Chapter One
Introduction and literature review

1.1General Introduction:

In chronic kidney disease (CKD) Red cell production due to the Erythropoietin deficiency is too low and causes development of anemia in this situation. In addition to anemia, platelet count also seems to be affected by renal disorder too(Akbar ,*et al*.,2013).

In (CKD) both bleeding and thrombotic complications are observed. Early stages of chronic kidney disease are typically associated with a prothrombotic tendency, whereas in its more advanced stage patients also suffer from a bleeding diathesis .Bleeding tendency of patients is characterized by haemorrhagic symptoms and by prolongation of bleeding time. The cause of bleeding in this group of patients has been elaborated in the past and the pathogenesis seems multifactorial. It is suggested that abnormal platelet function Is a major contributor (Esther,*et al.*,2012).

The mean platelet volume (MPV), a readily available indicator of platelet activation and function, is a useful predictive and prognostic biomarker of cardiovascular and cerebrovascular disease (CVD) in chronic kidney disease (CKD) .MPV it is associated with a variety of prothrombotic and proinflammatory diseases. Larger platelets are more likely to aggregate and release greater quantities of adhesive molecule (Ju ,*et al*., 2014).

Platelet large cell ratio (PLCR) Counting of large (young or immature) PLT. Recent evidence has shown that larger platelets are more reactive per unit

Volume than smaller platelets and are more likely to aggregate, leading to thrombosis. Large platelets seem to be an independent risk factor for myocardial infarction, and platelet size is one predictor of recurrent myocardial infarction

And death (Sharpe,*et al.*,1994) .

1.2 Literature review:

1.2.1Haematopoiesis

Is the formation of blood cellular components. All cellular blood components are derived from haematopoietic stem cells. In a healthy adult person, approximately 10^{11}–10^{12} new blood cells are produced daily in order to maintain steady state levels in the peripheral circulation(Tao,*et al*.,2010)

Haematopoietic stem cells (HSCs) reside in the medulla of the bone (bone marrow) and have the unique ability to give rise to all of the different mature blood cell types and tissues. HSCs are self-renewing cells(Morrison,*etal.*,2006).

1.2.1.1 Blood cells are divided into three lineages:

Erythroid cells are the oxygen carrying red blood cells**,** lymphocytes are the cells of adaptive immune system, myelocytes which include granulocytes, megakaryocytes and macrophages and are derived from common myeloid progenitors, are involved in as innate immunity, adaptive immunity, and blood clotting(Fernández, *etal*.,2013).

1.2.1.2 S Sites of haematopoesis (human) in pre- and postnatal periods:

In developing embryos, blood formation occurs in aggregates of blood cells in the yolk sac, called blood islands. As development progresses, blood formation occurs in the spleen, liver and lymph nodes. When bone marrow develops, it eventually assumes the task of forming most of the blood cells for the entire organism (Fernández, *etal*.,2013).

However, maturation, activation, and some proliferation of lymphoid cells occurs in secondary lymphoid organs (spleen, thymus, and lymph nodes). In children, haematopoiesis occurs in the marow of the long bones such as the femur and tibia. In adults, it occurs mainly in the pelvis, cranium, vertebrae, and sternum(Fernández, *etal*.,2013).

1.2.1.3 Extramedullary hemopoiesis:

In some cases, haematopoietic function occur in the liver, thymus, and spleen if necessary. This is called extramedullary haematopoiesis. It may cause these organs to increase in size substantially(Georgiades, *et al.*,2002).

During fetal development, since bones and thus the bone marrow develop later, the liver functions as the main haematopoetic organ. Therefore, the liver is enlarged during development(Georgiades, *et al.*,2002).

1.2.2 Platelets (Thrombocyte)

1.2.2.1 Definitionofplatelet:

Platelets are small anucleate cell fragments that have a characteristic discoid shape and range from 1 to 3 μm in diameter. Platelets are formed from the cytoplasm of megakaryocytes (MKs), their precursor cells, which reside in the bone marrow (Pease, 1956).

1.2.2.2Megakaryocytes (MKs) and Platelet Production:

Platelets are derived from bone marrow megakaryocytes, which are large cells with multilobated nuclei and abundant finely granular light gray-blue cytoplasm. With the Size (50–100 μm) and account for ~0.01% of nucleated bone marrow cells (Nakeff and Maat, 1974).

To assemble and release platelets, MKs become polyploid by endomitosis (multiple long processes called proplatelets. An MK may extend 10–20 proplatelets, then fragments of cytoplasm break off into platelets (Richardson, *et al.*, 2005).

As platelets develop, they receive their granule and organelle content as streams of individual particles transported from the MK cell body (Italiano, et al., 1999).

CThis processis regulated mainly by thrombopoietin(produced predominantly in the liver and have critical role in megakaryocyte growth and differentiation) (Kern,2002).

1.2.2.3 Ultra structure of resting platelet:

1.2.2.3.1 Peripheral zone:

Responsible for adhesion and aggregation.Consists of fluffy glycocalyx coat, cytoskeleton and platelet membrane. Contains absorbed coagulation factors I, V, VIII, XI, XII, receptors for ADP, thrombin, vWF, collagen, fibrinogen, fibrin, fibronectin, epinephrine, thrombospondin, thromboxane A2, prostacyclin, epinephrine, serotonin and glycosyltransferase(Escolar,*etal*.,1991).

1.2.2.3.2 Sol-Gel zone:

Responsible for contraction and support microtubule system. Contains the connecting system called the open canalicular system and the dense tubular system (McNicoland Israels,1999).

1.2.2.3.3 Organelle zone:

Contains the dense body system, non-metabolic ADP, serotonin, catecholamine's, calcium, alpha granules; platelet factor 4, platelet mitogenic factor, fibrinogen, beta thromboglobulin, lysosomal granules,mitochondria and glycogengranules (White,1998).

1.2.2.4 Function of platelet:

The main function of platelets is to contribute to hemostasis, the process of stopping bleeding at the site of interrupted endothelium. They gather at the site and unless the interruption is physically too large, they plug the hole (Weyrich and Zimmerman, 2004).

In addition to being the cellular effector of homeostasis, platelets are rapidly deployed to sites of injury or infection, and potentially modulate inflammatory processes by interacting with leukocytes and by secreting cytokines, chemokines, and other inflammatory mediators. Platelets also secrete platelet-derived growth factor (PDGF)(Wagner and Burger, 2003).

1.2.2.5 Process of platelet in hemostasis:

1.2.2.5.1Platelet adhesion:

Endothelial cells are attached to the subendothelial collagen by von Willebrand factor (vWF). when the endothelial layer is disrupted, collagen and vWF anchor platelets to the subendothelium. Platelet GP1b-IX-V receptor binds with vWF and GPVI receptor binds with collagen(Dubois, *et al*.,2006).

