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Determination of Anaemia Subtypes in Children from Six to Ten years old in Almesiktab Hospital

A thesis submitted for partial fulfillment of the degree of M.Sc. in medical Laboratory Sciences (Hematology)

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قال تعالى:-

وَوَصَمَّيْنَا الْإِنسَانَ بِوَالِدَيْهِ إِحْسَاناً حَمَلَتْهُ أُمُّهُ كُرْهاً وَوَضَعَتْهُ كُرْهاً وَحَمْلُهُ وَفِصَالُهُ ثَلَاثُونَ شَهْراً حَتَّى إِذَا بَلَغَ أَشُدَّهُ وَبَلَغَ أَرْبَعِينَ سَنَةً قَالَ رَبِّ أَوْزِعْنِي أَنْ أَشْكُرَ نِعْمَتَكَ الَّتِي أَنْعَمْتَ عَلَيَّ وَعَلَى وَالِدَيَّ وَأَنْ أَعْمَلَ صَالِحاً تَرْضَاهُ وَأَصْلِحْ لِي فِي ذُرِّيَتِي إِنِي تُبْتُ إِلَيْكَ وَإِنِي مِنَ الْمُسْلِمِينَ ﴾

سورة الأحقاف الآية (١٥)

Dedication

To the most person in my life who decorates

My sky her continuous brilliance .

Dear Mother,,,

You always be To unique source of inspiration to success,

the person who did not, and will not repeated for ever.

Dear Father ",

Your fingetprint in my life will never disappear.

And also to Dears:

My Brother.

My Sister.

My girls.

Acknowledgment

I would like to express my gratitude –after Allah –to my supervisor, Who guided me and patiently throughout this work .

Dr. Mohammed Osman Ali

Great thanks to every member in the gripping university.

I would like to extend my deep respect and great

Fluffiness

To the teaching staff in university of shendi ,Faculty of laboratory medicine, especially the department of hematology

List of abbreviation

Abbreviation	Term
Hb	Hemoglobin
RBCs	Red Blood Cells
MCV	Mean Cell Values
FL	Fillitometer
CBC	Complete Blood count
TIBC	Total Iron Binding Capacity
G6PD	Glucose 6 Phosphate Dehydranase
LDH	Lactate dehydrogenase
IDA	Iron Deficiency Anemia
RDV	Red Distribution Widlth
НСТ	Haematocrit

Abstract

Background: Anemia is a major health problem world wide. Because and socioeconomic problem, the prevalence of anemia is higher in developing countries, children are most vulnerable group of anemia the aim of the present study was to determine severity and type of anemia in children in Almesktab hospital aged from 6 to 10 years old.

Methodology:

A cross-sectional was conducted in April to August 2018, using pretest questionnaire for parent for data collection. Laboratory assessment of anaemia performed through venous blood collection on EDTA, the tested for complete blood count and serum iron.

Diagnosis of anemia was based on Hb level and clinical presentation, classification of anemia was done by clinical finding, complete blood picture, with peripheral smear and serum iron.

Results:

Regarding the Severity of anemia, the results of this study revealed that, (30%)had mild anemia, while (60%)had moderate anemia, and (10%) have severe anemia.

Also the results showed that (75%) of anemic children had normocytic normochromic, while (25%) had microcytic hypochromic.

Also the results of this study revealed that the anemia was found in (54%) of female and (46%) of male.

Conclusion:

The end contain the conclusion of the type of anemia that found that(75%)of aemic children had normocytice normochromic , while(25%) had microcytic hypochromic aemia .

ملخص البحث

فقر الدم من المشاكل الصحية التي تواجه العالم والأطفال من الفئات الضعيفة القابلة للإصابة بفقر الدم، وفى دراسة أجريت في مستشفى المسيكتاب بمحلية شندي للأطفال الذين لديهم أفقر الدم في الفئات العمرية من عمر ستة إلى عشرة سنوات.

وتم جمع عينة دم وريدي من الأطفال المصابين بفقر الدم , وتم تشخيص الأنيميا بعمل فحص الدم الكامل وقياس نسبة الحديد للأطفال الذين لديهم أنيميا نقص الحديد وعمل مسحة دم لتحديد وأشكال كريات الدم الحمراء.

من النتائج (٣٠%) لأطفال لديهم أنيميا بسيطة، (٣٠%) من الأطفال لديهم أنيميا متوسطة و(١٠%) لديهم أنيميا حادة. (٣٢%) من الذكور لديهم أنيميا نقص الحديد و(٦٨%) من الإناث لديهن أنيميا نقص الحديد.

أما عن أنواع فقر الدم بين الأطفال كانت أعلى نسبة انتشار (٢٥%) من طبيعية الحجم سوية الصبغة (Normocytic normochromic anemia), (٢٥%) من صغيرة الحجم ناقصة الصبغة Microcytic hypochromic anemia

وكانت نسبة انتشار فقر الدم بين الذكور (٤٦%) بينما كانت النسبة بين الإناث (٤٥%) . في الختام يجب تعليم الأمهات عمل موازنة في الغذاء بين مصدر حيواني ونباتي لتقليل نسبة انتشار فقر الدم لدى الأطفال.

