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Reference Values of Blood Count among Healthy Adult Sudanese Females

القيم المرجعية لتعداد خلايا الدم في الإناث البالغات الأصحاء السودانيات

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

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بِسْمِ اللَّهِ الرَّحْمَنِ الرَّحِيمِ

يقول الله تعالى :

{يَا أَيُّهَا النَّاسُ إِن كُنْتُمْ فِي رَيْبٍ مِّنَ الْبَعْثِ فَإِنَّا خَلَقْنَاكُمْ مِّن ثُرَابٍ ثُمَّ مِّن نُّطْفَةٍ ثُمَّ مِّن عَلَقَةٍ ثُمَّ
مِّن مُّضْغَةٍ مُّخَلَّقَةٍ وَغَيْرِ مُّخَلَّقَةٍ لِّنُبَيِّنَ لَكُمْ وَنُقَرِّرَ فِي الْأَرْحَامِ مَا نَشَاءُ إِلَىٰ أَجَلٍ مُّسَمًّى ثُمَّ نُخْرِجُكُمْ
طِفْلًا ثُمَّ لَتَبَلِّغُوهُنَّ أَشَدَّكُمْ وَمِنْكُمْ مَّن يَتَّقَىٰ وَمِنْكُمْ مَّن يُرَدِّ إِلَىٰ أَرْذَلِ الْعُمُرِ لِكَي لَا يَعْلَمَ مِ بَعْدِ
عِلْمٍ شَيْئًا وَتَرَى الْأَرْضَ هَامِدَةً فَإِذَا أَنزَلْنَا عَلَيْهَا الْمَاءَ اهْتَزَّتْ وَرَبَتْ وَأَنْبَتَتْ مِنْ كُلِّ زَوْجٍ
بَهْجَةٍ}.

سورة الحج الآية رقم (5)

Dedication

To whom breastfed me to love and compassion

to a symbol of love and healing balm

To pure whiteness heart(my mother).

To whom doses of the cup blank to give me a drop of love

and happiness, To whom work to give me a moment to

Pave the way for science, To who remove the thorns out of my way, To the Big Heart(my father).

to my life Basils (my Brothers).

Now open the sails to raise the anchor for the ship to start running on one dark sea that is the sea of life, there is no light in this darkness except candle of distant memory , To those I loved them and loved me(my friends).

my sincere thanks to my supervisor DR. **Fathia Adam Mohammed Salih** for her helping and skill full guides.

To ..

The absolutely necessary person. The gentle **reader**.

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Allah enabled me to conduct this study by his blessing therefore thanks for my god **Allah** firstly and lastly.

Thanks for all **people**, they were very helpful .

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ABSTRACT

Descriptive cross-sectional study is conducted to determine the normal reference values of hemoglobin concentration, hematocrit, red blood cells count, red blood cell indices, white blood cells and blood platelets counts for adult healthy females. 625 apparently healthy adult females from most common tribes in Sudan were selected. EDTA anti coagulant venous blood samples (3 ml) was collected from each subject. Complete blood count (CBC) was measured using automated hematological analyzer (Sysmex kx- 21N). Analysis was done in ALAMAL National Hospital. The ages of the subjects ranged from 18 to 68 years. A questionnaire and informed consent was filled by each subject. The results showed that the Hb concentration range was (12.04 ± 1.32) g/dl. HCT $(38.56 \pm 4.81)\%$, RBCs count $(4.65 \pm 0.45) \times 10^6/\mu\text{L}$, MCV (82.82 ± 7.18) , MCH (26.07 ± 2.40) pg, MCHC (31.22 ± 2.10) g/dl, WBCs count $(6.49 \pm 1.95) \times 10^3/\mu\text{L}$, blood Platelet count range was $(280.7 \pm 73.0) \times 10^3/\mu\text{L}$.

المستخلص

دراسة وصفية مقطعية أجريت لتحديد القيم المرجعية لخضاب الدم , حجم الخلايا المعبأة, الخلايا الحمراء ومؤشراتها , والأرقام القياسية لخلايا الدم الحمراء , الخلايا البيضاء والصفائح الدموية للإناث السودانيات الأصحاء البالغات. وكان العدد الإجمالي للعينات 625 عينة من الإناث السودانيات الأصحاء ظاهرياً من القبائل الأكثر شيوعاً في السودان. جمعت عينة من الدم الوريدي بمقياس (ثلاثة مل) في (E.D.T.A) المضادة للتخثر من كل مشارك. وتم قياس تعداد الدم الكامل باستخدام محلل الدم الآلي (sysmex KX-21N). تم إجراء التحليل المعمل في مستشفى الأمل الوطني. كانت تتراوح أعمار المشاركين من 18 إلى 68 عاماً. تم ملا استبيان وإقرار المشاركة من قبل كل مشارك. أظهرت النتائج أن خضاب الدم (12.04 ± 1.32) جم/دل. حجم الخلايا المعبأة (38.56 ± 4.81) %. عدد كريات الدم الحمراء هو (4.65 ± 0.45) $\times 10^6$ /ميكروليتر. متوسط حجم الخلايا (82.82 ± 7.18) فيمتوليتر. متوسط سعة الخلية من خضاب الدم (26.07 ± 2.40) بيكو جرام. متوسط تركيز خضاب الدم في الخلية (31.22 ± 2.10) جم/دل. عدد كريات الدم البيضاء هو (6.49 ± 1.95) $\times 10^3$ /ميكروليتر. عدد الصفائح الدموية (280.7 ± 73.0) $\times 10^3$ /ميكروليتر.

