of the virus and clear but non countable plaques were seen. The 2nd well was inoculated with 10^{-2} dilution and 0.2 ml of inoculum after an adsorption time of 4 hours followed by 7 days of incubation at $36^{\circ}\text{C} \pm 1^{\circ}\text{C}$, and clear countable plaques were seen.

Incubation Time Period:-

Results of standardization of incubation time showed that plaque development occurred best after 7 days of incubation. (Table-6)

On 5th day of incubation the plaque development was poor as very small sized plaques were seen. On 7th day clear and distinct plaques were observed.

Table 6:- Effect of Time Period of Incubation on Plaque Development.

Incubation period	Results
5 days	Small sized plaques; poor development.
7 days	Clear distinct plaques.

After standardization of the different parameters for the establishment of the plaques assay on Vero cells, a standardized procedure was followed for the demonstration of the plaques assay. The final results were recorded and photographed. Image1 represents the appearance of plaques of Yellow fever virus in a 6-well tissue culture plates. The first well is inoculated with 10^{-1} dilution of the virus and no clear plaques were seen. The 2nd well was inoculated with 10^{-2} dilution and 0.2 ml of inoculum after an adsorption time of 4 hours followed by 7 days of incubation at $36^{\circ}\text{C} \pm 1^{\circ}\text{C}$, and clear countable plaques were seen.

Vaccine titration: The final results recorded after standardization of different parameters of Plaque assay method.

Table 7:- Vaccine titre values obtained by PFU Method.

S. No.	Titre values of test performed (Log ₁₀ PFU/0.5ML)
1.	4.50
2.	4.49
3.	4.40
4.	4.47
5.	4.46
6.	4.43
MEAN	4.45
S.D.	0.37

Test titre values obtained by PFU method are given in Table-7. The lowest and the highest titres were 4.40 log PFU/ 0.5ml and 4.50 log PFU/ 0.5ml, respectively with mean of 4.45; standard deviation (SD) of 0.037. The titres obtained from PFU method were all above the minimum requirement for Yellow fever vaccine potency *i.e.*, 4.24 log PFU/ 0.5ml.

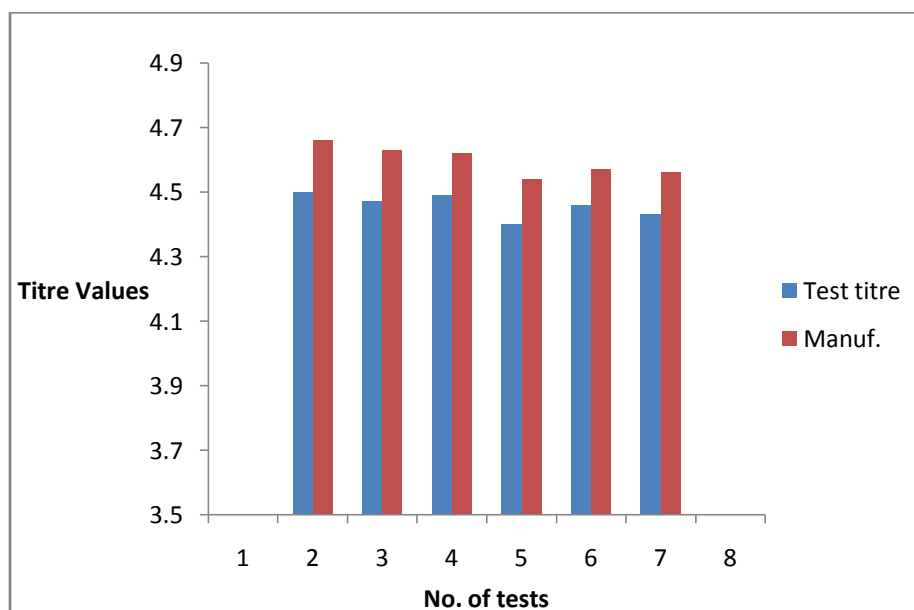
Table 8:- Vaccine titre along with IS titre obtained by PFU Method

S. No.	Titre values of test performed (Log10 PFU/ 0.5ml)		Titre values of IS (Log10 PFU/ 0.5ml)
Batch	A	B	
1.	4.50	4.47	4.72
2.	4.49	4.46	4.67
3.	4.40	4.43	4.69

Test was performed with two different batches of vaccine, A and B respectively, along with the test of IRS with PFU method. One sample from each batch was simultaneously titrated with IRS sample. The results obtained are given in Table-8.

Table 9:- Vaccine titre and Manufacturer's titre values.

S. No.	Titre values of test performed (Log10 PFU/ 0.5ml)	Titre values of manufacturer (Log10 PFU/ 0.5ml)
1.	4.50	4.66
2.	4.49	4.63
3.	4.40	4.62
4.	4.47	4.54
5.	4.46	4.57
6.	4.43	4.56
MEAN	4.45	4.59
S.D.	0.037	0.046
CORR.	0.916	

**Graph 1:-** The different results of titration of two batches A and B compared with the titre values of manufacturer.

6 different vials of Yellow fever 17D vaccine of two different batch numbers (A and B), were titrated for potency testing and were compared with the titre values of manufacturer. The results obtained are given in Table-9 and GRAPH-1. The lowest and the highest titres were 4.40 log PFU/ 0.5ml and 4.50 logs PFU/ 0.5ml, respectively with mean of 4.45; standard deviation (SD) of 0.037; and correlation coefficient (CORR.) was 0.91. The titres obtained from PFU method were all above the minimum requirement of Yellow fever vaccine potency *i.e.*, 4.24 log PFU/ 0.5ml. The assay was found to be reproducible.

Discussion:-

Vaccine potency of Yellow fever is routinely assay by Mouse LD₅₀ method (*in-vivo* method). Modern techniques in virology have shown that cell culture techniques are useful for potency and stability tests of many viral vaccines ^[14]. The potency test carried out in mice can be replaced by an alternative cell culture technique (*in-vitro*) for Yellow fever vaccine ^[13].

However, Mouse LD₅₀ method is sensitive with a long observation period *i.e.*, 21 days and involves large number of animals (8 for each dilution). On the other hand plaque technique is quite sensitive, accurate, less time consuming method for titration. The PFU method gives result in 7 days, and has no use of animals. Many problems of taking care of mice, their food, and breeding etc., and problems faced due to premature death, variation in animal immune status, sensitivity to particular immunological products and variability in results can be avoided.

Keeping in mind the guidelines of RRR, for use of laboratory animals, an attempt has been made through this study to establish Plaque Assay method by replacing MouseLD₅₀ method for potency testing, in CRI, Kasauli. As *in-vitro* Plaque Assay is already established as equivalent method to *in vivo* MouseLD₅₀ method, by WHO and also recommended in IP ^{[11][12]}.

The manufacturers of Yellow fever vaccine across the world are presently using PFU method to establish the titre of Yellow fever vaccine. In India, the titration of Yellow fever vaccine is being done by using mouse LD₅₀ or CCID₅₀ method. So to harmonize the methodology (as per the ICH guidelines) an attempt to adapt PFU method is made.

Different parameters were standardized for the plaque assay of Yellow fever vaccine on Vero cell line. The plaque assay was carried out in 6-well tissue culture plate. The first parameter was to standardize the cell count of Vero cells to be used per well of the 6-well tissue culture plate. The cell count which gave a complete monolayer formation after overnight incubation was the criteria for the selection. For that, varying number of cells, ranging from 2×10^5 to 5×10^5 per ml, were added to the individual wells of the 6-well tissue culture plate. The plate was incubated at $36^\circ\text{C} \pm 1^\circ\text{C}$ for overnight.

The cell count of approximately $3-4 \times 10^5$ cells/ml were found suitable as the cell number lower than this could not give a complete monolayer formation while on the other hand the cell count higher than $3-4 \times 10^5$ cells/ml, produced a confluent monolayer after overnight incubation, and produced a thick cell sheet and more than 50% of the cells were found rounded.

Standardization of different parameters for establishment of plaque assay was the next step of the study, such as dilution of the virus inoculum, size of inoculum, concentration of methyl cellulose and incubation time etc.

The dilution of the virus which could give at least 4.24 log₁₀ pfu/ 0.5ml was also standardized. The dilutions from 10⁻¹ to 10⁻⁵ were tested and finally 10⁻² dilution was selected, as it gave the desired pfu/ml.

In case of 10⁻¹ there is high pfu/ml and the entire sheet was found detached due to excessive infection as shown in Image1. At 10⁻³ dilution of the virus, very less number of plaques was seen. This may be due to low number of virus particles.

After the standardization of dilution of the virus, inoculum size of the virus was also standardized. For this, three different inoculum sizes were tested; 0.2ml, 0.3ml, and 0.4ml/well. Out of these, inoculum size of 0.2ml was found to be suitable for the study. With 0.3ml inoculum no major differences was observed. However, with inoculum size of 0.4ml, no plaques were seen due to excessive degeneration of cell line. So Yellow fever virus suspension adjusted to contain 4.24 log₁₀ pfu/ 0.5ml, and the inoculum size of 0.2ml/well of a 6-well tissue culture plate were finally selected for the plaque assay.

The effect of different adsorption time on the attachment and development of plaques was also studied. Two different adsorption times were studied *i.e.*, 1 hour and 4 hours. At 1 hour, no plaque development occurred as shown in Image-2 and 3. Best results were obtained at 4 hours of adsorption time and it was selected for the present study.

In order to localize the plaques, different concentrations of methyl cellulose were incorporated in the overlay media (MEM with 2% FBS) *i.e.*; 0.8%, 1%. The final concentration of 1% was found optimum for the plaque assay, as the Vero cells did not show any abnormal behaviour at this concentration. 1% MC just maintained the semi-solid nature of the overlay media, which is quite necessary for the localization of the plaques. The incubation period for the development of plaques was also standardized, and incubation period of 7 days was found to be adequate.

After standardization of different parameters, the reproducibility of the assay was also tested and was found to be good. The plaques were seen as clear, distinct and uniformly distributed. The titration of Yellow fever vaccine was performed on 6 different vials of two different batches along with the IRS and compared with the titre values of manufacturer potency assay. The results show titre above lower limit of 4.24 log PFU/0.5ml. The range of titre results was from 4.40 to 4.50 log PFU/0.5ml with mean of 4.45; SD of 0.037; and Corr. of 0.91.

Conclusion:-

The conclusions of the present study are:-

The plaque formation of Yellow fever 17D vaccine virus is affected by the inoculum size, dilution of virus, duration of adsorption, temperature and concentration of overlay media (MC).

From present study it can be concluded that plaque assay using Vero cell line can be used for potency testing. Mouse LD₅₀ method is sensitive with a long observation period *i.e.*, 21 days and involves large number of animals (8 for each dilution). On the other hand plaque technique is quite sensitive, accurate and less time consuming method for titration.

It is also concluded that PFU method as potency assay of Yellow fever vaccine is reproducible with a mean titre of 4.45; SD of 0.037; and Corr. Coff. Was 0.91. However, further studies are required for the establishment of plaque assay method for the potency testing of Yellow fever vaccine.

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RESEARCH ARTICLE

TO STUDY THE EFFECTIVENESS OF STRUCTURED TEACHING PROGRAMME ON KNOWLEDGE REGARDING CERVICAL CANCER AMONG WOMEN IN SANGANER DISTRICT, JAIPUR.

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Key words:-

Effectiveness, Structured teaching programme (STP), Knowledge, Women, Cervical cancer.

Abstract

Background: Most of the women keep struggling with cervical cancer because of barriers such as early detection and timely treatment which can be changed by adequate knowledge about the disease.

Aim: To assess effectiveness of Structured teaching programme (STP) on knowledge regarding cervical cancer among women in Sanganer district, Jaipur.

Objective: To assess pre-test and post-test knowledge regarding cervical cancer among women and find out association between knowledge regarding cervical cancer with their selected demographic variables.

Methodology: The design explains one group pre test post test design, quasi experimental design to measure the effectiveness of the structured teaching programme on knowledge regarding cervical cancer among rural women in selected area of Jaipur.

Result: Findings of study reveals that mean pre test knowledge score about cervical cancer is 9.65 and post test mean knowledge score is 17.87, structured teaching programme was found to be effective in increasing the knowledge regarding cervical cancer. No significant relationship was found between knowledge regarding cervical cancer and selected demographic variables in Pre test but in post test significant association between education and knowledge score were found. So more educated people had more knowledge score in post test.

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Introduction:-

Approximately 132,000 new cases are diagnosed and 74,000 die annually in **India**, accounting to nearly 1/3rd of the global **cervical cancer deaths**. Cervical cancer, mainly caused by Human Papillomavirus infection, is the leading cancer in Indian women and the second most common cancer in women worldwide. Though there are several methods of prevention of cervical cancer, prevention by vaccination is emerging as the most effective option. Early detection and timely treatment can reduce the burden of cervical cancer in India cancer

Background of the study:-

Cervical cancer is the major and devastating death for cause to young girls and their mothers, mortality and morbidity worldwide with an estimated take control of their health and help them protect themselves against cervical cancer. But most of them keep struggling with the disease because of lack of knowledge about the disease.

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Need of the study:-

70% or more cases found are of stage 3 or higher at diagnosis. Establishment of baseline indicators to monitor and evaluate cancer control programs, require data on incidence, clinical stage at presentation, and survival rate which concluded that all programs will be effective if they are able to persuade women with abnormal screening tests to attend for diagnosis and treatment, and facilities for treatment. Women, living in rural community need knowledge about the cervical cancer for early detection, screening and treatment about cervical cancer. A teaching programme for the women living in rural area will help them to know about etiological risk factor, signs symptoms, diagnosis and treatment aspects of cervical cancer which will help in early diagnosis and prompt treatment.

Aim and objective:-

- To assess pretest knowledge regarding cervical cancer among women.
- To find association between knowledge regarding cervical cancer with their selected demographic variables.
- To assess post test knowledge regarding cervical cancer among women.

Material and Methods:-

One group pre test_ post test was used on a sample of 60 women aged between 25 to 45, who were resident of Mangyawas of Sanganer . Subjects were enrolled by convenient sampling and data was collected by self reporting .

Tools:-

The questionnaire consisted of two sections. Section A consists of the questionnaire on demographic information of the respondent. Section B consists of 30 questions on knowledge of women regarding cervical cancer. The reliability coefficient (Pearson's co-efficient) was $r=0.739$. Each correct item was given a score of one and incorrect response a score of zero. The maximum scoring possible was 30 and minimum was zero. The scoring was categorised as

0—10 --- Poor

11---20 --- Satisfactory

21 -- 30 --- Good

Data Collection:-

Data was collected from the women aged between 25 to 45 years and residing in locality, Mangyawas of community health center of Sanganer. The study was conducted from 13 June 2015 to 26 June 2015. Prior permission was obtained through letter from Medical Officer of C.H.C of Mangyawas. Ethical permission was granted by committee in B.M.C.H.R.C, Jaipur

Result:-

Finding of study reveals that more than half of the women had poor knowledge regarding cervical cancer in pre test but of them none had in post test, i.e. score ranging from 01-10. Data revealed that 36.66% have satisfactory knowledge regarding cervical cancer in pre test were as in post test it raised to 85%, had scoring range from 11-20. None had scoring range from 21-30 in pre test and it reached to 9 in post test.

Table I:- Pre test Post test scoring of knowledge regarding cervical cancer N=60

SL. No	Scores	Pre test		Post test	
		Frequency	Percentage	Frequency	Percentage
1	01-10	38	63.33	0	0
2	11-20	22	36.66	51	85
3	21-30	0	0	9	15

Comparison of Pre test and Post test Knowledge score of samples

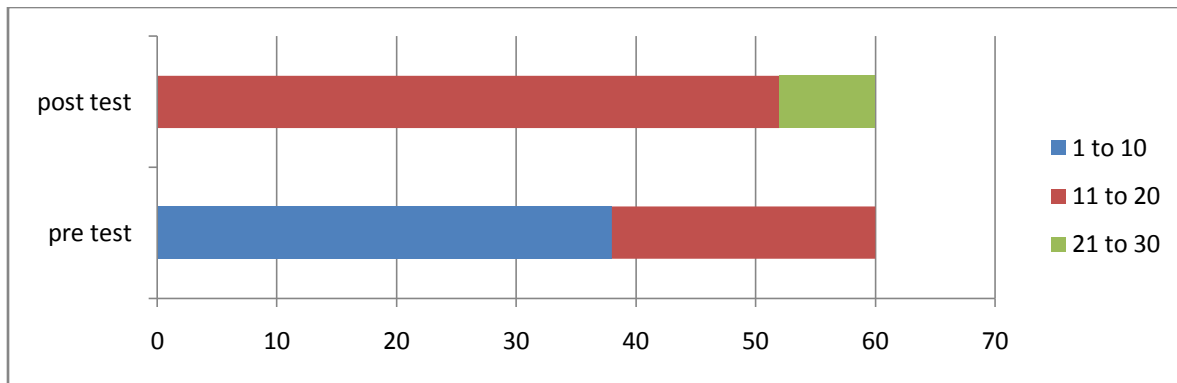


Figure 1:- Bar diagram showing comparison of pre test and post test knowledge s

Table Iii:- Effect of structured teaching programme N=60

Knowledge score	Mean score	Mean difference	Standard deviation Of difference	t value
Pre test	9.63	8.21	2.53	3.239
Post test	17.85			

t = 3.239, p = 5.52628E-31

p = 5.52628E-31 < 0.05 (level of significance) thus, shows that structured teaching programme is effective in increasing the knowledge regarding cervical cancer.

Table Iv:- Association between post test knowledge score and demographic variable N=60

S.No	ITEMS		11-20	21-30	Chi square	Table value	Result
1	Age in years	a)25-30	14	4	$\chi^2=0.990008$	7.81	No association
		b)31-35	12	2			
		c)36-40	19	2			
		d)41-45	6	1			
2	Educational Status	a)Secondary	39	2	$\chi^2=6.3725$	5.99	Significant association
		b)Senior secondary	10	6			
		c)Graduate	2	1			
		d)Post Graduate	0	0			

Discussion:-

The finding on the existing knowledge regarding cervical cancer among women shows that more than half of the women have poor knowledge score none of them have good. Mean knowledge score of pre test is 9.633 \pm 1.99. The finding is comparable to study by Nwankwo. K in 2010 where Nigerian women from urban and rural area were included in the study. Only 15.5% of the respondents were aware of availability of cervical cancer screening services. Only 4.2% had ever done Pap smear test. Hindrance in the use of available cervical cancer screening services were lack of knowledge (49.8%) and the feeling that they had no medical problems (32.0%).

The study shows that there is no significant relation between knowledge regarding cervical cancer and Age groups, Educational Status, Occupational status, Marital Status, Age of first intercourse, Number of children, Use of contraceptive pills, Pattern of Menstruation and Family history of Cervical Cancer. This also supports the finding of the study by Lee EE on Korean American women. It was founded that women never undergone Pap smear in the preceding 3 years irrespective of the variables as age, marital status, income, knowledge of early detection method for cervical cancer, and perceived beliefs about benefits of and barriers to receiving Pap tests. Components were suggested for the groups of Korean American women who never undergone Pap smear and for those who have not had one in the preceding 3 years. The common intervention strategies that aim to increase knowledge and perceived benefit and to decrease perceived barriers to receiving Pap tests.

Conclusion:-

From the researcher point of view screening is the first step for fighting against this deadly disease and until and unless this knowledge is propagated in women no program will be effective in control of cervical cancer. The structured teaching programme on cervical cancer is effective tool as demographic variables show little role in post test knowledge score. Women were ignorant about their health and have less knowledge regarding cervical cancer, so awareness camp should be organised to promote screening test. 'Cervical cancer' is curable, if detected early and in terminal stages it is the quality and not the quantity of life that really matters and is possible only by education and knowledge enhancement by awareness camps.



RESEARCH ARTICLE

INFLUENCE OF RESISTANCE TRAINING ON POWER AMONG UNDER GRADUATE STUDENTS.

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Key words:-

Resistance training, explosive strength, under graduate college female students

Abstract

The present study was to find out the effect of resistance training on power parameter among under graduate students. To achieve the purpose twenty under graduate female college students ($n = 20$) were selected as subjects and their age ranged between 18 and 25 years. They were divided in to two equal groups, each group consisted of ten ($n = 10$) subjects. The group I underwent resistance training (RTG), and group II acted as a control (CG) who did not exposed any special training apart from their regular activities. The training period of this study was limited to three days in a week for eight weeks. Explosive strength was selected as a criterion variable of this study and it was measured by using vertical jump. The analysis of covariance (ANCOVA) was applied as a statistical tool. In all cases 0.05 level of confidence was fixed to test the significant, which was considered as in appropriate. It was concluded from the result of the study that there was a significant improvement ($p \leq 0.05$) on explosive strength due to resistance training compared to control group.

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Introduction:-

Resistance training is a form of exercise that improves muscular strength and endurance. During a resistance training workout, you move your limbs against resistance provided by your body weight, gravity, bands, weighted bars or dumbbells. Some exercise machines can also be used for resistance training (<https://www.verywell.com/what-is-resistance-training>). Resistance training is the use of resistance to muscular contraction to build the strength, anaerobic endurance and size of skeletal muscles. Resistance training is based on the principle that muscles of the body will work to overcome a resistance force when they are required to do so. Resistance training has been general categorized into major type with different objectives: "strength type" and "hypertrophy-type". The former consists of high intensity exercises with two repetition and long rest periods between sets. This type of regimen is used for gaining strength. On the other hand, the hypertrophy type regimen consists of moderate intensity exercises with higher repetition and shorter rest periods between sets. This type of regimen has been thought to be effective in gaining muscle size (Kraemer, 2001). Recently, the use of a single particular type of regimen throughout the training period has become less common: instead, multiple training regimens are used in a periodized fashion, because it is thought that they course greater strength gains in muscles (Marx, 2001). When do the resistance training repeatedly and consistently, the muscles become fit. (<https://www.betterhealth.vic.gov.au/health/resistance-training-health-benefits>).

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Power is defined as the amount of work performed per unit of time. Power is an element of skill-related fitness that is needed to excel in athletic performance. Increased strength does not always translate into increased power. For example, a strong upper body lifts a high amount of weight. However a strong upper body does not always have the ability to throw a shot put very far if enough speed cannot be generated (www.livestrong.com.Sports and Fitness). Although power is a fundamental part of human relations, little is known about power in daily life. We studied the everyday experience of power by surveying individuals multiple times over 3 d regarding their subjective feelings of power and positional power (www.hydrol-earth-syst-sci.net). Power dynamics were a common, though not constant, experience. Rather than power being concentrated among a few individuals, almost half of participants reported sometimes holding high-power positions. People's feelings of power did not always map onto the positions they held. Most variability in power was related to people's changing situations rather than their stable traits. Low power negatively affected mood and cognition. Contrary to negative stereotypes of power holders, higher power was associated with greater feelings of responsibility (Smith & Hofmann, 2016).

Materials and Methods:-

The present study effort was made to find out the effect of resistance training on explosive strength of under graduate female college students. To achieve these purpose twenty (n = 20) under graduate female students were chosen as subjects from Physical Education Department, Calicut University, Kerala and their age ranged between 18 and 25 years. They were divided into two equal groups, each group consisted of ten (n = 10) subjects. The group I underwent resistance training (RTG) and group II acted as a control (CG) who did not exposed any special training apart from their regular activities. The training period of this study was limited to three days in a week for eight weeks. Explosive strength was selected as a criterion variable and it was measured by using vertical jump. For every training programmer there would be a change in various structures and system in a human body. The experimentation was subjected to eight weeks of resistance training. The training programme was scheduled for one session per day between 6 am to 7 am. Training was given under the direct supervision of the investigator. The progression of load was followed during experimentation. The following exercises were used to resistance training and performed with progressive methods squat, power clean, front squats, bench press, overhead press, dead lift, barbell row, pendlay row. The exercises were arranged from simple to complex.

Table-I:- Percentage of Intensity, Repetition and Sets of Resistance Training

Group I	Components	Weeks			
		I & II	III & IV	V & VI	VII & VIII
Resistance	Intensity	60%	70%	80%	90%
	Repetition	8 to 10	6 to 8	4 to 6	2 to 4
	Sets	2	2	3	3

Data Analysis:-

Mean and Standard deviation were calculated for explosive strength for each training group. And the data analyzed by using analysis of covariance (ANCOVA). Statistical significance was fixed at 0.05 levels.

Results:-

Table – II:- Analysis of Covariance on Explosive Strength of Resistance Training group and the Control Group

Test		RTG	CG	SOV	SS	df	MS	F ratio	
Pre test	Mean	21.9	22.1	B	.200	1	.200	.067	
	SD	1.6	1.8	W	53.80	18	2.9		
Post test	Mean	26.3	23.3	B	45.00	1	45.0	7.62*	
	SD	2.44	4.71	W	106.20	18	5.90		
Adjusted Post test	Mean	26.4	23.2	B	49.50	1	49.50	11.11*	
				W	74.95	17	4.40		

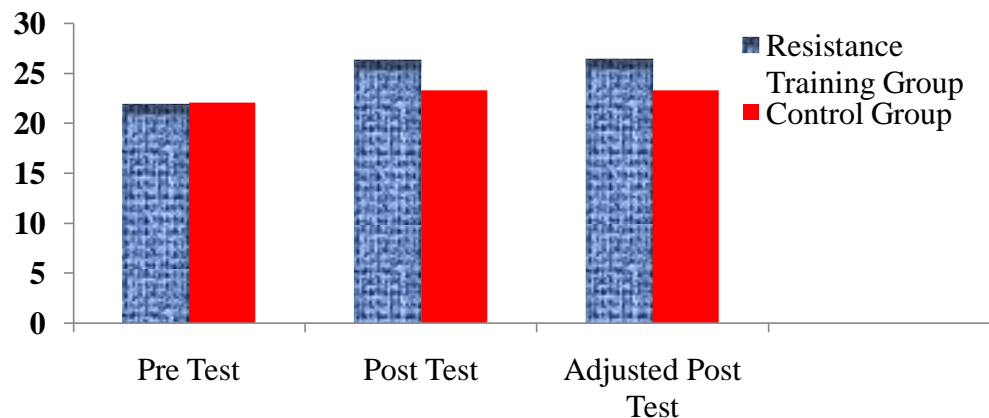
*Significant at 0.05 level of confidence

(The table value required for significance at 0.05 level of confidence with df 1 and 18 and 1 and 17 are 4.41 and 4.45 respectively)

Table II shows that the pretest mean values on explosive strength for the resistance training group and the control group are 21.9 and 22.1 respectively. The obtained F ratio of .067 for the pre test mean is less than the table value 4.41 for df 1 and 18 required for significance at 0.05 level. The post tests mean of resistance training and control group are 26.3 and 23.3 respectively. The obtained F ratio of 7.62 for post test mean is higher than the table value 4.41 for 1 and 18 required for significance at 0.05 level. The adjusted post test mean of resistance training group and the control group are 26.4 and 23.2 respectively. The obtained F ratio of 11.11 for adjusted post test mean is higher than the required table value 4.45 for df 1 and 17 required for significant at 0.05 level.

The result of the study indicated that there was a significant difference between the adjusted post test mean of the resistance training and the control group on explosive strength at 0.05 levels. The pre, post and adjusted post test mean values of the resistance training and the control group on explosive strength is graphically represented in the figure 1.