1.2.2.5.2 Platelet activation

1.2.2.5.2.1Induction

Platelet activation begins seconds after adhesion occurs. It is triggered when collagen from the subendothelium, and/or tissue factor from the media and adventitia bind with their respective receptors on the platelet (Dubois, *et al*.,2006).

1.2.2.5.2.2 Coagulation facilitation

Move of the negatively charged phospholipids from the inner to the outer platelet membrane surface. These phospholipids then bind the tenase and prothrombinase complexes, two of the sites of interplay between platelets and the coagulation cascade. Calcium ions are essential for the binding of these coagulation factors (Dubois, *et al*.,2006).

1.2.2.5.2.3 Morphology change

Intraplatelet calcium concentration increases, stimulating the interplay between microtubule/actin filament complex with the platelet cell membrane and open canalicular system (OCS)(Matarrese, *et al.*,2009)

The continuous changes in shape from the unactivated to the fully activated platelet is best seen on scanning electron microscopy. Activated platelets secrete the contents of their granules through their canalicular systems to the exterior(Matarrese,*etal*.,2009)

1.2.2.5.2.4 GPIIb/IIIa activation

Thromboxane A2 synthesis increases during activation it is secreted and acts on both its own thromboxane receptors (the so-called "out-in" mechanism), and those of other platelets. These receptors trigger intraplatelet signaling, which converts GPIIb/IIIa receptors to their active form to initiate aggregation(Yip, *et al*.,2005).

1.2.2.5.3 Platelet aggregation:

Aggregation begins minutes after activation, and occurs as a result of turning on the GPIIb/IIIa receptor, which allows these receptors to bind with vWF or fibrinogen, there are 50–100 of these receptors per platelet(Yip, *et al.*,2005).

When any one or more of at least nine different platelet surface receptors are turned on during activation, intraplatelet signaling pathways cause existing GpIIb/IIIa receptors to change shape and thus become capable of binding(Yip, *et al.*,2005).

1.2.2.5.4 Platelet-coagulation factor interactions

In addition to interacting with vWF and fibrin, platelets interact with thrombin, Factors X, Va, VIIa, XI, IX, and prothrombin to complete clot formation via the coagulation cascade, many studies suggested platelets express tissue factor(Ahmad, *et al*.,1992).

1.2.2.6 Symptoms of platelet disorders:

Spontaneous and excessive bleeding can occur because of platelet disorders. This bleeding can be caused by deficient numbers of platelets, dysfunctional platelets(Murakawa, *et al*.,1992).

All of the following suggest platelet bleeding, not coagulation bleeding: the bleeding from a skin cut such as a razor nick is prompt and excessive, but can be controlled by pressure; spontaneous bleeding into the skin which causes a purplish

stain named by its size: petechiae, purpura, ecchymoses(van Genderen, *et al.*,1996).

Bleeding into mucous membranes causing bleeding gums, nose bleed, and gastrointestinal bleeding; menorrhagia, intraretinal, and intracranial bleeding(van Genderen, *et al.*,1996).

1.2.2.7 Disorder of platelet:

1.2.2.7.1Thrombocytopenia

Is defined as a platelet count less than the lower limit of the reference range(Kern, 2002).

1.2.2.7.1.1 Causes of Thrombocytopenia:

1.2.2.7.1.1.1Pseudo thrombocytopenia (artifactual):

Platelet clumping**,** Platelet satellitism(Kern, 2002).

1.2.2.7.1.1.2 Inherited:

Thrombocytopenia–absent radii (TAR) syndrome, wiskott-Aldrich syndromem may-Hegglin anomaly, bernard-Soulier syndrome, Gray platelet syndrome**(**Laidlaw,*etal.*,2012**)**

1.2.2.7.1.1.3 Congenital non-inherited:

Intrauterine viral infection, maternal drugs or medications: (thiazide diuretics), maternal ITP or other immunologic diseases, neonatal alloimmune thrombocytopenia (Kern,2002).

1.2.2.7.1.1.4 Acquired:

Immune: Idiopathic,Infections: viruses (EBV, CMV, HIV), bacteria, riskettsiae, Mycoplasma, Lymphoproliferative disorders, Autoimmune (collagen vascular) diseases Post-transfusion purpura(Kern, 2002).

Non-immune:Infections ,Disseminated intravascular coagulation(DIC),Thrombotic thrombocytopenicpurpura(TTP),Hemolyticuremicsyndrome(HUS),Preeclampsia/e

clampsia and the HELLP syndrome Massive transfusion,Gestational thrombocytopenia(Kern, 2002).

1.2.2.7.1.1.5 Platelet sequestration in the spleen:

Hypersplenism: usually associated with anemia and/or leucopenia (Kern, 2002).

1.2.2.7.2 Thrombocytosis (Thrombocythemia):

Thrombocytosisis defined as a platelet count exceeding the upper limit of the reference range. either Primary thrombocythemia(may be associated with thrombosis or bleeding) or Thrombocytosis secondary to some other condition (reactive thrombocytosis) and is not associated with an increased risk of thrombosis or other complication.(like infection, inflammation, iron deficiency anemia) (Kern, 2002).

1.2.2.8 Measurement of platelet:

Platelet count is measured either manually using a hemacytometer, or by placing blood in an automated platelet analyzer using electrical impedance(Girling,1962). The normal range (99% of population analyzed) for platelets in healthy individual is 150,000 to 400,000 per cubic millimeter (a mm^3 equals a microliter) or 150–400 $\times 10^9$ per liter(Ross,*etal*.,1988). Platelet function is evaluated by bleeding time test , platelet aggregation test (Duke, 1910).

1.2.2.8.1 Additional platelet parameters: mean platelet volume (MPV), platelet distribution width (PDW), platelet large cell ratio(PLCR).

Circulating platelets vary in both size and functional activity. Large platelets are probably younger, more reactive and able to produce thrombogenic factors. The Sysmex KX-21N haematology analyzer provides platelet count and platelet indices which are calculated from the platelet size distribution histogram. Platelet parameters supply clinically useful information if methodologic problems are taken into consideration. Mean platelet volume is calculated by dividing the plateletcrit

by the platelet count. MPV is decreased in conditions associated with under production of platelets Such as Bone marrow aplasia .

MPV is of particular value in the presence of :

Thrombocytopenia: A High MPV with low platelet count indicates platelet destruction such as: Immune thrombocytopenia Pre-eclampsia Sepsis Some hereditary platelet disorders e.g. Bernard Soulier Syndrome. A Low MPV with low platelet count indicates hypersplenism or marrow underproduction of platelets, such as: Aplastic anemia Cytotoxic drug therapy Some hereditary platelet disorders e.g. Wiskott Aldrich Syndrome.

Thrombocytosis: A Low MPV with a high platelet count suggests a reactive thrombocytosis as seen in: Infection Inflammation Malignancy A High MPV with a high platelet count is more suggestive of primary thrombocytosis associated with myeloproliferative disorders.