List of content

	Page No
الآية	I
Dedication	II
Acknowledgements	III
Abstract	IV
المستخلص	V
List of Contents	VI
List of Tables	VIII
List of figures	VIII
List of Abbreviations	IX
Chapter One: Introduction and Literature Review	
1.1 Introduction	1
1.2 Literature Review	3
1.2.1 Hemoglobin	4
1.2.2 Hematocrit	5
1.2.3 Red blood cell	5
1.2.4 Red blood indices	6
1.2.5 White blood cells	7
1.2.6 Blood Platelet	11
1. 3 Rational	14
1.4 Objective	15
1.4.1 General Objective	15
1.4.2 Specific Objectives	15

Chapter Two: Materials and Methods	
2. Materials and Methods	16
2.1 Study design and time	16
2.2 Study area	16
2.3 Study Population	16
2.4 Inclusion criteria	16
2.5 Exclusion criteria	16
2.6 Sample size and Procedure	16
2.7 Ethical Considerations	18
2.8 Data collection	18
2.9 Statistical analysis	18
Chapter Three: Results	
3. Results	19
Chapter Four: Discussion, Conclusion and Recommendations	
4.1 Discussion	22
4.2 Conclusion	24
4.3 Recommendation	25
References	26
Appendices(1)	29
Appendices(2)	31
Appendices(3)	32

List of tables

Table	Title	Page No
Table (3.1)	shows Hematological parameters of study group	19
Table (3.3)	highest frequency of tribes in study group	21

List of figures

Fig	Title	Page No
figure(3.2)	frequency of tested group according to age	20

List of Abbreviations

NR	Normal reference values
CBC	complete blood count
RBCs	Red blood cells
HB	Hemoglobin
HCT	Hematocrit
MCV	Mean cell volume
MCH	Mean cell hemoglobin
MCHC	Mean cell hemoglobin concentration
WBCs	White cells
PLT	blood Platelets counts

Chapter One

Introduction and Literature Review

Chapter One

Introduction and Literature Review

1.1 Introduction

A reference range is usually defined as the set of values 95 percent of the normal population falls within (that is, 95% prediction interval). It is determined by collecting data from vast numbers of laboratory tests. A number of factors affect hematological values in apparently healthy individuals. These include the technique and timing of blood collection, transport and storage of specimens, differences in the subject's posture when the sample is taken, prior physical activity, or whether the subject is confined to bed. Variation in the analytic methods used may also affect the measurements. These can all be standardized (Weed.1963).

More problematic are the inherent variables as a result of sex, age, occupation, body build, genetic background, and adaptation to diet and to environment (especially altitude). These factors must be recognized when establishing physiologically normal values. Furthermore, it is difficult to be certain in any survey of a population for the purposes of obtaining data from which normal ranges may be constructed that the “normal” subjects are completely healthy and do not have nutritional deficiencies (especially iron deficiency, which is prevalent in many countries), mild chronic infections, parasitic infestations and the effects of smoking. Hematological values for the normal and abnormal will overlap, and a value within the recognized normal range may be definitely pathological in a particular subject. For these reasons the concept of “normal values” and “normal

ranges” has been replaced by reference values and the reference range, which is defined by reference limits and obtained from measurements on the reference population for a particular test. The reference range is also termed the reference interval. Ideally, each laboratory should establish a databank of reference values that take account of the variables mentioned earlier, so that an individual's result can be expressed and interpreted relative to a comparable apparently normal population, insofar as normal can be defined (Lewis.2006).

Health and disease can only be distinguished by accurate and reliable reference values of a particular laboratory test. It is now a proven fact that there is considerable variation in hematology reference intervals depending on the demographic and pre analytical variables. There are evidences that values provided by manufacturers do not have appropriate application for all populations. Moreover, reference ranges provided by different laboratory manuals and books also do not solve this problem (Bain.2002).

