Measurement of Red Cell Distribution Width in End Stage Renal Disease Patients in Alribat Hospital in Sudan

قياس عرض توزيع خلايا الدم الحمراء لدى مرضى الفشل الكلي المزمن في مستشفى الرباط في السودان

A dissertation submitted for partial fulfillment for M.Sc degree in haematology

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Dedication

To my mother

The love and kindness umbrella

The sacrifice and my support in my live

To all my lovers

To all patients suffering from renal failure

Noah
Acknowledgment

In the beginning my thanks to almighty Allah who give me the strength and patience to accomplish this work. Great gratefulness and sincerity should be offered to Dr. Mubarak Alsaeed Al karsani, the supervisor of this work for his advice valuable appreciated help, support and guidance during this study. Also Dr. Abu Elgasim Abass Awad Elkareem for support and help.

My appreciation and thanks are also extended to the staff of dialysis center at Alribat Hospital.

Also my thanks to all patients with end stage renal disease, finally thanks to the teaching staff of Sudan University for Science and Technology Faculty of medical laboratory Science.
Abstract

This case control study was carried out at Alribat university hospital in the renal dialysis unit to measure red cell distribution width of patient with end stage renal disease.

Aim of this study to measurement of red cell distribution width in the patients of End stage renal disease in haemodialysis unit.

84 samples were collected from the patients with end stage renal disease (ESRD), also 66 samples were collected from apparently healthy as controls individuals, 2.5 ml of blood was collected from each patient and control, automated haematological analyzer (sysmex KN-21N) was used to measure red cell distribution width. Informative demographic data of sex, age were collected using a questionnaire during the period of March to May 2014.

Age in this study was divided into three groups 20-40, 40-60 and 60-80.

The study result revealed that red cell distribution width was significant elevated on the end stage renal disease patient the mean value of patient with end stage renal disease 15.1% . the mean of RDW in healthy people (as control) was 13.1%.

The study concluded that there is significant effect of RDW on the end stage renal disease patient.

No significant effect of age on RDW of end stage renal disease patient. No significant effect of sex on RDW of end stage renal disease patient.
مستخلص البحث

هذه دراسة مقارنة لمجموعات ضابطة مع حالات مرضية تم إجراؤها بمستشفى الرباط الجامعي
في وحدة الغسيل الكلوي لقياس عرض توزيع خلايا الدم الحمراء لمرضى الفشل الكلوي المزمن
الخاضعين للغسيل الكلوي.

الغرض من الدراسة هو قياس عرض توزيع خلايا الدم الحمراء للمرضى الذين يعانون من
مرض الفشل الكلوي المزمن تحت الغسيل في وحدة غسيل الكلى.

تم اخذ 84 عينة دم من المرضى الذين يعانون من الفشل الكلوي المزمن و66 عينة تم اخذها من
أشخاص أصحاء كمجموعة ضابطة وجمع 2.5 مل من الدم من المرضى والأصحاء واستعمل
المحلل الالكتروني (sysmex KX-21N) لقياس تعداد الدم الكلي وقياس عرض توزيع خلايا
الدم الحمراء وتم جمعها من خلال استبيان المعلومات الديموغرافيه حول الجنس والعمروفرة
الغسيل في الفترة الزمنية من مارس الى مايو 2014.

نتيجة الدراسة اشارت إلى ان متوسط قيمة عرض توزيع خلايا الدم الحمراء لمرضى الفشل
الكلوي المزمن كان 15.1% وانه اعلى من متوسط قيمة عرض توزيع خلايا الدم الحمراء
للاصحاء كمجموعة ضابطة الذي كان 13.1%.

وقد قسمت الدراسة وفق الاعمار الى ثلاث مجموعات هي (20-40), (40-60), (60-80).