Normal Platelet Count and High MPV inducate Chronic Myeloid Leukemia or Hyperthyroidism, Low MPV may found in Chronic Renal Failure (Pya,Mml,2009)

The platelet distribution width refers to the width of the size distribution curve in fL established at the 20% height level of the peak (Figure 1.1). The platelet large cell ratio corresponds to the number of cells above the 12-fL threshold divided by the total platelet count (Figure 1)(Briggs and Machin ,2012). A high P-LCR or PDW indicates peripheral immune destruction of platelets (Kaitio ,*et al.*, 2005).also indicate presence of active large platelet.

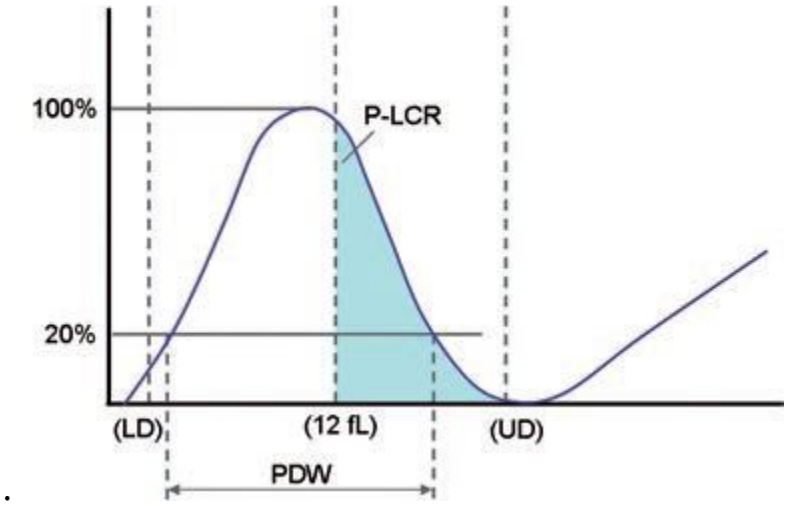

Figure 1.1 Platelet Distribution Widthand Platelet Large Cell ratio.

Abbreviations: PDW = plateletdistribution width; PLCR= platelet large cell ratio; LD / UD = lower / upperdiscrimination for platelet size distribution

1.2.2.9 Transfusion therapy with platelet:

1.2.2.9.1 Indications

Platelet transfusion is most frequently used to correct unusually low platelet counts, either to prevent spontaneous bleeding (typically at counts below (10–15)×10^9/L) or in anticipation of medical procedures that will necessarily involve some bleeding. For example, in patients undergoing surgery, a level below 50×10^9/L is associated with abnormal surgical bleeding (Roback,*et al.*,2011).Platelets may also be transfused when the platelet count is normal but the platelets are dysfunctional, such as when an individual is taking aspirin or clopidogrelFinally, platelets may be transfused to address severe hemorrhage. Platelet transfusion is contraindicated in thrombotic thrombocytopenic purpura (TTP) (Roback, *et al.*,2011).

1.2.2.9.2 Collection

Platelets are either isolated from collected units of whole blood and pooled to make a therapeutic dose, or collected by platelet apheresis(Högman,1992).

Apheresis platelets are collected using a mechanical device that draws blood from the donor and centrifuges the collected blood to separate out the platelets and other components to be collected. The remaining blood is returned to the donor (Högman,1992).

1.2.2.9.3 Storage

Platelets collected by either method have a very short shelf life, typically five days.platelets are stored under constant agitation at 20–24 °C (68–75.2 °F) (Högman,1992).

1.2.2.9.4 Delivery to recipients

Platelets do not need to belong to the same A-B-O blood group as the recipient or be cross-matched to ensure immune compatibility between donor and recipient unless they contain a significant amount of red blood cells (RBCs) (Schoenfeld,*etal*.,2006)

Prior to issuing platelets to the recipient, they may be irradiated to prevent transfusion-associated graft versus host disease or they may be washed to remove the plasma (Schoenfeld,et al.,2006)

The change in the recipient's platelet count after transfusionistermedthe"increment" and iscalculatedbysubtractingthepretransfusionplateletcountfromtheposttransfusion platelet count. When recipients fail to demonstrate an adequate post-transfusion increment, this is termed platelet transfusion refractoriness**(**Schoenfeld,*et al*.,2006**)**

1.2.2.10 Wound therapy:

Platelets release platelet-derived growth factor (PDGF), a potent chemotacticagent; and TGF beta, which stimulates the deposition of extracellular matrix; fibroblast growth factor, insulin-like growth factor 1, platelet-derived epidermal growt factor, and vascular endothelial growth factor. Local application of these factors increased concentrations through Platelet-rich plasma (PRP) is used as an adjunct in wound healing (Gawaz and Vogel,2013).

1.2.3 Chronic Kidney Disease:

1.2.3.1 The Kidneys:

The kidneys are paired' bean shaped organs located retroperitoneally on either side of spinal column(Micheal,*et al*.,2005). Each kidney of adult human weight about 150 grams(Guyton and Hall,2006) .

1.2.3.2 Renal anatomy:

Macroscopically a fibrous capsule of connective tissue encloses each kidney. Two regions can be clearly discerned an outer region called the cortex and an inner region called the medulla. The pelvis can also be seen. It is basin like cavity at the upper end of the ureter in to which newly formed urine passes. The bilateral ureters are thick walled canals, connecting the kidneys to the urinary bladder (Micheal ,*et al*.,2005).

The nephrons are functional units of the kidney that can only be seen microscopically. Each kidney contains approximately 1million nephrons. Each nephron is complex apparatus comprised of five basic parts:

1.2.3.2.1 The glomerulus:

A capillary surrounded by the expanded end of renal tubule known as Bowman's capsule. Each glomerulus is supplied by an afferent arteriole carrying the blood in and efferent arteriole carrying the blood out. The efferent arteriole branches in to peritubular capillaries that supply the tubules (Micheal ,*et al*.,2005).

1.2.3.2.2 The proximal convoluted tubules:

Located in the cortex, receive filtrate from the glomerular spaces. Convolution increases the tubular length and therefore contact between the luminal fluid and the proximal tubular cells, thus facilitating more solute reclamation than would occur if the loops were shorter(Philip D.M, *et al.*,1994).

1.2.3.2.3 The long loop of Henle:

It comprised of thin descending limb which spans the medulla. And the ascending limb, which is located in both the medulla and the cortex, comprised of a region that is thin and thick (Micheal ,*et al.*,2005).

1.2.3.2.4 The distal convoluted tubules:

Located in the cortex, important for fine adjustment of luminal fluid, lie near the afferent arterioles with the juxta glomerular apparatus between them. The production of renin by the latter is modified by flow in these blood vessels(Philip , *et al.*,1994).