In daily health related practice in Sudan, it is important to have hematological reference values for diagnosis orientation. The hematological reference values were determined many years ago for the Caucasian's populations. Analysis by different statistical techniques and variations with age and sex.

Recently, several authors tried to establish reference values in hematology for African countries. However there are some discrepancies from one study to another which may be related to different factors such as age, sex, geographic origin, altitude, and ethnic origin (Lugada. 2004).

Moreover, many constitutional hemoglobin abnormalities (thalassemia, sickle cell disease and hemoglobin C) or pathologic conditions (malaria, HIV, HBV and HCV viral infections), influence the hematological values (Saathoff et al. 2008).

1.2.Literature Review

A complete blood count (CBC), also known full blood count (FBC), or full blood exam (FBE), is a blood panel requested by a doctor or other medical professional that gives information about the cells in a patient's blood, such as the cell count for each cell type and the concentrations of various proteins and minerals. A scientist or lab technician performs the requested testing and provides the requesting medical professional with the results of the CBC. The cells that circulate in the blood stream are generally divided into three types white blood cells (leukocytes), red blood cells (erythrocytes), and platelets (thrombocytes). Abnormally high or low counts may indicate the presence of many forms of disease, and hence blood counts are amongst the most commonly performed blood tests in medicine, as they can provide an overview of a patient's general health status. A CBC is routinely performed during annual physical examinations in some jurisdictions Procedure. A phlebotomist collects the sample through venipuncture, drawing the blood into a test tube containing an anticoagulant (EDTA, sometimes citrate) to stop it from clotting. The sample is then transported to a laboratory. Sometimes the sample is drawn off a finger prick using a Pasteur pipette for immediate processing by an automated counter. A complete blood count (CBC) is a blood test used to evaluate your overall health and detect a wide range of disorders, including anemia, infection and leukemia. A complete blood count test measures several components and features of your blood, including Red blood cells, which carry oxygen .White blood cells, which fight infection .Hemoglobin, the oxygen-carrying protein in red blood cells. Hematocrit , the proportion of red blood cells to the fluid component, or plasma, in your blood.Platelets, which help with blood clotting (Gomella.2013).

1.2.1. Haemoglobin

Also spelled hemoglobin and abbreviated Hb or Hgb, is the iron-containing oxygen-transport metallo protein in the red blood cells of all vertebrates. Hemoglobin in the blood carries oxygen from the respiratory organs (lungs or gills) to the rest of the body (i.e. the tissues). There it releases the oxygen to permit aerobic respiration to provide energy to power the functions of the organism in the process called metabolism. Hemoglobin has an oxygen-binding capacity of 1.34 mL O₂ per gram (Dominguez. 1981).

This increases the total blood oxygen capacity seventy-fold compared to dissolved oxygen in blood. The mammalian hemoglobin molecule can bind (carry) up to four oxygen molecules (Costanzo.2007).

Hemoglobin is involved in the transport of other gases. It carries some of the body's respiratory carbon dioxide (about 10% of the total) as carbaminohemoglobin, in which CO₂ is bound to the globin protein. The molecule also carries the important regulatory molecule nitric oxide bound to a globin protein thiol group, releasing it at the same time as oxygen (Epistien.1998).

Hemoglobin is also found outside red blood cells and their progenitor lines. Other cells that contain hemoglobin include the A9 dopaminergic neurons in the substantia nigra, macrophages, alveolar cells, and mesangial cells in the kidney. In these tissues, hemoglobin has a non-oxygen-carrying function as an antioxidant and a regulator of iron metabolism. Hemoglobin and hemoglobin-like molecules are also found in many invertebrates, fungi, and plants. In these organisms, hemoglobin's may carry oxygen, or they may act to transport and regulate other things such as carbon dioxide, nitric oxide, hydrogen sulfide and sulfide.

A variant of the molecule, called leghemoglobin, is used to scavenge oxygen away from anaerobic systems, such as the nitrogen-fixing nodules of leguminous plants, before the oxygen can poison the system (Biagilio. 2009).

1.2.2. Hematocrit

Hematocrit is a blood test that measures the percentage of the volume of whole blood that is made up of red blood cells. This measurement depends on the number of red blood cells and the size of red blood cells. The hematocrit is almost always ordered as part of a complete blood count (CBC). This test may order to investigate Anemia, Diet deficiency, Leukemia and Other medical condition (Biagilio.2009).

