Various Investigations Which Help to Differentiate Between Iron Deficiency And Iron Deficiency Anaemia In Patients With Hookworm Infection Diagnosed by Doing Endoscopy

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Abstract:
**Objective:** Anaemia is commonly reported to occur in hookworm infection. Hence a detailed study was done to know about the various investigations which help to differentiate between iron deficiency and iron deficiency anaemia in patients with hookworm infection diagnosed by doing upper gastro-intestinal endoscopy.

**Methods:** A study of 1307 patients who had undergone upper gastro-intestinal endoscopy for a period of 5 years and one month from May 2009 to May 2014 was carried out. In all the patients found to have hookworms in duodenum, haemoglobin estimation was done to know about the presence or absence of anaemia. But in addition to haemoglobin estimation, serum ferritin and transferrin saturation are the most important investigations which help to differentiate between iron deficiency and iron deficiency anaemia. In all the patients with hookworm infection with normal haemoglobin without anaemia, serum ferritin and transferrin saturation could not be done. In one patient with mild anaemia, mean corpuscular volume or MCV was low. But in one patient with severe hookworm infection with multiple hookworms in duodenum, in addition to peripheral smear, mean corpuscular volume or MCV and haemoglobin estimation, serum ferritin and transferrin saturation were also done. The results were found as given below.

**Results:** Out of these 1307 patients, 14 patients found to have hookworms in duodenum were taken into consideration for our study. Out of these 14 patients, 5 patients had normal haemoglobin without anaemia. But since serum ferritin and transferrin saturation could not be done in these 5 patients, the status of iron deficiency could not be assessed in these 5 patients without anaemia. In one patient with mild anaemia (haemoglobin 11.7 g%) due to hookworm infection, mean corpuscular volume or MCV was low. But since serum ferritin and transferrin saturation could not be done in this patient, the status of iron deficiency anaemia could not be assessed in this patient. Low serum ferritin and transferrin saturation along with normal haemoglobin indicate iron deficiency without anaemia. Low serum ferritin and transferrin saturation along with low haemoglobin indicate iron deficiency anaemia. But in one patient with severe hookworm infection with multiple hookworms in duodenum, peripheral smear showed severe microcytic hypochromic anaemia, mean corpuscular volume or MCV was low, haemoglobin was low, serum ferritin and transferrin saturation were found to be very low. Hence this patient could be labelled as having iron deficiency anaemia.

**Conclusion:** Hence patients with low serum ferritin and transferrin saturation along with low haemoglobin could be labelled as having iron deficiency anaemia. But low serum ferritin and transferrin saturation along with normal haemoglobin indicate iron deficiency without anaemia.

**Keywords:** serum ferritin and transferrin saturation, haemoglobin, mean corpuscular volume or MCV, peripheral smear, iron deficiency anaemia

I. Introduction

Anaemia is commonly reported to occur in hookworm infection (1-17). Hence a detailed study was done to know about the various investigations which help to differentiate between iron deficiency and iron deficiency anaemia in patients with hookworm infection diagnosed by doing upper gastro-intestinal endoscopy.

II. Materials and Methods

This study was conducted in the department of general surgery, AarupadaiVeedu Medical College And Hospital, Puducherry. A study of 1307 patients who had undergone upper gastro-intestinal endoscopy for a period of 5 years and one month from May 2009 to May 2014 was carried out. In each of these 1307 patients, the first and second part of duodenum were carefully examined to find out the presence of hookworms. In all the patients found to have hookworms in duodenum, haemoglobin estimation was done to know about the presence or absence of anaemia. But in addition to haemoglobin estimation, serum ferritin and transferrin saturation are the most important investigations which help to differentiate between iron deficiency and iron deficiency anaemia. In all the patients with hookworm infection with normal haemoglobin without anaemia, serum ferritin...
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and transferrin saturation could not be done. In one patient with mild anaemia, mean corpuscular volume or MCV, mean corpuscular haemoglobin or MCH and mean cell haemoglobin concentration or MCHC were also done in addition to haemoglobin estimation. But serum ferritin and transferrin saturation could not be done in this patient. In one patient with severe hookworm infection with multiple hookworms in duodenum, in addition to peripheral smear, mean corpuscular volume or MCV and haemoglobin estimation, serum ferritin and transferrin saturation were also done. Anaemia is defined as haemoglobin <12g/dl or <12g% (18,19). Mild anaemia is taken as haemoglobin 10 to 12g/dl or g%, moderate anaemia is taken as haemoglobin 7 to 10g/dl or g% and severe anaemia is taken as haemoglobin <7g/dl or g% (20). The results were found as given below.