Figure 1:- The pre, post and adjusted post test mean values of resistance training group and the control group on explosive strength



Discussion:-

This study revealed that there was a significant improvement on explosive strength due to resistance training compared to control group. The result of this study is much related to other studies conducted before. Matavuji *et al.* (2001) conducted his study to evaluate the effects of resistance training on elite junior basketball players. The result of the study was proved that the training positively influenced the vertical jumping height of the selected subjects after the training duration. Markovic (2007) conducted his study to evaluate the effect of resistance training among the selected male subjects. The result of the study indicated that, the training was positively influenced the vertical jumping ability of the subjects as compared with the control groups. Martinez *et al.* (2011) included 40 women and 38 men medium level sprinters in their study. The resistance training was conducted for the duration of 8 weeks and reached the conclusion that the training was significantly improved the jumping ability of the both men and women subjects. Young *et al.* (1995) conducted his study to effects of resistance training on strength, power, among women. The result of the study was proved that the progressive resistance exercise can produce substantial increases in muscle strength and in power for women. Jeffrey *et al.* (2002) conducted his study to evaluate the effect of heavy- vs. light-load jump squats on the development of strength and power. The result of the study was proved that the investigation indicates that training with light-load jump squats results in increased movement velocity capabilities and those velocity-specific changes in muscle activity may play a key role in this adaptation. Moktar *et al.* (2008) conducted his study to evaluate of concurrent endurance and circuit resistance training sequence on muscular strength and power development. The result of the study was proved that the resistance circuit training alone induced strength and power improvements that were significantly greater than when resistance and endurance training were combined, irrespective of the intersession sequencing. So the present study clearly mentioned that there was a significant improvement made on explosive strength due to systematic of resistance training.

Conclusion:-

The resistance training can be improved explosive strength during the age between 18 and 25 years of under graduate female college students. The results of the study indicated that there was a significant improvement on explosive strength due to eight weeks of resistance training. From the result we recommended that the resistance training is very suitable to improve explosive strength.

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RESEARCH ARTICLE

TEACHING ENGLISH AS A SECOND LANGUAGE IN INDIA: A STUDY.

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Abstract

It is to note that today's world considers English as a global language because there has never been a language so widely spread or spoken by many people as English. English is one of the primary languages that is taught almost everywhere in the world. It has a lot of extended reach and influence around the world. The India has a tremendous amount of need to learn English as a second language. Every educational system has certain objectives which aim at bringing about desirable changes in pupil. In order to bring about those changes, the institutions arrange learning experience. The success of learning can be judged only in terms of the changes brought about by this experience and evaluation.

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Introduction:-

It is significant to note that English has become a world language rather than the language of only the English speaking countries such as the UK and the USA because the number of the people who use English as a means of communication exceeds much more than the number of the people who speak it as their mother tongue. In the case of English in India, more than two centuries, India has been directly and indirectly had influence of the language, English on all the fields, such as Education, Medical Science, etc. It examines the present status of English as a second language in India. This article lists different phases of learning English, defines language and identifies the factors that affect second language learning. It also asserts that it is essential for a language teacher to understand what a student is intending to say or communicate. This understanding forms the basis for a teacher to select a proper approach, method and technique that would be useful to his or her students. This paper also discusses different problems faced by teacher and learners of English in India. It also explains the need for a teacher to be resourceful and knowledgeable to meet the grave situation and promote better standards of achievement.

Teaching English As A Second Language:-

The Council of Chief State School Officers (CCSSO), U.S., defines English language proficiency in this way: A fully English proficient student is able to use English to ask questions, to understand teachers, and reading materials, to test ideas, and to challenge what is being asked in the classroom. Four language skills contribute to proficiency as follows:

1. Reading - the ability to comprehend and interpret text at the age and grade appropriate level.
2. Listening - the ability to understand the language of the teacher and instruction, comprehend and extract information, and follow the instructional discourse through which teachers provide information.
3. Writing - the ability to produce written text with content and format fulfilling classroom assignments at the age and grade-appropriate level.

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4. Speaking - the ability to use oral language appropriately and effectively in learning activities (such as peer tutoring, collaborative learning activities, and question/answer sessions) within the classroom and in social interactions within the school. Hence, the teacher should keep in mind while teaching English as a second language to the students.

English As A Second Language In India:-

English in India is a question of linguistic centralism while the other Indian languages lead to linguistic regionalism...A foreign languages existing so firmly and distinctly has posed a problem to the country .Yet the language problem became more complicated without any practical solution .In the process of teaching-learning ,the teacher should try to understand the student first .Then only he/she can enable the students to understand his/her teaching .Theory with practice on some of the teaching topics ,may enable the students to understand the concept easily. The growing modernization and internationalism in the world prevented us from doing away with the English language .Besides ,Indian languages are associated with tradition and are understood to be anti-modern .Therefore , a complete switchover to the Indian languages would lead to educational chaos and complete isolation from the developments on the international arena .We could not risk this because of the cultural ,social ,political and economic reasons .Hence ,this situation requires an urgent solution .

The only resolution that could be thought of was a need for coexistence of English with Indian languages .As a result, we had to define the role of English in India and its relationship with Indian languages .As a result, we had to define the role of English in India and its relationship with Indian languages .Further, the role of English was strengthened and consolidated as English was recognized and perceived as:

- The language of knowledge(science and technology)
- The language of liberal, modern thinking
- A window on the world
- The language of library

Thus, the three language formula came in to existence. This policy was proposed in 1956 by the central advisory board on education and was adopted at the Chief Ministers conference in 1961. The policy aimed at making English an integral part of the school education in India .this naturally restricted the learning and use of Hindi and the students started learning English as second language.

Common problems affecting teaching / learning english as a second language in india:-

There are so many factors that affect the teaching-learning process in India. The students in India can be categorized into two; the one is having the regional language as medium of study from the primary level and the other is having English as the medium of study. Hence, the problem of teaching English as a second language, to the Indian students starts from the pre schooling. Teaching of English in India has always been in difficult circumstances .This is so because of the large population of the country, bleak economic conditions, the cultural diversities, etc

Excess strength in class:-

With our large population we do not find any class where students number is less than 60.As the student in the class are always heterogeneous, there is hardly any time to pay attention to the weak or creative child .Even the average child do not get enough experience in the use of language .There is no scope for individual attention. This certainly, hinders the intellectual growth.

Lack of exposure:-

Most of the Indian students are exposed to their mother tongues .They do not get adequate opportunities either to listen to or speak in English .Because of the poor social and economic backgrounds ,they neither get enough exposure to English outside the classroom to improve themselves.

Lack of motivation:-

As students do not find any immediate need for English, their interest naturally lacks .Similarly, as there is no immediate reward for their achievement, the interest decreases. The poverty and the insecure sociological conditions also force them to neglect the language. Learning a foreign language is not an easy task and the much needed exposure and practice are not available to achieve fluency in the use of the language.

Lack of expert teacher:-

Good teachers of English are found in small number in India. Hence, not having a good teacher of English is common experience of all the students of English. The teachers of ENGLISH are either not trained by the teachers of English in India but not by the native speakers. Even the materials and methodology used in these training programs are outdated. As such, it is the same limited experience that is shared. There is hardly any scope to improve intuitive and spontaneous knowledge of the language. With all problems, discussed above, the standard of English in our country is deplorably low. Hence, a lot of responsibility is thrown on the shoulders of the teachers to make their classes interesting and learning of English fruitful. They have to gear up to meet the demands of the situation and successfully perform their duties.

Conclusion:-

This paper intends to bring facts that in our country about 75% of the students are from rural areas and they are coming through regional language medium schools. Keeping in mind the standards of the students and based on their background, we have to design the syllabus and adopt methods to test their English language proficiency. Therefore, it is necessary to go for a detailed discussion as to whether the existing curriculum is fulfilling the need of the hour and suitable to the students in achieving their goals. Taking into consideration of all the above points and undergoing a detailed analysis, a real solution may be found out for the betterment of the students as well as society.

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RESEARCH ARTICLE

SCREENING STUDY OF THE RELATIONSHIP BETWEEN TOXOPLASMOSIS AND ABORTION IN HAIL REGION, SAUDI ARABIA.

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Abstract

Background: Toxoplasmosis is one of the most common parasitic infections with variable prevalence in different countries caused by *Toxoplasma gondii*. The importance of toxoplasma mainly due to the risk of disease transmission during pregnancy and suggested its role in abortion. Chronic toxoplasmosis causes repeated abortion especially at first trimester.

Objective: This study aims to determine the proportion of women with Toxoplasmosis who experienced recurrent abortion among 2 years 1432 and 1433 and the relationship between the incidence of Toxoplasmosis and recurrent abortion in Hail with the effect of treatment.

Subjects and method: Data was collected from Maternity and Children's Hospital in Hail as a representative case in Hail, Saudi Arabia about women who had suffered from recurrent abortion (3 or more) among 2 years 1432 and 1433 and the seroprevalence of *Toxoplasma gondii* antibodies in pregnant women.

Results: indicate that the percentage of abortion due to toxoplasmosis in relation to total abortion due to different causes among 2 years (1432 & 1433) was found to be 2.85%. High percentage was found at first trimester (62.1%) followed by second trimester (31%) and then third trimester (6.9%). There is no significant difference between percentages either in year 1432 or 1433. High percentage of abortion due to *Toxoplasma gondii* was found in participant women with age of 25-45 years old.

Conclusion: We can conclude that it is of great importance to avoid raw or undercooked meat, preventing close contact with cats and early treatment from toxoplasmosis.

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Introduction:-

Toxoplasmosis is a parasitic infection caused by *Toxoplasma gondii* (*T. gondii*) with variable prevalence in different countries. Some studies have suggested its role in abortion (Ebadi et al., 2011). There are three main routes of

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toxoplasma transmission through ingestion of raw or undercooked meats, exposure to oocyst-infected cat feces, and vertical transmission. (Elmore et al., 2010).

Detection of IgM antibody in a woman who had abortion indicates the possible role of *T. gondii* in abortion phenomenon in Ahfaz, Southwest of Iran 2015 (Saki et al., 2015). The seroprevalence of *T. gondii* antibodies in pregnant women varies from the 6.1 to 75.2 % based on the geographical region (Elsheikha, 2008). This prevalence is 7.6% in India, 15-41% in Africa, 21% in Pakistan and Bangladesh, 31% in Ireland, 33% in Caribbean, 55% in France and 46% in other European countries (Remington et al., 2006). Other authors found that (53.14%) of women with recurrent abortion and (69.35%) of women with neonatal deaths tested positive IgM antibody against Toxoplasma. Although spontaneous abortion due to congenital toxoplasmosis is well known, it is yet controversial that to what extent toxoplasmosis can cause habitual abortion (Ebadi et al., 2011). Also Toxoplasma antibodies detected by positive serological tests tend to be higher with increased age and parity it seems that they have no role in habitual abortion, the study conducted between January 2000 and May 2001 at King Hussein Medical Center (Qublan et al., 2002). Toxoplasmosis can be involved in etiology of abortion and stillbirth (Ghasemi et al., 2016). The results indicate that the Type II *T. gondii* is associated with ovine abortion (Chessa et al., 2014). Other study indicates that chronic toxoplasmosis, most probably is significant cause of repeated abortion, also found that toxoplasmosis has a significant relation to abortion at first trimester (Sahwi et al., 1995). Also a higher percentage of antitoxoplasma IgG antibodies positivity in women with repeated abortion than the control group (Ebadi et al., 2011). Other study presents evidence of Toxoplasma infection in Quasseem, Saudi Arabia and directs the attention that toxoplasmosis is an important cause of repeated abortion there (El-Sebai, 1991). There is a relatively high seroprevalence of acute Toxoplasma infection in pregnant women, with the potential for transmission of the parasite to the fetuses (Al-Mohammad et al., 2010). Other study mentioned that one of every three pregnant women in Aydin was at risk of toxoplasmosis at the first trimester of their pregnancy (Ertug et al., 2005). Another study found that the seroprevalence increased significantly with age and was higher in the lower socioeconomic strata (Rossa et al., 2008).

Mwambe, et al. (2013) found that in *Mwanza, Tanzania* about 30.9% were sero-positive for *T. gondii*-specific antibodies. Infection was higher in urban than those from rural communities (41.5% versus 22.0%). Also, employed women were more likely to get infection than peasants (40.0% versus 25.9%). The risk of infection was increased by 7% yearly also with increased woman's age.

Therapies are based primarily on spiramycin because of the relative lack of toxicity and high concentration achieved in the placenta (Han et al., 1999). The effect of treatment with spiramycin on 122 women with recurrent abortions and 55 women with neonatal deaths who had tested positive for IgM antibody, 115 (94.26%) in current abortion group and 35(63.64%) in neonatal death group delivered normal babies (Zargar et al., 1999). Another study recommended that spiramycin treatment be started as soon as possible once the diagnosis of maternal Toxoplasma infection during pregnancy is proved or strongly suspected, because a prolonged time interval between onset of infection and start of treatment seems to be associated with the presence of severe fetal lesions at the time of prenatal diagnosis (Hohlfeld et al., 1989). Another study found a weak evidence for an association between early treatment and reduced risk of congenital toxoplasmosis (Leproust et al., 2007). Spiramycin may reduce the risk of materno-fetal transmission by 60%, but is in the present doses without influence on an already infected fetus. Pyrimethamine in combination with sulfonamides eradicates more effectively parasites in the placenta, and also in the fetus (Stray-Pedersen, 1992). The early start of treatment resulted in a significant reduction in the number of severely affected infant (Foulon et al., 1999).

Aim of the study:-

This study aims to determine the proportion of women with Toxoplasmosis who experienced recurrent abortion among 2 years 1432 and 1433. Also to discover whether there is a relationship between the incidence of Toxoplasmosis and recurrent abortion in Hail and the effect of treatment on this proportion.

Subjects and methods:-

Data were collected about women who had suffered from recurrent abortion among 2 years (Muharam 1432 to Dhu Al-hijah 1433) by making a survey of the medical records in Maternity and Children's Hospital. The data collected were related to the woman age, Number of pregnancies, Number of abortions, causes of abortions, exposure to treatment (duration and type).

Data were analyzed by using the SPSS software package (Chicago, IL, USA), and the chi-square test was used to compare proportions of cases in Toxoplasmosis infection. A statistically significant difference was assumed when the $p < 0.05$.

Results:-

Data was collected from Maternity and Children's Hospital in Hail as a representative case in Hail, Saudi Arabia. In table 1, the number of pregnant women among years 1432 and 1433 H was about 15319 women, about 50% in each year. The number of newborns among the same 2 years was 11252. The number of cases admitted to abortion department was 4057, and the number of abortion due to different causes was 1014 (6.62%); 7.28% in 1432 and 5.98% in 1433 in relation to the total number of pregnant women. Percentage of abortion due to toxoplasmosis in relation to total abortion due to different causes among years (1432 & 1433) was found to be 2.85% (2.90% for 1432 and 2.80% for 1433).

Table 2 showed the seropositivity of anti-toxoplasma among 2 years, for IgG (86.2%), IgM (0%) and Mixed IgG & IgM (13.8%). In year 1432, 1433 the percentage of positive IgG (93.8% & 76.9%), IgM (0%) and Mixed IgG & IgM (6.2% & 23.1%) respectively. Women with positive antibodies for toxoplasmosis have abortion at different gestational age among years (1432 & 1433 H). High percentage was found at first trimester (62.1%) followed by second trimester (31%) and then third trimester (6.9%). There is no significant difference between percentages either in year 1432 or 1433 H (Fig 1).

Table 2 showed also the frequency of abortion due to toxoplasmosis among 2 years (1432 & 1433) was found to be 69% for women that have abortion for 1-2 times and 13.8% (3-4 times) and 17.2% (≥ 5 times). These percentages have some variations among years (1432 & 1433), women with abortion 1-2 times (62.4% & 76.9%) and abortion for 3-4 times (18.8% & 7.7%) and abortion for ≥ 5 times (18.8% & 15.4%) respectively (Fig 2).

Table 3 showed that the age of participant women varies from < 16 to > 45 years old. High percentage of abortion due to toxoplasma gondii was found in participant women with age of 25 – 45 years old. From age of 25 – 34 years old was found to be (41.4%) and from 35-45 years old was found to be 44.8% among 2 years 1432 & 1433. Low percentage was found in women with age of < 16 – 24 years old (14.8%) and zero % in women with age > 45 years old (Fig 3). Causes of toxoplasmosis among years 1432 and 1433 H was found to be due to cats (3.5%) and due to other causes (96.5%) like uncooked meat, water and others (table 3). Treatment with spiramycin or pyrimethamine in combination with sulfonamides eradicates more effectively parasites in the placenta, and also in the fetus (data not shown).

Discussion:-

The importance of toxoplasma from the perspective of public health is mainly due to the risk of disease transmission during pregnancy (Elsheikha, 2008). Between 350 and 1,000 HIV-infected persons with seropositive for toxoplasmosis and have impact of congenital and AIDS-related symptomatic toxoplasmosis on morbidity and mortality in Kyrgyzstan (Minbaeva et al., 2013).

In this study, the number of abortion due to different causes was 1014 (6.62%), 7.28% in 1432 and 5.98% in 1433 in relation to the total number of pregnant women. Percentage of abortion due to toxoplasmosis in relation to total abortion due to different causes among 2 years (1432 & 1433 H) was found to be 2.85% (2.90% for 1432 and 2.80% for 1433). Elsheikha et al., (2008) said that the seroprevalence of toxoplasma antibodies in pregnant women varies from 6.1% to 75.2% based on the geographical region. In pregnant women worldwide, there are seroprevalences of toxoplasmosis from 7% to 51.3% and in women with abnormal pregnancies and abortions the seroprevalences vary from 17.5% to 52.3% (Galvan-Ramirez, et al., 1995). The prevalence of anti-Toxoplasma antibodies in Japan was 10.3%, and it was significantly higher in women aged above 35 years (Sakikawa et al., 2011), and in pregnant women in Tanzania (Mwambe, et al., 2013). In year 2012, congenital toxoplasmosis was estimated in France at 2.58 per 10,000 live; births and 204 cases of congenital toxoplasmosis were observed (Nogareda et al., 2014).

Our finding reported that, the sero-positivity of anti-toxoplasma among years (1432, 1433), for IgG (86.2%), IgM (0%) and Mixed IgG & IgM (13.8%). Our result was higher than the result of Vado-Solis et al. (2013). They found in Yucatan, Mexico that, 55.2% of the samples were positive to IgG, (3.4%) to IgM, (8.6%) to IgG and IgM. It is also higher than that reported by Ashraf et al. (2010), they found 41.90 % were seropositive for anti *T. gondii* IgG and 11.88 % for anti *T. gondii* IgM. It is also higher than that reported by Al-Harhi, et al. (2006) among pregnant

women in Makkah (Saudi Arabia) was (29.4%) for anti-Toxoplasma IgG, (5.6%) for anti-Toxoplasma IgM. Other authors reported that among pregnant women in Hebron district (Palestine) was (27.9%) for anti-IgG and (17.6%) for anti-IgM (Nijem and Al-Amleh, 2009). Other authors reported lower prevalence of Toxoplasma IgG (17.9%) and Toxoplasma IgM (12.8%) in Gaza city (Al-Hindi, 2009).

In Accra, Ghana, Seropositivity was found to be 51.2 % pregnant women for IgG anti-*T. gondii* antibodies, but 0% for mixed IgG and IgM, (Ayi, et al., 2016). The result of Murata et al., (2016) indicate that serum of pregnant women were positive for IgG (50%), and for IgM (91.9%).

This study found that high percentage of abortion was found at first trimester (62.1%) followed by second trimester (31%) and then third trimester (6.9%). This result was in agree with the result of Zargar, et al. (1998) who found that 48% of the abortions had occurred in first trimester followed by 36% and 16% in second and third trimester respectively.

In the current study, the frequency of abortion due to toxoplasmosis among years (1432 & 1433 H) was found to be 69% for women that have abortion for 1-2 times and 13.8% (3-4 times) and 17.2% (≥ 5 times).

In this study, high percentage of abortion due to toxoplasma gondi was found in participant women with age of 25 – 45 years old. From age of 25 – 34 years old was found to be (41.4%) and from 35-45 years old was found to be 44.8% among 2 years 1432 & 1433. Low percentage was found in women with age of <16 – 24 years old (14.8%) and zero % in women with age >45 years old. Our findings were in agree with the study carried out in Makkah Al-Mukarramah, the highest IgG and IgM seroprevalence were among participants aged 35 to 43 years (48.8% and 12% respectively) (Al-Harthi, et al., 2006). And also in agree with the study of Ashraf et al. (2010) in Sana'a, Yemen, participants women aged 31-40 years (45.3%), then age group 21-30 years 42.1%. found that toxoplasma antibodies detected by positive serological tests tend to be higher with increased age (Qublan et al., 2002). In Japan, the overall prevalence of anti-Toxoplasma antibodies was significantly higher in women aged above 35 years (Sakikawa et al., 2011).

Caroline Paquet, RM et al. (2013) stated that, spiramycin should be offered for fetal prophylaxis if pregnant women were infected but the infection not reached the fetus. Combined therapy of pyrimethamine, sulfadiazine, and folic acid should be started with toxoplasmosis infected women and fetus. Repeat testing should be performed within 2 to 3 weeks, if acute infection is suspected starting therapy with spiramycin immediately without waiting for the repeat test results. Moreover, treatment of pregnant women can modify the results of serological tests (Rodrigues, et al., 2014; Pomares and Montoya, 2016).

In this study, causes of toxoplasmosis among 2 years 1432 and 1433 was found to be due to cats (3.5%) and due to other causes (96.5%) like uncooked meat, water and others. On the other hand, Al-Hindi (2009) said that animal breeding was reported for 33.3% of the aborted women. A history of raw meat intake, exposure to oocyst-infected cat feces, and vertical transmission was identified to be a risk factor related to toxoplasmosis (Skariah et al., 2010; Elmore et al., 2010; Sakikawa et al., 2011).

Recommendations:-

Our study showed a moderate seroprevalence of *T. gondii* antibodies among pregnant women in Hail, KSA. Hence we recommend the implementation of health extension program among pregnant women in order to prevent primary infection during pregnancy. Screening of women for Toxoplasma antibodies before and during pregnancy and periodic surveillance of the disease among population all over the country is also recommended. It is of great importance to avoid raw or undercooked meat, handling raw meat safely and preventing close contact with cats. Also, the early start of treatment with spiramycin or pyrimethamine in combination with sulfonamides resulted in a significant reduction in the number of severely affected infant.

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Table 1:- Correlation between the total numbers of pregnant women, number of abortion due to several causes, number of cases admitted to abortion department and the number of abortion due to *Toxoplasma gondi* in Maternity and Children's Hospital in Hail, Saudi Arabia.

Year of Investigation		Number of pregnant women	Number of newborns	Number of cases admitted to Abortion Department	Number of Abortion due to different causes	Number of Abortion due to <i>Toxoplasma gondi</i> .
1432 H	First 6 months	3734	2704	1030	266 [7.12%]	11 (4.14%)
	Last 6 months	3823	2726	1097	284 [7.43%]	5 (1.76%)
	Total	7557	5430	2127	550 [7.28%]	16 (2.90%)
1433 H	First 6 months	4012	3020	982	268 [6.68%]	5 (1.87%)
	Last 6 months	3750	2802	948	196 [5.23%]	8 (4.08%)
	Total	7762	5822	1930	464 [5.98%]	13 (2.80%)
Total 1432 & 1433		15319	11252	4057	1014 [6.62%]	29 (2.85%)

[] Percentage change from number of pregnant women.

() Percentage change from number of Abortion due to different causes.

Table 2:- Seropositivity of Anti- *Toxoplasma* IgG and IgM in relation to the gestational age of abortive women due to *Toxoplasmosis* infection and frequency of abortion among 2 years (1432 & 1433 H) in Maternity and Children's Hospital in Hail, Saudi Arabia.

Year of Investigation		Gestational age			Antibodies For Toxoplasmosis			Frequency of Abortion			Total
		1st Trimester	2 nd Trimester	3 rd Trimester	+ ve IgG only	+ ve IgM only	+ ve IgG + ve IgM	1 -2	3- 4	≥ 5	
1432 H	First 6 months	7 (63.6%)	4 (36.4%)	0 (0%)	10 (90.9%)	0 (0%)	1 (9.1%)	8 (72.7%)	1 (9.1%)	2 (18.2%)	11
	Last 6 months	3 (60%)	1 (20%)	1 (20%)	5 (100%)	0 (0%)	0 (0%)	2 (40%)	2 (40%)	1 (20%)	5
	Total	10 (62.5%)	5 (31.3%)	1 (6.2%)	15 (93.8%)	0 (0%)	1 (6.2%)	10 (62.4%)	3 (18.8%)	3 (18.8%)	16
1433 H	First 6 months	5 (100%)	0 (0%)	0 (0%)	4 (80%)	0 (0%)	1 (20%)	3 (60%)	1 (20%)	1 (20%)	5
	Last 6 months	3 (37.5%)	4 (50%)	1 (12.5%)	6 (75%)	0 (0%)	2 (25%)	7 (87.5%)	0 (0%)	1 (12.5%)	8
	Total	8 (61.5%)	4 (30.8%)	1 (7.7%)	10 (76.9%)	0 (0%)	3 (23.1%)	10 (76.9%)	1 (7.7%)	2 (15.4%)	13
Total 1432 & 1433		18 (62.1%)	9 (31.0%)	2 (6.9%)	25 (86.2%)	0 (0%)	4 (13.8%)	20 (69%)	4 (13.8%)	5 (17.2%)	29

() Percentage of cases from the total number of women with abortion due to *Toxoplasma gondi* infection.

Table 3:- Correlation between causes of abortion, women gestational age and participant's age in Maternity and Children's Hospital in Hail, Saudi Arabia among 2 years (1432 & 1433H).

Year of Investigation		Gestational Age			Abortive Women Age				Causes of Abortion		Total
		1st Trimester	3 rd Trimester	2nd Trimester	<16 - 24	25 - 34	35- 45	>45	Cats	Others	
1432 H	First 6 months	7 (63.6%)	4 (36.4%)	0 (0%)	2 (18.2%)	4 (36.4%)	5 (45.4%)	0 (0%)	0 (0%)	11 (100%)	11
	Last 6 months	3 (60%)	1 (20%)	1 (20%)	0 (0%)	1 (20%)	4 (80%)	0	1 (20%)	4 (80%)	5
	Total	10 (62.5%)	5 (31.3%)	1 (6.2%)	2 (12.5%)	5 (31.2%)	9 (56.3%)	0	1 (6.2%)	15 (93.8%)	16
1433 H	First 6 months	5 (100%)	0 (0%)	0 (0%)	0 (0%)	3 (60%)	2 (40%)	0 (0%)	0 (0%)	5 (100%)	5
	Last 6 months	3 (37.5%)	4 (50%)	1 (12.5%)	2 (25%)	4 (50%)	2 (25%)	0 (0%)	0 (0%)	8 (100%)	8
	Total	8 (61.5%)	4 (30.8%)	1 (7.7%)	2 (15.4%)	7 (53.8%)	4 (30.8%)	0 (0%)	0 (0%)	13 (100%)	13
Total 1432 & 1433		18 (62.1%)	9 (31.0%)	2 (6.9%)	4 (14.8%)	12 (41.4%)	13 (44.8%)	0 (0%)	1 (3.5%)	28 (96.5%)	29

() Percentage of cases from the total number of women with abortion due to *Toxoplasma gondi* infection.



Fig 1:- Women with different gestational age of abortion due to Toxoplasma infection among years (1432 & 1433 H) in Maternity and Children's Hospital in Hail, Saudi Arabia.

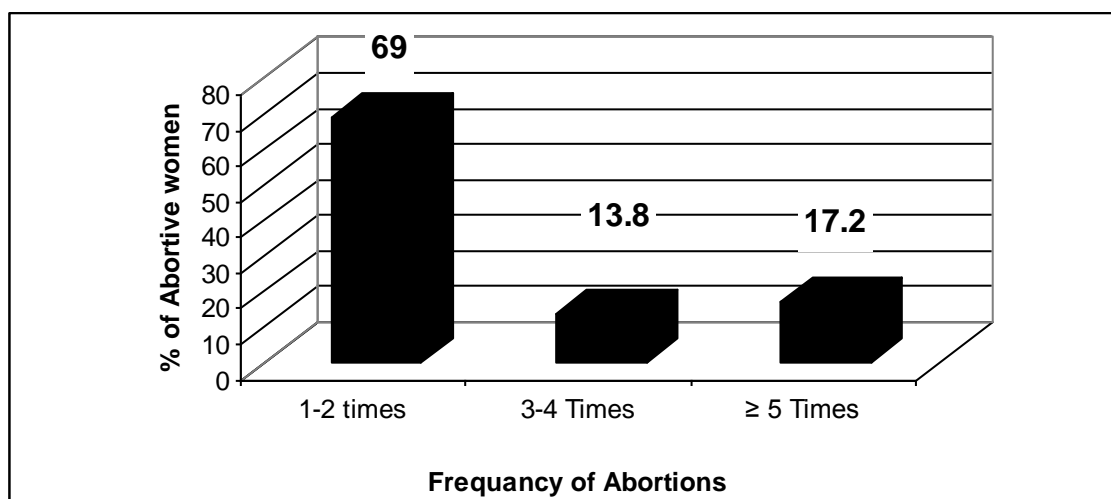


Fig 2:- Frequency of abortion due to Toxoplasma infection among 2 years (1432 & 1433 H) in Maternity and Children's Hospital in Hail, Saudi Arabia.

