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RESEARCH ARTICLE

ANTIBACTERIAL EFFICACY OF PLANT EXTRACTS OF *OCIMUM SANCTUM* L.

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Abstract

The antibacterial efficacy of aqueous and ethanolic extracts of leaf, stem and root of *Ocimum sanctum* L. were studied against 5 known pathogenic gram positive and gram negative bacteria (namely, *Staphylococcusaureus*, *Clostridium bovis*, *Salmonella typhi*, *Klebsiellapneumoniae* and *Escherichia coli*) by cup diffusion method with a control antibiotic (chloramphenicol). The aqueous and ethanolic extracts of *O. Sanctum* L. were found to be effective against all the bacteria cultured and tested in laboratory. The extracts of leaf, stem and root were most effective against *S. typhi* showing zone of inhibition (ZI) =11 mm, 10.90 mm and 10.75 mm (ethanolic) respectively and ZI = 10.75 mm, 10.60 mm and 10.55 mm (aqueous) respectively, followed by *C. bovis* and *S. Aureus*, which were at par with control (chloramphenicol). Minimum Zone of Inhibition for leaf, stem and root extracts were recorded against *E. coli* showing ZI = 4.00 mm, 4.20 mm, 3.75 mm (ethanolic) respectively and ZI = 3.75 mm, 3.70 mm, 3.50mm (aqueous) respectively. The results indicate that leaf extract of *Ocimum sanctum* L. was most effective as it was most potent against all the pathogens tested. This significant potential of *Ocimum sanctum* L. makes it an effective antibacterial agent and can be further explored for their antibacterial efficacy. This work may be useful to find out new antibacterial drugs having no side-effects.

Introduction:-

Antibiotics are very important with respect to healthcare. However, due to indiscriminate use of antibiotics, pathogens develop resistance against them. Investigations are needed to develop safe drug from the botanicals used in traditional system of medicines. Plant extracts are potential sources of novel antimicrobial compounds especially against bacterial pathogens **(Kumar *et al.*, 2009, Sharma *et al*, 2011)**. The medicinal value of *O. Sanctum* has been known for a long time. Systematic and scientific investigation is needed to find out the antibacterial efficacy of *O. sanctum* and to know its potentiality as an antibacterial agent **(Akerele O, 1993; Chopra *et al.*, 1956; Amadioha *et al.*, 1998)**.

O. sanctum L. is an aromatic plant with two main morphology i.e green leaved and purple leaved of the family LAMIACEAE, cultivated in India and Nepal. It is native of eastern world, widespread and sacred plant used from time immemorial for prevention of diseases either in fresh or dried form. It is an erect, much branched subshrub, 30–60 cm tall with hairy stems. Leaves (2.5 to 7 cm) are acute, oblong in shape, green or purple in colour and pubescent on both sides with minute dotted glands having aromatic odour and slight pungent taste. Flowers are purple

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colouredand arranged in racemose inflorescence. The fruits are like nutlets having four, small seeds which are oval, flattened and reddish black in colour **(Manandhar NP, 2000)**.

Material and Methods:-

Internationally accepted standard laboratory techniques were carried out to screen and identify antibacterial activities of aqueous and ethanolic extract. The brief procedure is as follows.

Collection of Plant Material:-

Fresh plant parts were collected randomly from the garden of Botany Department, A.N College, Patna and were washed under running tap water and distilled water, air dried, homogenized to fine powder, and stored in tightened light-protected containers for experimentation.

Preparation of the Extracts:-

Ethanolic and aqueous extracts of leaf, stem and root were prepared from coarsely powdered material using soxhlet apparatus. After 24 hrs., it was filtered through 4 layers of muslin cloth and centrifuged at 5000 rpm for 10 min. The supernatant was collected and the solvent was evaporated. The crude extract diluted with 5% of DMSO (dimethyl sulfoxide) to make the final volume one-tenth of the original volume and stored at 4°C in air tight bottles for further studies.

Test Microorganisms:-

The bacterial strains studied are *Staphylococcusaureus, Clostridium bovis, Salmonella typhi, Klebsiellapneumoniae* and *Escherichia coli*. These test organisms were collected from the Department of Microbiology, PMCH, Patna. Cultures of microorganisms for experimentation were maintained at 4°C on nutrient agar slants (Beef extract- 3.0g; peptone-5.0g; agar-15.0g). Each of the microorganisms was freshly cultured prior to susceptibility testing by transferring them into a separate sterile test tube containing nutrient broth and incubated overnight at 37°C. A bacterial loop was used to remove each colony separately from pure culture and transfer it into nutrient broth (distilled water-1000ml).

