بسم الله الرحمن الرحيم

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Determination of Platelets Count and Indices as Predictive Markers for vascular Occlusion among Hypertensive Sudanese Patients at Military Hospital - Omdurman

تحديد عدد ومؤشرات الصفائح الدموية كعلامة ماقبل الاصابة بانسداد الاوعية الدموية للمرضي السودانيين المصابين بارتفاع ضغط الدم بمستشفى السلاح الطبي بامدرمان

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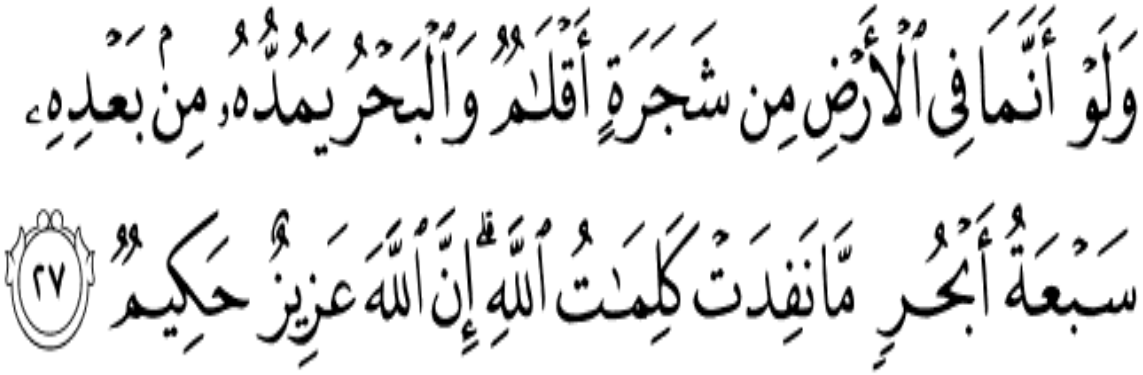
الآيــة


وَلَوْ أَنَّمَا فِي ٱلْأَرْضِ مِن شَجَرَةٍ أَقْلَٰمٌ وَٱلْبَحْرُ يَمُدُّهُۥ مِنۢ بَعْدِهِۦ
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سورة لقمان الايـة27

Dedication

To my lovely parents

Candles, lit me on a way

To my Darling husband

My companion through my life

To my beloved kids

The joy of my life

To my wonderful sisters and brothers

My precious treasure

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Abstract

This was an analytical case control study conducted in Military Hospital Omdurman aimed to measure the changes in platelet count and platelet indices (PDW, MPV, and P-LCR) in Sudanese patients with hypertension.

Platelet counts and indices are potentially useful markers for Platelet activation and in early diagnosis of thromboembolic diseases and play a critical role in the pathophysiology of thrombotic events, myocardial infarction and peripheral vascular disease.

The study was conducted on 70 patients with age range (30-85) of both sexes with hypertension and 70 subjects as control group. Five mls of blood sample was collected in container contain EDTA anticoagulant. Platelet counts, platelet distribution width (PDW), mean platelet volume (MPV) and platelet large cell ratio (P-LCR) was measured by using an automated blood cell counter (Sysmex KX-21N). Data collected was analyzed by using Statistical Package for Social Science (SPSS) computer program version 11.5.

There was no statically significant difference among hypertensive patients in platelet count (294 ±103 vs 269 ±74×10^9/L) *p.v* 0.1, PDW (11.7 ±1.9 vs 11.8±1.6FL) *p.v* 0.6, MPV (9.6 ± 0.9 vs 9.7± 1.2 FL) *p.v* 0.5 and P-LCR (22.3±7.2 vs 22.5±6.4 %) *p.v* 0.8 respectively.

There was statically significant correlation between platelet count, PDW, MPV, P-LCR with *p.v*< 0.01 in cases and controls. Negative correlation was observed between platelet count and PDW, MPV and P-LCR. Compared with Positive correlation was observed between PDW, MPV and P-LCR.

The study concluded that no significant effect of hypertension on platelet count and indices. There was correlation between platelet counts, PDW, MPV, and P-LCR.

الملخص

هذه دراسة حالة تحليلية مقارنة أجريت في مستشفى السلاح الطبيامدرمان هدفت لقياس التغيرات في عدد الصفائح الدموية و مؤشرات الصفائح الدموية (عرض توزيعها، متوسط حجمها ونسبة الصفائح الدموية الخليةالكبيرة) في المرضى السودانيين الذين يعانون من إرتفاع ضغط الدم .

تعداد الصفيحات ومؤشراتها يمكن أن تكون علامات مفيدة لنشاط الصفائح الدمويةوفي التشخيص المبكر للأمراض الانصمام الخثاري وتلعب دورا حاسما في الفيزيولوجيا المرضية من أحداث الجلطات, احتشاء عضلة القلب وأمراض الأوعية الدموية الطرفية.

أجريت الدراسة على 70 مريضا من الفئة العمرية (30-85) من كلا الجنسين مع ارتفاع ضغط الدم و 70 شخص كمجموعة ضابطة. خمسة مل من عينة الدم جمعت في حاوية معقمة تحتوي على مانع التجلط EDTA. عدد الصفائح الدموية ، عرض توزيع الصفائح الدموية ، متوسط حجم الصفائح الدموية و نسبة الصفائح الدموية الخلية كبيرة تم قياسها باستخدام عداد خلايا الدم الآلي . تم تحليل البيانات التي جمعت باستخدام برنامج الحزم الإحصائية للعلوم الاجتماعية النسخة 11.5.

لا يوجد فرقذو دلالة احصائية بين مرضى ارتفاع ضغط الدم و المجموعة الضابطة في عدد الصفائح الدموية($p.v$ 0.1)(294 ±103vs269 ±74×10^9/L)،عرض توزيع الصفائح الدموية (p.v = 0.6) (11.7 ±1.9vs11.8±1.6FL)،متوسط حجم الصفائح الدموية($p.v$ 0.5)(9.6 ± 0.9vs FL9.7± 1.2)، نسبة الصفائح الدموية الخلية كبيرة ($p.v$ 0.8)(22.3±7.2vs22.5±6.4 %)

وجد ارتباط ذو دلالة احصائية يبن عدد الصفائح الدموية وعرض توزيع الصفائح الدموية، متوسط حجم الصفائح الدموية ونسبة الصفائح الدموية الخلية كبيرة($p.v$<0.01) .

وجد ارتباط عكسي بين عدد الصفائح ومؤشراتها مقارنة بارتباط طردي بين عرض توزيع الصفائح الدموية، متوسط حجم الصفائح الدموية و نسبة الصفائح الدموية الخلية كبيرة.

خلصت الدراسة الى أنه لا يوجد تأثير معتبر لمرض ارتفاع ضغط الدم على تعداد و مؤشرات الصفائح الدموية. وجد ارتباط بين عدد الصفائح الدموية و بين عرض توزيع الصفائح الدموية، متوسط حجم الصفائح الدموية و نسبة الصفائح الدموية الخلية كبيرة.

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Abbreviations

ADP: Adenine Diphosphate.

ATP: Adenine Triphosphate.

BP: Blood Pressure.

CAMP: Cyclic Adenosine Monophosphate.

CBC: Complete Blood Count.

cDNA ComplementaryDeoxyribo Nucleic Acid.

CHF: Congestive Heart Failure.

CVD: Cardiovascular Disease.

DBP: Diastolic Blood Pressure.

DIC: Disseminated Intravascular coagulation

DNA: Deoxyribo Nucleic Acid.

EDTA: Ethylene Diamine Tetra Acetic acid.

FL: Femto- Liter.

GP: Glycoprotein**.**

HF: Heart Failure.

HLA: Human Leukocyte Antigen.

HPA: Human Platelet Antigen.

HT: Hypertension.

5-HT: 5-hydroxytryptamine.