وقد وجد أنه لا يوجد علاقة بين عمر المريض واختلاف قيم عرض خلايا الدم الحمراء وانه
لا يوجد اختلاف مؤثر في تلك القيم تبعا للجنس لمرضى الفشل الكلوي المزمن في وحدة غسيل
الكلى.
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Abbreviations

CBC: Complete Blood Count
CKD: Chronic Kidney Disease
CSF: Colony Stimulating Factor
CV: Coefficient of Variation
EDTA: Ethyle Diamine Tetra Acetic acid
ESRD: End Stage Renal Disease.
HSC: Haematopoietic Stem Cell
G-M CSF: Granulocyte Colony Stimulating Factor
GFR: Glomerular Filtration Rate
RCDW: Red Cell Distribution Width
SD: Standard Deviation
SCF: Stem Cell Factor
CHAPTER ONE

INTRODUCTION AND LITERATURE REVIEW
CHAPTER ONE
Introduction and literature review

1. Introduction

1.1 Blood Components:
Blood is specialized body fluid that delivers necessary substances to the body. Cells-such as nutrient and oxygen-and transports waste products away from the same cells (Franklin, et al.2009).
Invertebrates, it composed of blood cells suspended in liquid called blood plasma, which constitutes 55% of blood fluid, is mostly water (91% by volume), and contains dissolved proteins, glucose, mineral ions, hormones, carbon dioxide (plasma being the main medium for excretory product transportation), platelets and blood cells themselves. The blood cells present in blood are mainly red blood cells (also called RBCs or erythrocytes) and white blood cells, including leukocyte and platelets. The most abundant cells in vertebrate blood are red blood cells. These contain hemoglobin, an iron-containing protein, which facilitates transportation of oxygen by reversibly binding to this respiratory gas and greatly increasing its solubility in blood. In contrast, the carbon dioxide is almost entirely transported extracellular dissolved in plasma as bicarbonate ion (Franklin, et al.2009).

1:2 Literature Review:
Blood circulated around the body through blood vessels by the pumping action of the heart. In animals with lungs, arterial blood carries oxygen from the inhaled air to the tissues of the body, and venous carries carbon dioxide, a waste product of metabolism produced by cells, from the tissues to the lungs to be exhaled. (Franklin, et al.2009). Blood performed many important functions within the body including; Supply of oxygen to tissues (bound to hemoglobin, which is carried in red cells) Supply of
nutrient such as glucose, amino acid, and fatty acid (dissolved in blood or bound to plasma proteins (e.g., blood lipids). Removal of waste such as carbon dioxide, urea, and lactic acid. Immunological function, including circulation of white blood cells, and detection of foreign material by antibodies. Coagulation, which is one part of the body's self-repair mechanism (blood clotting after an open wound in order to stop bleeding). Messenger functions, including the transport of hormone and the signaling of the damage. Regulation of body PH. Regulation of core body temperature. Hydraulic function (Franklin, et al. 2009).

1.2.1 Red blood cells:
Human red blood cells (also referred to as erythrocytes) are the most common type of blood cells and the vertebrate organism's principal means of delivering oxygen (O2) to the body tissues via the blood flow through the circulatory system (Erich, et al. 1995). They take up oxygen in lungs or gills and release it while squeezing through the body's capillaries. These cells cytoplasm is rich in hemoglobin, an iron-containing biomolecule that can bind oxygen and is responsible for the blood's red cells (Erich, et al. 1995).

In humans, mature red blood cells are flexible biconcave disks that lack a cell nucleus and most organelles. 2.5 million new erythrocyte and produced per second (Erich, et al. 1995). The cells develop in the bone marrow and circulate for about 100-120 days in the body before their component are recycled by macrophages. Each circulation takes about 20 second. A proximately a quarter of the cells in the human body are red blood cells (Pierigè, et al. 2008).

1.2.2 Haematopoiesis:
Haematopoiesis is the formation of blood cellular components. All cellular blood components are derived from haematopoietic stem cell in healthy adult person, approximately $10^{11} \text{ to } 10^{12}$ new blood cells are
produced daily in order to maintain steady state levels in the peripheral circulation (Alenzi, et al.2009).

