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Evaluation of Complete Blood Count of Children with Protein Energy Malnutrition at Khartoum State in Omdurman Pediatric and Albulk Pediatric Hospital between May to September 2015

قياس تعداد الدم الكامل في الاطفال الذين يعانون من مرض سوء التغذية في ولاية الخرطوم
مستشفى ام درمان للاطفال ومستشفى البلك للاطفال في الفترة من مايو الي سبتمبر 2015

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الآية

بسم الله الرحمن الرحيم

قال تعالى :

اللَّهُ نُورُ السَّمَاوَاتِ وَالْأَرْضِ ۗ مَثَلُ نُورِهِ كَمِشْكَاةٍ فِيهَا مِصْبَاحٌ ۗ الْمِصْبَاحُ فِي
زُجَاجَةٍ ۗ الزُّجَاجَةُ كَأَنَّهَا كَوْكَبٌ دُرِّيٌّ يُوقَدُ مِنْ شَجَرَةٍ مُبَارَكَةٍ زَيْتُونَةٍ لَا شَرْقِيَّةٍ وَلَا
غَرْبِيَّةٍ يَكَادُ زَيْتُهَا يُضِيءُ وَلَوْ لَمْ تَمْسَسْهُ نَارٌ ۗ نُورٌ عَلَىٰ نُورٍ ۗ يَهْدِي اللَّهُ لِنُورِهِ مَنْ
يَشَاءُ ۗ وَيَضْرِبُ اللَّهُ الْأَمْثَالَ لِلنَّاسِ ۗ وَاللَّهُ بِكُلِّ شَيْءٍ عَلِيمٌ ﴿٣٥﴾

صدق الله العظيم

سورة النور الآية 35

Dedication

To the candle which burns to light my life

My Mother

To the one whom I live for making his dream true

My Father

To special person who inspired and give me meaning of
being

My

And special thanks to my husband

To those who have made it possible

My Teacher

To whom encouraged me

My Brothers and Friends

Acknowledgment

Thanks to Allah would be before anything as without Allah will, this work would not be completed.

We are heartily thankful to my supervisor, Dr. Nadia Madni, for her encouragement, guidance and support from the initial to the final level enabled me to develop and of the understanding of the subject.

Abstract

This was prospective case control study, was conducted to determine changes of complete blood count (CBC) among children under 5 years old children with protein energy malnutrition (PEM), the study took place in Omdurman pediatric Hospital and AL bulk Pediatric Hospital in Khartoum state between May to September 2015.

Three ml of venous blood sample were collected from 80 of children affected with protein energy malnutrition under five years, (58.8% of children were males and 42.2% were females) and 20 control samples were collected from normal children under five years (58.7% were males and 41.3% were females, using disposable syringes and added slowly in K₃- EDTA container for CBC analysis.

The study showed statistically significant decrease in the mean of HB (8.4 ± 2 , 12.6 ± 1.2), RBC ($4.1 \pm 1. \pm 7$, 4.7 ± 0.2), PCV (26.6 ± 5.3 , 40.1 ± 3.6), MCV (72.6 ± 13.4 , 88.1 ± 4.2) and MCH (22.8 ± 5.6 , 27.8 ± 0.8) respectively in children with protein energy malnutrition when compare to control. (*p.value* 0.000) for all parameters.

There was no statistically significant between MCHC of children with protein energy malnutrition compare to control (31.3 ± 3.3 , 31.3 ± 3.3) (*p.value* 0.9).

The study showed statistically significant increase in the mean of white blood cell count (13.2 ± 3.8 , 7.3 ± 1.5), lymphocyte (53.4 ± 13.9 , 27.5 ± 4.5) and platelet (384.2 ± 163.7 , 265.8 ± 55.1) and decrease in the mean of neutrophil (39.3 ± 13.7 , 61.1 ± 4) in children with protein energy malnutrition compared to control (*p.value* 0.000) for all parameters.

I conclude from this study low level of HB, RBCs, PCV, MCV, MCH, normal level in MCHC and neutrophil, and high level WBCs,

lymphocyte and PLTs, in patient with protein energy malnutrition when compare to control.

المستخلص

كان هذا المحتملين دراسة مراقبة حالة، أجريت لتحديد التغيرات في تعداد الدم الكامل (CBC) بين الأطفال دون سن 5 سنوات من العمر الأطفال يعانون من سوء التغذية البروتين والطاقة (بيم)، أخذت الدراسة في مستشفى أم درمان الأطفال و آل الأكبر مستشفى الأطفال في ولاية الخرطوم بين مايو-سبتمبر. العام 2015.