1.2.3.2.5 Collecting ducts:

Formed by two or more distal convoluted tubules as they pass back down through the cortex and the medulla to collect the urine that drains from each nephron. Collecting ducts eventually merge and empty their contents in to renal pelvis(Micheal ,*et al.*,2005).

1.2.3.3 Renal failure:

Failure of renal function may occur rapidly , producing the syndrome of Acute Renal Failure (ARF), or develops insidiously, often over many years producing Chronic Renal Failure (CRF) (William and Stephen ,2004) .

1.2.3.4 Chronic Renal Failure (CRF) / Chronic Kidney Disease (CKD):

Chronic kidney disease (CKD), also known as chronic renal disease (CRD), is a progressive loss in renal function over a period of months or years. It is differentiated from acute kidney disease in that the reduction in kidney function must be present for over 3 months.

Chronic kidney disease is identified by a blood test for creatinine. Higher levels of creatinine indicate a lower glomerular filtration rate and as a result a decreased capability of the kidneys to excrete waste products(National Kidney Foundation (2002)) .

1.2.3.5 Signs and symptoms

CKD is initially without specific symptoms and is generally only detected as an increase in serum creatinine or protein in the urine. As the kidney function decreases:

*Blood pressure is increased due to fluid overload and production of vasoactive hormones created by the kidney via the RAS (renin-angiotensin system), increasing one's risk of developing hypertension and/or suffering from congestive heart failure

*Urea accumulates, leading to azotemia and ultimately uremia (symptoms ranging from lethargy to pericarditis and encephalopathy). Urea is excreted by sweating and crystallizes on skin ("uremic frost").

*Potassium accumulates in the blood (known as hyperkalemia with a range of symptoms including malaise and potentially fatal cardiac arrhythmias). Hyperkalemia usually does not develop until the GFR falls to less than 20-25 mL/min/1.73 m^2, at which point the kidneys have decreased ability to excrete

potassium. Hyperkalemia in CKD can be exacerbated by acidemia (which leads to extracellular shift of potassium) and from lack of insulin.

*Erythropoietin synthesis is decreased

*Fluid volume overload — symptoms may range from mild edema to life-threatening pulmonary edema

*Hyperphosphatemia — due to reduced phosphate excretion, which follows the decrease in glomerular filtration. Hyperphosphatemia is associated to increased cardiovascular risk, being a direct stimulus to vascular calcification.(Hruska, *et al.*,2008)

*Hypocalcemia — due to 1,25 dihydroxyvitamin D_3 deficiency. The 1,25 dihydroxyvitamin D_3 deficiency is due to stimulation of fibroblast growth factor-23(Bacchetta ,*et al.*,2012) . Osteocytes are responsible for the increased production of FGF23, which is a potent inhibitor of the enzyme 1-alpha-hydroxylase (responsible for the conversion of 25-hydroxycholecalciferol into 1,25 dihydroxyvitamin D_3).

o Later this progresses to secondary hyperparathyroidism, renal osteodystrophy and vascular calcification that further impairs cardiac function.

*Metabolic acidosis, due to accumulation of sulfates, phosphates, uric acid etc. This may cause altered enzyme activity by excess acid acting on enzymes and also increased excitability of cardiac and neuronal membranes by the promotion of hyperkalemia due to excess acid (acidemia) .(Adroguéand Madias , 1981). Acidosis is also due to decreased capacity of generating enough ammonia from the cells of the proximal tubule.

*Iron deficiency anemia, which increases in prevalence as kidney function decreases, and is especially prevalent in those requiring haemodialysis. It is multifactoral in cause but includes increased inflammation, reduction in Erythropoietin, hyperuricemia leading to bone marrow suppression.

*People with chronic kidney disease suffer from accelerated atherosclerosis and are more likely to develop cardiovascular disease than the general population. Patients afflicted with chronic kidney disease and cardiovascular disease tend to have significantly worse prognoses than those suffering only from the latter.

*Sexual dysfunction is very common in both men and women with chronic kidney disease. A majority of men have a reduced sex drive, difficulty obtaining an erection and reaching orgasm, and the problems get worse with age. A majority of women have trouble with sexual arousal, and painful periods and problems with performing and enjoying sex are common(Vecchio ,*et al.*,2010) .

1.2.3.6 Causes

The three most common causes of CKD are diabetes mellitus, hypertension, and glomerulonephritis. Together, these cause approximately 75% of all adult cases.

Historically, kidney disease has been classified according to the part of the renal anatomy that is involved.

- Vascular, includes large vessel disease such as bilateral renal artery stenosis and small vessel disease such as ischemic nephropathy, hemolytic-uremic syndrome and vasculitis
- Glomerular, comprising a diverse group and subclassified into
    - Primary Glomerular disease such as focalsegmentalglomerulosclerosis and IgA nephropathy (or nephritis)
    - Secondary Glomerular disease such as diabetic nephropathy and lupus nephritis

- Tubulointerstitial including polycystic kidney disease, drug and toxin-induced chronic tubulointerstitial nephritis and reflux nephropathy
- Obstructive such as with bilateral kidney stones and diseases of the prostate
- On rare cases, pin worms infecting the kidney can also cause nephropathy.

1.2.3.7 Diagnosis:

It is important to differentiate CKD from acute renal failure (ARF) because ARF can be reversible. Abdominal ultrasound, in which the size of the kidneys is measured, is commonly performed. Kidneys with CKD are usually smaller (< 9 cm) than normal kidneys, with notable exceptions such as in diabetic nephropathy and polycystic kidney disease. Another diagnostic clue that helps differentiate CKD from ARF is a gradual rise in serum creatinine (over several months or years) as opposed to a sudden increase in the serum creatinine (several days to weeks). If these levels are unavailable (because the patient has been well and has had no blood tests), it is occasionally necessary to treat a patient briefly as having ARF until it has been established that the renal impairment is irreversible.

Additional tests may include nuclear medicine MAG3 scan to confirm blood flows and establish the differential function between the two kidneys. DMSA scans are also used in renal imaging; with both MAG3 and DMSA being used chelated with the radioactive element Technetium-99.

1.2.3.7.1 Stages:

All individuals with a glomerular filtration rate (GFR) <60 mL/min/1.73 m^2 for 3 months are classified as having chronic kidney disease, irrespective of the presence or absence of kidney damage. The rationale for including these individuals is that reduction in kidney function to this level or lower represents loss of half or more of the adult level of normal kidney function, which may be associated with a number of complications(National Kidney Foundation (2002)) .

All individuals with kidney damage are classified as having chronic kidney disease, irrespective of the level of GFR. The rationale for including individuals with GFR > 60 mL/min/1.73 m^2 is that GFR may be sustained at normal or increased levels despite substantial kidney damage and that patients with kidney damage are at increased risk of the two major outcomes of chronic kidney disease: loss of kidney function and development of cardiovascular disease. (National Kidney Foundation (2002)) .

