Chapter one

Introduction and literature Review

1.1 Introduction:

Platelets are extremely small and discoid, 3.0×0.5 mm in diameter ,with mean volume 7-11 fl. Produced in bone marrow by fragmentation of the cytoplasm of megakaryocyte, one of the largest cell in the body .

the normal platelet count is approximately 250×10^9cell/L(range 150-400×10^9 cell/L) and the normal platelet life span is 7-10 days (Hoffbrand ,2006).

The platelet is responsible for initiation of the hemostatic mechanisms that repair injury to the vascular endothelium. The four major platelet functions include Platelet adherence, Platelet activation and secretion, Platelet aggregation ,Interaction with coagulation factors (Deutsch and Tomer , 2006).

Malaria is a parasitic disease that involves high fevers, shivering chills, flu-like symptoms and anemia. This infection is caused by a parasite known as Plasmodium, transmitted by the Anopheles **mosquitoes**. After infection, the parasites enter the bloodstream and infect the red blood cells. The parasites multiply inside the red blood cells, which then break open to infect more red blood cells. The first symptoms typically occur 10 days to 14 days after the infection sets in.

Malaria is caused by one of plasmodium spices ;plasmodium falciparum ,p.vivax ,p.ovale and p. malaria.

Malaria incidence in Sudan was estimated to be about 9 million episodes in 2002 and the number of deaths due to malaria was about 44,000. 2,877,000 dalys were lost in sudan in 2002 due to malaria mortality, episodes, anaemia and neurological

sequelae. children under five years of age had the highest burden. Males had the highest incidence and mortality, but females lost more DALYs (WHO,2003).

Platelets play a critical role in the pathogenesis of malarial infections by encouraging the sequestration of infected red blood cells within the cerebral vasculature. But platelets also have well-established roles in innate protection against microbial infections. **(Hoffman,1995*).***

1.2 Literature review

1.2.1 Blood Platelets

1.2.1.1 Pletlate production

Platelets are anucleate, discoid cells, roughly 2–3 μm in diameter that function primarily as regulators of hemostasis, but also play secondary roles in angiogensis and innate immunity. Although human adults contain nearly one trillion platelets in circulation that are turned over every 8–10 days, our understanding of the mechanisms involved in platelet production is still incomplete. Platelets stem from large (30–100 μm) nucleated cells called megakaryocytes that reside primarily in the bone marrow,production of

Plateles is control by thrombopoietin *(Harrison et al, 1989).*

1.2.1.2 Pletlate structure

platelets are anuclear cytoplasmic fragments, they contain a number of organelles, such as mitochondria, microtubules, Dense Body, etc. These organelles can be divided into three defined zones that possess a unique function *(Harrison et al , 1989)*.

The three zones of organelles is :

1-Peripheral zone:

Responsible for platelet adhesion and aggregation and consist from :

A- Glycocalyx which Contains glycoprotein receptors:

- GPIb binds von Willebrand's factor needed for platelet adhesion to collagen.
- GPIIb/IIIa bind fibrinogen needed for aggregation.
- Bind ADP and thrombin, promoting aggregation.
- Factors I, V, VIII on surface, involved in 2^{o} hemostasis *(Harrison, et al ,1989).*

B- Plasma membrane:Exposed on platelet activation.

- Layer called PF3 (platelet factor) surface for interaction of plasma coagulation factors. (Levin , 1997).

- Initiation of formation of thromboxane $A_{2.}$ This stimulates aggregation and vasoconstriction (Levin , 1997).

2- Sol-Gel Zone:

The term Cytoskeleton is often used to describe this Structural or Sol-Gel zone: Responsible for platelet retraction/contraction functions and platelet shape (Machlus , Thon , 2014).

3-Organelle zone:

- Responsible for storage and platelet release functions and metabolic activities of platelets, consist from :

- Granules (Dense bodies, alpha granules, lysosomal granules and microperoxisomes.)

- Mitochondria.

- Glycogen.

Platelets contain three morphologically distinct types of storage granules; Alpha granules, dense granules, lysosomes containing acid hydrolase (Machlus , Thon , 2014).

The contents of the dense body granules are**;**

-Adenosine diphosphate (ADP).

-Adenosine triphosphate (ATP).

-Calcium.

-Catecholamines (epinephrine, norepinephrine).

-Serotonin.

-Pyrophosphate.