تم جمع ثلاثة مل من عينة من الدم الوريدي من 80 من الأطفال المصابين بسوء التغذية البروتين والطاقة دون سن الخامسة، (كانت 58.8% من الأطفال الذكور، وكانت 42.2% للإناث)، وجمعت 20 عينة مراقبة من الأطفال العاديين في ظل خمس سنوات (58.7% كانت الذكور و 41.3% من الإناث، وذلك باستخدام المحاقن وأضاف ببطء في K3- EDTA حاوية للتحليل CBC.

وأظهرت الدراسة انخفاض ملحوظ إحصائيا في متوسط 2 ± 8.4 HB، 12.6 ± 1.2 ، 7 ± 1.1 RBC و 0.2 ± 4.7 ، 26.6 ± 5.3 PCV، 40.1 ± 3.6 ، 72.6 ± 13.4 MCV، 4.2 ± 88.1 وصحة الأم والطفل 5.6 ± 22.8 ، 27.8 ± 0.8 على التوالي في الأطفال الذين يعانون من سوء التغذية البروتين والطاقة عند مقارنة السيطرة عليها. ($p.value 0.000$) لجميع المعلمات.

لم يكن هناك دلالة إحصائية بين MCHC من الأطفال الذين يعانون من سوء التغذية البروتين والطاقة مقارنة للسيطرة على 3.3 ± 31.3 ، 3.3 ± 31.3 (0.9) ($p.value$).

وأظهرت الدراسة زيادة significant إحصائيا في متوسط عدد خلايا الدم البيضاء 53.4 ± 13.9 ، 1.5 ± 7.3 ، 3.8 ± 13.2 ، 4.27 ± 5 lymphocyte، والصفائح الدموية 265.8 ± 384.2 ، 55.1 ± 163.7 و انخفاض في متوسط neutrophil 39.3 ± 13.7 ، 4 ± 61.1 في الأطفال الذين يعانون من سوء التغذية البروتين والطاقة مقارنة مع الشاهد ($p.value 0.000$) لجميع المعلمات.

أنا نستنتج من هذا المستوى الدراسي المنخفض من HB، كرات الدم الحمراء، PCV، حجم الكرية، المستوى العادي في MCHC والعدلة، والكريات البيضاء على مستوى عال، لمفاوية و PLTS، في المريض مع نقص البروتين والطاقة عند مقارنة السيطرة عليها

Abbreviations

AIDS	Acquired immune deficiency syndrome
BMI	Body mass index
CBC	Complete blood count
EDTA	Ethyl dimen tetra acetic acid
HB	Haemoglobin
MCH	Mean cell haemoglobin
MCHC	Mean cell haemoglobin concentration
MCV	Mean cell volume
PCV	Packed cell volume
PEM	Protein energy malnutrition
PLT	Platelets
RBC	Red blood cell
SPSS	Statistical package of social science
WBC	White blood cell

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Chapter One

Introduction and Literature review

Chapter one

Introduction and Literature Review

1.1 Introduction

Blood performs many important functions within the body including: supply of oxygen to tissues (bound to hemoglobin, which is carried in red cells), Supply of nutrients such as glucose, amino acids, and fatty acids (dissolved in the blood or bound to plasma proteins (e.g., blood lipids)), removal of waste such as carbon dioxide, urea, and lactic acid. Immunological functions, including circulation of white blood cells, and detection of foreign material by antibodies, coagulation, the response to a broken blood vessel, the conversion of blood from a liquid to a semi-solid gel to stop bleeding, messenger functions, including the transport of hormones and the signaling of tissue damage, regulation of body pH and regulation of core body temperature (Alberts, 2012).

The World Health Organization (WHO) defines malnutrition as "the cellular imbalance between the supply of nutrients and energy and the body's demand for them to ensure growth, maintenance, and specific functions." The term protein-energy malnutrition (PEM) applies to a group of related disorders that include [marasmus](#), kwashiorkor, and intermediate states of marasmus-kwashiorkor (Abobakar *et al.*, 2012).

