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# Iron status of pregnant women at first antenatal booking in Mbarara University Teaching Hospital

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## SUMMARY

An assessment of iron status was made on 96 pregnant women and 29 non-pregnant, non-lactating menstruating women of comparable age group as controls. Anaemia (haemoglobin < 110 g/l) was present in 84.4% of the pregnant women and in 48.3% of the control group. Iron deficiency (serum ferritin < 12.0  $\mu$ g/l) was present in 51.1% of the pregnant group and 37.9% of the control group. Prevalence of anaemia with iron deficiency was 54.7% in anaemic pregnant women. Serum ferritin correlated significantly with low haemoglobin (P < 0.05). Median serum ferritin declined progressively until 31 weeks of gestation. Preliminary studies on their dietetics showed that low animal protein consumption and poor dietary iron bioavailability were associated with anaemia.

# INTRODUCTION

Iron deficiency is now recognized as the most common nutritional deficiency state in developing countries and the most prevalent cause of anaemia<sup>1</sup>. Iron deficiency can be without anaemia or can coexist with anaemia (iron deficiency anaemia). Iron deficiency without anaemia represents the least severe stage of iron depletion and is identified by a serum ferritin level less than 12  $\mu$ g/l<sup>2</sup>. With continued loss of iron, red cell production decreases resulting in iron-deficiency anaemia identified by haemoglobin concentration lower than normal for the person's age or sex group. It represents a later stage of the deficiency. Pregnant women are at a high risk of developing nutritional anaemias because of their increased requirements for haematopoietic nutrients to meet the needs of the growing fetus<sup>3</sup>. Increased daily requirement for iron, especially in the last trimester of pregnancy, cannot be met by diet alone but also partly from maternal reserves. Poor consumption of haem iron and promoters of nonhaem iron absorption, and frequent pregnancies, lower these reserves<sup>4</sup>.

Correspondence to: Gertrude N Kiwanuka E-mail: must@uga.healthnet.org There is little information on the extent of iron deficiency and iron deficiency anaemia in Uganda. The purpose of this study was to establish the iron status of a group of pregnant women in Mbarara, Uganda.

#### MATERIALS AND METHODS

A total of 96 pregnant women studied [mean age $\pm$ standard deviation (SD) =  $23.4\pm5$  years] reporting for first antenatal booking at the routine antenatal clinic of Mbarara University Teaching Hospital (MUTH) were studied. Gestation range was 20–32 weeks. Mean parity was  $2.5\pm2.3$ . All were not on iron supplements. Twenty-nine non-pregnant, non-lactating menstruating women (mean age $\pm$ SD= $23.6\pm5.3$  years) were studied to establish the normal values of haemoglobin concentration. Socio-economic and biodata of the subjects were recorded on questionnaires.

Informed consent was obtained from each subject before they were included in the study. Clinical examination was done on pregnant women to detect anaemia which was classified as either severe, moderate or mild. A stool sample from every subject was examined for helminthic infections. Ova were detected microscopically by the direct saline method<sup>5</sup>.

Blood (10 ml) was drawn from each subject by venipuncture. One millilitre of this was used to determine haemoglobin concentration within 5 h after blood collection. This was by the cyanmethaemoglobin method<sup>6</sup> using a photoelectric colorimeter calibrated and checked at regular intervals against a standard solution of cyanmethaemoglobin. Serum fractions were collected in sterile polystyrene tubes by centrifugation, frozen at  $-20^{\circ}$ C for subsequent serum ferritin and serum iron assays. Serum ferritin was determined in duplicate using the Ferritin ELISA method (Beasley NMR, 1995, unpublished). Since serum ferritin approaches a log-normal distribution, semi-logarithm paper was used for log transformation of values. Serum iron was determined using a colorimetric method<sup>7</sup>. Statistical analysis was carried out with  $\chi^2$  test, *t*-test, z-test and multiple correlation coefficient.

#### RESULTS

The mean and standard deviation for haemoglobin, serum ferritin and serum iron levels for pregnant women and menstruating women are given in Table 1. Anaemia was defined as haemoglobin concentration of less than 110 g/l for pregnant women and less than 120 g/l for menstruating women. According to this definition, 84.4% of pregnant women and 48.3% of menstruating women were anaemic.

