Sudan University of Science and Technology

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Platelets Count and Indices as possible predictors for pre-eclampsia in Sudanese Women in Khartoum State Maternity Hospitals

عد ومؤشرات الصفائح الدموية عواملاً ممكنة للتنبؤ لضغط الدم الجنيني لدي السودانيات بمستشفيات الولادة في ولاية الخرطوم

A dissertation Submitted in Partial Fulfillment of the Requirement for the Degree of M.Sc in Medical Laboratory Science(Hematology and Immunohematology)

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سَنُرِيهِمْ آيَاتِنَا فِي الْآفَاقِ وَفِي أَنفُسِهِمْ حَتَّىٰ يَتَبَيَّنَ لَهُمْ أَنَّهُ الْحَقُّ ۗ أَوَلَمْ يَكْفِ بِرَبِّكَ أَنَّهُ عَلَىٰ كُلِّ شَيْءٍ شَهِيدٌ

صدق الله العظيم

سورة فصلت: الآية 53

Dedication

To the soul of my father.

To my mother for her care, support by all means and understanding

To my kind brothers and beautiful sisters.

To my friends, colleagues and teachers, and everybody who loves me.

Acknowledgements

Firstly, thanks to my God for giving me strength, patience, and courage to conduct this study. My gratitude and deepest thanks to my supervisor Dr. Nazik Elmalaika Obaid Seid Ahmed for her advice, great effort, patience, and encouragement to conduct this study.

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I also wish to thank the doctors and nursing staff of Omdurman Maternity Hospital, Ibrahim Malik Hospital and Academy Charity Teaching Hospital for their co-operation.

Finally, thanks to all the participants who donated the samples, and all the people who helped me in doing this work.

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Abbreviations

ADP	Adenosine diphosphate
ATP	Adenosine triphosphate
CBC	Complete blood count
DIC	Disseminated intravascular coagulation
DNA	Deoxyribonucleic acid
EDTA	Ethylene di-amine tetra- acetic acid
Hb	Haemoglobin
HELLP	Hemolysis, elevated liver enzymes, and low platelet
LD	Lower discriminator
MPV	Mean platelet volume
PCT	Platelet-crit
PDW	Platelet distribution width
P-LCR	Platelet large cell ratio
PLR	Platelet lymphocyte ratio
RBCs	Red blood cells
RPL	Red blood cells to platelets ratio
SPSS	Statistical package for social science soft ware
TXA2	Thromboxane A2
UD	Upper discriminator
vWF	von Willebrand’s factor
WBCs	White blood cells

Abstract

Pre-eclampsia is one of pregnancy complications that is characterized by hypertension and proteinuria after the 20th week of gestation. The exact etiology of pre-eclampsia is unknown, but it is a common complication of pregnancy that is associated with high maternal morbidity and mortality.

This is an analytical case control study carried out among pregnant women with pre-eclampsia (cases) and normal pregnant women (control) at Omdurman Maternity Hospital, Ibrahim Malik Hospital and Academy Charity Teaching Hospital from April to July 2015.

The aim of the study was to determine the level of platelets count and platelets indices among Sudanese pregnant women with pre-eclampsia compared with the level among normotensive pregnant women and to identify whether there is any correlation between severity of pre-eclampsia and platelet count, mean platelet volume (MPV), platelet distribution width (PDW), and platelet large cell ratio (P-LCR). Also To calculate lymphocyte to platelet and red cells to platelet ratios and correlate it with degree of preeclampsia and to find out whether there is any association between gravidity, education, and family history of pre-eclampsia with the development of pre-eclampsia.

In this study 120 pregnant women were informed about the study and agreed for participation,

60 of them worked as cases (pregnant women with pre-eclampsia) and the other 60 worked as controls (disease-free pregnant women), Control pregnant women were matched to cases in age group. A short questionnaire was used to identify age, residence, education, occupation, gravida, parity, gestational age, history of hypertension, history of diabetes mellitus, family history of Pre-eclampsia, other chronic or infectious diseases, blood pressure, and proteinuria.

Blood samples (2.5 ml) were collected from the participants in EDTA anticoagulant container. All samples were tested using hematology analyzer

(Sysmex KX-21N model- Japan). Data were analyzed using Statistical Package for Social Science software (SPSS), and P. value was considered statistically significant at 0.05.

Significant differences were found between the two studied groups (cases and control) with regard to platelet count, mean platelet volume (MPV), platelet distribution width (PDW) and platelets large cell ratio (P-LCR) (P. value 0.00) and between severe and mild cases (P. value 0.02, 0.005,0.008, 0.005), respectively.

The mean level of red blood cells to platelets ratio (RPR) was found to be significantly high among pregnant women with severe pre-eclampsia (P. value 0.002), whereas, the difference between the means levels of platelet to lymphocyte ratio (PLR) of mild and severe type of pre-eclampsia was found to be insignificant (P. value= 0.122).

This study suggested that, platelet count, mean platelet volume (MPV), platelet distribution width (PDW), platelets large cell ratio (P-LCR), and red blood cells to platelets ratio (RPR) can serve as early monitoring markers for the severity of pre-eclampsia (P. value 0.02, 0.005, 0.008, 0.005, 0.002, respectively). It also revealed that primigravida and family history of pre-eclampsia were considered as one of the main risk factors for developing or experiencing pre-eclampsia (OR: 1.9, 17.7, respectively). Furthermore, educational level and family history of pre-eclampsia were found to be associated with the developing pre-eclampsia (P. value= 0.001 and 0.000, respectively).

المستخلص

ضغط الدم الجنيني هو أحد المضاعفات التي تحدث أثناء الحمل والذي يتميز بارتفاع ضغط الدم ووجود فائض بروتين في البول بعد الاسبوع العشرين من الحمل. لا يوجد سبب معروف ومحدد لحدوث المرض ولكنه من مضاعفات الحمل المعروفة والمرتبطة بمعدلات عالية من الاصابات والوفيات.

هذه دراسة تحليلية تم اجراؤها بين النساء الحوامل المصابات بضغط الدم الجنيني (الحالات) والنساء الحوامل الطبيعيات (المجموعة الضابطة) في مستشفي أمدرمان للولادة، مستشفي ابراهيم مالك و مستشفي الأكاديمية الخيري التعليمي في الفترة من ابريل الي يوليو 2015.

الهدف من الدراسة هو قياس عدد ومؤشرات الصفائح الدموية لدي السودانيات المصابات بضغط الدم الجنيني ومقارنتها مع مستواها لدي الحوامل الطبيعيات، وكذلك تحديد ما اذا كانت هناك علاقة بين شدة المرض وعدد الصفائح الدموية، متوسط حجم الصفيحة الدموية (MPV)، عرض توزيع الصفائح الدموية (PDW) و (P-LCR). ايضا حساب نسبة الخلايا الليمفاوية للصفائح الدموية ونسبة خلايا الدم الحمراء للصفائح الدموية وربطها بشدة المرض. ايضا اكتشاف ما اذا كان هناك علاقة بين عدد الولادات، التعليم، ووجود تاريخ عائلي للأصابة بالمرض بحدوث المرض.

أخذت 120 عينة من النساء الحوامل بعد اخطارهن بهذه الدراسة وأخذت موافقتهن علي المشاركة ، 60 من الحالات المصابة بالمرض و 60 امرأة حامل غير مصابات بالمرض(المجموعة الضابطة) وتم مقابلة المجموعتين في الفئة العمرية.

استخدم استبيان مختصر لتحديد العمر، السكن، مستوي التعليم، الوظيفة، عدد مرات الحمل، عدد الولادات، عمر الحمل، تاريخ الاصابة بضغط الدم، تاريخ الاصابة بمرض السكري، وجود تاريخ عائلي للاصابة بضغط الدم الجنيني، أمراض أخري مزمنة أو معدية، مقياس ضغط الدم و وجود فائض بروتين في البول.

تم سحب عينة دم (2.5 مل) في حاوية تحتوي علي مانع تجلط (ثنائي الايثلين رباعي حمض الخل) من كل مشاركة. تم استخدام جهاز يعمل أتوماتيكيا لتحليل الدم الكامل (سسمكس). تم تحليل البينات احصائيا، وأخذت قيمة P للدلالة المعنوية عند 0.05.

وجد أن هناك اختلاف ذو دلالة معنوية بين المجموعتين تحت الدراسة(الحالات المصابة والمجموعة الضابطة) فيما يتعلق بعدد الصفائح الدموية(PC) ، متوسط حجم الصفيحة الدموية (MPV) ، عرض توزيع الصفائح الدموية (PDW) و (P-LCR) بنسبة معنوية (0.00)، وكذلك بين الحالات المعتدلة والحالات الشديدة للمرض بنسبة معنوية (0.02, 0.005, 0.008, 0.005) علي التوالي.

وجد في هذه الدراسة ان هناك اختلاف ذو دلالة معنوية بين متوسط نسبة كريات الدم الحمراء الي الصفائح الدموية (RPR) لدي الحالات الشديدة من المرض بنسبة معنوية (0.002) بينما لم يوجد اختلاف ذو دلالة معنوية بين متوسط نسبة الخلايا الليمفاوية للصفائح الدموية(LPR).

أثبتت هذه الدراسة أن تعداد الصفائح الدموية(PC) ، متوسط حجم الصفيحة الدموية (MPV) ، عرض توزيع الصفائح الدموية (PDW) ،(P-LCR) ونسبة كريات الدم الحمراء الي الصفائح الدموية (RPR) يمكن أن تستخدم كمؤشرات للدلالة علي شدة المرض بنسبة معنوية (0.02, 0.005, 0.016, 0.005،0.002) علي التوالي.

أثبتت ايضا ان الحمل للمرة الأولي ووجود تاريخ عائلي للأصابة بالمرض تعتبر من العوامل الرئيسية المهيئة للاصابة بالمرض لدي الحوامل في المستقبل بنسبة ارجحية (OR) (1.9 و 17.7) علي التوالي. علاوة علي ذلك وجد أن مستوي التعليم ووجود تاريخ عائلي للاصابة بالمرض له علاقة بحدوث المرض بنسبة معنوية (0.001 و 0.000) علي التوالي.

CHAPTER ONE

INTRODUCTION AND LITERATURE REVIEW

Chapter one

1. Introduction and literature review

1.1 Introduction:

Pre-eclampsia is one of pregnancy complications that occur after the twentieth week of gestation (Freitas *et al.*, 2013). The exact etiology of pre-eclampsia is unknown, but a genetic predisposition and association with the presence of thrombophilia (tendency to clot) has been proposed (Sibai, 2010).

Approximately 6% to 8% of all pregnancies are complicated by pre-eclampsia, most of them are less than 20 or greater than 30 years age (McCrae, 2007). It is associated with high maternal morbidity and mortality, and it is a common complication of pregnancy that causes intrauterine fetal growth retardation (Eiland *et al.*, 2012).

The disease affects multiple organ systems and it originates in the placenta and causes variable problems to both mother and fetus (Redman and Sargent, 2005).