The WHO estimates that by the year 2015 the incidence of malnutrition will have decreased to 17.6%. Currently, 113.4 million children are affected by protein energy malnutrition "PEM" as measured by low weight for age, the majority of these cases living in developing countries with "70%" of these children in Asia particularly south central region and "26%" in Africa, more than half of young children in south Africa have "PEM" which is 6.5 time prevalenc in

the western hemisphere, In sub –Sahara Africa 30% of children have "PEM"(Bench *et al.*,2010).

Protein energy malnutrition results in various change in the body including change in haematological profile of the body. Low red cell count resulting in anaemia has always been constant feature of protein energy malnutrition and may be normochromic normocytic ,microcytic hypochromic, or macrocytic. The anaemia of malnutrition may be attributable to various factors such as iron deficiency, and/or reduced red cell production in adaptation to a smaller lean body mass. Erythropoietin deficiency, deficiencies of vitamins (folic acid,B12) or trace elements (copper,zinc), infections and chronic disease have also been implicate(Saka *et al.*,2012)..

White cell changes seen in protein energy malnutrition varies and such changes have been attributed to various factors. These include the synergist relationship which PEM has with infections and thymic atrophy seen in children with PEM (Saka *et al.*, 2012).

1.2 Literature review

1.2.1 Hemopoiesis

Hemopoiesis means the formation of blood cells which is determined by the interaction of multiple genes and involves cytokines and other protein factors. During the first few weeks of embryonic life, the formation of blood cells takes place in the yolk sac. Later, until the sixth or seventh month of fetal development, the liver and spleen are the major hematopoietic organs. By the time of birth, more than 90% of all new blood cells are formed in the bone marrow. During infancy and childhood, the marrow of all bones contributes to hematopoiesis. During adult life, hematopoietic marrow is restricted to certain bones (e.g., pelvic bones, vertebral column, proximal ends of the femur, skull, ribs, and sternum). Even in these areas, a proportion of the marrow cavity consists of fat. During periods of hematopoietic stress (e.g., in severe hemolytic anemias and in some myeloproliferative disorders), the fatty marrow as well as the spleen and liver can resume the production of blood cells. This situation is called extramedullary Hematopoiesis (Munker *et al.*, 2007).

1.2.1.1 Erythropoiesis

Red blood cells are specialized cells that deliver oxygen to tissues and remove carbon dioxide from the human body. Erythropoiesis, “making of red cells,” involves many different genes and gene products that lead to the production of the mature cell.

Erythropoiesis begins at the level of the multipotent stem cell, which then undergoes commitment and differentiation. Listed as follows are the stages of erythroid differentiation:

1. Stem cell.

2. Burst-forming unit, erythroid (BFU-E); immature erythroid progenitor.
3. Colony-forming unit, erythroid (CFU-E); more mature erythroid progenitor.
4. Proerythroblasts, erythroblasts, normoblasts (morphologically recognizable red cell precursors, they still have a nucleus, multiply by cell division, and progressively decrease in size as hemoglobin content increases).
5. Reticulocytes; mature red blood cells (Munker *et al.*, 2007).

1.2.1.2 The Red Blood Cell

The normal erythrocyte has a diameter of about 8 μm and a biconcave disc form that provides the red cell with a maximum surface-for-gas exchange as well as optimal deformability. The bipolar lipid layer of the red cell membrane is stabilized on the inner side by the attachment of the structural proteins actin and spectrin. Defects of these proteins lead to hemolytic anemia. The outer layer is covered with mucopolysaccharides that form part of the structure of blood group antigens. The *N*-acetylneuraminic acid found in these glycoproteins results in a negative charge of the cell surface. (Munker *et al.*, 2007).

Because red cells have lost their nuclei, they are no longer capable of synthesizing proteins, including enzymes. Red cells remain viable and functional for an average of 120 days. The necessary energy for red cell metabolism is supplied by the Embden-Meyerhof pathway, which generates adenosine triphosphate by metabolizing glucose to lactate. This anaerobic process also results in the formation of nicotinamide-adenine dinucleotide, which is essential for the reduction of methemoglobin to functionally active hemoglobin (Munker *et al.*, 2007).

1.2.1.3 Hemoglobin

Hemoglobin is the molecule responsible for the transport of oxygen.

Under physiological conditions, three types of hemoglobins exist:

- Hemoglobin A ($\alpha_2 \beta_2$): major adult hemoglobin (96–98%).
- Hemoglobin F ($\alpha_2 \gamma_2$): predominant during fetal development, 60–80% at birth, 0.5–0.8% during adult life.
- Hemoglobin A₂ ($\alpha_2 \delta_2$): normally 1.5–3%.