HTN: Hypertension.

ITP: Idiopathic thrombocytopenic purpura.

LVH: Left Ventricular Hypertrophy.

MI: Myocardial Infarction.

MPC: Mean Platelet Component.

MPD: Myeloproliferative Disorder.

MPV: Mean Platelet Volume

NAIT: Neonatal Alloimmune Thrombocytopenia.

PCDW: Platelet Component Distribution Width.

PCT: Plateletcrit.

PDGF: Platelet-Derived Growth Factor.

PDW: Platelet Distribution Width.

PF4: Platelet Factor 4.

PFA: Platelet Function Assay.

PI 3 kinase Phosphatidyl Inositol 3-kinase.

PLA2: Phospholipase A2.

P-LCR: Platelet Large Cell Ratio.

PTP Post-Transfusion Purpura.

RNA: Ribo nucleic Acid.

SBP: Systolic Blood Pressure.

SPSS: Statistical Package for Social Science.

TAT: Thrombin-Antithrombin Complex.

TPO: Thrombopoietin.

TSH: Thyroid Stimulating Hormone.

TTP : Thrombotic thrombocytopenic purpura.

TXA2: Thromboxane A2.

VLA-2:	Very Late Antigen-2.
vWF:	von WillebrandFactor.
US :	Untied State.

Chapter One

Introduction and Literature Review

Chapter one

1. Introduction and Literature review

1.1. Introduction:

Platelets are highly complex, a nucleate cells, a fragment of megakaryocyte cytoplasm which circulates in the blood and participates in hemostasis (Bain and Gupta, 2003). They are the derivative of bone marrow megakaryocytes. Routinely a well-prepared peripheral blood film is used for evaluation of platelet number, size, distribution, and structure under the light microscope. One of the drawbacks of microscopic method is possibility of artifact that may lead to misdiagnosis. With availability of automated cell counters more precise information can be gathered. They give almost similar result to that of microscopy. Their use has been largely increase in developed as well as developing countries. (Shah *et al.*, 2013)

Hypertension, also known as high or raised blood pressure, is a condition in which the blood vessels have persistently raised pressure. Blood is carried from the heart to all parts of the body in the vessels. Each time the heart beats, it pumps blood into the vessels. Blood pressure is created by the force of blood pushing against the walls of blood vessels (arteries) as it is pumped by the heart. The higher the pressure the harder the heart has to pump (WHO, 2016).

Globally cardiovascular disease accounts for approximately 17 million deaths a year, nearly one third of the total of these complications of hypertension account for 9.4 million deaths worldwide every year**.** Hypertension is responsible for at least 45% of deaths due to heart disease, and 51% of deaths due to stroke (WHO, 2013).

Hypertension (HT) is a significant risk factor for cardiovascular and cerebrovascular events including heart attack and stroke. The Reno vascular system is commonly affected by elevated blood pressure in patients with Hypertension. Platelet activation plays a critical role in the pathophysiology of thrombotic events and HT-related target organ damage. Platelets retain their original and inactive form while circulating through vessels with intact endothelium. Because of the endothelial damage due to high blood pressure, platelets adhere to collagen released from the subendothelial tissue. Platelets are activated by increases in platelet adhesion and aggregation. Activated platelets exhibit degranulation, swelling, and increases in mass and volume. Platelet indices such as mean platelet volume (MPV), plateletcrit (PCT), and platelet distribution width (PDW) are easily calculated via routine blood counts. MPV reportedly increases in HT and its associated complications as well as in diabetes mellitus, hyperlipidemia, and coronary artery disease. There is an inverse relationship between MPV and cardiovascular outcome (Ates *et al.,* 2015).

PCT was recently accepted as an indicator of platelet activation, and it reportedly increases in cardiovascular diseases. PCT is calculated by using the following formula: PCT=platelet count×MPV/10^7. PDW—another indicator of platelet activation—is thought to be associated with inflammation and atherosclerosis. PDW levels increase in acute coronary syndrome, Alzheimer's disease, and rheumatological diseases (Ates *et al.,* 2015). With improvement in the technologies, advancement occurs in all fields including medicine. Automated cell counters are widely used for diagnosis of different diseases. Hematocrits are important parameters that are used in routine practices. Now a day for analyzing platelet abnormalities, plateletcrits are utilized. Among them Platelet counts, mean platelet volume (MPV) and platelet distribution width (PDW) are important parameters (Shah *et al.*, 2013)

1.2. Literature Review:

1.2.1. Platelets:

Platelets are produced predominantly by the bone marrow megakaryocytes as a result of budding of the cytoplasmic membrane. Megakaryocytes are derived from the haemopoetic stem cell, which is stimulated to differentiate to mature megakaryocytes under the influence of various cytokines, including Thrombopoietin. Once released from the bone marrow young platelets are trapped in the spleen for up to 36 hours before entering the circulation, where they have a primary hemostatic role. Their normal lifespan is 7-10 days and the normal platelet count for all age groups is $150\text{-}450\times10^{9}$/L. The mean platelet diameter is 1-2µm and the normal range for cell volume (MPV) is 8-11 fl. Although platelets are non-nucleated cells, those that have recently been released from the bone marrow contain RNA and are known as reticulated platelets. They normally represent 8-16% of the total count and they indirectly indicate the state of marrow production (Provan *et al*, 2003). The mean DNA content of megakaryocytes is at least eight times that of other somatic cells. One megakaryocyte can produce at least several thousand platelets. The formation and release of platelets is related to a preformed structure in the cytoplasm of megakaryocytes, the so-called "demarcation membrane system". Megakaryocytes are mainly found in the bone marrow but can transit to many organs, including the lung, where part of the platelet release occurs. Under physiological conditions, the serum levels of TPO are low at normal or elevated platelet counts and high in individuals with low platelet counts **(**Munker *et al.*, 2007)

1.2.1.1. Thrombopoietin:

Although the existence of a substance responsible for controlling platelet production (thrombopoietin, TPO) had been postulated for many years, all attempts to isolate it had failed. In 1992, the proto-oncogene c-mpl was cloned, leading to the identification of TPO. In 1994, five groups were able to either purify TPO or clone the cDNA (Gresele *et al.*, 2002).

1.2.1.2. Platelet structure:

Platelets are extremely small and discoid, 3.0 x 0.5 µm in diameter, with a mean volume 7-11 fL. The glycoproteins of the surface coat are particularly important in the platelet reactions of adhesion and aggregation which are the initial events leading to platelet plug formation during hemostasis. Adhesion to collagen is facilitated by glycoprotein Ia (GPIa). Glycoproteins Ib (defective in Bernard-Soulier syndrome) and IIb/IIIa (defective in thrombasthenia) are important in the attachment of platelets to von Willebrand factor (vWF) and hence to vascular sub endothelium where metabolic interactions occur. The binding site for IIb /IIIa is also the receptor for fibrinogen which is important in platelet-platelet aggregation. The plasma membrane invaginates into the platelet interior to form an open membrane (canalicular) system which provides a large reactive surface to which the plasma coagulation proteins may be selectively absorbed. The membrane phospholipids (previously known as platelet factor 3) are of particular importance in the conversion of coagulation factor X to Xa and prothrombin (factor II) to thrombin (factor IIa).The platelet contains three types of storage granules: dense, α and lysosomes. The more frequent specific α granules contain a heparin antagonist (PF4), platelet-derived growth factor (PDGF), β-thromboglobulin, fibrinogen, vWF and other clotting factors. Dense granules are less common and contain adenosine

diphosphate (ADP), adenosine triphosphate (ATP), 5-hydroxytryptamine (5-HT) and calcium. Lysosomes contain hydrolytic enzymes and peroxisomes contain catalase. During the release reaction described below, the contents of the granules are discharged into the open canalicular system. Platelets are also rich in signaling and cytoskeletal proteins which support the rapid switch from quiescent to activation that follows vessel damage (Hoffbrand *et al.,* 2006).