-Magnesium *(De Botton et al ,2002).*

The content of Alpha Granules are :-

- Fibrinogen
- Fibronicotin
- plasminogen
- platelet factor 4 (Pf4)
- platelet drived Growth factor (PDGF)
-Von willbrand factor (vWF)*(De Botton et al,2002).*

1.2.1.3 Pletlate function

- Surveillance of blood vessel continuity
    1-Checks endothelial lining for gaps and breaks
    2-Fill-in small gaps caused by separation of endothelial cells
- Formation of primary hemostatic plug
- Surface for coagulation factors to make secondary hemostatic plug
- Aid in healing injured tissue
- Provides the reaction surface for some coagulation system reactions, as well as platelet factor 3 (PF_3) which is platelet phospholipid (Kuter ,1996).

1.2.1.4 Formation of primary heamostatic plug

- Once the platelets "normal" environment is changed, they become activated or adhesive (Kuter , 1996).

Three stages of plug formation:

- Stage 1: pletlate adhesion

Platelets attach to non-platelet surfaces, such as collagen fibers in the sub endothelium then Platelets move from the blood vessels and into the tissues and Exposed to surfaces in the tissues causes them to bind to collagen with the presence of von Willebrand factor (vWF) and glycoprotein IbIX, making a bridge formation, which triggers a shape change

- Stage 2: pletlate aggregation

Chemical changes cause platelets to aggregate and stick to one another.

Newly arriving platelets become activated by agonists ,exposed of GPIIb/IIIa sites bind fibrinogen then Fibrinogen + activated platelets serves as a bridge between two platelets ,Calcium must be present.

- Stage 3:pletlate secreation and release.

Requires ATP, Platelets release contents of their granules, causing vasoconstriction Granules trigger a secondary aggregation which is irreversible.

- Final stage : stabilization of clot

primary hemostatic plug formation , thrombus formation , platelets release Factor V , expose factor III, accelerating coagulation cascade , promote activation of clotting factors (Hoffbarnd *et al*.,2006).

1.2.2 Platelet Indices

Platelet indices are potentially useful markers for the early diagnosis of thromboembolic diseases.

Platelet indices is : mean platelet volume (MPV) ,platelet distribution width (PDW) and plateletcrit (PCT).

An increase in both mean platelet volume (MPV) and platelet distribution width (PDW) due to platelet activation, was suggested due to : Platelet swelling ,Pseudopodia formation .

MPV and PDW are simple platelet indices, which increase during platelet activation *(Machlus et al , 2014).*

1.2.2.1 Mean Platelet Volume (MPV)

MPV is a machine-calculated measurement of the average size of platelets found in blood and is typically included in blood tests as part of the CBC .

A- MPV is higher when there is destruction of platelets.

- This may be seen as in inflammatory bowel disease.
- In immune thrombocytopenic purpura (ITP).
- In myeloproliferative diseases and Bernard-Soulier syndrome.
- It may also be related to pre-eclampsia, and recovery from transient hypoplasia.

B- Low MPV values seen in :

- Primarily with thrombocytopenia when it is due to:
- Impaired production as in aplastic anemia.

Standrd MPV range is 7.5 to 13 fimtoliter (fl) *(Bessman et al ,1981).*

Platelet size correlates with platelet reactivity; larger platelets have greater prothrombotic potential. Elevated platelet size (mean platelet volume) is associated with increased platelet aggregation, increased expression of adhesion molecules,

and elevated risk of cardiovascular and peripheral arterial diseases(Bath and Butterworth ,1996).

Interestingly, a study of patients with acute coronary disease found a direct association between α_2-integrin chain expression and mean platelet volume, suggesting that expression levels of integrin $\alpha_2\beta_1$ may be involved in the regulation of platelet size (***Kunicki et al , 2012***).

1.2.2.2 Platelet distribution width (PDW)

PDW is simple, practical and specific marker of activation of coagulation.

PDW is a more specific marker of platelet activation, since it does not increase during simple platelet swelling (martin *et al*.,1998).

PDW has been receiving attention due to its usefulness for distinguishing between reactive thrombocytosis and thrombocytosis associated with myeloproliferative disorder (Martin *et al*., 1998).