The hemoglobin molecule has a molecular weight of 64,500KD and consists of four polypeptide chains, each carrying a heme group. The heme synthesis starts with the amino acid glycine. Later, porphobilinogen, uroporphyrinogen, coproporphyrinogen, and protoporphyrin are formed as intermediate steps. Iron (Fe^{+2}) is supplied from serum transferrin and combines with protoporphyrin to form heme. One heme molecule then binds with one globin chain to form the hemoglobin molecule that avidly binds oxygen. The release of oxygen from red cells into tissue is strictly regulated. (Munker *et al.*, 2007).

1.2.1.4 Granulopoiesis

The blood granulocytes and monocytes are formed in the bone marrow from a common precursor cell. In the granulopoietic series progenitor cells, myeloblasts, promyelocytes and myelocytes form a proliferative or mitotic pool of cells while the metamyelocytes, band and segmented granulocytes make up a post-mitotic maturation compartment. Large numbers of band and segmented neutrophils are held in the marrow as a 'reserve pool' or storage compartment (Hoffbrand *et al.*, 2006).

The bone marrow normally contains more myeloid cells than erythroid cells in the ratio of 2 : 1 to 12 : 1, the largest proportion being neutrophils and metamyelocytes. In the stable or normal state, the bone marrow storage compartment contains 10-15 times the number of granulocytes found in the peripheral blood. Following their release from the bone

marrow, granulocytes spend only 6-10 hours in the circulation before moving into the tissues where they perform their phagocytic function. In the bloodstream there are two pools usually of about equal size: the circulating pool (included in the blood count) and the marginating pool (not included in the blood count) (Hoffbrand *et al.*, 2006).

It has been estimated that they spend on average 4-5 days in the tissues before they are destroyed during defensive action or as the result of senescence (Hoffbrand *et al.*, 2006).

1.2.3 white blood cells (leukocytes)

The white blood cells (leucocytes) may be divided into two broad groups: the phagocytes and the immunocytes. Granulocytes, which include three types of cell-neutrophils (polymorphs), eosinophils and basophils-together with monocytes, comprise the phagocytes. Only mature phagocytic cells and lymphocytes are found in normal peripheral blood. The function of phagocytes and immunocytes in protecting the body against infection is closely connected with two soluble protein systems of the body: immunoglobulins and complement (Hoffbrand *et al.*, 2006).

1.2.1.3 Platelet production (Thrombopoiesis)

Platelets are produced predominantly by the bone marrow megakaryocytes as a result of budding of the cytoplasmic membrane. Megakaryocytes are derived from the haemopoietic stem cell, which is stimulated to differentiate to mature megakaryocytes under the influence of various cytokines, including thrombopoietin. Once released from the bone marrow young platelets are trapped in the spleen for up to 36 hours before entering the circulation, where they have a primary hemostatic role (Provan, 2003).

1.2.4 protein energy malnutrition

Protein energy malnutrition(PEM), is defined as spectrum of disease arising as a result of an absolute, or relative deficiency of calories and or protein in the diet (Murray, 2008).

It is the most important risk factor for illness and death, with hundreds of millions of young children affected (Muller and Krawinkel, 2005).

Protein-energy malnutrition (PEM) may be present at any time during the human life cycle, but it is more common in the extreme ages that is, during infancy/childhood and in the elderly (Paddon *et al.*, 2008).

Protein energy malnutrition in children (PEM) is a pathologic depletion of the body's lean tissues caused by starvation, or a combination of starvation and catabolic stress (Paddon *et al.*, 2008).

It is the disease that develops when protein intake or energy intake, or both, chronically fail to meet the body's requirements for these nutrients (Njenga, 2014).

The underlying mechanisms include decreased food intake because of anorexia, decreased nutrient absorption, increased metabolic requirements and direct nutrient loss (Gonzalez and Torres, 2004).

Patients that lose 10–20 percent of their body weight may have moderate PEM. Losing 20 percent of body weight or more is generally classified as severe PEM (Gonzalez and Torres, 2004).