1.2.3 Haematopoietic stem cells (HSCs):
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. HSCs are self renewing: when they proliferate, at least some of their daughter cells remain as HSCs, so the pool of stem cells does not become depleted. The other Daughters of HSCs, (myeloid and lymphoid progenitor cells), however can each commit to any of the alternative differentiation pathways that lead to the production of one or more specific types of blood cells, but cannot self-renew. This is one of the vital processes in the body (Alenzi, et al.2009).

Locations: of haematopoiesis in developing embryos, blood formation occurs in aggregates of blood formation occurs in the spleen, liver and lymph node. When bone marrow develops, it eventually assumes the task of forming most of the blood cells for entire organism. However, maturation, activation, and some proliferation of lymphoid cells occur in secondary lymphoid organs (spleen, thymus, and lymph nodes). In children, haematopoiesis occurs in the bone marrow of the long bones such as femur and tibia. In adults, it occurs mainly in the pelvis, cranium, vertebrate, and sternum. In some vertebrates, haematopoiesis can occur wherever there is a loose stroma of connective tissue and slow blood supply, such as the gut, spleen, kidneys or ovaries. In some cases, the liver thymus, and spleen may resume their haematopoiesis function, if necessary. This is called extramedullary haematopoiesis. It may cause these organs to increase in size substantially. During fetal development, since bones and thus the bone marrow, develop later, the liver function as the main haematopoiesis organ. Therefore, the liver is enlarged during development (Alenzi, et al.2009).
1.2.4 Maturation:
As a stem cell matures it undergoes changes in gene expression that limit the cell types that it can become and moves it closer to a specific cell type. These changes can often be tracked by monitoring the presence of proteins on the surface of the cell. Each successive change moves the cell closer to the final cell type and further limits its potential to become a different cell type (Alenzi, et al.2009).

1.2.5 Determination:
Cell determination appears by the location of differentiation. For instance, the thymus provides an ideal environment for thymocytes to differentiate into variety of different functional of T cells. For the stem cells and other undifferentiated blood cells in the bone marrow, the determination is generally extoplained by the determination theory of haematopoiesis, saying that colony stimulating factors and other factors of the hematopoietic microenvironment determine the cells to follow certain path of cell differentiation (Alenzi, et al.2009).

This is the classical way of describing haematopoiesis. In fact, however, it is not really true. The ability of bone marrow to regulate the quantity of different cell types to produce is more accurately explained by a stochastic theory. Undifferentiated blood cells are determined to specific cell types by randomness. The hematopoietic microenvironment prevails upon some of the cells to survive and some, on the other hand, to perform apoptosis and die. By regulating this balance between different cells types, the bone marrow can alter the quantity of different cells to ultimately be produced (Alenzi, et al.2009).

1.2.6 Hematopoietic growth factors:
The proliferation and self-renewal of these cells depend on stem cell factor (CSF). Glycoprotein growth factor regulate proliferation and maturation of the red cells that enter the blood from the marrow, and cause cells in one or more committed cell lines to proliferate and mature. Three more factors that stimulate the production of committed stem cells are called colony-stimulating factors (CSF) and include granulocyte-macrophage CSF (GM-CSF), granulocyte CSF (G-CSF) and macrophage CSF (M-CSF). This stimulate match granulocyte formation and are active on either progenitor cells or end product of cells. Erythropoietin is required for myeloid progenitor to become erythrocyte. On the other hand, erythropoietin makes myeloid progenitor cells differentiate to megakaryocyte (thrombocyte formation cells) (Alenzi, et al.2009).

1.2.6 Transcription factor:
Growth factors initiate signal transduction pathway, altering transcription factors, that, in turn active genes that determine the differentiation of blood cells (Alenzi, et al.2009).