III. Results

Out of these 1307 patients, 14 patients found to have hookworms in duodenum were taken into consideration for our study. Out of these 14 patients, 5 patients had normal haemoglobin without anaemia.

1. Haemoglobin Values In All The 5 Patients Without Anaemia

5 patients had normal haemoglobin without anaemia. All the 5 patients had haemoglobin ≥12g% [haemoglobin 18g%, 17.6g%, 16g%, 14g%, 12.8g%]. But since serum ferritin and transferrin saturation could not be done in these 5 patients, the status of iron deficiency could not be assessed in these 5 patients without anaemia.

2. Detailed Investigations In The Patient With Mild Anaemia Due To Hookworm Infection.

In one patient with mild anaemia (haemoglobin 11.7g%) due to hookworm infection, mean corpuscular volume or MCV, mean corpuscular haemoglobin or MCH and mean cell haemoglobin concentration or MCHC were also done in addition to haemoglobin estimation. The results were found as given below.

a. Haemoglobin

In this patient with mild anaemia due to hookworm infection diagnosed by doing upper gastrointestinal endoscopy haemoglobin is 11.7g%.

b. Mean corpuscular volume or MCV

In this patient, mean corpuscular volume or MCV is 76.5 femolitres or fl [normal range 82-92 fl].

c. Mean corpuscular haemoglobin or MCH

Mean corpuscular haemoglobin or MCH is 27.9 picograms or pg [normal range 27-32pg] in this patient.

d. Mean corpuscular haemoglobin concentration or MCHC

Mean corpuscular haemoglobin concentration or MCHC is 36.6% [normal range 32-36%] in this patient.

In this patient with mild anaemia (haemoglobin 11.7g%) due to hookworm infection, mean corpuscular volume or MCV is low. But since serum ferritin and transferrin saturation could not be done in this patient, the status of iron deficiency anaemia could not be assessed in this patient.

3. Detailed Investigations In The Patient With Severe Hookworm Infection With Multiple Hookworms In Duodenum.

In one patient with severe hookworm infection with multiple hookworms in duodenum, in addition to peripheral smear, mean corpuscular volume or MCV and haemoglobin estimation, serum ferritin and transferrin saturation were also done. Mean corpuscular haemoglobin or MCH, mean corpuscular haemoglobin concentration or MCHC and serum iron were also done in this patient. The results were found as given below.

a. Low serum ferritin

In this patient with severe hookworm infection with multiple hookworms in duodenum, serum ferritin is very low 1.4 ng/ml [normal range 18-160 ng/ml].

b. Low transferrin saturation

In this patient with severe hookworm infection with multiple hookworms in duodenum, transferrin saturation is very low and is only 5% and its normal range is 20-50%.

c. Low serum iron
In this patient with severe hookworm infection with multiple hookworms in duodenum, serum iron is very low-20μg /dL [normal range 50 to 170μg/dL].

d.Lawhaemoglobin
In this patient with severe hookworm infection with multiple hookworms in duodenum, haemoglobin is very low-3.2g% [normal range 12 to 16g%].

e.Low mean corpuscular volume or MCV
In this patient with severe hookworm infection with multiple hookworms in duodenum, mean corpuscular volume or MCV is very low and is only 50.7 femolitres or fl [normal range 82-92 fl] indicating that the RBCs are very small with small volume and are microcytic.

f.Low mean corpuscular haemoglobin or MCH
In this patient with severe hookworm infection with multiple hookworms in duodenum, mean corpuscular haemoglobin also is very low -15 picograms or pg [normal range 27-32 pg] indicating that the RBCs are hypochromic and have low amount of haemoglobin.

g. Low mean corpuscular haemoglobin concentration or MCHC
In this patient with severe hookworm infection with multiple hookworms in duodenum, mean corpuscular haemoglobin concentration or MCHC also is low-29.9% [normal range 32-36%].

h. Peripheral smear examination
In this patient with severe hookworm infection with multiple hookworms in duodenum, peripheral smear examination showed severe microcytic hypochromic anaemia with anisopoikilocytosis.

Anisocytosis
An abnormal variation in cell volume is called anisocytosis.

Poikilocytosis
Irregularly shaped cells is called poikilocytosis.

