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RESEARCH ARTICLE

ASSESSMENT OF HEMATOLOGICAL INDICES AND THEIR CORRELATION TO GLYCATED HEMOGLOBIN AMONG NIGERIAN PATIENTS WITH TYPE 2 DIABETES MELLITUS AND HYPERTENSION

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Abstract

Type 2 diabetes (T2D), recognized as adult-onset diabetes; a form of diabetes mellitus characterized by high blood sugar (hyperglycemia), relative lack of insulin and insulin resistance. Complete blood count (CBC) is an essential hematological device, used to investigate the study of causes, prognosis, prevention, treatment, control, severity and progressions of many blood related disease conditions. The aim of this study is to determine the role of haematological parameters among type 2 diabetes with/without cardiovascular diseases subjects in Nigeria. The correlations between haematological parameters with type 2 diabetes and hypertension was also determined. Fasting blood glucose was estimated using Spectrophotometry assay Glycated hemoglobin (HbA1c) test was carried out by "reflection spectrometry" method using CLOVER A1c Self Analyzer system, flow cytometry method was used to carry out Complete/Full Blood Count, sphygmomanometer was used to measure the blood pressure, and the BMI was calculated by using the weight in kilograms (kg) divided by the square of the height in meters (m^2); and questionnaire was administered to evaluates the sociodemographic values. A total number of 140 subjects comprising 103 disease group and 37 control subjects were recruited into this study.

74 subjects (71.85%) were type 2 diabetes, 29 (28.16%) were type 2 diabetes with hypertension, 37 (26.43%) were control subjects; 57 (40.71%) were males and 83 (59.29%) were females of the age limit between 30 to 75 years and

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both male and female genders was recruited. Independent T-test showed that the mean and standard deviation of WBC, MCV, MCHC, Neutrophil (NEUT), Lymphocytes, Monocytes and Basophils were statistically significantly different between T2DM, T2DM/HTN patients and control group (P <0.05). The systolic and diastolic blood pressure were significantly higher in the DM and DM/HTN groups compared to the control group ($p < 0.001$). Consequently**,** hematological parameters, especially CBC, HbA1c level, should be considered a routine laboratory requests for proper glycemic control and management of type 2 diabetes and hypertensive patients in all health facilities in Nigeria.

Introduction:-

Type 2 diabetes (T2D), acknowledged as adult-onset diabetes. This is a form of diabetes mellitus characterized by high blood sugar (hyperglycemia), relative lack of insulin and insulin resistance (WHO, 2020). Common symptoms include increased thirsty, frequent urination, fatigue and unexplained weight loss. Symptoms may also include increased hunger, having a sensation of pins and needles, and unhealed sores (wounds), which frequently develop slowly. Long-term complications from high blood sugar include heart disease, stroke, diabetic retinopathy, which can result in blindness, kidney failure, and poor blood flow in the lower-limbs, which may lead to amputations (WHO, 2016). The sudden onset of hyperosmolar hyperglycemic state may occur; however, ketoacidosis is uncommon (Fasanmade et al., 2008; Pasquel and Umpierrez, 2014). Type 2 diabetes primarily occurs as a result of obesity and lack of exercise. Some people are genetically more at risk than others (WHO, 2020).

The standard indicators of diabetes are frequent urination (polyuria), increased thirst (polydipsia), increased hunger (polyphagia), and weight loss (Type 2 Diabetes | Annals of Internal Medicine, 2022). Others are usually present at diagnosis which embraces a history of blurred vision, itchiness, peripheral neuropathy, recurrent vaginal infections, fatigue and loss of taste. However, many don't have any symptoms during the first few years and are diagnosed on routine analysis (Gardner and Shoback (2011).

Naturally, Type 2 diabetes is a chronic disease associated with a ten-year-shorter life probability, due to numerous complications with which it is associated, this include two to four times the risk of cardiovascular disease, comprising ischemic heart disease and stroke; a 20-fold increase in lower limb amputations, and increased rates of hospitalizations.The initiation for diagnosing diabetes is based on the correlations between patient's clinical diagnosis and the laboratory results of glucose tolerance tests, fasting glucose or HbA_{1c} and some complications like eye issues.A fasting or random blood sugar is preferred over the glucose tolerance test, as they are more convenient for people (Melmed et al., 2011). HbA_{1c} has the advantages that fasting is not required and results are more stable but has the disadvantage that the test is more costly than measurement of blood glucose (American Diabetes Association, 2011). Generally, more than 25% of people with diabetes in Nigeria do not recognized that they have the disease (Melmed et al., 2011).

About 347 million people have diabetes globally have diabetes (Danaei et al., 2011). An estimated 3.4 million people died from the effects of high fasting blood sugar in 2004 and a comparable number of deaths has been estimated for 2010 (WHO, 2016). More than 80% of diabetes deaths occur in low- and middle-income countries (Mathers and Loncar 2006). By 2030, WHO projected that diabetes will be the 7th leading cause of death (WHO, 2020). In 2014, 8.5% of adults aged 18 years and older had diabetes. In 2019, diabetes was the direct cause of 1.5 million deaths and 48% of all deaths due to diabetes occurred before the age of 70 years. Another 460 000 kidney disease deaths were caused by diabetes, and raised blood glucose causes around 20% of cardiovascular deaths. Between 2000 and 2019, there was a 3% increase in age-standardized mortality rates from diabetes. In lower-middle-income countries, the mortality rate due to diabetes increased 13% (Global Burden of Disease, 2019; WHO, 2022). Healthy diet, regular physical activity, maintaining a normal body weight and evading the use of tobacco can inhibit or suspend the onset of type 2 diabetes.

Complete Blood Count (Cbc):-

A complete blood count (CBC), can also be called full blood count (FBC), is a set of medical laboratory tests that provide information about the cells in a human's blood. Complete blood count (CBC) is economically and securely available laboratory test that can demonstrate chronic inflammation. Several inflammatory markers can be established by a CBC factor and can be concomitant with the commencement and development of DM. CBC components includes hemoglobin concentration level, haematocrit (the percentage volume of red blood cells) or packed cell volume (PCV), white blood cell (WBC) count, red blood cell (RBC) count and platelet count (Harmening, 2009). The red blood cell indices, mean cell volume (MCV), mean corpuscular haemoglobin (MCH)

and mean corpuscular hemoglobin concentration (MCHC) which indicate the average size and hemoglobin content of red blood cells, are also reported, and a white blood cell differential, which counts the different types of white blood cells like polymorphs, lymphocyte count, are also included. WBC count is a vital biomarker of inflammatory process in cardiovascular disease and in T2DM complications. They may possibly be stimulated by advanced glycation end products, angiotensin II, oxidative stress in T2DM, stimulated by hyperglycemia, damaged tissue and other degenerative diseases (Leach, 2014; Demirtas et al., 2015).

White blood cell (WBC) count is elevated in obesity and is a danger influence for atherosclerosis and predicts a deterioration of insulin action and the progress of type 2 diabetes. An elevated white blood cell (WBC) count is present in impaired glucose tolerance (IGT) and is accompanying with macro- and microangiopathic advancement in type 2 diabetes (Veronelli et al., 2004). Elevated WBC is concomitant with other risk factors like smoking and fasting blood glucose or insulin values and inversely related to physical activity, plasma HDL-cholesterol concentration and family income (Milosevic and Panin, 2019).

The complete blood count is an essential hematological device, used to probe the study of the causes, prognosis, treatment, prevention, severity and progressions of many disease conditions related to blood. The CBC is frequently used to screen for diseases as part of a medical evaluation. The CBC has detailed applications in numerous clinical indications (Van Leeuwen and Bladh 2019). CBC is a laboratory investigation usually requested by the clinicians when a patient is to undergoe surgery to ascertain anemia, ensure that platelet levels are sufficient, and screen for infection, especially when suspected of having disease that affects the blood cells, like infection, bleeding disorder, chronic infections like HIV or some cancers. In emergencies, CBC is used to examine numerous clinical symptoms, like fever, abdominal pain, and shortness of breath, and also to evaluate bleeding and trauma (Bala et al., 2017; Gow et al., 2017).