Primary PEM results from a diet that lacks sufficient sources of protein and/or energy, while secondary PEM usually occurs as a complication of chronic diseases such as AIDS, cancer, chronic kidney failure, inflammatory bowel disease, and other illnesses that impair the body's ability to absorb or use nutrients or to compensate for nutrient losses (Hamer *et al.*, 2004).

protein-energy malnutrition (PEM) is a problem in many developing countries, most commonly affecting children between the ages of 6 months and 5 years. The condition may result from lack of food or from infections that cause loss of appetite while increasing the body's nutrient requirements and losses. Children between 12 and 36 months old are especially at risk since they are the most vulnerable to infections such as gastroenteritis and measles (Ayieko *et al.*, 2011)

1.2.4.1 Pathophysiology and clinical feature of PEM

PEM is characterized by atrophy and weakness of the skeletal muscles (including the respiratory muscles), reduced heart muscle mass, impaired wound healing, skin thinning with a predisposition to decubitus ulcers, fatigue, apathy and hypothermia.

The extracellular fluid compartment characteristically expands in PEM,

occasionally causing oedema (Njenga, 2014). Failure of pigments synthesis in hair and skin (e.g., hair colour may change and skin becomes hyperpigmented) due to lack of substrate (e.g., tyrosin) and coenzymes (Muller and Krawinkel, 2005).

The other essential aspects of severe protein-energy malnutrition are the fatty degeneration of the liver and heart. This degeneration is not just a sign of severe malnutrition; it also causes subclinical or overt cardiac insufficiency, especially when malnutrition is accompanied by oedema. If the myocardial insufficiency is not corrected, iatrogenic fluid and sodium overload quickly escalate it into cardiac failure (Kwena *et al.*, 2003). Another injurious aspect of PEM is the loss of subcutaneous fat, which markedly reduces the body's capacity for temperature regulation and water storage (Alam *et al.*, 2003). As a result, malnourished children become dehydrated, hypothermic and

hypoglycemic more quickly and severely than others (murugaiah *et al.*, 2014).

Severe protein–energy malnutrition is associated with atrophy of the mucosa

of the small bowel, leading to a loss of absorption as well as of digestion capacity (Alam *et al.*, 2003). Furthermore PEM is associated with chronic hypovolaemia, which leads to secondary hyperaldosteronism, and further complicates fluid and electrolyte balance (Kwena *et al.*, 2003). PEM affected children do not show signs of hyperkalaemia. This is because the development of muscular dystrophy mobilizes much of the body’s potassium, which is then lost through urine (Brewster, 2011).

1.2.4.2 types of protein energy malnutrition

1_kwashiorkor

Kwashiorkor, also called wet protein-energy malnutrition, it is a form of PEM characterized primarily by protein deficiency. This condition usually appears at the age of about 12 months when breast feeding is discontinued, but it can develop at any time during a child's formative years (fabiansen *et al.*, 2015). Kwashiorkor usually manifests with fluid retention (oedema) usually starting in the legs and feet and spreading, in more advanced cases, to the hands and face. Oedema may be detected by the production of a definite pit as a result of moderate pressure for 3 seconds with the thumb over the lower end of the tibia and the dorsum of foot, children with kwashiorkor may look “fat” due to oedema so that their parents regard them as well fed (fabiansen *et al.*, 2015).

There is hair discoloration or loss of pigmentation; curly hair becomes straight easily pluckable. Coloured, dark skin may become

dried and lighter in some places especially in the skin folds; outer layers of skin may peel off and ulceration may occur; the lesions may resemble burns (Cundiff and Harris, 2006). Children with Kwashiorkor are usually apathetic, miserable, and irritable.

2-Marasmic kwashiorkor

This is a severe wasting in the presence of oedema. It is a mixed form of PEM, and manifests as oedema occurring in children who may or may not have other signs of Kwashiorkor (fabiansen *et al.*, 2015).

3- Marasmus

Early marasmus occurs usually in the first year of life in children who have been weaned from breast milk or who suffer from weakening conditions like chronic diarrhoea. It is frequently associated with contaminated bottle-feeding in urban areas (ligarotimi ,2013).Primarily marasmus is caused by energy deficiency from prolonged starvation. It may also result from chronic or recurrent infections with marginal food intake (Abubakar *et al.* ,2012).

Marasmus is characterized by stunted growth and wasting of muscle and tissue. Wasting indicates recent weight loss, whereas stunting usually results from chronic weight loss. The major nutritional indicators underweight (low weight-for-age); and wasting (low weight-for-height). Of the three (3), wasting is the most dangerous and signifies acute malnutrition (Muller and Krawinkel, 2005).