1.2.7 Red Blood Cell Distribution width (RDW):
RDW is a measure of the variation of red blood cell (RBC) volume that is reported as part of a standard complete blood count. Usually red blood cells are a standard size of about 6-8 μm. Certain disorders, however, cause a significant variation in cell size. Higher RDW values indicate greater variation in size. Normal reference range in human red blood cells is 11.5-14.5%. If anemia is observed, RDW test results are often used together with mean corpuscular volume (MCV) results to determine the possible causes of the anemia. It is mainly used to differentiate an anemia of mixed causes from an anemia of a single cause. Deficiencies of Vitamin B₁₂ or folate produce a macrocytic anemia (large cell anemia) in which the RDW is elevated in roughly two-thirds of all cases; however, a varied size distribution of red blood cells is a hallmark of iron deficiency.
anemia, and as such shows an increased RDW in virtually all cases. In the case of a mixed iron and B$_{12}$ deficiency, there will normally be a mix of both large cells and small cells, causing the RDW to be elevated. An elevated RDW (red blood cells of unequal sizes) is known as anisocytosis (Evans, et al.1991).

An elevation in the RDW is not characteristic of all anemia's; anemia of chronic disease, hereditary spherocytosis, acute blood loss, a plastic anemia (anemia resulting from an inability of the bone marrow to produce red blood cells), and certain hereditary hemoglobinopathies (including some cases of thalassemia minor) all may present with a normal RDW (Evans, et al.1991).

1.2.7.1 Calculations:
The "width" in RDW is sometimes thought of as "misleading," since it in fact is a measure of deviation of the volume of RBCs, and not directly the diameter. However, "width" refers to the width of the volume curve (distribution width), not the width of the cells. Thus, it is a reasonably accurate term (Evans, et al.1991).

Mathematically the RDW is calculated with the following formula:

$$\text{RDW} = \left(\frac{\text{Standard deviation of MCV}}{\text{mean MCV}}\right) \times 100$$


1.2.7.2 Pathological Implications

Normal RDW:

When anemia is seen in the presence of a normal RDW, one must have a high suspicion towards thalassemia as the cause of anemia and a Mentzer Index should be done from the CBC report itself to confirm (Emerg, et al. 1991).
**High RDW:**

Iron Deficiency Anemia: usually presents with high RDW with low MCV
Folate and vitamin B12 deficiency anemia: usually presents with high RDW and high MCV (Emerg, et al. 1991).
Mixed Deficiency (Iron + B12 or folate) anemia: usually presents with high RDW with MCV being high, low or often normal range (Emerg, et al. 1991).

**1.2.8 End Stage Renal Disease (ESRD):**

ESRD is when the kidneys stop working well enough for you to live without dialysis or a transplant. This kind of kidney failure is permanent. It cannot be fixed. Most cases of ESRD are caused by diabetes or high blood pressure. Some problems you are born with, some reactions to medicines and some injuries can also cause ESRD. If you have ESRD, you will need dialysis or a kidney transplant to live (http://www.aakp.org/).

**1.2.8.1 Chronic kidney disease (CKD):**

Is when there is permanent damage to your kidney. Your kidneys may still work well enough for you to live, even if they have some damage. If your kidneys keep getting worse, CKD can lead to kidney failure (ESRD). This is when the kidneys do not work well enough for you to live. If this happens, you will need dialysis or a kidney transplant to live (http://www.aakp.org/).

**1.2.8.2 People Who Gets ESRD:**

Some people are more likely to have ESRD than others. Also some people more at risk for ESRD if you have and chronic kidney disease (CKD) and Injury or trauma to the kidneys and Major blood loss (http://www.aakp.org/).
1.2.8.3 Prevention of ESRD:
The best way to prevent ESRD is to prevent CKD. Diabetes and high blood pressure are the two leading causes of CKD. You can help to protect your kidneys by keeping these in control. Get your blood sugar and blood pressure checked often (http://www.aakp.org/).
You can also help protect your kidneys with other healthy habits. You should have regular check-ups with your doctor and eat a low-fat, low-salt diet and exercise most days of the week and avoid tobacco and drink alcohol only in moderation. Also, take medicines the way your doctor tells you. Talk to your doctor before you start any new medicine or supplement (http://www.aakp.org/).