HELLP syndrome is a variant of pre-eclampsia in which *h*emolysis, *e*levated *l*iver enzymes, and *l*ow *p*latelets are present. Women with pre-eclampsia who develop grandmal seizures have eclampsia (Fragento, 2006).

Among the clinical signs and symptoms, decrease platelets count is an important character and is associated with HELLP syndrome (Schroeder, 2002), due to generalized endothelial cell activation, caused by enhanced vascular endothelial growth factor release, expression of adhesion molecules, lipid peroxidation, or activation of metalloproteases. This then leads to activation of platelets and coagulation cascade (Rodger *et al.*, 2006).

In Sudan, where pre-eclampsia and eclampsia are among the main causes of obstetric complication, there is an extremely high maternal mortality (Ali and Adam, 2011). This high frequency of maternal morbidities and mortality needs improvement in obstetric care (Ali *et al.*, 2012). Several aspects of pre-

eclampsia in Sudan were investigated (Elhaj *et al*., 2015); however, there were no published research (to the best of our knowledge) on measurement of platelet count and indices in pre-eclamptic patients that carried out to find the role of platelet and platelet indices as predictors of its severity.

The aim of this study is to measure platelets count and indices among pre-eclamptic women, and to determine their value in predicting the severity of pre-eclampsia for early interventions.

1.2 Literature review:

1.2.1 Pre-eclampsia:

Pre-eclampsia is defined as the development of hypertension and proteinuria after the twentieth week of gestation. Specifically, systolic blood pressure of at least 140 mmHg or diastolic blood pressure of 90 mmHg must be present as well as proteinuria of at least 300 mg in a 24-hour period. The presence of nondependent edema is no longer included in the definition of preeclampsia (Frageneto, 2006).

1.2.1.1 Pathophysiology of preeclampsia:

The pathophysiological changes result in vascular lesions in uteroplacental vascular beds, peripheral vessels, and in various organ systems, such as the lungs, liver, brain, and kidneys. These pathophysiological abnormalities include:

1-Endothelial dysfunction

2- Inadequate maternal vascular response to placentation

3- Abnormal angiogenesis

4- Exaggerated inflammatory response with generalized vasospasm, activation of platelets, and abnormal hemostasis (Sibai, 2010).

Although the cause of preeclampsia remains unclear, there is evidence that increase in circulating soluble fms-like tyrosine kinase 1, and decrease placental growth factor- the binding of circulating soluble fms-like tyrosine kinase 1 to

placental growth factor and vascular endothelial growth factor– may have pathologic role in developing pre- eclampsia (Levine *et al*., 2004). Placental antiangiogenic factors are upregulated and disrupted in the maternal endothelium and cause:

1. Hypertension
2. proteinuria
3. glomerular endotheliosis
4. HELLP (hemoysis,elevated liver enzyme level,and low platelet count) syndrome
5. Cerebral edema (Young *et al.*, 2010).

1.2.1.2 Epidemiology of pre-eclampsia:

Systematic review by the World Health Organization as discussed by Jeyabalan (2013) indicates that hypertensive disorders account for 16% of all maternal deaths in developed countries, 9% of maternal deaths in Africa and Asia, and 26% of maternal deaths in Latin America and the Caribbean.

The risks for the baby are poor growth and prematurity. In developing countries neonatal mortality and morbidity is higher than in developed one, due to limited access to neonatal intensive care of public hospitals. 10%-15% of maternal death are associated with pre-eclampsia and eclampsia, and perinatal mortality is high following pre-ecampsia, and higher following eclampsia (Duley, 2009).

1.2.1.3 Hematological changes during pregnancy:

Pregnancy places extreme stresses on the haematological system and an understanding of the physiological changes that result is obligatory in order to interpret any need for therapeutic intervention. These changes are:

- **Physiological anemia**

Decrease in hemoglobin (Hb) concentration due to increased blood plasma volume by around 1250 ml, or 45% above normal.

- **iron deficiency anemia**

Up to 600 mg iron is required for the increase in red cell mass and a further 300 mg for the fetus. Despite an increase in iron absorption, few women avoid depletion of iron reserves by the end of pregnancy (Hoffbrand *et al*., 2006).

- **Folate deficiency**

Serum folate level decreases to about half the normal range due to increased folate requirements, along with poor diet, this leads to development of megaloblastic anemia (Hoffbrand *et al*., 2006).

- **Thrombocytopenia**

The platelet count falls by approximately 10% in an uncomplicated pregnancy. In approximately 7% of women this fall is more severe and can result in thrombocytopenia (platelet count <140 $x10^9$/L), and this known as incidental thrombocytopenia (Hoffbrand *et al*., 2006).

The hematologist is usually consulted in one of three scenarios that cause thrombocytopenia during pregnancy:

1. Pre-existing thrombocytopenia—most commonly, immune thrombocytopenia.
2. Decreasing platelet count or newly discovered thrombocytopenia in pregnancy, which may or may not be related to pregnancy.
3. Acute onset of thrombocytopenia in the setting of severe preeclampsia, the HELLP syndrome (hemolysis, elevated liver enzymes, low platelets) or acute fatty liver of pregnancy (Rajasekhar *et al*., 2013).

- **Haemostasis and thrombosis in pregnancy**

Pregnancy leads to a hypercoagulable state with consequent increased risks of thromboembolism and disseminated intravascular coagulation (DIC) due to changes in concentrations of procoagulant, anticoagulant, and anti-fibrinolytic factors. There is an increase in plasma factors VII,VIII, X, vWF, and fibrinogen, and in addition to the increase in procoagulant factors, there is a concurrent decrease in the anticoagulants proteins and antithrombin as well as a decrease in the activity of the fibrinolytic system, most likely due to an increase

in plasminogen activator inhibitors so fibrinolysis is suppressed (Hoffbrand *et al*., 2006; Rodger *et al*., 2006).

1.2.1.4 Classification of hypertensive disorders during pregnancy:

The hypertensive disorders of pregnancy are classified into:

- Chronic hypertension: which include patients with hypertension preceding, but exacerbated by pregnancy.
- Preeclampsia-eclampsia
- Preeclampsia superimposed on chronic hypertension: in which a sudden increase in proteinuria or a sudden increase in blood pressure in a woman with previously well controlled hypertension or in a woman with elevated blood pressure and proteinuria prior to 20 weeks of gestation.
- Gestational hypertension: which develop after 20 weeks of pregnancy but not associated with proteinuria.
- Chronic hypertension: in which hypertension persists for more than 12 weeks postpartum (McCrae, 2007).

1.2.1.5 Risk factors for pre-eclampsia:

Common risk factors are:

- Nulliparity
- African-American race
- Age more than 40 years

- Pre-eclampsia with previous pregnancy
- Diabetes
- Multiple gestation
- Lupus
- Chronic renal disease

- Obesity

1.2.1.6 Criteria for severe pre-eclampsia:

- Systolic blood pressure of at least 160 mmHg, or diastolic blood pressure at least 110 mmHg on two occasions at least 6 hours apart.
- Proteinuria of at least 5g in a 24 hours period.
- Oliguria.
- Pulmonary edema.
- Impaired liver function.
- Visual or cerebral disturbances.
- Epigastric or right upper quadrant pain.
- Thrombocytopenia.
- Intrauterine growth restriction (Fragento, 2006).

For the diagnosis of mild pre-eclampsia, a systolic blood pressure between 140 and 160 mmHg and a diastolic blood pressure between 90 and 110 mmHg, and proteinuria >1+ or
2+ on a urine dipstick was considered significant (Ramin, 2002).

1.2.1.7 Complications of preeclampsia and HELLP:

- Acute renal failure
- Pulmonary edema
- Abruptio placentae
- Hypertensive encephalopathy
- Postpartum hemorrhage
- Wound or intraabdominal hematomas
- Liver hematoma with possible rupture
- DIC and multiorgan failure, including liver, kidneys and lungs

- Neurologic-like eclampsia
- Ischemia, infarcts, edema and hemorrhage can also occur, as can cardiorespiratory arrest (Queenan *et al*., 2010).

1.2.1.8 Prevention and treatment of preeclampsia:

There is no single strategy that is of benefit in preventing the development of the disease, once the diagnosis of pre-eclampsia has been made, treatment options are limited, delivery of the fetus and placenta remain the only curative treatment (Norwitz *et al*., 2002).

Magnesium sulfate is administered to pre-eclamptic parturient for seizure prophylaxis.

Recent studies have demonstrated that it is more effective in preventing seizures than other drugs, including phenytoin and diazepam. While magnesium sulfate is administered to prevent seizures, it does produce other beneficial effects. A sustained decrease in systemic vascular resistance and an increase in cardiac index occur in pre-eclamptic patients receiving magnesium sulfate. This can lead to improved uteroplacental perfusion.

Parturients with severe hypertension will require antihypertensive therapy. Most obstetricians recommend treatment for systolic blood pressure >160 to 170 mmHg or diastolic blood pressure >105 to 110 mmHg. Intravenous hydralazine and labetalol are the most common antihypertensive agents used in pre-eclamptic patients (Fragento, 2006).

1.2.2 Platelets:

1.2.2.1 Definition:

Platelets are small anucleate cells that play a critical role in haemostasis and thrombosis.

Platelets are extremely small and discoid in shape, with dimensions in the human of approximately 3.0 × 0.5 μ m and a mean volume of 7 – 11 fL. This shape and small size enables platelets to be ‘ marginated’ by the red blood cells to the edge of the vessel, placing them next to the endothelial cells and therefore in the correct position to respond to vascular damage. Platelets are abundant in the circulation, with number in humans usually in the range $150 – 400 \times 10^9$ / L. Platelets are formed from megakaryocytes, one of the largest cells in the bone marrow, reaching more than 50 μ m in diameter. The nucleus of the megakaryocyte undergoes a process known as endomitosis that involves nuclear replication without cellular division, giving rise to DNA ploidy values that range from 4 *n* to 128 *n* . The reason why endomitosis occurs is not fully understood, but it may simply reflect the need to increase the DNA content to enable the cell to expand its protein synthetic capacity to generate 2000 – 3000 platelets per megakaryocyte. In addition, it allows cell growth and differentiation to occur without interruption by nuclear and cell divisions (Hoffbrand *et al*., 2011).

Platelets are central to primary hemostasis. They are responsible for the initial closure of the defect in the vessel wall through the formation of the platelet plug. Important pro- and anticoagulant factors are stored within platelet granules and these are released into the microenvironment around vessel injury. In secondary hemostasis, platelets provide the membrane surface to which the activated clotting factors bind, leading to increased enzyme efficiency and increased thrombin generation.(Shaughnessy *et al*., 2005).

1.2.2.2 Platelet structure:

1. Glycocalyx: Is the outer membrane surface. It is rich in glycoproteins, which serve as membrane receptors:

a. Glycoprotein Ib is the receptor for vonWillebrand's factor (vWF) in the presence of ristocetin.

b. Glycoproteins IIb and IIIa are receptors for vWF and fibrinogen and are exposed by stimulation of thrombin or adenosine diphosphate (ADP).

c. Glycoprotein Va is the receptor for thrombin.