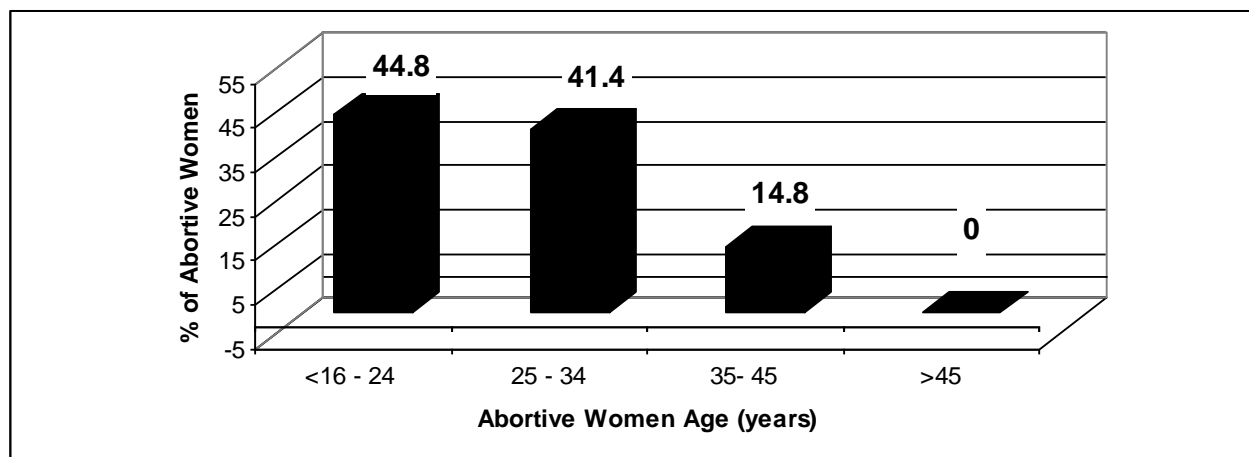


Fig 3:- Abortive Women Age due to Toxoplasma infection among 2 years (1432 & 1433 H) in Maternity and Children's Hospital in Hail, Saudi Arabia.

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RESEARCH ARTICLE

STUDY ON THE BACTERIAL ISOLATES FROM HAND SWAB SAMPLES OF HEALTH CARE WORKERS AND ANTI-BIOTIC SENSITIVITY PATTERN , IN A TERTIARY CARE HOSPITAL , LUCKNOW

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Abstract

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Introduction:-

The contaminated hands of health care workers (HCW), play an important role in transmission of hospital infection, and is a common pattern seeing in most healthcare settings[1]. An important cause of morbidity and mortality among hospitalized patients worldwide, is Healthcare-associated infections (HCAIs), also known as nosocomial infections[2]. In developed countries, HCAs have been reported to affect 5%-15% of hospitalized patients and 9%-37% of those admitted to intensive care units (ICUs), while in developing countries , prevalence rates have been estimated to be between 14.8% and 19.1%[3]. The single most important measure to prevent cross-transmission of microorganisms and to reduce the rate of nosocomial infection is to maintain hand hygiene[4]. The landmark study carried out by Semmelweis in 1884 , demonstrated that the simple act of hand washing could save lives especially when health care workers do it routinely and thoroughly[5]. The organisms transmitted can be harmful for both patients and health-care workers ,and these organisms are sometimes resistant to antimicrobial agents (AMA) .Still, health-care workers(HCW), do not practice hand washing[6].

To reduce such infections in hospital, a targeted surveillance culture to identify asymptomatic carriers of multidrug resistant bacteria and subsequent isolation and treatment should be done. To predict possible risk of infection with multi-drug resistant bacteria in health-care settings , the sensitivity patterns of isolates should also be studied.

This study was therefore undertaken in a TERTIARY CARE HOSPITAL , LUCKNOW as part of a wide scale hospital infection control surveillance programme. It seeks to ascertain the proportion of bacterial colonization of the isolates from the hands of healthcare workers who are in regular contact with patients, and to determine multidrug resistant isolates for possible intervention.

Materials and Methods:-

This study was carried out in Era's Lucknow Medical College & Hospital, Lucknow, which is a tertiary care hospital , in the month of August 2015. Ethical clearance was taken from the Institutional Ethics Committee.

A total of 60 hand swab samples were collected from 60 healthcare workers of the Hospital. Of all the healthcare workers, 50 (83.3%) were female nurses and 10 (16.7%) were male nurses, which were choosen randomly. Sterile cotton wool swabs, which were pre- moistened in sterile normal saline, were used to swab the interdigital spaces, dorsal and ventral aspect of hands of the participants. The media used in this study were Blood Agar, MacConkey

Agar and Mueller Hinton Agar. The swabs from hands were cultured on Blood agar and MacConkey agar plates. The plates were incubated at 35°C for 24-48 hrs. The bacterial isolates were identified using bacteriological procedures, involving microscopy, morphology, and biochemical tests.

All detected bacterial isolates were tested for antimicrobial susceptibility test by the standard Kirby-Bauer disc diffusion method, according to the Clinical Laboratory Standard Institute (CLSI) guidelines. The test organism was picked up with a sterile loop, suspended in peptone water and incubated at 37°C for 2 h. The turbidity of the suspension was adjusted to 0.5 McFarland's standard. It was then spread on the surface of Mueller-Hinton Agar (MHA) plate using sterile cotton swab. After drying the plates (37°C for 30 minutes) antibiotic discs were applied by sterile forceps. *Staphylococcus aureus* ATTC 25923 and *Escherichia coli* ATTC 25922, *P. aeruginosa* ATCC 27853 were used as controls during the study.

Results:-

A total 60 hand swab sample, were collected from 60 healthcare workers of Era's Lucknow Medical College & Hospital, Lucknow, among which 50 (83.3%) were female nurses and 10(16.7%) were male nurses. Of the 60 samples analysed, 48 (80%) yielded bacterial growth, while 12 (20%) showed no bacterial growth. Out of the 48 isolates, 36 (75%) were Gram negative, and 12(25%) were Gram positive organisms. Also out of total 50(83.3 %) females nurses, 30(60 %) had Gram negative and 8 (16 %) had gram positive bacteria; while out of 10(16.7 %)male nurses, 6(60%) had Gram negative and 4 (40%) had Gram positive bacteria.(Table1)

TOTAL NO. (%)	TOTAL GRAM NEGATIVE	TOTAL GRAM POSITIVE
FEMALE NURSES- 50 (83.3)	30 (60%)	8 (16%)
MALE NURSES – 10 (16.7)	6 (60%)	4 (40%)

TABLE 1:- (Distribution of Gram negative and Gram positive bacteria among female and male nurses)

Also out of total 38 bacterial isolates in female nurses,30(78.9 %) were Gram negative and 8 (21.05 %) were gram positive bacteria ;while out of 10 bacterial isolates in male nurses, 6 (60%) were Gram negative and 4 (40%) Gram positive bacteria.(Table 2)

SOURCE		TOTAL NMBER OF ISOLATES	GRAM NEGATIVE	GRAM POSITIVE
HAND SWABS	FEMALE NURSES	38	30(78.9%)	8(21.05%)
	MALE NURSES	10	6(60%)	4(40%)

TABLE 2:- (Distribution of Bacteria among nurses, out of total bacterias isolated)

Of the 60 samples analyzed, bacterial isolates were seen in 48. Out of total 48, organisms were identified as 22(45.8%) *Escherichia coli*, 4(8.3 %) *Pseudomonas aeruginosa* ,10(20.8 %) *Klebsiella pneumoniae* and 12 as *Staphylococcus spp*, of which 8(16.6 %) were *Staphylococcus aureus* and 4 (8.3 %) were *Coagulase Negative Staphylococcus(CONS)*.(Table 3)

TOTAL BACTERIAL ISOLATES	48
<i>Escherichia coli</i>	22(45.8%)
<i>Pseudomonas aeruginosa</i>	4(8.3%)
<i>Klebsiella pneumoniae</i>	10(20.8%)
<i>Staphylococcus aureus</i>	8(16.6%)
CONS	4(8.3%)

Table 3- Different bacterias isolated in the study

Out of 22(45.8 %) *Escherichia coli* isolated, 20(90.9%)were in female nurses and 2(9.1%)were in male nurses. Out of 4(8.3 %) *Pseudomonas aeruginosa*, all the 4(100%)were in female nurses and none was found in male nurse 0(0%).

Out of 10(20.8 %) *Klebsiella pneumoniae*,6(60%)were in female nurses and 4(40%) were in male nurses.

12were *Staphylococcus spp*, of which 8(16.6 %) were *Staphylococcus aureus* and 4 (8.3 %) were CONS.

Out of 8(16.6%) *Staphylococcus aureus*, 6(75%) were in female nurse and 2(25%) in male nurses.

Out of 4 (8.3 %)CONS, 2(50%) were in female nurses and 2(50%) were in male nurses. (Table 4)

ORGANISMS	ORGANISMS FOUND IN FEMALE NURSES	ORGANISMS FOUND IN MALE NURSES
<i>ESCHERICHIA COLI</i> 22(45.8%)	20(90.9%)	2(9.1%)
<i>PSEUDOMONAS AERUGINOSA</i> 4(8.3%)	4(100%)	0(0%)
<i>KLEBSIELLA PNEUMONIAE</i> 10(20.8%)	6(60%)	4(40%)
<i>STAPHYLOCOCCUS AUREUS</i> 8(16.6%)	6(75%)	2(25%)
<i>CONS</i> 4(8.3%)	2(50%)	2(50%)

Table 4- Distribution of total Bacteria isolated among female and male nurses

Out of total 50(83.3%) female nurses, *Escherichia coli* were isolated from 20(40 %) female nurses, *Pseudomonas aeruginosa* were isolated from 4(8 %) female nurses, *Klebsiella pneumoniae* were isolated from 6(12 %) female nurses; and *Staphylococcus spp* were isolated from 8(16 %) female nurses ; of which 6(75%) were *Staphylococcus aureus* and 2(25 %) were *CONS*.

Out of total 10(16.7%) male nurses, *Escherichia coli* were isolated from 2(20 %) male nurses, *Pseudomonas aeruginosa* were isolated from 0(0%) male nurse, *Klebsiella pneumoniae* were isolated from 4(40 %) male nurses; again, *Staphylococcus spp* were isolated 4(40 %) male nurses ; of which 2(50%) was *Staphylococcus aureus* and 2(50 %) was *CONS*.(Table 5).

TOTAL FEMALE NURSES 50(83.3%)	<i>ESCHERICHIA COLI</i> 20(40%)	<i>PSEUDOMONAS AERUGINOSA</i> 4(8%)	<i>KLEBSIELLA PNEUMONIAE</i> 6(12%)	<i>STAPHYLOCOCCUS AUREUS</i> 6(75%),OUT OF 8(16%)	<i>CONS</i> 2(25%),OUT OF 8(16%)
TOTAL MALE NURSES 10(16.7%)	2(20%)	0(0%)	4(40%)	2(50%),OUT OF 4(40%)	2(50%), OUT OF 4(40%)

Table 5- Distribution of bacterial isolates among total female and male nurses

The proportion of gram negative bacteria was high 36(75%),when compared to gram positive bacteria 12(25%),out of total 48 isolates. The antibiotic sensitivity pattern were as follows:-

Escherichia coli showed 100% sensitivity to Ciprofloxacin, Doxycycline, Amikacin, Gentamycin, Levofloxacin ;but were resistant to Amoxycillin/ clavulanic acid(81.8%), Ceftriaxone(72.7%)and Co-Trimoxazole(63.6%).Total ESBL producers were 72.72%.

Pseudomonas aeruginosa showed 100% sensitivity to Ciprofloxacin, Levofloxacin, Piperacillin/Tazobactam, Imipenem; but were resistant to Ceftazidime(75%), Cefepime(50%), Gentamycin(25%), Cefotaxime(75%).

Klebsiella Pneumoniae showed 100% sensitivity to Ciprofloxacin, Doxycycline, Amikacin, Gentamycin, Levofloxacin ; but were resistant to Amoxycillin/ clavulanic acid(60%), Ceftriaxone(60%) and Co-Trimoxazole(50%).Total ESBL producers were 60%.

Staphylococcus aureus showed 100% sensitivity toAmikacin; but were resistant to Cefoxitin, Levofloxacin(12.5%),Amoxycillin/ clavulanic acid (25%),Ceftriaxone(62.5%),Clindamycin(25%),Erythromycin(50%),Ciproflaxacin(50%). Total MRSA was 12.5%. *CONS* showed 100% sensitivity to Cefoxitin, Levofloxacin, Amikacin; but were resistant to Amoxycillin/ clavulanic acid (25%),Ceftriaxone(75%),Clindamycin(25%),Erythromycin(50%),Ciproflaxacin(50%). All of the *CONS* were Methicillin susceptible.(Table 6,7,8).

ORGANISMS	AMC	CIP	DO	AK	G	LE	CTR	COT	CAZ	CPM	PIT	IPM	CTX
<i>ESCHERICHIA COLI</i> -22(45.8%)	4(18.18%)	22(100%)	22(100%)	22(100%)	22(100%)	22(100%)	6(27%)	8(36%)					
<i>PSEUDOMONAS AERUGINOSA</i> -4(8.3%)		4(100%)			3(75%)	4(100%)			1(25%)	2(50%)	4(100%)	4(100%)	1(25%)
<i>KLEBSIELLA PNEUMONIAE</i> -10(20.8%)	4(40%)	10(100%)	10(100%)	10(100%)	10(100%)	10(100%)	4(40%)	5(50%)					

Table:6- Antibiotic Sensitivity Pattern Of Gram Negative Bacteria

ORGANISMS	AMC	CIP	AK	LE	CTR	CX	CD	E
<i>STAPHYLOCOCCUS AUREUS</i> - 8(16.6%)	6(75%)	4(50%)	8(100%)	7(87.5%)	3(37.5%)	7(87.5%)	6(75%)	4(50%)
<i>CONS</i> - 4(8.3%)	3(75%)	2(50%)	4(100%)	4(100%)	1(25%)	4(100%)	3(75%)	2(50%)

Table : 7- Antibiotic Sensitivity Pattern Of Gram Positive Bacteria

ORGANISMS	AMC	CIP	DO	AK	G	LE	CTR	COT	CAZ	CPM	PIT	IPM	CTX	CX	CD	E
<i>ESCHERICHIA COLI</i> -22(45.8%)	4(18.18%)	22(100%)	22(100%)	22(100%)	22(100%)	22(100%)	6(27%)	8(36%)								
<i>PSEUDOMONAS AERUGINOSA</i> -4(8.3%)		4(100%)			3(75%)	4(100%)			1(25%)	2(50%)	4(100%)	4(100%)	1(25%)			
<i>KLEBSIELLA PNEUMONIAE</i> -10(20.8%)	4(40%)	10(100%)	10(100%)	10(100%)	10(100%)	10(100%)	4(40%)	5(50%)								
<i>STAPHYLOCOCCUS AUREUS</i> - 8(16.6%)	6(75%)	4(50%)		8(100%)		7(87.5%)	3(37.5%)							7(87.5%)	6(75%)	4(50%)
<i>CONS</i> -4(8.3%)	3(75%)	2(50%)		4(100%)		4(100%)	1(25%)							4(100%)	3(75%)	2(50%)

Table :8- Antibiotic Sensitivity Pattern Of Both Gram Positive And Gram Negative Bacteria

(AMC=30mcg, CIP=5mcg, DO=30mcg, AK=30mcg, G=10mcg, LE=5mcg, CTR=30mcg, COT=25mcg, CAZ=30mcg, CPM=30mcg, PIT=100/10mcg, IPM=10mcg, CTX=30mcg, CX=30mcg, CD=2mcg, E=15mcg).

Discussion:-

A total of 60 healthcare workers, which included 50 female nurses and 10 male nurses, were included in this study, which were chosen randomly. Out of the 60 hand swabs samples analysed, 48 (80%) yielded bacterial growth, while 12 (20%) showed no bacterial growth.

Out of the 48 isolates, 36 (75%) were all Gram negative, and 12 (25%) were gram positive organisms. Of 48 isolates, organisms were identified as 22 (45.8%) *Escherichia coli*, 4 (8.3%) *Pseudomonas aeruginosa*, 10 (20.8%) *Klebsiella pneumoniae* and 12 as *Staphylococcus* spp, of which 8 (16.6%) were *Staphylococcus aureus* and 4 (8.3%) were *Coagulase Negative Staphylococcus* (CONS). Among the gram negative organisms, 16 (72.72%) ESBL producers were *Escherichia coli* and 6 (60%) ESBL producers were *Klebsiella pneumoniae*. Out of 8 *S. aureus*, 1 (12.5%) was MRSA. Among CONS, all of them were Methicillin sensitive.

This can be compared to a study carried out by Maheshwari *et al.* [7], which included 70 HCWs, comprising 20 doctors, 20 nurses, 20 operation theatre (OT) technicians and 10 were laboratory technicians, from whom a total of 140 swabs were collected; and the organisms which were isolated were *S. aureus* in 13 (18.6%) and *coagulase negative staphylococcus* in 44 (63%).

Out of 13, *S. aureus* isolates 6 (46.2%) were MRSA. Gram negative bacteria (GNB) were isolated in 22 (31.5%), out of which 15 (68.2%) were ESBL producers. Among the 15 ESBL producers, 9 were *Klebsiella pneumoniae*, 5 were *Escherichia coli* and 1 was *Proteus mirabilis*.

Sarfraz *et al.* [8] conducted a similar type of study, comparing doctors and nurses of clinical department; and doctors and staff of non-clinical department. Organisms isolated were *Staphylococcus aureus*, *Acinetobacter baumannii*, *Pseudomonas aeruginosa*, *Coagulase negative Staphylococcus* spp., *Acinetobacter lwoffii*, *Escherichia coli* and *Corynebacterium* spp., from hands of clinical staff (doctors + nurses). Organisms isolated from non-clinical staff was *Acinetobacter lwoffii*, *Coagulase negative Staphylococcus* spp., *Staphylococcus aureus*, *Acinetobacter baumannii*, *Pseudomonas aeruginosa* and *Micrococcus* spp.

Survana Sande *et al.* [9], also showed in their study, a high rate of bacterial colonization in hand swab samples of HCWs, and proved the importance of hand washing, by showing very low rate of bacterial colonization in hand swab samples after hand washing. In their study, out of total 150 samples collected from Nursing staffs before hand washing, growth of microorganisms was observed in 107 (71.3%) samples and no growth in 43 (28.7%) samples. No growth was obtained in 128 (85.3%) samples and single type of growth in 22 (14.7%) samples, after hand washing.

Of the 60 samples analyzed in our study, maximum samples showed growth of *Escherichia coli*. *E. coli* is Gram-negative, aerobic and facultative anaerobic bacteria [10]. *E. coli* is an emerging nosocomial pathogen causing serious problems in health care settings [11]. This species leads to both intestinal and extraintestinal infections in humans and many animals. Currently, six major groups of intestinal pathogenic *E. coli* (IPEC) have been recognized: enteropathogenic *E. coli* (EPEC), enteroaggregative *E. coli* (EAEC), Shiga toxin-producing *E. coli* (STEC), enterotoxigenic *E. coli* (ETEC), enteroinvasive *E. coli* (EIEC), and diffusely adherent *E. coli* (DAEC).

Three types of extraintestinal pathogenic *E. coli* (ExPEC) are also recognised, including neonatal meningitis-causing *E. coli* (NMEC), sepsis-causing *E. coli* (SEPEC), and uropathogenic *E. coli* (UPEC), associated with meningitis in newborns, systemic infections, and urinary tract infections (UTIs), respectively [12].

In our study, *Escherichia coli* showed 100% sensitivity to Ciprofloxacin, Doxycycline, Amikacin, Gentamycin, Levofloxacin; but were resistant to Amoxycillin/clavulanic acid (81.8%), Ceftriaxone (72.7%) and Co-Trimoxazole (63.6%). Total ESBL producers were 72.72%.

Pseudomonas aeruginosa showed 100% sensitivity to Ciprofloxacin, Levofloxacin, Piperacillin/Tazobactam, Imipenem; but were resistant to Ceftazidime (75%), Cefepime (50%), Gentamycin (25%), Cefotaxime (75%).

Klebsiella Pneumoniae showed 100% sensitivity to Ciprofloxacin, Doxycycline, Amikacin, Gentamycin, Levofloxacin; but were resistant to Amoxycillin/clavulanic acid (60%), Ceftriaxone (60%) and Co-Trimoxazole (50%). Total ESBL producers were 60%.

Staphylococcus aureus showed 100% sensitivity to Amikacin, (87.5%) to Cefoxitin and Levofloxacin, (75%) to Amoxicillin/ clavulanic acid, (75%) to Clindamycin, (50%) to Erythromycin, (50%) Ciprofloxacin, and least sensitive to Ceftriaxone (37.5%). 1 (12.5%) was MRSA.

CONS were (100%) sensitive to Cefoxitin, Amikacin, Levofloxacin; (75%) to Amoxicillin/ clavulanic acid, Clindamycin; (50%) to Erythromycin, Ciprofloxacin, and least sensitive to Ceftriaxone (25%). All of the *CONS* were Methicillin sensitive.

In a study carried by Edem EN *et al.* [13], the antibiotic sensitivity pattern of the *Staphylococcus aureus* showed 60% to Oxacillin, 80% to Clindamycin, 43.3% to Erythromycin, 40% to Ceftriaxone, 77% to Ciprofloxacin, 23.3% to Trimethoprim Sulphamethoxazole and 73.3% to Amoxicillin clavulanic acid. 13% of the *Staphylococcus aureus* were found to be inducible clindamycin resistant. The antibiotic sensitivity pattern of *Staphylococcus epidermidis* showed 56.2%, 75%, 19%, 81.3%, 12.5% and 68.8% to Erythromycin, Clindamycin, Ceftriaxone, Ciprofloxacin, Trimethoprim Sulphamethoxazole and Amoxicillin clavulanic acid, respectively. 6.25% of the *Staphylococcus epidermidis* were found to be inducible Clindamycin resistant. *Escherichia coli* showed 100% sensitivity to Ciprofloxacin, Ceftriaxone, Gentamycin, Ceftazidime and Cefotaxime, but were resistant to Amoxicillin clavulanic acid and Cefpodoxime. It was non-ESBL producing. *Proteus mirabilis* showed 100% sensitivity to Gentamycin, Ceftriaxone, Amoxicillin clavulanic acid and Cefotaxime. It was ESBL producing.

MRSA isolates showed 0%, 50%, 91%, 33.3% and 75% sensitivity to Oxacillin, Erythromycin, Clindamycin, Ceftriaxone, Ciprofloxacin, Trimethoprim Sulphamethoxazole and Amoxicillin clavulanic acid, respectively, and 16.6% of the MRSA isolates were inducible Clindamycin resistant.

Chaka *et al.* [14], in their study took samples from dominant hands of staff nurses, pediatric residents and medical interns and their cell phones. *Staphylococcus aureus* strains isolated from hand swabs were resistant to oxacillin, vancomycin and ceftazidime in 46%, 24% and 44% respectively. The resistance pattern of *Staphylococcus aureus* from cell phone isolates were 51.6%, 14% and 51% respectively for oxacillin, vancomycin and ceftazidime. *CONS* isolated were also resistant to commonly prescribed antibiotics.

Kumar *et al.* [15] in their study showed that almost 25% of healthcare workers are stable nasal carriers and 30%-50% of them possess the bacteria in their hands. Tammelin *et al.* [16] also showed in their study that 50.7% of healthcare workers carry bacteria in their nose and 26.3% in their hands.

Several studies have shown that most of the bacteria that cause nosocomial infections are those that have developed resistance to antibiotics used in treating those [17].

Conclusion:-

Since the group of individuals under this study were healthcare workers, their interaction and exposure to hospital environment could cause major risks in transmitting to hospital patients and spreading nosocomial infections.

So, by simple measure of hand washing, various infections can be controlled.

Healthcare workers should wash their hands regularly with antiseptic soap, or disinfect the hand by rubbing with alcohol solution.

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RESEARCH ARTICLE

GASTROPROTECTIVE EFFECT OF A NEW FORMULATED MILK TABLET ON ETHANOL-INDUCED GASTRIC MUCOSAL INJURY IN RATS.

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Abstract

FM is a newly formulated milk tablet that provides the supplementary food such as milk and honey, also fortified it with high nutritive value components (bee pollen, palm pollen, Royal jelly, Propolis, Ginseng, and Bee pollen) that have many remarkable biological activities such as antioxidant, antibacterial and anti-inflammatory features. The current study aimed to investigate the potential gastroprotective effects of FM against ethanol-induced gastric injury in male albino rats. The experimental set included four groups; Control group, an ulcer control group treated with absolute ethanol (10 ml/kg), FM pretreated groups at doses (157 and 314 mg/kg per oral). The gastric secretion was collected for assessment of pH. The involvement of malondialdehyde (MDA), catalase (CAT), nitric oxide (NO) and tumor necrosis factor- α (TNF- α), alanine aminotransferase (ALT), aspartate aminotransferase (AST), alkaline phosphatase (ALP), Uric acid, and creatinine were also measured. Histopathological studies were undertaken to evaluate the influence of the treatment on the gastric wall mucosal layer. FM suppressed gastric inflammation induced as a result of ethanol administration by decreasing of tumor necrosis factor- α (TNF- α), nitric oxide (NO) and neutrophil infiltration. Meanwhile, FM halted gastric oxidative stress via reduction of MDA level with concomitant enhancement of the activity of a potent antioxidant CAT. These favorable actions were associated with attenuation of the severity of ethanol gastric mucosal injury as evidenced by the morphological and histological observations. Together, these findings postulated the potent gastroprotective action of FM against ethanol-induced gastric injury which was mediated via suppression of gastric inflammation and oxidative stress besides boosting of the antioxidant defense.

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Introduction:-

Gastrointestinal ulcer disease poses a serious economic load as a treatment for the ulcers account to at least 10% of the total cost of treating digestive disorders (Brown and Everhart, 1994; Sandler *et al.*, 2002). Peptic ulcer is a common disorder of the entire gastrointestinal tract. Moreover, about 8 to 10% of the global population affected by peptic ulcers, approximately 5% of them suffer from gastric ulcers (Calam and Baron, 2001). The ulcers that affect

the gastrointestinal system are usually aggravated by a disproportion between destructive and defensive factors in the stomach (Calam and Baron, 2001). The major causes of peptic ulcer disease are stress, alcohol, chronic use of non-steroidal anti-inflammatory drugs (NSAIDs), cigarette smoking, genetic predisposition, diet, and *Helicobacter pylori* infection (Nemeroff, 1982).

Alcohol consumption has been commonly associated with gastric mucosal lesions including gastritis, gastric ulcer, and even gastric carcinoma (Franke *et al.*, 2005). The mechanisms underlying ethanol-induced gastric ulcer have not been fully defined. Yet, mounting evidence has indicated that oxidative stress, proinflammatory cytokines and apoptosis play crucial roles in its pathogenesis (Al Batran *et al.*, 2013; Mei *et al.*, 2012; Sangiovanni *et al.*, 2013). Ethanol is widely used to induce experimental gastric ulcer in animals (Loguercio *et al.*, 1993). The experimental model of ethanol-induced gastric injury mimics several features of the human condition and thus provides a mean for assessing agents with potential anti-ulcer actions along with their implicated mechanisms for gastric protection (Liu *et al.*, 2012; Salga *et al.*, 2012). The administration of these drugs has been associated with several adverse effects such as nausea, constipation, gynecomastia and impotence that limit their use (Halabi *et al.*, 2014). Besides, most of these drugs, lack an effect on other factors involved in ulcer disease and therefore, do not achieve all treatment goals. Thus, the search for effective agents with fewer side effects has been regarded as an effective strategy for the management of gastric ulcers (Antonisamy *et al.*, 2014; Halabi *et al.*, 2014). Previously, agents with antioxidant features have displayed beneficial actions in protecting against alcohol-induced gastric ulcer (Antonisamy *et al.*, 2014; Liu *et al.*, 2012; Mei *et al.*, 2012). Hence, there is a strong need for more effective antioxidant, and safe anti-ulcer agents with different protective mechanisms from natural resources.