عمل كشف دوري للأطفال لديهم فقر الدم من أجل تقليل مضاعفات فقر الدم لدى الأطفال.

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Chapter One

Introduction Justification Objectives

1-1 Introduction

Erythropoiesis is process which produces red blood cell (erythrocyte). It is stimulated by decreased oxygen, in circulation, which is detected by the kidney which secrete the hormone erythropoietin. ⁽¹⁾

This hormone stimulate proliferation and differentiation of red cell precursors, which activates increased erythropoiesis in the hemopoietic tissue, ultimately producing red blood cell (erythrocyte) in human this usually occurs with in red bone marrow. In the early fetus, erythropoiesis take place in mesodermal cells of the yolk sac by the third or fourth month, erythropoiesis moves to liver. ⁽²⁾

After seven months, erythropoiesis occurs in the bone marrow.

Increased level of physical activity can cause an increase. However, in human with certain diseases and in some animals, erythropoiesis also occurs outside bone marrow, with in the spleen or liver.

This is termed extramedulary erythropoiesis. ⁽³⁾ Key steps in red blood differentiation include condensation of red blood material, production of hemoglobin. There are six type of hemoglobin in developing human the embryonic Gower-1, Gower -11, fetal hemoglobin and normal (HbA) and HbA₂. ⁽⁴⁾

Anemia is defined as decrease concentration of hemoglobin and RBCs mass compare with age matched control.

Anemia in children defined as hemoglobin (Hb) concentration establish cut of level, the world health organization (WHO) has suggested level Hb below which said to present, this level are < 11g/dl in children aged 6 – 50 months, < 11.5 g/dl in children aged 5 – 11 years and 12g/dl in order children aged 12 – 14 years. Anemia is classified as mild degree Hb 9 – 11 g/dl, moderate 7.0 – 9.5 g/dl, sever Hb 4.0 - 7.0 g/dl.⁽⁵⁾

A common etiology classification of anemia in children in three causative group of anemia, one anemia cause by blood loss cause by trauma and gastro intestinal bleeding, second anemia cause by decreased production of RBCs include iron deficiency and lack vitamin B12 and thalassemia and number neoplasms of bone marrow, third anemia cause by increased break down of RBCs cause number of genetic condition, such as sickle cell anemia, infection malaria and certain Autoimmune disease.⁽⁶⁾

General symptoms are often vague and may include feeling tired, weakness shortness of breath .⁽⁷⁾

Anemia in children is commonly encountered by the family physician multiple cause exist, but through history and physical examination, and limited laboratory evaluation. ⁽⁸⁾

Anemia one of the most serious public health problem affecting in both developing and industrial country, it is reported than estimated 3 - 5 billion people are anemia.⁽⁹⁾

Anemia in children is particular interest since impairs their mental. ⁽¹⁰⁾, physical and social development, it cause negative effect in poor school performance. ⁽¹¹⁾

1-2 Justification

Anemia is considered a public health problem in children in world, because it affect development growth, and impaired leering process.

This study important to detection for type of anemia in children for type of anemia in children and diagnosis and treat early because it effect in childhood performance, in order to gain the attention to this important health problem

1-3 Objectives

1-3-1 General objective:

To determine the prevalence of different type of anemia in children from six to ten years in Almesiktab hospital.

1-3-2 Specific objectives:

- 1. To asses the knowledge of mother about type of anemia found in her children and how to treat anemia.
- 2. To determine the severity of the anemia in this group.
- 3. To investigate the effect of age of prevalence of anemia.
- 4. To evaluate the effect of sex on prevalence of anemia.

Chapter Two

Literature Review

2 Literature review

2-1 Anemia

Anemia is world wide public health that affect both developed and developing countries. ⁽¹²⁾

The groups most affected pregnant women, children under the age of five years and, to lesser extent, school age children .⁽¹³⁾

The who criterion (hemoglobin <11g/dl was used diagnosis anemia in children) $(^{(14)}$

Anemia is condition that result in sufficient red cell or hemoglobin in the blood hemoglobin is the iron, rich protein that oxygen in side red blood cell and give blood it's red color.

2.2 Anemia in children:

Including iron deficiency anemia from lower than normal amount of iron in blood.

- Vitamin deficiency anemia due to lower than normal amount of a certain vitamins, like B.₁₂.
- Folate (megaloblastic anemia).
- A plastic anemia which occurs when the bone marrow doesn't produce enough red blood cells.
- Hemolytic anemia, a condition in which the body destroys red blood cell.
- Abnormal from of hemoglobin leading to the premature of destruction of red blood cells.
- Sickle cell anemia, an inherited disorder characterized by abnormal crescent shape red blood cell.
- Thalassemia, an interrelated disorder in which the body produce an abnormal form of hemoglobin teaching to premature destruction of red blood cell⁽¹⁵⁾.