The main sign is a severe wasting and the child appears very thin and has no fat. Most of the fat and muscle mass have been expended to provide energy. There is severe wasting of the shoulders, arms, buttocks and thighs, with no visible rib outlines. There is no oedema(swelling that pits on pressure) of the lower extremities (Brewster ,2011).

Clinical aspects typically include a triangular face, extended abdomen (from muscular hypotonia) and anal or rectal prolapse (from loss of perianal fat) (fabiansen *et al.*, 2015).

1.2.4.3 Laboratory studies

The WHO recommends the following laboratory tests:

- Blood glucose
- Examination of blood smears by microscopy or direct detection testing
- Hemoglobin
- Urine examination and culture
- Stool examination by microscopy for ova and parasites
- Serum albumin
- HIV test (This test must be accompanied by counseling of the child's parents, and strict confidentiality should be maintained.)
- Electrolytes.

Significant findings in kwashiorkor include hypoalbuminemia (10-25 g/L), hypoproteinemia (transferrin, essential amino acids, lipoprotein), and hypoglycemia. Plasma cortisol and growth hormone levels are high, while insulin secretion and insulinlike growth factor levels are decreased.

The percentage of body water and extracellular water is increased. Electrolytes, especially potassium and magnesium, are depleted. Levels of some enzymes (including lactase) are decreased, and circulating lipid levels (especially cholesterol) are low. Ketonuria occurs, and protein-energy malnutrition may cause a decrease in the urinary excretion of urea because of decreased protein intake. In both kwashiorkor and marasmus, iron deficiency anemia and metabolic acidosis are present. Urinary excretion of hydroxyl proline is diminished, reflecting impaired growth and wound healing. Increased urinary 3-methylhistidine is a reflection of muscle breakdown and can be seen in marasmus. Malnutrition also causes immune

suppression, which may result in false-negative tuberculin skin test results and the subsequent failure to accurately assess for tuberculosis (Harima *et al.*, 2010).

And other test include Detailed dietary history, growth measurements, body mass index (BMI), and a complete physical examination are indicated. Sensitive measures of nutritional deficiency in children include height-for-age or weight-for-height measurements less than 95% and 90% of expected, respectively, or greater than 2 standard deviations below the mean for age. In children older than 2 years, growth of less than 5 cm/y may also be an indication of deficiency (Harima *et al.*, 2010).

1.3 Previous studies

Previous study in Sudan by Mohammed and Mirgani in 2011 evaluated hematological parameters in children under five years with protein energy malnutrition, and the result of complete blood count was conclude that low level of Hb, RBC, PCV, MCV, MCH, and normal MCHC, associated with high WBC and platelet. Hematological status of test group was significantly altered compared to control. (Mohammed and mirgani, 2011).

Other previous study done in north central Nigeria by Saka *et al* in 2012 evaluated hematological parameters in children with protein energy malnutrition under five years, and the result confirm the present of anemia due to low level of Hb, RBC, PCV, MCV, MCH, and also present of infection due to high level of white blood cells, and the result also reported the normal level of MCHC and platelet (Saka *et al.*, 2012).

1.4 Rationale

Malnutrition is one of health problems in developing countries in particular..Poor nutritional status of children is a major health problem throughout the developing world, and the underlying cause for 35% of child deaths and 11% of the total global disease burden(Ahmed *et al*,2013).

The global burden of childhood mortality, morbidity and under nutrition is now increasingly concentrated in the most deprived. The WHO estimates that by the year 2015 the incidence of malnutrition will have decreased to 17.6%, (Benach *et al*,2010). Protein energy malnutrition results in various change in the body including change in haematological profile of the body. Low red cell count resulting in anaemia has always been constant feature of protein energy malnutrition and may be normochromic normocytic ,microcytic hypochromic, or macrocytic, and white cell changes seen in protein energy malnutrition varies and such changes have been attributed to various factors.

Maternal and child health centers are often not available in many areas in Sudan.

1.5 Objectives

1.5.1 General objective

To study haematological parameters in children with protein energy malnutrition.

1.5.2 Specific objectives

-To compare complete blood count of children with protein energy malnutrition and aberrantly healthy children .

-To measure the difference in level of HB ,PCV ,RBC, MCV, MCH, MCHC,WBC and PLTs count between children with protein energy malnutrition and healthy one.

Chapter Two

Materials and Methods

Chapter two

Materials and methods

2.1 Study design

This was a prospective case control study, conducted at Omdurman Pediatrics Hospital and ALbulk Pediatrics Hospital at Khartoum state. Study was carried out during the period from May to September 2015.