The loss of protein in the urine is regarded as an independent marker for worsening of renal function and cardiovascular disease. Hence, British guidelines append the letter "P" to the stage of chronic kidney disease if there is significant protein loss.

Stage 1

Slightly diminished function; kidney damage with normal or relatively high GFR (≥90 mL/min/1.73 m^2). Kidney damage is defined as pathological abnormalities or markers of damage, including abnormalities in blood or urine test or imaging studies. (National Kidney Foundation (2002)) .

Stage 2

Mild reduction in GFR (60–89 mL/min/1.73 m^2) with kidney damage. Kidney damage is defined as pathological abnormalities or markers of damage, including abnormalities in blood or urine test or imaging studies.

Stage 3

Moderate reduction in GFR (30–59 mL/min/1.73 m^2). British guidelines distinguish between stage 3A (GFR 45–59) and stage 3B (GFR 30–44) for purposes of screening and referral.

Stage 4

Severe reduction in GFR (15–29 mL/min/1.73 m^2) Preparation for renal replacement therapy.

Stage 5

Established kidney failure (GFR <15 mL/min/1.73 m^2, permanent renal replacement therapy (RRT), or end stage renal disease (ESRD) (National Kidney Foundation (2002)) .

1.2.3.7.2 NDD-CKD vs. ESRD

The term non-dialysis dependent CKD, is a designation used to encompass the status of those persons with an established CKD who do not yet require the life-supporting treatments for renal failure known as renal replacement therapy (including maintenance dialysis or renal transplantation). The condition of individuals with CKD, who require either of the 2 types of renal replacement therapy (dialysis or transplantation), is referred to as the end-stage renal disease (ESRD). Hence, the start of the ESRD is practically the irreversible conclusion of the NDD-CKD. Even though the non-dialysis dependent status refers to the status of persons with earlier stages of CKD (stages 1 to 4), patients with advanced stage of CKD (Stage 5), who have not yet started renal replacement therapy are also referred to as NDD-CKD.

1.2.3.8 Screening

Screening those who neither have symptoms or risk factors for chronic kidney disease is not recommended.(Qaseem, *,et al*.,2013) Those who should be screened include: those with hypertension or history of cardiovascular disease, those with diabetes or marked obesity, those aged > 60 years, subjects with indigenous racial origin, those with a history of renal disease in the past, as well as subjects who have relatives who had kidney disease requiring dialysis. Screening should include calculation of estimated GFR/1.73 m^2 from the serum creatinine level, and measurement of urine-to-albumin creatinine ratio in a first-morning urine specimen as well as dipstick screen for hematuria. **(**Johnson, and Davied.,2011)Guidelines for nephrologist referral vary among different countries. Nephrology referral is

useful when eGFR/1.73m^2 is less than 30 or decreasing by more than 3 mL/min/year, when urine albumin-to-creatinine ratio is more than 30 mg/g, when blood pressure is difficult to control, or when hematuria or other findings suggest either a primarily glomerular disorder or secondary disease amenable to specific treatment. Other benefits of early nephrology referral include proper patient education regarding options for renal replacement therapy as well as pre-emptive transplantation, and timely workup and placement of an arteriovenous fistula in those patients opting for future hemodialysis.

1.2.3.9 Treatment:

The presence of chronic kidney disease confers a markedly increased risk of cardiovascular disease, and people with CKD often have other risk factors for heart disease, such as hyperlipidemia. The most common cause of death in people with CKD is therefore cardiovascular disease rather than renal failure. Aggressive treatment of hyperlipidemia is warranted.(Chauhan andVaid ., 2009).

Apart from controlling other risk factors, the goal of therapy is to slow down or halt the progression of CKD to stage 5. Control of blood pressure and treatment of the original disease, whenever feasible, are the broad principles of management. Generally, angiotensin converting enzyme inhibitors (ACEIs) or angiotensin II receptor antagonists (ARBs) are used, as they have been found to slow the progression of CKD to stage 5(Ruggenenti , *et al*.,1999). Although the use of ACE inhibitors and ARBs represents the current standard of care for patients with CKD, Currently, several compounds are in development for CKD. These include, but are not limited to, bardoxolone methyl, olmesartan medoxomil, sulodexide, and avosentan.

Replacement of erythropoietin and calcitriol, two hormones processed by the kidney, is often necessary in people with advanced disease. Guidelines recommend treatment with parenteral iron prior to treatment with erythropoietin. A target

hemoglobin level of 9–12 g/dL is recommended . (Clement, *et al.*,2009) Phosphate binders are also used to control the serum phosphate levels, which are usually elevated in advanced chronic kidney disease.

When one reaches stage 5 CKD, renal replacement therapy is usually required, in the form of either dialysis or a transplant.

1.2.3.10 Prognosis:

The prognosis of patients with chronic kidney disease is guarded as epidemiological data has shown that all cause mortality (the overall death rate) increases as kidney function decreases. The leading cause of death in patients with chronic kidney disease is cardiovascular disease, regardless of whether there is progression to stage 5(Perazella and Khan .,2006).

While renal replacement therapies can maintain patients indefinitely and prolong life, the quality of life is severely affected(Francisco and Piñera .,2006). increases the survival of patients with stage 5 CKD significantly when compared to other therapeutic options; however, it is associated with an increased short-term mortality due to complications of the surgery. Transplantation aside, high intensity home hemodialysis appears to be associated with improved survival and a greater quality of life, when compared to the conventional three times a week hemodialysis and peritoneal dialysis(Pierratos ,*et al.*,2005).

1.2.3.10.1 Cancer risk:

Patients with end-stage renal disease are at increased overall risk for cancer. This risk is particularly high in younger patients and gradually diminishes with age. (Maisonneuve,*et al.*,1999).

1.2.4 Platelet abnormalities and chronic kidney disease

Patients with end-stage kidney disease are prone to develop complications due to derangements in two opposite directions of the haemostatic proces: bleeding and clotting.

Bleeding disorders result from insufficient platelet function, inefficient coagulation and/ or excessive activation of the fibrinolytic system(Jalal ,*et al.*,2010) Bleeding and clotting problems are clinical relevant as fatal bleeding episodes such as prolonged bleeding from the dialysis fistula, gastrointestinal bleeding or cerebral haemorrhage can occur. A prothombotic status is associated with an increased number of cardiovascular events or recurrent thrombosis of the dialysis access with insufficient dialysis quality (Jalal ,*et al.*,2010) . Pathogenesis of bleeding in uraemia is considered to be multifactorial and involves the coagulation cascade, the fibrinolytic system, the platelets, the endothelium and the vessel wall with its extracellular matrix. However, major defects involve the so-called primary haemostasis, i.e. platelet adhesion and aggregation, because abnormalities in plateletplatelet and platelet–vessel wall interactions are of crucial importance. The relationship between all these components is influenced by uraemic toxins and metabolic compounds accumulating during renal insufficiency. Structural changes in the vessel wall related to arteriosclerosis, due to impaired calcium and phosphate metabolism resulting in increment of vessel wall calcifications, may also influence the proneness to coagulation activation (Glorieux ,*et al.*,2009).