The primary function of milk is to meet the complete nutritional requirements of human, making it a nutrient dense food. The recognition of milk's richness in nutrients that play a key-role in human physiology has in recent years led to a wide variety of novel applications in dairy technology (Livney, 2010).

New formula of healthier milk tablet aims to improve nutritional status and health. An optimum formula of milk tablet was formulated to minimize cost and maximize nutritional value. Providing the supplementary food such as milk and honey and fortified it with a high nutritive values component (Skim milk powder, honey, bee pollen, palm pollen, Royal jelly, propolis and ginseng). The advantages of new formulated milk tablets with high nutritive value are long shelf-life, convenience, and suitable for the distribution to remote areas. Skimmed milk powder is widely used due to its easy storage facility and immediate availability on hand in all conditions. Milk is a very important substitute for food in most circumstances. So using of skimmed milk powder is safer in such conditions as it contains all the nutritional constituents of calcium, proteins, many vitamins and carbohydrates and no fat.

Most people think of honey as excellent food, but some others consider it as medicine (Zaghloul *et al.*, 2001), but only recently its antioxidant property has come to limelight. With increasing demand for antioxidant supply in the food, honey is becoming important source of antioxidant since it is rich in flavonoids and phenolic acids and other antioxidants including glucose oxidase, catalase, ascorbic acid, carotenoid derivatives, and organic acids. The antioxidants have several preventative effects against different diseases like cancer, cardiovascular diseases, inflammatory disorders, neurological degeneration, wound healing, infectious diseases and aging, which led to search for foods rich in antioxidants (Khalil *et al.*, 2010). Beside all previous benefits for honey it could be as a natural binder for dairy product ingredients.

Bee pollen consider as a valuable therapeutic product greatly appreciated by the natural medicine because of its potential medical and nutritional applications. It demonstrates a series of actions such as anti-inflammatory, hepatoprotective, and immune-stimulating. The use of bee pollen as food supplement has been proposed to ameliorate various inflammatory diseases (Martín-Muñoz *et al.*, 2010). However, there are very few studies about using bee pollen in food and dairy industry. On the other side, palm pollen grains are a good economic nutritional source can be used as human food supplements (Fernández, 1983).

Royal jelly (RJ) is a secretion of the cephalic glands of nurse bees and serves as the most important part of honeybee larvae diet, playing a major role in caste differentiation (Moritz and Southwick, 1992). RJ, one of the most effectual and beneficial remedy for human beings, is widely used both in folk and in official medicine and it is a controversial dietary supplement. Due to its complex composition (water, proteins, lipids, carbohydrates, amino acids, mineral salts, vitamins, enzymes, hormones, and natural antibiotics), RJ has a multitude of pharmacological activities: antioxidant, hypoglycemia, hepatoprotective, antitumor, antibiotic, and anti-allergic (Mărghitaș, 2008). Moreover,

propolis acts as an antibacterial factor. In addition, propolis is a powerful antioxidant (Özcan and Ayar, 2003). These effects are due to its high concentration of phenolics and other antioxidant compounds. Ginseng, the root of *Panax* species, is a well-known folk medicine. It has been used as traditional herbal medicine in China, Korea and Japan for thousands of years and today it is a popular and worldwide used natural medicine. The active ingredients of ginseng are ginsenosides, which are also called ginseng saponins (Kamal *et al.*, 2016). Research suggests that the bioactive components of ginseng, such as ginsenosides and polysaccharides, can decrease oxidative stress, which is a mechanism associated with reduced chronic disease development (Sun, 2007; Uno and Nicholls, 2010).

Yet, the impact of this newly formulated milk tablet (FM) on gastric ulcer has not been previously explored. Thus, the present study aimed to investigate the potential gastroprotective actions of FM in ethanol-induced gastric injury in rats.

Materials and Methods:-

Chemicals

All common chemicals used were purchased from one of the following suppliers Sigma Co. (St. Louis, MO, USA). All other reagents were of the highest grade commercially available. (All chemicals used in the study were of analytical grade).

Experimental Animals

Adult male rats, weighing (150±20) g. They were kept under observation for about 15 days before the onset of the experiment to exclude any infection. The chosen animals were housed in plastic cages with good aerated covers at 25°C ± 0.5°C as well as 12 h light/dark cycles. Animals were allowed free access to water and were supplied daily with a standard diet. The study design was approved by Ain Shams University Research Ethics Committee.

Preparation of new formulated milk tablet (FM)

The ingredients of FM consist of skim milk powder with different proportion as 50, 55, 60, 65 and 70% in parallel with honey proportion as 28, 23, 18, 13 and 8%, respectively then added bee pollen 10%, palm pollen 5%, Royal jelly 5%, propolis 1% and ginseng 1% for each formula. Preparation of FM was prepared in the Dairy Technology Department Animal Production Research Institute. All ingredients were mixed then compressed into tablet form. Milk tablet samples were packed into airtight plastic bags and stored in the refrigerator for further analysis (AOAC, 2006). The chemical composition of milk tablet ingredients and Formulations of milk tablet from mixture design was shown in tables (1, 2). In addition, chemical composition of selected formula of new formulated milk tablet was illustrated in Table (4).

Mucosal injury induction

Gastric ulceration was induced in the animals according to the procedure described by (Huang *et al.*, 2014). Briefly, rats were deprived of food but had free access to water 24 h prior to ulcer induction then, they were administered, with a single oral dose of 75% ethanol (10 ml/kg body weight). Two hours after ethanol administration, animals were sacrificed.

Animal grouping and treatments

Thirty two male rats were randomly divided into four groups (n = 8 per group): Group 1 (normal control) animals received only distilled water. Rats in group 2 (ulcerated control) were given only 75% ethanol (10 ml/kg). Group 3 and 4 animals were given FM at doses 157 mg/kg and 314 mg/kg, respectively, for 4 weeks, then at day 29, 75% ethanol (10 ml/kg) was given orally to each animal to induce gastric ulceration. Two hours after ethanol administration, all rats were sacrificed and stomachs were removed, opened along the greater curvature and gastric content was drained into a centrifuge tube.

Blood Samples preparation

Blood was collected from sacrificed rat in vacutainer and centrifuged 3000 rpm for 10 min. Plasma samples were collected and stored at -80 until used for biochemical analysis.

Collection of gastric juice for determination of gastric pH

Gastric juice from each animal was centrifuged at 1000 rpm for 10 minutes after adding 5 ml distilled water to remove any solid debris and the volume of the supernatant was measured. The supernatant was then assayed for determination of the pH of gastric juice using a pH meter (Sabui *et al.*, 2015).

Preparation of stomach homogenate for biochemical investigations

The gastric tissue was homogenized in 10ml ice-cold phosphate buffer (PBS, 100 mM, pH 7.4). The homogenized tissue (1g of tissue /10ml PBS) was centrifuged at 4000rpm for 15 minutes. The supernatant was removed and stored at -80°C for determination of oxidative stress markers and cellular defenses markers.

Assessment of oxidant/antioxidant activities

The determination of gastric mucous membrane lipid peroxides, expressed as malondialdehyde (MDA), was carried out according to the thiobarbituric acid assay (Liu *et al.*, 2012). Using the TBA commercial kit (spectrum diagnostics Egyptian company of biotechnology, Cairo, Egypt). Catalase (CAT), and nitric oxide (NO) levels were tested in the gastric tissue homogenate using the CAT and the NO assay kits (spectrum diagnostics Egyptian company of biotechnology, Cairo, Egypt), following the manufacturer's instructions.

Assessment of TNF- α level

The plasma TNF- α level was measured using the Rat TNF-alpha (TNF- α) ELISA kit (KOMA BIOTECH INC., Seoul, Korea) according to the manufacturer's instructions.

Assessment of the liver and kidney functions

The functioning of the liver was assayed by evaluating the alanine aminotransferase, aspartate aminotransferase, alkaline phosphatase, markers of hepatocellular damage, were established by colorimetric assay using a commercial kit (spectrum diagnostics Egyptian company of biotechnology, Egypt). The functioning of the kidney was assayed by evaluating the creatinine and uric acid by colorimetric assay using a commercial kit (spectrum diagnostics Egyptian company of biotechnology, Egypt).

Histological evaluation of gastric injury

From all experimental groups, the stomach of each rat was removed, opened along its greater curvature and rinsed with saline to remove the gastric contents. The stomach was stretched on a piece of cork with the mucosal surface facing upward and was then photographed with a digital camera. The histological evaluation of the gastric injury was performed on the specimens of the glandular stomach of three rats randomly selected from each group of the experiment. In brief, stomachs were fixed in the aqueous Bouin's fluid, embedded in paraplast, and sections were made at a thickness of 5-7 μ m and stained with hematoxylin and eosin (Humason, 1972).

Statistical analysis

Statistical evaluation was conducted with Instat Program GraphPad. Software, Inc, San Diego, USA, version 3.6, Copyright©1992-2003 Results were expressed as mean \pm SEM. The results were analyzed for statistical significance by one way ANOVA followed by Tukey-Kramer multiple comparison post-test. Values of $p < 0.05$ were regarded as significant.

Results:-

Sensory acceptability Consumer acceptance test for products form and mixture design experiment:-

Consumer acceptance test was performed at the Animal Production Research Institute as presented in Table (3) it could be observed that the rate between the milk powder and honey had a highly effect in all sensory attributes. Formulations (E) and (A) showed the lowest scores in the overall acceptability, it was recorded 70 and 76 points. Highest percent of skim milk powder cooperates with the lowest percent of honey in the formula (E) affected tablets properties as hard tablets with weak and pail flavor. On the other hand the lowest percent of skim milk powder and the highest percent of honey as formula (A) gave weak form and body and over sweetness flavor. More acceptance scores recorded by formulas (B) and (D) respectively. From the same table it could be observed that formula (C) gained the highest scores owing to its balance in the all nutritive value and overall sensory acceptability.

Effect of FM on PH:-

Fig.1a shows the effect of (FM) on gastric secretions of ethanol ulcerated rats. EtOH administration caused significant ($p < 0.05$) increase in pH value as compared to control group. Pre-treatment with the FM at both doses produced insignificant increase in pH value when compared with ulcerated control rats. While a significant increase was observed ($p < 0.001$) in both doses as compared to control group.

Effect of (FM) on biochemical parameters of stomach tissue:-

The gastroprotective effect of 157mg/kg and 314mg/kg doses of (FM) on the ethanol-induced gastric ulcer was estimated with various gastric parameters. There were remarkable changes in the gastric parameters of ulcerated and (FM) pre-treated groups as compared with the control animals.

Evaluation of Antioxidant and enzymatic activities:-

The effect of (FM) on the ulceration process in ethanol-induced ulcer was evaluated by the oxidative stress marker level, MDA, catalase activity and gastric NO (Fig.1).

Administration of ethanol triggered an oxidative stress as indicated by significant increase of MDA level ($p < 0.001$) as compared to control group. Pretreatment with both concentrations of (FM) significantly protect the gastric tissue against the oxidative stress as evidenced by significant ($p < 0.05$, $p < 0.001$ respectively) lowering of MDA as compared to ulcer control rats. On the other hand, the gastric ulcer induced group showed a significant increase ($p < 0.001$) in catalase activity in comparison to the normal control. Pretreatment with FM, showed a significant ($P < 0.05$) reduction in the catalase activity at the low dose, and a significant increase at the high dose ($P < 0.05$) as compared to ulcer group. While, both doses of (FM) significantly significant elevate ($p < 0.001$) the enzyme level as compared to control group. Rats subjected to EtOH administration (ulcer control rats) showed significantly decreased NO content ($p < 0.05$) as compared to normal control rats. (FM) pretreatment at both doses significantly increased NO content ($p < 0.001$, $p < 0.001$ respectively) as compared to ulcer group.

Evaluation of plasma TNF- α level:-

Administration of ethanol resulted in a robust inflammatory response as indicated by a marked elevation of serum TNF- α ($p < 0.001$) as compared to the control group (Fig.1c). Pretreatment with (FM) in both doses significantly ($P < 0.001$) lowered the levels of TNF- α when compared to ethanol ulcer group.

Evaluation of liver function:-

EtOH administration did not significantly affect the levels of AST, ALT, and ALP as compared to control group (Fig.2a). (FM) pretreatment at high dose significantly ($p < 0.05$, Fig.2a) decrease AST level as compared to control rats. In addition, both doses of (FM) showed a significant ($p < 0.01$, $p < 0.05$, respectively) decrease of ALP level as compared to ulcer control rats. (Fig.2a).

Evaluation of kidney function:-

EtOH administration did not significantly affect the levels of creatinine, and uric acid as compared to control group (Fig.2b). (FM) pretreatment at low dose significantly ($p < 0.05$, Fig.2b) increased the uric acid level as compared to ulcer control rats, while no significant change was observed in creatinine level at both doses of (FM) as compared to ulcer control rats (Fig.2b).

Microscopic Evaluation of the gastric injury:-

The morphological observations of the stomach of the control group showed that it is pink in colour without any hemorrhagic areas and its mucosa is thrown into prominent longitudinal folds, rugae, (Fig. 3a). Moreover, the ethanol induced gastric injury in the ulcer group that include hyperemia and multiple linear hemorrhagic red bands of different size along the glandular portion of the stomach indicating that the gastric ulcer was completely formed (Fig. 3b). In addition, the FM pre-treatment groups exhibited a dose-dependent protective effect against ethanol-induced gastric injury, with a marked decrease in the gastric mucosa injury and a significant ulcer inhibition compared with the ulcer group at the standard dose and maximal ulcer inhibition at the double standard dose (Figs. 3c and 3d).

The histological observations of the gastric mucosa of the control group revealed that it consists of the lining epithelium, lamina propria and muscularis mucosae (Fig. 4a). The lining epithelium is formed of simple columnar epithelial cells (Fig. 4b). In addition, the lamina propria consists of connective tissue that contains numerous straight tubular gastric glands (Fig. 4a). These glands are lined by three main types of cells; the luminal lining epithelium, oxyntic and peptic (chief or zymogenic) cells (Figs. 4b and 4c). Moreover, the muscularis mucosae is built of smooth muscle fibers (Fig. 4a).

The ethanol induced extensive gastric mucosal lesions in the ulcer group which extended to the two internal thirds or the entire mucosal thickness (Fig. 5a). These lesions included highly necrotic mucosa, epithelial lifting, mucosal

hemorrhage and leukocyte infiltration in the lamina propria and submucosa (Figs. 5a-5c). Moreover, some mucosal cells showed necrosis (Fig. 5d) and others possess pyknotic nuclei (Fig. 5e).

The mucosal damage induced by absolute ethanol was significantly decreased by FM pre-treatment and the mucosa was relatively intact. FM has been shown to exert the cytoprotective effects in a dose-dependent manner. The rats pre-treated with standard diet of FM showed markedly better reductions in gastric lesion and comparatively better protection of the gastric mucosa, (Figs. 6a-6c), as proven by very superficial injury only, (Fig. 6d), and reduction of leukocyte infiltration (Fig. 6a). Moreover, rats treated with a double standard dose of FM showed more or less normal histological architecture of the gastric mucosa (Fig. 7a) and demonstrated significantly better mucosal protection and absence of leukocyte infiltration (Figs. 7b and 7c).

Discussion:-

Milk made formulations provide a simple, novel and patient friendly approach for the delivery of ionized as well as unionized hydrophobic drugs. Incorporation of milk in the formulation results in solubilization of drugs with enhanced pharmacokinetic properties. Taste masking and gastroprotective properties also prove it a promising carrier for drugs. Further in-vitro studies are yet to be carried out to elucidate the colloidal stability of resulting emulsion, particle size and distribution of milk component (Kamal *et al.*, 2016).

The current study highlights, for the first time, the protective role of FM, a new formulated milktable, against ethanol-induced gastric injury in rats. Ethanol is commonly used for inducing ulcer in the experimental rats and leads to intense gastric mucosal injury. Furthermore, alcohol causes indirect destructive effects via the recruitment of leukocytes which drives oxidative stress, inflammatory responses and apoptosis (Arab *et al.*, 2015).

In the present investigation, FM treatment markedly enhances the gastric mucosa and attenuates the gastric injury such as hemorrhage and hyperemia. These results suggest that FM has a protective action by reducing the gastric mucosal injury exposed to ethanol.

Additionally, secretion of mucus is supposed to be such a vital protective factor against gastric injuries. Previous reports suggested that mucus delays the diffusion of protons, which would aid in maintaining a favorable pH at the epithelial apical surface (Schreiber and Scheid, 1997). The pH gives an idea of the level of acidity and volume of gastric secretions. Low pH value is an indicator of decreased hydrogen ion concentration in gastric juice. This has been linked to the pathogenesis of ulcer and gastric damage in experimental animals (Lüllmann *et al.*, 2000). The present study showed that FM neutralized stomach juice pH to (7.30 ± 0.33 , 7.24 ± 0.77 ; respectively) by significantly increasing ($P < 0.001$) of the pH value of the gastric contents at both doses.

The involvement of oxidative stress in the pathogenesis of ethanol-induced gastric injury has been confirmed by several studies. One of the possible mechanisms of EtOH induced acute gastric mucosal injury is thought to arise as a result of extreme generation of free radicals (Schlorff *et al.*, 1999). The free radicals have a high tendency to react rapidly with lipids, thus, triggering lipid peroxidation that can lead to disorganization of the membrane and consequently reduces its fluidity (Recknagel and Ghoshal, 1996).

In harmony with these mentioned data, the current study showed that, ethanol administration initiated gastric oxidative stress and significantly increased the MDA level, a byproduct of lipid peroxidation, as compared to control group. While, this elevation of MDA level was reversed by pretreatment with FM in both doses ($p < 0.05$, $p < 0.001$ respectively) suggesting a reduction in the lipid peroxidation activity upon the oral administration of FM.

Catalase, a highly reactive enzyme, plays a protective antioxidant effect against the harmful actions of lipid peroxidation by reacting with H_2O_2 to form water and molecular oxygen (Zakaria *et al.*, 2015). Data of the current study demonstrated that FM combated oxidative stress and enhanced the antioxidant status in animals with ethanol gastritis as revealed by the reduction of MDA levels. This was accompanied with a significant ($P < 0.001$) increase of CAT in both doses. These findings highlight the premise that the antioxidant features of the components of this new formulated milktable are implicated in the alleviation of ethanol gastric injury. In fact, scavenging of ROS has been considered as one of the mechanisms involved in the healing of ulcers (Mei *et al.*, 2012), which confirmed that FM anti-ulcerogenic properties are partially based on its antioxidant function.

Emerging evidences support the concept that the elevation of NO levels is a protective mechanism against the damage of gastric mucosa (J'uniore *et al.*, 2014; Nordin *et al.*, 2014). It is considered one of the most important

defensive endogenous mediators in the gastric mucosa and a potent chain-breaking antioxidant in free radical-mediated lipid Peroxidation (O'Donnell *et al.*, 1997; Halabiet *et al.*, 2014). Additionally, NO plays a vital role in maintaining the gastric mucosal alkaline production (Calatayud *et al.*, 2001), and inhibiting neutrophil aggregation (Wallace *et al.*, 1997).

According to our results, EtOH administration significantly ($P < 0.05$) decreased NO content in the stomach. Similarly, as reported by (Abdulla *et al.*, 2009; Goswami *et al.*, 2011). So, one of the protective way against harmful effect of ethanol on gastric mucosa is try to keep the normal level of nitric oxide to stop infiltration of the neutrophil.

The present study showed that the NO reduction induced by ethanol was significantly attenuated by FM that increase gastric NO level in both doses ($P < 0.001$). This result is consistent with the present histological results that have revealed protection of the gastric mucosa and inhibition of leukocyte infiltration in rats pre-treated with FM. Similar results were reported by Kobayashi *et al.*, (2001) who stated that the inhibition of neutrophil infiltration in the ulcerated gastric tissue is a protective effect against mucosal lesions. The neutrophils are a main source of inflammatory mediators, therefore, suppression of neutrophil infiltration during inflammation was found to improve gastric ulcer healing.

On the other hand, it has been suggested that gastric ulcers are considered to be the manifestation of an inflammatory response (Kang *et al.*, 2014). It is characterized by the migration of polymorphonuclear leukocytes and macrophages in the ulcer area. Migrated macrophages release proinflammatory cytokines such as TNF- α and this upregulates leukocyte recruitment and transmigration in gastric inflamed areas. Our experimental results are in line with these previous data. Ethanol ingestion upregulated the inflammatory response as evidenced by the increase of gastric proinflammatory TNF- α ($P < 0.001$). These findings are consistent with previous reports (Liu *et al.*, 2012; Li *et al.*, 2013). TNF- α has been closely linked to gastric inflammation via activation of immune cells, generation of other proinflammatory mediators and overexpression of NF- κ B (Li *et al.*, 2013). TNF- α also subdues gastric microcirculation around ulcerated mucosa and thus delays its healing (Hasgulet *et al.*, 2014).

Interestingly, FM significantly suppressed gastric TNF- α in both doses ($P < 0.001$). Thus, the reduction of TNF- α which was accompanied with an elevation of gastric NO, has been viewed as a successful strategy for the management of gastric injury. Meanwhile, the above findings reinforced the histopathological results that described attenuation of neutrophil infiltration, edema and hemorrhage by new formulated milk tablet in ethanol-induced ulcer in rat.

The liver is the main metabolic organ for ingested alcohol; therefore, the series of events that occur in the liver following alcohol intake may predispose the organ to damage. Thus, biochemical markers in the liver and plasma have been widely used as effective methods for the early diagnosis of liver diseases and to determine the functional components for prevention of hepatic lesions. In contrast to the previous studies (Gramenziet *et al.*, 2006; Purohit *et al.*, 2009) that reported an elevated plasma activity of AST and ALT in alcohol-treated rats, our results demonstrate that AST, ALT, and ALP activity was insignificantly induced by ethanol ingestion. This result could be attributed to that observation of plasma AST and ALT induced by ethanol have been dependent on the animal strain, feeding period and method, and/or dosage of ethanol (Baldiet *et al.*, 1993). Nevertheless, our data show that pretreatment with high dose of FM lowered the activity of serum AST ($P < 0.05$). Notably, the present data showed that pretreatment with low dose of FM significantly ($P < 0.05$) increase in uric acid and creatinine levels. This could be an indicative of the renal cellular injury due to kidney dysfunction and with alteration in membrane permeability.

Conclusion:-

In conclusion, Based on the above mentioned data, oxidative stress has been involved in the development of ethanol-induced gastric injury where a store of reactive oxygen species (ROS) generated by leukocyte activation triggers mucosal damage via lipid peroxidation and via the antioxidant defenses depletion. Activation of neutrophils is associated with an unregulated inflammatory response with an increased gastric generation of proinflammatory cytokines, including tumour necrosis factor- α (TNF- α). The current study highlights evidences for the anti-ulcer effect of new formulated milk tablet (FM) could be attributed to the its marked antioxidant features abolish lipid peroxidation and preserve gastric antioxidant defenses. It also acts by boosting of NO biosynthesis accompanied reduction of neutrophil infiltration with the suppression of the proinflammatory cytokine TNF- α .

Table 1:- Chemical composition of the new formulated milk tablet (FM) ingredients.

Chemical composition	Fat	Protein	Carbohydrate	Ash	Moisture
Skim milk powder	0.90	36.0	48.0	8.5	03.0
Honey	01.5	12.0	16.0	0.6	58.0
Bee pollen	06.0	20.0	40.0	2.2	07.5
Palm pollen	20.5	31.5	14.0	4.5	28,8
Royal jelly	03.0	17.0	18.0	2.0	57.0
Propolis	15.5	25.0	28.0	4.5	10.0
Ginseng	1.2 0	12.0	75.0	7.0	11.0

Table 2:- Formulations of the new formulated milk tablet(FM) from mixture design.

Formulation	Skim Milk powder (%)	Honey (%)	Other ingredients*(%)
A	50	28	22
B	55	23	22
C	60	18	22
D	65	13	22
E	70	8	22

* Other ingredients: bee pollen, palm pollen, Royal jelly, propolise and gensing

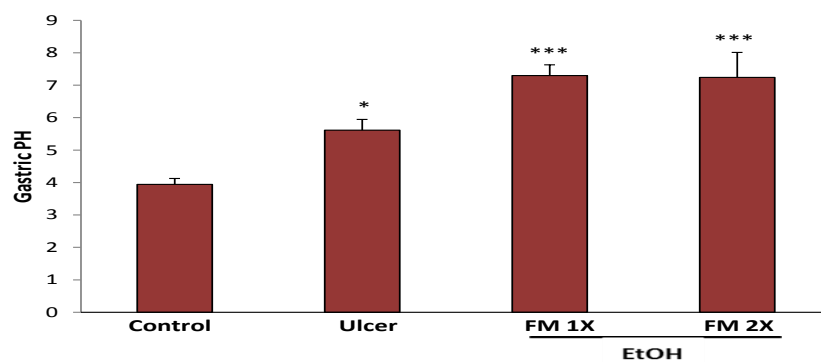
Table 3:- Sensory evaluation of the new formulated milk tablet (FM) formulation.