1.2.8.4 Tests for ESRD:
Blood pressure check and Urine test and Blood test for eGFR (http://www.aakp.org/).

1.2.9 Principle of automation technique:
The blood cells are counted in systems on either aperture impedance (voltage _plus) or light_ scattering technology (electro optical counting) It began as counting instrument and has developed into very sophisticated analyzers capable of producing simultaneous count of all the three blood cells type, hemoglobin values ad red cell indices (Suttor, et al.1995). Variation in cell size, as well as being able to provide data on differential white cell count. The most commonly used type in hematology laboratories is the impedance cell counters, firstly describe by Wallace coulter in 1956 {coulter W.H after that the insurgent has been developed that they can produce data related to} (Suttor, et al.1995).

1.2.10 Aperture impedance technology:
Depend on the fact that red cells are conductors of electricity while certain diluents are good conductors, this difference from the basis of the
counting system used in Beckman _coulter, Sysmex, Abbot, Roche and a number of other instruments (Suttor, et al.1995).

For cell counting blood is highly diluted in a buffered electrolyte solution. The rate of this diluted sample is controlled by mercury siphon (as in original coulter system) or by displacement of tightly fitting piston. this measurers the volume of the sample passing through an aperture tube of specific dimension, by constant source of electricity direct current is maintained between two electrodes, one in the sample baker or the chamber surrounding the aperture tube, and another inside the aperture tube. As a blood cell is carried though the aperture, it displaces, it some of the conducting fluid and increase the electrical resistance, this produce change in potential between the electrodes the height of the pluses produced indicates the volume of the cells passing through (Suttor, et al.1995).

The puises can be displayed on and oscillograph screen. The pulses are led to a threshold circuit provide the minimal pulse height which will count (Suttor, et al.1995).

The impedance principle is still the most commonly used method even through it is unable to provide consistently accurate platelet counting in thrombocytopenia, this is due to progressive loss of linearity an, increase influence of back ground noise and sample debris (Suttor, et al.1995).

The main problem is that platelet can't be discriminated from other cellular particles and precipitate that cause similar signals, that main non platelet particles include fragment of RBCs, WBCs, especially of myeloblast (M2, M5) and lipid or protein aggregate. In addition, platelet may not be counted if they are too big small to fit into the window reserved for normal platelet size (Dickerhoff, et al.1995), Komiyama, et al.1997, Throm, et al. 1990).
1.2.11 Light Scattering Method (electro _optical counting):
The other method counting depends on light scattering these work on a similar principle reverse dark field microscope, the blood cells counted by means of electro optical detector (Koht, et al. 1996). The diluted cells flow through the aperture so that the cell pass, in single file, in front of a light source, light is scattered by the cell passing through light beam, scatter light is detected by photomultipliers which converts it into electrical impulses which accumulated and counted (Nisshiyahma, et al. 2005).

Electro_ optical detectors are employed for red cell sizing and counting in Bayer_ technician system, and for white cell differential counting in a number of other instruments. The optical method using two dimension of light scatter are less prone to these problems but there are still some circumstances in which optical analyses may be erroneous. About 70% of sample with an abnormal platelet distribution width showed discrepant value between automated and manual visual counting (Nisshiyahma, et al. 2005).