2.3 Etiological of anemia in children:

2.3.1 Decreased or impaired production of red blood cell due to:

- Bone marrow damage, infiltration and trophy which occur in leukemia, aplastic anemia, pure red cell aplastic and multiple myeloma.
- Decreased erythropoietin, which occurs in inflammatory process, anemia of chronic disease and hypothyroidism.
- Defect in globin synthesis.
- Vitamin S and mineral deficiency which occur in iron, vitamin B_{12} deficiency ad folic acid deficiency.
- In effective erythropoiesis, which occurs in congenital dyserthropoietic anemia.⁽¹⁶⁾

2.3.2 Increased cell red destruction:

(A) Intrinsic defect with in the red blood cell which occur in:

- Hereditary membrane defect spherocytosis and stomatocytosis.
- Hereditary enzyme defect like glucose- 6- phosphate dehydrogenase deficiency and pyruvate kinase.
- Hereditary defective globin synthesis like B. thalassemia major E disease.
- Hereditary Hemoglobin like sickle cell disease.

(B) Extra corpuscular cause:

Non immune acquired hemolytic anemia.

- Chemical e.g. toxin, venom.
- Infection e.g. parasitic infection (malaria).

(C) Extra corpuscular causes:

Immune hemolytic anemia's:

- Iso immune antibodies.
- Auto immune antibodies.

(D) Mis cell aneous:

- Anemia of liver disease.
- Methemoglobinemia.

2.3.3 Anemia due to blood loss:

Overt blood loss is important cause of anemia and through history should be taken establish weather epistaxis or bleeding from different site of the body under different cause . ⁽¹⁷⁾

2.4 Anemia classification RBCs size:

Anemia classification based on size RBCs:

Anemia is usually classified based on RBCs size as measured by the mean corpuscular volume (MCV), anemia can be microcytic (MCV) typically less than 80 UM^3 (80 FL), normocytic (80 to 100 UM^3) [80 to 100 FL] or macrocytic (greater than 100 UM^3 [100 FL]).

2.4.1 Microcytic anemia:

It most common form caused by reduce dietary in take. It is easily treatment with supplemental iron and early intervention may prevent later loss of cognitive function of children less common cause of microcytosis are thalassemia and lead poising. If the anemia is severe or un responsive to iron therapy, the patient should be evaluate for gastro internal blood loss, tests used in the evaluation of microcytic anemia include serum iron studies, lead level, and hemoglobin electrophoresis.

2.4.2 Normocytic anemia:

May be caused by chronic disease, hemolysism or bone marrow disorder, workup normocytic anemia is based on bone marrow function as determined by reticulocyte count. If the reticulocyte count is elevated, the patient should be evaluated for blood loss or hemolysis low reticulocyte count suggests aplasia or bone marrow disorders.

2.4.3 Macrocytic anemia:

Common include vitamin B_{12} and folate level, and thyroid function testing. A peripheral smear can provide additional information in patient with any morphology .⁽¹⁸⁾

2.5 Sign and symptom:

In children sign and symptoms in anemia in children are due to anemia . ⁽¹⁹⁾

It self or the disorder causing anemia most commonly children with anemia report nonspecific symptom of a feeling of weakness, fatigue, general malaise and sometime poor concentration.

They may also report dyspnea (shortness of breath), in very severe anemia, the body may compensate for lack of caring capability of the blood by increasing cardiac output.

The children may have symptoms related to this, such as palpitations angina pectoris on intermittent claudication of the legs symptoms of heart failure $.^{(20)}$

Symptom referable to the nervous system these symptoms include faintness, head ache, banging in the ears .⁽²¹⁾

Signs may divide in general and specific, the general sign pallor of mucous membranes which occurs if hemoglobin level is less than 9 - 10 g/dl skin colour is not reliable sign .⁽²²⁾

The specific signs are associated with particular types of anemia e.g. koilony chio (spoon nails) with iron deficiency, jaundice with hemolytic megaloblastic anemia's e.g. ulcers with sickle cell and other hemolytic one miss bone deformities with thalassemia major or other sever congenital hemolytic anemias ⁽²³⁾

2.6 Diagnosis of Anemia in children:

2.6.1 Physical examination:

It is important but will be urremorkable in most children with anemia finding that suggest chronic anemia include irritability, pollar, (usually not seen until hemoglobin levels are loss than 7g/dl, glossit is children with acute anemia often clinical finding including jaundice, tachypnea, splenomeyaly hemataria and congestive heart failure). ⁽²⁴⁾

2.6.2 Laboratory diagnosis:

Anemia is defined as decreased concentration of hemoglobin and RBCs mass compared with age matched control in screening situations, such as the one year checkup only a hemoglobin level is usually obtained when anemia is encountered during this screening the specimen should be upgraded to complete blood count (CBC). Physician should first look at the mean corpuscular volume (MCV), which allows placement of the anemia in to one of the standard classification of microcytic, normocytic and macrocytic.

After narrowing the differential diagnosis based on MCV, the clinical can proceed with additional diagnostic work up.⁽²⁵⁾

The next step of the anemia work-up should include a peripheral smear and measurement of reticulocyte count pathologic finding on the peripheral smear can indicate the etiology of the anemia based on red cell morphology.

The reticulocyte cout (or percentage) helps distinguish a hyproductive anemia (decreased RBC production) from destructive process (increased RBC destruction).

