Sudan University of Science and Technology

Collage Graduate Studies

Iron Profile and Hemoglobin Concentration among patients with Chronic Renal Failure on Hemodialysis in Khartoum

قياس مستوي الحديد و تركيز خضاب الدم لدى المرضى الذين يعانون من الفشل الكلوي المزمن تحت الإستصفاء الدموي في الخرطوم

A Dissertation Submitted in Partial Fulfillment of the Requirement for the Award of the Degree of M.Sc In Hematology and Immunohematology

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الآية

بسم الله الرحمن الرحيم

قال تعالى:-

يَرْفَعِ اللَّهُ الَّذِينَ آمَنُوا مِنْكُمْ وَالَّذِينَ أُوتُوا الْعِلْمَ دَرَجَاتٍ ۚ وَاللَّهُ بِمَا تَعْمَلُونَ خَبِيرٌ

صدق الله العظيم

سورة المجادلة الآية 11

DEDICATION

I dedicate this work to:

My parents who are supporting me and gave us hope and wished to success

My sister and my brothers

My husband and my kids

My teacher who supported and supervisor

My college who gave me possibility of completing this dissertation

Everyone who has helped me to learn new things and reach this level

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Thanks are first and last to allah who enable me to conduct this study

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All love to my kids (egbal and abubaker) and my husband (Mohamed).

Abstract

Anaemia is common in patients with chronic renal failure specially in patients on haemodialysis. An important factor in successful treatment of anaemia by giving erythropoietin stimulating agent combined with iron supplement .so, iron status must be monitored regularly to ensure maintain of adequate iron supplement Without adverse effect of excess iron(Allen and Nissenson,1999).

This study aimed to assess body iron status and measurement of haemoglobin concentration in Sudanese patients at end stage chronic renal failure treated with regular haemodialysis,iron supplement and recombinant erythropoietin, and correlate iron profile with gender, age and duration of dialysis.

A total of Hundred Sudanese patients (80 chronic renal failure patients under haemodialysis and 20 healthy control) were Included in this study from dialysis department in Khartoum states (Omdurman military teaching hospital). 53% of patients enrolled in the study were male and 47% were female, their age ranged between 18 to 85 years. Data collected using structured interview questionnaire. Serum iron, serum ferritin and total iron binding capacity measured using DIRUICS-T240 auto chemistry analyzer. haemoglobin concentration was measured by using automated haematological analyzer (sysmex).Data analyzed using statistical package for social sciences (spss).

64% of patients were found with high s.ferritin level with more than 400Mg/L,The mean was(615±245.8).and high serum iron with mean (86.39±52.148). Seventy two of patients out of hundred were found to have anaemia,the mean of haemoglobin was (8.96±1.5), .this study showed lowered level of TIBC with mean (176.25±69.89,p-value0.000).

The present study showed that the serum iron and serum ferritin of those under haemodialysis patients were significantly higher compared to the control group with p value 0.001 and 0.000 as respectively.

There were no influence of patients age or gender on iron profile.

In conclusion patients with chronic kidney disease who were treated with intravenous iron are prone to iron over load

مستخلص البحث

فقر الدم شائع عند المرضى اللذين يعانون من الفشل الكلوي المزمن وخاصه في المرضى اللذين يحتاجون الى الاستصفاء الدموي . يعتبر اعطاء الاريثروبيتين بالاضافه الى الحديد التكميلي عامل مهم في نجاح علاج فقر الدم. لذلك يجب متابعه حاله الحديد بانتظام لضمان الحفاظ على كميه كافيه من الحديد دون التاثير سلبا بسبب ذيادته.

تهدف هذه الدراسه الى تقييم حاله الحديد وقياس كميه خضاب الدم لدى المرضى السودانيين الذين يعانون من الفشل الكلوي المزمن والذين يعالجون بالاستصفاء الدموي,الاريثروبويتين, والمكمل الحديدي, ثم ربط النتائج مع عمر ونوع المرضى ومده الاستصفاء الدموي. تم تضمين مائه مريضا في هذه الدراسه من قسم الغسيل الكلوى بولايه الخرطوم من مستشفى ام درمان التعليمي العسكري (ثمانون مريضا يعانون من الفشل الكلوي المزمن ويخضعون للاستصفاء الدموي و عشرون اخرون لايعانون من الفشل الكلوي). 53% من المرضى الذين شملتهم الدراسه من الذكور و47% كانوا من الاناث. ولقد كانت اعمارهم تتراوح بين 18الى 85 عاما. تم جمع البيانات عن طريق الاستبيان. تم قياس الحديد المصل وفرتين المصل والقدره الكامله على ربط الحديد باستخدام جهاز تحليل الكيمياء الآلي DIRUICS-T240 auto chemistry وقد تم قياس كميه خضاب الدم باستخدام محلل الدم الالي (سيسمكس) .كما تم تحليل البيانات باستخدام الحزم الاحصائيه للعلوم الاجتماعيه.

أظهرت الدراسة أن 64% من المرضى كان مستوى الفرتين المصلي لديهم مرتفعا اكثر من 400 مايكروجرام/لتر وكان المتوسط (615±245). كما أظهرت الدراسه أن مستوى الحديد المصل كان مرتفعا حيث كان المتوسط (86.39±52.148). كما وجد أن 72من المرضى من مجموع مائه يعانون من فقر الدم, حيث كان المتوسط 8.96±1.5. وقد وجد ايضا أن القدره الكامله على ربط الحديد كانت منخفضه حيث كان المتوسط 176.25±69.89. وبالتالي خلصت هذه الدراسة إلى أن مستوى الحديد المصل وفرتين المصل للمرضى اللذين يتم علاجهم بالاستصفاء الدموي تكون مرتفعه مقارنه بالمرضى الاخرين الذين لايخضون للاستصفاء الدموي. حيث كانت الفروق ذات دلالة إحصائية 0.001 للحديد المصل و0.000 للفرتين المصل وقد وجد ايضا ان نوع المرضى واعمارهم ليس له تاثير على محتوى الحديد لديهم. وبذلك خلصت هذه الدراسة إلى أن المرضى الذين يعانون من الفشل الكلوي المزمن تحت الاستصفاء الدموي والذين يعالجون بالحديد عرضه لزياده نسبه الحديد.

List of abbreviation

abbreviation	Term
ACD	Anaemia of chronic disease
CKD	Chronic kidney disease
CRF	Chronic renal failure
CAPD	Continuous ambulatory peritoneal dialysis
D.W	Distilled water
DMT.1	Divalent metal transported
ECF	Extra cellular fluid
EDTA	Ethylene diamin tetra acetic acid
GFR	Glomerular filtration rate
HD	Hemodiafiltration
HF	Hemofiltration
HB	Haemoglobin
HLA	Human lecuocoyte antigen
MCV	Mean corpuscular hemoglobin

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Chapter one

Introduction and literature review

1.1Introduction:

Chronic renal failure (CRF) also called chronic kidney failure, chronic renal insufficiency, or uremia is a slowly progressive loss of renal function over period of months or years and defined as an abnormally low glomerular filtration rate (GFR).

GFR that lead to severe illness and requires some form of renal replacement therapy such as dialysis is called end-stage renal disease (Levey *et al*, 2005).

chronic kidney disease (CKD) is complex disease impacting more than twenty million individual in the united state .number of prevalent CKD patient will continue to rise, reflecting the growing elderly population and increasing numbers of patient with diabetes and hypertension. progressive of CKD is associated with a number of serious complication ,including increased incidence of cardiovascular disease , hyper lipidemia, anemia and metabolic bone disease . CKD patient should be assessed for the presence of these complication and received optimal treatment to reduce their morbidity and mortality (Thomas *et al* ,2009).