1.2.1.3. Platelet-specific antigens:

Platelets carry ABH, Lewis, li and P antigens, as well as HLA class I and several so-called platelet-specific antigens. The HLA class I antigens are predominantly HLA-A and HLA-B; HLA-C is only weakly expressed on platelets. In some individuals, HLA-A and HLA-B are barely, or not at all, detectable on the platelet surface. HLA antibodies are the single most important cause of immunological refractoriness to random donor platelet transfusions although platelet-specific antibodies occur in 3–9% of cases, usually in association with HLA antibodies, and are also responsible for post-transfusion purpura (PTP) and neonatal alloimmune thrombocytopenia (NAIT) and, very occasionally, febrile transfusion reactions. Over 20 antigens have been described as platelet specific,. These include the antigens of the various HPA (human platelet antigen) systems, which are composed of a high-incidence ‘a’ allele (e.g. *HPA-1a*) and a low-incidence ‘b’ allele (*HPA-lb*). Some of the HPA antigens are not truly platelet specific; the HPA-l antigens are also found on endothelial cells, which might contribute to the severity of NAIT and PTP caused by anti-HPA- 1a. The HPA-5 antigens have been described on activated T cells and probably also on endothelial cells, on the very late antigen-2 (VLA-2) membrane protein interactions (Hoffbrand *et al.,* 2005).

1.2.1.4. Normal hemostasis:

The platelet membrane has integral glycoproteins essential in the initial events of adhesion and aggregation, leading to formation of the platelet plug during hemostasis. Glycoprotein receptors react with aggregating agents such as collagen on the damaged vascular endothelial surface, fibrinogen, and von Willebrand factor to facilitate platelet-platelet and platelet-endothelial cell adhesion. The major glycoproteins are the Ib-IX complex, whose main binding protein is von Willebrand factor, and IIb/IIIa which specifically binds fibrinogen. Storage organelles within the platelet include the “dense” granules which contain nucleotides, calcium and serotonin, and _ granules containing fibrinogen, von Willebrand factor, platelet-derived growth factor and many other clotting factors. Following adhesion, the platelets are stimulated to release the contents of their granules essential for platelet aggregation. The platelets also provide an extensive phospholipid surface for the interaction and activation of clotting factors in the coagulation pathway (Provan *et al.*, 2003).

1.2.1.5. Platelet physiology:

Platelet function

The main function of platelets is the formation of mechanical plugs during the normal hemostatic response to vascular injury. In the absence of platelets spontaneous leakage of blood though small vessels may occur. The immobilization of platelets at the sites of vascular injury requires specific platelet-vessel wall (adhesion) and platelet-platelet (aggregation) interactions. The blood flow conditions determine the specific receptor ligand interactions (Hoffbrand *et al.*, 2006).

Platelet adhesion:

The initiating event following vascular damage is platelet adhesion to exposed sub endothelial matrix proteins. The platelet glycoprotein (GP) receptors which mediate adhesion are dependent on the rate of shear. Under the intermediate to high shear conditions found in arterioles, this event is strictly dependent on von Willebrand factor (vWF) and its receptor, the GPIb–IX–V complex. However, at the lower rates of shear found in the venous circulation and in the static conditions frequently used for experimental purposes, adhesion can occur directly to other sub endothelial matrix proteins such as collagen and fibrinogen, although vWF also supports this event in these vessels. In both cases, adhesion is strengthened considerably through activation of platelet surface integrins, which leads to an increase in affinity for their adhesive ligands. Adhesion applies also to recruitment of circulating platelets into the thrombus. vWF, exposed on the surface of the growing thrombus, also plays a fundamental role in this process, most notably at the high rates of shear that exist within arterioles and in diseased vessels. The platelet-bound vWF that supports these events is derived from plasma and via secretion from platelet α-granules. Adhesion to the growing thrombus is supported by binding of fibrinogen to the integrin αIIbβ3, a process that is more correctly termed aggregation (Hoffbrand *et al.*, 2005).

Platelet activation:

Normal platelet function results from several platelet activities. Platelet adhesion to subendothelium is the initial response to vascular injury, and is mediated by the plasma adhesion protein, von Willebrand's factor (vWF) and its platelet receptor, glycoprotein (GP) Ib. Platelet-platelet interaction (aggregation) is mediated by plasma fibrinogen and its platelet receptor, GP IIb-IIIa. Following platelet

activation, secreted ADP amplifies platelet aggregation to generate the platelet thrombus. All of these platelet functions (adhesion, aggregation, and secretion) can be measured in the laboratory, and deficient platelet GP function can be inferred or quantitated. Screening tests for platelet function such as the bleeding time are not recommended and the utility of other screening tests such as the PFA-100 is unproven (Bennett *et al.,* 2007).

The activation of the platelets causes some morphological alterations: the activated platelets seem larger by becoming spherical in shape and forming pseudopodia. An increased PDW is an indication of the anisocytosis and activation of platelets (Konca *et al*, 2014) .Extracellular nucleotides are released from several sources, including sympathetic nerves, platelets, endothelial and inflammatory cells. Adenosine diphosphate (ADP) is stored in high concentration in platelets and this nucleotide is released in response to platelet activation. It is well known that ADP is a pro-aggregating agent and also activates platelet function during arterial thrombosis. ADP elicits platelet aggregation through the activation of the P2Y1 and P2Y12 receptors expressed by platelets. ADP has been shown to elicit vasorelaxation, as well as to cause vasoconstriction, through the activation of purine receptors in the vascular endothelium and smooth muscle cells, respectively (Giachini *et al.*, 2014)

Platelet aggregation:

Aggregation is used to describe cross-linking of platelets through binding of fibrinogen, or other bivalent or multivalent ligands such as vWF to the integrin αIIbβ3 on adjacent cells. In resting platelets, the integrin αIIbβ3 exists in a low-affinity conformation that is unable to bind to vWF or fibrinogen at the concentrations found within plasma (although it is able to bind to immobilized

forms of these two ligands under static or low shear conditions). Upon platelet activation, so-termed 'inside-out' signals from other receptors cause αIIbβ3 to undergo a conformational change that increases its affinity for fibrinogen, vWF and other RGD (arginine–glycine–aspartate)-containing ligands, including fibronectin and CD40 ligand (CD40L). Binding of fibrinogen and other ligands to αIIbβ3 promotes 'outside-in' signals that rein force platelet activation. The integrin can be activated through elevation of Ca^{2+} and by activation of protein kinase C, rapIb and phosphatidylinositol 3-kinase (PI3 kinase) (Hoffbrand *et al.,* 2005).

.Platelet release reaction and amplification:

Primary activation by various agonists induces intracellular signaling, leading to the release of α and δ- granules. α -Granule contents play an important role in platelet aggregate formation and stabilization and, in addition, the ADP released from dense granules plays a major positive feedback role in promoting platelet activation. TXA2 is the second of the two major platelet positive feedback loops important in secondary amplification of platelet activation to firm a stable platelet aggregate. It is formed *de novo* upon activation of cytosolic phospholipase A2 (PL_{A2}) which is the rate limiting step. This liberates arachidonic acid from the membrane phospholipids, and is metabolized by cyclooxygenase to TXA2. Thromboxane A2 not only potentiates platelet aggregation, but also has powerful vasoconstrictive activity. The release reaction is inhibited by substances that increase the level of platelet cAMP. One such substance is the prostaglandin prostacyclin (PGI2) which is synthesized by vascular endothelial cells. It is a potent inhibitor of platelet aggregation and prevents their deposition on normal vascular endothelium (Hoffbrand *et al.,* 2006).