2. Microtubule and micro filaments: These provide an active means of platelet Contraction to squeeze out the contents of the cytoplasmic granules.

a. Microtubules form the submembranous band around the circumference of the cell and structurally support the normal discoid-shaped platelets.

b. The contractile microfilaments (thrombosthenin) contain actin and are closely related to the microtubule.

3. The open canalicular system: Provides direct communication between intracellular and extracellular compartments.

4. A dense tubular system: Forms a circle within the microtubule. This system serves as

a. A site for arachidonic acid metabolism.

b. A calcium-sequestering pump that maintains platelet cytoplasmic calcium levels.

5. Mitochondria: Are responsible for energy production.

6. Glycogen granules: Provide energy substrate.

7. Alpha (α) granules contain contact-promoting factors, including:

a. Platelet fibrinogen.

b. Platelet-derived growth factor.

c. von Willebrand's factor (factor VIII:R).

d. β-Thromboglobulin.

e. Platelet factor 4 (heparin-neutralizing).

f. Fibronectin.

8. Dense granules contain nonprotein factors including:

a. Adenosine diphosphate (ADP)

b. Adenosine triphosphate (ATP)

c. 5-Hydroxytryptamine (5-HT; or serotonin)

d. Calcium

9. Other specific organelles include lysosomes which contain hydrolytic enzymes, and

peroxisomes which contain catalase (Hubbard, 2010)

1.2.2.3 Platelets functions:

Platelets are involved in the interaction with the vascular endothelium to stop and prevent bleeding after vascular injury (primary hemostasis).

Approximately 30 percent of the platelets that have been released from the bone marrow into the circulation are sequestered in the microvasculature or in the spleen as functional reserves (Harmening, 2005).

The main functions of platelets are:

1-Adhesion

Platelets adhere to the exposed subendothelial connective tissue, which release factors that cause platelets activation, shape transformation, and aggregation. These factors are:

a. Collagen

b. Fibronectin

c. vWF (factor VIII:R)

d. Thrombin

e. ADP

2- Activation and release reaction

Tissue platelet activators cause the platelet to change shape from discoid to spherical.

Dense and α-granules undergo internal contraction and centralization. The complete process is calcium dependent.

a. The exposure of surface membrane receptors to vWF and fibrinogen results in cytoplasmic calcium ionization, stimulation of ATP generation, and activation of the actin monomers in the micro filaments.

b. Contractions result in a centralization of the cytoplasmic granules and a release of their contents through the canalicular system.

3-Agreggation

Plug formation, or secondary aggregation, is primarily stimulated by thrombin and thromboxane A2 (TXA2).

a. Membrane-binding of vWF and collagen to platelet receptors unmasks membrane phospholipid A2, which is the precursor of arachidonic acid, and is important for the production of TXA2 and other prostaglandins. Phospholipid A2 is unmasked by the binding of vWF and collagen to platelet receptors on the membrane.

b. TXA2 inhibits adenylate cyclase [thus, it inhibits the formation of cyclic adenosine

monophosphate and liberates sequestered calcium into cytoplasm.

Calcium causes further cytoplasmic contraction, release of granule contents, and platelet aggregation.

c. Thrombin enzymatically cleaves fibrinogen to form fibrin, which is necessary to stabilize the platelet plug.(Hoffbrand *et al*, 2006; Hubbard, 2010).

Tests of platelets functions:

Blood collection needs to be optimal with non-traumatic venepuncture, rapid transport to the lab with storage at room temperature and testing within a maximum of 6h (Harmening, 2005).

a- Platelet count:

Normal range 150–450 $\times 10^9$/L. Adequate function is maintained even when the count is <1⁄3 normal level, but progressively deteriorates as it drops. With platelet counts <20 × 10^9/L there is usually easy bruising, petechial haemorrhages (although more serious bleeding can occur).

b- Platelets morphology:

Large platelets are biochemically more active; high mean platelet volume (MPV) is associated with less bleeding in patients with severe thrombocytopenia, and can be counted by new analysers.

c- Platelet adhesion: Rarely performed in routine lab practice.

d- Platelet aggregation:

Performed on fresh sample using aggregometer but poor correlation with bleeding tendency except in specific circumstances, e.g. Glanzmann's thrombasthenia, Bernard–Soulier syndrome (Provan *et al.*, 2004).

e- Bleeding time:

Is an effective in vivo screening test of platelet function by timing the length it takes for platelets to plug broken capillaries after a small cut is made in the forearm.

Reference values are approximately 3 to 8 minutes, and increased bleeding times are seen in:

(1) Patients taking drugs with antiplatelet action (e.g., aspirin).

(2) Patients with von Willebrand's disease.

(3) Patients who suffer from congenital platelet abnormalities.

(4) Patients with platelet counts lower than 100,000/mm3 (Hubbard, 2010).

1.2.2.4 Platelet pathophysiology:

1.2.2.4.1 Quantitative platelet disorders:

- **Thrombocytopenia:**

Is characterized by a decrease in the number of circulating platelets (i.e., <100,000/mm3) which result from the following conditions:

A- Defective production in the bone marrow due to:

(1) Decreased numbers of megakaryocytes

(2) Ineffective platelets production

B- Disorders of distribution and dilution these conditions include:

(1) Splenic pooling, which is seen with splenomegaly and hypersplenism

(2) Hypothermia

(3) Dilution in the circulation by transfused stored blood

C- Thrombocytopenia due to consumption of platelets occurs in the following disorders:

(1) Thrombotic thrombocytopenia purpura.

(2) Toxicity due to snake venoms

(3) Tissue injury

(4) Obstetric complications

(5) Neoplasms

(6) Bacterial and viral infections

(7) Intravascular hemolysis

(8) Hemolytic uremic syndrome

(9) Vasculitis (as seen with systemic lupus erythematosus).

(10) Disseminated intravascular coagulation (DIC)

D-Thrombocytopenia due to destruction of platelets occurs in the following disorders:

(1) Idiopathic (immunologic) thrombocytopenia purpura.

(2) Posttransfusion purpura

(3) Drug-induced thrombocytopenia

E- Heparin-induced thrombocytopenia is observed in more than 10% of patients who undergo heparin therapy.

F- Thrombocytopenia associated with human immunodeficiency virus infection (Hubbard, 2010).

- **Thrombocytosis:**

Is characterized by an increase in circulating platelet counts >450,000/mm3.

(a) Essential thrombocytosis

It is the result of a primary bone marrow disorder, and. Patients will have increased bleeding tendencies because of possible accompanying functional abnormalities. As seen in patients with the following disorders:

(1) Hodgkin's lymphoma

(2) Polycythemia vera

(3) Myelofibrosis

(4) Chronic myelogenous leukemia

(5) Thrombocythemia

(b) Secondary thrombocytosis is a secondary response most commonly associated with the following disorders:

(1) Iron-deficiency anemia

(2) Chronic inflammatory disease

(3) Splenectomy

(4) Rebound thrombocytosis, which may occur after a platelet depletion through a massive blood loss (Hubbard, 2010).

1.2.2.4.2 Qualitative platelet abnormalities:

1-Surface membrane defects that are genetically acquired as seen in the following disorders:

(a) Glanzmann thrombasthenia

(b) Bernard-Soulier syndrome

2- Abnormalities in the granular fraction of the platelet

- Defects in dense granule

These could be seen in the following disorders:

(a) Hermansky-Pudlak syndrome

(b) Chediak-Higashi syndrome

(c) Wiskott-Aldrich syndrome

- α-Granule deficiencies are rare platelet functional abnormalities in which both aggregation and release properties are diminished.
- Storage pool disease, also known as Grey platelet syndrome is a congenital platelet disease which associates thrombocytopenia and aggregation abnormalities.

3. Deficiencies of thromboxane generation: can occur because of a genetic deficiency of the cyclo-oxygenase enzyme (Hubbard, 2010).

1.2.3 The effect of pre-eclampsia on platelets count and indices:

The platelet activation is a major cause of accelerated platelet clearance in this disorder, and this argument is supported by increased plasma levels of the platelet α-granule proteins β-thromboglobulin and platelet factor 4, as well as the increased levels of thromboxane A2 metabolites in the urine of preeclamptic patients. The activation of coagulation system and generation of thrombin leads to platelets activation with accelerated clearance of activated platelets. The platelets contact with exposed subendothelium underlying the injured placental vasculature, increased level of von Willebrand factor (vWF), as well as adhesive proteins such as fibronectin also cause platelet activation. Patients with preeclampsia also display microangiopathic hemolytic anemia due to red cell fragmentation, due to shearing of red cells on fibrin strands in the microvasculature or placental circulation (McCrae, 2007).

Platelets when activated increase number and size of pseudopodia which in turn increase the size of platelets leading to large platelets formation. The mean platelet volume (MPV), and platelet distribution width (PDW) are platelet indices that increased during platelet activation (Vagdatli *et al*., 2010).

The MPV is measured by clinical hematology analyzers and it is used as a marker for platelet function and activation, and it is increased in certain vascular risk factor states, such as hypercholestrolaemia, diabetes mellitus, and pre-

eclampsia. An elevated MPV predicts a poor outcome following myocardial infarction and the development of pre-eclampsia (Bath and Butterworth, 1996).

The PDW is calculated, by the hematology analyzers, by measuring the width of the size distribution curve (in femtolitre (fl)) at the 20% level when the peak distribution curve is taken as 80% or 100% (Briggs *et al*., 2007).

The automated blood count shows an increased MPV and PDW when there is increased platelet consumption or destruction and a low MPV when there is failure of bone marrow output (Bain, 2006).

1.2.4 Previous and relevant studies :

Boriboonhirunsarn *et al* study showed that MPV in severe preeclamptic women is statistically greater than in normal pregnant women (Boriboonhirunsarn *et al*., 1995).

A previous longitudinal study in Turkey provided evidence that mean platelet volume (MPV) gradually increased in pregnant women affected by pre-eclampsia compared to women with normal pregnancies (Dundar *et al*., 2008).

Other study in Brazil found that lower platelets count were observed in sever pre-eclampsia compared to normal pregnant and to non pregnant women (Freitas *et al*., 2013).

A study was conducted among normal and pre-eclamptic pregnancies, and they found that there was increase in mean platelet volume (MPV) in patients with pre-eclampsia and suggested that mean platelet volume (MPV) may serve as early monitoring marker for the severity of pre-ecampsia (Han *et al*.,2014).

Recently, a study was conducted in Korea and the researcher found that, as the disease progressed, there was decrease in platelet count, and increase in mean platelet volume (MPV) and platelet distribution width (PDW). Also they suggested that PDW can serve as a candidate marker for predicting the severity of pre-eclampsia (Yang *et al*., 2014).

