

## Relations between Iron Deficiency Anemia and Anemia from Hookworms Parasites

Eman Hussein Jafer, Jamal A. AL Jabbar Attawi and Taghreed khudhur Mohammad  
Medical Technology Institute in AL-Mansour, Middle Technical University, Baghdad, Iraq.

E-mail: [rusul.aljabbar@yahoo.com](mailto:rusul.aljabbar@yahoo.com)

Accepted: 18/5/2015

### Summary

This research aimed at identifying the relationship of iron-deficiency anemia caused by insufficient dietary intake and the iron-deficiency anemia caused by parasitic worms such as hookworms. Whole blood was drawn from 40 specimens; 20 males and 20 females, normal healthy controls with age ranges 8-50 year. Blood samples were collected from 80 patients with symptoms of anemia, with age range from 10-50 year. After fecal examination, they were divided into two groups: Group one, iron-deficiency anemia with non-parasitic; and group two, iron-deficiency anemia with parasitic. Blood samples were divided into two container, one for the hemoglobin, other for serum ferritin and elements of iron, zinc in tubes without anticoagulants. Results demonstrated a significant decrease in the levels of serum iron, serum ferritin, and hemoglobin in male and female patients (in group one without parasitic worms) as compared with control groups. Furthermore (in two groups with and without parasitic worms), a significant decrease in the level of serum zinc in male and female patients as compared with control group. A significant decrease in the levels of serum iron, ferritin and hemoglobin were observed in male and female patients with iron-deficiency anemia caused by parasitic hookworms compared with control group. The worm burden was classified as light, moderate and heavy as estimated by egg counts per gram of faeces, so results showed the median increase with developing of iron deficiency anemia from parasitic hookworms.

**Keywords:** Iron-deficiency anemia, Hemoglobin, Parasitic hookworms, Ferritin, iron, zinc.

### Introduction

Iron-deficiency anemia (IDA) is a common anemia, low red blood cells (RBCs) or hemoglobin levels (Hb) caused by insufficient dietary intake and absorption of iron, and/or iron loss from bleeding which can originate from a range of sources such as the intestinal, uterine or urinary tract (1). Anemia can result when the body does not make enough red blood cells and bleeding causes loss of red blood cells more quickly than they can be replaced. The most significant cause of iron-deficiency anemia in developing world patients is parasitic hookworms, worms cause intestinal bleeding, which is not always noticeable in faeces (2). Iron-deficiency anemia is characterized by the sign of pallor (reduced oxyhemoglobin in skin or mucous membranes), and the symptoms of fatigue, lightheadedness, and weakness (3). A diagnosis of iron-deficiency anemia then requires further investigation as to its cause. It can be caused by increased iron demand/loss or decreased iron intake (4). Furthermore, interactions of different trace elements with iron determine the relationship between

changes in trace element status in the organism and development of IDA (5). Increases in content of antagonistic to iron trace elements such as zinc and calcium, which impair iron absorption or its physiological impact, can lead to development of IDA (6).

Parasitosis: There are no specific symptoms or signs of hookworm infection, they give rise to a combination of intestinal inflammation and progressive iron/ protein-deficiency anemia. Larval invasion of the skin might give rise to intense, local itching, usually on the foot or lower leg, which can be followed by lesions that look like insect bites, can blister (ground itch) and last for a week or more (7). Additionally, cough and pneumonitis may result as the larvae begin to break into the alveoli and travel up the trachea, then once the larvae reach the small intestine of the host (8). Major morbidity associated with hookworm is caused by intestinal blood loss, iron deficiency anemia, and malnutrition (9). The results from adult hookworms in the small intestine ingesting blood, rupturing erythrocytes, and degrading hemoglobin in the host (10). A lack of iron in the diet and an inability to absorb

iron: The body normally gets the iron it requires from foods, iron that is poorly absorbed (non-heme iron), they can become iron deficient over time, iron from food is absorbed into the bloodstream in the small intestine, especially the duodenum and proximal ileum. Many intestinal disorders can reduce the body's ability to absorb iron. In cases where there has been a reduction in surface area of the bowel, such as in celiac disease, inflammatory bowel disease or post-surgical resection the body can absorb iron, but there is simply insufficient surface area (11).