Formulation	Body & Texture (50)	Flavor (30)	Color (10)	Sweetness (10)	Total (100)
A	40	22	7	7	76
B	42	25	8	8	83
C	48	28	9	9	94
D	45	25	8	8	86
E	40	18	7	5	70

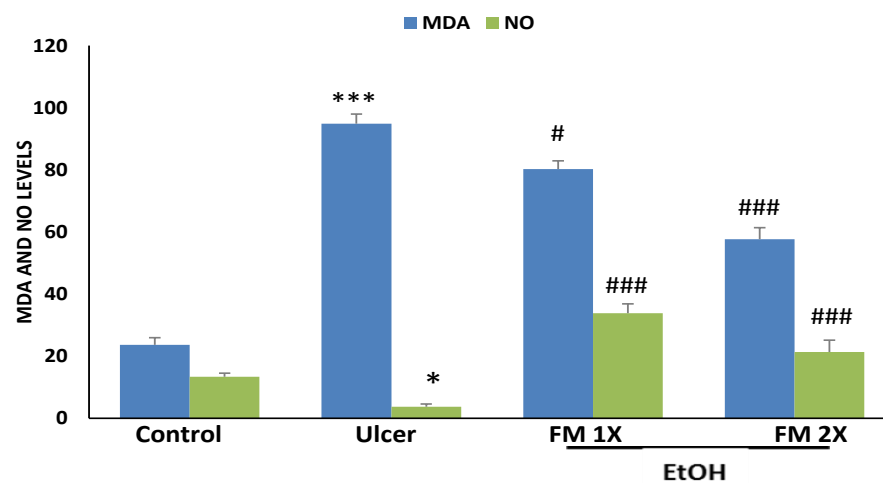
Table 4:- Chemical composition of the selected formula (C) of new formulated milk tablet (FM)

Constituent (%)	FM Formula
Fat	4.20
Protein	33.85
Carbohydrate	40.75
Ash	12.55
Moisture	7.65

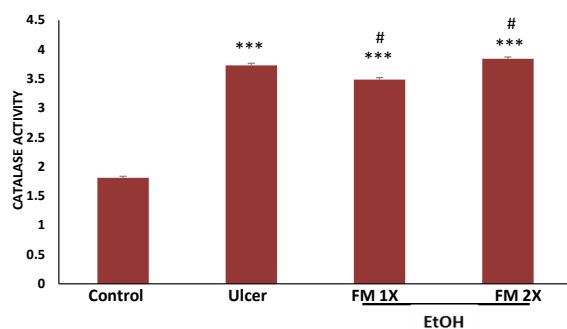
(a)



(b)



(c)



(d)

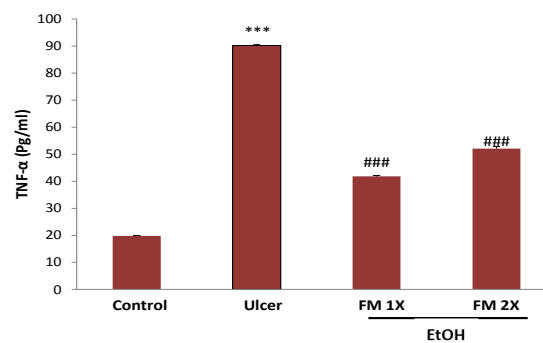
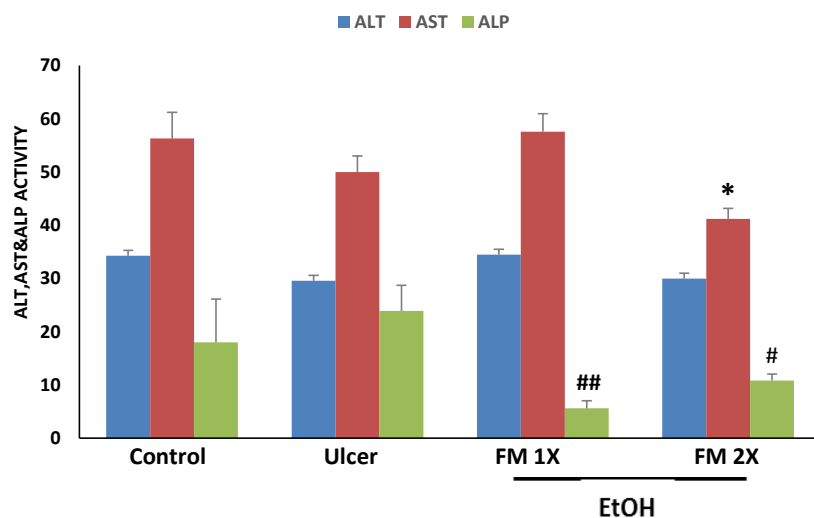


Fig.1:- Effects of FM on gastric pH (a), MDA (nmol/g tissue) and NO (μmol/g tissue) (b), CAT (U/L) (c) and Plasma level of TNF-α (d) in EtOH induced gastric ulcer. The data represent the means ± SEM. * $P < 0.05$, *** $P < 0.001$. # Significantly different from the respective ulcer group at # $P < 0.05$, ### $P < 0.001$. FM-1X (157mg/kg), FM-2X (314mg/kg).

(a)



(b)

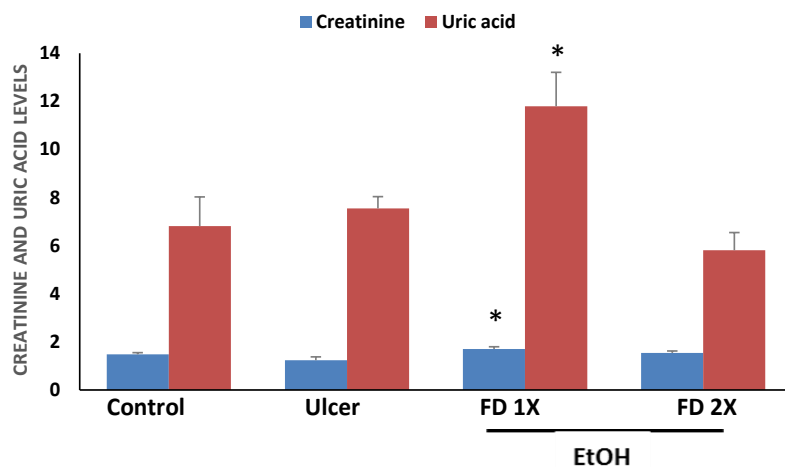


Fig.2:- Effects of FM on the activity of ALT, AST, and ALP (U/L) (a). Creatinine, and uric acid levels (b) in EtOH induced gastric ulcer. The data represent the mean \pm standard error. * $P < 0.05$.# Significantly different from the respective ulcer group at [#] $P < 0.05$, ^{##} $P < 0.01$.FM-1X(157mg/kg), FM-2X (314mg/kg)

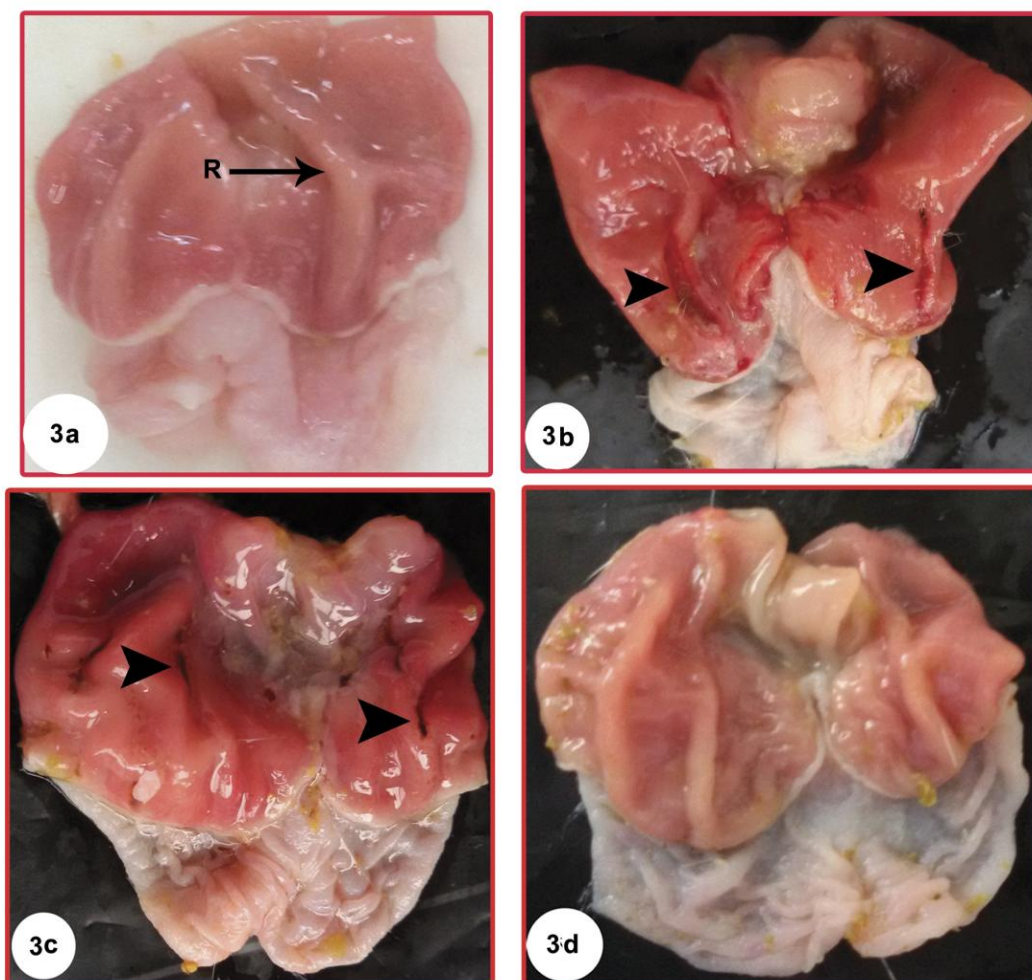


Fig. 3:-Photographs of the stomach of rats (a) the control group showing normal mucosa with rugae, (b) the ulcer group showing linear hemorrhagic bands (arrow heads), (c) the FD pre-treated group (standard diet) showing few hemorrhagic areas (arrow heads) and (d) the FD pre-treated group (double standard diet) showing normal stomach architecture.

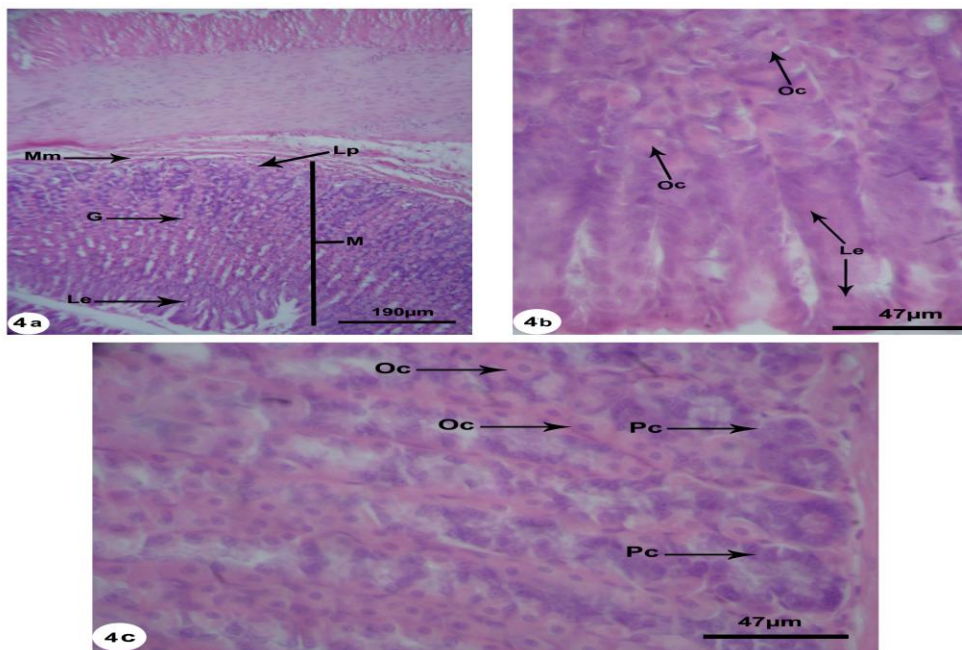


Fig. 4 (a-c):-photomicrographs of the stomach of rats of the control group showing normal mucosal architecture. The gastric mucosa (M), lining epithelium (Le), lamina propria (Lp), muscularis mucosae (Mm), gastric gland (G), oxyntic cells (Oc) and peptic cells (Pc).

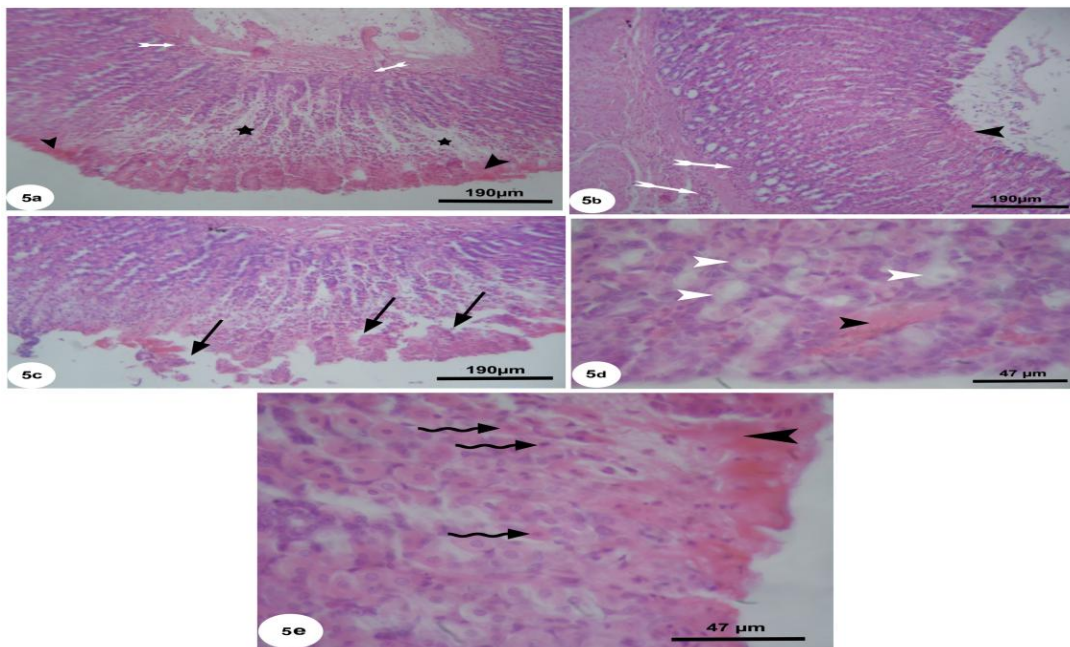


Fig. 5 (a-e):-photomicrographs of the stomach of rats of the ulcer group showing the gastric mucosal lesions. The necrotic mucosa (asterisk), mucosal hemorrhage (black arrow head), leucocytes infiltration (white arrow), epithelial lifting (black arrow), cell necrosis (white arrow head) and pyknotic nuclei (curved arrow).

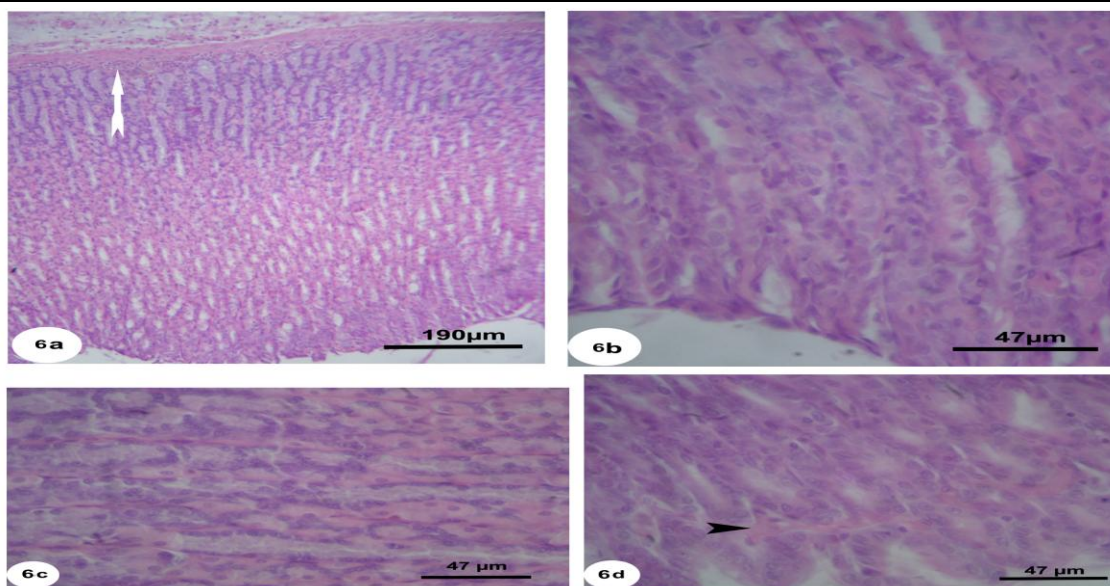


Fig. 6 (a-d) :-photomicrographs of the stomach of rats of the FD pre-treated group (standard diet) showing normal mucosal architecture with superficial gastric lesions. The leucocytes infiltration (white arrow) and mucosal hemorrhage (black arrow head).

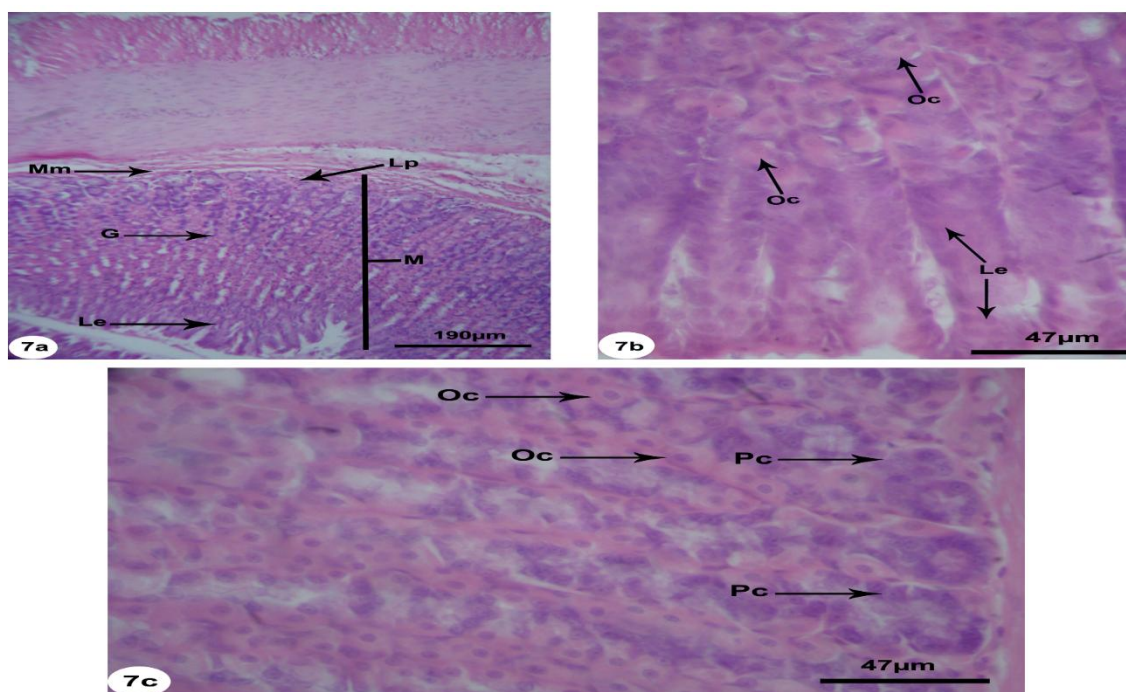


Fig. 7 (a-c) :-photomicrographs of the stomach of rats of the FD pre-treated group (double standard diet) showing normal mucosal architecture.

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RESEARCH ARTICLE

Clinical profile of Central Nervous System Tuberculosis in Kanniya kumari medical college.

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Abstract

Background: Tuberculosis of the central nervous system is an important cause of morbidity and mortality. Once a disease mainly of childhood, it is now seen at any age and is equally frequent in adults. They may present as Meningitis, Tuberculoma, Potts Paraplegia, Intrapinal Granuloma or Arachnoiditis. This study was conducted to analyse various clinical presentations of CNS tuberculosis and their sequelae. **Methods :** An observational study was conducted in 30 patients admitted in Kanniyakumari Government Medical College from Jan 2016 to December 2016 with CNS manifestations of Tuberculosis. Computed tomography(CT), electroencephalography (EEG) and clinical findings were taken. **Results:** In the present study, the most common CNS presentation was Tuberculous Meningitis followed by Tuberculoma. Common age group being between 15-19 yrs. Aseptic meningitis had a good prognosis compared to Obstructive Hydrocephalus which had a bad prognosis. In this series, complete recovery was 66.6%, mortality was 13.4% and Sequelae in 20%. **Conclusion:** CNS tuberculosis is being reported more often nowadays due to its awareness and improved diagnostic modalities. Resistance to drugs has added a new challenge. Morbidity and mortality can be reduced by a great extent with early recognition and timely treatment

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Introduction:-

Both as a cause of death and disability, Tuberculosis of the Central Nervous System is of great clinical importance. In children the condition most often arises as a consequence of primary or miliary tuberculosis, but in adults it usually develops in individuals with known tuberculosis elsewhere, especially in the lung. Once a disease mainly of childhood, it is now seen at any age and is equally frequent in adults. The pattern of Tuberculous infection of the Nervous System is changing. In an American epidemiological study of extrapulmonary tuberculosis, up to 10% of cases showed CNS involvement¹, whereas CDC data showed that 6.3% of extrapulmonary cases (1.3% of total tuberculosis cases) had CNS TB². A study done at Taiwan, 1.5% of TB deaths between 1997 and 2001 were attributable to CNS disease, a percentage that had increased from previous years³.

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Risk factors for CNS tuberculosis include age (children > adults) HIV-coinfection, malnutrition, recent measles in children, alcoholism, malignancies, the use of immunosuppressive agents in adults and disease prevalence in the community⁴. The clinical presentation is determined by the nature of the pathological process. It commonly takes the form of a meningitis, but encephalopathy (encephalitis) can also occur. At other times, extra medullary Tuberculosis can produce secondary neurological lesions or tuberculosis disease is confined to spinal cord. Seizures, either focal or generalised, may occur during acute illness or months after treatment⁵.

Materials and Methods:-

An observational study was conducted in 30 patients admitted in Kanniyakumari Govt. Medical College from Jan 2016 to December 2016 with CNS manifestations of Tuberculosis. CT Scan was done for all 30 patients. EEG (Electro Encephalogram) was done for 7 Patients

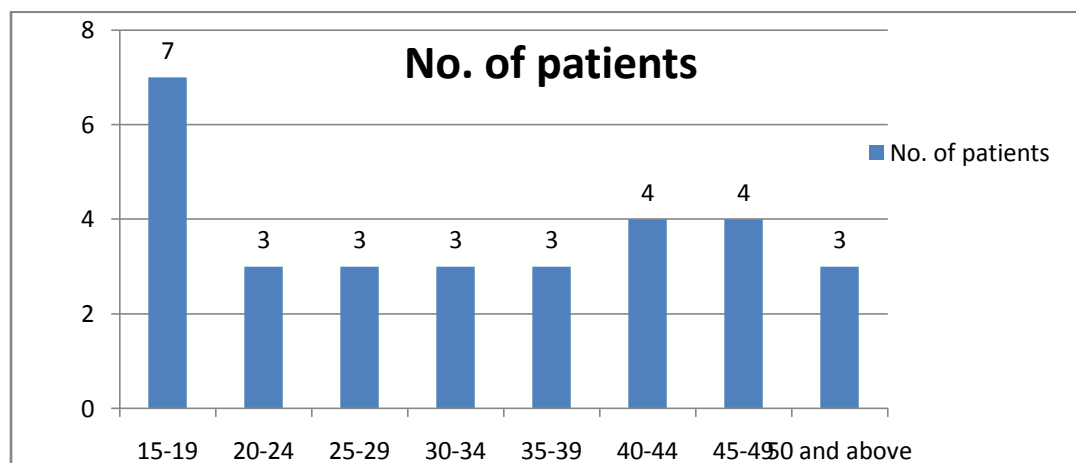
Following the clinical, CSF and CT Scan, study patients were analysed for

1. Age and Sex Incidence
2. Mode of presentation
3. Symptom analyses
4. Neurological Sequelae

Observation

Table – 1:- Age Incidence

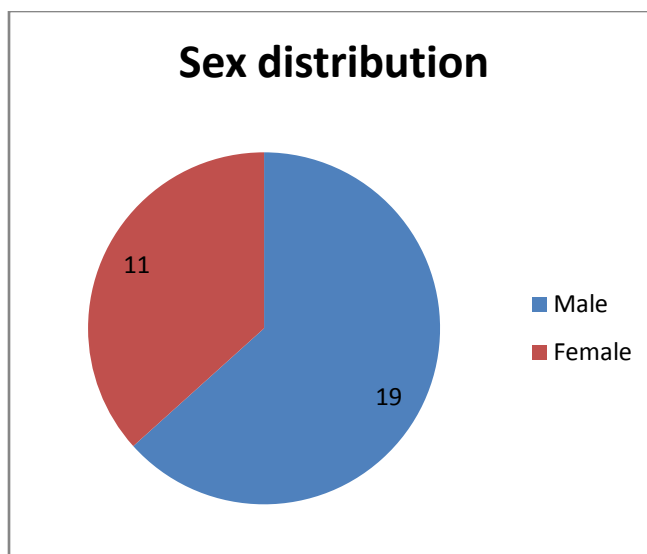
Age group	No. of patients
15 – 19	7
20-24	3
25-29	3
30-34	3
35-39	3
40-44	4
45-49	4
50 and above	3



In the present study, the vulnerable age group of CNS TB belonged to 15-19 years carrying 23% of the total patients.

Table – 2:- Sex distribution

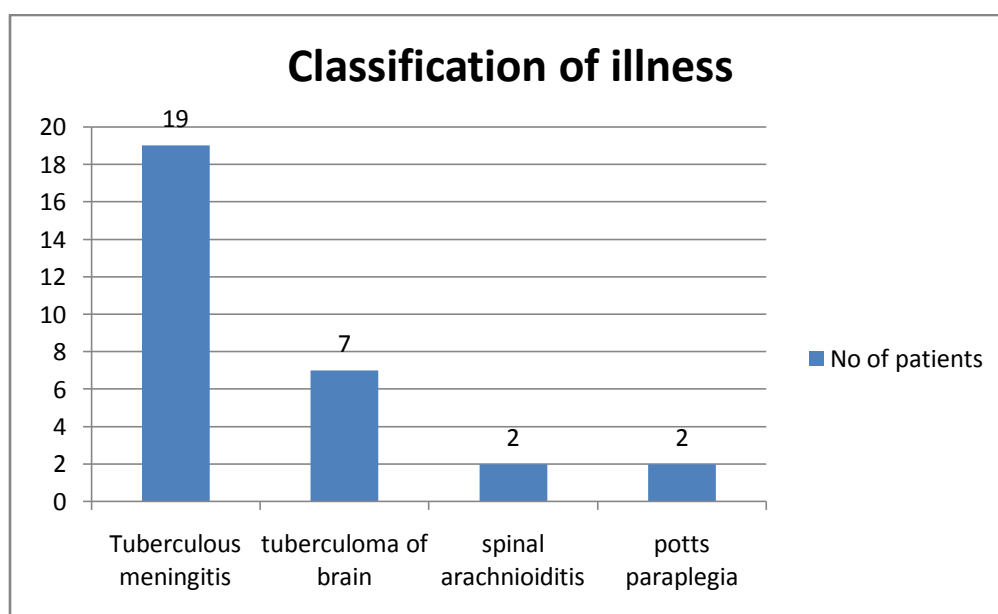
Sex	No. of patients	Percentage
Male	19	64
Female	11	36



In the present study, 64% were males and 36% were females.

Table -3:- Classification Of The Illness

Classification	No. of patients	Percentage
Tuberculous meningitis	19	63.3
Tuberculoma of brain	07	23.3
Spinal Arachnoiditis	2	6.6
Potts Paraplegia	2	6.6



In the present study of 30 patients, 19 patients, (63.3%) belongs to the class tuberculous meningitis, 7 Patients (23.3) belongs to the class tuberculoma of brain, 2 patients (6.6%) belongs to the group spinal arachnoiditis and 2 patients belongs to the group pott's paraplegia (6.6%)

Table -4:- Symptom Analysis

Major Classification	Symptom	No.of patients
TB Meningitis	Fever	18
	Headache,Vomiting	17
	Altered sensorium	6
	Double vision	5
	Photophobia	4
	Convulsion	3
	Weakness of limbs	2
	Numbness of extremities	1
	Retention of Urine	1
	Dysphagia	1
Tuberculoma of Brain	Fever	3
	Convulsion	6
	Headache and Vomiting	5
	Numbness limbs	2
	Weakness of limbs	2
	Numbness and weakness of lower limbs	2
	Numbness and weakness of lower limbs	2
	Retention of urine	1

Table 4:- Symptom Analysis

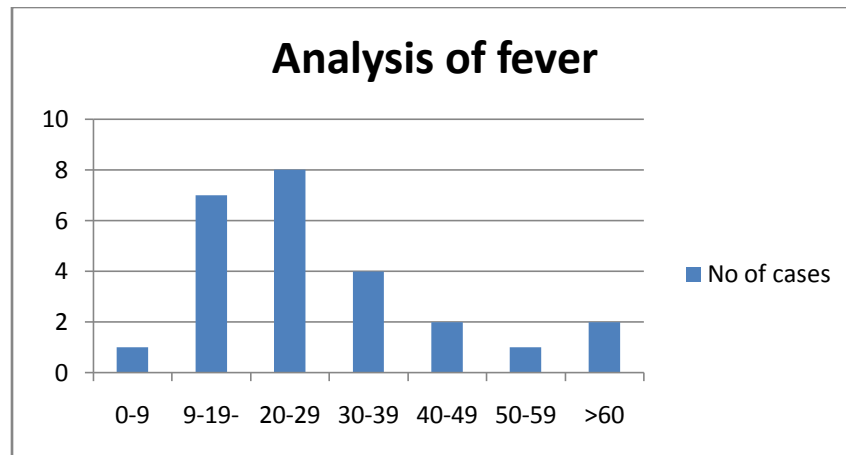
In patients with tuberculous Meningitis, fever associated with headache, vomiting was the main presentation, presented in 89.4%. Photophobia was present in 4 cases who also had features of rise in intracranial tension. Numbness in the right half of the body was presented in one case. Weakness of right upper and lower limbs was presented in one case of Tuberculoma of brain.