Mild thrombocytopenia frequently occurs in uraemia, suggesting inadequate platelet production, overconsumption or increased clearance. However, thrombocytopenia which is severe enough to cause bleeding is very rare. The haemodialysis procedure may itself cause thrombocytopenia through the interaction of blood components with the dialysis membranes that may activate complement (e.g. cuprophane) or from heparin (used as anticoagulant) which

occasionally may induce thrombocytopenia through an immunologic mechanismIn addition, a reduced percentage of reticulated platelets has been reported in patients with haemodialysis treatment, indicating reduced production. (Tassies ,*et al*.,1995). In chronic renal disease, impaired erythropoietin secretion leads to a decrease in platelet count (Suresh,*et al.*,2012). The detection of receptors for erythropoietin in megakaryocytes is understandable, because erythropoietin levels can affect platelet level and because of extensive homology between erythropoietin and thrombopoietin, erythropoietin act as the major humoral regulator of platelet mass (Suresh,*et al*.,2012). In previous study obtained by Forbes,*et al*,2013) condcted that Both thrombocytosis and thrombocytopenia are commonly seen in patients with end-stage renal disease (ESRD), and the role of antiplatelet agents in these patients is, at best, controversial. Relative thrombocytosis platelet count >300 has been linked with severity of cardiovascular disease in the CKD population and increase death rate. The investigators here linked this relative thrombocytosis with a reduction in iron stores, which may also be a key player (Forbes, et *al*, 2013). Conversely, patients with thrombocytopenia and CKD are also commonly seen. A reduction in platelet count during the course of a dialysis session is recognized, alongside platelet activation and degranulation, which is attributed to exposure to the dialysis membrane and the roller pump and results in platelet–lymphocyte aggregates. Thrombocytopenia may also be seen as part of the syndrome of heparin-induced thrombocytopenia in dialysis patients, whereby the generation of the platelet-factor 4/ heparin complex triggers antibody formation. The complex then binds to the antibodies, cross-reacts with platelet surface receptor activation and aggregation, further PF4 release and formation of procoagulant factors and thrombin. It has been suggested that the presence of these antibodies is an independent predictor of cardiovascular morbidity and mortality (Forbes,*et al*,2013).

Furthermore, in patients with chronic kidney disease, platelets and the coagulation system could be activated in atherosclerotic vessels, contributing to the formation of venous thrombosis at different vessel site (Piazza ,*et al*.,2011) .

1.3 Objectives:

1.3.1 General objective:

To measure platelet count, platelet indices(mean platelet volume (MPV), platelet distribution width (PDW) and platelet large cell ratio (PLCR)) in chronic kidney disease patients.

1.3.2 Specific objectives:

- To estimate the platelet count and platelet indices (MPV,PDW, and PLCR) in chronic kidney Disease patients compared to normal control.
- To determine the relation between platelet count , platelet indices(MPV,PDW, and PLCR) and concentration of serum creatinine in chronic kidney Disease patients.
- To determine the relation between platelet count , platelet indices(MPV,PDW, and PLCR) and duration of disease in chronic kidney Disease patients
- To determine if there is any difference of platelet count, platelet indices(MPV,PDW, and PLCR) in patients on haemodialysis and patients not on haemodialysis.
- To estimate the level of creatinine to determine severity of disease.

1.4 Rationale:

Chronic Kidney disease is associated with bleeding problems, indicated by decreased platelet aggregation and prolonged bleeding times.

Recent evidence has shown that larger platelets are more reactive per unit volume than smaller platelets and are more likely to aggregate, leading to thrombosis. Large platelets seem to be an independent risk factor for myocardial infarction which occur in the patients of CKD. and platelet size is one predictor of recurrent myocardial infarction and death.

Platelet disorders are relatively common in the general practice of medicine. In order to assist in clarifying the cause of these disorders and help in some instances with diagnosis ,in this study we sought to assess the impact of CKD on platelet count and indices(MPV,PDW, and PLCR) ,and then may help to establish secondary preventive medication in individual patients.

Chapter Two

Material and Method

2.1Study design:

This is descriptive cross sectional study conducted from February August 2014. At East Nile modern hospital in hematology and clinical department. Aimed to measured platelet count, mean platelet volume, platelet distribution width ,platelet large cell ratio and creatinine in chronic kidney disease patients (case) and non chronic kidney disease individual (control).

2.2 Study population:

The study includes 75 diagnosed chronic kidney disease patients and 75 age and sex matched healthy individuals as control group.

2.2.1 Inclusion criteria:

- Diagnosed chronic kidney disease patients who had serum creatinine > 2.0 mg/dl.
- Confirmed cases of chronic kidney disease patients, who were on haemodialysis.
- Confirmed cases of chronic kidney disease patient, who were not on haemodialysis.
- Non chronic kidney disease individual as control group for comparing.

2.2.2 Exclusion criteria:

- Patients on CKD had recent blood loss and transfusion.
- Patients had recent or previous thrombosis.
- Patient had recent infection that are known to affect the parameter we investigate.
- Patients non cosent for investigation also excluded.

2.3 Sample collection:

Blood sample were collected from anticubital vein from each case pre haemodialysis(for patients on dialysis) and control groups under sterile condition using sterile disposable syringe and 2.5 ml blood drained into EDTA vaccontainerand 2.5 ml drained into heparin vaccontainer.

2.4 Data collection:

Data collected through questionnaires(sex, age, duration of the disease, therapy type and others disease)

2.5 Methodology:

2.5.1 Measuring of platelet count, MPV , PDW and PLCR by Sysmex KX-21N:

2.5.1.1 Principle:

Blood sample is aspirated, measured to a predetermined volume, diluted at the specified ratio, then fed into each transducer. The transducer chamber has a minute hole called the aperture. On both side of the aperture, there are the electrodes between which flows direct current. Blood cells suspended in the diluted sample pass through the aperture, causing direct current resistance to change between the electrodes. As direct current resistance changes, the blood size is detected as electric pluses. Blood cell size is calculated by counting the pulses, and a histogram of blood cell sizes is plotted by determining the pulse sizes. Also, analyzing a histogram makes it possible to obtain various analysis data.(Sysmex,1999).

2.5.1.2 Reagent:

Cell pack

Stromatolyser-WH

2.5.1.3 Method:

Well mixed EDTA blood was aspirated from the sample probe into the sample rotar valve, then 4µl of blood measured by sample rotor valve was diluted into 1:50 with 1.996ml of diluted and brought to the mixing chamber as diluted sample, then out of the1:500 dilution sample,40µl is measurd by the sample rotor valve, diluted into1:25000 with1.960ml of dilutent, then transferred to the RBC\PLT transducer chamber. then 250µlfrom this was aspirated through the aperture.then was counted by detection method .