2.2 Study population

Children with protein energy malnutrition under five years old .

2.2.1 Inclusion criteria

Children with age below 5 years old and newly diagnosed with protein energy malnutrition and before receiving treatment were included.

2.2.2 Exclusion criteria

Any child with co-morbidities, and any child with history of pre term labour.

2.3 Sample size

Eighty blood sample of children with protein energy malnutrition collected and 20 sample of normal children collected in K₃-ethyle dimen tetra acetic acid container.

2.4 methods of data collection

A questionnaire was filled for each patient by parents.

2.5 Sample collection

Three ml of venous blood were collected using disposable syringes and added slowly to K3- EDTA containers for CBC analysis.

2.6 Methodology

Sysmex kx-21N haematological analyzer was used to measure HB,PCV,RBC count,MCV,MCH,MCHC,WBC count and PLTs count.

2.6.1 Principle of sysmex autoanalyzer

The kx-21N employs three detector blocks and two kinds of reagents for blood analysis. WBC count was measured by WBC detector block using detection method. RBC and PLTs count were taken from RBC detector block , also using the DC detection method. The hemoglobin detector block measure hemoglobin concentration using the non cyanidehemoglobin method. A blood sample was aspirated , measured to predetermined volume , diluted at the specified ratio , then fed into each transducer. The transducer chamber has a minute hole called aperture. On both side of the aperture, there are the electrodes between which flows direct current . blood cells suspended in the diluted sample pass through the aperture , causing direct current resistance to change between the electrodes .As direct current resistance change , the blood cell size is detected as electric pulses.

Blood cell count is calculated by counting the pulses, and histogram of blood cell sizes is plotted by determining the pulse sizes. Also, anything a histogram makes it possible to obtain various analysis data. For hemoglobin determination the non-cyanide hemoglobin method was used. Blood hemoglobin was converted rapidly to oxyhemoglobin. The later absorbance was measured in the Hb flow cell at 555 nm wavelength against diluent and the concentration of Hb was calculated.

Sysmex KX-21N calculates the RBC indices (mean corpuscular volume) MCV,mean corpuscular hemoglobin MCH, and mean

corpuscular Hb concentration from red cell count (RBC), hemoglobin concentration (Hb) and packed cell volume (PCV).

2.6.2 Quality control

All quality control for machine done in instructed manner. The dialyzer, weekly and monthly maintenance and calibration used to ensure assurance. Then before using the apparatus one of the last samples was reanalyzed for delta check.

2.7 Ethical consideration

Study protocol was approved by Ethical Committee of Medical Laboratory Science, Sudan University for Sciences and Technology, written informed consent was taken for parents.

2.8 Data analysis

Data were collected manually and analysis was performed using computerized program (SPSS) version 20.

Chapter Three

Results

Chapter three

Results

Eighty malnutrition patients under 5 years of age were included with mean age (18.46±1.2), (58.7% were males and 41.3 were females), compare with 20 of normal children with mean age (19.45±1.8) (60% were males and 40% were females). The result was showed decrease in the mean of weight in children with protein energy malnutrition compared with control (6.4±1.5, and 12.4±1.9 respectively). **Table(3.1):** Distribution of study groups

according to age , sex and weight:

Variable	PEM	Control
Age mean ±SD(months)	18.46±1.2	19.45±1.8
Sex male	80 (58.7%)/47	20 (60%)/12
Female	80 (41.3%)/33	20 (40%)/8
Weight(kg)	6.4±1.5	12.4±1.9

The study showed statistically significant decrease in the mean of (Hb, RBC, PCV, MCV and MCH) in study groups compared with control. (*p, value* 0.000)(table 3.2).

The study showed no statistically significant of MCHC in PEM when compare to control. (*P. value* 0.9)(table 3.2)

Table3(3.2): Red blood cells parameters of study group (HB, RBC, PCV, MCV, MCH and , MCHC)

Haematological parameters	PEM mean \pmSD	Control mean \pmSD	<i>P.value</i>
Haemoglobin(%)	8.4 \pm 2	12.6 \pm 1.2	0.000
Red blood cell($\times 10^6$ cell/mm ³).	4.1 \pm 1.7	4.7 \pm 0.2	0.000
Haematocrit(%)	26.6 \pm 5.3	40.1 \pm 3.6	0.000
Mean cell volume(fl)	72.6 \pm 13.4	88.1 \pm 4.2	0.000
Mean cell haemoglobin(pg)	22.8 \pm 5.6	27.8 \pm 0.8	0.000
Mean cell haemoglobinconcentration(%)	31.3 \pm 3.3	31.4 \pm 1.1	0.9

The mean of (WBC, lymphocyte, and PLTs) was increased in PEM, compared with control, and decrease in the mean of neutrophil.(table3.3) .

