INTRODUCTION

White blood cells: The name "White blood cell "derives from the fact that after centrifugation of a blood sample, the white cells are found in the Buffy coat, a thin layer of nucleated cells between the sedimented red blood cells and the blood plasma, which is typically white in color. The scientific term leukocyte directly reflects this description, derived from Greek leukos - white, and kytos– cell (Bain BJ -1996). White blood cells, or leukocytes, are cells of the immune system defending the body against both infectious disease and foreign materials.

Several different and diverse types of leukocytes exist, but they are all produced and derived from a multipotent cell in the bone marrow known as a hematopoietic stem cell. Leukocytes are found throughout the body, including the blood and lymphatic system (Bain BJ -1996).

There are several different types of white blood cells. They all have many things in common, but are all different. One primary technique to classify them is to look for the presence of granules, which allows the differentiation of cells into the categories granulocytes and a granulocytes (Amador 1975).

Granulocytes (polymorph nuclear leucocytes)
Leukocytes characterized by the presence of different staining granules in their cytoplasm when viewed under light microscopy. These granules are membrane-bound enzymes, which primarily act in the digestion of endocytosed particles. There are three types of granulocytes: neutrophils, basophils, and eosinophils, which are named according to their staining properties (Amador 1975).

A granulocytes (mononuclear leucocytes)
Leukocytes characterized by the apparent absence of granules in their cytoplasm. Although the name implies a lack of granules these cells do contain non-specific azurophilic granules, which are lysosomes. The cells include lymphocytes, monocytes, and macrophages (Amador 1975).
Leucopoiesis: The blood granulocytes are formed in the bone marrow from a common precursor cell (stem cells). In the granulopoietic series progenitor cells, myeloblasts, promyelocytes form a proliferative or mitotic pool of cells while the metamyelocyte, band and segmented granulocytes make up a post mitotic maturation compartment (Amador 1975).

The bone marrow normally contains more myeloid cells than erythroid cells in the ratio of 2:1 to 12:1 the largest proportion being neutrophil and metamyelocytes. In the stable or normal state, the bone marrow storage compartment contain 10 -15 times the number of granulocyte found in the peripheral blood (Dacie 2006). Following their release from the bone marrow granulocyte spend only 6 – 10 hours in the circulation before moving into the tissue where they perform their phagocytic function. It has been estimated that they spend an average 5 – 6 days in the tissues before they are destroyed during defensive actions or as the result of senescence.

Control of leucopoiesis: The granulocytes series arises from bone marrow progenitors cells which are increasingly specialized. Many growth factors are involved in this maturation process, including interleukin–1 (IL–1), IL–3, IL–5, IL–6, IL–11, granulocyte-macrophage-colony-stimulating factor (GM-CSF), G-CSF and monocyte CSF.

The growth factor stimulates proliferation and differentiation and also affect the function of the mature cells on which they act (e.g. phagocytosis, superoxide generation and cytotoxicity in the case of neutrophils phagocytosis, cytotoxicity and production of other cytokines by monocytes. Increased granulocytes and monocytes production in response to an infection is induced by increased productions of growth factors from stromal cells and T- Lymphocytes stimulated by endotoxin, IL – 1 or tumor necrosis factor TNF (Daniel Catovsky - 2005).

Neutrophils: This cell has a characteristic dense nucleus consisting of between two to five lobes and pale cytoplasm with an irregular outline containing many fine pink – blue (azurophilic) or grey – blue granules (Daniel Catovsky - 2005).

The granules divided into

Both types of granules are lysosomal in origin, the primary contain myeloperoxidase, acid phosphates and other acid hydrolyses, the secondary contains collagenase, lactoferrin and lysozyme, the life span of neutrophil in the blood is only about 10 hours and days in the spleen and other tissue (Daniel Catovsky - 2005).

Neutrophil precursors: The earliest recognizable precursor is the myeloblast a cell of variable size, which has a large nucleus with fine chromatin and usually 2-5 nucleoli.

The cytoplasm is basophilic and no cytoplasmic granules are present, the bone marrow contains up to 4% of myeloblast. Myeloblast give rise by cell division to promyelocytes which are slightly larger cells and have developed primary granules in the cytoplasm. These cells then produced myelocytes which have specific or secondary granules, the nuclear chromatin is now more condensed and nucleoli are not visible (A.V. Hoffbrand – 2005).