2.5.1.4 Result calculation:

MPV(fl)= $\frac{\text{PCT(platelet}-\text{crit)}}{PLT(X1O\text{^}3\backslash\mu L)}$ X1OOO

PDW(fl): is distribution width on 20%frequency level with the peake taken as100%

PLCR: Fixed discriminator at 12 fl divided by total number of platelet.

2.5.1.5 Refrence range:

PLT count=150-450x10^3\µl

MPV=8.5-12.5fl

PDW=9 – 14 fl

PLCR=15-35%

2.5.2 Count of platelet from thin blood film staining by Ral-555:

2.5.2.1 Staining principle:

The mechanism by which certain structural components of a cell stain with a certain dye whereas other similar structures do not, although staining with other dyes, depends on complex differences between dyes, how they interact with each other, as well as the pH of the stain and the cellular micro environment. Acidic structures pick up the basic dye, methylene blue, which as its name implies is blue

in colour. In contrast basic or alkaline structures bind acidic dyes, in this case eosin which is pink.

2.5.2.2 Reagents :

Solusion (1): Alcholic fixative.

Solution(2): Eosin.

Solution(3): Methylen blue.

2.5.2.3Method:

Manual smears were made by placing a drop of blood on one side of a glass slide, and spreading this by rapidly moving a second glass slide or spreader across the first slide at an angle .after dry a good quality smear were staining as follow:

- Slid was Dipped 1 minute in solution 1
- Excess solution was removed into filter paper.
- Slid was Dipped 2X1 second solution 2
- Excess solution was removed into filter paper.
- Slid was Dipped 2X1 second solution 3
- The slide was washed briefly by distilled water.
- The slid was allowed to dry in open air.
- The slide was examined under x100 immersion objective microscope.

2.5.2.4 Result:

Count platelets(small 2.5μm in diameter ,discoid shape, purple color) in ideal area. counting platelet in 10 field and then get the mean ,divided mean x20000 to get numer of platelet / µl.

2.5.3 Measurement of Creatinine level byRoche/Hitachi cobas c systems:

2.5.3.1 Test principle:

Buffered kinetic Jaffé reaction without deproteinization,

In alkaline solution creatinine reacts with picrate to form a yellow-red adduct .The rate of the dye formation (color intensity) is directly proportional to the creatinine concentration in the specimen and is measured photometrically,(Cobas c system manual,2008) .

2.5.3.2 Reagents :

R1 Potassium hydroxide: 900 mmol/L; phosphate: 135 mmol/L; pH ≥13.5, preservative; stabilizer

R2/R3 Picric acid: 38 mmol/L; pH 6.5; non reactive buffer

2.5.3.3 Method:

Centrifuged heparin zed blood(in vaccotainer tube) was putted in the cobas system then the system take 13µl from R1 +77 µl from diluents and17µl from R3+30 µl from diluents and10µl from the sample to start the work,increase kinetic method,read at wavelength 570nm.

2.5.3.4 Result:

Roche/Hitachi cobas c systems automatically calculate the analyte

Concentration of each sample.

Conversion factors: µmol/L x 0.0113 = mg/dL

µmol/L x 0.001 = mmol/L

2.5.3.5 Refrence range:

Adults: for

Females 44-80 µmol/L (0.50-0.90 mg/dL).

Males 62-106 µmol/L (0.70-1.20 mg/dL).

2.6 Ethical consideration:

The research approved at the level of hematology department research committee of post graduate studies (SUST).

All patient informed (verbal informed consent) about research before collection of samples.

2.7 Data analysis:

Data was analyzed by SPSS computer program, version 14.0 the significant level set at ($\leq$0.05).

Chapter Three

Results

Table (3.1) show A total of 150 subjects age (22--90years) of mean (52.43±18.4years),male 44(58.7%), female 31 (41.3%) was studied for their haematological variables (Platelets count, MPV, PDW, P-LCR), Out of total 75 were patients well diagnosed with CKD as a case study group with mean of creatinine level(7.91 ± 4.8mg/dl), and had disease for period (1.8±1.7 year). and the other 75 were healthy individual have been selected as control group ,mean of creatinine for them(1.4±1.3mg/dl). Table (3.2) Show that there was insignificance difference in (Platelets count, MPV, PDW, P-LCR), between CKD patients and control group . (p-value 0.37,0.06, 0.44, 0.192respectively) Table (3.3) Show that insignificant different in (Platelets count, MPV, PDW, P-LCR), between CKD patients with s.creatinine less than 6 mg\dl and CKD patients with s.creatinine more than 6mg\dl (P-value 0.26, 0.53, 0.53, 0.69 respectively).Table (3.4) Show that there was insignificance difference in (Platelets count, MPV, PDW, P-LCR), between Duration group.(p-value 0.76, 0.49, 0.789, 0.59 respectively)

Table (3.5**)** Show that insignificant different in (Platelets count, MPV, PDW, P-LCR), between CKD patients on haemodialysis and CKD patients were not on haemodiaylisis (P-value0.46, 0.858,0.74 ,0.92 respectively)

Table (3.1) Baseline Characteristics of the Study Population:

Characteristics	Case	Control
Number	75	75
Mean of Age / years Mean±SD	52.43 ± 18.4	52.43 ± 18.4
Male	44 (58.7%)	44 (58.7%)
Female	31 (41.3%)	31 (41.3%)
Mean of Duration of the diseases / years Mean±SD	1.8±1.7	-
Mean of Creatinine mg/dl Mean±SD	7.91 ± 4.8	1.4±1.3

Table (3.2) Comparison of mean platelet count and mean platelet indices (MPV, PDW and PLCR) between cases and controls:

Parameters	Mean ± SD		P value
	Case (N = 75)	Control (N = 75)	
Platelets count $x10^3/\mu l$	274.01±102.5	286.60±76.5	0.37
MPV / fl	9.27±1.1	9.57±0.6	0.06
PDW / fl	11.58±2.3	11.85±1.8	0.44
PLCR %	21.07±8.0	22.50±4.7	0.19

* The mean difference is significant at the 0.05 level.