In this patient with severe hookworm infection with multiple hookworms in duodenum, peripheral smear showed severe microcytic hypochromic anaemia, mean corpuscular volume or MCV was low, haemoglobin was low, serum ferritin and transferrin saturation were found to be very low. Hence this patient could be labelled as having iron deficiency anaemia. In this patient serum iron is also very low.

The following features increase the post-test probability of a diagnosis of iron deficiency anaemia being correct: reduced mean cell haemoglobin, hypochromia (decreased haemoglobin concentration in RBC), increased variation in cell size (Red cell width or RDW), an abnormal variation in cell volume (anisocytosis) irregularly shaped cells (poikilocytosis) and reduced mean cell haemoglobin concentration.

In this patient with severe hookworm infection with multiple hookworms in duodenum also, mean corpuscular haemoglobin is very low, mean corpuscular haemoglobin concentration also is low, peripheral smear showed an abnormal variation in cell volume (anisocytosis) and irregularly shaped cells (poikilocytosis).

IV. Discussion

1. Importance of serum ferritin and transferrin saturation in the diagnosis of iron deficiency anaemia

a. Low serum ferritin and transferrin saturation along with low haemoglobin indicate iron deficiency anaemia.

The patients having anaemia (haemoglobin<12g%) with mean corpuscular volume or MCV< 78 cuμ or fluorescent serum ferritin < 12 ng/ml, transferrin saturation < 16% and microcytic hypochromic RBCs on peripheral smear were labelled as having iron deficiency anaemia (21).

Hence our patient with severe anaemia with hemoglobin -3.2g%, mean corpuscular volume or MCV 50.7 femolitres or fl, serum ferritin 1.4 ng/ml, transferrin saturation 5% and microcytic hypochromic RBCs on peripheral smear has iron deficiency anaemia.

b. Low hemoglobin alone without the values of serum ferritin and transferrin saturation cannot be labelled as iron deficiency anaemia.

Our patient with mild anaemia with hemoglobin-11.7g%, mean corpuscular volume or MCV-76.5 femolitres or fl cannot be labelled as iron deficiency anaemia as serum ferritin, transferrin saturation and peripheral smear was not done for the patient. Hence low serum ferritin and transferrin saturation along with low haemoglobin,
low mean corpuscular volume or MCV and microcytic hypochromic anaemia in peripheral smear only indicate iron deficiency anaemia. Low hemoglobin and low mean corpuscular volume or MCV alone without the values of serum ferritin and transferrin saturation cannot be labelled as iron deficiency anaemia.

2. Differences between iron deficiency and iron deficiency anaemia

Iron deficiency has generally been defined as an absence of iron stores (very low serum ferritin) while iron deficiency anaemias defined as the absence of iron stores (very low serum ferritin) but with a haemoglobin concentration below a defined threshold.

Iron deficiency is defined as serum ferritin <13 microg/l and iron deficiency anaemia as serum ferritin <13 microg/l and haemoglobin<5th percentile in iron replete pregnant women(22). A serum ferritin below 15 – 20 micrograms/L in a person with microcytic anaemia confirms iron deficiency anaemia.

Low serum ferritin and low transferrin saturation with normal haemoglobin, normal mean corpuscular volume [MCV] and normocytic normochromic RBCs on peripheral smear indicate iron deficiency without anaemia.

Low serum ferritin and low transferrin saturation with low haemoglobin, low mean corpuscular volume [MCV] and microcytic hypochromic RBCs on peripheral smear indicate iron deficiency anaemia.

a. Iron deficiency without anaemia or iron deficiency alone or latent iron deficiency

Iron deficiency without anaemia (ID-A) is generally considered to correspond to a degree of dietary iron deficiency sufficient to deplete ferritin stores and to decrease iron concentrations in some tissues, but not sufficient to reduce hemoglobin to the point of anemia. Patients with normal haemoglobin, but with low serum ferritin were considered as suffering from latent iron deficiency (23).

b. Iron deficiency with anaemia or iron deficiency anaemia

But our patient with severe anaemia had iron deficiency sufficient to deplete ferritin stores and to decrease iron concentrations in some tissues and also had very low haemoglobin (haemoglobin<3.2g%) and hence had iron deficiency with anaemia or iron deficiency anaemia.

Serum ferritin is a surrogate marker of stored iron (23). When serum ferritin is low without low haemoglobin, it is called latent iron deficiency, whereas when there is also low hemoglobin, it is iron deficiency anaemia (23). Hence serum ferritin provides the most useful indirect estimate of a person’s iron stores in both latent iron deficiency and iron deficiency with anaemia.