The myelocytes give rise by cell division to metamyelocytes, non dividing cells, which have an indented or horseshoe shaped nucleus a cytoplasm filled with primary and secondary granules. Neutrophil forms between metamyelocytes and fully mature neutrophil are termed band, stab, or juvenile. These cells may occur in normal peripheral blood. They do not contain the clear fine filamentous distinction between nuclear lobe, which is seen in mature neutrophil (A.V. Hoffbrand – 2005).

Eosinophils: These cells are similar to neutrophil, except that the cytoplasmic granules are coarser and more deeply red staining and there are rarely more than 3 nuclear lobes. Eosinophil myelocytes can be recognized, but earlier stages are indistinguishable from neutrophil precursors. The blood transit time for eosinophils is longer than for neutrophils. They enter inflammatory exudates and have a special role in allergic responses, defense against parasites and removal of fibrin formed during inflammation (A.V. Hoffbrand – 2005).

Basophils: These cells are only occasionally seen in normal peripheral blood. Basophil are chiefly responsible for allergic and antigen response by releasing the chemical histamine causing inflammation. The nucleus is bi- or tri-lobed, but it is hard to see because of the number of coarse granules which hide it. They are characterized by their large blue granules (A.V. Hoffbrand – 2005).

Monocytes: These are usually larger than other peripheral blood leucocytes and possess a large central oval or indented nucleus with clumped chromatin. The abundant cytoplasm stains blue and contain many fine vacuoles, giving a ground – glass appearance.

The monocytes precursors in the marrow (monoblast, promonocytes) are difficult to distinguish from myeloblast and monocytes. Monocytes spend only a short time in the marrow and after circulating for 20 to 40 hours, leave the blood to enter the tissue where they mature and carry out the principle function. Their extravascular lifespan after their transformation to macrophages may be also long as several months or even years (A.V. Hoffbrand – 2005).
**Lymphocytes:** Lymphocytes are immunologically competent cells that assist the phagocytes in defense of the body against infection and other foreign invasion. In postnatal life, the bone marrow and thymus are the primary lymphoid organs in which lymphocytes develop. The secondary lymphoid organs in which specific immune responses are generated are the lymph nodes, spleen and lymphoid tissues of the alimentary and respiratory tracts (Daniel Catovsky - 2005). By their appearance under light microscope there are two broad categories of lymphocytes namely the large granular lymphocytes. Most but not all large granular lymphocytes are more commonly known as natural killer cells. The small lymphocytes are the T- cells and B- cells. The formation of lymphocytes is known as lymphopoiesis, B- cells mature in the bone marrow and circulate in the peripheral blood until they undergo recognition of antigen. While T- lymphocytes migrated to the thymus where they differentiate into mature T cells during the passage from the cortex to the medulla. They live weeks to several years, which are very long compared to other leukocytes. T&B cells are the major cellular component of the adaptive immune response. The T cell is involved in cell mediated immunity where as B cells are responsible for humoral immunity (Daniel Catovsky - 2005).

The function of the T&B cell is to recognize the specific antigen, during processes known as antigen presentation. The B cell responds to pathogens by producing large quantity of antibodies which then neutralize foreign objectives (Bacteria, Viruses).

In response to pathogens some T cells called helper T cells produce cytokine that direct the immune response while other T cells called cytotoxic T cell produce toxic granules that induce the death of pathogen infected cells (Daniel Catovsky - 2005). Natural killer cells (NK) are cytotoxic CD 8+ cells that lack the T- cell receptor (TCR) and also they are a part of innate immune system and play a major role in defending the host from both tumor and virally infected cells. N.K distinguishes infected cells and tumors from the normal and infected cells by recognizing alteration in the level of a surface molecule called major histocompatibility complex class I.

They are large cells with cytoplasmic granules and typically express surface molecules CD16 (Fc receptor), CD56 and CD57. NK cells are activated in response to family of cytokines called interferons. Activated NK cells release cytotoxic granules (cell killing) which then destroy the altered cells. They were named (NK cell) because of the initial notion that they don’t require prior activation in order to kill cells, which are missing Major Histocompatibility Complex I (MHC I) (Daniel Catovsky - 2005).

**MATERIAL AND METHODS**

**Study area:** The study was carried out in the Central blood bank, Wad Medanni teaching hospital. Wad Medanni is the capital of Gezira state, it is considered one of the largest states in Sudan with an area of 35.304 km and a population of 4 millions.

The Central Blood Bank provides blood donation services to 4 governmental hospitals and other special hospitals in Wad medanni. About 1600 to 1700 donors attend the central blood bank monthly. Different types of blood components (whole blood, packed red cells, platelets, fresh frozen plasma) are prepared from whole blood using large refrigerated centrifuges. All donors are selected according to the accepted criteria for donation, including age, weight, physical and medical examination and screening for viral infections (hepatitis B, C and HIV) and the test for syphilis. A haemoglobin level assessment is performed by copper sulphate method and donors are reported as fit for donation if a drop of blood sinks in a copper sulphate solution, of a certain specific gravity.