Clot Formation and Retraction:

The highly localized enhancement of ongoing platelet activation by ADP and TXA2 results in a platelet plug large enough to plug the area of endothelial injury. In this platelet plug the platelets are completely degranulated and adherent to each other. This is followed by clot retraction which is mediated by GPIIb/IIIa receptors which link the cytoplasmic actin filaments to the surface bound fibrin polymers (Hoffbrand *et al.*, 2006).

1.2.1.6. Tests of platelet function

The most valuable investigation is platelet aggregometry which measures the fall in light absorbance in platelet-rich plasma as platelets aggregate. Initial (primary) aggregation is caused by an external agent, the secondary response to aggregating agents released from the platelets themselves. The five external aggregating agents most commonly used are ADP, collagen, ristocetin, arachidonic acid and adrenaline. The pattern of response to each agent helps to make the diagnosis. Flow cytometry is now increasingly used in routine practice to identify platelet glycoprotein defects. In the PFA-l00 test, citrated blood is aspirated through a capillary tube onto a membrane coated with collagen/ADP or collagen/adrenaline. Blood flow is maintained. Platelets begin to adhere and aggregate, primarily via VWF interactions with GPIb and GPIIb/IIIa, resulting in occlusion of the aperture. The PFA-l00 analysis may give false negative results with relatively common platelet defects. Full platelet function tests and vWF screening may be required to exclude abnormal platelet function, even if the PFA-l00 test is normal (Hoffbrand *et al.*, 2006).

1.2.1.7. Platelet disorder:

1.2.1.7.1. Quantitative platelet disorder:

●Thrombocytosis:

Thrombocytosis is defined as a platelet count >450× 10^9/L. May be due to a primary myeloproliferative disorder (MPD) or a secondary reactive feature (Provan *et al.,* 2004).

Familial thrombocytosis:

Familial thrombocytosis is a rare condition with an autosomal dominant inheritance. It can result from a mutation in the promoter of *TPO*, the gene that encodes thrombopoietin, the mutation leading to an aberrantly stable messenger RNA. In another family it resulted from a dominant activating mutation in the *MPL* gene that encodes the thrombopoietin receptor (Bain, 2006).

Essential thrombocythaemia:

Like polycythemiaVera and idiopathic myelofibrosis, essential thrombocythaemia is one of the groups of clonal conditions known as the myeloproliferative disorders. A persisting platelet count 600×10^9/l is the central diagnostic feature, but other causes of a raised platelet count need to be excluded before a diagnosis of essential thrombocythaemia can be made (Provan *et al.,* 2003)

●Thrombocytopenia:

Thrombocytopenia is defined as platelet count <150× 10^9/L. Although there is no precise platelet count at which a patient will or will not bleed, most patients with a count >50 × 10^9/L are asymptomatic. The risk of spontaneous hemorrhage increases significantly <20 × 10^9/L (Provan *et al,* 2004). The platelet count is performed to detect thrombocytopenia, which is defined as a platelet count of less than 150,000/ μL. The test is usually performed as part of an automated blood cell

profile and can be considered as reliable down to a platelet count of 30,000/μL. The finding of an unexpected thrombocytopenia should be confirmed by a review of the peripheral blood smear. The possibility of the existence of red blood cell fragments or of a pseudothrombocytopenia may provide clues or further evaluation of the patient (Munker *et al.*, 2007).

Congenital Thrombocytopenias:

Congenital thrombocytopenia may be inherited or due to a pathological process, e.g. infection or exposure to an antibody or a toxic substance, occurring during intrauterine life. It may be caused by failure of production or increased consumption or destruction of platelets (Bain, 2006).

Autoimmune ('idiopathic') thrombocytopenic purpura (ITP):

Autoimmune or 'idiopathic' thrombocytopenic purpura is an acquired condition in which platelet survival is reduced by the presence of platelet directed autoantibodies (Bain, 2006).

Thrombocytopenia Due to Infections:

Infections are a prominent source of thrombocytopenia, including sepsis from any cause. Two mechanisms are common: DIC or Platelet Adhesion to damaged endothelium or both. Immune-Mediated; Immune destruction may also occur in bacterial and fungal infections, but is most prominent in viral infections (Beck, 2009).

Thrombotic thrombocytopenic purpura (TTP):

TTP is a systemic disorder, usually of unknown cause, in which micro thrombi in multiple organs lead to platelet consumption and renal and cerebral manifestations (Bain, 2006).

1.2.1.7.2. Qualitative Platelet Disorders:

In these disorders, platelet dysfunction is associated with a normal platelet count.

- Bernard-Soulier Syndrome.
- Glanzmann's thrombasthenia.
- Storage pool Disorder.
- Gray plateletSyndrome.
- Scott's syndrome. (Bennett *et al.*, 2007).

Defective platelet functionthis may occur as a complication of viral haemorrhagic fevers, liver cirrhosis, alcoholism, leukemias, paraproteinaemias, uraemia and treatment with certain drugs (e.g. aspirin and non-steroidal anti-inflammatory drugs) (Cheesbrough, 2006)

1.2.1.8. Platelet Indices:

Thromboembolic diseases are among the major cause of mortality in developed countries. Early diagnosis of progressive activation of coagulation can help manage these diseases successfully. A significant list of reliable markers have been investigated recently, concerning activation of coagulation, such as prothrombin fragment 1+2, thrombin-antithrombin complex, and platelet activation, such as β-thromboglobulin (β-TG) or soluble platelet P-selectin**.** However, laboratory measurement of these indices is laborious and expensive. Additionally, the above mentioned indices cannot be included in routine laboratory tests. Several investigators have used a series of platelet indices measured by hematology analyzers, given the fact that platelet activation causes morphologic changes of platelets. The mean platelet volume (MPV) is probably the most extensively studied platelet activation marker **(**Vagdatli *et al*, 2010)

Recently, novel platelet indices such as mean platelet component (MPC) and platelet component distribution width (PCDW) have been investigated as prospective platelet activation markers. However, not all hematology analyzers examine these indices. The present effort for finding simple and widely used platelet activation indices focused on the fact that platelet activation causes morphologic changes of platelets, including both the spherical shape and pseudopodia formation. Platelets with increased number and size of pseudopodia differ in size, possibly affecting platelet distribution width (PDW). The possibility whether platelet activation increases MPV and PDW as expected was examined. Moreover the issue whether pseudopodia formation could cause specific changes, supporting the differential diagnosis between platelet activation and other causes of platelet swelling was assessed **(**Vagdatli *et al.*, 2010)

1.2.1.8.1. Normal value of platelet indices:

The normal range of platelet indices shows in(Giovanetti *et al.*, 2011) as PDW in male (9.4-18.1) and female (9.8 - 18.0), MPV in male (8.5 - 12.4) and female (8.1 - 12.4) and P-LCR (14.3 – 44) in male and (10.7 - 45.0) in female.

1.2.1.9. Platelets and Hypertension:

Platelets are the smallest of the blood cells, yet they are one of the main players during the process of thrombus formation (thrombogenesis). Furthermore, the traditional belief that the endothelium exists simply to provide an inert interface between the blood and the vessel wall is no longer accurate.Indeed, the endothelium produces a large number of substances that affect blood flow and in turn are affected by changes in the blood and the pressure of blood flow (Lip, 2003). Despite many therapeutic advances that have lead to increasingly effective

antihypertensive drug treatments, the precise pathophysiological mechanisms of hypertension and its complications are still poorly understood. In hypertension, the delicate balance between the vasodilators and the vasoconstrictors is upset, leading to changes that then take place in the vascular beds, setting up a vicious cycle that further maintains the high blood pressure. There is also increasing evidence that platelets and the endothelium, which both get activated in hypertension, have a crucial role in the increased thrombotic tendency seen in hypertension. Indeed, despite exposure of the blood vessels to high pressures, the main complications of hypertension (that is, myocardial infarction and stroke) are paradoxically thrombotic in nature rather than hemorrhagic—"the thrombotic paradox of hypertension" or "Birmingham paradox." Certainly, increasing clinical and laboratory evidence suggests that hypertension per se may confer a prothrombotic or hypercoagulable state, with abnormalities of coagulation, platelets, and the endothelium—in fulfillment of Virchow's triad for thrombogenesis (Lip, 2000).