A low reticulocyte count may indicate bone marrow disorder or plastic crisis, while a high count generally indicate a hemolytic process or active blood loss⁽²⁶⁾ If after analysis of initial laboratory finding, the diagnosis is still unclear, other confirmatory studies may required. Test to determine if the MCV is too low include serum iron level, total iron binding capacity (T1BC) and lead level, serum ferritin is levels are the first to decrease in patient with iron deficiency and are sensitive and specific.

If hemolysis is suspected, a direct coombs test, G6PD assay, hemoglobin electrophorses, and lactate dehydrogenase (LDH), Haptoglobin and bilirubin (indirect) determination may help to confirm the diagnosis for the anemia child with an elevated MCV, the physician should test the vitamin B12, folate and thyroid. Stimulating hormone levels.

Other test for diagnostic confirmation include an RBC enzyme panel to diagnosis hereditary spherocytosis, hemoglobin isoelectric focusing to diagnose hemoglobin variants and cytogenetic studies .⁽²⁷⁾

In certain circumstances. Such as a suspected hematological malignancy, a bone marrow aspiration may be indicated.

2.7 Consequences of anemia in children:

Iron deficiency is veiled to the most important cause of anemia among children and is attributable to poor nutritional iron in take and low iron bioavailability. ⁽²⁸⁾ Other factors, including folate and vitamin deficiencies malaria infection. ⁽²⁹⁾ Hook worm infection are also associated with childhood. ⁽³⁰⁾

This is the disturbance of physical and mental development often irreversible in infants and children, of least resistance to infection, tiredness and decreased physical and intellectual lead to poor school performance. ⁽³¹⁾

2.8 Treatment of anemia in children:

Anemia treatment varies depending on the specific diagnosis. Anemia related to blood may require surgery to stop source of bleeding in the case of celiac disease, dietary modification are necessary to avoid gluten.

Treatment option, for other type of anemia vary:

- Iron deficiency, iron supplements and if necessary blood transfusion.
- Vitamin deficiency anemia B12 injection and folic Acid supplement's.
- A plastic anemia blood transfusion to boost red blood cell level.
- Anemia related to autoimmune disorders drugs that suppress the immune system.
- Anemia associated with bone marrow disease medication, chemotherapy or bone marrow transplants.
- Hemolytic anemia: spleen removal surgery, drugs that suppress the immune system.
 - Sickle cell anemia: blood transfusion folic acid supplements, bone marrow transplant or drugs.

• Thalassemia: blood transfusion, folic acid supplement.

2.8 Previous study:

Anemia is one of the major public health problem that affect the world's total population widely. ⁽³²⁾

Anemia is known to affect people belonging to all age group particularly women of child bearing age and children.

The WHO Global database on anemia 1993 - 2005, estimated the prevalence of anemia worldwide at 25% with higher percentage noted in developing country 43%. ⁽³³⁾

In absolute number anemia affect 293 million children out of which 89 million live India.

India is one of the countries with very high prevalence of nutritional anemia among children in the world. ⁽³⁴⁾

In developing countries, the prevalence of anemia among school age children is 40% and it is classified as severe public health problem. ⁽³⁵⁾

The problem is alarming in Subaran Africa countries such as Kenya 48.9%.⁽³⁶⁾ Mali 55.8%.⁽³⁷⁾ and Tanzania 79%.

Lack of awareness among the mother but of the problem coupled with their low educational status poor nutritional practices and un healthy food habits, low iron bioavall ability of diet. ⁽³⁸⁾

Malaria and parasite infection and additional factor associated with lower.

Mouneke vueta (2011): Determined the prevalence, etiology and outcome of severe anemia in children aged 6 to 11 years in Abakaliki in Nigeria. Out 1450 children, 140 children had sever anemia, the finding of this study show that the prevalence rate of severe anemia was 9.7% malaria was commonest cause of severe anemia 64% other common cause include sickle cell anemia 9 - 3%, septicemia 13.6%, and malnutrition 7.1%.⁽³⁹⁾

Gur E et al (2005): `Determined the prevalence of old anemia out risk factor associated with anemia among primary school children in Istanbul.

1581 student between 6 to 10 years (52.1%, 798 student) were male and (49.7% - 733 student) were female, from 14 primary school located in seven different region in Istanbul a complete blood count was done by automatic cell count measure prevalence of anemia.

Kappor Gneglas et al (1992): Found 35% anemia prevalence among 296 children 7 to 10 years old in Nopur (India).

Of which 69.2% had mild anemia. 30.8% moderate anemia and non children had sever anemia, they found significant association between anemia and the socioeconomic state of children, their parent, literacy

A.A Adish et al (1999): The result revealed that of children from 7 - 10 years old in northern Ethiopia, reported that prevalence of iron deficiency (ID) and iron deficiency anemia (IDA) was high 2 - 4 year old and prevalence rate is decreased as age increased.

The prevalence of anemia, iron deficiency and IDA in 997 raral children was 24.9%, 8.5% and 4 respectively.

The study also found iron deficiency and IDA, with out a relation were higher among boys than in girls.

There was no relationship between iron deficiency or IDA . And some variable such as birth rate mother education, the majority of anemia cases in this study were normocytic.