**Study population:** Apparently healthy male Blood donors attending the Central Blood Bank (500 donors).

**Selection criteria:** Donors were selected according to the accepted criteria for donation.
- Age between 18- 60 years.
- Weight: 50 Kg (110 pounds) and more.
- Haemoglobin : 12.5 g/dl - 17.5 g/dl

Donors were selected with clinical examination (abdominal, cardiopulmonary), pulse and blood pressure were measured, VDRL, hepatitis B,C and HIV were screened.

**Exclusion criteria**
- All donors should be clinically in a good health, subject with any disease symptoms and signs should be excluded.
- Any person taking medications.

**Study design:** Descriptive, prospective cross sectional study was conducted in wad Medanni central blood bank, during the period from 16/03/2009 to 26/12/2009.

**METHODS**

**Sample collection:** A total of 500 apparently healthy adult male donors were screened for White blood cell count. This analysis was conducted at the Wad Medanni central blood bank, department of pathology (medical laboratory) and the central laboratory of the Wad Medanni teaching hospital. Venous Blood samples were taken from an antecubital vein by a 5ml syringe. The site of collection was cleaned using 70% alcohol and left to dry. An elastic tourniquet was applied if needed to the arm. 2.5 ml of blood was taken in a container with 0.05ml (K2 EDTA) as an anticoagulant with a
concentration of 1.5-2.2 mg/ml and then the sample gently mixed. The blood samples were tested within 2 hours of sample collection using an automated blood cell counters (sysmex KN21 analyzer) with a flow cytometry using a laser light to perform white blood count. It is calibrated by a standardized commercially prepared calibrators (Dacie 2006).

**Making a blood film:** Manual spreading of blood films using frosted glass slides were performed. The frosted glass slides were clean and free of grease. A drop of blood was placed near one end of the slide and spreader was applied at an angle of 45, in front of the drop of blood, making a thin blood film using a cover glass as spreader and allowed to dry. Then they were labelled with the donor number and date of sample collection. The films were then fixed in absolute methanol for 10-20 minutes. The films were placed horizontally on the staining rack and flooded with Leishman's stain and left for 4 minutes.

A double volume buffer was added with gentle blowing over the surface without touching the film surface. The films were left for another 8 minutes and then washed off with buffered distilled water. The back of the slide was cleaned using cotton dipped in alcohol and then left to dry (Dacie 2006).

**Examination of the blood films:** The identification of the specimen was checked and matched with the white blood cells report. The films were examined macroscopically to confirm adequate spreading followed by microscopic examination. A low power field (10 objective) to assess the quality of the stain and (40 objective) to determine the suitable area for blood film examination (Dacie 2006). The white blood cells report was examined and an assessment of their number, size, morphology and presence of aggregate were also evaluated.

The aim of this study was to detect the white blood cells count in 500 apparently healthy male donors selected according to the accepted criteria for donation including age, weight, physical and medical examination. All donors were subjected to screening for viral infections (hepatitis B, C and HIV) and the test for syphilis. The White blood cell count was estimated using an automated cell counter (sysmex KN 21).

**Statistical analysis:** The results were analyzed using statistical software package of social sciences (SPSS) version 17 and descriptive data were expressed as means.

**Ethical clearance:** Ethical clearance was obtained from the University of Gezira ethical committee and blood bank authority. Verbal informed consent was obtained from all donors.

**RESULTS**

The white cells count mean value was found to be 5.64 +/- 1.7 standard deviation with a minimum value 2.1 and maximum value 14.1, with 77 cases ranged from 2.1 to 3.9, 372 cases ranged from 4 to 8, 46 cases ranged from 8.1 to 11 and 5 cases ranged from 11.1 to 14.1. The differential white blood count was performed manually based on counting 100 cells and expressed as percentage. This manual differential white blood cells were correlated with the automated differential white blood cells.

| Table 1: Mean, minimum, maximum values and standard deviation(SD) for the white blood count values in 500 apparently healthy Sudanese male donors. |
|---|---|---|---|---|
| Number of sample | Minimum value | Maximum value | Mean value | Standard Deviation |
| WBCs | 500 | 2.1 x 10^9/L | 14.1 x 10^9/L | 5.696 | 1.7989 |