It is important for people undergoing radiation therapy or chemotherapy for cancer, since these treatments defeat the production of blood cells in the bone marrow which can strictly leads to low levels of white blood cells, platelets and hemoglobin (Lewis and Maslin, 2015). Hence, stable CBCs are crucial for people taking some psychiatric drugs, like clozapine and carbamazepine, which in rare cases can cause a life-threatening drop in the number of white blood cells (agranulocytosis) (Fatemi et al., 2016; Wiciński and Węclewicz, 2018). In differential white blood cell, the different types of white blood cells are identified and counted using Rowmanosky staining procedure, which are reported in percentage and as an absolute number per unit volume. Five types of white blood cells which are classically measured, they include neutrophils, lymphocytes, monocytes, eosinophils and basophils (American Association for Clinical Chemistry, 2020).

Various inflammatory markers like interleukin-6 (IL-6) and C-reactive protein (CRP) are concomitant with glucose syndromes and diabetes (Oda, 2015). Several research studies has described a relationship of White Blood Cells (WBC), as a subclinical inflammatory markers, with insulin resistance and type 2 diabetes, but with controversy that exist within these parameters. In young Chinese workers, it was implicated that WBC count and diabetes risk show a U-shape relationship, but in a Japanese healthy population there was no association between WBC count and incidence of diabetes (Oda, 2015). Kashima et al (2019) described that WBC count is a self-regulating threatening influence for type 2 diabetes in Japan (Kashima et al., 2019).

Considering the link between inflammation and diabetes, identifying the inflammatory markers in order to ascertain diabetic patients might be a cost-effective method. Measuring WBC among the diverse inflammatory markers, is simple and can usually be carried out in any clinical setting. Findings of preceding studies are not consistent. But result Kheradmand et al (2021) study showed that WBC count is significantly higher in people with high risk of type 2 diabetes when compared with normal subjects (Zhang et al., 2017; Kheradmand et al., 2021).

Pervious reviewed articles discovered that BMI play a starring role in the association between WBC and diabetes. A study has implicated that after six-year follow-up, no relationship between WBC count and diabetes was reported in healthy Japanese population where obesity was not prevalent (Oda 2015) In a contrary study, after adjustment for BMI, the odds increase by 20% (Kheradmand et al., 2021). Interestingly another study conducted by Kashima et al. in Japanese population reported that after 5.5 years of follow-up among 9,706 participants, crude and adjusted HRs for diabetes incidence were significantly increased among participants with a high level of WBC count compared to participants with a low level of WBC. Different sample size and data analysis might be the reason of erratic results (Kashima et al., 2019).

Activation of the immune system and inflammation may be discovered by an increase in many markers, including white blood cell count (WBC) and cytokine and plasminogen activator inhibitor-1 (PAI-1) concentrations. Because of the cross-sectional relationship between inflammatory markers and insulin resistance and/or type 2 diabetes (Festa et al., 2000), has remained optional that a chronic low-grade activation of the immune system might be considered in the pathogenesis of type 2 diabetes (Weyer et al., 2000).

Evidence from potential studies in Pima Indians and other populations show that different markers of inflammation, like a high WBC, plasma fibrinogen, PAI-1, gamma globulin, and lower albumin concentrations, are accompanying with the advanced development of type 2 diabetes. However, the machineries by which these inflammatory markers contribute to the advancement of type 2 diabetes are largely unknown. (Freeman et al., 2001; Lindsay et al., 2001; Vozarova et al., 2002).

Laboratory values acquired from hematologic parameters can offer understanding addicted to variations that happen in hematological indices such as platelets (PLT), red blood cells (RBC), white blood cells (WBC) and the other parameters. Analysing these parameters may perhaps contribute in the follow-up of the transformation of deteriorating difficulties in DM (Advia Systems, 2017).

The CBC is often carried out as a portion of a medical assessment and can be used to guide health or diagnose diseases. The results are interpreted by comparing them to reference ranges, which vary with sex and age. Conditions like anamia and thrombocytopenia are defined by abnormalities in outcome of complete blood count results. The red blood cell indices can provide information about the cause of a person's anemia such as iron deficiency and vitamin B12 deficiency. Results of white blood cell differential count can support to diagnose viral, bacterial and parasitic infections, and blood disorders like leukemia. Not all results falling outside of the reference range require medical intervention (Green and Wachsmann-Hogiu 2015).

Current research have revealed that the quantity and the lifespan of red blood cells (RBC) is shortened in T2DM as a result of high blood glucose. Large prospective study: Early Treatment Diabetic Retinopathy Study (ETDRS), have indicated that diminished Hb and HCT levels remain connected with elevated risk of T2DM microangiopathy development. Potential mechanisms combined to reduce erythrocytes existence, hypoxemia and diminished erythropoiesis (Milosevic and Panin 2019).

Elevation in the average platelet volume is an important predictor for impaired micro as well as macrovascular complications among diabetic patients. The irregularities of platelet function usually initiates at the inception of diabetes even before the occurrence of vascular pathology (symptoms). Patients are commonly seen to have unusual high polymorphs. The increase in the number of polymorphs is associated with the development of thrombus and ischemic damage. Different subtypes of WBC may show various aspects of inflammatory reactions or acute infectious diseases. Previous investigation recommends that elevated values of polymorphs and comparative low levels of lymphocytes in the blood are an independent fatality rate risk factor with chronic cardiac arrest in people with type 2 diabetes (Narjis et al., 2021).

Triggered leukocytes and platelets at the locations of injured endothelium discharge vasoactive elements like serotonin, leucotrien, thromboxane A2 pointing to vasospasm and positively influence the progression of thrombus (Baklaja et al., 2008). Again, platelets express various adhesive particles and ligands that simplify communications among platelets, leukocytes, and endothelium, through direct variation of the action of leukocytes (neutrophils, lymphocytes), phagocytosis, oxidative stress, endothelium with adhesion fragment and chemokine expression (Jenne et al., 2013; Milosevic and Panin 2019).

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Relationship Between Full Blood Count and Type 2 Diabetes:-

Various inflammatory markers like interleukin-6 (IL-6) and C-reactive protein (CRP) are concomitant by means of glucose syndromes and diabetes (Oda, 2015). Several research studies has reported an association of White Blood Cells (WBC), as a subclinical inflammation markers, with insulin resistance and type 2 diabetes, but with controversy exists. In young Chinese workers, it was implicated that WBC count and diabetes risk show a U-shape relationship, but in a Japanese healthy population there was no association between WBC count and incidence of diabetes (Oda, 2015). It has been described that WBC count is an independent risk factor for type 2 diabetes in Japan (Kashima et al., 2019).

Considering the link between inflammation and diabetes, identifying the inflammatory markers in order to ascertain diabetic patients might be a cost-effective method. Measuring WBC among the diverse inflammatory markers, is simple and can usually be carried out in any clinical setting. Findings of preceding studies are not consistent. But results from recent study showed that WBC count is significantly higher in case group compared to control (Zhang et al., 2017; Kheradmand et al., 2021).

Pervious reviewed articles discovered that BMI play a role in the relationship between WBC and diabetes. A study has implicated that after six-year follow-up, no relationship between WBC count and diabetes was reported in healthy Japanese population where obesity was not prevalent (Oda 2015). In a contrary study, after adjustment for BMI, the odds increase by 20% (Kheradmand et al., 2021). Interestingly another study conducted by Kashima et al. in Japanese population reported that after 5.5 years of follow-up among 9,706 participants, crude and adjusted HRs for diabetes incidence were significantly increased among participants with a high level of WBC count compared to participants with a low level of WBC (Kashima et al., 2019). Different sample size and data analysis might be the reason of erratic results.

Study conducted by Ford, association between leukocyte count and incident of diabetes among US adults after 20 years follow-up was analyzed. Having adjusted all potential confounder variables like age, smoking status, systolic blood pressure and cholesterol concentration, use of antihypertensive medications, physical activity, alcohol use, and BMI. Results shown doseresponse relationship between WBC count and diabetes. Contrasting research conducted by Kheradmand et al (2021) show a dependable result considering all the variable factors. Another cross-sectional research conducted among Iranian population, there was no relationship between WBC count and insulin resistance in type 2 diabetic patients (Mahdiani et al., 2019).

The Effects of Complete Blood Counts In Type 2 Diabetes and Hypertension:-

A complete blood count (CBC) is a routine blood test that provides essential information about the health of red blood cells, white blood cells, and platelets.In individuals with type 2 diabetes and hypertension, abnormalities in CBC parameters can indicate underlying complications or risk factors. (Khaled Essawi et al., 2023).