Blood loss: Blood contains iron within red blood cells (RBCs), so blood loss leads to a loss of iron; there are several common causes of blood loss. Women with menorrhagia (heavy periods) are at risk of iron-deficiency anemia because they are at higher-than-normal risk of losing a larger amount blood during menstruation than is replaced in their diet. Chronic blood loss within the body, such as from a colon polyp or gastrointestinal cancer can cause iron-deficiency anemia (12 and 13).

### Materials and Methods

Whole blood was drawn from 40 specimens (20 male and 20 female) normal healthy controls with age ranges 8-50 year. Another blood samples were collected from 80 patients with symptoms of iron-deficiency anemia from East-Baghdad, these samples were divided into the following diagnostic categories: Group one: 40 patients 19 males and 21 females, age ranged from 10-50 year with iron-deficiency anemia from non-parasitic worms, characterized by symptoms with iron-deficiency anemia like constipation, tinnitus, sleepiness and hair loss. Group two: 40 patients 19 males and 21 females, age ranged from 10-50 year with iron-deficiency anemia from parasitic worms, characterized by estimating the egg counts per gram of faeces, and symptoms like constipation, breathlessness, and very pale yellow skin. During the period between November 2013 to November 2014, the blood samples were transported to the Al yarmouk teaching hospital. These samples were divided into two container, one for the hemoglobin, other for serum ferritin and elements iron, zinc in tubes without

anticoagulants. First one (Hb) of whole blood samples were measured with an automatic cell counter (Beckman Coulter). Serum was separated from the second blood samples by centrifuge at 3000 r.p.m, for 15 min and the resulting sera were placed into tubes and were used for determination of iron, zinc, and ferritin. For the analysis of elements in blood were measured with absorption spectrophotometer. The method of ferritin was measured by ELISA (14). Faecal samples were collected to detect parasites by using formalin technique, followed by iodine staining and microscopy examination the presence of hookworms. Technique was employed to determine the intensity of hookworm infections, as estimated by egg counts per gram of faeces (15).

Data expressed as means  $\pm$ SD. Student's t-test is used to evaluate differences between the groups. For all tests ( $P \leq 0.05$ ) is considered statistically significant, the intensity of hookworms infections was quantitatively estimated as ova per gram of faeces and was categorized into three main categories: light, moderate and heavy infections. The distribution of egg counts were not normally distributed, hence presented as median.

### Results and Discussion

A sufficiently low hemoglobin (Hb) by definition makes the diagnosis of anemia, if the anemia is due to iron deficiency, one of the first abnormal values to be noted on a complete blood count (CBC), as the body's iron stores begin to be depleted (16). The blood smear of a patient with iron deficiency shows many hypochromic (pale and relatively colorless) and rather small RBCs (17). The most common cause of iron deficiency anemia is chronic gastrointestinal bleeding from non-parasitic causes, such as gastric ulcers, duodenal ulcers or gastrointestinal cancer (18).

In group one the results showed a significant decrease in measuring the level of hemoglobin in male patients ( $125 \pm 3.22$ ) g/L and in female patients ( $118 \pm 3.66$ ) g/L compared to control groups as in (Tables, 1 and 2). There are close relations between the metabolism of different trace elements including iron based on antagonistic or synergistic interactions, low concentration of

zinc may be considered as contributing factors for IDA due to known synergistic interactions of both trace elements with iron-participation of zinc in hemoglobin synthesis and its essentiality in erythropoiesis (19). One of known links is at the level of common intestinal transporters for iron and other divalent metals, iron deficiency predisposes to metabolic imbalances and respective changes in trace element status (20). These changes in zinc status are frequently explained by coexisting deficiencies of iron and zinc due to common dietary sources of both micronutrients and decreasing their intestinal absorption by the same dietary factors (21). Results showed a significant decrease in the level of serum iron in males (11.6±1.52) Umol/L and in females (11.5±1.83) Umol/L