3. Iron deficiency without anaemia or latent iron deficiency

The patients without anaemia and having haemoglobin ≥12gm%, MCV > 78 μm or fl, transferrin saturation <16%, ferritin <12 ng/ml and normocytic normochromic RBCs on peripheral smear were categorized as having latent iron deficiency or iron deficiency without anaemia (21).

Out of the 14 patients with hookworms in duodenum taken into consideration for our study, 5 patients did not have anaemia (haemoglobin ≥12g%). But serum ferritin, transferrin saturation, mean corpuscular volume or MCV and peripheral smear were not done for these 5 patients without anaemia. But the patients without anaemia and having haemoglobin ≥12g% can be labelled as iron deficiency without anaemia or latent iron deficiency only if serum ferritin, transferrin saturation, mean corpuscular volume or MCV and peripheral smear were done and only if serum ferritin is < 12ng/ml [normal range 18-160 ng/ml] and transferrin saturation is <16% [normal range 20-50%] and only if mean corpuscular volume or MCV is >78 μm or fl and peripheral smear showed normocytic normochromic RBCs.

Hence none of these 5 patients without anaemia (haemoglobin ≥12g%) with hookworms in duodenum taken into consideration for our study can be labelled as having iron deficiency without anaemia or latent iron deficiency since only haemoglobin was done for these 5 patients and serum ferritin, transferrin saturation, mean corpuscular volume or MCV and peripheral smear could not be done for these 5 patients without anaemia.

Hence even in patients without anaemia (haemoglobin ≥12g%) serum ferritin, transferrin saturation, mean corpuscular volume or MCV and peripheral smear should be done to diagnose iron deficiency without anaemia or latent iron deficiency.

V. Conclusion

1. The patients having anaemia (haemoglobin <12g%) with mean corpuscular volume or MCV < 78 μm or fl, serum ferritin < 12ng/ml, transferrin saturation <16% and microcytic hypochromic RBCs on peripheral smear were labelled as having iron deficiency anaemia.
2. Hence our patient with severe anaemia with low hemoglobin (3.2g%), low mean corpuscular volume or MCV (50.7 femolitres or fl), low serum ferritin [1.4 ng/ml], low transferrin saturation [5%] and microcystic hypochromic RBCs on peripheral smear has iron deficiency anaemia.

3. Low hemoglobin and low mean corpuscular volume or MCV alone without the values of serum ferritin and transferrin saturation cannot be labelled as iron deficiency anaemia. Hence our patient with mild anaemia with low hemoglobin (hemoglobin 11.7g%), low mean corpuscular volume or MCV (76.5 femolitres or fl) cannot be labelled as having iron deficiency anaemia since serum ferritin, transferrin saturation was not done for the patient.

4. The patients without anaemia and having haemoglobin ≥12g%, MCV > 78 cuμ or fl, ferritin <12ng/ml, transferrin saturation <16% and normocytic normochromic RBCs on peripheral smear were categorized as having iron deficiency without anaemia (lateral iron deficiency).

5. Hence none of the 5 patients without anaemia (haemoglobin ≥12g%) with hookworms in duodenum in our study can be labelled as having iron deficiency without anaemia (lateral iron deficiency) since only haemoglobin was done for these 5 patients and serum ferritin, transferrin saturation, mean corpuscular volume or MCV and peripheral smear could not be done for these 5 patients without anaemia.

6. Hence low serum ferritin and transferrin saturation along with low haemoglobin, low mean corpuscular volume or MCV and microcystic hypochromic anaemia in peripheral smear indicate iron deficiency anaemia.

7. But low serum ferritin and transferrin saturation along with normal haemoglobin, normal mean corpuscular volume or MCV and normocytic normochromic RBCs on peripheral smear indicate iron deficiency without anaemia (lateral iron deficiency).

8. Hence low serum ferritin and transferrin saturation occur both in iron deficiency without anaemia and in iron deficiency anaemia. Low serum ferritin and transferrin saturation along with normal haemoglobin indicate iron deficiency without anaemia. Low serum ferritin and transferrin saturation along with low haemoglobin indicate iron deficiency anaemia.

9. Hence in addition to haemoglobin estimation, serum ferritin and transferrin saturation are the most important investigations which help to differentiate between iron deficiency without anaemia (lateral iron deficiency) and iron deficiency anaemia.

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