The processes of thrombogenesis and atherogenesis are also intimately related.Many components of the coagulation and fibrinolytic pathways are primary and secondary predictors of cardiovascular events (Ridker, 1994). The close association of these markers with cardiac outcomes and common cardiovascular risk factors raises the distinct possibility that such indices are not merely markers or consequences of thrombosis, but may significantly contribute to the pathogenesis of arterial thrombotic disease (Lip, 2003). It is reported that high grade inflammatory diseases present with low levels of MPV, but low-grade inflammatory conditions present with high levels of MPV (Konca *et al.*, 2014).

1.2.2. Hypertension:

Hypertension, also known as high or raised blood pressure, is a condition in which the blood vessels have persistently raised pressure. Blood is carried from the heart to all parts of the body in the vessels. Each time the heart beats, it pumps blood into the vessels. Blood pressure is created by the force of blood pushing against the walls of blood vessels (arteries) as it is pumped by the heart. The higher the pressure the harder the heart has to pump (WHO, 2016). Normal adult blood pressure is defined as a blood pressure of 120 mm Hg when the heart beats (systolic) and a blood pressure of 80 mm Hg when the heart relaxes (diastolic). When systolic blood pressure is equal to or above 140 mm Hg and/or a diastolic blood pressure equal to or above 90 mm Hg the blood pressure is considered to be raised or high. Most people with hypertension have no symptoms at all; this is why it is known as the "silent killer". Sometimes hypertension causes symptoms such as headache, shortness of breath, dizziness, chest pain, palpitations of the heart and nose bleeds, but not always (WHO, 2015).

1.2.2.1. Diagnosis of high blood pressure:

Signs and symptoms:

There are usually no symptoms or signs of hypertension, and thus it is called the "silent killer". Since humans are completely unaware of excessive blood pressure, it is only through measurements that it becomes detected. The exception is malignant hypertension, which can cause headache, congestive heart failure, stroke, seizure, papilledema, renal failure and anuria (Baker, 2005).

Table (1.1) show hypertension stages: (Baker, 2005).

	Systolic (mmHg)		Diastolic (mmHg)
Normal	<120	AND	<80
Pre-hypertension	120-139	OR	80-89
Stage I (moderate)	140-159	OR	90-99
Stage II (sever)	>160	OR	>100

1.2.2.2 Types of hypertension:

There are two types:

1. **Primary or essential hypertension** (97-98%) has no clear underlying and without an identifiable cause but appears to be the result of an interplay of complex genetic and environmental factors (Baker, 2005).

2. **Secondary hypertension** about 10% of hypertension is secondary to some identifiable cause such as steroids, renal vascular disease, renal parenchymal disease, pregnancy related, pheochromocytoma, Cushing's syndrome and coarctation of the aorta or primary hyperaldosteronism to name a few (Baker, 2005).

1.2.2.3. Mechanisms in primary hypertension

Several pathophysiological mechanisms contribute to the development of primary hypertension. The factors include: genetics, high salt intake, low physical activity, Obesity, insulin resistance, Renin – angiotensin system, Sympathetic nervous system, intrauterine nutrition and low birth weight (Kenning *et al.*, 2014).

1.2.2.4 Causes of secondary hypertension:

Secondary causes account for only a small percentage of all cases of hypertension, but their detection can be important to determine appropriate intervention. Some lifestyle risk factors like obesity and excessive alcohol use may also contribute to hypertension and treatment resistance but are not usually classified as secondary causes. Features suggestive of secondary hypertension include abrupt onset of symptomatic hypertension, hypertensive crisis and sudden loss of blood pressure control after previous stability on drug therapy and drug resistance hypertension (Kenning *et al.*, 2014).

These are unusual but are important because the cause may be curable:

- Endocrine causes: Cushing syndrome, Conn syndrome, Pheochromocytoma, Hyper / Hypothyroidism, Acromegaly, Hyperparathyroidism and Exogenous hormones, e.g. contraceptive pills, glucocorticoids (Ilyas, 2009).

-Renal causes: Glomerulonephritis, Diabetic nephropathy, polycystic kidney disease, and renal artery stenosis (Ilyas, 2009).

-Other causes: Coarctation of the aorta, Pregnancy associated hypertension, Alcohol and Acute stress (Ilyas, 2009).

1.2.2.5. Complication:

Consequences of long-standing hypertension causes accelerated atherosclerosis, which in turns leads to all of the biological fallout of this disease. Some consequences include: stroke, coronary artery disease, myocardial infarction, aneurysmal and occlusive aortic disease. Long-standing hypertension also causes the heart to remodel and undergo a process of hypertrophy (left ventricular hypertrophy or LVH). Hypertrophy can lead to diastolic dysfunction, which can

lead to congestive heart failure (CHF) since the heart is too stiff to relax properly. The stiffened heart requires elevated filling pressures, and this can worsen the dysfunction. Long-standing hypertension can also cause the heart to dilate and lose its ability to pump during systole (systolic congestive heart failure). Lastly, the kidneys are injured by long-standing hypertension and this is a significant cause of renal failure in the U.S (Kenning *et al.*, 2014).

Elevated systemic blood pressure results in high intravascular pressure, ischemic strokes and peripheral vascular disease, are related to thrombosis rather than hemorrhage .Therefore it is important to investigate whether antithrombotic therapy also prevents vascular dysfunction, which is a hallmark of hypertension. (Giachini *et al.*, 2014).

1.2.2.6. Initial laboratory studies:

Initial lab screen should include 12-lead electrocardiogram, urinalysis, fasting blood glucose or HbA1c, serum sodium, potassium, creatinine (with estimated or measured glomerular filtration rate), calcium and lipid profile (total cholesterol, high-density lipoprotein cholesterol, low-density lipoprotein cholesterol and triglycerides). Additional laboratory and diagnostic studies may be required in individuals with suspected secondary hypertension and/or evidence of target organ damage. Other tests may be ordered at the discretion of the clinician such as complete blood count, chest x-ray, uric acid or TSH (Chobanian *et al.,* 2004)

1.2.2.7. Treatment:

Benefits of Lowering Blood Pressure:

In clinical trials, antihypertensive therapy has been associated with reductions in stroke incidence, averaging (35–40) %; myocardial infarction (MI), averaging (20–25) %; and HF, averaging >50. It is estimated that in patients with stage I hypertension (SBP 140–159 mmHg and/or DBP 90–99 mmHg) and additional

cardiovascular risk factors, achieving a sustained 12 mmHg reduction in SBP over 10 years will prevent 1 death for every 11 patients treated. In the added presence of CVD or target organ damage, only nine patients would require such BP reduction to prevent one death (Chobanian *et al.*, 2004)

1.2.2.8. Hypertension crisis:

Uncontrolled hypertension can progress to a hypertensive crisis defined as a systolic blood pressure _180 mm Hg or a diastolic blood pressure _120 mm Hg. Hypertensive crisis can be further classified as a hypertensive urgency or hypertensive emergency depending on end-organ involvement including cardiac, renal, and neurologic injury.The prompt recognition of a hypertensive emergency with the appropriate diagnostic tests and triage will lead to the adequate reduction of blood pressure, ameliorating the incidence of fatal outcomes that may include myocardial infarction (MI), stroke, renal failure, coma, and death. There may be signs and symptoms associated with a hypertensive crisis, or its manifestation may be silent. Silent HTN crisis has been found to be especially common in young black men. In general, there is no precise BP rise for the presentation of a hypertensive emergency. Instead, the specific symptoms imply the presence of end-organ damage. These symptoms include chest pain (myocardial ischemia or MI), back pain (aortic dissection), dyspnea (pulmonary edema or congestive Heart failure), neurologic symptoms, seizures, or altered consciousness (hypertensive encephalopathy) (Rodriguez *et al.*, 2010).