2 patients of TB Meningitis in the course of their treatment developed spinal arachnoiditis in the form of lower limb weakness and numbness after 45 days.

2 patients had pott's paraplegia. They presented as weakness and numbness of lower limbs and bladder retention developed in one case in the later period

Table 5:- Analysis Of Fever

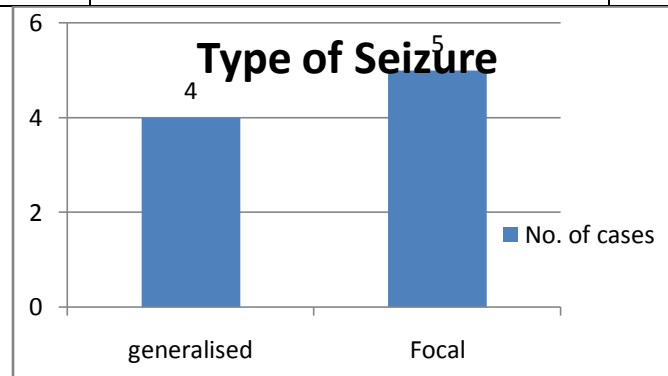
Duration of days	No.of cases	percentage
0-9	1	3.3
10-19	7	28
20-29	8	32
30-39	4	16
40-49	2	8
50-59	1	3.3
>60	2	6.6



In the present study, higher number of cases had duration of fever for 2-3 weeks. 2 Cases (6.6%) had duration of fever for a longer period, more than 60 days.

Table 6:- Types Of Seizures

Type	No.of cases	percentage
Generalised	4	13.3
Focal	5	16.6



In the present study generalized seizures were present in 4 patients(13.3%) , Focal seizures were present in 5 patients (16.6%). Generalised seizures were present invariably in Tuberculous meningo encephalitis patients and focal seizures were present in patients with tuberculoma of brain.

Table – 7:- Neurological Presentation And Sequelae

Type of presentation	No.of Cases	Recovery	Sequelae
Tuberculous meningitis	7	7	-
Meningo encephalitis	4	3	Dead (1)
Basal meningitis	1	-	Dead (1)
Brainstem lesion (quadripareisis)	1	-	Dead (1)
Cranial Nerve Palsy	5	4	Optthalmoplegia(1)
Monoparesis (TB meningo encephalitis)	1	1	-
Spinal Arachnoiditis	2	-	Paraplegia(2) Optic atropy(1)
Tuberculoma	7	5	Dead(1) Hemiplegia(1)
TB Spine	2	-	Paraplegia(2)

In the present study 7 patients of tuberculous meningitis recovered after treatment without any sequelae. 4 patients developed meningo encephalitis with fits and altered sensorium, and 1 patient died during the illness. 1 patient

developed right lower limb monoparesis, but recovered 4 patients had third nerve palsies who had not good recovery. 1 patient had combination of third, sixth and seventh nerve palsies and features of basal meningitis and died during the illness. One case presented with focal neurological damage as Brainstem lesion with features of Quadriplegia and dysphagia and died during the illness.

Fundus findings included:-

Papilloedema in 6 Patients

2 patients of tuberculous meningitis

4 patients of Tuberculoma Brain

Optic atrophy in 1 patient (Tuberculous Arachnoiditis)

Discussion:-

Tuberculosis remains a serious medical problem in economically depressed countries. Tuberculous meningitis constitutes 50-55% of all bacterial meningitis⁶.

Sex Incidence:-

Among the sex incidence, males rank foremost. 19 out of 30 cases were males (63%) and 11 out of 30 cases (36%) were female. previous studies favoured incidents among males are more than females (Udani et al)⁶

Classification:-

In the present study of 30 patients, 19 patients (63.3%) belongs to the class Tuberculous meningitis, 7 patients (23.3%) had tuberculoma of Brain. Two patients (6.6%) had pott's paraplegia. Among the extrapulmonary tuberculosis, TB Meningitis has a higher incidence. (Tandon et al)⁷

Tuberculous Meningitis:-

Onset : In the present study the onset is acute in 4% and subacute in 30% previous studies favoured subacute onset in adults 35% (Dastur, Udani)⁸

Fever : Fever was observed invariably in all patients 95.7% It is of low grade pyrexia and fever associated with headache and vomiting is presented in 89.4% Fever is absent in 10% (Tandon et al)⁷

Convulsion : Convulsion was present in 3 cases (10%) in the present study

Licoll et al study⁹ said convulsion are frequent below 2 yrs (17%) and less frequent in adults (9.5%)

Neurological Presentation

Signs of meningeal irritation like neck stiffness and kernigs sign positive was present invariably in all patients with tuberculous meningitis.

Hydrocephalus

Raised intracranial pressure is present at all stages of the disease. Papilledema is seen in 40% which may lead to optic atrophy. Mactier et al study of 79 cases in Australia incidence of obstructive hydrocephalus in 2.8%¹⁰. In the present series 2 patients (6.6%) had obstructive hydrocephalus and one patient (3.3%) had communicating hydrocephalus.

Cranial Nerve Palsy

20 – 30% may develop cranial nerve palsies (Dastur et al)⁸. As per Tandon et al⁷ – 10 to 25%. The sixth nerve is the most affected followed by third, fourth and Seventh⁴. In the present series 4 patients had 6th nerve palsy, One patient had third nerve palsy. All of them recovered. One patient had Combined third, Sixth and Seventh nerve palsy who died during the illness.

Focal Neurological Deficit

One case presented with Quadriplegia and dysphagia, died during the illness probably due to brain stem lesion.

Tuberculoma Of The Brain:-

In the present study, 7 cases (23.3%) were found to have tuberculoma of brain. The Incidence of tuberculomas among intracranial space occupying lesions varies from 3.3 – 17.6% in different series since 1970.

Papilledema:-

Presented in 4 patients with tuberculoma (57.1%). Reported incidence by Tandon study⁷ is 87%

Tuberculous Arachnoiditis:-

In the present study 2 cases of tuberculous meningitis (6.6%), in the course of their treatment later presented as spinal arachnoiditis. They showed good recovery, but they carried a morbidity of paraparesis as their sequelae, but they are on follow up further.

Potts Paraplegia:-

In the present study 2 cases had presented with potts paraplegia(6.6%)

Both of them were males, below 40 years and both of them, the thoracic spine was involved.

The incidence of potts paraplegia is around 10% of CNS tuberculosis¹¹

Conclusion:-

1. In the present series of 30 cases during the period from January 2016 to December 2016, 19 patients (63.3%) were tuberculous meningitis, 7 Patients (28.3%) were tuberculoma of brain, 2 patients (6.6%) were tuberculous Arachnoiditis and 2 patients were pott's paraplegia (6.6%)
2. Males are more affected than females.
3. 15-19 years were vulnerable age group for TBM in adults
4. Minimum period of fever for 2 weeks is present in a majority of cases
5. In TBM patients, Who had presented as 'Aseptic Meningitis' had good recovery without any sequelae.
6. Patients presented with obstructive hydrocephalus had a bad prognosis
7. Optic atrophy was observed in a patient with Tuberculous Arachnoiditis
8. CT Scan finds an important role in the diagnosis of tuberculoma of Brain
9. Successful management of TBM depends on early diagnosis and effective antituberculous therapy
10. In this series complete recovery was 66.6%, mortality was 13.3% and Sequale in 20%

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RESEARCH ARTICLE

IMPORTANCE OF COMPUTER VISION FOR HUMAN LIFE

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Abstract

Computer Vision (CV) is a discipline that studies methods to reconstruct, interpret and comprehend a 3D scene from its 2D pictures in terms of the homologies of the constitution gift in the scene. For the implementation of CV, a man or woman should have the skills of more than a few different subjects. In this paper we will be able to be aware of concerning the Computer imaginative and prescient, then we will speak about the advantages and downsides of CV. Then we will be able to examine the imaginative and prescient of a human with Computer imaginative and prescient. On this paper, we will be able to talk about various strategies of CV and in the end, we will talk about quite a lot of functions of CV in quite a lot of fields.

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Introduction:-

People utilize their eyes and their brains to detect their general surroundings for all intents and purposes. CV is the science that intends to give a comparable ability to a machine or Computer. CV (picture comprehension) is teaches that arrangements with 2D pictures and reproduces, decipher and comprehend a 3D scene from it as far as the properties of the structures exhibit in the pictures. CV is the study with the programmed extraction, investigation, and comprehension the helpful data from a solitary picture or an arrangement of pictures. It includes the advancement of a hypothetical and

Algorithmic premise to accomplish programmed visual comprehension. A definitive objective of CV is to demonstrate, recreate and surpass the capacity of human vision utilizing computer programming and equipment at various levels. It needs information of various subjects like software engineering, electrical designing, arithmetic, science, psychological science, and physiology.

Advantages of computer vision:-

1. Simple & Faster processes: - Speedy computer systems replace lengthy visual tests.
2. Reliability: - Cameras and computers, as opposed to a human eye, in no way get tired.
3. Accuracy: - By using utilizing computer imaginative and prescient the completing of the tip product increases to a large extent.
4. A wide variety of use: - It has an awfully huge range of applications.
5. Price discount: - Time is saved on people and devices, therefore misguided merchandise are eliminated.
6. No boundaries like human perception.
7. Do not must have instruments embedded, bodily printed or externally hooked up to objects specified for detection.
8. Picture shooting gadgets are convenient to mount, do away with, substitute and upgrade.

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9. Upgrading photo sensors doesn't require upgrading tags, identifiers or transponder devices.

Disadvantages of computer vision:-

1. Data processing and analytics is intensive and requires a lot of computation resources and memory.
2. Main technical barriers are its robustness in the face of fixing the environment.
3. Illumination variation further complicates the design of effective algorithms on account that of changes in shadows being cast.

Computer Vision Vs Human Vision:-

In object classification, it was known that the human brain tactics visible know-how in a semantic area traditionally, i.e. Extracting the semantically significant elements equivalent to line segments, shape, boundaries etc. In any case, by late data handling methods, these sorts of components can't be recognized by PCs heartily so that in PC vision it's still hard to prepare visual data as people do. PCs need to prepare visual data in information space framed by the vigorously distinguishable yet less important components, for example, hues, surfaces, and so on. In this way, the handling philosophy in PCs is entirely not the same as that in individuals.

Different Techniques of Computer Vision:-

There are various systems of CV like:

Registration and Stitching:-

Image Stitching is the method of editing the point of view of portraits and blending them in order that the photos can be aligned seamlessly.

Snapshot stitching system will also be divided into three main categories, In First category is, Image Registration entails matching points in a collection of images or making use of direct alignment methods to search for snapshot alignments that minimise the sum of absolute variations between overlapping pixels.

In Second Category is, Image Calibration targets to minimise variations between excellent lens units and the camera-lens combo that was used, optical defects akin to distortion, and exposure differences between pictures, camera response and chromatic aberrations.

In Third Category is, Photo mixing entails executing the changes found out within the calibration stage, mixed with remapping of the portraits to an output projection. Colours are adjusted between photographs to catch up on exposure differences. Snapshots are blended collectively and seam line adjustment is done to diminish the visibility of seams between images.

Video Processing:-

Video processing is a specified case of sign processing, which most often employs video filters and the place the input and output signals are videos records or video streams. Video processing systems are utilised in television units, VCRs, DVDs, video codec, video avid gamers, and so forth.

Detection and Tracking:-

Identification and following of autonomously moving items from the scene are vital components in video observation. On account of a moving camera, the recognition and following issues are naturally more mind boggling, since the camera movement incorporates a movement in all pixels of the picture.

A typical approach for recognizing moving districts depends on the adjustment because of the camera movement utilizing parametric movement models and characterizing moving pixels as the ones that have not been balanced out. This functions admirably when the seen can be viewed as planar, or when the movement of the camera is dish or zoom.

Visual Inspection:-

Regardless of technological developments in inspection approaches the principal and primary inspection/scan manner is visual inspection.

Visible inspection is an extraordinarily mighty method, and it should be the main method incorporated in any powerful quality manage application. It has been shown many times that, “visual Inspection” performed by adequately informed inspectors, the outcome in the discovery of the large majority of these defects which might simplest be found out later by using some more high-priced non-harmful experiment procedure. Even as visual inspection is confined to materials surface the handiest examination.

Pattern and Character Recognition:-

Spatial sample recognition, of which the recognition of alphanumeric characters is a subclass, is a principal and functional trouble. More efficient coding of transmitted pictorial knowledge and extra efficient utilisation of humanly produced information could outcome from its solution.

The pattern realisation has been mentioned as the challenge of a significant code to a recognisable structure in a suite of signals. The signals are the outcome of a metamorphosis from a visible picture area to an electrical illustration of this field.

Photo Enhancement:-

In computer photographs, the process of improving the nice of a digitally stored snapshot by way of manipulating the snapshot with the application. It's rather effortless, for illustration to make a picture lighter or darker, or to increase or lower distinction. Evolved photo enhancement application also helps many filters for alternating images in more than a few methods. Packages specialised for snapshot enhancement are in many instances referred to as photo editors.

Applications of Computer Vision:-

The CV is being utilized today as a part of a wide assortment of true applications, as:

1. Optical character realisation (OCR): Studying handwritten postal codes on letters and Automatic Number Plate Recognition (ANPR).
2. Desktop Inspection: Rapid parts inspection for quality confirmation utilising stereo vision with particular enlightenment to quantify resistances on flying machine wings or auto body parts or searching for deformities in steel castings utilizing X-beam vision.
3. Retail: Object acknowledgment for mechanized checkout paths.
4. 3D model building (Photogrammetry): Fully mechanized development of 3D models from airborne photos utilized as a part of frameworks, for example, Bing Maps.
5. Medical Imaging: Registering pre-agent and intra-agent symbolism or performing long haul investigations of individuals' cerebrum morphology as they age.
6. Automobile defence: Detecting startling deterrents like people on foot in the city, under conditions where dynamic vision strategies, for example, radar don't function admirably.
7. Fit transfer: Merging Computer Generated Imagery (CGI) with no frills footage by following element focuses in the source video to gauge the 3D camera movement and state of nature. Such strategies are generally utilized as a part of Hollywood (e.g. in films like Jurassic World), they additionally require the utilization of exact tangling to embed new components amongst forefront and foundation components.
8. Movement seizes (mocap): Using retro –reflective markers saw from different cameras or other vision-based systems to catch on-screen characters for Computer animation.
9. Surveillance: Monitoring for gate crashers, investigating thruway activity, and observing pools for suffocating casualties.
10. Fingerprint attention and biometrics: For programmed get to validation and additionally scientific applications.
11. Stitching: Turning covering photographs into a solitary flawlessly sewed display.
12. Exposure bracketing: Merging various exposures taken under testing lighting conditions (solid daylight and shadows) into a solitary flawlessly uncovered picture.
13. Morphing: Turning a photo of one of your companions into another, utilizing a consistent transform move.
14. Image founded walkthroughs: exploring a huge accumulation of photos, for example, the inside of your home, by flying between various photographs in 3D.
15. Face detection: For enhanced camera center and also more significant picture looking.
16. Visible authentication: robotically logging family individuals onto your residence computer as they sit down in front of the webcam.

Conclusion:-

We got here to grasp that CV has a big scope in future with its titanic advancement corresponding to it's a self-discipline that reviews how one can reconstruct, interrupt and realise a 3D scene from its 2d pics in phrases of the residences of the structure gift in the scene. For the implementation of CV, we came to understand that an individual will have to have the talents of quite a lot of different subjects. In this paper we have now learned about the COMPUTER imaginative and prescient, then we've discussed the advantages and downsides of CV. Then we've got when compared the vision of human with CV. In this paper, we've got also mentioned more than a few procedures of CV and eventually we now have mentioned quite a lot of functions of CV in various fields.

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RESEARCH ARTICLE

KNOWLEDGE, ATTITUDE AND PRACTICE OF DIETARY AND LIFESTYLE HABITS AMONG MEDICAL STUDENTS IN HAIL UNIVERSITY, SAUDI ARABIA.

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Abstract

Knowing the medical students' knowledge, attitude and practices of nutritional and lifestyle habits helps to enhance the nutrition of the community, which will consequently lead to a healthier society, as they will constitute the main body of future physicians and professionals. We aimed to evaluate their nutrition knowledge, their eating habits and overall perception regarding importance of healthy eating habits and lifestyle habits of medical students of Hail University, Kingdom of Saudi

Arabia . A cross-sectional study included 207 students between ages 19-24 years from basic and clinical levels in the faculty of medicine. A self-reported questionnaire was employed to assess perceived attitude and barriers to healthy practices by the study participants. Overall most medical students were aware of importance of minerals and vitamins in diet yet they were not practicing it adequately in their daily life but the majority of the students were not aware of the composition of balanced diet, improving nutrition knowledge, attitude and dietary practices through nutritional education may help to prevent many nutrition related diseases.

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Introduction:-

During the 20th century, the leading causes of death shifted from infectious to chronic diseases: Cardiovascular disease, cancer, and diabetes are now among the most prevalent, costly, and preventable of all health problems.[1] These diseases have been strongly associated with unhealthy lifestyle habits, including inappropriate nutrition, lack of exercise, smoking, alcohol consumption, caffeine overuse, and improper sleeping habits.[2]

Recently, the Kingdom of Saudi Arabia has faced marked changes in the demographics, socioeconomics, and public health. This increasingly higher economic development of Saudi Arabia has resulted in a rapid Westernization of lifestyle habits. [3] The Westernization of dietary habits and lifestyle reflects the national increasing trend towards consumption of macronutrients including total food, fats, animal products, and refined foods. [4,5] In contrast, this new trend in the Saudi dietary patterns is accompanied by low consumption of vegetables and fruits, which are some of the main sources of essential micronutrients. [6,7] Changes in lifestyle and socioeconomic status in KSA have also had a significant impact on Saudis' physical activity. With the availability of modern techniques and entertainments, life has become more sedentary. The college students; representing the young age population of community, for different reasons are prone to eat unhealthy foods and to have bad health habits during their college

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years, and it is often assumed that the medical students have a greater knowledge about healthy lifestyle and dietary habits when compared to non-medical students. However there is no evidence to indicate that this knowledge translates into maintaining good health practices [8].

In Saudi Arabia, there are few studies reflecting medical students knowledge and perception of healthy lifestyle habits, so this study was conducted to assess knowledge, attitude and practice of medical students in University of Hail, Hail, Saudi Arabia regarding lifestyle and dietary habits.

Methods:-

Study design:-

A cross-sectional survey was conducted from 2016 October 18th to 2017 March 28th involving 207 participants from medical students enrolled at University of Hail, Hail, KSA.

Study participant:-

A total of 207 students out of 439 medical students in UOH were selected by convenience sampling method. Include all male and female students from basic and clinical years.

Electronic Informed consent of participants were properly obtained before data collection.

Data collection:-

The Knowledge, Attitude and Practice (KAP) of dietary habits and exercise were assessed by an electronic questionnaire which contained three questions about nutrition knowledge, four questions about attitudes, four questions about dietary practice, and four questions about the exercise were completed by the students.

Data Analysis:-

The collected data was coded and analyzed by using descriptive statistics via SPSS , and planned in on deferent charts abstraction.

Results:-

A total of 207 students participated of which females were higher (58.8%) compared to males (41.5%).44% were from basic years and 56.1% were from clinical years. The mean age for male and female student subgroups was 19-24 years respectively. Figure (1)(2)

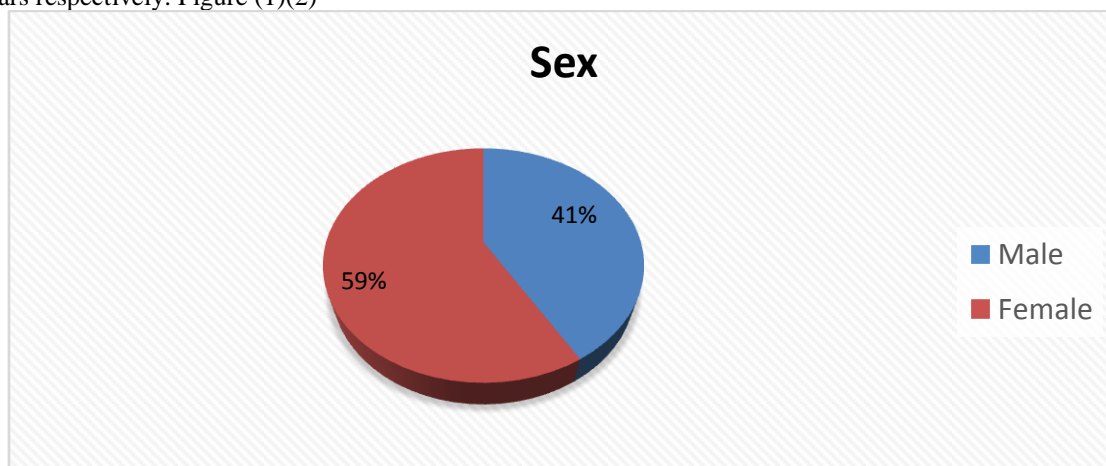
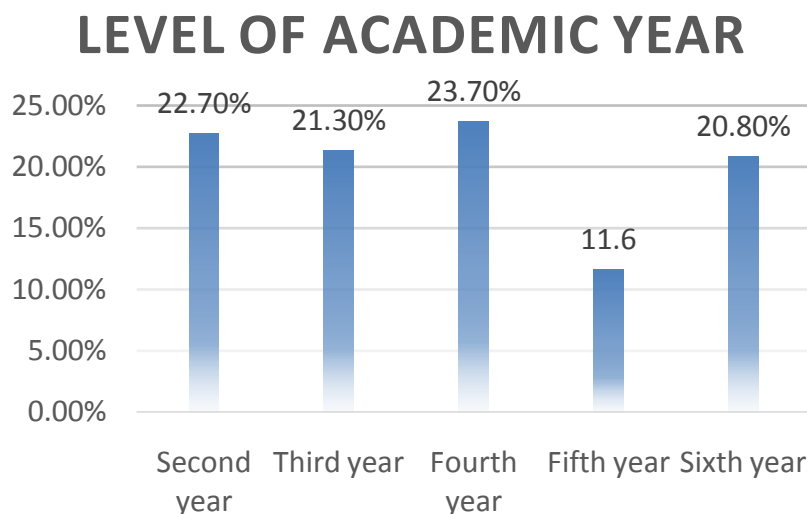


Figure 1:-

**Figure 2:-**

Nutritional knowledge of the study participants is summarized by their answers to three questions in **Table (1)**. Overall, only 11.6% of the students were aware of the composition of balanced diet, 69.6% were aware of the importance of fruits and vegetables, and 73.4% were aware of the meaning of organic food.

Table 1:- Nutritional knowledge among the study cohort (N=207).

	No. (%) of respondents answered correctly	No. (%) of respondents answered incorrectly
Composition of balanced diet	11.6%	79.4%
Importance of fruits and vegetables	69.6%	30.3%
Meaning of organic food	73.4%	26.5%

When the students were asked about how would they rate their nutrition Separately, in females only (2.4%) percent of 125 students rated their nutrition excellent, (14.6%) answered good and most of them (50%) answered quite good. Where the remaining of students their answers varies between poor (25.6%) and very poor (7.3%).(Figure3)

In males, (2.4%) percent of 82 students rated their nutrition excellent, only (9.6%) answered good and (52%) answered quite good, (28%) answered poor, where (8%) answered very poor. So males and females have no big difference in diet rating.(Figure3)

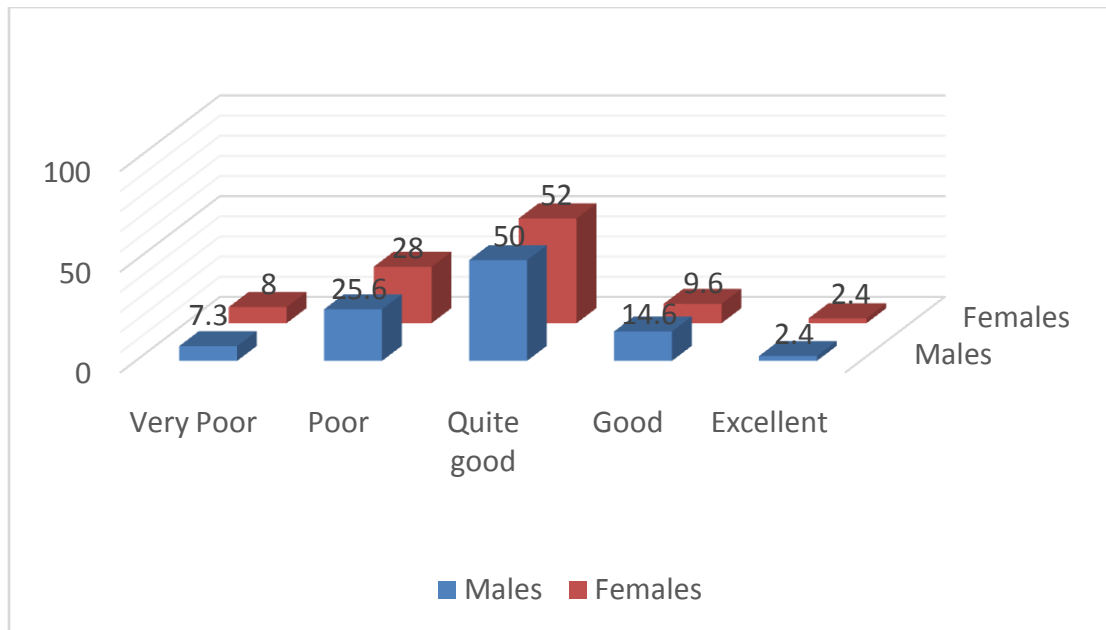


Figure 3:-

Although 69.6% knew about the importance of consuming fruits and vegetables only 11.1% admitted that they eat fruits and vegetables daily, and 21.3% eat it monthly, the rest of students are in between. Figure (4)

Importance of Fruits and vegetables

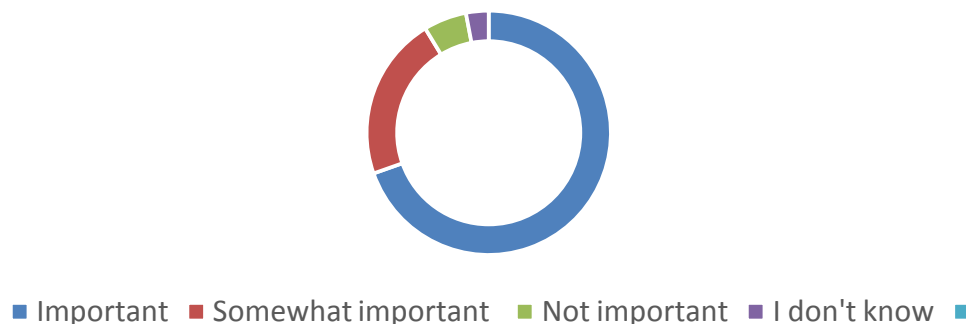


Figure 4:-

About half of the students (54.1%) knew their daily calorie need, but the majority (84.5%) do not calculate their calories whereas 15.5% calculate their calories. About 35.3% of students based all their meals around starchy foods, 28% have only one starchy meal, 31.4% have two starchy meals and 5.3% does not count on starchy foods in any of their meals. Percentage of students who consume sugary snacks daily is 33.3%, and 13% of students consume it in monthly manner. The consumption of pre-prepared food among study cohort is 21.7% daily, and only 15.5% monthly.