compared to control groups as in (Tables, 1 and 2). A significant decrease in the level of serum zinc in males (11.23±4.41) Umol/L and in females (11.25 ± 1.75) Umol/L was observed compared to control groups as in (Tables, 1 and 2). The remainder of iron is stored for later use in all cells, but mostly in the bone marrow, liver, and spleen. These stores are called ferritin complexes, a low serum ferritin is the most sensitive lab test for iron deficiency anemia, however serum ferritin can be elevated by any type of chronic inflammation (22). Reduced level of ferritin (27.31±2.11) ng/ml in males and (25.13 ±2.28) ng/ml in females as in (Tables, 1 and 2) showed a significant decrease in serum ferritin in male and female patients compared to control groups.

**Table, 1: Some parameters for iron deficiency anemia without parasitic worms and control groups in males.**

Males	IDA	Controls	P value
	Mean±SD	Mean±SD	
Hemoglobin (g/L)	125±3.22	145±7.08	0.024*
Serum ferritin (ng/ml)	27.31±2.11	37.75±3.07	0.032*
Serum iron (Umol/L)	11.6±1.52	28.71±2.55	0.040*
Serum zinc (Umol/L)	11.23±4.41	16.96±1.06	0.036*

\*Significant using t-test for two independent means at 0.05 level of significance

**Table, 2: Some parameters for iron deficiency anemia without parasitic worms and control groups in females.**

Females	IDA	Controls	P value
	Mean±SD	Mean±SD	
Hemoglobin (g/L)	118±3.66	135± 6.70	0.023*
Serum ferritin (ng/ml)	25.13±2.28	35.26±3.05	0.025*
Serum iron (Umol/L)	11.5±1.83	28.33±2.25	0.037*
Serum zinc (Umol/L)	11.25±1.75	16.36±1.08	0.012*

\*Significant using t-test for two independent means at 0.05 level of significance

Group two, adult hookworms tend to inhabit the upper small intestine. Bleeding due to hookworm infection occurs in the upper small intestine and some of the constituents from the blood are reabsorbed further down in the gastrointestinal tract. Therefore, it is possible that the reabsorption of iron may be impaired and causes dysentery and chronic faecal blood loss (23). Reduced level of hemoglobin (120±2.22) g/L in males and (115±3.01) g/L in females as in (Tables, 3 and 4), showed a significant decrease in (Hb) levels in male and female patients compared to control groups. Also a significant decrease in

the level of serum iron in males (10.9±1.34) Umol/L and in females (10.5±1.26) Umol/L was observed compared to control groups as in (Tables, 3 and 4). Reduced level of ferritin (22.33±3.51) ng/ml in males and (21.41 ±3.56) ng/ml in females as in (Tables, 3 and 4) showed a significant decrease in serum ferritin in male and female patients compared to control groups. A significant decrease in the level of serum zinc in males (11.20 ±4.31) Umol/L and in females (11.21±1.71) Umol/L was observed compared to control groups as in (Tables, 3 and 4).

**Table, 3: Some parameters for iron deficiency anemia with parasitic worms and control groups in males.**

Males	IDA	Controls	P value
	Mean±SD	Mean±SD	
Hemoglobin (g/L)	120±2.22	145±7.08	0.021*
Serum ferritin (ng/ml)	22.33 ±3.51	37.75±3.07	0.030*
Serum iron (Umol/L)	10.9±1.34	28.71±2.55	0.041*
Serum zinc (Umol/L)	11.20±4.31	16.96±1.06	0.024*

\*Significant using t-test for two independent means at 0.05 of significance

**Table, 4: Some parameters for iron deficiency anemia with parasitic worms and control groups in females**

Females	IDA	Controls	P value
	Mean±SD	Mean±SD	
Hemoglobin (g/L)	115±3.01	135± 6.70	0.045*
Serum ferritin (ng/ml)	21.41 ±3.56	35.26±3.05	0.039*
Serum iron (Umol/L)	10.5±1.26	28.33±2.25	0.011*
Serum zinc (Umol/L)	11.21±1.71	16.36±1.08	0.014*