1.2Iron deficiency in patient with renal failure:

The anemia of renal failure is caused by lack of sufficient quantities endogenous erythropoietin with availability of recombinant human erythropoietin (rHuEPO), however ,it has become apparent that to achieve a given target , hematocrit requires proper management of iron replacement , as well as the administration of rHuEPO. the iron deficiency , either absolute or functional will occur in most, if not all patient on hemodialysis receiving (rHuEPO) because of the increased demand for iron driven by the accelerated erythropoiesis that occurs with exogenous rHuEPO administration , coupled with ongoing blood losses from dialyzer and tubing , blood sampling , gastrointestinal blood loss and blood loss at the time of dialysis needle placement and removal blood loss is less of a

problem in patient on peritoneal dialysis, but poor iron intake and increased demand for iron are also seen ,the latter in patient receiving rHuEPO (Allen and Nissenson ,1999). The anemia on patient with CKD is profound in addition to the well known symptoms of fatigue , dizziness ,and shortness of breath ,anemia has been associated with more sever adverse outcomes ,such as cardiovascular complication including left ventricular hypertrophy and congestive heart failure .hypoxia caused by anemia stimulate the rennin –angiotensin-aldosterone system and contribute to renal vasoconstriction .other general complication associated with anemia include reduced cognitive function and mental acuity , impaired quality of life , and the need for blood transfusions. (O'mara, 2008).

1.3. Literature Review:

1. 3.1Urinary system:

The organs of the body that are concerned with formation of urine and its elimination from the body are referred to as urinary organs.

They consist of the right and left kidneys in which urines is formed, the right and left ureters, the urinary bladder (Singh,2008).

1.3.2 The definition and microscopic structure of the kidney:

The kidney are two reddish brown organ situated high up on the posterior abdominal wall, one on each side of the vertebral colum.(Snell, 2012).

The kidney is composed of about 1million functional units , the nephrons, and smaller number of collecting tubules.The collecting tubules transport urine through the pyramids to renal pelvis giving them their striped appearanceThe tubules are supported by small amount of connective tissue, Containing blood vessels, nerves and lymph vessels (Waugh and Grant,2001) .

1.3.3 The function of the kidney:

The kidneys have three major functions:

Excretion of waste, Maintenance of extracellular fluid (ECF) volume and composition and Hormone synthesis .(Marshall and Bnagert,2008).

1.4 The kidney in chronic kidney disease:

During the initial evaluation of patient with chronic kidney disease (CKD), physician routinely measured kidney size. commonly measured via renal ultrasound, kidney size measurement is of fundamental clinical importance for at least three reasons (i) it facilitates the diagnosis of kidney disease aetiology for example, large kidney with bilateral renal cyst may suggest polycystic kidney disease, and small kidney may denote the presence of chronic glomerulonephritis. (ii) it help to decide whether to biopsy kidney.(iii) the measurement of kidney size is useful to assess renal prognosis ,small atrophic kidney are thought to be associated with poor renal prognosis.(Kariyanna *et al*,2010).

1.5 Renal failure:

Renal failure result when the kidney cannot remove the body's metabolic waste or perform their regulatory function.The substance normally eliminated in urine accumulate in the body fluid as result of impaired renal excretion, affecting endocrine and metabolic function as well as fluid, electrolyte and acid –base disturbance. Renal failure is systemic disease and final common pathway of many different kidney and urinary tract disease (Brunner and Suddarth's,2014).

1.6 Renal replacement therapy:

The use of renal replacement therapy necessary when the kidney can no longer remove wastes, maintain electrolytes and regulate fluid balance.

This can occur rapidly or over long period of time and need for replacement therapy can be acute (short term) or chronic (lon11g term). The main renal replacement therapies include the various type of dialysis and kidney transplantation (Brunner and Suddarth's,2014).

1.7 Dialysis:

Among the end stage renal disease therapies, the choice made will depend on what is available. One of the main decisions to be whether the patient will be coming to clinic for regular dialysis (hemodialysis) or whether he or she would prefer the independence of dialyzing at home using either a home hemodialysis or peritoneal dialysis (Daugirdas *et al*,2015).

1.7.1 Definition of dialysis:

Dialysis is process of separating macromolecules from iron and low molecular weight compound in solution by the difference in their rates of diffusion through

a semipermeable membrane. Crystalloids pass readily through this membrane, but colloid pass very slowly or not at all. Dialysis procedure include hemodialysis (HD), hemodiafiltration (HDF) and peritoneal dialysis (PD) (Burtis *et al*,2008).

1.7.2 Hemodialysis:

The objective of hemodialysis are to extract toxin nitrogenous substances from blood and to remove excess fluid. Hemodialysis is used for patient who are

actually ill and require short-term dialysis for days to weeks until kidney function resumes and for patient with advanced CKD. hemodialysis prevents death but does not compensate for the loss of endocrine or metabolic activities of the kidneys.A dialyzer (also referred to as an artificial kidney) is a synthetic semi permeable membrane through which blood is filtered to remove uremic toxins and desired amount of fluid. In hemodialysis the blood, toxin and nitrogenous wastes, is diverted from the patient to machine via the use of a blood pump to the dialyzer, where toxins are filtered from the blood and the blood returned to the patient (Brunner and Suddarth's,2014).

1.7.3 Peritoneal dialysis:

Due it simplicity, PD offers patient a heme-based therapy with very little requirement for special water system and simple equipment setup time. The percentage of patients choosing PD over hemodialysis is about 12% in the United States and 20%-30% in Canada. There are two type of PD for patient to consider:

1. Continuous ambulatory peritoneal dialysis (CAPD), where the pt performs manual exchange 4 or 5 times per day.
2. Automated peritoneal dialysis where apatient hook up to machine at night and exchanges are carried out automatically while the pt sleep (Daugirdas *et al*, 2015).

1.7.4 Hemodiafiltration:

HDF is a method of dialysis that combines HD and hemofiltration(HF). It offers the advantages of both HD and HF in a single therapy.

The replacement fluid, previously supplied in autoclaved bag, is now generated "online" from concentrated bicarbonate and uses 20to30L of water per session.

The result is that HDF provide 10% to 15% increase in urea clearance compared with HD as well as increased middle molecule clearances (Burtis *et al*,2008).

1.8 Iron:

Iron is essential element for many living organism. In human it is required for oxygen transportation and electron transfer reaction. Approximately two-third of the body iron is found in erythrocyte and further 15% is in muscle and cellular enzymes. The remaining iron is excess to needs and stored primarily as ferritin or hemosiderin in the liver and within macrophages in the reticulo endothelial system (Lam,2013).

1.8.1 Iron absorption:

Organic dietary iron is partly absorbed as haem and partly broken down in the gut of inorganic iron. Absorption occurs through the duodenum. Haem is absorbed through receptor, yet to be identified, on the apical membrane of the duodenal enterocyte. Haem is then digested to release iron. In organic iron absorption is favored by factor such as acid and reducing agent that keep iron in the gut lumen in (fe+2)ferrous rather than (fe+3)ferric state. DMT.1is involved in transfer of iron from lumen of the gut across the enterocyte microvilli. Ferro protein at the baso lateral surface control exit of iron from the cell into plasma .the amount of iron absorbed is regulated according to the body need.

Table (1.1) iron absorption:

Factor favoring absorption	Factor reducing absorption
Haem iron	In organic iron
Ferrous form (fe+2)	Ferric form (fe+3)
Acid (Hcl, vitamin C)	Alkalis-antacid, pancreatic secretion
Solubilizing agent e.g. Sugar and amino acid.	Precipitating agent, phosphates, tea.
Reduced serum hepcidin	Increased serum hepcidin
Infective erythropoiesis	Decreased erythropoiesis
Pregnancy	Inflammation
Hereditary haemochromatosis	_

(Hoffbrand and Moss,2016).

1.8.1.1 Mechanism regulating iron absorption:

The iron stores in the body are regulated by intestinal absorption. Intestinal absorption of iron is itself regulated process and the efficacy of absorption increases or decreases depending on the body requirement of iron.

The dietary, which exist mostly in ferric form, is converted to the more soluble ferrous form which is readily absorbed.

The ferric form is reduced to ferrous by the action of acid in stomach, reducing agent such as ascorbic acid and cystein.

Entry of (Fe+3) into mucosal cell may be aided by an enzyme on the brush-border of the enterocyte (the enzyme possesses ferric reductase activity also). The ferrous iron is then transported in the cell by divalent metal transported (DMT1) in the

intestinal cell, the iron may be (a) stored in to ferritin in these individuals who have adequate plasma iron concentration.