Table3(3.3): white blood cells and platelets parameters of study group

<i>P.value</i>	Control mean \pmSD	PEM mean \pmSD	Haematological parameters
0.000	7.3 \pm 1.5	13.2 \pm 3.8	White blood cell($\times 10^3$ cell/mm ³)
0.000	27.5 \pm 4.5	53.4 \pm 13.9	Lymphocyte(%)
0.000	61.1 \pm 4	39.3 \pm 13.7	Neutrophils(%)
0.000	265.8 \pm 55.1	384.2 \pm 163.7	Platelet($\times 10^3$ cell/mm ³)

Chapter four

Discussion, Conclusion, and Recommendations

Chapter four

4.1- Discussion

It is recognized that the vast majority of deaths among children in the developing world is associated with malnutrition. Protein energy malnutrition results in various changes in the body including change in hematological profile of the body. Low red cell count resulting in anemia has always been constant feature of protein energy malnutrition and may be normochromic normocytic, microcytic hypochromic, or macrocytic.(Saka *et al.*, 2012).

Our present study confirms statistically significant anemia in patients with protein energy malnutrition compared to the control. There is a decrease in the mean of the HB, RBC, PCV, MCV, MCH, in children with protein energy malnutrition compared with the mean of control. The anemia is microcytic hypochromic, this results agreed to the results reported by El Nawawy, *et al.*, who reported decrease in the mean of HB, RBC, PCV, MCH, but showed no change in the mean of MCV (El Nawawy *et al.*, 2002). This study agreed with this reported by Shaikh *et al.*, who reported that the anaemia affected patients with protein energy mal nutrition is iron deficiency anaemia (Shaikh *et al.*, 2013) . This study disagree with study conducted by Yaikhomba *et al.*, who conclude that Vitamin B12 deficiency was more common than iron and folate deficiencies in patients with severe acute malnutrition, (Yaikhomba *et al.*,2015).

In current study there is increase mean of total WBC, lymphocyte and decrease in the mean of neutrophil count in PEM when compared to control. This study agreed with study conducted by Borelli *et al.*, who studied the literature of protein energy malnutrition and conclude that Leucopenia and leucocytosis are situations that have been reported in

malnutrition, as it is usually accompanied by infectious processes or chronic disease(Borelli *et al*, 2004).This result is disagree with study conducted by Ugwuja *et al* which show depression of total white blood cell count in protein energy malnutrition patient compared to the control, he related this depression of WBC to Zinc and copper deficiency(Ugwuja *et al*.,2007).

In this study there was statistically significant increase in the mean of platelets in children with PEM when compared to control , this result disagree with study conducted by Uner *et al*, which showed reduction in the mean of platelet count in patient with protein energy malnutrition ,the number of platelet return to normal value when patients caught the normal weight (Uner *et al*.,2001).

4.2 Conclusion

The mean of HB, RBC, PCV, MCV, MCH, were significantly decreased in protein energy malnutrition compared with control.

The mean of MCHC was insignificant in protein energy malnutrition compared with control.

The mean of WBC, lymphocyte, and PLT were significantly increased in protein energy malnutrition compared with control.

4.3 Recommendations

- Patients with protein energy malnutrition should be followed up and complete blood count should be done regularly.
- In order to get more informative data, the large sample size should be increased in related subsequent researches.
- A further study about iron profile level (serum iron and serum ferritin) in patient with protein energy malnutrition should be conducted.

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Appendix I

Questionnaire:

Sudan University of Science and Technology

Colleges of Graduate Studies

Evaluation of Complete Blood Count in Children with Protein Energy Malnutrition at Khartoum State in Omdurman Pediatric and Albulk Pediatric Hospital between May to September 2015

NO-----DATE----/-----

Age;-----

Phone;-----Address-----

Weight-----

RESULT;

CBC;

Hb-----PCV-----RBCs count-----PLT count-----

MCV-----MCH-----MCHC-----

-TWBCs-----lymphocyte-----neutrophil-----

Appendix II



Automated hematology analyzer (sysmex KX-21N)