\*Significant using t-test for independent means at 0.05 of significance

The worm burden was classified as light, moderate and heavy as estimated by egg counts per gram of faeces, so results showed the range (4-18 EPG), Eggs Per Gram, (EPG) and increase egg counts per gram of faeces as worm burden was classified as light (6) EPG, moderate (12) EPG, and heavy (16) EPG, respectively, and the median increase with developing of iron deficiency anemia from parasitic hookworms as in (Table, 5). The relations between iron-deficiency anemia and anemia from hookworms parasites were decrease in the levels of serum iron, ferritin, zinc and hemoglobin in male and female patients compared with control groups, and there was a decrease in the levels of serum iron, ferritin, zinc and hemoglobin in male and female patients in group two with parasitic worms more than in group one without parasitic worms as in (Tables, 1 and 3) for male patients and (Tables, 2 and 4) for female patients.

**Table, 5: Intensity of hookworm infections.**

Intensity of infections	No	%	Median/EPG
Light	42	52.5	6
Moderate	36	45	12
Heavy	2	2.5	16

No, Number examined. EPG, Eggs per Gram.

The diagnosis of iron deficiency anemia will be suggested by appropriate history (e.g. anemia in a menstruating woman or an athlete engaged in long-distance running), the

presence of occult blood (i.e. hidden blood) in the stool. For example known celiac disease can cause malabsorption of iron (24). A travel history to areas in which hookworms are endemic may be helpful in guiding certain stool tests for parasites or their eggs.

The present study is useful to know that the relation between iron deficiency anemia and anemia from hookworms parasites decrease in the levels of serum iron, ferritin, zinc and hemoglobin in two groups with and without parasitic worms. In conclusion, high prevalence of iron deficiency anemia in individuals was observed in rural and remote areas, some were daily wage earners working in rubber or unskilled laborers in factories or construction sites.

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## العلاقة بين فقر الدم الناجم عن نقص الحديد وفقر الدم الناجم عن الطفيليات الكلابية

أيمان حسين جعفر و جمال عبد الجبار عطاوي و تغريد خضر محمد

المعهد الطبي التقني المنصور، بغداد، العراق.

E-mail: [rusul.aljabbar@yahoo.com](mailto:rusul.aljabbar@yahoo.com)

### الخلاصة

هدفت الدراسة إلى معرفة العلاقة بين فقر الدم الناجم عن نقص الحديد بسبب نقص الغذاء اليومي وفقر الدم الناجم عن نقص الحديد الذي تسببه الطفيليات الكلابية. أخذت 20 عينة دم من الرجال الأصحاء و20 عينة دم من النساء الأصحاء كمجموعة تحكم سوية وبعمر 8-50 سنة. كما أخذت 80 عينة دم من الأشخاص الذين ظهرت عليهم أعراض فقر الدم وبعد فحص البراز وحسب وجود البيوض قسمت إلى مجموعتين: الأولى فقر الدم الناجم عن نقص الحديد من أمراض الديدان الطفيلية. أظهرت النتائج (في المجموعة الأولى) انخفاضاً معنوياً في مستويات الحديد، الفيريتين والهيموغلوبين (نقص كريات الدم الحمراء) في مرضى الرجال والنساء مقارنة مع مجموعة التحكم فضلاً عن (وجود الطفيليات أو لا في مجموعتي المرضى) انخفاض معنوي في مستوى الزنك في مرضى النساء والرجال مقارنة مع مجموعة التحكم. كذلك انخفاض معنوي في مستويات الحديد والفيريتين والهيموغلوبين في مرضى الرجال والنساء لمرض فقر الدم الناجم عن نقص الحديد والذي تسببه الديدان الطفيلية الكلابية (في المجموعة الثانية) مقارنة مع مجموعة التحكم. قسمت عينات البراز إلى خفيفة ومتوسطة وثقيلة، وهي قياس كمية البويضات في كل غرام من البراز. نستنتج من الدراسة زيادة متوسط كمية البويضات مع نمو فقر الدم الناجم عن نقص الحديد الذي تسببه أمراض الديدان الطفيلية.

**الكلمات المفتاحية:** فقر الدم الناجم عن نقص الحديد، خضاب الدم، الطفيليات الكلابية، فريتين، حديد، زنك.