Determination of the PDW reference range is fundamental Comment in

Mean platelet volume (MPV) and platelet distribution width (PDW) are useful in the differential diagnosis of aplastic anemia and idiopathic thrombocytopenic purpura , an increased PDW is an indication for the anisocytosis of platelets.

Standard PDW ranges from 9 to 14 fL *(martin et al.,1998*).

1.2.2.3 Platelet crit (Pct)

Plateletcrit is an effective screening tool for detecting platelet quantitative abnormalities (martin *et al.*,1998).

Normal platelet count have a Plateletcrit within the range of 0.20-0.36%.

1.2.2.4 Normal Values

- The average of Mean platelet volume was 8.13 fl.

-Plateletcrit- 0.23%.

-Platelet distribution width – 56.6% in normal platelet counts.

-Sensitivity and specificity for detection of :

-Thrombocytopenia at Plateletcrit cut off range of 0.20 – 0.36% is 97 and 80%-Thrombocytosis is 94 and 98% *(Martin et al.,1998).*

1.2.3 Malaria

Malaria is one of the most common **infectious** diseases and a great public health problem worldwide, particularly in Africa and south Asia. yearly, there are an estimated 250 million cases of malaria leading to approximately one million deaths, mostly in children under five years of age. The organism that causes the most dangerous form of malaria is a microscopic **parasite** called plasmodium falciparum (Talaro, 2012).

1.2.3.1 Plasmodium species

There are four type of plasmodium species that cause Malaria in humen:-

A- Plasmodium falciparum

Plasmodium falciparum is a **protozoan** **parasite**, one of the species of ***Plasmodium*** that cause **malaria** in humans. It is transmitted by the female ***Anopheles*** **mosquito**. Malaria caused by this species (also called malignant or falciparum malaria) is the most dangerous form of malaria, with the highest rates of complications and mortality. As of the latest **World Health Organization** report in 2014, there were 198 million cases of malaria worldwide in 2013, with an estimated death of 584,000 (WHO , 2014).

It is much more prevalent in sub-Saharan Africa than in many other regions of the world; in most African countries, over 75% of cases were due to *P. falciparum*, whereas in most other countries with malaria transmission, other, less virulent plasmodiam species predominate. Almost every malarial death is caused by *P.* falciparum (WHO , 2008).

B- Plasmodium vivax

Plasmodium vivax is a **protozoal parasite** and a **human pathogen**. The most frequent and widely distributed cause of recurring (Benign tertian) **malaria**, P. vivax is one of the four species of **malaria** parasites that commonly infect humans It is less virulent than **Plasmodium falciparum**, the deadliest of the four. but vivax malaria can lead to severe disease and death due to **splenomegaly** (a pathologically enlarged **spleen)**(Davidson , 2004).

C- Plasmodium ovale

Plasmodium ovale is a species of **parasitic protozoa** that causes tertian **malaria** in humans. It is one of several species of Plasmodium parasites that infect humans including Plasmodium falciparum and Plasmodium vivax which are responsible for most malarial infection. It is rare compared to these two parasites, and substantially less dangerous than P. falciparum. (Davidson , 2004).

P. ovale has recently been shown by genetic methods to consist of two subspecies, **P. ovale curtisi** and **P. ovale wallikeri**.

D- Plasmodium malaria

Plasmodium malarie is a **parasitic protozoa** that causes **malaria** in humans. While found worldwide, it is a so-called "benign(4) malaria" and is not nearly as dangerous as that produced by *P. falciparum* or *P. vivax*. It causes **fevers** that recur at approximately three-day intervals (a *quartan fever*), longer than the two-day (tertian) intervals of the other malarial parasite , hence its alternate name is quarten fever and quarten malaria (Davidson , 2004).

1.2.3.2 Life cycles:

*Plasmodium life cycle h*as two hosts: mosquitoes and humans.

Sexual reproduction takes place in the mosquito and the parasite is transmitted to humans when the mosquito takes a blood meal *(Kayser et al , 2005).*

A- life cycle in humen

The mosquito injects *Plasmodium* into a human in the form of *sporozoites*.

The sporozoites first invade liver cells and asexually reproduce to produce huge numbers of merozoites which spread to red blood cells where more merozoites are produced through more asexual reproduction.

Some parasites transform into sexually reproducing *gametocytes* and these if ingested by a mosquito continue the cycle (*Kayser et al , 2005).*

B- Life cycle in mosquitoes:

Gametocytes ingested by a mosquito combine in the mosquito's stomach to produce zygotes , These zygotes develop into motile elongated *ookinites*.