A ferroxidase converts the absorbed ferrous iron to the ferric form, which then combines with apoferritin to form ferritin or (b) transported to transport protein at the basolateral cell membrane and released into the circulation (Arora and Kapoor,2012).

1.8.2. Iron metabolism:

Most of the iron is present in the oxygen carring protein of the red blood cell-hemoglobin. Iron turnover is also dominated by the synthesis and break down of hemoglobin.Haem is synthesized in the nucleated red cell in the bone marrow. Haem break down take place in the phagocytic cell, largely these in the spleen, liver and bone marrow. Iron is released from haem by haem oxygenase and is largely reused for haem synthesis. Every day about 30mg of iron are used to make new hemoglobin, and most of these is obtained from the breakdown of old red cells. Relatively little iron is lost from the body (about 1mg/dl in men), and these losses are not influenced by body iron content or requirement of the body iron.The body iron content is maintained by variation in the amount of iron absorbed. In the women, menstruation and child birth increase iron losses to about 2mg/dl.Iron absorption may not increase sufficiently to compensate for the iron losses and these may lead to the development of iron deficiency anaemia (Lewis *et al*, 2006).

1.9 Hemoglobin:

Hb is predominant protein in the red blood cell and is responsible for transporting oxygen, carbon dioxide and protein between the lung and tissue (Bhagaran and Eun Ha,2011). Hemoglobin (mw 64,500dalton) is composed of haem (consisting

of iron and protoprophyrin) and globin.The globin portion of the molecule consist of four or two pairs of polypeptide chain. One haem group is bound to each poly peptide chain. Hemoglobin is not homogenous and normally different variant exist such as A, A2, F, Gower1, Gower 2and Portland. The last three are present in varying proportion during fetal and adult life.

The relative proportion of different hemoglobin are adult-Hb A97%, Hb A2 2-5%, and HbF 0.5%. newborn –Hb F 80% and Hb A20%.

Hemoglobin A (Hb A), the principle hemoglobin of adult, consist of pair each of alpha and beta, poly peptide chain and it's structure is designated as alpha2 beta2.Fetal hemoglobin (HbF) the predominant hemoglobin in fetal life, contain pair of alpha and pair of gamma chain .two type of gamma chain are distinguished which are different amino acid either (glycine or alanine) at position 136. During the embryonic life, there are three hemoglobin Gower1,Gower2 and Portland.

1.9.1 Hb reference range:

Adult males: 13.0-17.0 g/dl, Adult female (non pregnant): 12.0-15.0g/dl, Adult female (pregnant): 11.0-14.0g/dl,Children 6-12 years: 11.5-15.5g/dl,Children 6 month -6years: 11.0-14.0g/dl,Infant 2-6 months 9.5-14.og/dl andNew born 13.6-19.6g/dl (Kawthalkar,2013).

1.10 Anaemia :

Anaemia is decreased in the total amount of red blood cell (RBCs) or hemoglobin in the red blood cell .or lowered ability of the blood to carry oxygen. or it defined as decreased in the erythrocyte content or oxygen- carring capacity of blood as consequence of drop in packed cell volume (PCV), red cell count, or hemoglobin concentration to less than the lower limit of the reference interval .Anaemia is most

common blood condition in the U.S. it affect about 3-5 million American.Women, young children and people with chronic disease are at increased risk of anaemia. Certain forms of anaemia are hereditary and infant may be affected from the time of birth, women in the child bearing years are particulary susceptible to iron deficiency anaemia because of the blood loss from the menstruation and the increased blood supply demand during pregnancy.Older adult also may have greater risk of developing anemia because poor diet and other medical condition .Anaemia has usually been classified as mild, moderate, sever and very sever based on PCV values and microcytic , normocytic and macrocytic and hypochromic, normochromic and hyper chromic based on mean corpuscular volume (MCV)and mean corpuscular hemoglobin concentration (MCHC) (Padalino et al ,2016).

1.10.1Classification of anaemia:

Three group are distinguished

1. Microcytic , hypochromic(MCV<78fl).
2. Normocytic,normochromic.
3. Macrocytic(MCV>100).

1.10.1.1Microcytic anaemia:

measurement of serum iron, total iron binding capacity and ferritin, bone marrow aspirate with staining for iron ,stool for malabsorption, endoscopic examination with biopsy.

1.10.1.2 Macrocytic anaemia:

recognition of the morphological red cell changes of alcohol excess and liver disease is vital, as is increased polychromasia of reticul ocytosis .where macrocytic megaloblastic erythroid maturation is demonstrated.

1.10.1.3.Secondary anaemia:

The haematologist's principle task exclude a primary haematological disorder such as secondary tumer, directly involving the marrow.These anaemia may be complicated by other factor such as blood loss or folate deficiency.

1.10.1.4. A plastic anaemia:

Bone marrow aspirate and trephine biopsy, ham's test for PNH, urine for haemosidrine, neutrophil alkaline phosphatase , vitamin B12 and folate level , chromosom studies radiology of hand and for arm if fanconi's anaemia is suspected, viral studies HLA typing if bone marrow transplantation is consideration and colony culture.

1.10.1.5.Haemolytic anaemia:

A haemolytic process may be suspected by the presence of red cell abnormalities, a reticulocytosis and an increased unconjugated bilirubin level (Dacie and Lewis,1995).

1.10.2 Symptom of anaemia:

The symptoms of anaemia depend upon the degree of reduction in the oxygen – carrying capacity of the blood, change in the total blood volume, the rate in which these change occurs ,the degree of severity of the under lying disease contributing

to anaemia, and the power of the cardiovascular and hematopoietic system to recuperate and compensate (Maakaron and Joseph,2016)

1.11 Iron deficiency:

1.11.1Definition and history:

Iron deficiency is the state in which the content of iron in the body is less than normal. it occurs in varying degree of severity. Iron deficiency and iron deficiency anaemia are common nutritional and hematological disorder in north America and worldwide (Beutler *et al*, 2011).

The term of iron deficiency can be applied to lake of iron that is sever enough to impair the production of red blood cell put not necessary to the extent that the hemoglobin concentration falls below the normal reference range .

It affects roughly a third of the world's population, half the cases are due to iron deficiency. It is major and global public health problem that affect maternal and child mortality, physical performance and referral to health-care professionals. Children age 0-5 years, women of child bearing age and pregnant women are particularly at risk. Several chronic disease are frequently associated with iron deficiency anaemia-notably chronic kidney disease, chronic heart failure, cancer and inflammatory bowel disease (Lopez et al, 2016).

In infant and young children iron deficiency is most commonly due to insufficient dietary iron. In young women it is most often the result of blood loss in menstruation or as result of pregnancy. In older adult bleeding may be from the gastrointestinal tract, as form of hemorrhoid, bleeding peptic ulcer, colon cancer or angiodysplasia. It may result from uterine leiomyomas or carcinoma or renal tumor. Iron deficiency has adverse affects on activity of numerous enzyme and in

infant can result in impairment of growth and intellectual development. The haematologic features of iron deficiency are non specific and too often confused with other causes of microcytic anaemia such as thalassemias, chronic disease, renal neoplasm and other disorder. A low serum ferritin concentration is an excellent indicator of iron deficiency .iron deficiency anaemia is most advance stage of iron deficiency. It characterized by decreased or absent iron store, low serum iron concentration, low transferrin saturation and low hemoglobin concentration or hematocrit level. In certain rare disorder, such as idiopathic pulmonary hemosiderosis or paroxysmal nocturnal hemoglobin urea (Beutler *et al* , 2011).

1.11.2 Clinical Manifestation of iron deficiency:

The clinical picture varies greatly from one case to another, and it is produced both by the anaemia itself and by the lack of iron, which is essential for cellular energy metabolism. Symptoms depend greatly on the speed of onset of anaemia. Thus, ID can be detected in an Asymptomatic individual or in person with symtoms that include general weakness, Fatigue, irritability, poor concentration, headache, and intolerance to exercise. Some iron deficient patients, with or without anaemia, might have alopecia, atrophy of lingual papillae, or dry mouth due to loss of salivation. Other symptoms, such as weakness or digging fingernails (koilonychia), chlorosis, or the syndromes of plummer- Vinson or Paterson –kelly (dysphagia with esophageal membrane and atrophic glossitis).(Bermejo and Lopez,2009).