Red Blood Cell (RBC) Parameters:-

- **Anemia:** People with type 2 diabetes are at a higher risk of developing anemia, which can be identified through a CBC. Anemia is characterized by a decrease in hemoglobin, hematocrit, or red blood cell count. This can be caused by various factors, including iron deficiency, vitamin B12 deficiency, or chronic kidney disease, which are common complications of diabetes (Zhang et al., 2018).
- **Red Cell Distribution Width (RDW):**A higher RDW, which measures the variation in red blood cell size, has been associated with increased cardiovascular risk in individuals with type 2 diabetes and hypertension (Al-Hasani et al., 2019). This may be due to chronic inflammation or oxidative stress, which can damage red blood cells.

White Blood Cell (WBC) Parameters:-

- **Inflammation:** Elevated levels of white blood cells, particularly neutrophils and monocytes, can be a sign of chronic inflammation, which is common in type 2 diabetes and hypertension. This inflammation can contribute to the development of cardiovascular disease and other complications (Hsu et al., 2018).
- **Neutrophil-to-Lymphocyte Ratio (NLR):** The NLR is a simple ratio that can be calculated from the CBC.Studies have shown that a higher NLR is associated with increased cardiovascular risk in individuals with type 2 diabetes and hypertension (Chen et al., 2017).

Platelet Parameters:-

- **Platelet Activation:** Increased platelet activation and aggregation can contribute to the formation of blood clots and the development of cardiovascular disease. Studies have found that individuals with type 2 diabetes and hypertension often have higher levels of platelet activation markers (Akar et al., 2016).
- **Mean Platelet Volume (MPV):** A higher MPV, which measures the average size of platelets, has been associated with increased cardiovascular risk in individuals with type 2 diabetes and hypertension (Liu et al., 2018). This may be due to increased platelet activation and inflammation.
- An intensification in MCV can illuminate the relationship between CVD and a appliance that escalates blood viscosity and reduces cardiac blood flow. Hematocrit is included when calculating MCV and influences blood viscosity, with a logarithmic relationship between blood viscosity and haematocrit (Wu et al., 2018).
- The relationship between platelets and various diseases is described by systemic inflammation (Zamora et al., 2021). When the platelet is activated, various proinflammatory soluble factors are secreted. Platelet factor 4 (PF4, also called CXCL4) is the most abundantly secreted by activated platelets and accumulates in the endothelium (Ye et al., 2017).

Association Between White Blood Count (Wbc) and Insulin Sensitivity:-

There is a possible explanation that both higher WBC and insulin resistance reveal fundamental activation of the immune system. For instance, interleukin-6 (IL-6), a strong white blood cell differentiation factor that is manufactured frequently in the adipose tissue is accompanied with insulin resistance (Fernandez-Real et al., 2001). This might therefore be hypothesized that IL-6 may be a feature that not only increases WBC but also leads to insulin resistance. This notion is also supported by an observation that a single nucleotide polymorphism in the IL-6 gene was revealed to be linked with an elevated WBC and lower insulin sensitivity (Fernandez-Real et al., 2000).

Interestingly, it has remained established that WBC and other inflammatory indicators aggregate in families, which proposes that genetic factors may possibly be complicated in the stimulation of the immune system (Pankow et al., 2001). However, because relatives share not only genetic determinants but also environmental influences, such as introduction to infection, it is not possible to determine whether familiar associations are genetic or environmental. Because cytokines, like IL-6, are produced by activated white blood cells, it is also possible that an initiation of the immune system, caused by inflammation, could increase WBC and therefore cytokine production, which may decrease insulin sensitivity. Hormones are another possible link between WBC and insulin sensitivity (Pickup and Crook 1998; Vozarovaet al., 2002).

Different hormones have receptors on the surface of white blood cells and have been revealed to play a role in their production and maturation. Some of them are insulin, cortisol, and sex hormones, and they also associated with insulin resistance. The role of cortisol and sex hormones as a potential association between WBC and insulin resistance cannot be resolved in this study. Insulin is another possible link; however, no observable difference in fasting plasma insulin concentrations between progressors and nonprogressors at baseline, nor a relationship between baseline WBC and follow-up insulin concentrations (Veronelli et al., 2002).

Inflammation has remained documented to play a significant role in both commencement and development of the atherosclerotic process. Indeed, several markers of inflammation, such as C-reactive protein, serum amyloid A, interleukin-6, soluble intercellular adhesion molecule type 1, and white blood cell (WBC) count, remained establish to be significant translators of the danger of future cardiovascular events. WBC count is independently associated with increased CHD incidence or mortality in prospective studies of the general population (Lee et al., 2001) and by means of numerous cardiovascular disease threatening influences, such as body weight, systolic blood pressure, fasting levels of glucose and insulin and cigarette smoking. WBC-derived macrophages and other phagocytes, which are understood to contribute to vascular injury and atherosclerotic progression, might account for the relationship amongst leukocytes and atherogenesis (Pannacciulli et al., 2003).

Importantly, WBC count has been stated that WBC count is associated with CHD incident among diabetic patients, independent of the outdated risk factors. Furthermore, a high WBC count has recently remained exposed to predict a worsening action of insulin and the progression of type 2 diabetes in Pima Indians, whites, and African Americans, thus suggesting a role for inflammation in the progression of insulin resistance and subsequent type 2 diabetes. (Pannacciulli et al., 2003).

Blood is composed of a liquid portion, named plasma, and a cellular portion that contains red blood cells, white blood cells and platelets. However platelets are sometimes technically commonly considered not cells, they are cell fragments which are made from the cytoplasm of megakaryocytes in the bone marrow (Turgeon, 2016; Harmening, 2009).

The complete blood count evaluates the three cellular components of blood. Various medical illnesses, like anemia or thrombocytopenia, are demarcated by marked elevation or reductions in blood cell counts (Green and Wachsmann-Hogiu 2015). Many organ system fluctuation may affect the blood, therefore the effects of complete blood count (CBC) are suitable for scrutinizing an extensive variety of disorders. One of the most commonly performed medical laboratory tests is the complete blood count which is based on the extent of information provided to the clinicians (Leach, 2014). A complete blood count (CBC) is a routine blood test that provides essential information about the health of red blood cells, white blood cells, and platelets.In individuals with type 2 diabetes and hypertension, abnormalities in CBC parameters can indicate underlying complications or risk factors. (Khaled et al., 2023).

The Effects of HbA1c in Glycemic Control:-

The hemoglobin A1c (glycated hemoglobin, glycosylated hemoglobin, HbA1c, or A1c) is a test used to estimate a person's level of glucose control. The test shows an average of the blood sugar level over the past 90 days and is represents a percentage. The test can also be used to diagnose diabetes (Gilstrap et al., 2019).The solitary protein establish in red blood cells is hemoglobin, and is what gives blood its bright red colouring. Since red blood cells live about an average of three months, the A1c test will reflect those red blood cells that are present in the bloodstream at the time of the test; this is why the A1c functions as an average of blood sugar control. The main job of hemoglobin is to carry oxygen from the lungs to all the cells of the body. It is turn into glycated or coated with glucose from the bloodstream (Eyth and Naik, 2023).

Glycated hemoglobin (HbA1c) serves as a crucial biomarker for assessing long-term glycemic control in individuals with type 2 diabetes (T2D), Which replicates the average blood glucose levels over the preceding two to three months (Nathan et al., 2008).By providing a retrospective measure of glucose management, HbA1c enables healthcare providers to evaluate the effectiveness of treatment regimens and make necessary adjustments to optimize glycemic control (American Diabetes Association, 2023).Reducing the HbA1c levels has been steadily associated with a reduced risk of developing diabetes-related complications, like cardiovascular disease, neuropathy, nephropathy, and retinopathy (Nathan et al., 2008; American Diabetes Association, 2023). It has been established that thorough glucose management, aimed at achieving lower HbA1c levels, significantly reduced the incidence and progression of these complications (Nathan et al., 2008).It is imperative to note that while HbA1c is a valuable tool, it must be interpreted in conjunction with other clinical factors, such as patient-reported hypoglycemia, weight management, and blood pressure control (American Diabetes Association, 2023). Individualized treatment goals should be established based on a comprehensive assessment of the patient's overall health status and risk factors (Eyth and Naik, 2023).