1.2.3 Red blood cell

Red blood cells (RBCs), also called erythrocytes, are the most common type of blood cell and the vertebrate organism's principal means of delivering oxygen (O_2) to the body tissues via blood flow through the circulatory system. RBCs take up oxygen in the lungs or gills and release it into tissues while squeezing through the body's capillaries. The cytoplasm of erythrocytes is rich in hemoglobin, an iron-containing biomolecule that can bind oxygen and is responsible for the red color of the cells. The cell membrane is composed of proteins and lipids, and this structure provides properties essential for physiological cell function such as deformability and stability while traversing the circulatory system and specifically the capillary network. In humans, mature red blood cells are flexible and oval biconcave disks. They lack a cell nucleus and most organelles, in order to accommodate maximum space for hemoglobin. Approximately 2.4 million new erythrocytes are produced per second in human adults, quarter of the cells in the human body are red blood cells. In order to carry hemoglobin into close contact with the tissues and for successful gaseous exchange, the red cell, 8 μ m in diameter,

must be able to pass repeatedly through the microcirculation whose minimum diameter is 3.5 μm , to maintain hemoglobin in a reduced (ferrous) state and to maintain osmotic equilibrium despite the high concentration of protein (hemoglobin) in the cell .(Hoffbrand et al.2006).

Red blood cells are also known as, red cells. Red blood corpuscles (an archaic term), haematids, erythroid cells or erythrocytes (from Greek *erythros* for "red" and *kytos* for "hollow vessel", with *-cyte* translated as "cell" in modern usage).The functional lifetime of an erythrocyte is about 100–120 days, during which time the erythrocytes are continually moved by the blood flow push (in arteries), pull (in veins) and a combination of the two as they squeeze through micro vessels such as capillaries Increase in red blood cells known as polycythemia, result from Dehydration, Lung (pulmonary) disease, Kidney or other tumor that produces excess erythropoietin, Smoking, Genetic causes (altered oxygen sensing, abnormality in hemoglobin oxygen release)and Polycythemia vera a rare disease. Decrease in red blood cells known as anemia result from Acute or chronic bleeding, RBC destruction (e.g., hemolytic anemia, etc),Nutritional deficiency (e.g., iron deficiency, vitamin B12 or folate deficiency),Bone marrow disorders or damage, Chronic inflammatory disease and Kidney failure. (Lang.2008).

1.2.4 Red blood indices

1.2.4.1 MCV

Decrease Mean Corpuscular Volume Indicates RBCs are smaller than normal (microcytic), caused by iron deficiency anemia or thalassemias, for example. Increase Indicates RBCs are larger than normal (macrocytic), for example in anemia caused by vitamin B12 or folate deficiency (Lewis.D.2006).

1.2.4.2 MCH

Mean Corpuscular Hemoglobin Mirrors MCV results; small red cells would have a lower value. Mirrors MCV results, macrocytic RBCs are large so tend to have a higher MCH (Lewis.2006).

1.2.4.3 MCHC

Mean Corpuscular Hemoglobin Concentration May be low when MCV is low; decreased MCHC values (hypochromia) are seen in conditions such as iron deficiency anemia and thalassemia. Increased MCHC values (hyperchromia) are seen in conditions where the hemoglobin is more concentrated inside the red cells, such as autoimmune hemolytic anemia, in burn patients, and hereditary spherocytosis, a rare congenital disorder(Lewis. 2006).

1.2.5 White blood cells

Also called leukocytes or leucocytes, are the cells of the immune system that are involved in protecting the body against both infectious disease and foreign invaders. All leukocytes are produced and derived from a multi potent cell in the bone marrow known as a hematopoietic stem cell. Leukocytes are found throughout the body, including the blood and lymphatic system.Five different and diverse types of leukocytes exist. These types are distinguished by their physical and functional characteristics. Monocytes and neutrophils are phagocytic (La Fleur.2008).

The number of leukocytes in the blood is often an indicator of disease, and thus the WBC count is an important subset of the complete blood count. The normal white cell count is usually between 4 and $11 \times 10^9/L$. In the US this is usually expressed as 4,000–11,000 white blood cells per microliter of blood. They make up

approximately 1% of the total blood volume in a healthy adult. An increase in the number of leukocytes over the upper limits is called leukocytosis, and a decrease below the lower limit is called leukopenia. The name "white blood cell" derives from the physical appearance of a blood sample after centrifugation. White cells are found in the buff, a thin, typically white layer of nucleated cells between the sedimented red blood cells and the blood plasma. The scientific term leukocyte directly reflects its description. It is derived from the Greek word leuko- meaning "white" and kytos meaning "hollow vessel", with -cyte translated as "cell" in modern use. The buffy coat may sometimes be green if there are large amounts of neutrophils in the sample, due to the hem-containing enzyme myeloperoxidase that they produce (Bruce.2002).

1.2.5.1 Neutrophil

Neutrophils are the most abundant white blood cell, constituting 60-70% of the circulating leukocytes (Bruce. 2002).

They defend against bacterial or fungal infection. They are usually first responders to microbial infection; their activity and death in large numbers forms pus. They are commonly referred to as polymorph nuclear (PMN) leukocytes, although, in the technical sense, PMN refers to all granulocytes. They have a multi-lobed nucleus, which consists of three to five lobes connected by slender strands (Salandin.2012).