These charts shows practices of physical exercises among the study participants (cohort study) . Those who are physically active represent 45.1% of students, 48.6% of them exercise for about 1-2 hours weekly, 22.9% exercise for 2-4 hours weekly, 16.5% exercise for 4-6 hours weekly and only 11.9% exercise for more than 7 hours weekly. Figure (5)

ACTIVE HOURS (WEEKLY)

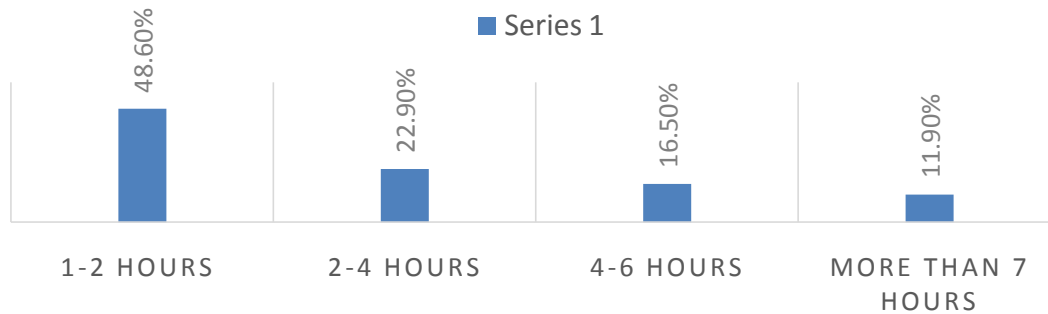


Figure 5:-

When the students were questioned whether their daily routine involve physical activity, most of them 31% stated that they perform more than one type of Exercise and the same percent perform brisk walking only . 15% run, 6% climb stairs as exercise, 8% do high intensity workouts, and only 1% swim. 8% of students perform other types of exercise like weight lifting or self-defense. Figure (6)

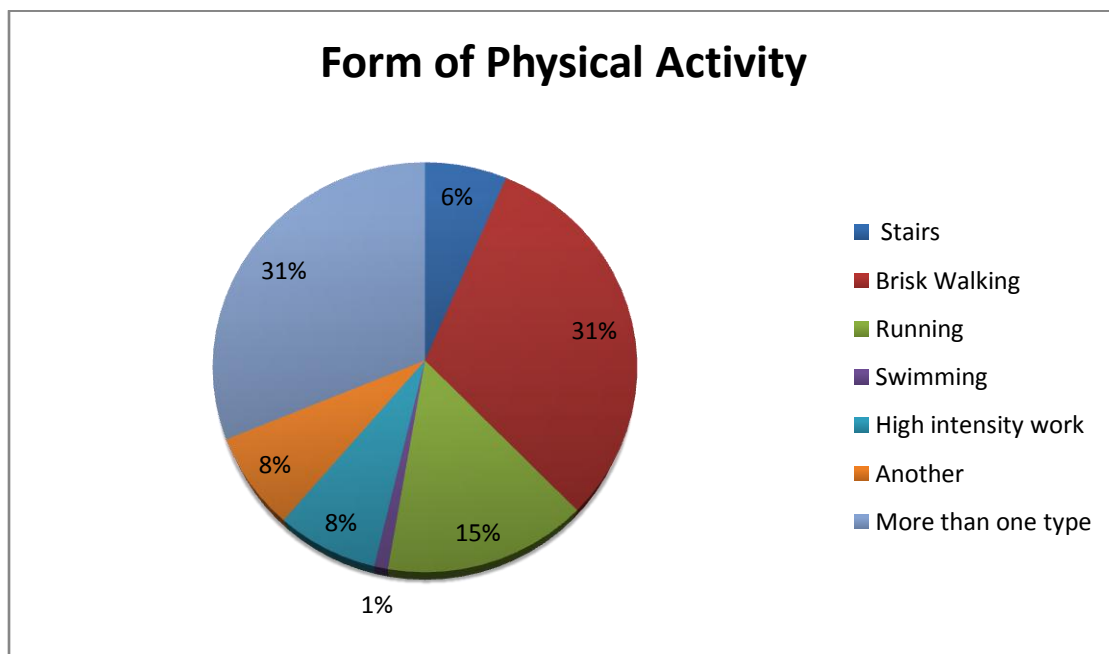


Figure 6:-

Students responded that they practice physical exercises in more than one place, including home about 42.2% of them, 18.3% exercise in public open area, also 18.3% exercise at the gym, only 6.4% exercise at the university campus and 14.7% exercise in more than one of these places .

Concerning attitude towards healthy lifestyle, most of students 29% prefer to eat burger and French fries than other meals, 27.5% of them prefer kabsa (rice and meat) with green salad, 22.7% prefer grilled chicken with white rice and steamed vegetables, 12.1% chose steak with mushroom sauce and mashed potato, and 8.7% prefer green salad only. Regarding fat consumption 58% of students would choose low fat products, 30% would prefer fat free products, 12.1% would prefer full fat products (Figure (7)).

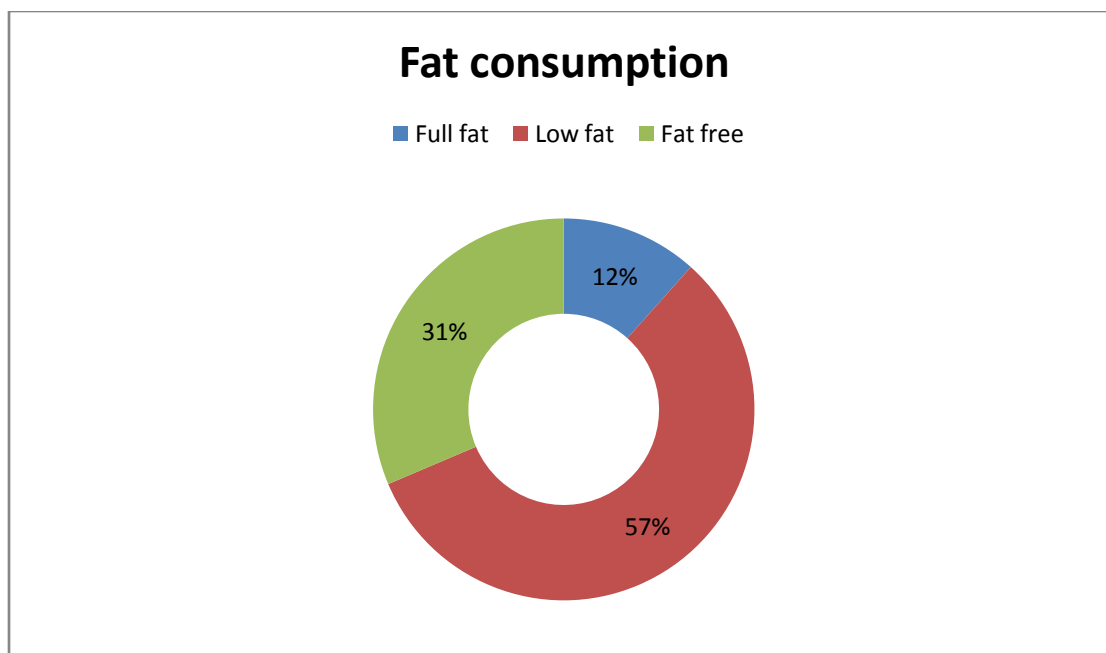
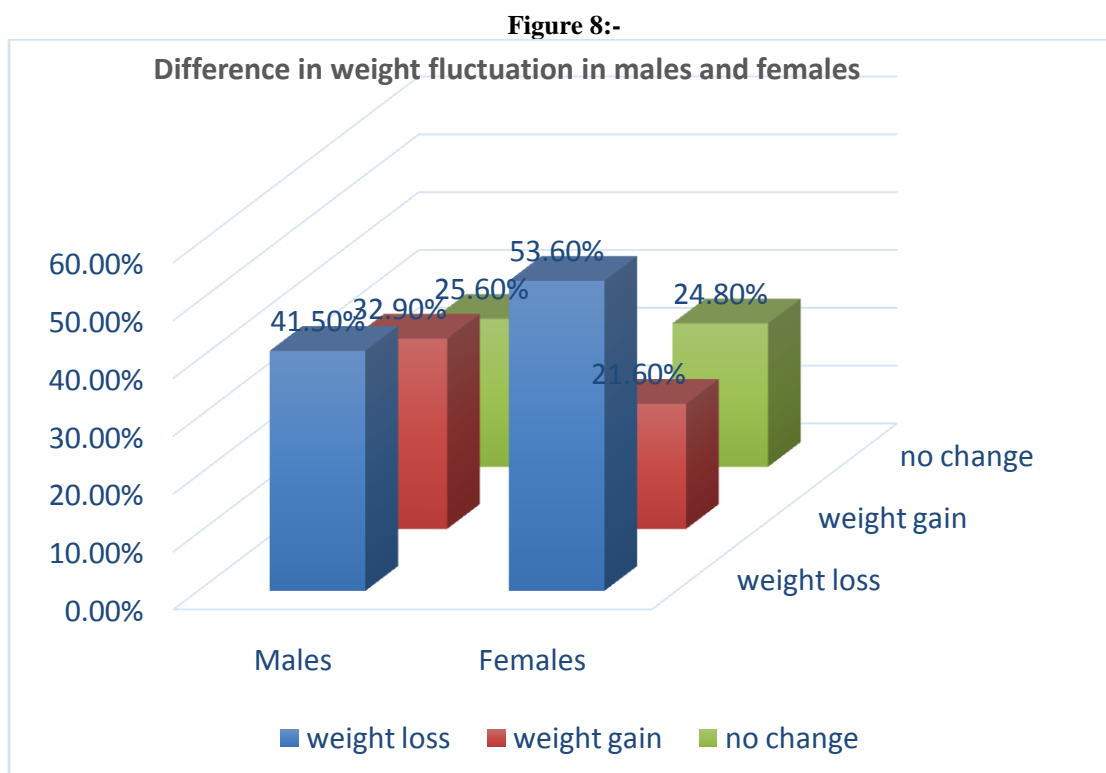


Figure 7:- Regarding weight fluctuations, (53.60%) of females and (41.5%) of males loss their weight during last year. (21.6%) of females and (32.9%) of males gain more weight in last year. The remaining of them have no weight charges.



Feigure 8:-

When females who follow special diet questioned about their diet, only (10.4%) follow the diet always and regular. and majority of them (36.8 %) follow it sometimes (Table2). And regarding type of diet in females' population most of them preferred low fat diet (Table3).

Where in Males (11.0%) follow the diet always and regular, (32.9%) follow it sometimes (Table1) . And about type of diet it varies, where most of them preferred low fat diet and others preferred low calories diet.(Table3).

Table 2:- Male & Females who follow special diet.

Males Population	Frequency	Percentage
1 (Always)	9	11.0%
2 (Sometimes)	27	32.9%
3 (Rare)	16	19.5%
4 (Never)	30	36.6%
	Total = 82	Total = 100%

females Population	Frequency	Percentage
1 (Always)	13	10.4%
2 (Sometimes)	46	36.8%
3 (Rare)	11	8.8%
4 (Never)	55	44.0%
	Total = 125	Total = 100%

Table 3:- Males & Females in type of diet.

Males Population	Frequency	Percentage
1 (Low carbohydrates diet)	7	8.5%
2 (Low calories diet)	18	22.0%
3 (low fat diets)	23	28.0%
4 (None)	34	41.5%
	Total = 82	Total = 100%

Females Population	Frequency	Percentage
1 (Low carbohydrates diet)	14	11.2%
2 (Low calories diet)	22	17.6%
3 (low fat diets)	31	24.8%
4 (None)	58	46.4%
	Total = 125	Total = 100%

Discussion:-

The general Saudi diet is largely dependent on carbohydrates, fats and animal sources of protein. so this study was held to assess whether being in a medical field has increased the knowledge and improved the lifestyle of the medical students in University of Hail or not.

When it comes to knowledge, our study found some variations, majority of the students (79.4%) were not aware of the composition of balanced diet, unlike other KAP held at Jeddah study shows.[9]

Meanwhile (69.6%) knew the importance of minerals and vitamins, which goes in parallel with the previously mentioned KAP study.

Depending on the study results, it seems like the knowledge students earned has no significant role in improving their lifestyle. Despite the majority were aware about importance of minerals and vitamins, less than half the study cohort (40.6%) consume fruits and vegetables on regular basis, contrary to medical students at University of Dammam.[10]

Recently, the interest in adopting a healthy lifestyle and counting calories has significantly increased among young Saudis, but we did not notice this in our study where only 15.5% of the study cohort stated they count their daily

calorie intake. This could be contributed to the ignorance of the true concept of healthy lifestyle and the new applications that counts calories and facilitates committing to a calculated and balanced diet.

Physical activity defined by the WHO as bodily movement produced by skeletal muscles that require energy expenditure and known to has significant benefit to the human body. In this study cohort lower than half of the sample are physical active (45.9%) by deferent forms of activity for 1-2 hours per week in the majority. In a study held at Dammam only 35.5% of their sample are active physically [11]. This is may be due to the lack of time and stressful academic life of the medical students .

With deferent types of diet or physical activity almost have of the study sample (48.8%) loss weight during last year where in the study held at Jeddah only (38%) of their sample [9].

Being a medical student makes them busy most of the time so they would not be able to cook everyday and as the university facilities do not provide healthy food so these factors make it likely to have unhealthy dietary choices among our study cohort. The majority of the students about 30% eat fast food twice or thrice weekly, and about 19.3% do it once a week, although about 33.3% eat cakes, chocolate and drink soda daily, yet 28.5% do it twice or thrice weekly, but more than 88% of the student admitted that they will choose low fat or fat free products instead of full fat products if it's available for them.

Other study held at Dammam have shown that the majority of the students (91.3%) admitted eating fast foods, most of them do it twice or less per week, and about 25% of the students; most of them are males (85%) do it 3 times or more per week, 77.4% of the students are consuming soft drinks slightly higher in females versus male (85% and 67% respectively), furthermore 30% of them do it 3 times or more per week. [10]

Conclusion:-

Overall most medical students were aware of importance of minerals and vitamins in diet yet they were not practicing it adequately in their daily life but the majority of the students were not aware of the composition of balanced diet, improving nutrition knowledge, attitude and dietary practices through nutritional education may help to prevent many nutrition related diseases.

Despite the interest in adapting a healthy lifestyle and counting calories has significantly increased among young Saudis, but we did not notice this in our study, this may be contributed to the ignorance of the true concept of healthy lifestyle and the new applications that counts calories and facilitates committing to a calculated and balanced diet. Or may be due to lack of time as the learning system in the college of medicine, Hail University is very tough.

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RESEARCH ARTICLE

A NOTE ON STABILITY ANALYSIS OF COMPARTMENTAL MATHEMATICAL MODEL FOR THE SPREAD OF MALARIA.

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Mathematical modeling, Malaria disease, equilibrium, stability, basic reproduction number

Abstract

In this study, we present a compartmental model for the spread of malaria in a population where group of individuals were vaccinated. The purpose of this paper is to analyze the transmission dynamics of Malaria by using the compartmental model, including ordinary differential equations for human host and mosquito vector populations. A parallel system is obtained, which has two equilibriums: a disease-free equilibrium and an endemic equilibrium. The stability of the equilibrium points is verified by the basic reproduction number R_0 . Asymptotically stable solution is obtained only for disease-free equilibrium and results are presented graphically.

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Introduction:-

Malaria is most common in Africa. The disease prevails in warmer climates and in areas where there is abundance of humidity and rain. According to latest WHO estimates released in December 2015, there were 214 million cases of malaria in 2015 and 438,000 deaths. At present five known species of plasmodium exists which are; *P falciparum*, *P vivax*, *P ovale*, *P malariae* and *P knowlesi* among them *P. falciparum* is responsible for most fatal infection and is widespread in the tropics. Clinical symptoms of malaria include: high fever, muscle and joint aches, headache, vomiting, chills, sweating and anemia etc. Young children and travellers (who do not have developed immunity) are at high risk of malaria. In highly endemic areas people develop a degree of acquired immunity which is boosted-up by new infections. Immunity lasts for a certain period in the absence of new infection. The duration of infection depends on many factors such as degree of infection, method and time of treatment, resistivity of parasite to drugs and biology of host.

Mathematical modeling has been frequently used in epidemiology and several other fields. Epidemiological modeling is very helpful in identifying important model parameters and suggests improvements in models for future predictions.

We modified the model of NidhiNirwani, R. Khandelwal and V.H. Badshah by performing the stability analysis. The purpose of this study is better understanding the dynamics of malaria.

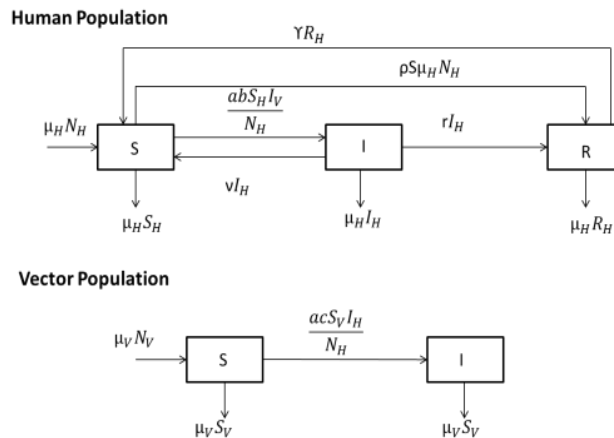
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Description of Model:-

Let N_H and N_V denote human and mosquito population respectively with total population at time t . We assume that human and mosquito population has constant size with birth and death rates equal to μ_H and μ_V .

The human population of size N_H is composed of Susceptible S_H , Infective I_H and Recovered R_H whereas vector population consists of Susceptible S_V and Infective I_V .



We say group of people ρ , $0 \leq \rho \leq 1$ of newborns host population be vaccinated. Assume that vaccine is not perfect and let the effectiveness of vaccine is s , then $\mu_H(1 - \rho s)N_H$ newborns remain susceptible, and $\mu_H \rho s N_H$ directly being removed to R_H . The governing equations are:

Human population:-

$$\begin{aligned} \frac{dS_H}{dt} &= \mu_H(1 - \rho s)N_H - \frac{ab S_H I_V}{N_H} + v I_H + \gamma R_H - \mu_H S_H \\ \frac{dI_H}{dt} &= \frac{ab S_H I_V}{N_H} - v I_H - r I_H - \mu_H I_H \\ \frac{dR_H}{dt} &= \mu_H \rho s N_H + r I_H - \gamma R_H - \mu_H R_H \end{aligned} \quad (1)$$

Vector Population:-

$$\begin{aligned} \frac{dS_V}{dt} &= \mu_V N_V - \frac{ac S_V I_H}{N_H} - \mu_V S_V \\ \frac{dI_V}{dt} &= \frac{ac S_V I_H}{N_H} - \mu_V I_V \end{aligned} \quad (2)$$

The human population of size N_H is formed of susceptible S_H , infective I_H and recovered R_H where vector population is composed of S_V and I_V .

Table 2.1:- Description of parameters of the model.

Variables	Interpretation
a	The average infection rate on man by single mosquito.
b	The proportion of bites on man that produce an infection.
c	The probability that a mosquito becomes infectious.
γ	The per capita rate of loss of immunity in human hosts.
r	The rate at which human hosts acquire immunity.
v	The rate of recovery of human hosts from the disease.

Using $S_H + I_H + R_H = N_H$ and $S_V + I_V = N_V$, eqs (1) and (2) become

$$\begin{aligned}\frac{dS_H}{dt} &= \mu_H(1 - \rho s)N_H - \frac{abS_H I_V}{N_H} + \nu I_H + \Upsilon(N_H + S_H + I_H) - \mu_H S_H \\ \frac{dI_H}{dt} &= \frac{abS_H I_V}{N_H} - \nu I_H - r I_H - \mu_H I_H \\ \frac{dI_V}{dt} &= \frac{acI_H(N_V - I_V)}{N_H} - \mu_V I_V\end{aligned}\quad (3)$$

Writing the eq (2.3) in population proportion

$$\begin{aligned}S_h &= \frac{S_H}{N_H}, I_h = \frac{I_H}{N_H} \text{ and } I_v = \frac{I_V}{N_V}, \\ \frac{dS_h}{dt} &= \mu_H(1 - \rho s) - ab\phi S_h I_v + \nu I_h + \Upsilon - \Upsilon(S_h + I_h) - \mu_H S_h \\ \frac{dI_h}{dt} &= ab\phi S_h I_v - \nu I_h - r I_h - \mu_H I_h \\ \frac{dI_v}{dt} &= acI_h(1 - I_v) - \mu_V I_v\end{aligned}\quad (4)$$

Where $\phi = \frac{N_V}{N_H}$ is the ratio of host and vector population.

Further we rescale t by ac and let $x = S_h, y = I_h$ and $z = I_v$

$$\begin{aligned}\frac{dx}{dt} &= \mu(1 - x) - \sigma x - \eta x z + k y + \varphi - \varphi y \\ \frac{dy}{dt} &= \eta x z - (k + m)y \\ \frac{dz}{dt} &= y(1 - z) - \omega z\end{aligned}\quad (5)$$

Where

$$\mu = \frac{\mu_H}{ac}, \pi = \rho s, \eta = \frac{b\phi}{c}, m = \frac{\mu_H + r}{ac}, \omega = \frac{\mu_V}{ac}, k = \frac{\nu}{ac}, \sigma = \frac{\mu_H + \Upsilon}{ac}, \varphi = \frac{\Upsilon}{ac}$$

Euler Method:-

Values of parameters used were $\beta = 0.000318$ and $\mu = 0.0175$ and the initial conditions were $S(0) = 460, I(0) = 12, R(0) = 0$

The Euler method

$$\begin{cases} w_0 = \alpha \\ w_{i+1} = w_i + hf(t_i, w_i) \end{cases}, \quad i = 0, 1, 2, \dots, N-1$$

The time interval was [0 90] and $N=10$ so

$$h = \frac{b-a}{N}$$

$$h = \frac{90-0}{10} = 9$$

$$t_0 = 0, t_1 = 9, t_2 = 18, t_3 = 27, \dots, t_{10} = 90$$

$$w_0 = \begin{pmatrix} S(0) \\ I(0) \\ R(0) \end{pmatrix} = \begin{pmatrix} 460 \\ 12 \\ 0 \end{pmatrix} = \begin{pmatrix} w_{1,0} \\ w_{2,0} \\ w_{3,0} \end{pmatrix}$$

Iteration-1 for $i=0$

$$w_1 = w_0 + hf(t_0, w_0)$$

$$w_1 = \begin{pmatrix} w_{1,0} \\ w_{2,0} \\ w_{3,0} \end{pmatrix} + hf(0, w_0)$$

$$w_1 = \begin{pmatrix} 460 \\ 12 \\ 0 \end{pmatrix} + 9 \begin{pmatrix} -0.00038 \\ -0.20962 \\ 0.21 \end{pmatrix}$$

Steady State And Equilibrium Points:-

The system (5) has a disease-free equilibrium point $E_0 (\frac{\mu(1-\pi)+\varphi}{\sigma}, 0, 0)$ and an endemic equilibrium point $E_1(x_e, y_e, z_e)$, where

$$\begin{aligned} x_e &= \frac{(k+m)[\mu(1-\pi)+\varphi+(m+\varphi)\omega]}{\eta(m+\varphi)+\sigma(k+m)} \\ &= \frac{(v+\mu_H+r)[ac\{\mu_H(1-\rho s)+\gamma\}+(\mu_H+r)\mu_V]}{ac[ab\phi(\mu_H+r+\gamma)+(\mu_H+\gamma)(v+\mu_H+r)]} \\ y_e &= \frac{\eta[\mu(1-\pi)+\varphi]-\sigma(k+m)\omega}{\eta(m+\varphi)+\sigma(k+m)} \\ &= \frac{a^2bc\phi[\mu_H(1-\rho s)+\gamma]-(\mu_H+\gamma)(v+\mu_H+r)\mu_V}{ac[ab\phi(\mu_H+r+\gamma)+(\mu_H+\gamma)(v+\mu_H+r)]} \\ z_e &= \frac{\eta[\mu(1-\pi)+\varphi]-\sigma(k+m)\omega}{\eta[\mu(1-\pi)+\varphi+(m+\varphi)\omega]} \\ &= \frac{a^2bc\phi[\mu_H(1-\rho s)+\gamma]-(\mu_H+\gamma)(v+\mu_H+r)\mu_V}{ab\phi[ac\{\mu_H(1-\rho s)+\gamma\}+(v+\mu_H+r)\mu_V]} \end{aligned}$$

This has been obtained by setting the time derivatives of the Eq (5) equal to zero. Here the basic reproduction number R_0 is defined by

$$R_0 = \frac{\eta[\mu(1-\pi)+\varphi]}{\sigma(k+m)\omega} = \frac{a^2bc\phi[\mu_H(1-\rho s)+\gamma]}{(\mu_H+\gamma)(v+\mu_H+r)\mu_V}$$

And an endemic equilibrium $E_1(x_e, y_e, z_e)$ is stable when

$$R_0 = \frac{\eta[\mu(1-\pi)+\varphi]}{\sigma(k+m)\omega} > 0$$

$$w_1 = \begin{pmatrix} 459.99658 \\ 10.11342 \\ 1.89 \end{pmatrix}$$

Asymptotic Behaviour Of The Model:-

Theorem 1: If $R_0 < 1$, then the disease-free equilibrium is locally E_0 is locally stable and if $R_0 = 1$, E_0 is stable.

Proof:

To discuss the stability of the model, the governing dynamical system is

$$F_1 = \mu(1-\pi) - \sigma x - \eta xz + ky + \varphi - \varphi y \quad (6)$$

$$F_2 = \eta xz - ky - my \quad (7)$$

$$F_3 = y(1-z) - \omega z \quad (8)$$

The variation matrix of the above system is given by

$$J = \begin{bmatrix} -\sigma - \eta z & k - \varphi & -\eta x \\ \eta z & -k - m & \eta x \\ 0 & 1 - z & -y - \omega \end{bmatrix}$$

For disease free equilibrium $E_0 (\frac{\mu(1-\pi)+\varphi}{\sigma}, 0, 0)$ the variation matrix will be

$$J(E_0) = \begin{bmatrix} -\sigma & k - \varphi & \frac{-\eta[\mu(1-\pi)+\varphi]}{\sigma} \\ 0 & -k - m & \frac{\eta[\mu(1-\pi)+\varphi]}{\sigma} \\ 0 & 1 & -\omega \end{bmatrix}$$

The characteristic equation of it will be

$$(\sigma + \lambda) \left[\lambda^2 + (k+m+\omega)\lambda + (k+m)\omega - \frac{\eta[\mu(1-\pi)+\varphi]}{\sigma} \right] = 0 \quad (9)$$

By the above equation at eigenvalues, one can easily see that disease-free equilibrium E_0 is locally stable if

$$(k+m)\omega - \frac{\eta[\mu(1-\pi) + \varphi]}{\sigma} > 0$$

i.e., $R_0 < 1$

Stability of disease-free equilibrium:-

The assumed values of all the parameters are given in the table below:

Parameters	Values
μ	1.16
η	0.00492
π	0.009
σ	2.1724
ϕ	0.9
k	0.0026
ω	1
m	0.011

With initial conditions $(x(0), y(0), z(0)) = (0.5, 0.5, 0.4)$

After substituting the values in equation (4.4) we get;

$$(2.1724 + \lambda)(\lambda^2 + 1.0136\lambda) + 0.0089 = 0(10)$$

Where,

$$\lambda_1 = -2.1724$$

$$\lambda_2 = -0.0089$$

$$\lambda_3 = -1.00475$$

We see all Eigen values are real and negative, also $R_0 = 0.3413084471 < 1$, so the system is stable.