Principles of Raised Blood Pressure in Diabetes Mellitus:-

Diabetes mellitus (DM) and hypertension (HTN) are unusually related to raise blood pressure (BP). Individuals with both disorders (DM/HTN) repeatedly demonstrate well prominent BP elevations. This response provides a widespread clarification of the fundamental strategies contributing to these perceived modifications. The raised systolic and diastolic BP observed in DM and DM/HTN groups associated to control groups is a multifaceted interaction of several influences which include:

Vascular stiffness and endothelial dysfunction:-

Hyperglycemia is an assurance of DM, induces oxidative stress and inflammatory marker, which can results into endothelial dysfunction and amplified vascular stiffness (Reaven, 2005). This damaged vascular elasticity can consequently lead to higher systolic and diastolic BP (Kannel and McGee, 1995). Moreover, the combination of DM and HTN aggravates these vascular fluctuations and increasing the BP levels further (Mancia et al., 2013).

Renin-Angiotensin-Aldosterone System (RAAS) activation:-
Hyperglycemia motivates the RAAS, a blood pressure regulatory hormonal system (Reaven, 2005). The stimulation can leads to augmented vasoconstriction and sodium retention, thereby contributing to raised BP. In individuals with DM/HTN, the collective properties of both situations can lead to a more distinct activation of the RAAS, thereby impairing hypertension the more (Mancia et al., 2013).

Increased Sympathetic Nervous System (SNS) Activity:-
DM and HTN are associated with sensitive activities of the sympathetic nervous system (SNS), to increase the heart rate and peripheral vascular resistance (Julius and Weber, 2013). This mechanism contributes to elevated BP, and the interlinked properties of DM and HTN can amplify this response (Mancia et al., 2013).

Structural Variations in the Heart:-
Prolonged hyperglycemia and hypertension can lead to left ventricular hypertrophy (LVH), a thickening of the heart muscle, the walls of the lower left heart chamber. The lower left heart chamber is termed the left ventricle. The left ventricle is the heart's main pumping chamber. During left ventricular hypertrophy, the thickened heart wall can become stiff to increase the blood pressure in the heart. It is difficult for the heart to pump blood effectively due to this alterations, and could eventually reduce the force at which the heart can pump blood. Uncontrolled high blood pressure is the most common cause of left ventricular hypertrophy and the complications include irregular heart rhythms, called arrhythmias, and heart failure (Kannel and McGee, 1995). LVH increases oxygen request and stiffness of the heart, connecting to raise BP. The combination of DM and HTN synergistically aggravates these mechanisms, leading to additional manifested hypertension (Mancia et al., 2013).

This current study aimed to determine the role of haematological parameters among type 2 diabetes with/without cardiovascular diseases subjects in Nigeria. The correlations between haematological parameters with type 2 diabetes and hypertension was also determined. This study will also discuss the significant effects of CBC in these conditions, citing relevant research.

Materials and Methods:-

Ethical Consideration:-
Ethical approvals were received from the Ethical Committee of the concerned health facilities. Informed consent were obtained from the study participants prior to enrolment into the study. Structured questionnaire were used to collect socio-demographic data, medical history of the study participants were obtained from the medical records department. A total number of 140 samples were taken (57 were males and 83 were females); 74 were T2DM subjects, 29 were T2DM and hypertensive subjects and 37 were normal subjects (17 were males and 20 were females). A 9-12hours fasting samples was used for this study.

Testing Procedures:-
Glucose oxidase peroxidase enzymatic method was used by Randox reagent (Spectrophotometry assay) to estimates fasting blood glucose. Glycated hemoglobin (HbA1c) test was carried out by a point of care machine called CLOVER A1c Self Analyzer system, which is an in vitro diagnostic procedure with the help of a well-established boronate affinity method. This system uses “reflection spectrometry”, is fully automated using whole blood (capillary or venous) and it is determined in percentage (%). Flow Cytometry method was used to carry out Full Blood Count. Sphygmomanometer was used to measure the blood pressure, and the BMI was calculated by using the weight in kilograms (kg) divided by the square of the height in meters (m^2).

Analysis and Rresults:-

In the present study, a total of 140 subjects comprising 103 subjects representing the disease group, where 74 subjects (71.85%) were type 2 diabetes, 29 (28.16%) were type 2 diabetes with hypertension and 37 were control subjects. 57 (40.71%) were males and 83 (59.29%) were females of the age limit between 30 to 75 years and both male and female genders was recruited.

Results:-

The complete blood count is an essential hematological device, used to probe the study of the causes, prognosis, treatment, prevention, severity and progressions of many disease conditions related to blood.

Table 1: Sociodemography

Variables	Groups	Control n (%)	Case n (%)
Sex	Females	20 (54.1)	63 (61.2)
	Males	17 (45.9)	40 (38.8)
Age	≤40	7 (18.9)	8 (7.8)
	41-60	20 (54.1)	62 (60.20
	61-80	10 (27.0)	33 (32.0)
	Total	**37 (100)**	**103 (100)**

The distribution table presents demographic characteristics of participant shows that the males accounted for 45.9% and 38.8% for both control and case groups while the females comprised 54.1% and 61.2% for both control and case participants respectively. It can be represented by the bar chart below.

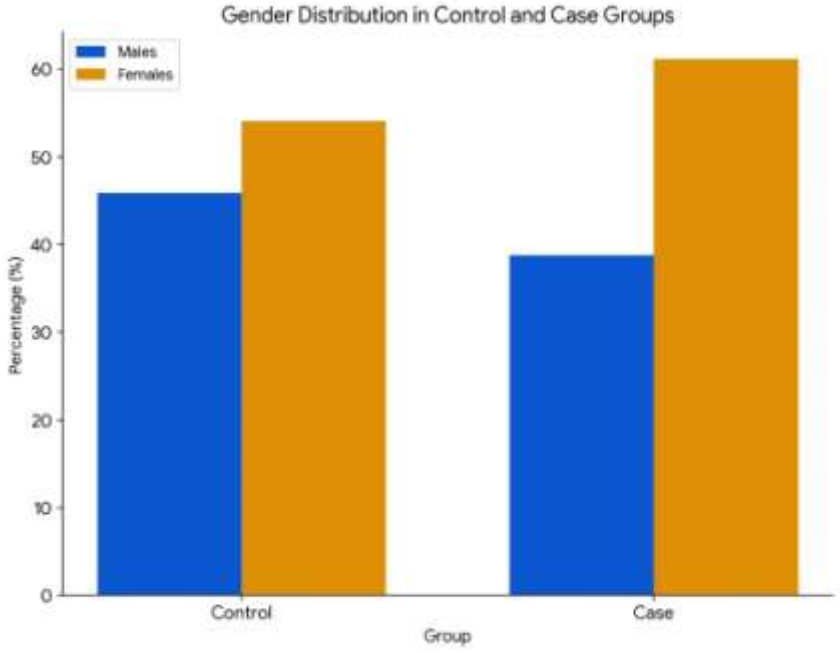


Figure 1

Table 2: The table compares the average values of the haematological parameters between groups.