The ookinites invade the mosquito's midgut wall where they ultimately produce sporozoites, which make their way to the salivary glands where they can be injected into a new human *(Kayser et al , 2005*).

1.2.3.3 Pathophysiology:

-Liver Stage **:** Human infection is initiated when sporozoites are injected with the saliva during mosquito feeding. The sporozoites enter the circulatory system and within 30-60 minutes will invade a liver cell. Host cell entry, as in all apicomplexa, is facilitated by the apical organelles. After invading the hepatocyte, the parasite undergoes an asexual replication. This replicative stage is often called exoerythrocytic (or pre-erythrocytic) schizogony (*Hoffman et al ., 2004*).

In *P. vivax* and *P. ovale* some of the sporozoites do not immediately undergo asexual replication, but enter a dormant phase known as the hypnozoite. This hypnozoite can reactivate and undergo schizogony at a later time resulting in a relapse. *(Hoffman et al ., 2004).*

-Blood Stage : Merozoites released from the infected liver cells invade erythrocytes. The merozoites recognize specific proteins on the surface of the erythrocyte and actively invade the cell in a manner similar to other apicomplexan parasites.

After entering the erythrocyte the parasite undergoes a trophic period followed by an asexual replication. The young trophozoite is often called a ring form due to its morphology in Geimsa-stained blood smears. As the parasite increases in size this 'ring' morphology disappears and it is called a trophozoite. During the trophic period the parasite ingests the host cell cytoplasm and breaks down the hemoglobin into amino acids. A by-product of the hemoglobin digestion is the malaria pigment, or hemozoin.These golden-brown to black granules have been long recognized as a distinctive feature of blood-stage parasites**.** *(Hoffman et al ., 2004).*

-Sexual Stage: As an alternative to schizogony some of the parasites will undergo a sexual cycle and terminally differentiate into either micro- or macrogametocytes. Gametocytes do not cause pathology in the human host and will disappear from the circulation if not taken up by a mosquito.

Gametogenesis, or the formation of micro- and macrogametes, is induced when the gametocytes are ingested by a mosquito. After ingestion by the mosquito, the microgametocyte undergoes three rounds of nuclear replication. The macrogametocytes mature into macrogametes. (*Hoffman et al ., 2004).*

The highly mobile microgametes will seek out and fuse with a macrogamete. Within 12-24 hours the resulting zygote develops into an ookinete. The ookinete is a motile invasive stage which will transverse both the peritrophic matrix and the midgut epithelium of the mosquitos.

Sporogony. After reaching the extracellular space between the epithelial cells and the basal lamina, the ookinete develops into an oocyst. The oocysts undergo an asexual replication, called sporogony, which culminates in the production of

several thousand sporozoites. This generally takes 10-28 days depending on species and temperature. Upon maturation the oocyst ruptures and releases the sporozoites which cross the basal lamina into the hemocoel (body cavity) of the mosquito (*Hoffman et al ., 2004).*

1.2.3.4 Symptoms of malaria:

A malaria infection is generally characterized by recurrent attacks with the following signs and symptoms:

-Moderate to severe shaking chills

-High fever

-Profuse sweating as body temperature falls

Other signs and symptoms may include:

-Headache ,vomiting ,diarrhea

Malaria signs and symptoms typically begin within a few weeks after being bitten by an infected mosquito. However, some types of malaria parasites can lie dormant in the body for months, or even years.

More severe symptoms of malaria might include:

-Hepatomegaly ,splenomegaly.,anemia ,jaundice ,dehydration (Kumar and clarek,1998).

1.2.3.5 Diagnosis of malaria:

- Thick and thin blood smears
- Rapid diagnostic tests (antigen testing)
- Molecular tests (**Polymerase chain reaction, PCR**)
- Antibody tests (serology)(WHO , 2006).

1.2.3.6 Effect of malaria on the immune system

Plasmodium parasites exist in various forms within the liver and blood but manage to escape from the immune system. This is because in most of its forms it resides within the liver and blood cells and is relatively invisible to immune surveillance.

Normally the RBCs undergo destruction in the spleen at regular intervals. Infected RBCs especially those with *plasmodium falciparum* escape this destruction by developing adhesive proteins on the surface of the infected blood cells, causing the blood cells to stick to the walls of small blood vessels. This leads to sequestering the parasite from passage through the general circulation and the spleen.