Hypertensive crises (76% urgencies, 24% emergencies) represented more than one fourth of all medical urgencies/emergencies. Hypertensive urgencies frequently present with headache (22%), epistaxis (17%), faintness, and psychomotor agitation (10%) and hypertensive emergencies frequently present with chest pain (27%), dyspnea (22%) and neurological deficit (21%). Types of end-organ damage

associated with hypertensive emergencies include cerebral infarction (24%), acute pulmonary edema (23%) and hypertensive encephalopathy (16%), as well as cerebral hemorrhage (4.5%) (Papadopoulos, 2010).

1.3. Rationale:

Platelets are involved in diseases responsible for the majority of disability and death worldwide, including myocardial infarction, stroke, peripheral vascular disease, cancer, and many infections. Platelets are also studied as a model in many areas of neurobiology, pharmacology, biochemistry and molecular biology (Gresele *et al.,* 2002).

Platelet indices are potentially useful markers for the early diagnosis of thromboembolic diseases. An increase in both mean platelet volume (MPV) and platelet distribution width (PDW) due to platelet activation, resulting from platelet swelling and pseudopodia formation was hypothesized **(**Vagdatli *et al.*, 2010).

Globally cardiovascular disease accounts for approximately 17 million deaths a year, nearly one third of the total of these complications of hypertension account for 9.4 million deaths worldwide every year**.** Hypertension is responsible for at least 45% of deaths due to heart disease and 51% of deaths due to stroke (WHO, 2013). Platelets have attracted increasing interest among clinicians and basic scientists over recent years, and are now known to play a part in many physiological and pathological conditions(Gresele *et al.,* 2002).

1.4. Objectives:

1.4.1. General Objective:

To determine of Platelets Count and platelet indices as predictive markers for pre-vascular occlusion among hypertensive Sudanese patients at Military Hospital Omdurman.

1.4.2. Specific Objectives:

- To measure platelets count and platelet indices among hypertensive patients and compare with non-hypertensive patients.
- To correlate between platelet count and PDW, MPV, P-LCR in hypertensive patient and non-hypertensive.
- To determine the effect of duration, gender, age and medication on platelet parameter (count, indices).

Chapter Two

Materials and Methods

Chapter Two

2. Materials and methods

2.1. Study design and duration:

The study was analytical case control and hospital based study. Conducted during a period from January to April 2016.

2.2. Study area:

The Study was conducted in military hospital Omdurman.

2.3. Study population:

The Sample size were70 venous blood, samples were collected from Sudanese hypertensive patients and 70 samples were collected from healthy individuals as normal control.

2.4. Inclusion criteria:

Both sexes those diagnosed with hypertension were included in the study.

2.5. Exclusion criteria:

Any one of study population with diabetes mellitus, renal problems, other cardiovascular disease and other disease that affect study parameter that to check the effect of hypertension alone on platelet count and platelet indices was excluded.

2.6. Data collection:

A structured questionnaire was designed to collect personal and medical information about the study group, including age, sex, duration of disease, medical condition and medication used.

2.7. Sample collection:

Five mls of the Blood were collected from the superficial vein in the anticupital fossa from the study population under sterile condition and collected using the following procedure.

2.8. Method of sample collection:

2.8.1 Requirement:

- EDTA container.
- Syringe.
- Cotton.
- 70% Alcohol.
- A tourniquet.

2.8.2 Procedure:

1. Participant was set up at right position for the collection.

2. The skin was cleaned by 70% alcohol and allowed to dry, to avoid stinging when the skin is penetrated.

3. A tourniquet was applied to the arm, tight sufficiently to distend the vein, but not so tightly to cause discomfort.

4. The needle was inserted, tourniquet removed and 5ml of the blood sample were collected incontainer with EDTA anticoagulant and mix gently.

5. Blood samples were mixed again before analyzed.

2.9. Test performed:

Complete blood count CBC was done using Sysmex Automated Hematology Analyzer KX 21N series SN B 2010.

2.9.1 PDW (PLT Distribution Width):

PDW is the distribution width on 20% frequency level with the peak taken as 100%. The unit applied is fL (femto = 10-15L).

2.9.2. MPV (Mean Platelet Volume)

MPV is calculated by the following formula:

MPV (fL) = (PCT (%) / PLT ($\times 10^3/\mu L$)) × 1000

Where PCT (%) represents the value weighted with PLT frequency and is called platelet-crit or platelet volume ratio.

2.9.3 P-LCR (Large Platelet Ratio):

This is the ratio of large platelets exceeding 12 fL discriminator and is calculated as the ratio of the particle count between the 12-fL fixed discriminator and Upper discriminator (UD) to the particle count between Lower discriminator (LD) and Upper discriminator (UD).

2.10. Principles of instrument (Sysmex):

2.10.1. Detection Principle:

This instrument performs blood cell count by DC detection method.

2.10.2. Direct Current Detection Method:

Blood sample is aspirated, measured to a predetermined volume, diluted at the specified ratio, and then fed into each transducer. The transducer chamber has a

minute hole called the aperture. On both side of the aperture, there are the electrodes between which flows direct current. Blood cells suspended in the diluted sample pass through the aperture, causing direct current resistance to change between the electrodes. As direct current resistance changes, the blood cell size is detected as electric pulses. Blood cell count is calculated by counting the pulses, and a histogram of blood cell sizes is plotted by determining the pulse sizes. Also, analyzing a histogram makes it possible to obtain various analysis data.

2.11. Ethical consideration:

The study was approved by Ethical Committee of the College Medical Laboratory Sciences, Sudan University of Science and Technology written informed consent was taken from each participant.

2.12. Statistical analysis:

The results were presented as mean ±standerd deviation. The Statistical analysis was performed using Statistical Package for Social Science (SPSS11.5). Means separation was performed using student t test to determine the effect of hypertension on platelet count and platelet indices. Data presented in form of tables and graphs P.value was considered statistically significant.

Chapter Three

Results

Chapter Three

3. Results

Seventy consented volunteers were enrolled in this study and according to sex distribution they were (22/70, 31%) male and (48/70, 69%) female in cases while in control group male (41/70, 59%) and (29/70, 41%) female (Table 3.1) with age group distribution (72/140, 51%) for (20-50 years) and (68/140, 49%) for less than 50 years(Table 3.2). Hypertensive patients have no significant different in the mean of platelet count, PDW, MPV and P-LCR when compared with controls with (*p.v* 0.1, *p.v* 0.6, *p.v* 0.5, *p.v* 0.8) respectively (Table 3.3). Both in cases and controls there was negative correlation between platelet count and PDW, MPV, P-LCR with (*p.v* 0.00, *p.v* 0.00, *p.v* 0.00) respectively and positive correlation was observed between PDW and MPV and P-LCR with (*p.v* 0.00, *p.v* 0.00, *p.v* 0.00) respectively. Most of patients with duration group less than 5 years (35/70, 50%), 5-10 years (17/70, 24%) and >10 years (18/70, 26%) (figure.1.3) and the duration of disease have no statistically significant on platelet count and PDW, MPV, P-LCR (*p.v* 0.8, *p.v* 0.2, *p.v* 0.3, *and p.v* 0.3) respectively (Table 3.4). There was no significant difference between patients whom take medication as Aspirin (Table 3.5), Amlodipine Table (3.7), Loscar (Table 3.6), Lostran (Table 3.9), Lisinopril (Table3.8) and Atenolol (Table 3.10) when compared with controls in platelet count and PDW, MPV, P-LCR.