1:2:12 Disadvantage of Automation Technique:
The instruments that depend on impedance principle are set to count only particles with proper size range for exclusion limit. Any cell or material larger or smaller than size exclusion limit will not be counted. Any object in the paper size range is counted, however, even if it is not a platelet. Electronic counting instruments sometime produce artificially wrong platelet count. It may arise from increase or decrease in counting time or volume, if the platelet and other blood cell pass through the counter at the same time, the instrument will not count the large cell because of the size exclusion limit, which will cause the instrument to accidentally miss the platelet. Clumps of the platelets will not be counted because clumps exceed the upper size exclusion limit for platelet. Re_ circulation of cells
and recounting, presence of non platelet particle called pseudo platelet such as bubble and anthers extraneous particles (Baker, et al. 1985).
If the sample has high white blood cell count, electronic counting may yield an unusually low platelet count because white blood cells may filter out some of the platelets before the sample counted. On the other hand, if the red cells in the sample have burst, their fragments will be falsely counted as platelets (Baker, et al. 1985).
To some extent most of these errors had been limited in automated counter by adjusting volume and time of counting using higher dilution, filter and sweep flow of counted cells eliminated much of these errors so that automated counters can give more reliable results than manual techniques (Suttor, et al. 1995, Frank, et al, 1989, ChanarianI, et al. 1989, Brown, et al. 1993).

1.2.13 Previous studies:
Croat Med J. Feb 2013 Reported that RDW could be an additive predictor for all-cause mortality in patients on chronic dialysis. Furthermore, RDW combined with sound clinical judgment improves identification of patients who are at increased risk compared to RDW alone.
Of 100 patients, 25 patients died during the follow-up period of one-year. Patients who died had significantly higher median [range] RDW levels.

1.2.14 Red cell Distribution Width and other studies of diseases:
Eur, et al. 2009 reported that Higher RDW levels at discharge were associated with a worse long-term outcome, regardless of hemoglobin levels and anemia status. Higher values of RDW were associated with an increased risk.
Webster, Castro in 1986 Jul-Aug; reported that was examined in 60 patients with sickle cell anemia (Hb SS), 28 patients with hemoglobin sickle cell (SC) disease, and seven patients with sickle cell-beta(+)
thalassemia (S-thal). All patients were adults and in the steady state of their disease. The RDW was greater in sickle cell patients than in 39 healthy, age and race matched controls without hemoglobinopathy (Hb AA). Patients with sickle cell anemia had higher mean RDW than those with Hb SC disease.
1:3 Rationales:
Chronic renal failure is considered as a major public health problem in Sudan and might cause many complications of high mortality and morbidity. Including anemia and others so the study to see the effect of red cell distribution width in the patient of end stage renal disease in haemodialysis unit and also in diagnosis of end stage renal disease.
Objectives:

1:4:1 General objective:
Measurement of red cell distribution width in end Stage renal disease (ESRD) patients.

1:4:2 Specific objectives:
. To measure the red cell distribution width.
. To correlate RDW to age group of end stage renal disease patients.
. To correlate RDW to sex of end stage renal disease patients.
CHAPTER TWO

MATERIALS AND METHODS
CHAPTER TWO
Materials and Methods

2 Methods:

2.1 Study design: This is a case control study.

2.2 Study areas: This study was conducted in Alribat hospital at renal dialysis unit

2.3 Study population: Patient with End stage renal disease
Renal failure already diagnosed as case

2.4 Study period: during the period of March to May 2014.

2.5 Inclusion criteria All patient with End stage renal disease available at the time of study

2.6 Exclusion criteria Non patient with End stage renal disease with known hematological parameter.

2.7 Sample size: Probability sampling of 84 patients with End stage renal disease under dialysis came to the Alribat hospital during the period of March to May 2014 were included. 66 healthy person as a control group.

2.8 Test performed: Independent T test.

2.9. Laboratory procedure: Complete blood count was done to measure red cell distribution width and Use of sysmexKX-21N.

2.9.1: Data analysis: Data obtained were analyzed using SPSS program, frequencies, means and independent T test were calculated.