These proteins are also thought to be the cause of complications caused by this type of malaria parasite. They are called PfEMP1, for *Plasmodium falciparum* erythrocyte membrane protein 1 and have a variety and diversity and thus cannot be targeted by the antibodies formed in the body *(Spring et al, 2009)*.

1.2.3.7 Effect of Malaria on platelet

Platelets, as well as regulating blood hemostasis, are an important component of the body's defense against invading microbial pathogens. platelets protect during malaria infection by binding Plasmodium-infected erythrocytes (IE) and killing the parasite within. more recent studies have now revealed the platelet plasmocidal factor, platelet factor 4 (PF4) and the red cell-expressed Duffy-antigen molecule as the central players in the parasite killing activity of platelets *(Hoffman et al , 2004)*.

1.2.4 The Protective Role for platelets in Malaria infection

In addition to their well-defined role in hemostasis, platelets are increasingly implicated in immunological processes, including direct pathogen-killing functions . Platelets share many properties with classical immune cells. They express receptors that bind host immune response modulators (e.g., antibodies and cytokines) and Toll-like receptors that bind microbial products. They also express the CD154 co-stimulatory molecule and influence the development of adaptive

immune responses. They also produce microbicidal products such as oxygen free-radicals and peptides.Iimportantly, their location in the circulation makes them ideal “sentinels” against any nascent infection. Platelet number and mass exceed that of all leukocytes in the circulation. Platelets respond to a variety of microbial cells by releasing immunomodulatory molecules and by directly killing microbial pathogens ***(Hoffman et al ., 2004).***

1.3 Previous studies:

Conducted study by Mohamed abdelrahman about evaluation of platelets count and platelet indices in patients with acute malaria in shendi locality, (Abdelrahman, 2008)The results of this study showed that 67% of patients with thrompocytopenia an mean platelet volume (MPV), and Plateletcrit (PCT) values were exhibited significant decrease, but there was no change in platelet distribution width (PDW).

Other study done in Pakistan which had high percent (85.5) % of patients with thrompocytopenia, The platelet indices in this study showed (90.2) % had low MPV and (81.3) % had low PCT (Sheikh ,2005).

In Nigeria study was done , the results showed that (77%) of patients with thrompocytopenia , (74%) are significant decreased in (MPV and PCT) , no significant diffrence in (PDW) (Marsh .1995) .

In india study was done , there is (81%) of patient with thrombocytopenia and low MPV ,PCT(lal ,2010).

1.4 Objectives

General Objective

To study platelets count and indeces in malaria patients.

Specific Objectives

1-To evaluate Platelets count and indeces (MPV , PDW , PCT) in malaria patients.

4-To study effects of age on platelets count and indeces in malaria patients .

5-To study effects of gender on platelets counts and indeces in malaria patients.

1.5 Rationale

Malaria is a serious, sometimes fatal disease caused by plasmodium species .

Plasmodium falciparum is the most dangerous form of malaria and called malignant malaria .

more than 40% are in risk to malaria in the world and more than 500 million people are infected by malaria yearly.(WHO, 2006).

Platelet are shown to be more active to kill the parasite infected red blood cell . platelets count and indices could be useful in diagnosis of malaria .

Malaria is associated with different degrees of reduction platelet count and alteration of platelet indices and may be used as probable indicator for malaria in endemic regions and therefore encourage the laboratory physicians for more depth search of the parasite microscopiclly .

Chapter Two

Materials and Methods

2.1 Study design

This study is an analytical case control study , conducted between January and April 2015 to evaluate platelet count and indices of malaria patients who visited dongola hospital , Northen state .

2.2 Study population and sample size

Seventy patients with malaria and thirty healthy volunteers as a control group were inrrolled in this study .

2.2.1 Inclusion criteria

patients with malaria attended Dongola Hospital from January to April 2015.

2.2.2 Exclusion Criteria

- Malaria patients under treatment .
- Malaria patients with other diseases .

2.3 Ethical consideration

Ethical clearance was obtained for this study ,and the sample was collected after consent of participants were informed about the procedure of blood collection and the aim and benefits of this study .

2.4 Tools of data collection

Data were collected using a personal interview questionnaire to the patients ,including age , gender , history of disease .