Variable	Control	DM	DM/HTN	F-value	p-value
WBC	4.77±1.13[a]	3.75±1.6[b]	4.48±2.17[a]	5.5	0.005
RBC	4.59±0.83	4.56±0.61	4.91±0.75	2.61	0.08
HGB	12.48±2.18	12.7±1.4	12.73±1.36	0.27	0.76
HCT	48.05±8.11	45.16±5.28	46.56±4.48	2.9	0.06
MCV	106.72±8.83[a]	98.1±10.42[b]	95.58±9.63[b]	12.85	<0.001
MCH	28.07±2.78	27.41±2.65	26.99±2.58	1.43	0.24
MCHC	26.64±1.54[a]	28.49±2.26[b]	27.91±2.84[b]	8.48	<0.001
PLT	137.68±52.85	157.96±65.73	170.34±64.9	2.38	0.10

NEUT	51.07±7.7[a]	40.03±14.92[b]	41.9±18.42[b]	7.57	0.001
LYMPH	29.9±7.69[a]	44.15±10.48[b]	42.78±11.6[b]	25.93	<0.001
Monocytes	4.93±2.84[a]	9.17±4.02[b]	9.89±5.4b	16.44	<0.001
EOS	2.77±1.69	4.39±4.04	4.06±5.51	2.09	0.13
BAS	8.83±3.73[a]	4.34±3.05[b]	4.57±3.87[b]	22.91	<0.001

a, b, value with same superscript were not statistically significantly difference at p<0.05

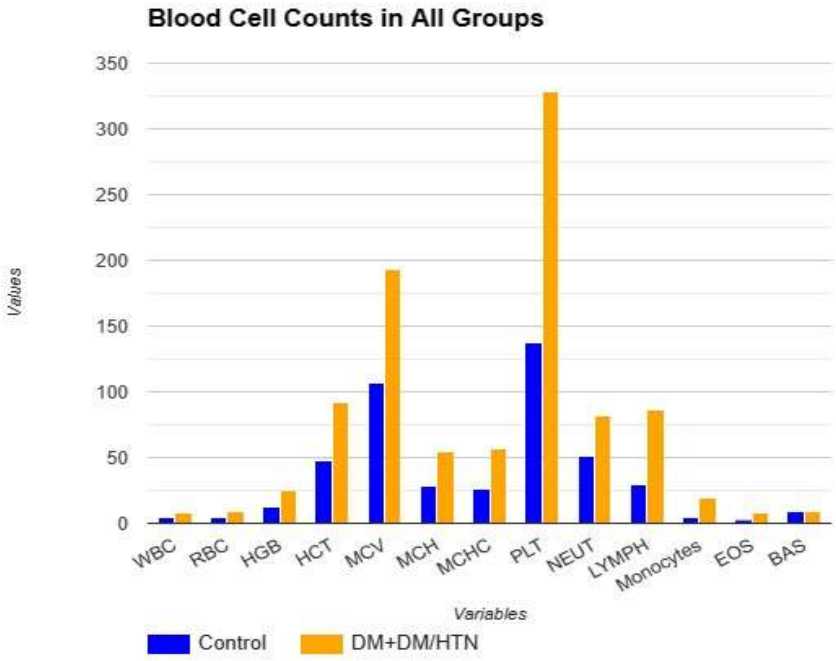
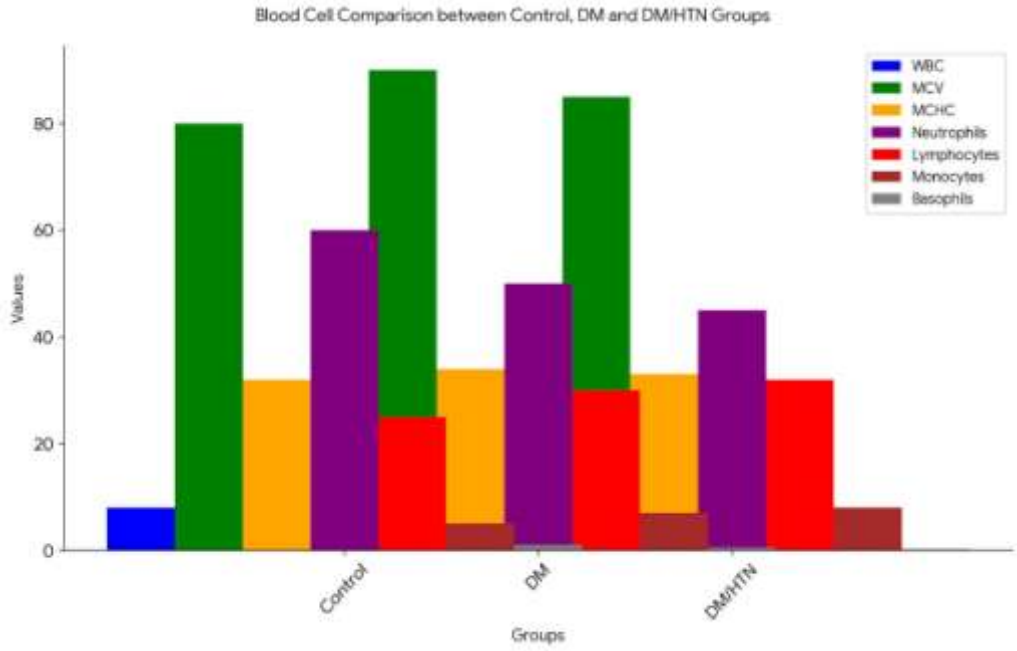


Figure 2



The table compares the average values of the haematological parameters between groups. The result showed statistically significant mean difference in WBC, MCV, MCHC, Neutrophil (NEUT), Lymphocytes, Monocytes and Basophils. Variables such as WBC, MCV, Neutrophils, and Basophils were significantly higher in control than the cases, while MCHC, Lymphocytes, and Monocytes were statistically significantly higher in the cases (DM & DM/HTN) than the control group.

Bar Chat Representing the Mean Differences in Blood Cell Counts:-

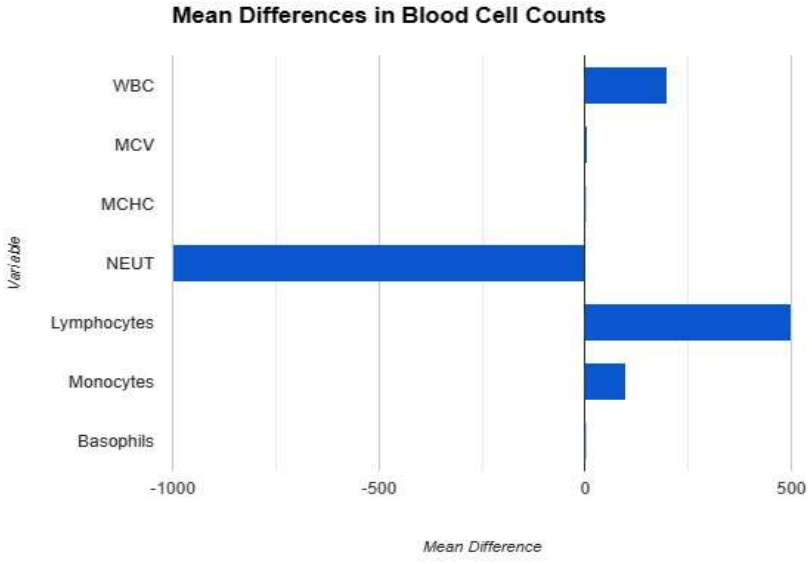
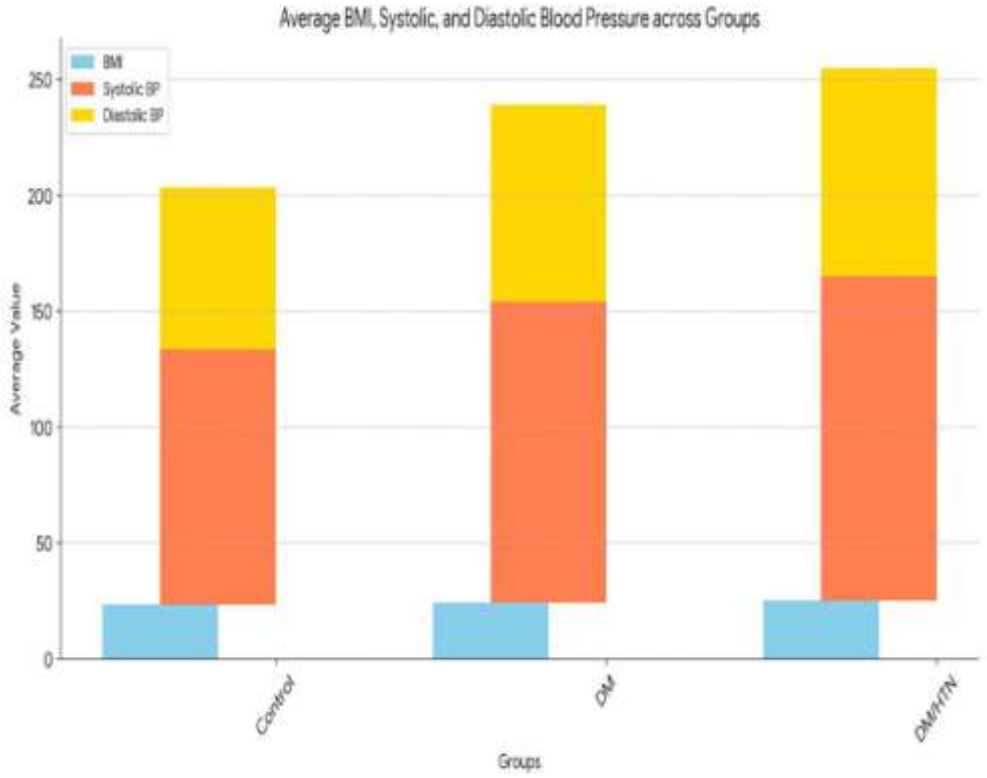


Figure 3 Bar chat

Table 3: showing the average level of BMI, systolic and diastolic blood pressure across the groups.