2.9.2: Ethical consideration: Samples were collected after the consent of participants who were informed about the procedure of blood collection and the aim of the study.
2.9.3 Principle:
RDW measurement using Sesmex KX21N depend on the fact that red cells are conductors of electricity while certain diluents are good conductors, this difference from the basis of the counting system used in Sysmex.
CHAPTER THREE

RESULTS
Chapter Three

Results

Table (3.1): Distribution of sex among the study group:

Table (3:1) show that males are 109 and females are 41 and total count is 150.

<table>
<thead>
<tr>
<th>Sex</th>
<th>Frequency</th>
<th>Percent</th>
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<tr>
<td>Male</td>
<td>109</td>
<td>72.7</td>
</tr>
<tr>
<td>Female</td>
<td>41</td>
<td>27.3</td>
</tr>
<tr>
<td>Total</td>
<td>150</td>
<td>100</td>
</tr>
</tbody>
</table>
Table (3.2): Mean, SD, maximum, and minimum of study parameters:

Table (3:2) show the mean and SD of age and mean and SD and also RDWS RDWCV mean and SD.

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Mean</th>
<th>SD</th>
<th>Maximum</th>
<th>Minimum</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age</td>
<td>43.1</td>
<td>17.4</td>
<td>85</td>
<td>5</td>
</tr>
<tr>
<td>RDWCV</td>
<td>14.3</td>
<td>3.1</td>
<td>44.2</td>
<td>11.5</td>
</tr>
<tr>
<td>RDWSD</td>
<td>44.5</td>
<td>6.2</td>
<td>72.4</td>
<td>13.7</td>
</tr>
</tbody>
</table>
Table (3.3): Relation between RDWCV and RDWSD in case and control.

Table (3.3) show that the RDWCV of case and control mean, SD and P value and also show RDWSD of case and control mean, SD and P value.

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Sample</th>
<th>N</th>
<th>Mean</th>
<th>SD</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>RDWCV</td>
<td>Case</td>
<td>84</td>
<td>15.1</td>
<td>3.7</td>
<td>0.000</td>
</tr>
<tr>
<td></td>
<td>Control</td>
<td>66</td>
<td>13.2</td>
<td>1.2</td>
<td></td>
</tr>
<tr>
<td>RDWSD</td>
<td>Case</td>
<td>84</td>
<td>47.2</td>
<td>6.7</td>
<td>0.000</td>
</tr>
<tr>
<td></td>
<td>Control</td>
<td>66</td>
<td>41.2</td>
<td>3.5</td>
<td></td>
</tr>
</tbody>
</table>
Table (3.4): Relation between RDWCV and RDWSD in male and females among case group

Table (3.4) show the relation of RDWCV and RDWSD in mean, SD and P value.

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Sample</th>
<th>N</th>
<th>Mean</th>
<th>SD</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>RDWCV</td>
<td>Male</td>
<td>60</td>
<td>15.4</td>
<td>4.3</td>
<td>0.301</td>
</tr>
<tr>
<td></td>
<td>Female</td>
<td>24</td>
<td>14.5</td>
<td>1.2</td>
<td></td>
</tr>
<tr>
<td>RDWSD</td>
<td>Male</td>
<td>60</td>
<td>47.1</td>
<td>7.5</td>
<td>0.835</td>
</tr>
<tr>
<td></td>
<td>Female</td>
<td>24</td>
<td>47.4</td>
<td>4.2</td>
<td></td>
</tr>
</tbody>
</table>
Figure (3.1): Correlation between RDWCV and age among the case group ($r=0.07$, $P$ value$=0.52$)
Figure (3.1): Correlation between RDWSD and age among the case group (r= 0.123, P value= 0.266)
CHAPTER FOUR

DISCUSSION, CONCLUSION AND RECOMMENDATIONS
Chapter Four
Discussion Conclusion and Recommendation