The haemoglobin test detected 54.7% of cases with iron-deficiency anaemia among anaemic pregnant women. Eighty-nine per cent of iron deficient pregnant women had anaemia. There was a significant correlation between serum ferritin and low haemoglobin (<110 g/l) values (R=0.33, P>0.05). Serum iron and serum

Indicator	Statistic	Pregnant women	Non-pregnant women	P values
Haemoglobin	Mean $\pm$ SD (g/l)	93+16	119+13	≪0.001
Serum ferritin	Geometric mean $(\mu g/l)$	13.6	17.9	< 0.001
Serum iron	Mean $\pm$ SD ( $\mu$ mol/l)	$5.6 \pm 4.0$	$9.1 \pm 3.8$	< 0.001

Table 1. Iron status indicators in pregnant and non-pregnant women

SD = Standard deviation

ferritin concentrations were not significantly correlated (P > 0.10).

Clinical examination detected only 48.1% of anaemic subjects (haemoglobin < 110 g/l) and 53.3% as nonanaemic. There was no significant association between clinical examination results and haemoglobin levels. The proportions of anaemic subjects detected by clinical examination and those detected by haemoglobin assay were significantly different (P < 0.0001). Using serum ferritin values only 53.7% of women clinically diagnosed as anaemic had depleted iron stores. There was no significant relationship between clinical anaemia and iron deficiency (P > 0.1).

The means of haemoglobin values of subjects of gestation periods 20–24, 25–28, and 29–32 weeks were 94, 90, 95 g/l, respectively. Their corresponding medians of serum ferritin were 12.5, 10.7, 9.0, respectively. Sixty-six per cent of the pregnant women reported for antenatal care for the first time after their 24th week of gestation. Serum ferritin concentrations in the two gestational groups (i.e. 20–24 and 25–32) were statistically different ( $\chi^2$ =4.89; P < 0.05). Median serum ferritin declined progressively until about 31 weeks of gestation (Figure 1).

Haemoglobin levels were significantly associated with frequency of animal protein consumption in the two groups of animal protein consumption (<twice/week and  $\ge$  twice/week) as shown in Table 2.

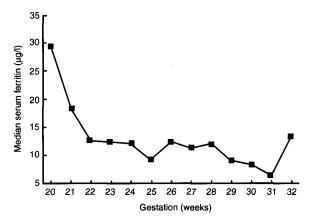


Figure 1. Median serum ferritin by gestation among pregnant women at Mbarara University Teaching Hospital (Uganda)

 Table
 2. Association
 between animal
 protein
 and

 haemoglobin (Hb) levels

II	Animal protein consumption		
Haemoglobin levels (g/l)	< Twice/week	$\geq Twice/week$	Total
Hb≥110	11	4	15
Hb<110	75	6	81
Total	86	10	96

 $\chi^2 = 5.047; P < 0.025$ 

Only 20 of 91 women were infected with helminths. Hookworm was the most common helminth (50% of infected cases). Other helminths were *Ascaris* species and *Trichuris trichura*. Sixty per cent of those infected with helminths had serum ferritin levels  $<12 \ \mu g/l$ . There was no statistical relationship between helminthic infection and haemoglobin levels ( $\chi^2 = 0.042$ ; 0.75 > P < 0.9). Helminthic infection and serum ferritin levels were not statistically related ( $\chi^2 = 0.944$ ; P = 0.20).

#### DISCUSSION

This study was based on a representative sample of women reporting for first antenatal care at MUTH and is not necessarily representative of the community as a whole. A big percentage (84.4%) of pregnant women studied were anaemic according to the World Health Organization (WHO) criteria used. This is comparable to that (78.4%) reported in Liberia<sup>8</sup>. Iron deficiency was the main cause of anaemia since 54.7% of anaemic pregnant women had iron-deficiency anaemia. Serum ferritin test, considered to be the most sensitive test, is used to assess iron status because it is directly proportional to the body level of iron stores9. However, in populations where iron deficiency coexists with infection, inflammation and chronic diseases, serum ferritin values may be in the normal range despite the presence of iron deficiency<sup>10</sup>. In our study it was not possible to include the transferrin saturation assay which would have, in combination with serum ferritin, improved the accuracy of iron status assessment. Nevertheless, the prevalence of iron deficiency observed is comparable to one reported in Zaire (Democratic Republic of Congo since 1997) (55.4%) where multiple indicators were used<sup>4</sup>. The geometric mean of serum

ferritin in the latter study was 10.7  $\mu$ g/l while that obtained in this study was 13.6  $\mu$ g/l.

In this study the pregnant group had low serum iron values compared to the non-pregnant group. This suggests iron deficiency within the pregnant group. The significant statistical difference between the geometric means of serum ferritin of the two groups (see Table 1) confirms iron deficiency in the pregnant group. Thus pregnancy reduces the iron stores of many women. This is supported by the mean haemoglobin concentration of the non-pregnant group which is within the accepted normal range. Lack of correlation between serum iron and serum ferritin of pregnant women was also observed by other workers<sup>11</sup>.