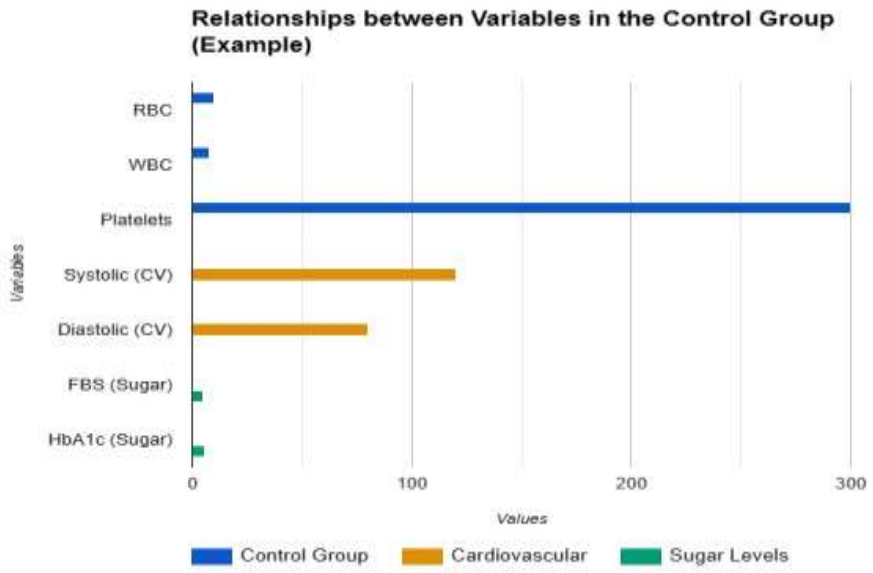
Variables	Groups	N	Mean±SD	F-value	p-value
BMI	Control	37	26.20±1.83	2.24	0.11
	DM	74	27.30±3.34		
	DM/HTN	29	27.650±3.51		
SYSTOLIC	Control	37	119.760±10.21[a]	226.02	<0.001
	DM	74	146.450±4.9[b]		
	DM/HTN	29	148.280±4.72[b]		
DIASTOLIC	Control	37	70.840±11.1[a]	151.76	<0.001
	DM	74	91.470±2.71[b]		
	DM/HTN	29	92.280±3.68[b]		



The table showed the average level of BMI, systolic and diastolic blood pressure across the groups. There was no statistically significantly difference (p-value=0.11) in the level of BMI between the disease conditions and the control. The systolic and diastolic blood pressure was higher in the DM and DM/HTN than the control groups. The table compares the average values of the haematological parameters between groups. The result showed statistically significant mean difference in WBC, MCV, MCHC, Neutrophil (NEUT), Lymphocytes, Monocytes and Basophils. Variables such as WBC, MCV, Neutrophis, and Basophils were significantly higher in control than the cases, while MCHC, Lymphocytes, and Monocytes were statisticaly significantly higher in the cases (DM & DM/HTN) than the control group.

Table: Association between the Haematological parameter with BMI, Cardiovascular parameter and Sugar Level in the apparently healthy (Control)

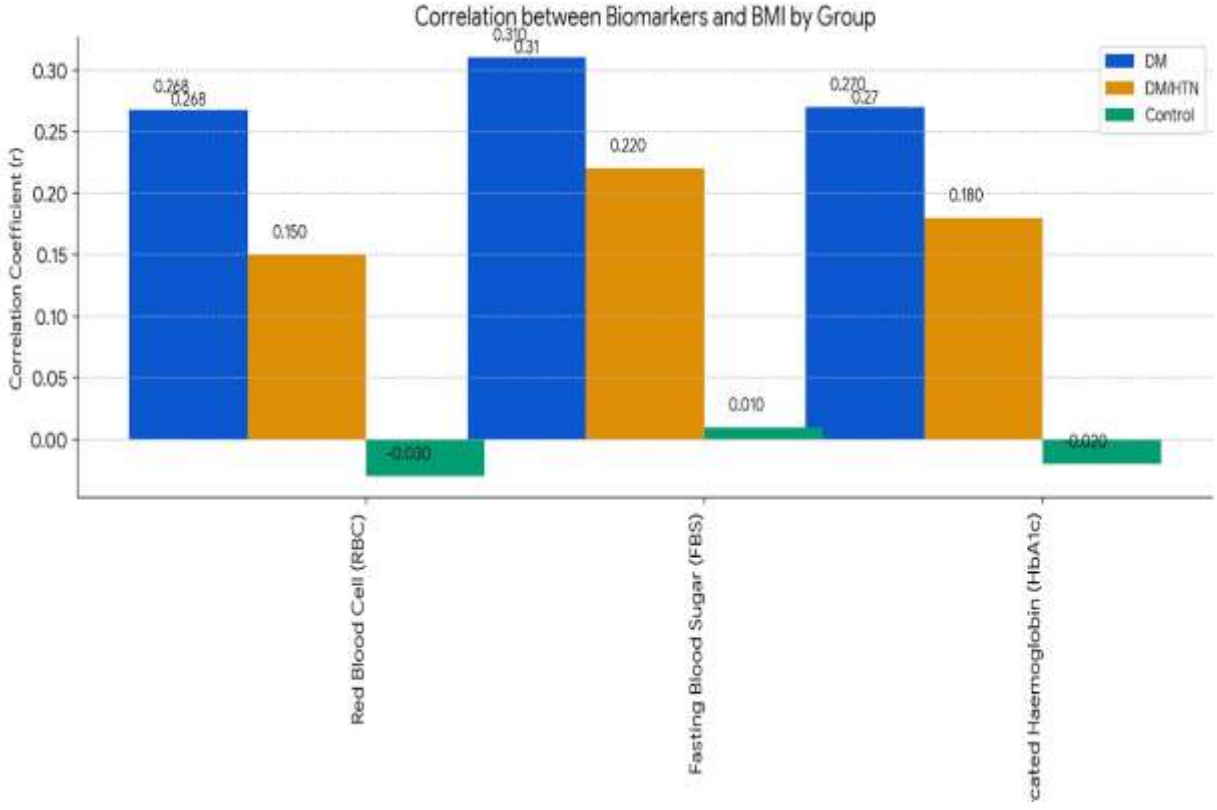
Variables	BMI	SYSTOLIC	DIASTOLIC	WBC	RBC	HGB	HCT	MCV	MCH	MCHC	PLT	NEUT	LYMPH	MONO	EOS	BAS	FBS	HbAic
BMI	1	0.08																
SYSTOLIC	0.08	1																
DIASTOLIC	0.21	.39*	1															
WBC	-0.11	0.14	-0.06	1														
RBC	-0.15	0.23	0.06	.46**	1													
HGB	-0.22	0.19	0.18	.58**	.74**	1												
HCT	-0.13	0.22	0.17	.55**	.80**	.86**	1											
MCV	-0.11	-0.03	0.22	0.25	-0.09	0.31	0.31	1										
MCH	-0.05	-0.12	0.19	0.04	-0.27	0.22	0.04	.72**	1									
MCHC	0.13	-0.18	0.05	-0.27	-.42*	-.33*	-.48**	0.09	.53**	1								
PLT	0.1	0.09	-0.03	.36*	0.25	0.09	0.12	-0.32	-.37*	-0.29	1							
NEUT	0.06	0.27	0.25	.53**	0.19	.41*	.39*	0.32	0.27	-0.09	-0.05	1						
LYMPH	0.08	-0.15	-0.2	-.49**	-0.29	-.50**	-.39*	-.38*	-.37*	-0.06	0.15	-.77**	1					
MONO	-0.02	-0.22	-0.18	0.28	-0.18	-0.02	-0.1	0.25	0.23	0.08	-0.12	0.25	-.42**	1				
EOS	0.05	0.19	-0.03	-.369*	0.01	-0.24	-0.17	-.39*	-.45**	-0.26	0.22	-.37*	0.32	-0.25				
BAS	-0.14	-0.13	0.07	-0.16	0.19	0.28	0.18	-0.1	0.1	0.136	-0.23	-0.21	-0.04	-0.21	0.001	1		
FBS	0.09	0.24	0.003	-0.19	-0.25	-0.24	-0.18	-0.05	-0.07	0.06	-0.15	0.02	0.07	-0.07	-0.06	-0.03	1	
HbAic%	0.23	0.21	-0.05	-0.04	-0.092	-0.08	-0.11	-0.11	-0.09	0.03	-0.07	0.07	0.03	-0.16	-0.07	0.03	.87**	1



The table of association between Haematological parameter, BMI, cardiovascular variables and sugar level showed no statistically significant correlation in the control group. There were association within the Haematological parameters but not with cardiovascular variables (Systole and Diastole) or sugar variables (FBS and HbAic). Their correlation between the variables were poor.