Hassen. F et al (2004): Reported that prevalence of anemia among 265 children between the group 7 to 11 years old of Bangladesh was 70.57% of which 30.57% mildly anemia and 12.83% severely anemia.

The prevalence of anemia increase with age and becomes maximum 78.57% in age group 11 years.

The frequency of mild anemia was displayed to maximum (38 - 46) by age group.

In this study, the category of moderate anemia with maximum (25 - 55 %) present in age group 9 years.

Osonrio MM. (2001): Determined the prevalence of anemia (35.3%) among children aged 6 – 3 years old in 6 – 10 years old in state pernambuco brazil.

The mean hemoglobin was 12 mg/dl, standard deviation was 0.915, while the hemoglobin values ranged between 8.9 g/dl and 15.2 g/dl.

This study also indicates that anemia slightly higher in girls 36.3% than boy (34%)m but no significant difference was found.

Cornet M, et al (1998); Conducted a research to assess the prevalence of anemia in school children with in ages of 5 to 10 years in southern Cameroon. Relevant history, complete physical ecamination, hemoglobin estimation and peripheral smear were done for two hundred school children. The result revealed that 56.5% of children had anemia.

A significantly higher number of girls were anemia all age (66.6%) and more girls were anemic (36.5%) At most all ages significantly more (65.2%) vegetarian children were anemic.

Hemoglobin showed arising trend with improved socio-economic group were anemic.

The prevalence of anemia was high (66.8%). In under nourished children, 20.29% were anemic in the well-nourished group. Clinical pallor was detected in 42% of total children while 56.5% were anemic as per hemoglobin estimation.

The commonest blood picture was microcytic hypochromic seen 54.86% followed by normocytic normochromic 42.47% and dimorphic picture was seen in 2.45% only the most common cause could nutritional 48.67% followed by different worm infestation in 17.69% only. ⁽⁴⁰⁾

Global anemia prevalence when examined for each physiological group using WHO global data on anemia report that most affected group school age between 6 - 11 years 33%. ⁽⁴¹⁾ in Asia prevalence of nutritional anemia is particularly

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high in countries such as Bangladesh (74-80%) Indonesia (37-73%) and India (34-69%). $^{(42)}$

It has been known to be important problem on most tropical countries. WHO global data know, that anemia duet iron deficiency affect approximately 30% of the word's population about 37% of school children from 6 - 11 years old.

Some studies conducted in Africa countries have show high prevalence of anemia also exists at school age from 6 - 10 years old, in single the prevalence of anemia in children was 30.7%.⁽⁴³⁾

In Nigeria, this proportion was 82.6% among children aged 6 to 10 years. ⁽⁴⁴⁾ Other studies conducted in other countries in children from 6 to 10 years in Mozambique (54%) and china (41%). ⁽⁴⁵⁾

Chapter Three

Materials and Method

3 Material and method

3.1 Study design:

A descriptive analytical, cross sectional study design was used to study type of anemia in children from 6 to 10 years old.

3.2 Study area:

This study was conducted in Almasktab hospital.

Almasktab village sitting north Shendi town.

3.3 Study population:

A total Anemic of 100 children aged between 6 - 10 years present in Almasktab hospital.

3.4 Inclusion criteria:

-Children age more than 10 years old .

-Children HB more than 15 gldl .

3.5 Exclusion criteria:

Children with Hb less than 10.0 g/dl but age more than 10 years old or less than 6 years old, Adult people.

3.6 Ethical consideration:

Children parent, were informed in their simple language about research and it's benefit, method of sample collection, and the consent was taken, the data was kept in highly security mode.

Also the hospital and ministration was informed.

3.7 Data collection:

EDTA anticoagulant blood sample (2.5 ml blood) was used to evaluate (CBC) complete blood count.

2.5 ml blood in plain container to obtain serum to measure serum iron. Thin blood film.

Questionnaire to mother children data collected on, (gender, age, skin color, prior history of anemia diagnosed by physician, in last and use therapy, dietary patients animal or plant).

3.8 Data analysis:

Data analysis was performed using statistical package for social sciences (SPSS).

Analyze categorical variable for the difference, while test was used for continuous variable.

All P. values presented are two tailed are considered statistically significant if P was less than 0.05.

Used description statistics (mean, frequencies, cross tabulation) were done to describe to study participant and determine the prevalence of anemia.

3.9 Data presentation:

Data will presented by tables and figures.

3.10 Laboratory procedure:

- 3.10.1 Method of sample collection.
- 3.10.1.1 Requirement:
 - 1. Plain container.
 - 2. Alcohol 70% ethanol.
 - 3. Cotton.
 - 4. Syringe.
 - 5. Tourniquet.
 - 6. K2 EDTA containers.

3.10.1.2 Procedure:

- 1. Patient was sat down right on an examination table.
- 2. The arm was posltioned on the arm rest so that the vein identified.
- 3. The skin was deaned 70% ethanol and allow to dry.
- 4. Tourniquet was applied to the arm.
- 5. 5ml of blood samples were taken from Superficial vein, 2. 5 put in container of K2 EDTA anti coagulant for CBC and 2.5ml of Blood in plain container to obtain serum for measurement serum iron.