Table (3.1) Show distribution of gender among cases and controls:

Sex	Case	Control
Male	22 (31%)	41(59%)
Female	48(69%)	29(41%)
Total	70(100%)	70(100%)

Table (3.2) Show distribution of age group among study volunteers:

Age groups	Frequency	Percent %
20-50	72	51
>50	68	49
Total	140	100

Table (3.3) Show effect of hypertension on platelet count and Platelet indices (PDW, MPV and P-LCR):

Parameter	Case	Control	*p.v*
Platelet Count	294 ±103	269 ±74	0.1
PDW	11.7 ±1.9	11.8±1.6	0.6
MPV	9.6 ± 0.9	9.7± 1.2	0.5
P-LCR	22.3±7.2	22.5±6.4	0.8

Values are in Mean±SD

Table (3.4) Show effect of duration of disease on platelet count and indices (PDW, MPV and P-LCR):

Variables	<5	5-10	>10	p.value
Platelet Count	304 ±110	297 ±103	284±102	0.8
PDW	11.5 ±2.3	12.1±1.9	11.0±1.6	0.2
MPV	9.6± 1.0	9.8 ± 1.1	9.3±0.9	0.3
P-LCR	22±7.6	24±8.0	20±6.6	0.3

Table (3.5) Show effect of Aspirin on platelet count and indices (PDW, MPV and P-LCR):

Variables	Aspirin intake	controls	*p.v*
Platelet Count	291 ± 107	269± 74	0.2
PDW	11.5 ± 2.1	11.8 ± 1.6	0.4
MPV	9.5 ± .8	9.7 ± 1.2	0.4
P-LCR	21.7 ± 6.7	22.5 ± 6.4	0.5

P.value <0.05 in significant

Table (3.6) Show effect of Loscar on platelet count and indices (PDW, MPV and P-LCR):

Variables	Loscar intake	controls	*p.v*
Platelet Count	308 ± 122	269 ± 74	0.2
PDW	10.7 ±2.1	11.8 ± 1.6	0.1
MPV	9.5 ± 1.3	9.7 ± 1.2	0.7
P-LCR	20.5 ± 10.2	22.5 ± 6.4	0.5

P.value <0.05 in significant

Table (3.7) Show effect of Amlodipine on platelet count and indices (PDW, MPV and P-LCR):

Variables	Amlodipine intake	controls	*p.v*
Platelet Count	292 ± 97	269 ± 97	0.1
PDW	11.7 ±1.6	11.8 ± 74	0.7
MPV	9.7 ± 0.8	9.7 ± 1.2	0.9
P-LCR	22.8 ± 6.5	22.5 ± 6.4	0.8

P.value <0.05 in significant

Table (3.8) Show effect of Lisinopril on platelet count and indices (PDW, MPV and P-LCR):

Variables	Lisinopril intake	controls	*p.v*
Platelet Count	282 ± 89	269 ± 74	0.6
PDW	10.9 ± 1.7	11.8 ± 1.6	0.1
MPV	9.4 ± 1.0	9.7 ± 1.2	0.5
P-LCR	20.8 ± 7.5	22.5 ± 6.4	0.4

P.value <0.05 in significant

Table (3.9) Show effect of Lostran on platelet count and indices (PDW, MPV and P-LCR):

Variables	Lostran intake	controls	*p.v*
Platelet Count	270 ± 60	269 ± 74	0.9
PDW	11.9 ± 2.0	11.8 ± 1.6	0.8
MPV	9.8 ± 0.9	9.7 ± 1.2	0.7
P-LCR	23.4 ± 7.2	22.5 ± 6.4	0.7

P.value <0.05 in significant

Table (3.10) Show effect of Atenolol on platelet count and indices (PDW, MPV and P-LCR):

Variables	Atenolol intake	controls	*p.v*
Platelet Count	271 ± 96	269 ± 74	0.9
PDW	10.7 ± 2.1	11.8 ± 1.6	0.1
MPV	9.1 ±1.3	9.7 ± 1.2	0.2
P-LCR	19.0 ± 9.9	22.5 ± 6.4	0.1

P.value <0.05 in significant

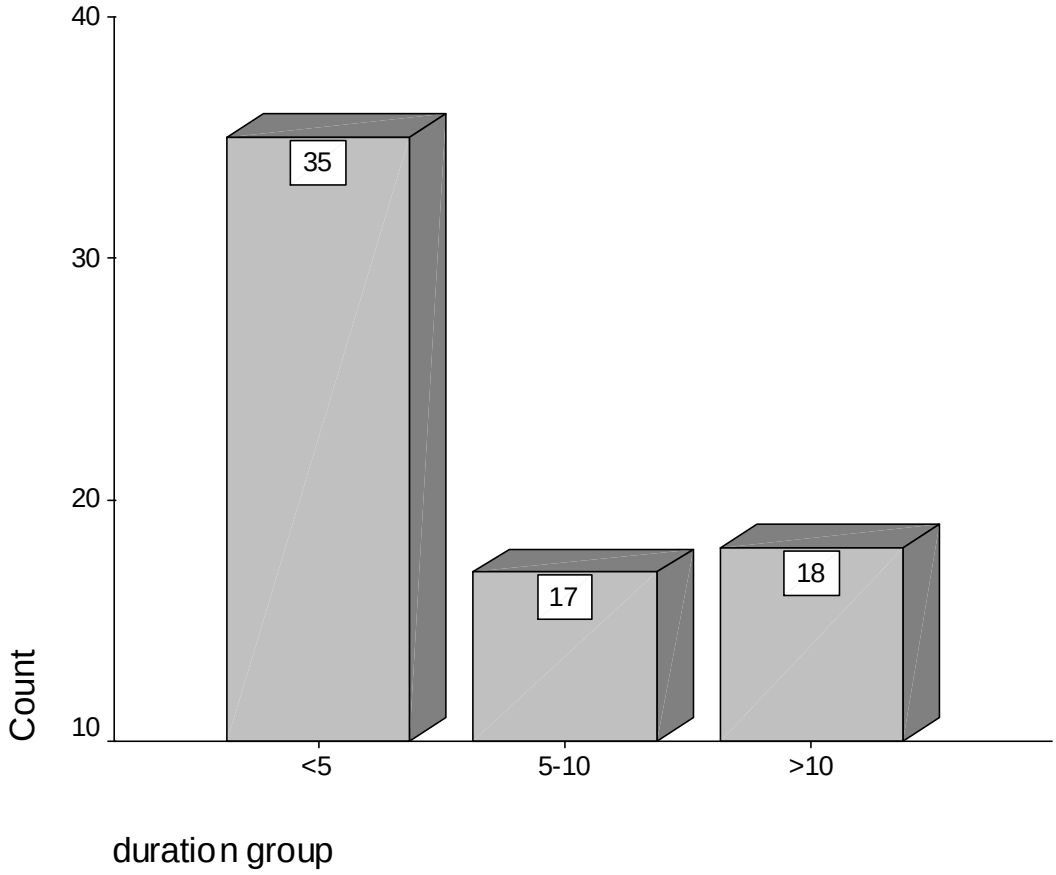

Figure (1.3) shows distribution of duration disease among cases.

Chapter Four

Discussion, Conclusion and Recommendation

Chapter four

4. Discussion, Conclusion, Recommendation

4.1. Discussion:

This is quantitative analytic case control study involve 140 subjects,70 as cases with hypertension and 70 normal individuals as control, for all those platelet count and indices (PDW, MPV, P-LCR) was measured.

Hypertension (HT) is a significant risk factor for cardiovascular and cerebrovascular events including heart attack and stroke (Ates *et al.*, 2015). In hypertension, the delicate balance between the vasodilators and the vasoconstrictors is upset, leading to changes that then take place in the vascular beds, setting up a vicious cycle that further maintains the high blood pressure. There is also increasing evidence that platelets and the endothelium, which both get activated in hypertension, have a crucial role in the increased thrombotic tendency seen in hypertension (Lip, 2002).