1.11.3Laboratory finding of iron deficiency anaemia:

1. Red cell indices and blood film

The red cell indices fall and they fall progressively as the anaemia become more severe. The blood film shows hypochromic,microcytic cell with occasional target cell and pencil-shaped poikilocytes the reticulocyte count is low in relation to the degree of anaemia. A dimorphic blood film is also seen in patient with iron deficiency anaemia who have received recent iron therapy and produced a population of new haemoglobinized normal –sized red cell and when the patient has been transfused. The platelets count is often moderately raised in iron deficiency.

2. Bone marrow iron:

In iron deficiency anaemia there is complete absence of iron from stores (macrophages) and from developing erythroblast.

3.Serum iron and total iron biding capacity:

The serum iron falls and total iron binding capacity (TIBC) rises

4.Serum ferritin:

In iron deficiency anaemia the serum ferritin is very low while a raised serum ferritin indicates iron over load or excess release of ferritin from damaged tissue or an acute phase response (e.g. in inflammation) (Hoffbrand and Moss,2016).

1.11.4 Treatment of iron deficiency anaemia:

Treatment of patient with iron deficiency anaemia should be focused on addressing the underlying cause of iron deficiency and replenishing iron stores. Increasing dietary iron intake alone is insufficient to treat cases of established iron deficiency. First line therapy involves oral iron replacement in doses of 100 to 200mg of elemental iron in two to three divided doses in adult and 3 to 6 mg/kg of element iron in children. Reticulocytosis should occur within 72 hours of therapeutic iron replacement and haemoglobin level should rise by about 20g/l every three weeks. It is recommended that iron supplementation continue for three to six months once the haemoglobin level has normalized to improve iron stores. gastrointestinal disturbance including nausea and constipation are responsible for the major side effects and in patient with mild iron deficiency the dosing internal can be increased to every second day to minimize these complication (Clarke and Dodds,2014).

1.12 Anaemia of chronic disease:

Anaemia of chronic disease (ACD), sometimes knownas anaemia of inflammation, is second most common form of anaemia worldwide and is seen in variety of condition including cancer, autoimmune condition and infection (Cullis,J., 2013).

1.12.1Condition associated with anaemia of chronic disorder:

1.Chronic infection

- Pulmonary infection: abscesses, tuberculosis, pneumonia.
- Sub acute bacterial endocarditis
- Pelvic inflammatory disease
- Osteomyelitis
- Chronic urinary tract infection

- Chronic fungal disease
- Meningitis
- Human immunodeficiency virus

2. **Chronic non infectous inflammation**

- Rheumatoid arthritis
- Rheumatoid fever
- Systemic lupus erythromatous
- Sever trauma
- Thermal injury
- Adjuvant disease
- Sterile abscesses

3. **Malignant disease**

- Carcinoma
- Hodgkin disease
- Lympho sarcoma
- Leukemia
- Multiple myeloma

4. **Miscellaneous**

- Alcoholic liver disease
- Congestive heart disease
- Ischemic heart disease

5. **Idiopathic** (Lee *et al*,1999).

Table(2.1): Haematological feature of anaemia of chronic disease

1)haemoglobin	Not less than 9g/dl
2) mean corpuscular volume	Normal or mildly redused(ususlly 77-82fl)
3) mean corpuscular haemoglobun	usually normal occasionally redused
4) serum iron	Redused
5) total iron binding capacity	Redused
6)transferring saturation	Mildly redused
7) serum ferritin	Normal or increased
8) ser um and urine hepcidin	Raised
9)C-Rreactiv protein	Usually raised
10)erythrocyte sedimentation rate	Usually raised

(Hoffbrand *et al*,2015).

1.12.2 Pathophysiological features of anaemia of chronic disease:

Anaemia of chronic disease is immune driven, cytokines and cell of the reticuloendothelial system induce change in iron hemostasis, the proliferation of erythroid progenitor cell, the production of erythropoietin, and the life span of red cell, all of which contribute to the pathogenesis of anaemia. Erythropoiesis can be affected by disease underlying anaemia of chronic disease through the infiltration of tumor cell into bone marrow or of micro organism, as seen in human immunodeficiency virus, infection, hepatitis and malaria. Tumor cell can produce pro inflammatory cytokines and free radicals that damage erythroid progenitor. Anaemia with chronic kidney disease share same of the characteristic of anaemia of chronic disease, although the decrease in the production of erythropoitein, mediated by renal Insufficiency and the anti proliferative effect of accumulating uremic toxin, contribute importantly. In addition, in patient with end stage renal

disease, chronic immune activation can arise from contact activation of immune cell by dialysis membranes, from frequent episodes of infection, or from both factor (Weiss and Goodnough, 2005).

1.13 Previous studies:

Thise study was agree with previous study in sudan and the result showed that the serum iron and serum ferritin of the patient under dialysis were significantly higher as compared to healthy control group (p value 0.000) while the total iron binding capacity in haemodialysis patient were significantly lower as compared to healthy control individual (p value 0.00). (Osman and Ibrahim,2017) .

In 2017 performed the study in Khartoum state among eighty patient with end stage chronic renal failure treated with haemodialysis ,81% were found to have anaemia, while high s.ferritin level with more than 800Mg/L were found in 71.2% of patient (Taha and Omer,2017).

Ateam lead by Zadeh in the university of California , analyzed data for 5years (January 2007-december 2011) about the serum ferritin level in new haemodialysis patients and the result showed that the serum ferritin levels are increased over time (Zadeh *et al*,2011).

In 2011 performed study in al ribat hospital to determine the morphological pattern of anaemia and iron status among anemic chronic renal failure patients. And the result showed that the mean of hemoglobin (9.4g/dl) of case group were significantly lower than means of control group (p value <0.05).and hightly significant level of serum ferritin with mean 621Mg/L when compared to the mean of control group with mean 73Mg/L, p value (<0.05). (Abd araheem,2011).

Other study done in 2011 to evaluate the iron profile in 40 haemodialysis patients and reported that 30% 0f patients were normal ,70% iron were overload.(Canavese *et al*,2004).

1.14 Rationale:

chronic renal failure is considered as important health problem all over the world and also in sudan. And the patients with CRF are frequently subjected to anaemia of chronic disease, and the some patients which are treated with haemodialysis were not monitoring for the iron status suffering to high serum iron and high serum ferritin in order to iron over load, and these cases were detected in sudan. The aim of this study is to assess the impact of chronic renal failure disease on the diagnosis of anaemia, it is also aimed to study the iron profile for the patient with CRF under the study.

1.15 Objectives

1. General objective

To assess the iron profile and haemoglobin concentration in the patient with chronic renal failure undergoing haemodialysis .

2 Spesific objective:

1. To perform the iron profile in the patients with chronic renal failure under the haemodialysis

2. To evaluate the effect of dialysis in prognosis of anaemia.

Chapter Two

2-Materials and Methods

2.1 Study design:

this is case control study aimed to measure the iron profile among haemodialysis patient.

2.2 Study area:

Omdurman –sudan,omdurman military teaching hospital, the haemodialysis department.

2.3 Study population:

80 patients with renal failure under haemodialysis and 20 normal individual . 2.7

2.4 Sample size:

Total of 100 cases were chosen 80 with CRF under haemodialysis and 20 normal individual

2.5 Inclusion criteria:

For case group any patient diagnosis with renal failure under haemodialysis , and control group which is any patient free from renal failure.

2.6 Exclusion criteria:

Some subjects refuse to sampling, and any patients with renal failure but not under haemodialysis.

2.7 Tool of data collection:

Information from individual (case, control) was collected by using structured questionnaire (age, sex, medical history, family history, take iron supplement (if yes oral or intravenous) and duration of illness.