The neutrophil count mean value was found to be 54.43 +/- 11.576 standard deviation with a minimum value 12% and maximum value 80%, the lymphocyte count mean value was found to be 41.20 +/- 11.497 standard deviation with a minimum value 16% and maximum value 88%. The eosinophil count mean value was found to be 3.18 +/- 2.269 standard deviation with a minimum value 1% and maximum value 16%, the monocyte count mean value was found to be 2.07 +/- 1.496 standard deviation with a minimum value 1% and maximum value 10% and the basophil count mean value was found to be 1.22 +/- 0.441 standard deviation with a minimum value 1% and maximum value 2%.

| Table 2: Mean, minimum, maximum values and standard deviation(SD) for the neutrophil % values in 500 apparently healthy Sudanese male donors. |
|---|---|---|---|---|
| Number of sample | Minimum value | Maximum value | Mean value | Standard Deviation |
| Neutrophils | 500 | 12% | 80% | 54.43 | 11.576 |
Table 3: Mean, minimum, maximum values and standard deviation(SD) for the lymphocyte % values in 500 apparently healthy Sudanese male donors.

<table>
<thead>
<tr>
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<th>Number of sample</th>
<th>Minimum Value</th>
<th>Maximum Value</th>
<th>Mean value</th>
<th>Standard Deviation</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lymphocytes</td>
<td>500</td>
<td>16%</td>
<td>88%</td>
<td>41.20</td>
<td>11.497</td>
</tr>
</tbody>
</table>

Table 4: Mean, minimum, maximum values and standard deviation(SD) for the eosinophils % values in 500 apparently healthy Sudanese male donors.

<table>
<thead>
<tr>
<th></th>
<th>Number of sample</th>
<th>Minimum Value</th>
<th>Maximum Value</th>
<th>Mean value</th>
<th>Standard Deviation</th>
</tr>
</thead>
<tbody>
<tr>
<td>Eosinophils</td>
<td>500</td>
<td>1%</td>
<td>16%</td>
<td>3.18</td>
<td>2.269</td>
</tr>
</tbody>
</table>

Table 5: Mean, minimum, maximum values and standard deviation(SD) for the monocyte % values in 500 apparently healthy Sudanese male donors.

<table>
<thead>
<tr>
<th></th>
<th>Number of sample</th>
<th>Minimum Value</th>
<th>Maximum Value</th>
<th>Mean value</th>
<th>Standard Deviation</th>
</tr>
</thead>
<tbody>
<tr>
<td>Monocytes</td>
<td>500</td>
<td>1%</td>
<td>10%</td>
<td>2.07</td>
<td>1.496</td>
</tr>
</tbody>
</table>

Table-6 Mean, minimum, maximum values and standard deviation(SD) for the basophil % values in 500 apparently healthy Sudanese male donors.

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<thead>
<tr>
<th></th>
<th>Number of sample</th>
<th>Minimum Value</th>
<th>Maximum Value</th>
<th>Mean value</th>
<th>Standard Deviation</th>
</tr>
</thead>
<tbody>
<tr>
<td>Basophils</td>
<td>500</td>
<td>1%</td>
<td>2%</td>
<td>1.22</td>
<td>0.441</td>
</tr>
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Microscopic examination: Hypersegmented neutrophils (1-2) observed in 15 cases (3%) with lymphocytes predominance in 159 cases (31.8%) and reactive lymphocytes (2-10) in 105 cases (21%), eosinophil count more than 4% detected in 72 cases (14%) and 15 cases (3%) with monocyte count more than 8%. No lymphoblast, myeloblast, promyelocytes, myelocytes, metamyelocytes detected in the blood films.

The neutrophil percentage less than 50 % observed in 165 donors (33%) and 231 donors (46.2%) and neutrophil percentage more than 70 % observed in 26 donors (5.2%) with lymphocyte percentage more than 40 % observed accompanied by reactive lymphocytes most probably due to chronic infection (Wilson -1991). The eosinophils percentage of more than 4% observed in 121 donors (24.2%), the bulk of this eosinophilia probably reflects asymptomatic parasitism (e.g. schistosomiasis). Monocyte percentage more than 8 % observed in 6 donors (1.2%) (Wilson -1991).

CONCLUSION AND RECOMMENDATIONS
1- The study revealed that significant number of study population had low white blood count.
2- The Sudanese population have lower white blood count compare with Caucasian population.
3- Significant numbers of Sudanese population were detected with low neutrophil, high lymphocyte and eosinophil count compare with Caucasian.
4- Similar study with increased sample size should be carried out to know the real reference range of Sudanese population.

REFERENCES