In this study statically no significant differences were observed on platelet count and platelet indices; PDW, MPV and P-LCR among hypertensive patients compared to control group (p.v =0.1, p.v=0.6, p.v =0.5, p.v =0.8) respectively.

(Giovanetti *et al*., 2011) in their study they found that there is no significant variations were observed between individuals who have hypertension and who were not or any consequences arising fromthe use ofhypertensive medications. Other study (Sunil, *et al*., 2004) shows treatment of uncomplicated hypertensive using amlodipine -based anti-hypertensive therapy results in reversal of platelet morphology abnormalities and indices of platelet activation.

The mean platelet count, MPV, and PDW were significantly higher in the proteinuria (+) group than in the proteinuria (-) group because proteinuria is

marker of renal problem as complication of hypertension (Target organ damage) (P<0.05) **(**Ates *et al*.,2015).

The present study disagrees with study done by (Yavuzkir M *et al*., 2014)) which observe that MPV increase in hypertensive patients. The reason for the MPV increase in the patients with hypertension could be due to the stimulation of platelet production in the bone marrow by hypertension-induced stress.

In this study there is statically significant correlation between platelet count and PDW, MPV, P-LCR with (*p.v* 0.00, *p.v* 0.00, *p.v* 0.00) in cases and (*p.v* 0.002, *p.v* 0.001, *p.v* 0.001) respectively in controls respectively. platelet count correlate inversely with PDW, MPV and P-LCR and positive correlation was observed between PDW, MPV and P-LCR with *p.v*< 0.01, that agree with (Yan *et al*., 2015) reported that the platelet count correlate inversely with PDW, MPV and P-LCR in healthy individual.

4.2. Conclusion:

In conclusion this study shows no significant changes in platelet count, PDW, MPV and P-LCR in hypertensive patients. Presence of negative correlation between platelet count and PDW, MPV, P-LCR while positive correlation was observed between PDW, MPV and P-LCR.

4.3. Recommendation:

- Platelet indices should be used as routine test for hypertensive patients to evaluate the susceptibility for thrombosis and platelet activation.
- The platelet indices are provide an important simple, practical, , effortless, and cost effective tool, which can be suitable for use in daily practice to predict an impending acute coronary event and other hypertension complications.
- More studies should be under taken with large sample size and special test for platelet activation and coagulation profile for hypertensive patients. Additional randomized controlled studies with long follow-up durations are needed before platelet indices can routinely be used for diagnosis and treatment.

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Appendices

Appendix (1)

جامعة السودان للعلوم والتكنولوجيا

كلية الدراسات العليا

برنامج ماجستير العلوم في المختبرات الطبية علم امراض الدم المناعة الدموية

استبيان لجمع العينات بغرض البحث

DETERMINATION OF PLATELET COUNT AND PLATELET INDICES IN WITH HYPERTENSIVE SUDANES PATIENTS AS MARKER FOR PRE- IVASCULAR OCCLUSION

Questionnaire

Serial number: ———— Name:————————————————————

Age: ———————— Resident :
————————————————————————————

Gender: male ◯ female ◯

Do you have hypertension? Yes ◯ No ◯

Duration: -————————————————

Medication:

Aspirin ◯ Amoldac (amlodipine) ◯ Loscar ◯ Atenolol ◯

Methyldopa ◯ Lisinopril◯ Lostran ◯ Other————————————

Do you have any otherdisease?DM ◯ cardiovascular disorder ◯

Renal problem ◯ other————————————————

Tests:

Platelets count ———— PDW————

MPV ————————P-LCR ————

Date: ——/ ——/—— signature:————————————

Appendix (2)

Informed consent

الموافقة المستنيرة

هذه الدراسة تهدف للتعرف علي مقدار نشاط الصفائح الدموية عند مرضي ارتفاع ضغط الدم من خلال معرفة عددها ومؤشراتها. نشاط الصفائح الدموية يؤدي الي تكوين جلطات داخل الاوعية الدموية و سيتم جمع عينة دم وريدي5 ملليتر ووضعها في حاوية بها مضاد تجلط وملء استبيان يساعد في الدراسة بمعرفة بعض المعلومات عن تاريخ المرض والعلاجات المستخدمة, ومن ثم يتم عمل فحص الدم الكامل للعينة واستخراج عدد ومؤشرات الصفائح الدموية من خلال هدا الاختبار .

أقر أنا _____

بالموافقة علي المشاركة في هذا البحث مع العلم بالمنافع والمضار المترتبة علي المشاركة واعلم بأنني لست ملزم \ة للمشاركة في هذا البحث وأن عدم موافقتي للمشاركة لا يؤثر علي الخدمة الطبية المقدمة لي مع التعهد بسرية المعلومات من قبل الباحث.

التوقيع:_____ رقم الهاتف:_____

Appendix (3)

Sysmex KX21N

Appendix (4)

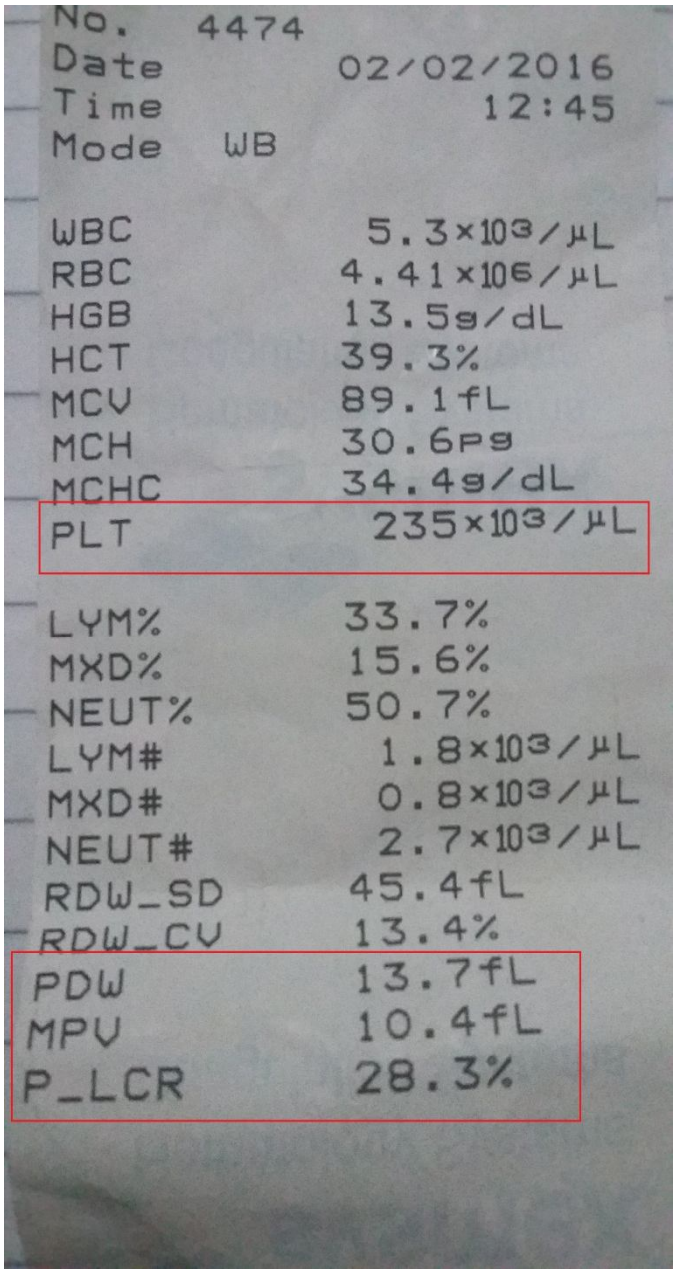

Figure of printed result parameters of study with label