2.8 Methodology:

2.8.1 Sample collection:

Venous blood sample (5ml) was collected using sterile disposable plastic syringe after cleaning the veinopuncture area with 70% ethanol ,the sample was divided into two part, (2.5) of blood was drawn into plain tube. Serum separated by centrifuging blood for 10 minute at 3000RPM and then labeled with data name and identification number of volunteer participating in this study. And stored frozen at 4c for 3 days to biochemical analysis of serum iron and serum ferritin and total iron binding capacity on DIRUI CS-T240 auto chemistry analyzer. The other (2.5) of blood was added into ethylene diamine tetra-acetic acid (EDITA) use to analyze hemoglobin concentration was analyzed by using blood counter (sysmexKx21).

to break down RBCs and release of hemoglobin .mentioned pulses converts in to digital number using in build calculator programmed and designed for RBCs,

2.8.2 Estimation of serum iron:

2.8.2.1 Principle of the method:

Transferring- bound ferric ions in the sample are released by guanidinium and reduced to ferrous by means of ascorbic acid. Ferrous iron react with ferrozine forming a coloured complex that can be measured by spectrophotometry

2.8.2.2 Procedure:

The reagent was bring to room temperature and Pipette into labeled test tube

	Reagent blank	Sample blank	Sample	Standard
D.W	200M L	-	-	-
Sample	-	200ML	200ML	-
Iron standard	-	-	-	200ML
reagent	-	1.0ml	-	-
Working reagent	1.0ml	-	1.0ml	1.0ml

(Iron- Ferrozine,Biosystems Reagents).

Mixed thoroughly and the tube was let stand for 5 minutes at room temperature, then the absorbance of the sample blank was read at 560nm against D.W, and the absorbance of the sample and standard were read.

2.8.2.3 Calculations:

The iron concentration in the sample is calculated using the following general formula

$\frac{sample-sample\ blank}{stanard}$×standard = sample

2.8.2.4 Reference values:

- Men: 65-175 Mg/dl
- Women: 50-170 Mg/dl. . (Iron- Ferrozine,Biosystems Reagents).

2.8.3 Estimation of serum ferritin:

2.8.3.1 Principle of the method:

Serum ferritin causes agglutination of latex particles coated with anti-human ferritin antibodies. The agglutination of the latex particles is proportional to the ferritin concentration and can be measured by turbidimetry.

2.8.3.2 Procedure:

1.The working reagent and the instrument were brought to 37c,and the instrument was zeroed with D.W. then was pipetteed into acuvette,

Working reagent	1.0ml
Standard(s) or sample	30ML

(Ferritin Latex,Biosystems reagents).

The cuvette was mixed and inserted into the instrument and stopwatch was started,

The absorbance was recorded at 540nm after 10seconds(A1) and after 5minutes(A2).

2.8.3.3Referrence values:

Children: 7-140Mg/l

Men: 20-250Mg/l

Women: 20-200Mg/l. (Ferritin Latex,Biosystems reagents).

2.8.4 Estimation of total iron binding capacity:

2.8.4.1 Principle of the method:

Excess of Fe+3 is added to the sample to saturate serum transferrin. Uncomplexed Fe+3 is precipitated with magnesium hydroxide carbonate and the iron bonded to protein in the supernatant is then spectrophotometrically measured.

2.8.4.2 Procedure:

Into labelled tube was pipetted,

Sample	0.5ml
Reagent(A)	1.0ml

(TIBC,Biosystem reagents).

Mixed thoroughly and let standed for 5-30minutes at room temperature, One spoonful of reagent (B) was added to each tube, Mixed thoroughly and the tube let standed for 30-60 minutes at room temperature. During time mix thoroughly several times, Centrifuged at a minimum of 3000r.p.m. for 10minutes,Carefully the supernatant was collected and the iron concentration in the supernatant was measured.

2.8.4.3 Calculations:

Total iron binding capacity (TIBC)

TIBC=iron concentration in the supernatant×3(dilution)

Iron saturation

$$\frac{100\times SERUM\ IRON\ CONCENTRATION}{TIBC} = \text{IRON SATURATION(\%)}$$

2.8.4.4 Reference values:

TIBC:

Infant 100-400Mg/dl

Adult 250-425Mg/dl.(TIBC,Biosystem reagents).

2.8.5 Estimation of hemoglobin concentration:

2.8.5.1 Principle of sysmex KX21 hematological analyzer:

Measurement of blood cells and hemoglobin concentration obtained by aspirating of small volume of well mixed (EDTA) blood by sample probe and mixed with isotonic diluents in nebulizer. Diluents aspiration delivered or RBCs aperture bath for providing information about RBCs and platelet. Other portion of aspirated sample induced into WBCs bath in which hemolytic reagent (stromatolyzer) added to break down RBCs and release of hemoglobin .mentioned pulses converts in to digital number using in build calculator programmed and designed for RBCs, WBCs count come portion o diluted sample delivered to in build hemoglobin meter at the same time, hence three values directly measured (RBCs, TWBCs and Hb) and displayed on (LCD).

2.8.5.2 Procedure of sysmex:

- The reagent needed was checked and power switch was turned.

- Self-auto rinse, and back ground check was automatically performed and vend (vend for analysis) will appear.

- Whole blood mode was selected.

- Sample number and patient name were entered.

- Sample was mixed sufficiently.

- The tube was set to the sample probe, and in that condition the start switch was pressed.

- When the sucking of the sample was done, the tube was removed.

- After that automatic analysis was done and the result was displayed in the screen.

2.9 Ethical consideration:

This study was approved by college of medical laboratory science and ethical committees, permission from hospital manger was taken before beginning. Every sample was collected after verbal approval by the volunteer. Provide privacy and confidentially for every participant.

2.10 Data analysis:

The data were analyzed by using statistical package for social sciences (spss version16.0) for windows version (7)using T-independent test and the data presented in tables and graphs.

Chapter three

Results

One hundred (100) venous blood sample were collected in EDTA and plain blood collection tube for the participant, 80 of them with chronic renal failure (80 case,42 male and 38 female) and 20 health individual (20 controle,11 male and 9 female).

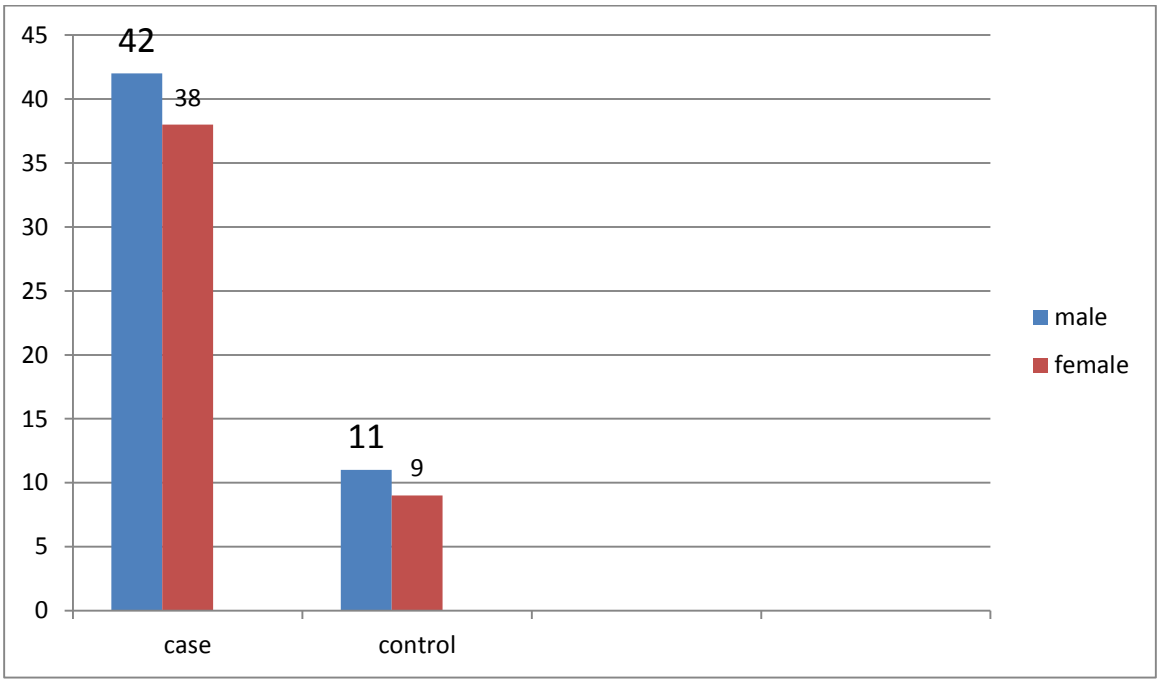

Figure3.1: Gender distribution among study groups.