This gives the neutrophils the appearance of having multiple nuclei, hence the name polymorph nuclear leukocyte. The cytoplasm may look transparent because of fine granules that are pale lilac when stained. Neutrophils are active in phagocytizing bacteria and are present in large amount in the pus of wounds.

These cells are not able to renew their lysosomes (used in digesting microbes) and die after having phagocytized a few pathogens (Wheater. 2002).

Neutrophils are the most common cell type seen in the early stages of acute inflammation. The life span of a circulating human neutrophil is about 5.4 days (Pillay.2010).

1.2.5.2 Eosinophil

Eosinophil's compose about 2-4% of the WBC total. This count fluctuates throughout the day, seasonally, and during menstruation. It rises in response to allergies, parasitic infections, collagen diseases, and disease of the spleen and central nervous system. They are rare in the blood, but numerous in the mucous membranes of the respiratory, digestive, and lower urinary tracts. They primarily deal with parasitic infections. Eosinophil's are also the predominant inflammatory cells in allergic reactions. The most important causes of eosinophilia include allergies such as asthma, hay fever, and hives; and also parasitic infections. They secrete chemicals that destroy these large parasites, such as hook worms and tape worms that are too big for any one WBC to phagocytize. In general, their nucleus is bi-lobed. The lobes are connected by a thin strand. The cytoplasm is full of granules that assume a characteristic pink-orange color with eosin staining (Pillay. 2010).

1.2.5.3 Basophil

Basophiles are chiefly responsible for allergic and antigen response by releasing the chemical histamine causing the dilation of blood vessels. Because they are the rarest of the white blood cells (less than 0.5% of the total count) and share physicochemical properties with other blood cells, they are difficult to study. They can be recognized by several coarse, dark violet granules, giving them a blue hue.

The nucleus is bi- or tri-lobed, but it is hard to see because of the number of coarse granules that hide it. They excrete two chemicals that aid in the body's defenses, histamine and heparin. Histamine is responsible for widening blood vessels and increasing the flow of blood to injured tissue. It also makes blood vessels more permeable so neutrophils and clotting proteins can get into connective tissue more easily. Heparin is an anticoagulant that inhibits blood clotting and promotes the movement of white blood cells into an area. Basophils can also release chemical signals that attract eosinophil's and neutrophils to an infection site (Pillay. 2010).

1.2.5.4 Lymphocyte

Lymphocytes are much more common in the lymphatic system than in blood. Lymphocytes are distinguished by having a deeply staining nucleus that may be eccentric in location, and a relatively small amount of cytoplasm. Lymphocytes include B cells Make antibodies that can bind to pathogens, block pathogen invasion, activate the complement system, and enhance pathogen destruction .T cellsCD4+ helper T cells T cells displaying co-receptor CD4 are known as CD4+ T cells. These cells have T-cell receptors and CD4 molecules that, in combination, bind antigenic peptides presented on major histocompatibility complex (MHC) class II molecules on antigen-presenting cells. Helper T cells make cytokines and perform other functions that help coordinate the immune response. In HIV infection, these T cells are the main index to identify the individual's immune system integrity.CD8+ cytotoxic T cells.T cells displaying co-receptor CD8 are known as CD8+ T cells. These cells bind antigens presented on MHC I complex of virus-infected or tumor cells and kill them. Nearly all nucleated cells display MHCI. $\gamma\delta$ T cells possess an alternative T cell receptor (different from the $\alpha\beta$ TCR found on conventional CD4+ and CD8+ T cells). Found in tissue more commonly than in blood, $\gamma\delta$ T cells share characteristics of helper T cells, cytotoxic T cells,

and natural killer cells. Natural killer cells are able to kill cells of the body that do not display MHC class I molecules, or display stress markers such as MHC class I polypeptide-related sequence A (MIC-A). Decreased expression of MHC class I and up-regulation of MIC-A can happen when cells are infected by a virus or become cancerous (Hoffbrand et al.2006).

1.2.5.5 Monocyte

Monocytes, the largest type of WBCs, share the "vacuum cleaner" (phagocytosis) function of neutrophils, but are much longer lived as they have an extra role, they present pieces of pathogens to T cells so that the pathogens may be recognized again and killed. This causes an antibody response to be mounted. Monocytes eventually leave the bloodstream and become tissue macrophages, which remove dead cell debris as well as attacking microorganisms. Neither dead cell debris nor attacking microorganisms can be dealt with effectively by the neutrophils. Unlike neutrophils, monocytes are able to replace their lysosomal contents and are thought to have a much longer active life. They have the kidney shaped nucleus and are typically a granulated. They also possess abundant cytoplasm. Once monocytes move from the bloodstream out into the body tissues, they undergo changes (differentiate) allowing phagocytosis and are then known as macrophages (Hoffbrand et al.2006).