On the other hand, some studies disagreed with the above studies, they found that there were no prognostic significance of complete blood count (CBC), platelets count, and mean platelet volume (MPV) on the presence and/or severity of pre-eclampsia (Ceyhan et *al*., 2006).

Another study in Turkey found that, increased mean platelet volume (MPV) was not a significant predictor for pre-eclampsia, and did not differ both between mild and severe pre-eclampsia, and pre-eclampsia and normotensive pregnant women (Altinbas *et al*., 2012).

Duckitt and Harrington (2005) in their study proved that nulliparity and family history of pre-eclampsia were strong risk factor almost increasing the risk of pre-eclampsia.

The mean platelet volume (MPV) and platelet to lymphocyte ratio (PLR) did not show statistically significant differences between patients with severe preeclampsia (PE), healthy pregnant and non-pregnant women (Yavuzcan *et al.*, 2014).

1.3 Justification:

Pre-eclampsia is an obstetric disorder that is associated with maternal and prenatal morbidities. Timely accurate diagnosis of the disease is very important and helpful in preventing the poor outcome of the disease.

Sudan is one of the developing countries, where pre-eclampsia is one of the main causes of obstetric complications with high maternal morbidities and mortality. Because of the importance of the disease for both mothers and babies along with the significant increase in the affected individuals among pregnant ladies, a reliable predictor for pre-eclampsia will play an important role in early prevention and intervention.

This study was conducted to determine the changes in platelets count and indices among pre-eclamptic women and to verify their value in predicting the severity of pre-eclampsia for early interventions, since they are simple and routinely done tests, with lower cost and greater accessibility in clinical laboratories.

1.4 Objectives:

1.4.1 General objective:

To determine the level of platelets count and platelets indices among Sudanese women with pre-eclampsia compared with the level among normotensive pregnant women in some Khartoum Maternity Hospitals.

1.4.2 Specific objectives:

1. To measure platelet count, mean platelet volume (MPV), platelet distribution width (PDW), and platelet large cell ratio (P-LCR) in pregnant women with pre-eclampsia and correlate it with those in pregnant women without pre-eclampsia.
2. To identify whether there is any correlation between severity of pre-eclampsia and platelet count, mean platelet volume (MPV), platelet distribution width (PDW), and platelet large cell ratio (P-LCR).
3. To calculate lymphocyte/platelet and red cells/platelet ratios and correlate it with the degree of preeclampsia.
4. To find out the possible association between age, education, and family history of pre-eclampsia with the development of pre-eclampsia.

CHAPTER TWO

MATERIALS AND METHODS

Chapter two

2. Materials and Methods

2.1 Study design:

This is an observational analytical case control study carried out among pregnant women with pre- eclampsia (cases) and normal pregnant women (control).

2.2 Study area and duration:

The study was carried out at Omdurman Maternity Hospital, Ibrahim Malik Hospital, and

Academy Charity Teaching Hospital. The data collection period was from April to July 2015.

2.3 Study population:

2.3.1 Inclusion criteria:

Pregnant women who attended the above mentioned hospitals with hypertension (a systolic blood pressure of at least 140 mmHg or diastolic blood pressure of 90 mmHg) and proteinuria (> 300 mg per 24 hours) after the twentieth week of gestation during the study data collection period (from April to July 2015) were enrolled in this study. Normotensive pregnant women who attended the hospitals for routine obstetric care were used as control. Control pregnant women were matched to cases in age.

2.3.2 Exclusion criteria:

Women with history of hypertension, diabetes, malaria, renal disease, and cardiovascular diseases were excluded from the study.

2.4 Data collection:

A predesigned questionnaire was used for collecting the required data from both pregnant women with pre-eclampsia (cases) and normal pregnant women (control). Variables in this questionnaire included age, residence, education, occupation, gravida, parity, gestational age, history of hypertension, history of diabetes mellitus, family history of Pre-eclampsia, other chronic or infectious diseases, blood pressure, and proteinuria. The questionnaire was administered by the researcher.

2.5 Sample size and sampling technique:

All incident cases that were found in the hospitals during data collection period were included. The control cases were randomly selected from pregnant women who attended the hospitals for routine obstetric care using simple random sampling from the list of all pregnant women who attended to the hospitals during the same time. A total of 120 samples (60 cases and 60 controls) were collected.

2.6 Blood Sampling:

About 2.5 ml of venous blood was collected once from both study group and control group by clean venepuncture, using a sterile disposable syringe, into commercially prepared concentration of Ethylene Di-amine Tetra-acetic Acid (EDTA) containers. Then each sample was mixed thoroughly and gently. All samples were tested within less than 6 hours of collection using hematology analyzer (Sysmex KX-21N model- Japan) which performs blood cell count by direct current detection method.

2.7 Methods:

2.7.1 Principle of hematological analyzer: Direct current detection method:

Blood sample was aspirated, measured to a predetermined volume, diluted at specified ratio, then fed into each transducer. The transducer chamber had a minute hole called the aperture. On both side of the aperture, there were the electrodes between which flowed direct current. Blood cells suspended in the diluted sample passed through the aperture, causing direct current resistance to change between the electrodes. As direct current resistance changed, the blood cell size was detected as electric pulses.

Blood cells count was calculated by counting the pulse, and a histogram of blood size was plotted by determining the pulse size (Sysmex Corporation, 2004).

2.7.2 Blood cell discrimination circuit:

White blood cells (WBC), Red blood cells (RBC), and Platelets (PLT) were discriminated and calculated by the following blood cell discriminator.

2.7.2.1 White blood cells discriminator:

As to WBC lower discriminator, the optimum position in 30 – 60 fl was automatically determined by the microcomputer. WBC was calculated from the particle counted more than this lower discriminator (LD).

2.7.2.2 Red blood cells discriminator:

As to RBC lower discriminator (LD) and upper discriminator (UD), the optimum position in 25 – 75 fl and 200 – 250 fl, respectively, were automatically determined by the microcomputer. RBC were calculated from the particle counted between this lower discriminator and upper discriminator.

2.7.2.3 Platelets discriminator:

As PLT lower discriminator (LD) and upper discriminator (UD), the optimum position in 2 – 6 fl and 12 – 30 fl, respectively, were automatically determined by the microcomputer. PLT were calculated from the particle counted between this lower discriminator and upper discriminator.

2.7.3 Platelets histogram:

Platelets histogram was analyzed using three discriminators: two discriminators (LD) and (UD) –determined automatically between 2 - 6 fl and between 12 – 30 fl, respectively- and the fixed discriminator at 12 fl. Regarding platelets histogram, check was made to see that there were no relative frequency errors at discriminators (LD) and (UD), distribution width error, and there was a single peak.

2.7.3.1 Platelets distribution width (PDW):

PDW is the distribution width on 20% frequency level with the peak taken as 100%. The unit applied was femtoliter (fl).

2.7.3.2 Mean platelet volume (MPV):

MPV was calculated by the following formula:

MPV (fl) = PCT (%) × 1000 / PLT (10^3/µl)

Where PCT (%) represented the value weight with platelet frequency and is called platelet- crit or platelet volume ratio.

2.7.3.3 Platelet large cell ratio (P-LCR):

This was the ratio of large platelets exceeding 12 fl discriminator and was calculated as the ratio of the particle count between the 12 fl fixed discriminator and upper discriminator (UD) to the particle count between (LD) and upper discriminator (UD) (Sysmex Corporation, 2004).

2.8 Ethical consideration:

Samples were collected following a written informed consent from each participant, and the questionnaire was easy to them, to give accurate and formative answers. Ethical approval was obtained from the Health Ministry of Khartoum State and a permission of the study conduct was taken from the administrates of the three hospitals.

2.8 Data analysis:

Data were analyzed using Statistical Package for Social Science soft ware (SPSS version 15), and the statistical tests used were Chi-square test as test of association, Spearman's rho Correlations, T. test as statistical tests of significance of differences between means of every two variables handled in this study and Odds ratio was calculated. P value was considered statistically significant at 0.05.

Normal values for platelets count ($\times 10^3/\mu l$), platelet distribution width (PDW(fl)), mean platelet volume (MPV(fl)), and platelet large cell ratio (P-LCR(%)) were used as statistical cut point to analyze these parameters in the two studied groups (Appendix IV).

CHAPTER THREE

RESULTS

Chapter Three

3. Results

This case-control study involved sixty pregnant Sudanese women with pre-eclampsia (cases) and other sixty disease- free pregnant women (control) who attended three maternity hospitals in Khartoum State.

Distribution of the participants of the two groups (cases and controls) according to their ages, showed that 40 (33.3%) of them were found to have been falling in the age group (22-26) years, whereas 7 (5.8%) of them were falling in the age group (37-42) years with an overall average age of 26 years and standard deviation, as a measure of variability (dispersion), of 6 years as seen in table 3.1.

Figure 3.1 showed that 117 (97.5%) of them were from Khartoum State. According to their level of education, it was noticed that 39 (32.5%) of both cases and controls were having basic education with an illiteracy rate 21.7% among the two groups (figure 3.2).

The results showed that 112 (93.3%) of the participants in the two groups (n=120) were house wives, whereas 5 (4.2%) of them were teachers (figure 3.3).

Table 3.2 shows the distribution of the participants of the two groups (cases and controls) according to the potential risk factors, where cases showed a higher level of education than the controls, 72 (60%) of them were found to have gravida 1 to 3, whereas 33 (27.5%) were having gravida 4 to 6, with an overall average gravida of 3, with slight differences in gravida among both cases and controls, 51 (42.5%) of both cases and controls were found to have zero parity (primigravida), whereas 34 (28.3%) of them were having a parity of 3 to 5 with an overall average parity of 2. Parity 1 to 2 and parity 3 to 5 were greater among controls compared with cases. It was found that family history of pre-eclampsia was present among 37 (30.8%) of the families of cases, compared with 5 (4.2%) among the families of the controls.

The study showed that the average systolic blood pressure among cases was found to be 165 mmHg compared with 112 mmHg among controls, with an overall average of 138 mmHg among both cases and controls. Average diastolic blood pressure was found to be 103 mmHg among cases compared with 74 mmHg among controls, with an overall average of diastolic pressure of 89 mmHg. Thirty (25%) of the cases showed proteinuria level of (+++), whereas 13 (10.8%) of them showed a level of (++++).

Severe pre-eclampsia was shown by 38 (63.3%) of the cases (n=60), whereas the mild type was seen among the rest (figure 3.4).

A vast number of significant differences between the two means of platelets count ($\times 10^3/\mu l$), platelet distribution width (PDW(fl)), mean platelet volume (MPV(fl)), and platelet large cell ratio (P-LCR(%)) between cases and control was highly significant (P-value= 0.00) as seen in tables 3.3 and among severe and mild cases (P. value 0.02,0.008,0.005,0.005), respectively, as seen in table 3.4.