The result showed that the age distribution among the study group (in case group ,39 of patients are adult and 41 are old) and (in control group10 patient are adult and 10 are old). Their aged ranged between 18-85years(adult group from 18 to 45years) and (old group from 46 to 85 years).

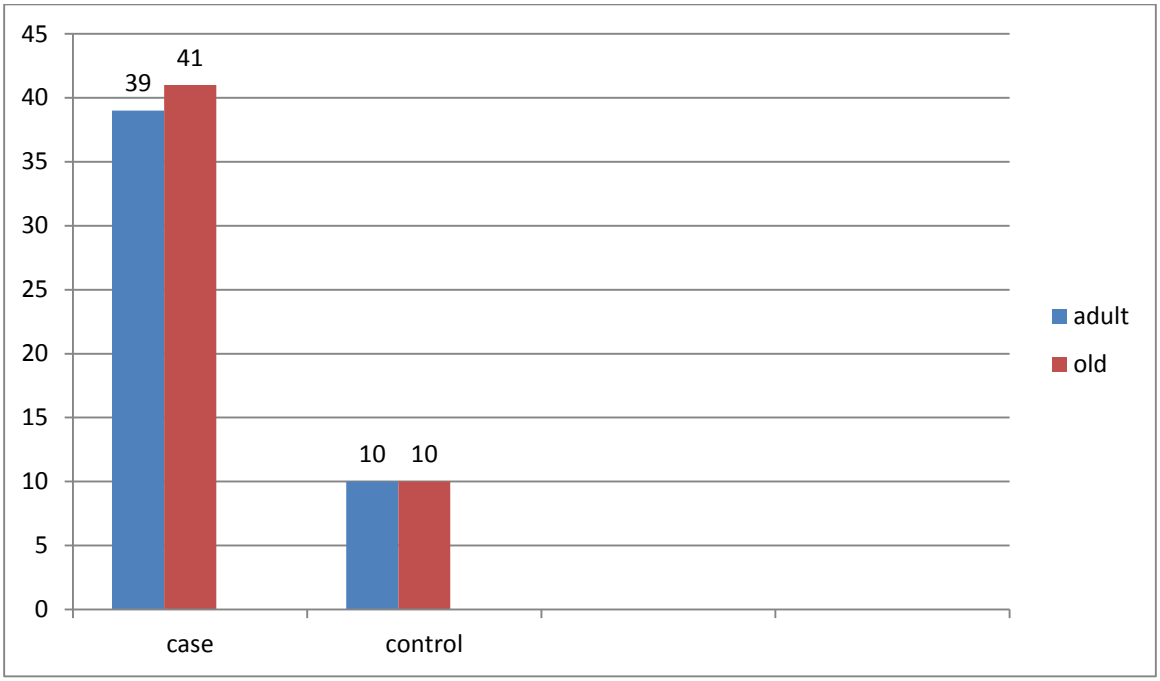

Figure3. 2 Age distribution among study group

The result show that the patients which take iron supplement (48% take iron supplement and 52% not take iron supplement).

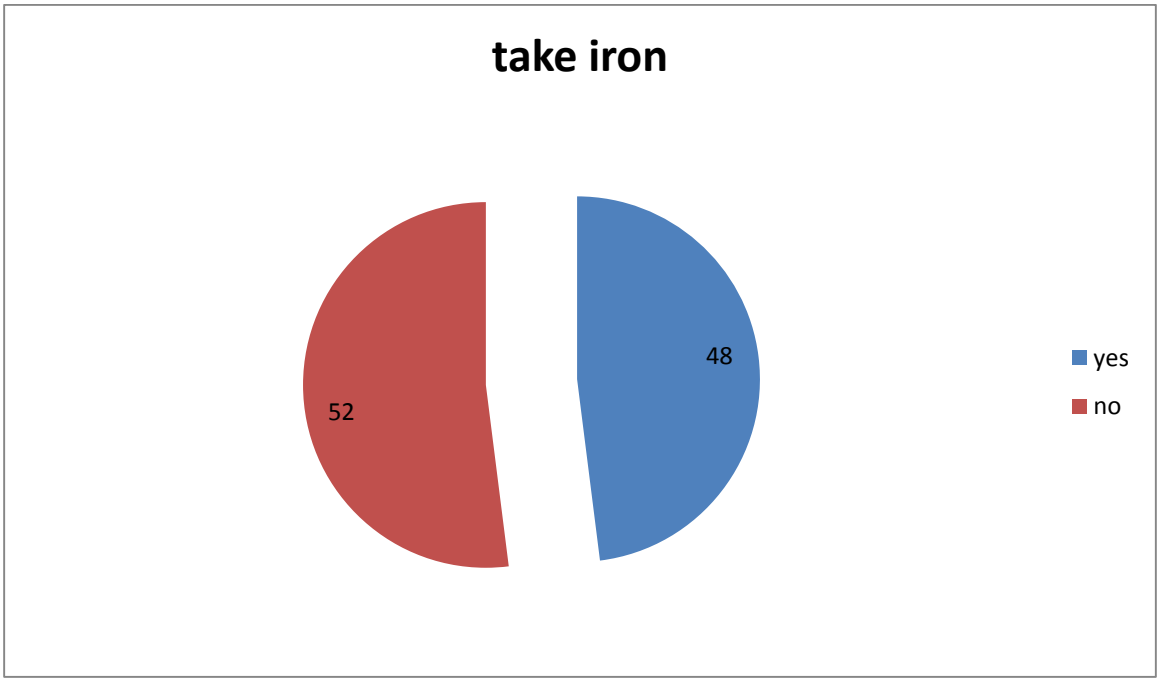

Figure3.3: Take iron supplement among the patients under the study.

The results show that 67.5% of the patients with CRF have short duration (the period of disease from 1-5years of illness) and 32.5% of patients have long duration (from 6-11years of illness).

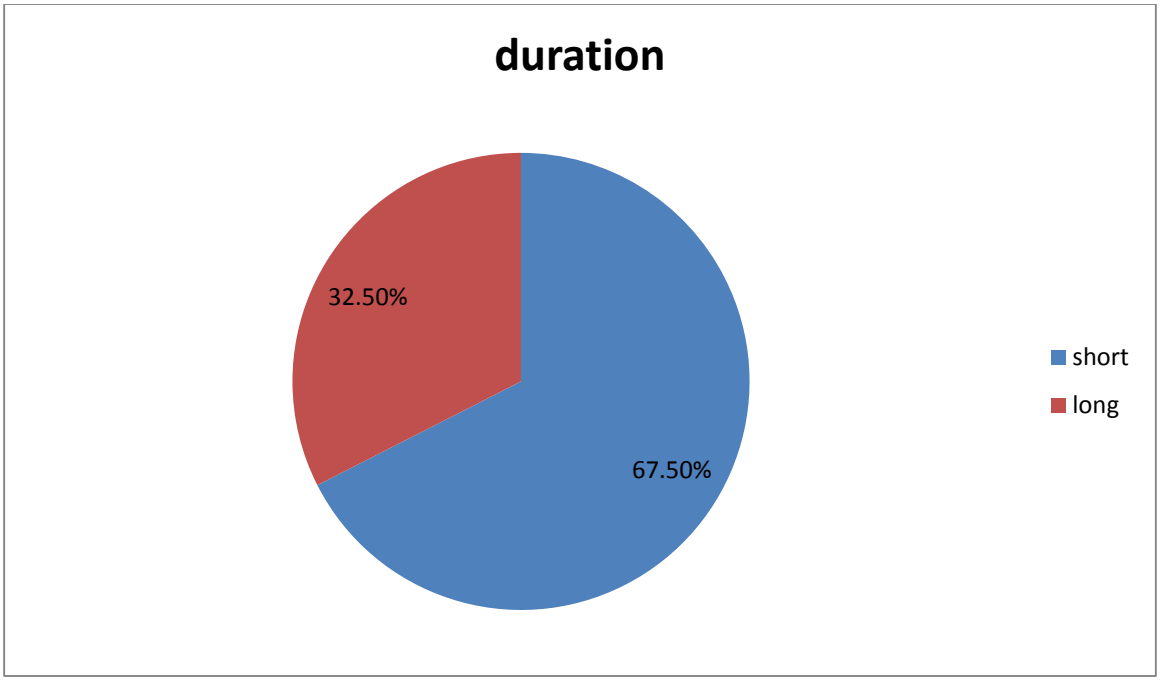

Figure 3.4: Duration of disease in the patient with CRF

The result show that the history of disease among the study group, 20% have history to CRF and 80% have no history to the disease.