1.2.6 Blood Platelets

Also called thrombocytes, are a component of blood whose function (along with the coagulation factors) is to stop bleeding by clumping and clogging blood vessel injury. Platelets have no cell nucleus, they are fragments of cytoplasm which are derived from the megakaryocytes of the bone marrow, and then enter the circulation. These inactivated platelets are biconvex discoid (lens-shaped)

structures 2–3 μm in greatest diameter. Platelets are found only in mammals, whereas in other animals (e.g. birds, amphibians) thrombocytes circulate as intact mononuclear cells. On a stained blood smear, platelets appear as dark purple spots, about 20% the diameter of red blood cells. The main function of platelets is to contribute to hemostasis, disorder of platelet function is a thrombocytopathy. Low platelet concentration is thrombocytopenia and is due to either decreased production or increased destruction.

Examples of causes of thrombocytopenia:-

- Viral infection (mononucleosis, measles, hepatitis).
- Rocky mountain spotted fever.
- Platelet auto antibodies.
- Drugs (acetaminophen, guanidine, sulfa drugs).
- Cirrhosis.
- Autoimmune disorders.
- Sepsis .
- Leukemia, lymphoma .
- Myelodysplasia.
- Chemo or radiation therapy.

Elevated platelet concentration is thrombocytosis and is either congenital, reactive (to cytokines), or due to unregulated production one of the myeloproliferative neoplasm's or certain other myeloid neoplasm's.

Examples of causes of thrombocytosis:-

- Cancer (lung, gastrointestinal, breast, ovarian, lymphoma).
- Rheumatoid arthritis, inflammatory bowel disease, lupus.

- Iron deficiency anemia.
- Hemolytic anemia.
- Myeloproliferative disorder as essential thrombocythemia (Laki.1972).

1.3 Rationale

Health and diseases can only be distinguished by accurate and reliable reference values of a particular laboratory test. It is now a proven fact that there is a considerable variation in hematology reference values depending on the demographic and pre analytical variables(Saathoff et al. 2008).

Sudan is a North African country, with an Altitude/ elevation 380 m (1247 ft). According to study done in 2008 Sudan population will be in 2015 about (38,435,252)individuals .Females in age between 18 and 68 about (87,98,059). (Population projection Sudan. 2008).

Until now, blood cells and red cells indices reference values have not been established. The values usually used are those of Caucasian populations. According to this context and for a better interpretation of hematologic results. Till now Sudanese physicians and doctors depend on the universal reference values of RBCs, WBCs, PLTs and hemoglobin in diagnosis of diseases. Few studies were done in this filed in specific population not representing all Sudanese population. Approximately most of Sudanese trips were included in this study for better screening and reliable results. In daily practice in Sudan, it is important to have hematological reference values for diagnosis orientation.

1.4 Objectives

1.4.1 General Objective:

To determine the normal values of Blood Cells, Haemoglobin and Red Blood Cell Indices among Adult Sudanese Females.

1.4.2 Specific Objectives:

1. To establish reference values for hemoglobin concentration and Hematocrit of adult Sudanese females.
2. To establish reference values for Red blood cells count and RBCs indices of adult Sudanese females.
3. To establish reference values for White blood cells count of adult Sudanese females.
4. To establish reference values for Blood Platelets count of adult Sudanese females.

Chapter Two

Materials and Methods

Chapter Two

Materials and Methods

2.1. Study design and time

Descriptive cross-sectional study, conducted during the period of March 2015 to October 2015.

2.2. Study area

The study was conducted in Khartoum State.

2.3. Study Population

Apparently healthy None hospitalized adult Sudanese females.

2.4. Inclusion criteria

Apparently Healthy Sudanese females age ranged (18-68) .

2.5. Exclusion criteria

Those whom complaining from any infections or diseases that may affect the parameters of CBC.

Those individual under 18 years old or above 68 years.

Pregnant women's.

2.6. Sample size and Procedure

625 sample were collected using dry, vacuum system, tourniquet was used to make the veins more prominent, blood samples (3ml) was collected in EDTA vacuum tube from each volunteer under septic condition.

2.6.1.Material

- 1- EDTA container .
- 2- Disposable syringes.
- 3- Tourniquet.
- 4- Cotton wool.
- 5- Sysmex Kx,21N.

2.6.2.Reagent

- 1-Cell pack (diluent).
- 2-Stromatolyser .
- 3-Cell clean .