3.10.2 Complete blood count:

CBC test is penal the measure (RBC) red blood cell, white blood cell (WBC), and platelets (PLT) for diagnosis of anemia, CBC provides critical information on size, volume, and shape of red blood cell, CBC result includes measurement of hemoglobin, hematocrit (HCT), mean corpuscular volume (MCV), and hemoglobin content.

These measurement are provide by any of the common automated counter. ⁽⁴⁶⁾ In addition to MCV, MCH, and MCHC, automated counter provide and index of distribution of red blood cell volumes termed red cell distribution (RDW). ⁽⁴⁷⁾ RDW is useful in early classification of anemia because it becomes abnormal in iron deficiency anemia (IDA) (high RDW) and normal RDW in thalassemia.⁽⁴⁸⁾ Through using three preliminary hydraulic system for WBCs, RBCs and platelet and hemoglobin, and display the mode of blood cell count result on (liquid crystal displayer) (LCD) with histogram and printed result in thermal paper.⁽⁴⁹⁾

3.10.2.1 Principle of sysmex 21 hematological analyzer:

Measurement of blood cell (RBCs, WBCs and platelet) and hemoglobin concentration obtained by aspiration of small volume of well mixed (K^2 EDTA) blood by sample probe and mixed with isotonic diluent in nublazer.

Diluent aspiration delivered to RBCs aperture both for providing information about RBCs and platelet.

Other protion of aspirated sample induced in to WBCs bath in which hemolytic reagent (stomatolyzer) added to break down RBCs and release of hemoglobin which measured in build colorimeter based on cyanomet hemoglobin method (HICN). The through three sensing aperture for each cell type, cell counted and size information generated in triplicate pulse acting to electronic conductivity mentioned pulses converts in to digital number using in build calculator programmed and designed for RBCs and WBCs counts.

Some portion of diluted sample delivered to in build hemoglobin meter at the sometime, hence three values directly measured (RBCs, TWBCs – Hb) and displayed on (LCD) other values red cell indices, leukocyte differential and

absolute count calculated from given information, the result printed out aced to the setting mode.

On the other hand platelet count and histogram determined from pulses acting to the platelet. ⁽⁵⁰⁾

Reagents and material provided by sysmex manufacture and contain.

- 1- Sample: well mixed K²EDTA blood.
- 2- Cell bank.
- 3- Stromatolyzer.
- 4- Detergent.
- 5- Cell cleaner.

3.10.2.2 Procedure of sysmex 21:

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

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

3- Whole blood mode was selected.

4- Sample number and patient name entered.

5- Sample was mixed sufficiently.

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

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

8- After that automatic analysis was done and the result was displayed in the screen. ⁽⁵¹⁾

3.10.3 Estimation of serum iron:

(colorimetric method)

3.10.3.1 Principle of serum iron:

Estimation of serum iron was performed using manual method (iron. FERROZINE) . $^{\rm (52)}$

Based on the fact that transferring bound ferric ion in sample and released by guanidinium and reduced to ferrous by means of an ascorbic acid. Ferrous iron

react with ferrozine forming a ferrozine forming a coloured complex that can be measured by colorimeter.

3.10.3.2 The procedure of estimation of serum iron:

Three test tube were labeled (sample. Sample blank. Standard).

1.0ml from working reagent was placed in tube labeled with the sample and standard.

1.0ml from reagent (A) was placed in the tube labeled with sample blank.

200ul from the sample was added to the tube labeled with sample and sample blank 200 μ l from standard was added to the tube labeled with the standard.

All the tubes were mixed end 1et stand for 5 minutes at room temperature.

Absorbance (A) for all tubes was read at 560nm against distilled water.

Concentration of serum iron in sample = blank

Absorbance of sample – Absorbance of sample blank A of standard blank

The reference value of serum iron in children: 50 to 120 µg/dl

- All samples with MCV less than 80 femtollter were processed for serum iron evaluation. ⁽⁵³⁾
- Samples with hemoglobin less than 11 g/dl, MCV below 80 μ m³ (<L), and serum iron less than 50 μ g/dl were considered iron deficiency anemia.
- Sample with MCV below 8 μ m³ (FL), serum iron below 50 μ g/dl, and hemoglobin with in normal value were considered iron.

3.10.4 Thin blood film:

3.10.4.1 Preparation of thin blood film:

- Equipment.
- Slides.
- Spreders.
- Blood sample.

- Procedure.

A small drop of blood was placed in the centre live of a slide about ICM from one end. Then, without delay, the spreader was placed in front of the drop at an angle of about 30° to the slide and was moved back to make contact with drop. The drop of blood spread quickly along the line of contact. With steady movement of the hand, the drop of blood was spread along the slide the spreader did not lift off until the last trace of blood was spread out. With a correctly sized drop the film was dried by air then stained.

3.10.4.2 Staining of thin blood film:

Equipment's:

- thin blood film.
- Staining rach.
- Leish man, stain.
- Buffer (tap water).