2.5 Samplling

One handred samples was collected from patients and healthy volunteers , 2.5 ml of venous blood was collected from each one using disposable syringes and sprit (70% alcohol) is use to sterilizing the area of collection , the blood is drawn in EDTA containers , measurement of platelets count and indices was determined

within two hours after collection of blood sample using BC -3000 plus auto hematology analyzer

2.6 Determination of platelet counts and indices by auto hematology analyzer (Sysmex)

BC -3000 plus auto hematology analyzer is a three part auto hematology analyzer able to run 19 parameters per sample including : hemoglobin level ,packed cell volume ,red cell concentration ,mean corpuscular hemoglobin ,mean cell volume, mean corpuscular hemoglobin concentration ,white blood cells .

platelets count .mean platelet volume , platelet distribution width , plateletcrit.

2.6.1 Platelets count measurement principle

Platelet is counted and sized by the impedance method , this method is based on the measurement of change in electrical resistance produced by a particle ,which in this case is a blood cell ,suspended in a conductive diluent as it passes through an aperture of known dimensions . an electrodes is submerged in the liquid on both sides of the aperture to create an electrical pathway .as each particle passes through the aperture ,a transitory change in the resistance between the electrodes is produced .this change produces a measurable electrical pulse .the number of pulses generated indicates the number of particles that passed through the aperture . the amplitude of each pulse is proportional to the volume of each particle. Each pulse is amplified and compared to the internal reference voltage channels, which only accepts the pulses of a certain amplified. If the pulse generated is above the platelet lower threshold, it is counted as an platelet .

2.7 Data analysis

Data obtained was analysed by statistical package of social science (SPSS .version -11) soft were program to compare means and P.values at 0.05 by Independent – sample T test.

Chapter Three

Results

The study was conducted to study platelet count and platelet indices in malaria patients who visited dongola hospital in Northen state . the study included handerd subjects , 70 were diagnosed with malaria 40 (57%) of them were males and 30 (43%) of them were females; 30 healthy volunteers were selected as a control 16 (53%)of them were males and 14 (47%) of them were females (Table 3.1).

Table 3.2 showed means and SD of age in patients(mean±SD :44±18.2)years and control group (mean±SD :39±17)years .

Age of 14% of subjects range (0-20) years and 33% (21-40) years and 34% (41-60) years ,16% of study group was (61-80) and 3% (81-100) years (Table 3.3).

Table 3.1 Percent of gender among patients and control groups

		Case	Control
Gender	Male	40 (57%)	16 (53%)
	Female	30(43%)	14 (47%)

Table (3.2) Means and SD of age among patients and control groups

	Case		Control	
Age	Mean	SD	Mean	SD
	44	18.2	39	17

Table (3.3) Distribution of age among study population

Age	Frequency	Percent%
0 - 20	14	%14
40 - 21	33	%33
60 - 41	34	%34
80 - 61	16	%16
100 - 81	3	%3

Table 3.4 showed that Mean platelets count of patients decreased significantly compared to controls (135±16.1×10^9 , 280$\pm$63.9×10^9cell/L P.value 0.000) ,mean platelet volume (MPV) in patients were showed significant decreased in patients compared to control group (7.68±.51 ,8.56$\pm$.62 fl P.value 0.000), platelet distribution width(PDW)was showed no significant increased in patients compared to control group (15.69±.86 , 15.57$\pm$.86 fl P.value 0.595) , Platelet crit (PCT) showed significant decrease in patients compared to control group (0.191±00.06 , 0.235$\pm$0.065% P. value 0.002) .

Table 3.5 showed frequency of thrombocytopenia among malaria patients ,(61.4%) of patients have thrombocytopenia ,(38.6%) were noral platelet count and no one with high platelet count.

Table 3.6 showed no effect of gender on platelet count ,MPV ,PDW and PCT .

Effect of age on platelet count ,MPV ,PDW and PCT showed in table 3.7 ,no correlation between age and platelet count ,MPV ,PDW and PCT .

Table 3.4 The blood platelets count ,MPV ,PDW ,PCT of study Population

Parameter	Groups	Means $\pm$ SD	p .value
Platelet count($\times 10^9$cell/L)	Patients	135 ± 16.1	0.000
	Control	280 ± 63.9	
MPV (fl)	Patients	7.68 ± 0.51	0.000
	Control	8.56 ± 0.62	
PDW (fl)	Patients	15.69 ± 1.05	0.595
	Control	15.57 ± 0.86	
PCT (%)	Patients	0.191 ± 0.067	0.002
	Control	0.235 ± 0.056	

Note :The Mean difference is significant at the P .value $\leq$ 0.05.