2.6.3.Procedure

The reagent for operation were checked ,then the power switch was turn on auto rise and background check were automatically performed then three level of control (low count, normal and high) were applied after selection whole blood mode of analysis sample number were introduced by pressing sample number keys then enter key was pressed ,after that sample was mixed carefully the tab bring in close contact with sample probe and the start key was pressed, after the required volume of blood were aspirated ,then tube was removed ,result was displayed in the screen and was printout.

2.6.4.Quality control of Sysmex

Hematology analyzer provide quick and accurate results in most situation. However ,false results related either to platelets or other parameter from complete

blood cells count may be observed in several instances , false low WBCs count may be observed because of observed because of agglutination in the presence of EDTA.

The sample size calculated with known population size as follow.

$$n = \frac{N}{1 + N(d)^2}$$

n=sample size.

N= population size.

d= degree of precision (0.04).

Sample size = $8798059 / 1 + 8798059 (0.04)^2 = 625$.

2.7. Ethical Considerations

A Study was approved from ethical committee of the Sudan University of Science and Technology, consents were taken from all the participations after they had been informed about the procedure of blood collection and aims of study .they were used that data will not be used for any other purpose than this study.

2.8. Data collection

Data were collected using structural interviewing questionnaire, which was designed to collect and maintain all information needed.

2.9. statistical analysis

Data were analyzed by SPSS statistical package of social science (version 11.5, SPSS) mean \pm standard deviation.

Chapter Three

Results

Chapter Three

Results

3. Results

In this study 625 Apparently healthy None hospitalized adult Sudanese females subjects were chosen for determination of normal values of hemoglobin concentration Hematocrit ,RBCs, RBCs indices total WBCs ,and blood platelets count in adult healthy Sudanese females during the period march to October 2015. And the table below show the results of this study(mean \pm standard deviation).

Table (3.1) complete blood count parameters of study group

Parameter	mean \pm standard deviation
HGB g/dl	12.04 \pm 1.32
HCT %	38.56 \pm 4.81
RBC $\times 10^6/\mu\text{L}$	4.65 \pm 0.45
MCV fl	82.82 \pm 7.18
MCH pg	26.07 \pm 2.40
MCHC g\dl	31.22 \pm 2.10
WBC $\times 10^3/\mu\text{L}$	6.49 \pm 1.95
PLT $\times 10^3/\mu\text{L}$	280.7 \pm 73.0

Young age groups were found healthier than the elder one so the largest number of participants in age group (18-28)



Fig (3.2) frequency of tested group according to age

The highest frequency in group of age 18-28 and the lowest frequency in group of age 59-68.

50 different Sudanese tribes were included in this study the table below show the highest frequency of tribes in study group.

Table (3.3) highest frequency of tribes in study group

Tribe	Frequency
Gallia	128
Shawiga	80
Dnagla	37
Mahas	30
Noba	29
Halfawien	27
Kawhla	25
Rofaiin	19
Hassania	16
Fur	15

Chapter Four

Discussion, Conclusion and Recommendations

Chapter Four

Discussion, Conclusion and Recommendations

4.1.DISCUSSION

The results of this study showed that the Hemoglobin concentration in adult females of Sudanese population (12.04 ± 1.3)g/dl is lower than the hemoglobin concentration in adult females of British population (13.75 ± 2.75) g/dl (Lewis .D, 2006).

Also the normal hemoglobin concentration in adult females of Sudanese population (12 ± 1.3)g/dl is lower than the normal hemoglobin concentration in the adult females of Saudis population (13.8 ± 1.1)g/dl (Alhazmi.2001).

The normal Red Blood Cells Count in adult females of Sudanese population (4.6 ± 0.4) $\times 10^6/\mu\text{L}$ is slightly higher than The normal Red Blood Cells Count in adult females of British population females ($4.3 \pm 0.5 \times 10^6/\mu\text{L}$), (Lewis. 2006).Also normal Red Blood Cells Count in adult females of Sudanese population (4.65 ± 0.45) $\times 10^6/\mu\text{L}$ slightly increase in lower values than germane population adult females (4.8 ± 0.6) $\times 10^6/\mu\text{L}$. (Badenhorst .1995).But higher than the Normal Red Blood Cells Count in adult females of Chinese population (4.39 ± 0.7) $\times 10^{12}/\text{L}$. (Cong.2011).Normal Red Blood Cells Count in adult females of Sudanese population (4.6 ± 0.4) $\times 10^6/\mu\text{L}$ is lower than Normal Red Blood Cells Count of adult females Saudis population (4.9 ± 0.59) $\times 10^6/\mu\text{L}$ (Alhazmi.2001).

While the normal red blood cells indices of this study is MCV(82.82 ± 7.18) fl, MCH(26.07 ± 2.40)pg and MCHC(31.22 ± 2.10 g\dl).