Procedure:

The film was placed on the staining rack the flooded by the stain for two minutes. Then the buffer was applied for additional eight minutes. After that the slide was washed well by the buffer and let to dry by air. The film was examined under the microscope .⁽⁵⁴⁾

Chapter Four

Results

4 Results

In this descriptive prospective cross sectional study a total of handred children with age group from (6–10years) old, was included to determine type of anemia. In table (4-1) the anemic children with age from (6–8 years) represented (70 %) ,while those with age from (9–10 years) was (30 %) .

According to the Figure (4-1) the prevalence of anemia in male was (46%), while in female in was (54%).

Also according to table (4-2) children with family history of anemia with frequency (16%), while children that have not family history was (84%) from anemic children.

Regarding complication of anemia about (60%) of children have loss of weight, while (13%) suffering of loss of weight and numbress, while (11%) of those have numbress only, and (10%) of children do not have any complication of anemia, as described in figure (4-2).

Also regarding the type of nutritional of children (16 %) depend on animal source, while (13%) of children were depended on plant source, and (53%) of those were depend both animal and plant source, as demonstrated in table (4-3). According to table (4-4), (5%) of anemic children with chronic disease, while (95 %) do not have any chronic disease.

Also according to the table (4-5) which describe sign and symptom of anemia in children, (1%) of children those have smooth tongue, while (22%) show pallor with smooth tongue, while (1%)from children have smooth tongue with change in finger, while (34%) from children with pallor only, and (42%) of anaemic children do not have any sign and symptom.

Also according to table (4 - 6) the children that transfused with blood was (3%), while those don't transfuse was (97 %).

Figure (4--4) determined the severity of anemia according WHO cut – off of hemoglobin, about (30%) of anemic children have mild anemia, while (60%) those had moderate anemia, and (10%) from children had severe anemia.

Figure (4--5) showed type of anemia according to RBCs morphology, Normocytic normochromic was represented (75%) of children, while microcytic hypochromic was represented (25%) of anaemic children.

According to table (4 -7) the measurement of serum iron in iron deficiency anemia gives low results in (32%) of male and (68%) of female .

4 Results

Table(41): Distribution	of age.
------------------	-----------------	---------

Age	Frequency	Percent
6-8 years	70	70.0%
9-10 years	30	30.0%
Total	100	100%

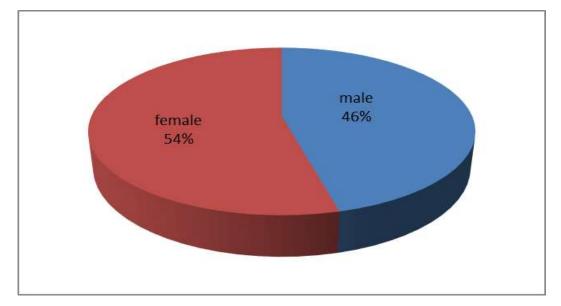


Figure (4-1): Gender distribution of anemic children .

	Frequency	Percent
Yes	16	16.0%
No	84	84.0%
Total	100	100%

Table (4-2)Family history of anemia.

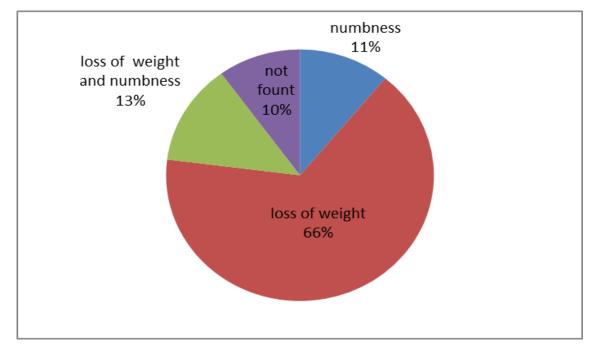


Figure (4-2): Complication of anemia in children

Type of	Animal9s	Vegetable	Together(Animals	Total
nutritional	source	source	- Vegetable)	
Percentage	16 (16.0%)	31 (31.0%)	53 (53.0%)	100(100%)

 Table (4-3): Type of nutritional of child

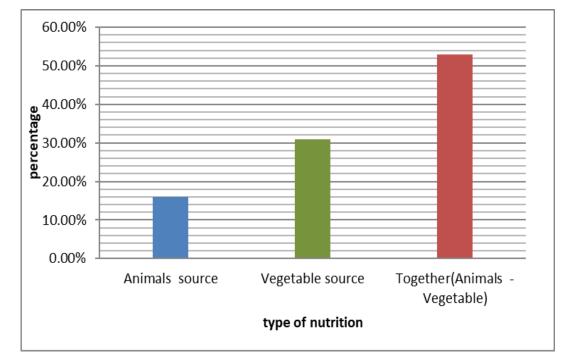


Figure (4-3): Type of nutritional of child

 Table (4-4) Chronic disease in children

	Frequency	Percent
Yes	5	5.0%
No	95	95.0%
Total	100	100%

 Table (4-5) Sign and symptom of anemia of children.

	smooth	pillar	Pillar	finger	not found	
sign and	tongue		and	clubbing		
U			smooth	and		Total
symptom			tongue	smooth		
				tongue		
Percentage	1(1.0%)	32(32.0%)	24(24.0%)	1(1.0%)	42(42.0%)	100(100%)

	Frequency	Percent
Yes	3	3.0%
No	97	97.0%
Total	100	100%

 Table (4-6) Past history of blood transfusion in children.