Preparation of media:-

The growth media prepared for experimentation were nutrient agar and nutrient broth as per following composition. Nutrient broth lacks agar. The medium was adjusted to pH 7.4 and sterilized by autoclaving at 15 lbs pressure and 121°C temperature for 15 min.

Preparation of Inoculum:-

Stock cultures were of gram positive and gram negative bacteria maintained at 4°C on slants of nutrient agar. Active cultures for experiments were prepared by transferring a loopful bacterial cells from the stock cultures to Erlenmeyer flask of nutrient broth followed by incubation with agitation at 37°C for 24 hrs. The bacterial cultures of gram positive and gram negative bacteria were maintained on nutrient agar medium. These microorganisms were allowed to grow at 35°C - 37°C temperature. A fresh inoculum of test microorganism in saline solution was cultured on freshly grown agar slant before every antibacterial assay by adjusting the concentration of microorganism in the medium using spectronic-20 colorimeter (Bausch and Lomb) set at 630 nm. Each organism was recovered for testing by sub culturing on fresh media. A loopful inoculum of each bacterium was suspended in 5ml of nutrient broth and incubated overnight at 37 °C. These overnight cultures were used for the experiment.

Estimation of Antibacterial Efficacy:-

Antibacterial efficacy of the different extracts was determined by cup diffusion method on nutrient agar medium **(Nair *et al.*, 2005)**. For the antibacterial efficacy test, glass Petri-plates of medium size (9 cm) was used. The Petri plates used for pouring were already sterilized and 15 ml of medium was allowed to set, which on cooling gave a layer of 2-3 mm thickness in each plate. Wells were made in nutrient agar plate using cork borer (5 mm diameter) and inoculums containing 10^6 CFU/ml of bacteria were spread on the solid plates with a sterile swab moistened with the bacterial suspension and fifty micro-litres of the working suspension/solution of *O. sanctum* extract (in the conc. 50mg/ml) and same volume of antibiotic chlormphenicol for control was filled in the wells with the help of micropipette. Plates were left for some time to allow the extract to diffuse in the medium incubated at 37°C for 24 h. Culture plates were examined for the evaluation of ZI and the data were recorded nex day (**Hammer *et al.*, 1999 and Evans WC 2002).**

Result and Discussion:-

Antibacterial efficacy revealed that *O. Sanctum* extract inhibited the growth of *S. aureus*, *C. bovis*, and *S. typhi*to higher extant. Highest affectivity was seen in the case of *S.typhi*. *K. Pneumonia* and *E. Coli* showed lower growth inhibition. It was observed that the leaf extract of *O. Sanctum* was more effective to inhibit all the test bacteria in comparison to that of stem and root. By the perusal of table 1 it was concluded that the ethanolic extract of leaf, stem and root shows better result in comparison to aqueous extract against all the test bacteria. The results revealed that the sensitivity of *S. typhi*was at par with a standard positive control "chloramphenicol". The 20 µg concentration of chloramphenicol inhibited the growth of *E.coli*, *S. typhi* and *K. pneumonia* and gave good results against *K. pneumonia* as compared to *E. coli* and *S. typhi.* **Conventry and Allan (2001)** also measured zones of inhibition of *O. sanctum* seed and leaf extract against *B. subtilis.* **Okemo*et al.* (2001)** have also reported that crude extract of *O. sanctum* plant was very effective against *Staphyloccousaureus*. Similar results were also recorded by **Kumar *et al.*, (2009)** on an Ayurvedic formulation i.earmycard powder. **Sharma *et al.*, (2011)** also studied the antibacterial property of *Terminaliachebula Retz* against some human pathogenic strains showing similar results.

Table 1:-Zone of Inhibition for different parts of *O. sanctum* L. against different test bacteria.