The Total white blood cells count in adult females of Sudanese population is higher than total white blood cells count in adult females of Black population of

the Witwatersrand (South Africa) being $(5.60 \pm 1.51) \times 10^3/l$. But lower than total white blood cells count in adult females British population $(7.0 \pm 3.0) \times 10^3/\mu L$ (Lewis.2006).

The total white blood cells count in adult females of Sudanese population is $(6.5 \pm 2) \times 10^3/\mu L$ is lower than the Total white blood cells count in adult females of Saudis population $(7.9 \pm 2.1) \times 10^3/\mu L$ (Alhazmi.2001).

This research confirms that the genes are related to variation of white count cells.

Blood Platelet count in Sudanese adult females population $(280.7 \pm 73) \times 10^9/l$ slightly lower than the blood Platelets counts of adult Black females population of the Witwatersrand (South Africa) $(317 \pm 64.0) \times 10^9 /l$. (Lewis.2006).

The mean blood Platelets count of adult females in Sudan $(280.7 \pm 73) \times 10^9/l$ is higher than The mean blood Platelets count of adult females of Togo $247 \times 10^9/L$ (Isrn;hematology. 2011).

Similar study was done in Khartoum north area in Sudan with sample size 200 male and 200 female, the results of females population are relatively close to results that obtained in this study RBCs $(4.6 \pm 0.4) \times 10^6/\mu L$, TWBCs $(5.3 \pm 1.1) \times 10^3/\mu L$ and blood Platelet $(277.8 \pm 72.4) \times 10^3/\mu L$ in females (Osman. 2013).

4.2.CONCLUSION

1.The Hemoglobin concentration in adult females of Sudanese population is lower than the hemoglobin concentration of British and of Saudis adult females population.

2.The normal Red Blood Cells Count in adult females of Sudanese population is higher than The normal Red Blood Cells Count of Chinese adult females But lower than the Red Blood Cells Count in adult females of Saudis population.

3.The normal total white blood cells count in adult females of Sudanese population is higher than the normal total white blood cells count in adult females of Black population of the Witwatersrand (South Africa). But lower than the normal Total white blood cells count in females of Saudis population.

4.The mean blood Platelets count of adult females in Sudan is higher than The mean blood Platelets count in females of Togo.

4.3.RECOMMENDATIONS

- Reference values of complete blood count parameters for males and children in Sudan should be done .
- Reference values of complete blood count parameters for pregnant females in Sudan should be done.
- Another study with larger sample size investigating all tribes in Sudan for complete blood count parameters reference values should be done.
- References values of all hematological parameters should be established for the Sudanese population.

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Appendices

Appendices (1)

Sudan University of Science and Technology

College of Graduate Studies

Questionnaire

Date:

ID NO:

Name:

Telephone NO:

Address:

Tribe:

Age group: 18 – 28 () 29 – 38 () 39– 48() 49 – 58() 59 – 68 ()

Medical history:-

Did you suffer from any one of these diseases?

	Yes	no
1- Chronic diseases	()	()
2- Genetic diseases	()	()
3- Hepatitis	()	()
4- Syphilis	()	()
5- HIV	()	()

Did you undergo to surgical operation during the last 12 month? () ()

Did you donate or receive blood during the last 6 month? () ()

Did you infected with malaria during the last month? () ()

Are you using any type of medication? () ()

If yes mention

Are you pregnant ? () ()

Clinical examination:-

General appearance

Blood pressure

Appendices (2)

Sysmex Kx,21N Principle

Sysmex Kx,21N the sysmex is hematology automated analyzer use to quickly perform full blood count and reticulocyte count .it is made by the sysmex corporation .Blood is sample and diluted and moves through a tube thin enough that cell pass by one at time ,because not everything about the cells can be measured at the same time ,blood is separated into number of different channels .As the cells pass through apertures the signals are transmitted in sequence to analog circuit and then to particle size distribution analysis circuits for conversion to cumulative cell size distribution data particle size distribution curves constructed and the auto discrimination level is then set by the microprocessor for each population, This floating threshold allow for discrimination of all population ,the cell count include the pulses between the lower and the upper auto discrimination level

Appendices (3)

بسم الله الرحمن الرحيم

جامعة السودان للعلوم والتكنولوجيا

كلية الدراسات العليا

ماجستير مختبرات طبية

تخصص علم أمراض الدم ومبحث المناعة الدموية

إقرار بالمشاركة

الاسم:

سوف يتم اخذ عينة من الدم (3 مل) من الوريد بواسطة حقنة طعن وذلك بعد مسح منطقة اخذ العينه بواسطة المطهر.

كل الأدوات المستخدمة لأخذ العينة معقمة ومتبع فيها وسائل السلامة المعملية.

أوافق أنا المذكور أعلاه اخذ عينه لإجراء الدراسة.

الإمضاء:

التاريخ:

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