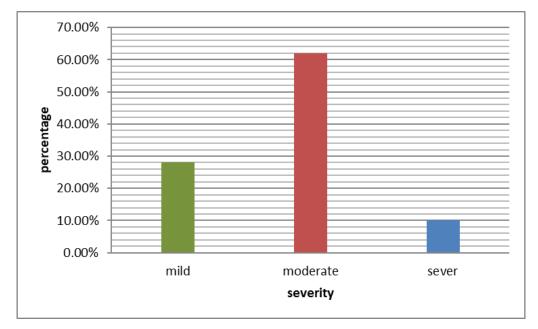
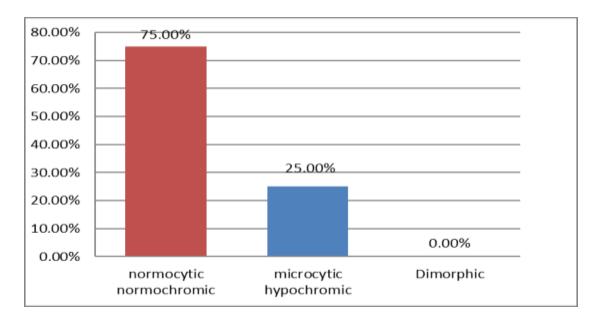


Figure (4- 4) Severity of anemia in children.



Figure(4-5) Type of anemia in children.

Table (4-7)	Discription	of iron detected	l acorrdig to gender .
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Sex	No. of children	Percentage
Male	8	32%
Female	17	68%

Chapter Five

Discussion Conclusion Recommendations

5-1 Discussion

The present study was conducted to determine type and severity of anemia and influence of various factor like sex, diet and family history of anemia. This prospective study conducted from April to August 2018, to determine the prevalence and type of anemia in children with age of (6 - 10 years).

In this study the prevalence of anemia in female was (54%) and (46%) in male, which show high prevalence of anemia in female than male. This results was similar to observation of study which conducted by kapoor and Anejce in India which show that the prevalence of anemia in female was (52%) and male was (48%). ⁽⁵⁵⁾. Also was similar to the study done In vellore district of tamil nadu Rajarath man, et al in India, which showed that the prevalence of anemia was (40%) in male, (60%) in female. ⁽⁵⁶⁾

Also the results of this study revealed that age the prevalence of anaemia in age from (6 - 8 years) was (70%), while in age group from (9 - 10 years) was (30%).We observed that there was decrease of percentage of anemia with increases age, this may be due increase nutritional substance intake with increasing of age. This results were similar to reported study done on children of south west Ethipia, in which prevalence of anemia in age of (5-8 years) was (68%) and in age of (8-10 years) was (32%).

The type of nutritional of children (16%) depend on animal source, (31%) depend on plant source, (53%) depend on plant and animal source, most of severe anemia occur in those children that depend on plant source in their nutrition, because it poor iron source.

Anemia was define according to world health organization cut. Off as Hb level < 11 g/dl under 15 years age, mild anemia was defined as hemoglobin level 10 – 10.9, and moderate defined as hemoglobin as 7 – 9.9 g/dl, and severe anemia defined as hemoglobin < 7 g/dl.⁽⁵⁸⁾

Regarding to the severity of anemia, the present study revealed that (30%) from children had mild anemia, while (60%) have moderate anemia, and (10%) from children had severe anemia.

Regarding to the types of anaemia according to the RBCs morphology, the present study showed that normocytic normochromic anaemia was found in (75%) from anemic children, this may be due to early iron deficiency when by there is still sufficient concentration of normal RBCs in circulation, other explain involves genetic adaptation documents inhibition of high altitude. Since the 1970 several report describe relatively low He concentration among Tibetan the question of genetic influence on the physiology of oxygen transport.⁽⁵⁹⁾

Other explain the bone marrow suppression in malarial infection it cause by high parasitemia or is reflect to another mechanism such as immune mediated red blood cell (RBCs) destruction. ⁽⁶⁰⁾

Microcytic hypochromic anaemia was (25%) of anemic children, in this condition iron deficiency anemia, the main reason of this type of anemia may be nutritional associated with irregular dietary habit.

In iron deficiency anemia measured of serum iron revealed that (32%) of male, and (68%) of female had low serum iron less than normal many 50 µg/dl.

5.2 Conclusion

By the end of this study we conclude that:

- Anemia was found in (70 %) of children in age of (6—8 years), while in age group of (8—10 years) was (30%).
- High prevalence of anemia in female than male.
- Majority type of anemia was normocytic normochromic anemia (75%), while microcytic hypochromic was (25 %).
- Measured serum iron was showed low than normal in (32 %) from male and (68%) from female.

5-3 Recommendations

- Health education for mothers about the importance of nutrition of children to prevent the occurrence of anemia.
- Further studies should be conducted with increasing sample size and study area to obtain accurate results.
- Make screening for children to reduce consequence of anemia in children.



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Appendix

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