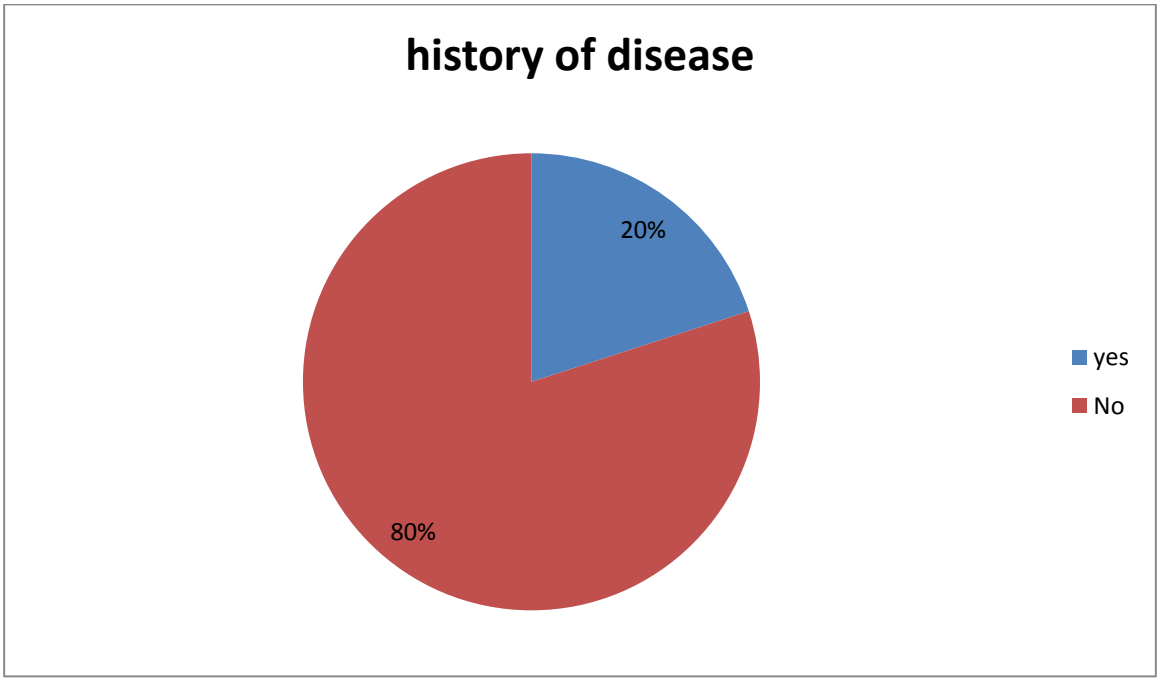

Figure3. 5: History of disease among the study group.

The results of the present study showed that the serum iron and serum ferritin of the patients under dialysis were significantly higher as compared to healthy control group (mean of S. iron in case=86.39, mean of S iron in control=66.0) (p.value 0.001).and (mean of S. ferritin in case= 615.54, mean of S.ferritin in control =29.55) (p.value 0.000).While the total iron binding capacity and hemoglobin in hemodialysis patient were significantly lower as compared to healthy control individual. (mean of TIBC in case =176.25, mean of TIBC in control =273.15).and (mean of HB in case= 8.96,mean of HB in control 13.29).(Pvalue0.000) for both as shown in table1.

Table3.3T. Test of study parameters in different study groups

	Group	N	mean	Std.deviation	p. value
s.iron	Case control	80 20	86.39 66.05	52.15 6.22	0.001
s.ferritin	Case control	80 20	615.54 29.55	245.82 5.61	0.000
TIBC	Case Control	80 20	176.25 273.15	69.89 13.39	0.000
HB	Case Control	80 20	8.964 13.290	1.51 0.55	0.000

The results of correlation of the study parameters and the gender of the patient showed no statistically significant association between gender of the patient and serum iron, serum ferritin, TIBC and hemoglobin with p.value(0.325,0.194,0.403,and 0.881) respectively as shown in table2 .

Table 3.4.Correlation between study parameters and gender of patients

	Gender	N	mean	Std.deviation	p.value
s. iron	Male female	53 47	86.47 77.34	51.98 41.57	0.325
s. ferritin	Male female	53 47	537.89 453.74	321.29 320.47	0.194
TIBC	Male female	53 47	201.47 189.04	79.06 67.67	0.403
HB	Male female	53 47	9.860 9.794	2.33 2.10	0.881

The result of the association of the study parameters and the duration of hemodialysis showed no statistically significant association between duration of the dialysis and s. iron, s. ferritin, TIBC and HB.with p.value(0.435,0.922,0.883,0.515) respectively as shown in table 3.

Table3.5.Correlation of study parameters and duration of dialysis

	Duration	N	mean	Std.deviation	p.value
s.iron	Short long	54 26	83.20 93.00	46.02 63.53	0.435
s.ferritin	Short long	54 26	617.43 611.62	259.25 220.09	0.922
TIBC	Short long	54 26	177.06 174.58	61.68 85.83	0.883
HB	Short Long	54 26	9.041 8.804	1.59 1.35	0.515

The correlation results between the study parameters and age of the patient showed no statistically significant association between the age of the patient under the study and the s.iron, s.ferritin, TIBC and HB with p.value(0.693,0.99,0.983and 0.561) respectively as shown in table4.

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Table3.6.Correlation of study parameters and age of patients

		N	Mean	Std.deviation	p.value
s.iron	Adult old	49 51	84.24 80.47	57.77 35.11	0.693
s. ferritin	Adult old	49 51	444.04 550.51	323.02 315.49	0.99
TIBC	Adult old	49 51	195.47 195.78	82.15 65.67	0.983
HB	Adult old	49 51	9.961 9.702	2.18 2.26	0.561

Chapter four

4. Discussion conclusion and recommendation

4.1 Discussion:

Chronic kidney disease (CKD) is an irreversible progressive reduction in renal function and main source of long term morbidity and mortality.This study was carried in Omdurman Military Teaching Hospital ,Sudan. And aimed to assess the iron profile and hemoglobin concentration among chronic renal failure in Sudanese patient undergoing hemodialysis. Its included 80 patient known diagnosed with chronic renal failure and 20 normal individual as control group. The results of the study showed increased level of serum iron (86.39±52.148,p value0.001) ,serum ferritin (615.54±245.8,p value0.000). and lowered the level of TIBC (176.25±69.8,