Patients number was 70 ,control was 30 subjects

Table (3.5) Frequency of thrombocytopenia among malaria patients

Platelet count	Frequency	Percent
Normal	27	38.6%
Low	43	61.4%
High	0	0
Total	70	100%

Table 3.6 blood platelet count ,MPV ,PDW and PCT accourding gender of patients

Parameter	Gender	Means±SD	P.value

Platelet count($\times 10^9$cell/L)	Males	135±23	0.238
	Femals	135±67	
MPV(fl)	Males	7.26±0.3	0.566
	Females	7.21±0.1	
PDW(fl)	Males	15.3±1.3	0.443
	Females	15.8±0.9	
PCT%	Males	0.192±0.52	0.139
	Females	0.194±0.61	

Table 3.7 effects of patients age on platelet count ,MPV,PDW and PCT

Parameter	Age					P.value
	0-20	21-40	41-60	61-80	81-100	
Platelet	141	138	139	135	141	0.221

count($\times 10^9$cell/L						
MPV (fl)	7.81	7.93	7.91	7.65	7.55	0.301
PDW(fl)	15.6	15.4	15.35	15.81	15.36	0.361
PCT%	0.186	0.189	0.193	0.195	0.187	0.486

Chapter Four

4.1 Discussion

In the present study, the analysis of the platelet count and platelet indices in patients with malaria revealed a high frequency of thrombocytopenia (61.4) and changes in MPV and PCT but no change in PDW. All subjects in this study were

free from other disease can affect on platelets count and platelet indices and did not take any medication .

In the result of current study mean of platelets count in patients is decreased compared to mean of platelets count in control group which consistent with previous studies that mentioned platelets count is deceased in malaria patients .

In this study mean of MPV is decreased in patients group compared to control group that means the malaria is affect on this parameter .

PDW in this study showed no significant differance in mean of patients compared to control group so it is clearly not affected by malaria.

PCT in this study showed decreased in mean of patients compared to control group.

In the result of this study showed that age of patients was not effected on platelet count ,MPV ,PDW and PCT .

The result of present study showed both male and females not effected on platelet count ,MPV ,PDW and PCT.

In this study Thrombocytopenia occurred in (61.4 %) of malarial cases this is lower in comparison to study done in Pakistan which had high percent (85.5%)of thrombocytopenia ,while this result agrees with other study done in shendi (north sudan) which report thrombocytopenia in (67%) of patients and significant decreased in MPV **,**PCT ,no change in PDW .

Other previous studies done in Nigeria (Marsh ,2005) , india (lal ,2010) agrees with this study ,thrombocytopenia ,decreased MPV ,PCT was occurred but PDW showed no significant difference .

4.2 Conclusion

1- platelets count , MPV and PDW of patients were significantly decreased compared to control group , no significant diffreance in PDW between patients and control .

2-Age of patiets were not affected platelets count and indeces.

3 Gender had no effects on platelets count and indeces.

4-Thrombocytopenia was observed among most of the patients .

5-Low Platelet counts and platelet indices can be used as indicator in malaria diagnosis.

4.3 Recomendation

1-Routine investigation of platelet count and platelet indices with investigation of malaria to confermation.

2- prevention against malaria infection is highly recommended throgh appropriate health polities.

3- Further studies recommended with using a large sample size and duration.

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Appindex (1)

Refrance values :

Parameter	Refrance value
Platelet counts	150-450cell/L
MPV	8.5-13 FL
PDW	9-14 FL
PCT	0.20-0.36 %

Appindex (2)

Sudan University of Scince and Technology

Collge of Graduate Studies

Department of Hematology

Study of Platelets Count and Indices in Malaria patients_Northen State

No : ( )

Age : ……

Gender : Male: ( ) Female: ( )

History of disease

Bleeding disease Yes ( ) No ( )

Renal disease Yes ( ) No ( )

Liver disease Yes ( ) No ( )

Cancers : ……..

Others : ……..

Investigation :

Platelets count ( )

MPV ( )

PDW ( )

PCT ( )