Chi square test, as a nonparametric test of association, showed that educational level and family history of pre-eclampsia as independent variables, were found to be associated with the developing pre-eclampsia as dependent variable, P-values were highly significant (P-value=0.001 and 0.000, respectively) as seen in table 3.5. On the other hand age, educational level, and family history of pre-eclampsia were not associated with the degree of pre-eclampsia as shown in table 3.6.

For red blood cell to platelet ratio (RPR), the difference between the means levels of mild and severe type of pre-eclampsia was found to be significant (P-value= 0.002) as shown in table 3.7. Red blood cell to platelet ratio (RPL) was found to be weekly correlated with the degree of pre-eclampsia as shown in table 3.8.

Application of Chi square test among cases and controls as regarding the Platelets count ($\times 10^3/\mu l$), mean platelet volume (MPV (fl)), platelets

distribution width (PDW(fl)), and platelets large cell ratio (P-LCR(%)) as a test of significance of differences between percentages, the following results were obtained, as shown in table 3.9. There was a significant difference between cases and controls regarding platelets count. Cases showed a high percentage of low platelets count than controls, P value (0.000) which is highly significant. The difference in percentages between cases and controls regarding platelets distribution width (PDW) was quite significant. High PDW (fl) was greater among cases than controls. None of the controls has shown high PDW (fl). Mean platelets volume (MPV (fl)) was significantly among cases than controls. Platelets large cell ratio (P-LCR (%)) was significantly among cases compared with controls where none of the controls had shown any high P-LCR (%).

Table 3.10 showed significant differences between the percentages of severe pre- eclampsia and low platelets count, P value (0.001). Those with severe pre-eclampsia showed low level of platelets count.

Application of Spearman's rho correlation coefficient between pre-eclampsia and the 4 measurements (variables) dealt with above, revealed the following association as shown on table 3.11:

- A weak negative correlation was found between pre-eclampsia and platelets count
- A weak positive correlation was found between pre-eclampsia and mean platelets volume (MPV).
- Pre-eclampsia was found to be positively and weakly associated with platelets large cell ratio (P-LCR).

Computation of odds ratio (OR) among cases and controls in regard to family history of pre-eclampsia and gravidity, revealed that those with family history of pre-eclampsia were more likely to develop the disease almost 18 times as those of no family history of pre-eclampsia, and primigravida ladies were more likely

to develop the disease almost twice time as multigravida one, as shown on table 3.12 and 3.13.

Table 3.1: Distribution of study sample according to age.

Cross tabulation		Type	
		Case (Pregnant with Preeclampsia)	Control (Healthy pregnant)
Age group (Years)	(17-21)	10 (8.30%)	16 (13.30%)
	(22-26)	22 (18.30%)	18 (15.00%)
	(27-31)	17 (14.20%)	18 (15.00%)
	(32-36)	5 (4.20%)	7 (5.80%)
	(37-42)	6 (5.00%)	1 (0.80%)
Study population = 120 (100%)			

Table 3.2: Distribution of the two studied groups according to the potential risk factors.

Cross tabulation		Case (Pregnant with Preeclampsia)	Control (Healthy pregnant)
Educational level	Illiterate	6 (5.00%)	20 (16.70%)
	Basic	21 (17.50%)	18 (15.00%)
	Secondary	14 (11.70%)	17 (14.20%)
	University	19 (15.80%)	5 (4.20%)
Gravidity	(1-3)	36 (30.00%)	36 (30.00%)
	(4-6)	15 (12.50%)	18 (15.00%)
	(7-9)	9 (7.50%)	6 (5.00%)
Parity	None	29 (24.20%)	22 (18.30%)
	(1-2)	11 (9.20%)	14 (11.70%)
	(3-5)	13 (10.80%)	21 (17.50%)
	(6-8)	7 (5.80%)	3 (2.50%)
Family history of Preeclampsia	Yes	37 (30.80%)	5 (4.20%)
	No	23 (19.20%)	55 (45.80%)
Study population = 120 (100%)			

Table 3.3: The effect of pre-eclampsia on complete blood count.

T-test		Mean	Std. Deviation	P-value
TWBCs ($\times 10^3/\mu L$)	Case (Pregnant with Preeclampsia)	12.7	4.2	0.000**
	Control (Healthy pregnant)	7.7	2.1	
RBCs ($\times 10^6/\mu L$)	Case (Pregnant with Preeclampsia)	4.3	0.7	0.445
	Control (Healthy pregnant)	4.2	0.5	
HGB (g/dL)	Case (Pregnant with Preeclampsia)	11.2	2.1	0.141
	Control (Healthy pregnant)	11.6	1.3	
HCT (%)	Case (Pregnant with Preeclampsia)	32.8	5.6	0.270
	Control (Healthy pregnant)	33.7	3.5	
MCV (fL)	Case (Pregnant with Preeclampsia)	76.9	6.4	0.001**
	Control (Healthy pregnant)	80.5	5.0	
MCH (pg)	Case (Pregnant with Preeclampsia)	26.3	3.2	0.003**
	Control (Healthy pregnant)	27.8	2.2	
MCHC (g/dL)	Case (Pregnant with Preeclampsia)	34.0	2.0	0.101
	Control (Healthy pregnant)	34.6	1.5	
PLT ($\times 10^3/\mu L$)	Case (Pregnant with Preeclampsia)	185.5	78.1	0.000**
	Control (Healthy pregnant)	257.7	66.6	
RDW_SD (fL)	Case (Pregnant with Preeclampsia)	45.4	4.7	0.001**
	Control (Healthy pregnant)	42.9	3.8	
RDW_CV (%)	Case (Pregnant with Preeclampsia)	16.2	2.8	0.000**
	Control (Healthy pregnant)	14.1	1.4	
PDW (fL)	Case (Pregnant with Preeclampsia)	15.5	3.5	0.000**
	Control (Healthy pregnant)	12.6	2.0	
MPV (fL)	Case (Pregnant with Preeclampsia)	10.8	1.1	0.000**
	Control (Healthy pregnant)	9.8	1.0	
P_LCR (%)	Case (Pregnant with Preeclampsia)	32.8	8.6	0.000**
	Control (Healthy pregnant)	24.1	7.0	

* T- Test P. value less than 0.05 that's considered as statistically significant.

Table 3.4: The effect of the grade of pre-eclampsia on complete blood count.

T-test	Preeclampsia	Mean	Std. Deviation	P-value
TWBCs ($\times 10^3/\mu L$)	Mild	13.1	5.2	0.574
	Severe	12.5	3.7	
RBCs ($\times 10^6/\mu L$)	Mild	4.2	0.7	0.760
	Severe	4.3	0.8	
HGB (g/dL)	Mild	11.0	1.9	0.669
	Severe	11.3	2.2	
HCT (%)	Mild	32.6	5.5	0.842
	Severe	32.9	5.7	
MCV (fL)	Mild	77.0	5.8	0.978
	Severe	76.9	6.8	
MCH (pg)	Mild	26.1	2.6	0.711
	Severe	26.4	3.6	
MCHC (g/dL)	Mild	33.8	1.5	0.508
	Severe	34.2	2.2	
PLT ($\times 10^3/\mu L$)	Mild	216.1	56.9	0.020**
	Severe	167.8	83.7	
RDW_SD (fL)	Mild	43.6	3.9	0.016**
	Severe	46.5	4.7	
RDW_CV (%)	Mild	15.3	2.0	0.041**
	Severe	16.8	3.0	
PDW (fL)	Mild	14.0	2.9	0.008**
	Severe	16.4	3.5	
MPV (fL)	Mild	10.3	1.0	0.005**
	Severe	11.1	1.1	
P_LCR (%)	Mild	28.7	8.0	0.005**
	Severe	35.1	8.2	

* T- Test P.value less than 0.05 that's considered as statistically significant.

Table 3.5 Univariate analysis for individual factors in the studied groups (cases and control).

Cross tabulation		Type		Pearson Chi-Square value	P. value
		Case	Control		
Age group (Years)	(17-21)	10 (8.30%)	16 (13.30%)	5.718	0.221
	(22-26)	22 (18.30%)	18 (15.00%)		
	(27-31)	17 (14.20%)	18 (15.00%)		
	(32-36)	5 (4.20%)	7 (5.80%)		
	(37-42)	6 (5.00%)	1 (0.80%)		
Educational level	Illiterate	6 (5.00%)	20 (16.70%)	16.226	0.001**
	Basic	21 (17.50%)	18 (15.00%)		
	Secondary	14 (11.70%)	17 (14.20%)		
	University	19 (15.80%)	5 (4.20%)		
Family history of Preeclampsia	Yes	37 (30.80%)	5 (4.20%)	37.509	0.000**
	No	23 (19.20%)	55 (45.80%)		

* Chi square p value less than 0.05 that’s considered as statistically significant

Table 3.6: Univariate analysis for individual factors according to the degree of pre-eclampsia.

Cross tabulation		Preeclampsia		Chi square value	p value
		Mild	Severe		
Age group (Years)	(17-21)	1 (1.7%)	9 (15%)	4.291	0.368*
	(22-26)	10 (16.7%)	12 (20%)		
	(27-31)	6 (10%)	11 (18.3%)		
	(32-36)	2 (3.3%)	3 (5%)		
	(37-42)	3 (5%)	3 (5%)		
Educational level	Illiterate	2 (3.3%)	4 (6.7%)	1.720	0.632*
	Basic	10 (16.7%)	11 (18.3%)		
	Secondary	4 (6.7%)	10 (16.7%)		
	University	6 (10%)	13 (21.7%)		
Family history of Preeclampsia	Yes	14 (23.3%)	23 (38.3%)	0.057	0.811*
	No	8 (13.3%)	15 25%)		

* Chi square p value more than 0.05 that's considered as statistically insignificant.

Table 3.7: Differences between means of platelet to lymphocyte ratio and red blood cell to platelet ratio in the studied cases.

Variables	Degree of Preeclampsia	N	Mean	Std. Deviation	P-value
PLR	Mild	22	102.9	40.6	0.122
	Severe	38	82.3	52.9	
RPR	Mild	22	20.7	5.9	0.002**
	Severe	38	31.9	15.8	

* T- Test P. value less than 0.05 that's considered as statistically significant.

Table 3.8: The association of red blood cell to platelet ratio and the degree of the disease.

Spearman's rho Correlations		Preeclampsia
RPR	Correlation Coefficient	0.245
	Sig. (2-tailed)	0.059
	N	60
	Strength	Weak
	Direction	Positive
Correlation insignificant at 0.05 level		

Table 3.9: Differences between percentages of platelet count and indices in the cases and controls.

Cross tabulation		Type		Chi square value	p value
		Case	Control		
PLT (×10^3/µL)	Low	22 (18.3%)	1 (0.8%)	23.720	0.000**
	Normal	38 (31.7%)	59 (49.2%)		
PDW (fL)	Low	1 (0.8%)	1 (0.8%)	12.131	0.002**
	Normal	48 (40%)	59 (49.2%)		
	High	11 (9.2%)	0		
MPV (fL)	Low	1 (0.8%)	5 (4.2%)	6.274	0.044**
	Normal	53 (44.2%)	54 (45%)		
	High	6 (5%)	1 (0.8%)		
P_LCR (%)	Low	1 (0.8%)	4 (3.3%)	7.883	0.019**
	Normal	53 (44.2%)	56 (46.7%)		
	High	6 (5%)	0		
	Study population = 120 (100%)				

* Chi square p value less than 0.05 that's considered as statistically significant.