Decline in mean haemoglobin of the different gestation periods is partly due to the fall of haemoglobin levels as pregnancy advances. A similar decline observed by other workers was explained as due to the existence of iron deficiency rather than to a mere physiological haemodilution<sup>12</sup>. The decline of serum ferritin as pregnancy advances (see Figure 1) is due to the dilution and rapid mobilization of maternal iron stores.

A large number of women report late for antenatal care (at the end of second and in third trimester). This delays identification of anaemic individuals making it difficult to institute corrective measures. A substantial number (51.9%) of women were classified as non-anaemic clinically (i.e. had false negative results). No subject was clinically diagnosed as severely anaemic whereas haemoglobin assay detected 21.9% of pregnant women with severe anaemia. Severe anaemia refers to haemoglobin concentration below 80  $\mu g/l^{13}$ . This study shows that detection of anaemia by clinical screening is an insensitive technique and thus unreliable.

Diet played a major role in the high frequency of iron deficiency and anaemia due to the poor iron intake. The typical staple diet (matoke, milled millet and maize, cassava, potatoes and rice) was poor in haem iron and promoters of non-haem iron. It contained a majority of foods with compounds which inhibit iron absorption (tannates and phytates). A significant relationship between animal protein consumption, particularly meat and fish, and anaemia showed that low consumption of animal protein was a major cause of anaemia.

Lack of association between hookworm infestation and haemoglobin levels may be due to the method used which did not measure parasite load. Significance of hookworm infestation is most relevant at high levels of infestation<sup>14</sup>.

This study shows a need to search for other causes of anaemia other than iron deficiency and diet. Effects of malaria, levels of vitamins  $B_{12}$  and folate, and chronic diseases should be investigated. Clinical examination at the antenatal clinic should be complemented by the haemoglobin test. There is also a need to sensitize

pregnant women on the importance of early antenatal services.

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#### REFERENCES

- 1 WHO. Nutritional Anaemia, Tech Rep Ser No. 53. Geneva: WHO, 1972
- 2 Cook JD, Finch CA. Assessing iron status of a population. Am J Clin Nutr 1979;32:2115-19
- 3 Calvo EB, Sosa EM. Iron status in non-pregnant women of child-bearing age living in Greater Buenos Aires. *Eur J Clin Nutr* 1991;**45**:215–20
- 4 Mashako L, Preziosi P, Nsibu C, Galan P, Kapongo C, Potier de Courcy, et al. Iron and folate status in Zairian mothers and their newborns. Ann Nutr Metab 1991;35:309-14
- 5 Cheesbrough M. Direct examination of faecal specimens. In: Medical Laboratory Manual for Tropical Countries. Oxford: Butterworth-Heinemann, 1987
- 6 Dacie JV, Lewis SM. Basic haematological techniques. In: *Practical Haematology*. Edinburgh: Churchill-Livingstone, 1991
- 7 Dawson DW, Hoffbrand AV, Worwood M. Investigation of megaloblastic and iron-deficiency anaemia. In: Dacie JV, Lewis SM, eds. *Practical Haematology*. Edinburgh: Churchill-Livingstone, 1991:412–13
- 8 Jackson RT, Jackson LC. Biological and behaviour contributors to anaemia during pregnancy in Liberia, West Africa. Human Biol 1987;59:585-97
- 9 Romslo I, Haram K, Sagen N, Augensen K. Iron requirement in normal pregnancy as assessed by serum ferritin, serum transferrin saturation and erythrocyte protoporphyrin determinations. Br J Obstet Gynaecol 1983;90:101-7
- 10 Prual A, Galan P, De Bernis L, Hercberg S. Evaluation of iron status in Chadian pregnant women: consequences of maternal iron deficiency on the haemotopoietic status of newborns. *Trop Geogr Med* 1988;40:1–6
- 11 Kumar N, Bhargava M, Sundara KR, Kumar S, Bhargava VL, Kumar R. Serum ferritin as a measure of iron stores in normal Indian subjects and pregnant women. *Nutr Res* 1989;9:999–1006
- 12 Kaufer M, Casanueva E. Relation of pre-pregnancy serum ferritin levels to haemoglobin levels throughout pregnancy. *Eur J Clin Nutr* 1990;44:709–15
- 13 Yip R. Iron deficiency: contemporary scientific issues and international programmatic approaches. J Nutr 1994;124: 1479 S-1490 S
- 14 Royston E. The prevalence of nutritional anaemia in women in developing countries: a critical review of available information. World Health Statistics Quart 1982;35:52-91