4:1 Discussion
This study involved a total of eighty four end stage renal disease patient, and sixty sex healthy persons as control. The study group responded to questionnaire, the frequency of the study participants in case and control groups according to sex the males109, females were 41.
The study showed that, the mean of RDW level in end stage renal disease patient is significantly higher than the mean of control group (P value < 0.05).
The study explained that, the RDW in Sudanese end stage renal disease was significantly higher than the values obtained from control group which similar to study in USA which obvious that the mean of RDW was significantly higher in end stage renal disease (Croat, et al. 2013).
In patients with chronic kidney disease, normochromic normocytic anemia mainly develops from decreased renal synthesis of erythropoietin. The anemia becomes more severe as the glomerular filtration rate (GFR) progressively decreases. No reticulocyte response occurs, red blood cell survival is decreased and there is an associated increased bleeding tendency due to uremia-induced platelet dysfunction.
Iron deficiency is also common in patients with chronic kidney disease (CKD). The iron deficiency may be absolute, often due to poor dietary intake or sometimes occult bleeding, or functional, when there is an imbalance between the iron requirements of the erythroid marrow and the actual iron supply. Iron deficiency leads to a reduction in formation of red cell hemoglobin, causing hypo chromic microcytic anemia. Other causes for anemia in chronic kidney disease include the presence of uremic inhibitors (eg, parathyroid hormone, inflammatory cytokines), reduced half-life of circulating blood cells and deficiencies of folate or vitamin B12.
4.2 Conclusion:

There is significant effect of RDW on the end stage renal disease patient.

No significant effect of age on RDW of end stage renal disease patient.

No significant effect of sex on RDW of end stage renal disease patient.
**4:3 Recommendations:**

This study can be purposeful in the diagnosis as laboratory finding on end stage renal disease patient.

More research is needed to elaborate on the important of RDW in end stage renal disease.

Important to know RDW with duration of disease for follow up of patients.

More research of end stage renal disease to know the kind of anemia.
References

Alenzi, FQ; Alenazi, BQ; Ahmad, SY; Salem, ML; Al-Jabri, AA; Wyse, RK (2009). "The haemopoietic stem cell: between apoptosis and self renewal.". The Yale journal of biology and medicine 82 (1): 7–18. PMID 19325941.

American Association of Kidney Patients (http://www.aakp.org/).


Nisshiyahma M, hayashi S, futsukaichi Y, suchi E, kurata Y. (2005). Fragmented red cells are the major cause of suriously hight platelet count in patients with fragmented red cells when counts are obtained by automated blood cell counter. Rinshbyori; (10): 898.
APPENDICES
Appendices
Appendix (1): questionnaire
Sudan University of science and technology
College of graduate studies
Department of hematology and immunohematology
Questionnaire

1-Patient name ...........................................

2-Age..........................................................

3- Tel.No....................................................

3-Gender
Male (  )  female (  )

4- Disease.....................................................

5-Duration of disease........................................

6- Result:
A- CBC
..........................................................
..........................................................
..........................................................

B- RDW CV..................................................

C- RDW SD..................................................

Worker sign..................................................
Appendix (2): Consent form.

بسم الله الرحمن الرحيم

جامعة السودان للعلوم والتكنولوجيا
كلية الدراسات العليا

دراسته لنيل درجة الماجستير

اسم/الرقم ..............................................................

الاسم/الرقم...

سيتم اخذ عينة من الدم بحجم 5 مل من الوريد بواسطة حقنة الطعن ، وذلك بعد تعقيم منطقة اخذ العين بواسطة مطهر.

جميع الأدوات المستخدمة لأخذ العين معقمه ومتبعة فيها كل سبل السلام المعمليه وليس هناك اثار جانبية للعملية.

ربما يحدث تورم وسيط في منطقة اخذ العين وسوف يزول بعد فترة قصيرة.

الغرض من اخذ العين هو البحث العلمي وسوف يسلم المريض نسخة من النتائج وسيتم الاحتفاظ بالنتائج في سريه تامة.

وافق انا المذكور أعلاه على اخذ عينة دم لأجراء الدراسة.

الامضاء..............................................................

التاريخ..............................................................