Table (3.3) Comparison of platelet count and platelet indices (MPV, PDW and PLCR) between patients had S.creatinine Less than 6 mg\dl and More than 6 mg\dl:

Parameters	Mean ± SD		P value
	Less than 6 mg\dl (N= 32)	More than 6 mg\dl (N= 43)	
Platelets count $x10^3$/µl	290.06 ± 112.1	262.07 ± 94.5	0.26
MPV / fl	9.37 ± 1.1	9.20 ± 1.2	0.53
PDW / fl	11.60 ± 2.2	11.57 ± 2.5	0.53
PLCR %	21.52± 7.7	20.76 ± 8.4	0.69

* The mean difference is significant at the 0.05 level

Table (3.4) Comparison of platelet count and platelet indices (MPV, PDW and PLCR) between patients had the disease less than 2 years and more than 2 years:

Parameters	Mean ± SD		P value
	Less than 2 years (N = 62)	More than 2 years (N = 13)	
Platelets count $x10^3/\mu l$	275.73 ± 102.7	265.85 ± 105.6	0.76
MPV / fl	9.32 ± 1.2	9.07 ± 0.8	0.49
PDW / fl	11.58 ± 2.4	11.77 ± 2.5	0.79
PLCR %	21.32 ± 8.4	19.96 ± 8.5	0.59

* The mean difference is significant at the 0.05 level

Table (3.5) Comparison of platelet count and platelet indices (MPV, PDW and PLCR) between patients on haemodialysis and not on haemodialysis:

Parameters	Mean ± SD		P value
	haemodialysis (N= 45)	Not on haemodialysis (N= 30)	
Platelets count $x10^3/\mu l$	266.80 ± 100.6	284.83 ± 106.2	0.46
MPV / fl	9.25 ± 1.2	9.30 ± 1.1	0.88
PDW / fl	11.67 ± 2.5	11.47 ± 2.2	0.74
PLCR %	21.00 ± 8.2	21.20 ± 7.9	0.92

* The mean difference is significant at the 0.05 level

Chapter Four

Discussion, Conclusion and Recommendation

4.1 Discussion

Chronic Kidney Disease (CKD) is a major health problem throughout the world (Hsu, et *al*., 2001). In Sudan, according to ministry of health records, the prevalence of renal failure is increasing through the few past years; approximately 70 to 140 new patients undergo dialysis each year (Mohamed,*et al*.,2008).This study was conducted to assess if platelet count and indices affects in CKD.

Our study revealed insignificant difference in platelet count among chronic kidney disease patients compare to normal control group. There was no relation between platelet count and level of serum creatinine. No significant difference according to duration of CKD. The present result showed insignificant difference in platelet count among patients who were on hemodialysis and those who were not on hemodialysis .this result in agreement to study of (Akinsola,*et al*.,2009) in Nigeria. And study of (Oluboyede and Williams.,1995) in Nigeria, and study of Arogundade,*et al*.,2006)also study of(Talwar,*et al*.,2002) in India.And study of(sharpe,*et al*.,1994) in Ireland.also study of(Mohamed ,*et al*.,2008) in sudan).Inaddition to study of (Nazrul,*et al*.,2010) in Bangladesh.On the other hand our finding in this study was in disagreement to study conducted by John,*et al*.,2012) , (Ezimah and Abijah,2004) in Maiduguri, (Suresh,*et al*.,2012) ,and (Shittu ,*et al*.,2013) in Nigeria. who found in their study significant reduction in platelet count in CKD patients but insignificant reduced in plalelet in current study and these may be due to use of Erythropoietin therapy in individual patients.because of the role of Erythropoietin potentites the effect of megakaryocyte colony stimulating factors,platelet activating- acetlyhydroase (PAF-AH) and paraoxonase (PON1).

Therefore, it can be observed that platelet count is highly variable in chronic kidney disease(Nazrul,*et al*.,2010).

Our study showed insignificant difference in mean platelet volume (MPV) among chronic kidney disease patients compare to normal control group. There was no relation between MPV and level of serum creatinine. No significant difference according to duration of CKD. The present result revealed insignificant difference in MPV among patients whom on hemodialysis and those who did not on hemodialysis .this is in agreement to study of (Bilen,*et al*.,2014),and study of (Marianne ,*et al*.,2013).on the other hand this result in disagreement to previous study obtain by (Zhang,*et al*.,2011) in Wuhan_china , (sharpe,*et al*.,1994) in Ireland ,and study conducted by (Ju,*et al*.,2014) in their study MPV significant increase in MPV. In current study insignificant result may be due to low number of subject under study.

Recent study showed insignificant differences in platelet distribution width (PDW) among chronic kidney disease patients compare to normal control group. There was no relation between PDW and level of serum creatinine. No significant difference according to duration of CKD. The present result revealed insignificant difference in PDW among patients whom on hemodialysis and those who did not on hemodialysis. our finding in PDW similar to study of (sharpe,*et al.*,1994 in Ireland), also agree to study of (Marianne ,*et al*.,2013).This result was in contrast to study obtain by(Zhang,*et al*.,2011,in Wuhan_china) whom found significant increase in PDW this finding supported that there were active large platelet in CKD patients. But in our study we were performed peripheral blood picture and no large platelet was observed.

Our study revealed insignificant differences in platelet large cell ratio (PLCR) among chronic kidney disease patients compare to normal control group. There was no relation between PLCR and level of serum creatinine. No significant

difference according to duration of CKD. The present result showed insignificant difference in PLCR among patients whom on hemodialysis and those who did not on hemodialysis. This is in agreement to study of (Marianne, et *al*., 2013).No more published data found in this parameter.

4.2 Conclusions:

This study concluded that:

- Insignificant difference in platelet count and platelet indices (MPV, PDW, and PLCR) in chronic kidney Disease patients compared to normal control.
- No relation between platelet count, platelet indices (MPV, PDW, and PLCR) and concentration of serum creatinine in chronic kidney Disease patients.
- NO relation between platelet count , platelet indices(MPV,PDW, and PLCR) and duration of disease in chronic kidney Disease patients
- Insignificant difference of platelet count, platelet indices (MPV, PDW, and PLCR) in patients on haemodialysis and patients not on haemodialysis.
- **4.3 Recommendations:**

  After completion this work the following were recommended:
- Management CKD patients according to clinical data and correlate with laboratory finding of platelet count and Indices.
- Increase sample size, and further studies are needed to elucidate the basis of this finding.
- Other research in platelet in CKD patients such as platelet function because there is evidence of an effect of ureamia on platelet function most common abnormality is prolongation in bleeding time also defective aggregation. To detect and follow up the complication of CKD to prevent risk of bleeding or thrombosis in these patients.

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Appendix

Sudan University of Science & Technology

College of Medical Laboratories Science

Measurement of platelet count and Indices(MPV,PDW,and PLCR)in CKD patients.

Questionnaire

Name:……..

Age:……

Gender: **Male ( )** **Female ( )**

Residence:……

Telephone Number:…….

Duration of chronic kidney disease:.....

Anyothersdisease:…….....

Do you on haemodialysis? Yes( ) No( )

If on haemodialysis ,How long?.....

Medication on use……..

If intake Medicatio,for how long?.....

Investigation:

Serum creatinine:……

Platelets count:…… **MPV**:… …… **PDW**:…… …. **P-LCR**:……

Comment on peripheral blood picture:……

بعد فهم محتويات هذا الاستبيان والغرض من إجراء البحث انا .....(او من ينوب عنه) اوافق على اخذ العينة.

Date:..... *Sig*:.....