Table 3.10: The association of the degree of pr-eclampsia and platelets count and indices.

Cross tabulation		Preeclampsia		Chi square value	p value
		Mild	Severe		
PLT (×10^3/µL)	Low	2 (3.3%)	20 (33.3%)	11.375	0.001**
	Normal	20 (33.3%)	18 (30%)		
PDW (fL)	Low	1 (1.7%)	0	3.522	0.172
	Normal	19 (31.7%)	29 (48.3%)		
	High	2 (3.3%)	9 (15%)		
MPV (fL)	Low	1 (1.7%)	0	5.400	0.067
	Normal	21 (35%)	32 (53.3%)		
	High	0	6 (10%)		
P_LCR (%)	Low	1 (1.7%)	0	5.400	0.067
	Normal	21 (35%)	32 (53.3%)		
	High	0	6 (10%)		

* Chi square p value less than 0.05 that's considered as statistically significant.

Table 3.11: The association of pre-eclampsia and the changes in platelet count and indices.

Spearman's rho Correlations		PLT ($\times 10^3/\mu L$)	PDW (fL)	MPV (fL)	P_LCR (%)
Preeclampsia	Correlation Coefficient	-0.435**	0.217	0.294*	0.294*
	Sig. (2-tailed)	0.001	0.096	0.023	0.023
	N	60	60	60	60
	Strength	Weak	Weak	Weak	Weak
	Direction	Negative	Positive	Positive	Positive
** Correlation is significant at the 0.01 level (2-tailed).					
* Correlation is significant at the 0.05 level (2-tailed).					

Table 3.12: The association of family history of pre-eclampsia and the studied disease.

Cross tabulation		Type		
		Case	Control	Total
Family history of Preeclampsia	Yes	37 (30.8%) A	5 (4.2%) B	42 (35%) A+B
	No	23 (19.2%) C	55 (45.8%) D	78 (65%) D+C
Total		60 (50%) A+C	60 (50%) B+D	120 (100%) A+B+D+C
Odds Ratio = ((a*d)/(c*b)) = 17.7 CIM (6.173-50.724)				

Table 3.13: The association of gravidity and the studied disease.

Cross tabulation		Type		
		Case (Pregnant with Preeclampsia)	Control (Healthy pregnant)	Total
Gravidity	Primigravida	28 (23.3%) A	19 (15.8%) B	47 (39.2%) A+B
	Multigravida	32 (26.7%) C	41 (34.2%) D	73 (60.8%) C+D
Total		60 (50%) A+D	60 (50%) B+D	120 (100%) A+B+C+D
Odds Ratio = ((a*d)/(c*b)) = 1.9 CIM (0.898-3.972)				

Risk factors (Family history of Preeclampsia and Gravidity,): Case (Pregnant with Preeclampsia) more likely to be exposed than Control (Healthy pregnant) with odd ratio 17.7 and 1.9, respectively.

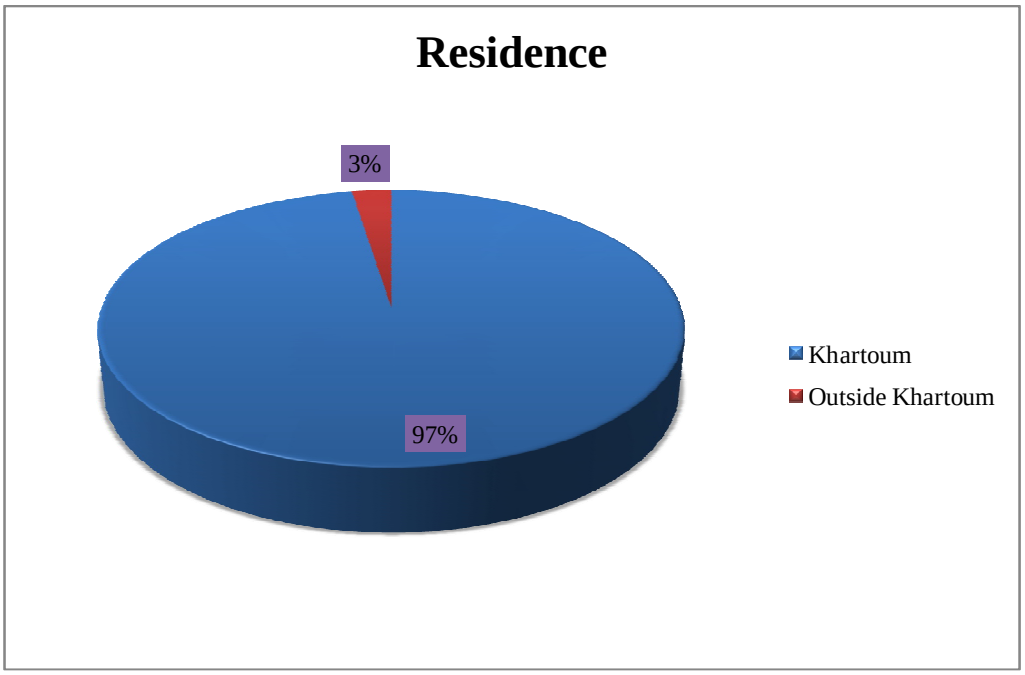


Figure 3.1: Distribution of the study sample according to residence.

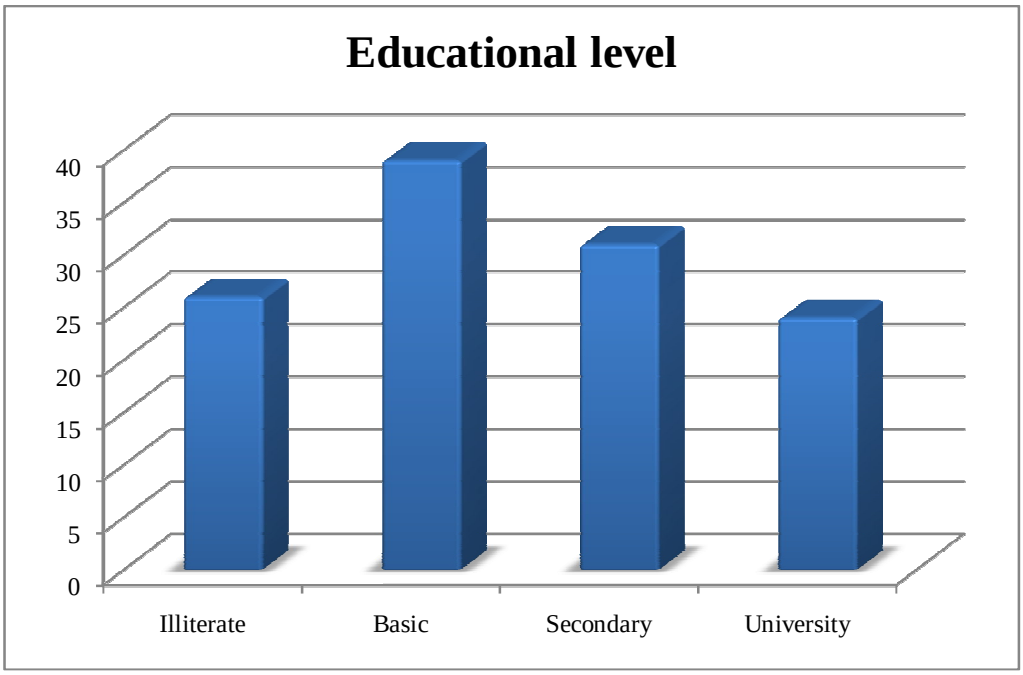


Figure 3.2: Distribution of the study sample according to educational level.

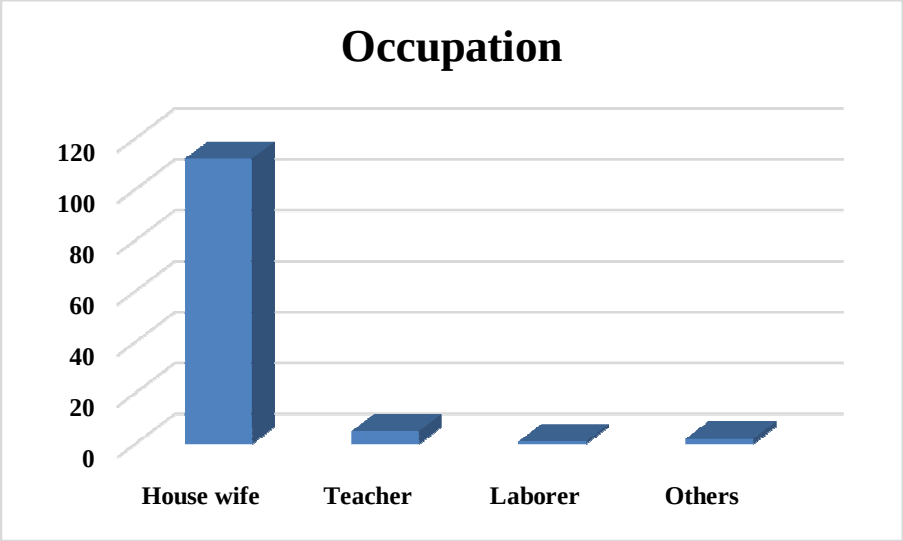


Figure 3.3: Distribution of the study sample according to occupation.

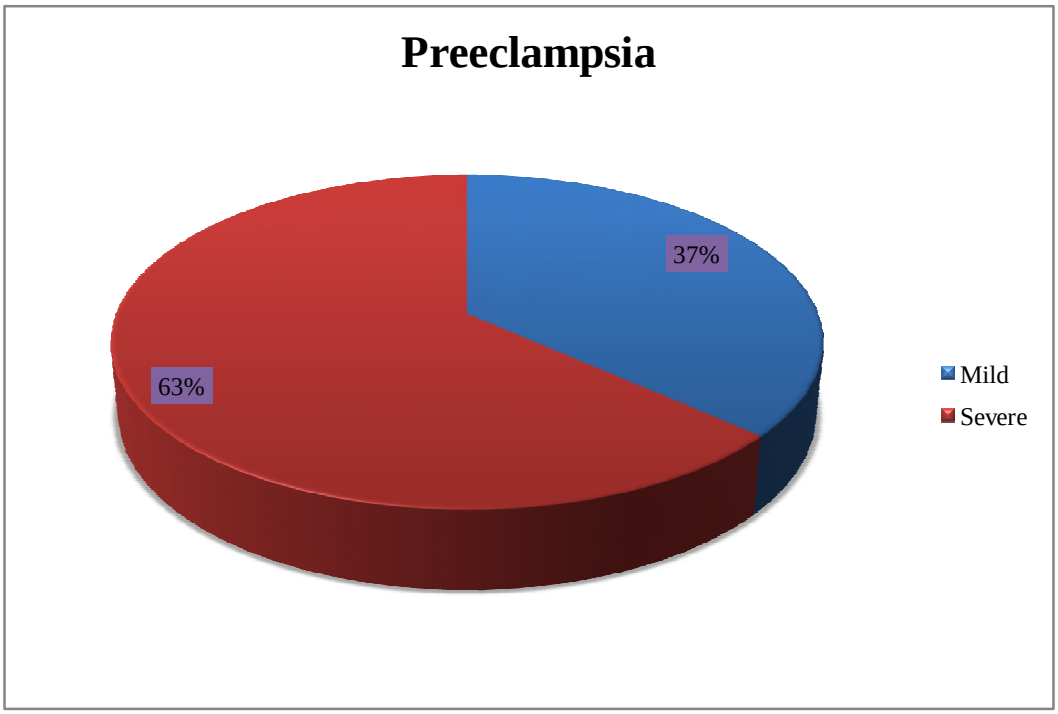


Figure 3.4: Distribution of pre-eclampsia patients according to degree of preeclampsia.