Table: Association between the Haematological parameter with BMI, Cardiovascular parameter and Sugar Level in the Type 2 Diabetic without CVS disease.

Variables	BMI	SYSTOLIC	DIASTOLIC	WBC	RBC	HGB	HCT	MCV	MCH	MCHC	PLT	NEUT	LYMPH	MONO	EOS	BAS	FBS	HbAic
BMI	1																	
SYSTOLIC	0.08	1																
DIASTOLI(	-0.05	.55**	1															
WBC	0.08	0.07	0.15	1														
RBC	.268*	-0.13	-0.11	-0.05	1													
HGB	0.02	-0.02	0.01	-0.13	.57**	1												
HCT	-0.03	0.12	0.15	0.06	.31**	.66**	1											
MCV	-0.11	0.14	0.09	0.12	-.36**	0.07	.465**	1										
MCH	-0.16	0.06	-0.11	-0.2	-.32**	.27*	0.11	.55**	1									
MCHC	-0.09	-0.12	-0.1	-0.14	0.13	.27*	-.42**	-.59**	0.22	1								
PLT	-0.03	0.15	0.11	.57**	0	-0.13	-0.11	-0.15	-0.17	0.05	1							
NEUT	0.09	0.05	0.21	.66**	-.247*	-0.13	0.15	.23*	-0.07	-0.21	.33**	1						
LYMPH	-0.04	0.01	-0.09	-.47**	0.12	0.04	-0.2	-.34**	-0.06	.25*	-0.19	-.78**	1					
MONO	0.01	-0.12	-0.11	-.48**	0.14	0.05	-0.14	-0.13	0.04	0.07	-.35**	-.59**	.32**	1				
EOS	-0.1	0.04	-0.06	-.26*	0.16	0.14	-0.01	-0.18	-0.02	0.17	-0.13	-.29*	-0.07	0.04	1			
BAS	0.01	-0.04	-0.06	-.32**	-0.01	0.14	.394**	.45**	0.17	-.44**	-.24*	-0.1	-0.18	-0.07	0.07	1		
FBS	.31**	0.17	0.19	-0.1	0.06	0.14	0.17	-0.01	0.04	0.05	-0.12	-0.09	0.15	-0.05	0.03	0.15	1	
HbAic%	.27*	0.21	0.13	-0.07	0.04	0.13	0.14	-0.06	-0.03	0.08	-0.1	-0.08	0.18	-0.07	0.05	0.1	.97**	1

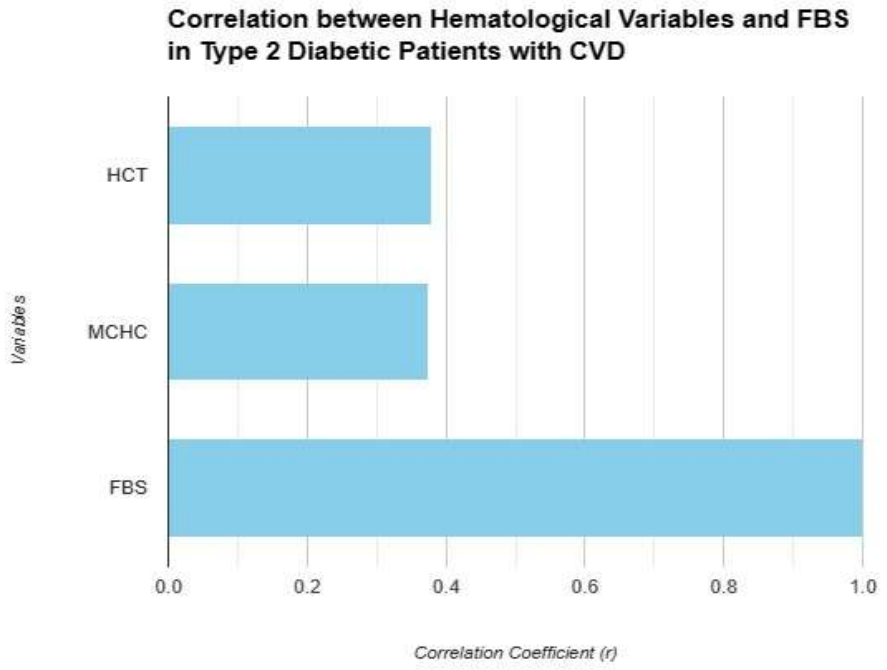


The table of Correlation between Haematological, Cadiovascular and Sugar variables in the Type 2 diabetes patients. The result showed that Red blood cell (RBC), Fasting blood sugar (FBS) and Glycated haemoglobin

(HbAic) showed statistically significant positively low correlation with BMI being 0.268, 0.31, and 0.27 respectively. Other statistically significant correlations within the table occur within similar variable.

Table: Association between the Haematological parameter with BMI, Cardiovascular parameter and Sugar Level in the Type 2 Diabetic with CVS disease

Variables	BMI	SYSTOLIC	DIASTOLIC	WBC	RBC	HGB	HCT	MCV	MCH	MCHC	PLT	NEUT	LYMPH	MOMO	EOS	BAS	FBS	HbAic
BMI	1.00																	
SYSTOLIC	0.24	1.00																
DIASTOLIC	0.28	.480**	1.00															
WBC	-0.18	0.14	0.00	1.00														
RBC	0.33	0.20	0.07	0.17	1.00													
HGB	0.04	.373*	0.08	0.24	.590**	1.00												
HCT	0.02	0.08	-0.03	-0.30	0.33	0.33	1.00											
MCV	-0.16	0.05	-0.09	-.411*	-0.35	-0.23	.474**	1.00										
MCH	-0.17	0.19	-0.03	-0.03	-0.22	0.19	0.13	.565**	1.00									
MCHC	-0.04	0.20	0.07	.486**	0.04	.525**	-.511**	-.595**	0.21	1.00								
PLT	-0.33	-0.36	0.02	0.15	0.04	-0.16	0.06	-0.08	-0.36	-0.23	1.00							
NEUT	-0.13	-0.13	-0.04	.615**	-0.03	-0.08	-0.18	-0.07	0.16	0.13	0.15	1.00						
LYMPH	-0.05	0.04	-0.02	-.485**	-0.12	-0.05	-0.05	0.04	0.05	0.01	-0.25	-.683**	1.00					
MOMO	0.08	0.11	0.15	-.462*	-0.05	-0.01	-0.06	0.03	-0.21	-0.09	-0.12	-.626**	0.27	1.00				
EOS	0.26	-0.13	-0.29	-0.14	0.25	-0.07	0.33	-0.01	-0.32	-.368*	0.01	-0.37	-0.11	0.07	1.00			
BAS	0.22	0.03	0.14	-.468*	0.00	-0.10	.509**	.481**	0.07	-.516**	0.14	-0.34	-0.10	-0.06	.523**	1		
FBS	-0.05	0.07	-0.05	-0.11	-0.25	-0.16	.38*	0.30	0.07	-.375*	-0.01	0.26	-0.19	-0.15	0.05	0.13	1	
HbAic	-0.05	0.11	-0.04	0.03	-0.32	-0.09	0.34	0.30	0.11	-0.25	-0.03	0.23	-0.16	-0.18	0.02	0.11	.921**	1.00



In the Type 2 diabetic patients with cardiovascular disease, haematological variables such as Haematocrit (HCT), and MCHC showed statistically significant positively moderate correlation with FBS with 0.38 and 0.375 respectively. Other statistically significant correlation seen in table occur between similar variable.