P value 0.000) and hemoglobin concentration (8.96±1.5,p value 0.000). And the present study showed that no statistically significant association between gender of the patient and S.iron,S.ferritin ,TIBC and HB with p value (0.32,0.19,0.40 and 0.88).and also showed no statistically significant association between duration of dialysis and S.iron,S.ferritin,TIBC and HB with p value (0.44,0.92,0.88,0.52).and also there is no statistically significant association between the age of the patient under the study and the S.iron,S.ferritin,TIBC and HB with p value (0.69,0.99,0.98 and 0.5)respectively.These study was agree with other study in sudan by Ibrahim,I.K and Osman,A.A and the result showed that the S.iron and S.ferritin of The patients under dialysis was significantly higher as compared to healthy control group (p value 0.000).While the TIBC were significantly lowered as compared to healthy control group (p value 0.000).(Osman A.A and Ibrahim,I.K,2017).And also agree with other study in sudan by Taha and Omer,R.T .in 2107 which are performed the study among eighty patients with end stage CRF treated with

haemodialysis ,81% were found to have anaemia, while high S.ferritin level with more than 800Mg/l were found in 71.2% of patients (Taha,Omer,R.T,2017).Other study done in California by team lead by Kalentar,K, Analyzed data for 5 years about s.ferritin level in haemodialysis patients and the result showed that the s.ferritin levels are increased over time (Kalentar.K,2017). Other study done by Canavese C,Bermago D and Ciccone G in 2004 who was estimate the iron profile in 40 transfused hemodialysis patients and reported that 30% of the patients were normal and 70% iron were over load.And also other study by Hassan,W.A. In 2011 performed study in Alribat hospital to determine the morphological pattern of anaemia and iron status among anemic CRF patients. And the result showed that the mean of haemoglobin (9.4g/dl) of case group were significantly lower than mean of control group(p value<0.05). and highly significant level of s.ferritin with mean 621Mg/l, p value (0.05).(Hassan, W.A,2011). In patient with chronic kidney disease the kidneys are damaged and stop to production of EPO which lead to the bone marrow decreased the production of red blood cells and that lead to anemia and the loss of blood during the hemodialysis process is also one caused of anemia.To avoid the anemia during hemodialysis need to frequent blood transfusion and iron supplement therapy which lead to iron over load.

4.2 Conclusion:

The study concluded that the chronic renal failure patients under haemodialysis presented with increase level of serum iron and serum ferritin while the total iron binding capacity and haemoglobin was decreased and these changed are not influenced by the duration of dialysis and gender of disease.

4.3 Recommendations:

- Patients with renal failure should received iron supplement and erythropoietin to avoid anaemia.
- Erythropoietin and haemoglobin must be regularly measured in chronic renal failure.
- The iron profile should be done every three month routinely to the patient with CRF under haemodialysis to monitor the iron status and avoid the hazard of blood transfusion and iron over load.

Reference

Abd araheem, W.,(2011), evaluation of haematological parameters and iron status among renal failure patients,Sudan.

Allen,R., and Nissenson,(1999)Iron deficiency in patient with renal failure, *kidney international*,**55(69)**, page18-21.

Arora,S. and Kapoor,R,K.(2012), Iron metabolism in human, *Department of biochemistry,ESI post graduate institute of medical science*, ISBN:967-953-15.1605-0.

Bermejo,F. and Lopez,S.G.,(2007),A guide to diagnosis of iron deficiency and iron deficiency anaemia in digestive diseases, *World Journal of Gastroenterology*, **15**(37):4638.

Beutler, E., Lichtman,M.A., Coller B., Kipps,T., Seligsohn,U.,(2011), Williams haematology, 6edition, New York,Mc graw-Hill , P447.

Bhagavan,N.V.and Eun-Ha,C.,(2011), Essential of Medical Biochemistry, 1st ed,USA, Elselvier,p355.

Brunner and Suddarth's,(2014),Text book of medical –Surgical nursing, 13th ed

,India, wolters kluwer ,P1535,1548.

Burtis,**C.A** ,Ashwood,E.R .and Bruns,D.E.,(2008),fundamental of clinical chemistry, 6th edition, USA, Saunders Elsevier, p652.

Canavese, C., Bergamo D and CicconeG,(2004),harmful iron overload in dialysis patients , *kidney international*,65(3),P1091-1098.

Clarke,L.,Dodds,A.J,(2014), iron deficiency anaemia,*Medicine today*,**15**(11): P41.

Cullis,J.(2013),anaemia of chronic disease, *clinical medicin*,**3**(2):193-6.

Dacie,J.V and Lewis,S.M.,(1995),Practical haematology, 8thed,USA, Churchill-livingstone, .P192,193.

Daugirdas,G.T., Blake,P.G. and Ing,T.(eds),(2015), handbook of dialysis, 5th ed, Philadelphia.Wolter Kluwer.p14,321.

Hoffbrand,A.V. and Moss,P.A.H.,(2016), hoffbrand's Essential Haematology,7th ed, UK,by John Wiley and Sons, p30.

Hoffbrand,A.V, Catovsky,D. and Tuddenham,E.G.D.(eds),(2015),Post Graduate Haematology, 5thed ,Australia, black well ,p1012.

Kariyanna,S.S, Light,R.P. and Agarwal,R., (2010). Alongitudinal study of kidney structure and function in adult, *nephrology dialysis transplantation*. **25**(4):1120-1126.

Kawthalkar,S.M.(2013). Essential of haematology.2 nd ed, India, Japee Brothers, P8,519.

Lam,Q.(2013). Common sense pathology,1st ed,Australian, cirrus media.p3.

Lee,G.R., Foerster,J., Lukens,J.,Paraskevas,F.andRodgers,G.M.,(1999),wintrobe's clinical haematology,10 thed, New York, Lippincott Williams and wilkins,p1012.
Levey, A.,Eckardt, K. and Tsukamoto Y. (2005),Definition and Classificaton of chronic kidney disease,*Kidne international*,**67**(6):2089-100.

Lewis,S.M., Bain,B.J. and Bates,I.,(2006). Dacie and lewis Practical Haematology

, 10thed, USA,Churchill living stone Elsevier,P132.

Lopez,A.,Cacoub,P., Macdougall,L.and peyrin,L.,(2016). Iron deficiency anaemia,*The lancet,* **387**(10021**)**, p907- 916.

Maakaron,A and Joseph ,M.(2016),Anaemia: practice essential pathology , etiology, 3rd ed,p 143.

Marshall,**W.J** and Bnagert ,S.K.,(2008). Clinical chemistry,6 ed,London,

Mosby Elsevier,p69.

O'mara,N.B.,(2008). anaemia in patient with chronic kidney disease ,*diabetes spectrum*, **21**(1),P12-19.

Osman,A.A. and Ibrahim,I.k,(2017) Assessment of iron profile among Sudanese patients with chronic renal failure undergoing haemodialysis ,*international journal of information research and review* ,**4**(8)p.4393-4395.
Padalino,B.,Rubino,G.,Lacinio,R.and Pettazzi,F.,(2016), Anew classification to diagnose type of anaemia in standard horses, *equine veterinary science,**3(4)**,P21-25*.

Singh,I,B.,(2008), The E ssential of Anatomy, 2nd ed, India, jaypee brother,p269.

Snell,R.,(2012).clinical anatomy by regions. 9th ed,USA, Lippincott Williams and wilkins,P207.

Taha and roa taha omer,(2017), iron level among patients with chronic renal failure on haemodialysis ,Khartoum,Sudan.

Thomas,R.,Kanso,A. and Sedor,J.R,(2009). Chronic kidney disease and its complications,*Prim Care Clin Office Pract*,35(2), p329.

Waugh,A., and Grant,A.,(2001), Anatomy and Physiology in Health and illness, 9th ed, London,WIT 4Lp, p314.

Weiss,G. and Goodnough,L.T,(2005),Anaemia of Cronic Disease ,*New England Journal Of Medicine*,**352**(10):1011-23.

Zedeh,K.K.,Rodriguez,R.A. and Humphreys,M.H.,(2004),Association between serum ferritin and measures of inflammation, nutrition and iron in haemodialysis patients, *Nephrology DialysisTransplantation,***19(**1) ,p141-149.

Appendix

Sudan university of science and technology

Collage of medical laboratory sciences

Questionnaire

Patient No( )

General information:

Age……..

Gender: male …….. female……

If hava a history of disease yes…… no…….

Duration of hemodialysis……..

take ion supplement yes……. No…….

if yes iv…….. oral……

اقر انا.....بالموافقه على اخذ عينه من الدم لاجراء فحص الحديد وقياس كميه الهيموجلوبين, علما بانه تم اطلاعي على جميع اهداف البحث.

التوقيع..... رقم الهاتف.....

Results:

HB concentration…….

Iron profile:

s.iron…… s.ferritin…… TIBC…….