CHAPTER FOUR

DISCUSSION, CONCLUSION, AND RECOMMENDATIONS

Chapter four

Discussion, Conclusion, and Recommendations

Discussion:

This case-control study involved 120 Sudanese pregnant women (60 with preeclampsia and 60 disease-free) who attended three maternity hospitals in Khartoum State during the study period.

The outcome of this study is that primigravida women had twice time chance to develop pre-eclampsia than multigravida women (Odds ratio 1.9). Family history of pre-eclampsia was present among the families of 30.8% of the cases, compared with only 4.2% of the control.

Family history of pre-eclampsia was considered as one of the main predisposing factors of developing or experiencing pre-eclampsia in the future and that was statistically proved in this study where the computed Odds ratio, as a risk factor was found to be very high (17.7). These findings agreed with controlled cohort studies which showed that the risk of pre-eclampsia was increased in women with nulliparity (Odds ratio 2.91,1.28 to 6.61) and family history of pre-eclampsia(Odds ratio 2.9, 1.7 to 4.93) (Duckitt and Harrington, 2005).

Proteinuria at the range of (++ - +++) was present among all cases where level of proteinuria of (+++) was dominant among cases (50%). Zhuang *et al* (2015) found that 84% of cases had proteinuria ≥ 0.3 g/ 24 hour (P value 0.000) which prove that proteinuria is an important diagnostic criterion for pre-eclampsia.

In this study it was found that the difference between the two means of mean platelet volume (MPV) among the cases and control was highly significant (P.value 0.00). On contrast, a study done in Turkey found that increased mean platelet volume (MPV) was not a significant predictor for pre-eclampsia and did not differ both between mild and severe pre-eclampsia, and pre-eclamsia and normotensive pregnant women (Altinbas *et al.*, 2012), whereas we were agreed

with another study which provided evidence that MPV gradually increases in pregnant women affected by pre-eclampsia compared to women with normal pregnancies (Dundar *et al.*, 2008).

Interestingly, there was significant difference between the two means of platelet distribution width (PDW) among the cases and control (P.value 0.00) and among the severe and mild cases (P.value 0.008). This is similar to a study done in Korea which suggested that PDW can serve as a candidate marker for predicting the severity of pre-eclampsia (Yang *et al.*, 2014). The same study proved that as the disease progresses, there is a decrease in platelet count which agrees with our study in which there was significant difference between the two means of platelet count among cases and control (P.value 0.00) and among severe and mild cases (P.value 0.02). Furthermore, red blood cell to platelet ratio (RPR) was found to be significantly high among patient with severe pre-eclampsia (P. value 0.002), but this was not found in literature review, whereas, the difference between the means levels of platelet to lymphocyte ratio (PLR) of mild and severe type of pre-eclampsia was found to be insignificant (P. value= 0.122), this agrees with Yavuzcan *et al* (2014) who found that PLR is not an effective marker for severe PE (P. value= 0.098).

Pre-eclampsia was found to be weakly and negatively associated with platelets count. Weak positive correlations were found between pre-eclampsia and both mean platelets volume (MPV) and platelets large cell ratio (P-LCR). So, low platelets count can be considered as an indicator of severe pre-eclampsia. This agrees with a study in Brazil which found that lower platelets counts were observed in severe pre-eclampsia compared to normal pregnant and to nonpregnant women (Freitas *et al.*, 2013). On the other hand we agreed with Boriboonhirunsarn *et al* (1995) who suggested that MPV in severe preeclamptic women is statistically greater than in normal pregnant women. Also we agreed whith Han *et al* (2014) who found that there was an increase in the mean platelet volume (MPV) in patients with pre-eclampsia and suggested that mean

platelet volume (MPV) may serve as an early monitoring marker for the severity of pre-eclampsia.

Conclusion:

1- Age, educational level and family history of pre-eclampsia were not associated with the degree of pre-eclampsia.

2- Primigravida ladies were more likely to develop pre-eclampsia than multigravida ones.

3- Family history of pre-eclampsia was found to be associated with the development of pre-eclampsia.

4- The difference between the two means of platelet count, mean platelet volume (MPV), platelet distribution width (PDW), and platelets large cell ratio (P-LCR), for cases and control were found to be highly significant.

5- A weak correlation was found between the degree of pre-eclampsia and platelet count (negative), mean platelet volume and platelet large cell ratio (positive).

6- The mean level of red blood cells to platelets ratio (RPR) was found to be significantly high among pregnant women with severe pre-eclampsia while platelet to lymphocyte ratio (PLR) was not.

Recommendations:

1- More prenatal care should be taken especially with primigravida women and pregnant women who have family history of the disease.
2- In women with pre-eclampsia, the platelet count can be used as a valid measurement tool to predict the progress of the disease.
3- Measurement of Platelet count and indices (MPV, PDW, and P-LCR) can be used as a useful marker for the prediction of pre-eclampsia.
4- Further community-based studies with large sample size are recommended to verify these findings.
5- Follow up for pre-eclamptic women after delivery should be taken and also for their babies. Neonatal intensive care should be found in all public hospitals.

REFERENCES

References:

Ali A.A. and Adam I. (2011). Lack of antenatal care, education, and high maternal mortality in Kassala hospital, eastern Sudan during 2005-2009. J Matern Fetal Neonatal Med. 24(8): 1077-8.

Ali A.A., Okud A., Khojali A. and Adam I. (2012). High incidence of obstetric complication in Kassaa Hospital, Eastern Sudan. Journal of Obstet Gynaecol. 32(2): 148-9.

Altınbas S., Togrul C., Orhan A., Yucel M. and Danısman N. (2012). Increased MPV is not a significant predictor for preeclampsia during pregnancy. Journal of Clinical Laboratory Analysis. 26(5): 403-6.

Bain B.J. (2006). Blood Cells - A Practical Guide. 4thEd. Malden: Blackwell: 242.

Bath P.M. and Butterworth R.J. (1996). Platelet size: measurement, physiology and vascular disease. Blood coagulation & fibrinolysis: an international journal in haemostasis and thrombosis, 7(2): 157-61.

Boriboonhirunsarn D., Atisook R., and Taveethamsathit T. (1995). Mean platelet volume of normal pregnant women and severe preeclamptic women inSiriraj Hospital. Journal of Medical Association of Thailand. 78(11): 586-9.

Briggs C., Harrison P. and Machin S.J. (2007). Continuing developments with the automated platelet count. International Journal of Laboratory Hematology. 29(2): 77- 91.

Ceyhan T., Beyan C., Başer I., Kaptan K., Güngör S. and Ifran A. (2006). The effect of pre-eclampsia on complete blood count, platelet count and mean platelet volume. Annals of Hematology. 85(5): 320-22.

Dacie J.V and Lewis S.M. (1995). Practical Haematology. 8th Ed. New York: Churchill. Livingstone: 12.

Duckitt K. and Harrington D. (2005). Risk factors for pre-eclampsia at antenatal booking: systematic review of controlled studies. BMJ (Clinical research ed.). 330(7491): 565.

Duley L. (2009). The Global Impact of Pre-eclampsia and Eclampsia. Seminars in Perinatology. 33(3):130-37.

Dundar O., Yoruk P., Tutuncu L., Erikci A.A., Muhcu M., Ergur A.R., Atay V.and Mungen E. (2008). Longitudinal study of platelet size changes in gestation and predictive power of elevated MPV in development of pre-eclampsia. Prenatal Diagnosis. 28(11):1052-6.

Eiland E., Nzerue C. and Faulkner M. (2012). Preeclampsia 2012. Journal of Pregnancy, 2012: 586578.

Elhaj E.T., Adam I., Alim A., Elhassan E.M. and Lutfi M.F. (2015). Thyroid Function/Antibodies in Sudanese Patients with Preeclampsia. Front Endocrinol (Lausanne). 6:87.

Fragento R.Y. (2006) The High-Risk Obstetric patient. In: Braveman F.R.(ed.). Obstetric and Gynecologic Anesthesia : The requisites. Philadelphia: Elsevier Mosby: 79-80.

Freitas L.G., Alpoim P.N., Komatsuzaki F., Carvalho M.d.and Dusse L.M. (2013). Preeclampsia: are platelet count and indices useful for its prognostic? Hematology (Amsterdam, Netherlands). 18(6): 360-4. Available at: http://www.ncbi.nlm.nih.gov/pubmed/23676885.

Han L., Liu X., Li H., Zou J., Yang Z., Han J., Huang W., Yu L., Zheng Y. and Li L. (2014). Blood coagulation parameters and platelet indices: changes in normal and preeclamptic pregnancies and predictive values for preeclampsia. PLoS One, 9(12):e114488.

Harmening D.M. (2005). Modren Blood Banking and transfusion practices.5th Ed. Philadelphia: F.A.Davis company: 14.

Hoffbrand A.V., Moss P.A.H,and Pettit J.E. (2006). Essential Hematology. 5th Ed. Malden: Blackwell: 269,352-4.

Hoffbrand A.V., Catousy D., Tuddenham E.G.D. and Green A.R. (2011). Postgraduate Haematology. 6th Ed. Oxford, UK: Wiley-Blackwell: 773.

Hubbard J.D. (2010). A concise Review of Clinical Laboratory Science. 2nd Ed. Philadelphia: Lippincott Williams& Wilkins: 52-9.

Jeyabalan A. (2013). Epidemiology of preeclampsia: Impact of obesity. Nutrition Reviews, 71(1): 18-25.

Levine R.J., Maynard S.E., Qian C., Lim K.H., England L.J., Yu K.F., Schisterman E.F., Thadhani R., Sachs B.P., Epstein F.H., Sibai B.M., Sukhatme

V.P.and Karumanchi S.A. (2004). Circulating angiogenic factors and the risk of preeclampsia. The New England journal of medicine. 350 (7): 672-83.