Plant Part	**Bacteria**				
	E. coli	***S. typhi***	***S. aureus***	***K. pneumonie***	***C. bovis***
Ethanol ext.	Z.I(mm)	Z.I(mm)	Z.I(mm)	Z.I(mm)	Z.I(mm)
Leaf	4.00	11.0	7.5	4.25	9.00
Stem	4.20	10.90	7.2	3.70	8.75
Root	3.75	10.75	7.5	3.90	8.70
Aqueous ext.	Z.I(mm)	Z.I(mm)	Z.I(mm)	Z.I(mm)	Z.I(mm)
Leaf	3.75	10.75	8.5	3.80	8.75
Stem	3.70	10.60	8.4	3.75	8.60
Root	3.50	10.55	8.2	3.25	8.50
Control	Z.I(mm)	Z.I(mm)	Z.I(mm)	Z.I(mm)	Z.I(mm)
Chloramphenicol	12.00	12.50	10.90	14.00	10.00

Z.I = Zone of Inhibition

The beneficial medicinal effect of plant materials recorded due to the secondary metabolites present in the plant although it is usually not attributed to a single compound but a combination of the metabolites. The medicinal actions of plants are unique to a particular plant species or group, consistent with the concept that the combination of secondary products in a particular plant is taxonomically distinct **(Parekh *et al.*, 2005)**. Plant essential oils and extracts have been used for many thousands of years, in food preservation, pharmaceuticals, alternative medicine and natural therapies.

In vitro studies in this work showed that the plant extracts inhibited bacterial growth but their effectiveness varies. The present study supports the traditional use of *O. sanctum* for the treatment of various diseases and infections **(Rastogi RP, Mehrotra BN. 2002)**. Further studies can be carried out for pharmacological evaluation and determination of specific components responsible for its antibacterial properties and its use in medicine.

Conclusion:-

It must be thus concluded that the *O. sanctum* has antibacterial efficacy. It is an important finding in the healthcare. It will improve our weapon quality against the pathogens. It is expected that using natural products as therapeutic agents will probably not elicit resistance in microorganisms. This can explain the rationale for the use of the plant in treating infections in traditional medicine. The plant could be a veritable and cheaper substitute for conventional drugs since the plant is easily obtainable and the extract can easily be made via a simple process of maceration or infusion. It is essential that research should continue to isolate and purify the active components responsible for its antibacterial properties and its use in medicines.

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Refrences:-

1. Akerele, O. (1993). Summary of WHO Guidelines for the Assessment of Herbal Medicines Herbal Gram., 22: 13-28.
2. A.C Amadioha (1998). *Antibacterial and antifungal components of plants*,Journal of Herbs, Spices and Medicinal Plants. 32: 20-26.
3. Chopra, R. N., Nayer, S. L. and Chopra, I. C., *Glossary of Indian Medicinal Plants*, CSIR, New Delhi, 1956. 45:65-68.
4. Conventry, Allan and Okemo*et al.* (2001), *Antimicrobial efficacy of medicinal herbs.* Journal of Ethnopharmacology.
5. Evans W.C. (2002). Trease and Evan's Pharmacognosy. 5th ed., Haarcourt Brace And Company, pp. 336.
6. Hammer K.A., Carson C.F., Riley T.V. (1999). *Antimicrobial efficacy of essential oils and other plant extracts*. J. Appl. Microbiol., 86(6): 985.
7. Kumar B.S.A, Lakshman K., Thirupati M.S., Jayaveera K.N., Naraynswamy, V.B., Sabemulla K., Nandeesh R. (2009). Free radical scavenging and antibacterial activities of Armycard powder (An Ayurvedic formulation). European bulletin of Drug Research.,17:5-9.
8. Manandhar N.P. (2000). *Plants and People of Nepal*. Timber Press, USA. p. 50.
9. Nair R., Kalariya T, Chanda S. (2005). *Antibacterial efficacy of some selected Indian medicinal flora.* Turk. J. Biol., 29: 41-47.
10. Parekh J., Jadeja D., Chanda S. (2005). *Efficacy of Aqueous and Methanol Extracts of Some Medicinal Plants for Potential Antibacterial Efficacy*. Turk. J. Biol., 29: 203-210.
11. Rastogi R.P., Mehrotra B.N. (2002). *Glossary of Indian Medicinal Plants*. National Institute of science communication, New Delhi, India.
12. Sharma A., Meena S. and Barman N (2011). *Efficacy of ethyl acetate and ether extract of Terminaliachebula Retz against some human pathogenic strains*. International journal of pharmatech research. 2011, 3.(2):724-727.