Stability of disease-free equilibrium:-

Taking equations (6) and (7); the variation matrix is given by

$$J_1 = \begin{bmatrix} -\sigma - \eta z & k - \varphi \\ \eta z & -k - m \end{bmatrix}$$

For disease free equilibrium $E_0 \left(\frac{\mu(1-\pi)+\varphi}{\sigma}, 0, 0 \right)$ the variation matrix will be

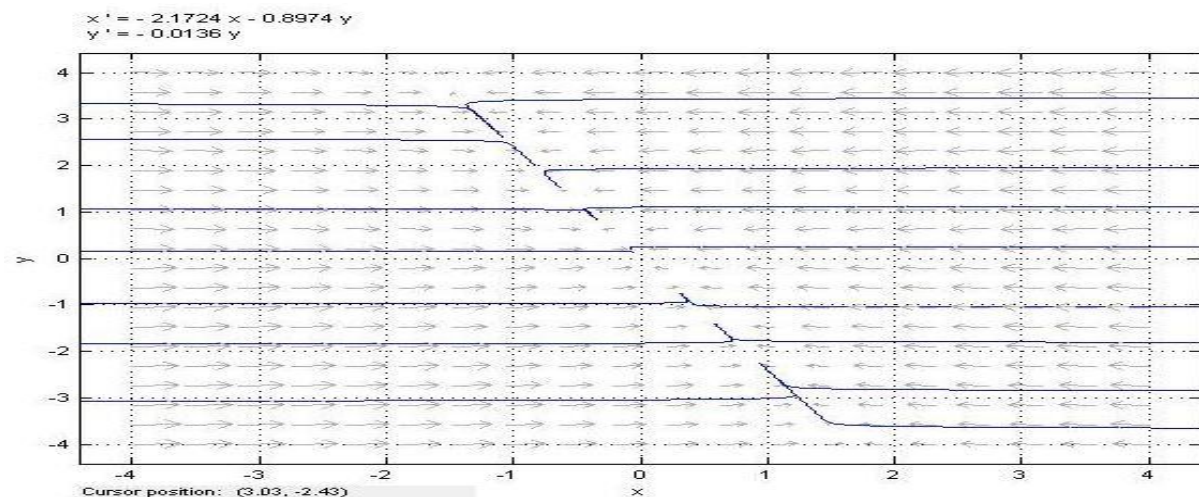
$$J_1 = \begin{bmatrix} -\sigma & k - \varphi \\ 0 & -k - m \end{bmatrix}$$

And putting the values from the table of disease-free equilibrium, we will get the following linear system;

$$\dot{x} = -2.1724x - 0.89747y$$

$$\dot{y} = -0.0136y$$

Now we take the graph of above equation through p-plane



Now, taking equations (6) and (7); the variation matrix is given by

$$J_2 = \begin{bmatrix} -k - m & \eta x \\ 1 - z & -y - \omega \end{bmatrix}$$

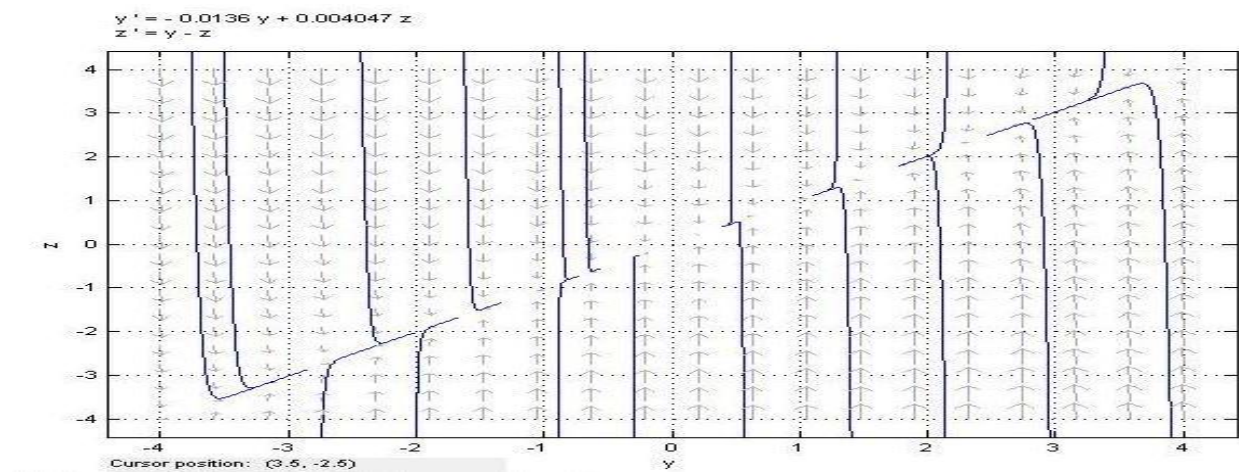
For disease free equilibrium $E_0 (\frac{\mu(1-\pi)+\varphi}{\sigma}, 0, 0)$ the variation matrix will be

$$J_1 = \begin{bmatrix} -k - m & \frac{\eta[\mu(1-\pi) + \varphi]}{\sigma} \\ 1 & -\omega \end{bmatrix}$$

And putting the values from the table of disease-free equilibrium, we will get the following linear system;

$$\begin{aligned} \dot{y} &= -0.0136y + 0.004047z \\ \dot{z} &= y - z \end{aligned}$$

Now we take the graph of above equation through p-plane



Now, taking equations (6) and (9); the variation matrix is given by

$$J_3 = \begin{bmatrix} -\sigma - \eta z & \eta x \\ 0 & -y - \omega \end{bmatrix}$$

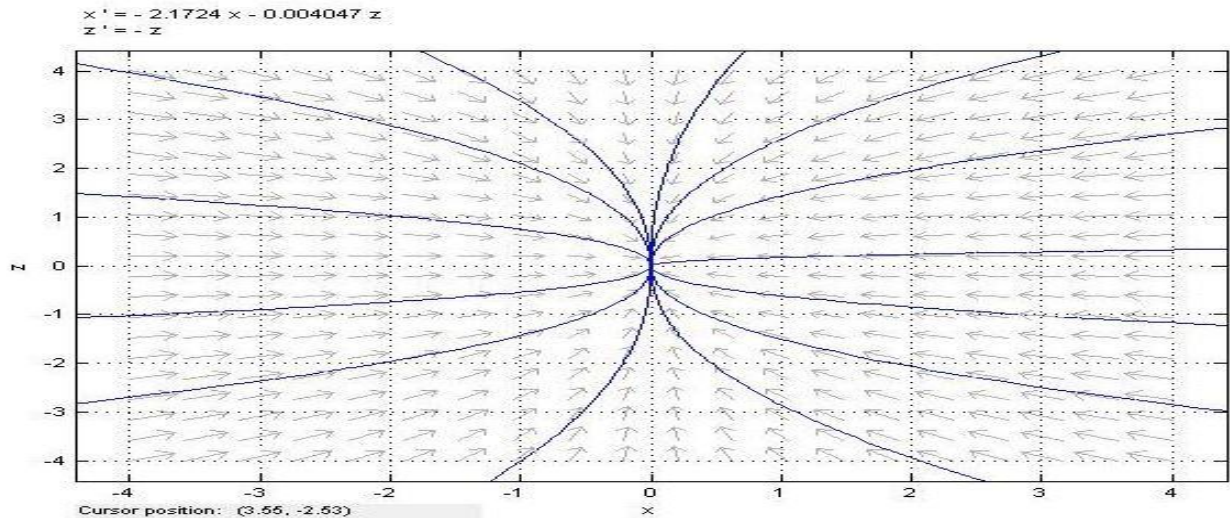
For disease free equilibrium $E_0 (\frac{\mu(1-\pi)+\varphi}{\sigma}, 0, 0)$ the variation matrix will be

$$J_1 = \begin{bmatrix} -\sigma & \frac{\eta[\mu(1-\pi) + \varphi]}{\sigma} \\ 0 & -\omega \end{bmatrix}$$

And putting the values from the table of disease-free equilibrium, we will get the following linear system;

$$\begin{aligned} \dot{x} &= -2.1724x + 0.004047z \\ \dot{z} &= -z \end{aligned}$$

Now we take the graph of above equation through p-plane



Conclusion

In this paper we present compartmental model for spread of malaria. An identical system is obtained which has two equilibrium points; a disease-free equilibrium and an endemic equilibrium. We discussed the stability of the disease-free equilibrium point numerically by computing Eigen-values and by using basic reproduction number. Further we perform stability analysis and graphically verify our results. All the graph indicate that the system is stable.

Future Work

The model presented here is multi-dimensional. In future we can perform stability analysis for endemic-equilibrium. Furthermore we can establish these results by using any of the numerical methods. Also we can work on its sensitivity analysis and parameter estimation.

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RESEARCH ARTICLE

STUDIES ON PRE-GERMINATION SEED TREATMENT OF A TRADITIONAL MEDICINAL PLANT -MADHUCA LONGIFOLIAL.

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Key words:-

Madhuca longifolia, pre-sowing treatment, seedling growth.

Abstract

Madhuca longifolia L (Family : Sapotaceae) is a medicinal and economic important tree. This study was carried out to know the effect of pretreatment on seed germination and seedling growth parameters of size graded *M. longifolia* seeds viz. large, medium and small. The result shows that the large and medium sized seeds treat with tap water is significantly increased the germination and seedling growth compared to control and other treatments.

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Introduction:-

To achieve success in any plantation programs a good start is essential from the germination stage. In order to that, many tree species germinate readily when subjected to favorable conditions of moisture, gaseous balance, light and temperature, but many other species possess degree of seed dormancy or have other germination problems. In order to ensure quick and even germination in nursery beds, artificial treatments to be attempted before sowing (Hanumantha et al., 2002). Trees have been considered as vital part of the life and their significance being described in epics. Many traditional societies all over the world value a large number of plant species for a wide variety of reasons viz., food, fibre, shelter, medicine, etc..

Madhuca longifolia L. is a commonly known as Mahua, or Butter nut tree. It is a medium sized deciduous tree distributed in Nepal, India and Srilanka (Saluja et al., 2011). *M. longifolia* seeds are of economic importance as they are good source of edible fats (Chatterjee and Pakrashi 2000). The medicinal properties attributed to this plant are stimulant, demulcent, emollient, heating and astringent (Awashti and Mitra 1967). The flowers have been traditionally used as cooling agent, tonic, aphrodisiac, and astringent, demulcent and for the treatment of chronic tonsillitis (Chandra 2001). The leaves are expectorant and also used for chronic bronchitis. The bark is used for rheumatism (Sunita and Sorojini, 2013). The root powder is used to cure diarrhea and other chronic fluxes (Chatterjee and Pakrashi 2000).

Since the present study was carried out to find out the effectiveness of size graded seeds in various pretreatment methods to understand the behavior of seed germination and seedling growth which will reduce the maintenance cost in nursery and result in good crop-stand.

Materials and Method:-

The freshly collected seeds of *M. longifolia* were sorted out into three groups namely large, medium and small on the bases of visual observation. The following treatments was given to the seeds

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- T0: Control (non-treated)
T1: Seeds were soaked in warm water (50-55°C) for 10 minutes,
T2: Seeds were soaked in warm water (50-55°C) for 15 minutes,
T3: Seeds were placed under running tap water for 12 hours,
T4: Seeds were placed under running tap water for 24 hours,
T5: Seeds were soaked in concentrated sulfuric acid for 10 minutes,
T6: Seeds were soaked in concentrated sulfuric acid for 20 minutes,
T7: Seeds were soaked in concentrated sulfuric acid for 1 hour,
T8: Seeds were soaked in concentrated nitric acid for 10 minutes,
T9: Seeds were soaked in concentrated nitric acid for 20 minutes,
T10: Seeds were soaked in concentrated nitric acid for 1 hour,
T11: Seeds were manually by rubbing on sand paper at the cotyledon ends,
T12: A small piece of seed coat was removed mechanically using razor blade.

After pretreatment by various methods (T1- T10), the seeds were thoroughly washed in tap water and were sown in 30cm X 11cm polythene bags containing sand medium kept at room temperature (28±2°C). The germination studies were carried out with five replicates and each replication consisted of fifty seeds of uniform size. The mechanically scarified (T11 and T12) seeds are sown immediately. Watering was done daily and germination was recorded on alternate days during 90 day period for experimentation. The statistical analysis was done by the method of Duncan's Multiple Range Test (DMRT).

Result and Discussion

In *M. longifolia*, large sized seeds treated with tap water (24 hours) were enhanced the higher percentage of germination when compared to other treatments. Whereas the medium and small sized seeds exhibited poor germination in all the treatments (Fig. 1). The continuous soaking of seeds in running tap water will leaches out inhibitory chemicals, thereby improving the germination of chemically dormant seeds (Bewley and Black 1994). The root and shoot length of large sized seeds exhibited more in 12 hours tap water treated seedlings followed by 24 hours treatment compared to control (Fig. 2 & 3). Mean number of days taken for germination of the seedlings was also less for large and medium sized seeds than the small size.

In the present study large sized seeds of *M. longifolia* treated with tap water for 48 hrs showed higher percentage of germination and seedling growth. This study is concordant to Naidu and Mastan (2001) in *Pterocarpus santalinus*. They observed that the seeds kept under running tap continuously for 6 days exhibits more germination.

Fig -1: Effect of pretreatment on seed germination of size graded seeds of *M. longifolia*

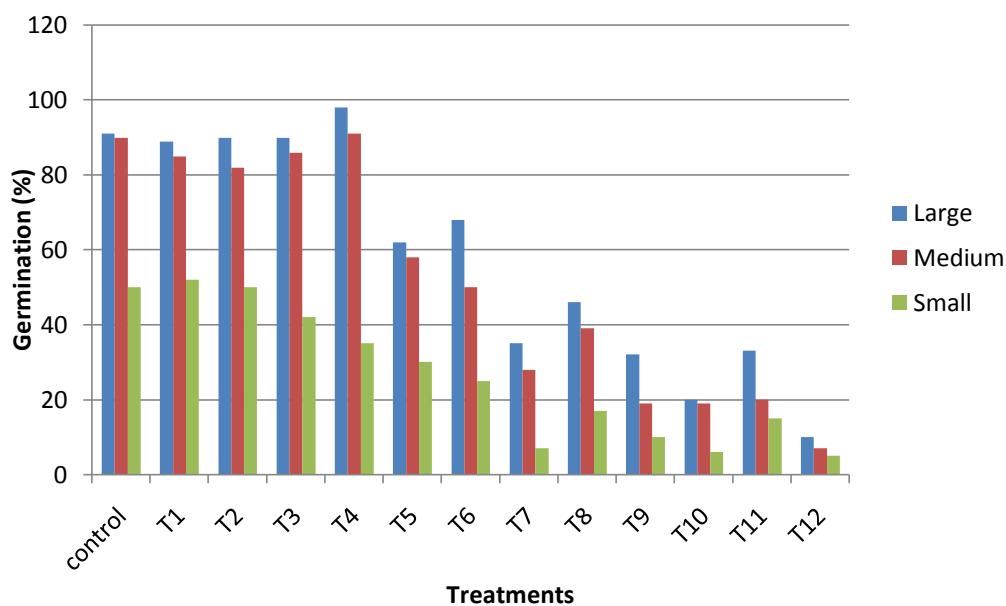
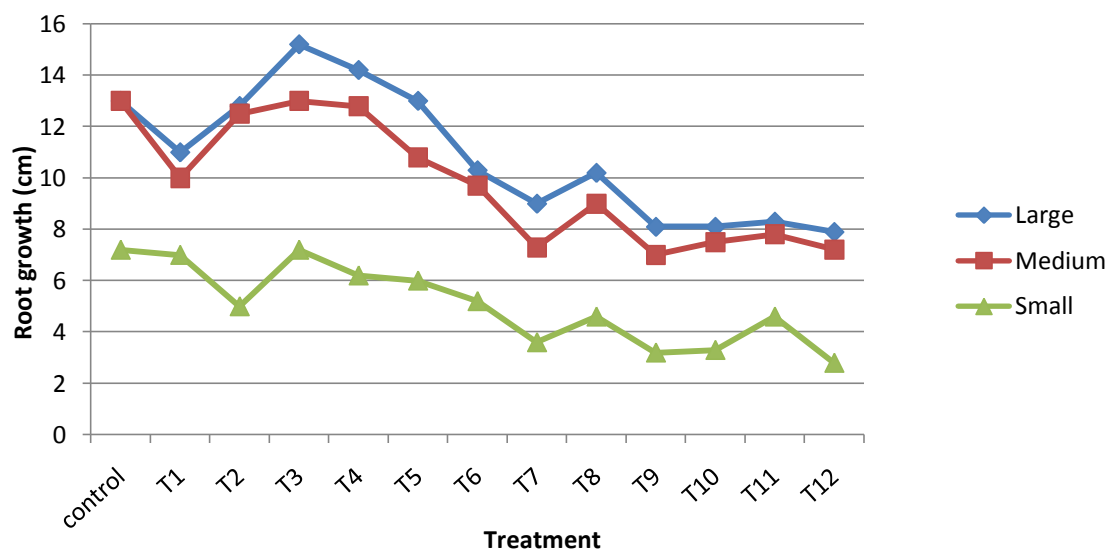
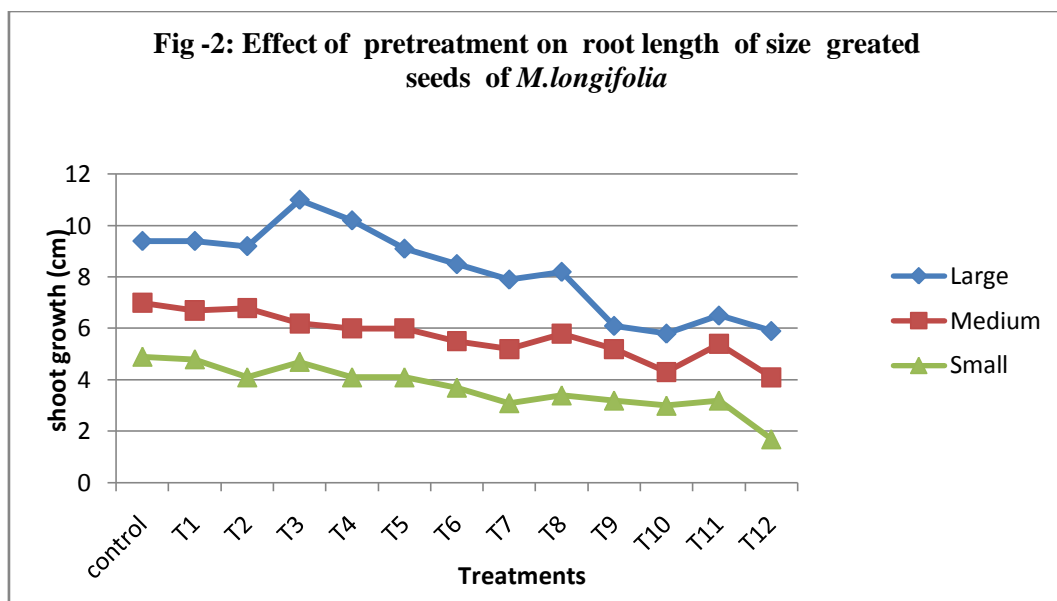


Fig -2: Effect of pretreatment on root length of size graded seeds of *M. longifolia*





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RESEARCH ARTICLE

EFFECT OF SEED SIZE ON SEED GERMINATION OF *AEGLE MARMELOS*, L. CORR., TAMILNADU INDIA.

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Key words:-

A. marmelos, Seed germination,
Seedling growth

Abstract

The present study was undertaken to examine the effect of physical characters seeds on physiological characters like seed germination and seedling growth of *A. marmelos* L. Corr. Seeds were size graded in to large, medium and small based on seed size (length and breadth) and weight. The result revealed that the size-graded seeds influence the seed germination and seedling growth. The large seeds gave maximum germination followed by medium and small size seeds.

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Introduction:-

Aegle marmelos (L.) Corr., is an endangered medicinal tree generally known as Bael belonging to the family Rutaceae. It is a medium sized deciduous and aromatic tree about 12-15m height, leaves trifoliate, aromatic, alternate. Leaflets ovate or ovate- lanceolate; greenish white flowers, Large, globose fruits, woody berry (Mazumder et al 2006). It is a popular medicinal plant in Ayurveda and siddha medicines to treat a variety of ailments (Venudevan 2013). Bael fruits are popular due to its medicinal and nutritional properties. Fruits juice is used to cure of diarrhea and dysentery, jaundice, ulcer and eczema (Nadkani 1954). The roots are useful for treating diarrhea and dyspepsia (Pallab Maity 2009). The tree is normally propagated through seeds and seeds required specific quality for better performance with uniformity on seedling production in nursery. However, seeds are exhibit wide varieties such as seed morphological characters as per the influence of development and maturation (Abdul- Baki and Baker, 1973) therefore the present study was conducted to evaluate the effect of seed size and weight on germination and initial growth in the nursery

Materials and Method:-

Physical characters:-

Mature fruits of *Aegle marmelos*, were collected during November to April in Coimbatore district, Tamilnadu, India. The seeds were extracted by macerating the fruits and washed in running water and then air dried at room temperature ($28 \pm 2^\circ\text{C}$). The seeds were graded into three size grades i.e., large, medium and small. Seed samples (100 seeds) were drawn from each grade and measured for length and breadth using vernier- scale. The length was measured from the base to the tip of the seed and recorded in cm. The breadth was measured at its broadest portion and expressed in cm. One hundred seeds were counted and its weight was recorded in grams using electronic top pan balance as per ISTA (1985) rules. Number of seeds per kilogram was also calculated.

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Physiological Characters:-

Hundred seeds of three different sizes (based seed size) were sown in 30/11 inch polythene bags with holes at the rate of one seed / bag. These bags were filled with sand + red soil (1:1). The weight of the medium was one kilogram in each bag for all cases. The optimum depth was 1.0 to 1.5 cm below the surface of the soil. There were four replications consisting of one hundred bags /replication. The bags were watered daily. Germination percentage was calculated using the following formula:

$$\text{Germination \%} = \frac{\text{No. of seed germinated}}{\text{Total no of seeds sown}} \times 100$$

The growth and biomass productions of seedlings were taken once in fifteen days from the date of germination up to 90 days. Ten seedlings in each replication at random were uprooted by cutting the polythene bags without damaging the roots were washed with water. The root and shoot lengths were measured and expressed in cm. The seedlings were dried in a hot-air oven at 60°C for 48 hours. After cooling, them in desiccators weighed in electronic top pan balance and expressed in grams.

Result and Discussion:-**Physical Parameter :**

The physical characteristics of seeds viz., seed length, seed breadth, 100 seeds weight, percentage of occurrence and No. of seeds / kg was observed and tabulated [Table-1].

The seeds of *A. marmelos* were oblong in shape and milky white in colour. They were size Graded into large, medium and small representing in the ratio of 2:4:1. Seed length, breadth and 100 seed weight were noticed to be more in larger seeds followed by medium and small. Number of seeds / Kg was lowest in larger seeds 5300 (± 237.1) followed by higher number in medium 6437 (± 515.3) and 8125 (± 772.9) in small. Similar seed morphological studies were made by Athaya (1985) in certain tree species growing in tropical dry deciduous mixed forests of central India. Roy [1985] in *Albizia lebbbeck*, Ponnammal *et.al.* , [1993] in *Hardwickia binnata* ,Suresh *et .al.*, [2003] in *Bassia longifolia* and Kathiravan(2004) in *Jatropha*.

Physiological characters:-

Seed grading is a continuous process in seed cleaning in which the seeds are graded according to their size or weight. The purpose of grading is to improve the physiological quality of the seed lot by eliminating small, empty and underdeveloped seeds

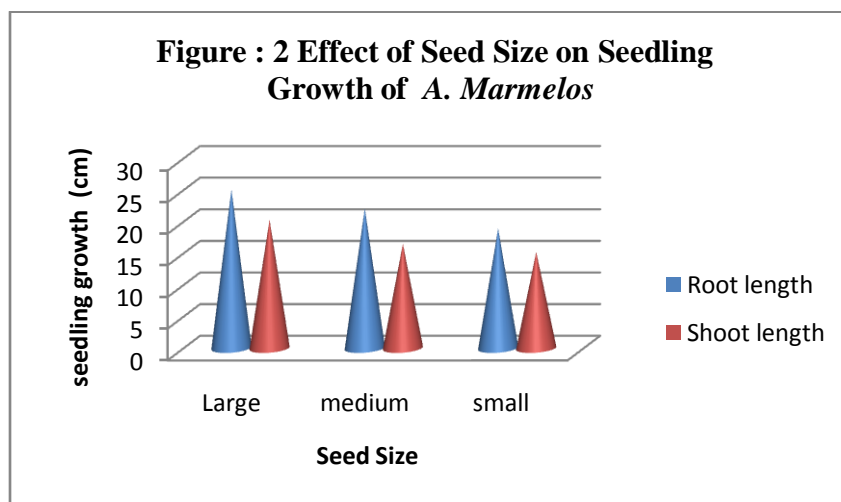
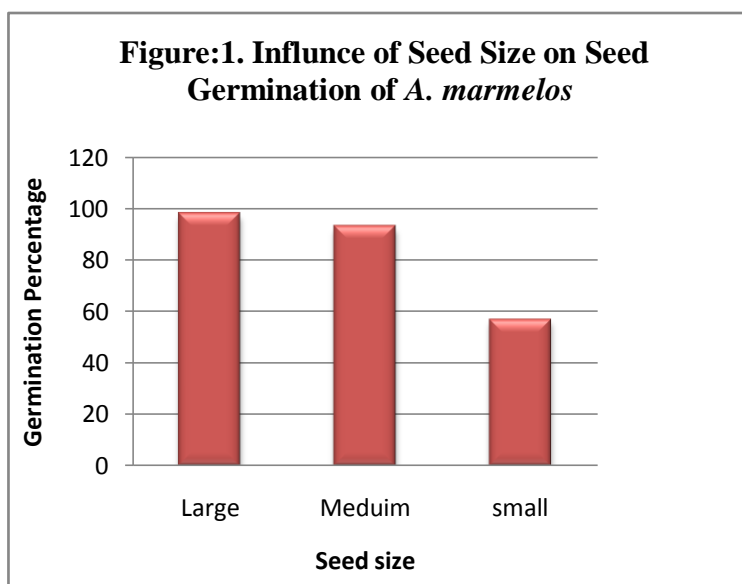
Among freshly collected seeds of *A. marmelos* germination was noticed 6- 10 days after sowing. The large sized seeds exhibited higher (98%) percentage of germination compared to that of medium and smaller sized seeds (Fig: 1.) This may be due to large quantity of stored food materials in the larger seeds, which contributed for best germination. The better germination of larger seeds also noticed in, *Aholo sercia* by Hillum (1990), *Syzigium cumini* by Ponnammal *et al.*, (1992) and *Hardwickia binnata* by Ponnammal *et al.*,(1993). *Dalbergia sissoo* by Yadav *et al.*, (1998), *Bassia longifolia* by Suresh *et al.*, (2003) and *Pinus wallichiana* by Ghildiyal and Sharma (2005)

In the growth performance of the seedlings (90 day old), root and shoot length was higher in large sized seeds (25.1 cm and 20.3 cm) followed by medium (22.1 cm and 16.5 cm) and small (18.9cm and 15.2cm) respectively. This result conformed that the bigger seeds giving rise to larger seedlings. Due to bigger embryo, which had more nutrients to support more growth has been reported by Ponnusamy *et al.*, (1991) in *Neem*. Similar findings were also reported by Chauhan and Raina (1980) in *Pinus roxburghii.*, Ponnammal (1992) in *Syzigium cumini* Arjunan *et al.*, (1995) in *Pongamia pinnata* and Dar *et al.* , (2002) in *Albizia lebbck*, *Acacia catechu* and *Pinus roxburghii*. In all these three sized seeds, the root growth was found to be faster than the shoot growth. After 90 days of seed germination, highest biomass production was noticed in seedlings from large seeds (0.186 g/plant) (Table.13).

Table 1:- Physical characters of *A. marmelos* seeds.

Seed size	100 Seed weight (g)	Seed length (cm)	Seed breadth (cm)	Occurrence (%)	No. of seeds / Kg	Seed Shape and Colour
Large	37.0 \pm 1.85	0.81 \pm 0.09	0.38 \pm 0.05	20-25	5300 \pm 237.1	Oblong Milky White
Medium	30.0 \pm 3.29	0.65 \pm 0.05	0.32 \pm 0.04	55-60	6437 \pm 515.3	
Small	22.0 \pm 2.94	0.48 \pm 0.06	0.25 \pm 0.07	18-22	8125 \pm 772.9	

\pm = Standard deviation



Conclusion:-

Seed is one of the most important inputs for forest nursery production. Based on the results of the present study, seed germination and seedling growth characters were higher in larger seeds. So this study concluded that the seed size and seed quality are positively related.

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