McCrae K. R. (2007) Thrombocytopenia in Pregnancy. In : Michelson A.D.(ed.).Platelets. 2nd ED. London: Elsevier: 928-30.

Norwitz E .R., Hsu C.D. and Repke J.T. (2002). Acute complications of preeclampsia. Clinical obstetrics and gynecology, 45(2): 308-29.

Provan D., Singer C.R.J., Baglin T. and Lilleyman J. (2004). Oxford Handbook of Clinical Haematology. 2nd Ed. New York: Oxford University Press: 370.

Queenan J.T., Hobbins J.C. and Spong C.Y. (2010). Protocol for High -Risk Pregnancies: An Evidence Based Approach. 5th Ed. London: Wiley- Blackwell: 422.

Rajasekhar A., Gernsheimer T., Stasi R. and James A.H. (2013). Clinical Practice Gide on Thrombocytopenia in Pregnancy. Washington, DC: American society of hematology: 2.

Ramin S.M. (2002). ACOG practice bulletin. Diagnosis and management of preeclampsia and eclampsia. Number 33, January 2002. American College of Obstetricians and Gynecologists. International Journal of Gynaecology and Obstetrics, 77(1): 67-75. Available at: http://www.ncbi.nlm.nih.gov/pubmed/21860314\nhttp://www.ncbi.nlm.nih.gov/pubmed/12094777

Redman C.W. and Sargent I.L. (2005). Latest advances in understanding preeclampsia. Science (New York, N.Y.). 308(5728): 1592- 4.

Rodger L.B., Frenkel E., Baker W. and Sorode R. (ed.) (2006) Hematological Complications in Obstetrics, Pregnancy, and Gynecology. New York: Cambridge University Press: 495.

Schroeder B.M. (2002). ACOG practice bulletin. Diagnosis and management of preeclampsia and eclampsia. Am Fam Physician. 66(2): 330-1.

Shaughnessy D.O., Makris M. and Lilllicarp D. (2005). Practical Hemostasis and thrombosis. Malden: Blackwell: 3.

Sibai B.M. (2010). Preeclampsia. In: Queenan J. T., Hobbins J. C. and Spong C. Y.(ed.). Protocol for High-Risk Pregnancies. 5th Ed. West Sussex, UK: Welly-Blackwell: 414.

Sysmex Corporation (2004). Automated Hematology analyzer: Operator Manual. Kobe, Japan: Sysmex Corporation.

Vagdatli E., Gounari E., Lazaridou E., Katsibourlia E., Tsikopoulou F.and Labrianou I. (2010). Platelet distribution width: A simple, practical and specific marker of activation of coagulation. Hippokratia. 14(1): 28-32.

Yang S.W., Cho S.H., Kwon H.S., Sohn I.S.and Hwang H.S. (2014). Significance of the platelet distribution width as a severity marker for the development of preeclampsia.

Yavuzcan A., Caglar M., Ustun Y., Dilbaz S., Ozdemir I., Yildiz E., Ozbilgeç S., and Kumru S. (2014). Mean platelet volume, neutrophil-lymphocyte ratio

and platelet-lymphocyte ratio in severe preeclampsia. Ginekol Polska. 85(3): 197-203.

Young B.C., Levine R.J. and Karumanchi S.A. (2010). Pathogenesis of preeclampsia. Annual review of pathology. 5: 173-192.

Zhuang X., Chen Y.Y., Zhou Q. and Lin J.H.(2015). Qualitative Analysis of Diagnostic Value of 24-h Proteinuria for Preeclampsia. . Chinese Medical Journal. 128(22): 2998-3002.

APPENDICES

Appendix I

Sudan University of Science and Technology

College of Graduate Studies

Questionnaire

Platelets Count and Indices as possible predictors for pre-eclampsia in Sudanese Women in Khartoum State Maternity Hospitals

1- Serial Sample No: ☐☐☐

2- Age: ☐☐

3- Residence: ☐

Khartoum: 1

Outside Khartoum: 2

4- Education: ☐

Illiterate: 1

Basic: 2

Secondary: 3

University: 4

5- Occupation: ☐

House wife: 1

Teacher: 2

Laborer: 3

Others: 4

6- Gravid ☐☐

7- Parity ☐☐

8- Gestational Age: ☐

First trimester: 1

Second trimester: 2

Third trimester: 3

Postpartum: 4 ☐

9- History of Hypertension:

Yes 1

No 2 ☐

10- History of Diabetes mellitus:

Yes 1

No 2 ☐

11- Other Chronic or Infectious Diseases:

Yes 1

No 2 ☐

12- Family History of Preeclampsia:

Yes 1

No 2

13- Blood Pressure: ……

14- Proteinuria: ……

Appendix II

بسم الله الرحمن الرحيم

جامعة السودان للعلوم والتكنولوجيا

كلية الدراسات العليا – برنامج الماجستير – مختبرات طبية

تخصص علم أمراض الدم ومبحث المناعة الدموية

إقرار موافقة علي إجراء بحث علمي

عدد ومؤشرات الصفائح الدموية كعوامل تنبؤ لضغط الدم الجنيني لدي السودانيات بمستشفيات الولادة في ولاية الخرطوم

الإسم:.....

سوف يتم أخذ عينة من الدم (2.5 مل) من الوريد بواسطة حقنة طعن وذلك بعد مسح منطقة أخذ العينة بواسطة المطهر. كل الادوات المستخدمةلأخذ العينة معقمة ومتبع فيها وسائل السلامة المعملية .

وانا اقر بأن هذه العينات سيتم تحليلها فقط لطلب البحث وأن إسمك سيبقي طي الكتمان.

أوافق انا المذكور أعلاه أخذ عينة لإجراء الدراسة

الإمضاء : .....

التاريخ: .....

تلفون : .....

Appendix III

بسم الله الرحمن الرحيم

Ministry of Health
Khartoum State
Directorate of
Research

Date:1/4/2015

النمرة وخ/وص/اع /اب

Ethical Clearance

This is to certify that the research entitled:

"measurement of platelet count and indices among Sudanese women with preeclampsia in Khartoum state maternity hospitals ", had been ethically approved by the ethical committee at the Ministry of Health, Khartoum State.
The principal investigator is : Faiza Ali Nugud Mohammed

Dr. Abdel Rahman Al Asha

Director of Research Department

Appendix IV

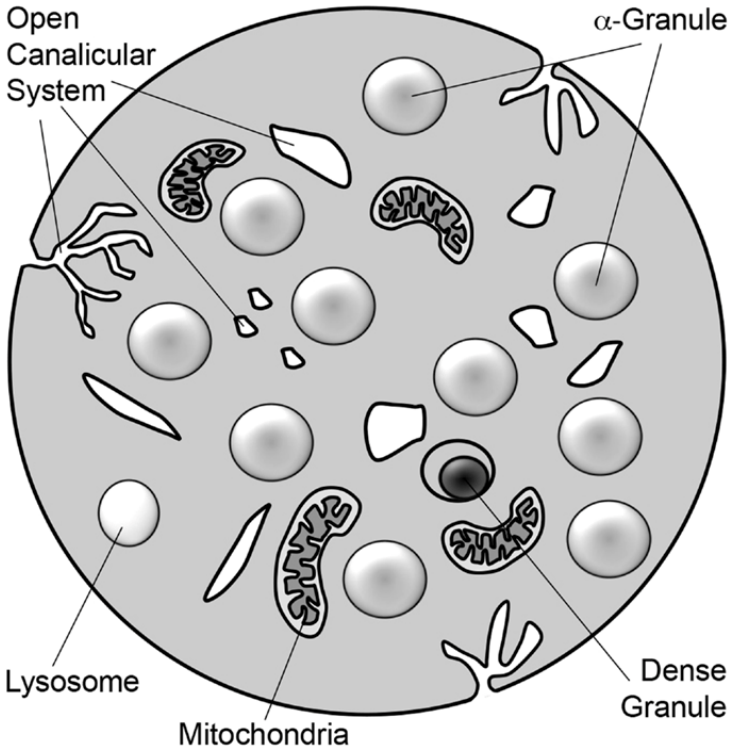
Reference range:

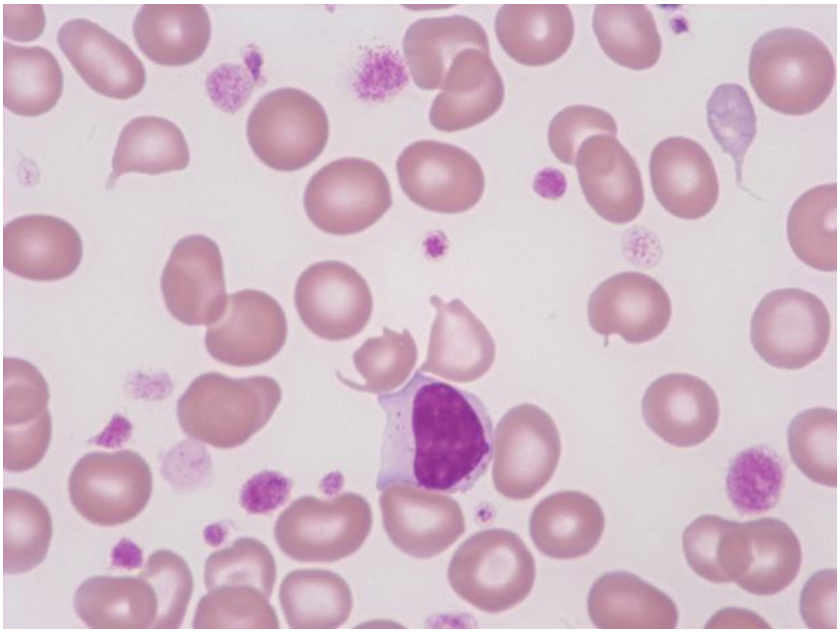
WBC (× 10^3/µl)	4.0 - 10.0
RBC (× 10^6/µl)	3.8 - 4.8
HB (g/ dl)	12.0 - 15.0
HCT (%)	36 - 46
MCV (fl)	83 – 101
MCH (pg)	27 – 32
MCHC (g/dl)	31.5 – 34.5
PLT (× 10^3/µl)	150 – 400
LYMPHOCYTE (%)	20 – 40
MXD (%)	2 – 10
NEUTROPHIL (%)	40 – 80
ABSOLUTE YMPHOCYTE (× 10^3/µl)	1.0 – 3.0
ABSOLUTE MXD (× 10^3/µl)	0.2 -1.0
ABSOLUTE NEUTROPHIL (× 10^3/µl)	2.0 – 7.0
RDW-SD (fl)	39 – 46
RDW-CV (%)	11.6 – 14
PDW (fl)	9.4 - 18.1
MPV (fl)	8.5 – 12.4
P-LCR (%)	14.3 – 44

(Dacie, 1995; Sysmex Corporation 2004)

Appendix V

Platelet structure:





Thrombocytosis, anisocytosis of the platelets and giant platelets

Sysmex KX-21N model- Japan.