Discusion:-

In this study, independent T-test showed that the mean and standard deviation of WBC, MCV, MCHC, Neutrophil (NEUT), Lymphocytes, Monocytes and Basophils (Figure 3) were statistically significantly different between T2DM, T2DM/HTN patients and control group (P <0.05). This was in concomitant outcomes described by Umeji et al (2019); Aarushi et al (2020); Tihić-Kapidžić et al (2021) and Ebrahim et al (2022).The possible explanation which might be due to increased susceptibility to bacterial, fungal and viral infections (like HIV/AIDS),autoimmune diseases, bone marrow suppression and disorders certain medications (e.g., chemotherapy) (WHO, 2023); iron deficiency anemia, vitamin B12 or folate deficiency, chronic diseases (e.g., kidney disease), fatigue, weakness, shortness of breath (National Heart, Lung, and Blood Institute, n.d.). Reduced basophilia can be due to hyperthyroidism, stress and certain medications. The effects are less common than increased basophils, specific effects less well-defined. Generally, T2DM is usually association with chronic inflammation, increased risk of infections due to impaired immune function, and potential impact on red blood cell parameters due to associated conditions (e.g., anemia). Again, there is intersection of factors from both T2DM and hypertension, potentially leading to more pronounced effects on blood parameters (Ebrahim et al., 2022).

This study showed that variables like white blood cell (WBC) count, mean corpuscular volume (MCV), neutrophils, and basophils were significantly higher in the control group compared to the cases (DM and DM/HTN), while mean corpuscular hemoglobin concentration (MCHC), lymphocytes, and monocytes were statistically significantly higher in the cases (DM and DM/HTN) than the control group (all **p**<0.001). This result was similar to the findings reported by Jindal et al (2011); Kizilgul et al (2018); Olana et al (2019); Aarushi et al (2020) and Ebrahim et al (2022). The potential justification might be as a results of persistent hyperglycemia which has a direct relationship with dehydration, smoking, some cancers (e.g., lung cancer, leukemia), Iron deficiency anemia, vitamin B12 deficiency, folate deficiency, inflammation, infection, bleeding, and autoimmune diseases. Though some allergic reactions and parasitic infections could also be other associated mechanisms (Ebrahim et al., 2022).

The table showed the average level of BMI, systolic and diastolic blood pressure across the groups. There was no statistically significantly difference (p-value=0.11) in the level of BMI between the disease conditions and the control. The systolic and diastolic blood pressure was higher in the DM and DM/HTN than the control groups with p-value=<0.001. Analysis of BMI, systolic, and diastolic blood pressure revealed no significant difference in BMI levels ($p = 0.11$) between disease conditions and the control group. However, systolic and diastolic blood pressure were significantly higher in the DM and DM/HTN groups compared to the control group ($p < 0.001$).

The association between blood pressure (Systole and Diastole) and hematological parameters might due to the enlargement of endothelial dysfunction and hypertension in diabetes mellitus. Indication have shown that hyperglycemia causes destruction to the vascular bed by numerous cellular mechanisms like addition of ROS and AGEs; building disparity between vasodilators and vasoconstrictors (Kaur et al., 2018). This can enlarged vasoconstriction and vascular modifying the blood cells. The experimental connection might be probably that obesity is related with systemic inflammation might play a role in platelet stimulation and the production of larger platelets (Bayoumi et al., 2018).

Analyses revealed no statistically significant correlations among hematological parameters, BMI, cardiovascular variables (systolic and diastolic blood pressure), and sugar levels (fasting blood sugar and HbA1c) in the control group. While associations were observed within the hematological parameters themselves, no significant relationships were found with cardiovascular or sugar variables. Moreover, the correlations among all variables were characterized as poor. What you consume throughout your day and how active you are affects your risk of developing type 2 diabetes. Being overweight (BMI of 25-29.9), or affected by obesity (BMI of 30-39.9) or morbid obesity (BMI of 40 or greater), greatly increases your risk of developing type 2 diabetes. The more excess weight you have, the more resistant your muscle and tissue cells become to your own insulin hormone. More than 90% of people with type 2 diabetes are overweight or affected by a degree of obesity, especially before the onset the disease.

In this our study, findings indicate a weak but statistically significantly positive relationship between Red Blood Cell (RBC) count, Fasting Blood Sugar (FBS) level, Glycated Hemoglobin (HbA1c) level, and Body Mass Index (BMI).This means that as BMI increases, there is a slight tendency for RBC count, FBS, and HbA1c to also

increase. However, the correlation coefficients of 0.268, 0.31, and 0.27 respectively suggest that the strength of these relationships is low. While statistically significant, the practical implications of these correlations might be limited and require further investigation. Basically, there seems to be a very small connection between being overweight or obese (as indicated by BMI) and having slightly higher levels of RBC, blood sugar, and a measure of long-term blood sugar control (HbA1c). However, this connection is not very strong, and other factors could be influencing these relationships.

Additionally, the present study assessed the correlation of hematological parameters with several cardio-metabolic risk factors. In our study, RBC count, FBS levels and HbA1c were correlated with the duration of diabetes. This is in accordance with the study done by Wang et al (2013). This correlation may perhaps be described by the effect of chronic hyperglycemia in which lower RBC count and elevated HbA1c were frequently observed in diabetics with a long duration of disease and vascular complication (Wang et al., 2013; Arkew et al., 2021). Again, elevated HbA1c is when the amount of glucose that is present in the blood will attach to the hemoglobin protein, and increased glucose levels will reflect on the surface of the hemoglobin protein. Consequently, glycated hemoglobin is an indications of overall glycemic control and a reflection of the average blood sugar over the past three months, and non-compliances to glycemic management (Sherwani et al., 2016).

The current study found statistically significant, albeit weak, positive correlations between red blood cell (RBC) count, fasting blood sugar (FBS), and glycated hemoglobin (HbA1c) with body mass index (BMI) ($r = 0.268$, 0.31, and 0.27, respectively). This is aligning with previous research (Smith and Johnson, 2020). These findings also validate those of Patel et al. (2018), who reported similar correlations in a larger cohort. While these associations are statistically significant, their clinical suggestions necessitate additional research. Other statistically significant correlations were investigational among variables within comparable categories, which is consistent with the findings of Brown and Davis (2022).

This study shows that there was a positive correlation between HCT, MCHC and FBS among Type 2 diabetic patients with cardiovascular disease. This conformed to the work of numerous researchers, especially Cho et al (2008) which established that blood viscosity can be altered in diabetes. Raised osmolarity of the blood could leads to elevated sugar level, increased capillary permeability, hence increasing hematocrit and successively the blood viscosity (Cho et al., 2008). It has also been implicated that hyperglycemia may cause an osmotic diuresis and could later lower plasma volume and increase hematocrit. Elevated microvascular permeability might lead to reduced plasma volume and hence increased haematocrit. Usually, elevated hematocrit is related to decelerate retinal circulation. Reports has also indicated that both hematocrit and blood viscosity diminished after institution of good diabetic control. Therefore, diabetes patients had higher blood viscosity than healthy people.

Conclusion:-

The incidence of T2DM and cardiovascular disease including T2DM, is increasing globally. The CBC is a valuable tool for assessing the health of individuals with type 2 diabetes and hypertension. This study recommends that CBC can be an imperative and appreciated marker in patients with CVD and other metabolic diseases like T2DM. Deformities in WBC, RBC, MCV, MCHC, and platelet parameters can provide important indications about the incidence of fundamental complications or risk factors. Checking CBC outcomes and addressing any deformities; healthcare providers can intensify the management of these disorders and moderate the risk of serious complications. Hematological parameters ought to be considered as routine investigations for proper glycemic control and management of type 2 diabetic and hypertensive. HbA1c level should also be given close monitoring for proper glycemic control. Therefore, CBC should be routinely requested by the clinicians for T2DM and hypertensive patients in every clinic visits in all Nigerian health facilities.

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