Nutritional Sciences 7(4), November, 2004, pp.218-222

Incidence of Iron Deficiency Anemia in Day Scholar University Girls as Affected by Socioeconomic Status

Nayab Batool, Saeed Ahmad Nagra[§] and Muhammad Imtiaz Shafiq
Biochemistry Section, Institute of Chemistry University of the Punjab, Lahore 54590, Pakistan

Iron deficiency anemia (IDA) is the world's most common nutritional problem. It is characterized by a low hemoglobin (Hb) level and low iron status. A study was conducted to investigate the incidence of iron deficiency anemia in day scholar girls belonging to different socioeconomic strata at Punjab University, Lahore. Iron status of the subjects was estimated by measuring Hb, hematocrit (Hct), red blood cell (RBC) count, mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), serum iron, serum ferritin, total protein and albumin. Results indicated that females belonging to low socioeconomic strata had lower values for Hb, Hct, RBC count, total protein and albumin. Serum iron, serum ferritin, MCV and MCH values fell within the normal range for all of the socioeconomic groups. However, serum iron and ferritin varied with socioeconomic status and higher-income groups had significantly higher serum iron and ferritin. It was concluded that anemia may develop due to poor intake and absorption of iron and that those in the low-income bracket are the most affected group.

Key words : Pakistan, Iron, University, Girls, Anemia

INTRODUCTION

Iron deficiency anemia (IDA) is the most common nutritional problem in both the developing and developed worlds.[1] Females are relatively more prone to the problem. Anemia is characterized as a "reduction in the number of red blood cells or the quantity of hemoglobin and volume of packed red cells per 100 mL of blood." The World Health Organization[2] has recommended the cut-points for diagnosing anemia in adults as 13 g/dl for men, 12 g/dl for menstruating women and 11 g/dl for pregnant women. For optimal health, the amount of iron obtained from the diet must replace the obligatory losses from the skin and gastrointestinal system. The iron content of the body is regulated by homeostatic mechanisms mainly through the control of absorption from the gut. Clinical manifestations of iron deficiency anemia include pallor, irritability, fatigue, tachycardia, sore or swollen tongue, enlarged spleen, pica, weakness, shortness of breath, headache and loss of appetite. The classic symptom of iron deficiency anemia is fatigue. Iron absorption may be retarded due to excessive intake of tea, which contains tannin, polyphenols present in certain vegetables, phytates in bran and calcium in dairy products. Absorption is augmented by the presence of reducing substances such as hydrochloric acid and ascorbic acid.[3]

In the absence of any infection, hemorrhage or parasitic infestations, IDA is directly related to iron intake. Aside from low iron intake, a common cause of iron deficiency anemia is parasitic infestations. The hookworm provokes damage to the villi, resulting in blood loss and anti-coagulant production, which promotes continued bleeding. Each worm can provoke the loss of up to 250 μl of blood per day. It is estimated that this disease affects over 200 million people in India and Pakistan (National Health Survey of Pakistan,[4] 1998).

IDA prevalence is generally high in women of reproductive age because iron requirements are high during this period owing to iron loss from menstruation and iron-poor diets. It has been estimated that 90 percent of all anemic individuals are found in developing countries, including Pakistan. According to the National Health Survey of Pakistan[4] (1998) 43-47 percent of rural women and 35-39 percent of urban women, aged 15-44 years, are anemic. It is believed that most of these anemia cases are nutritional and due to iron deficiency. In Punjab University hostels, the majority of students had hemoglobin levels below the standard, but the cause of anemia was not known.[5] The recommended dietary allowance (RDA) for iron in Pakistan is a minimum of 18 mg for pre-menopausal women. For the population as a whole, the recommended dietary allowance of iron is 15 mg per capita per day. Most women don't get the

Accepted : September 24, 2004
[§] To whom correspondence should be adressed.

suggested 18 mg of iron daily from their diet alone.

The present study is an attempt to assess iron intake and the magnitude of IDA in day scholar university girls belonging to different socioeconomic groups.

MATERIALS AND METHODS

1. Selection of Study Samples

Total study sample comprised 300 women between the ages of 20 and 23, studying in various teaching departments at the University of the Punjab, Lahore. Depending on the socioeconomic status of the subjects, the study sample was further classed as high, medium and low according to the method of Nagra *et al.*.[6] Each socioeconomic group consisted of 100 subjects. All of the students included in the study were healthy and apparently did not have any clinical manifestation.

2. Collection of Blood Samples

Five ml of venous blood was drawn from each subject with a pyrogen-free, disposable syringe between 10 and 11 am. Blood was transferred in heparin-containing blood-collecting tubes.

3. Assessment of Iron Intake

Seven-day diet record was used to estimate usual iron intake. Respondents were asked to record anything they consumed. The collected data was classed under cereals, vegetables, pulses, milk, eggs and meat. Using food composition tables, the amount of iron intake was calculated. The daily average of the total intake over 7 days was considered as the daily intake of iron.[7]

4. Biochemical Parameters

Blood was used to estimate hemoglobin,[8] hematocrit[8] and red blood cell count[9] (RBC count). Serum was prepared by centrifuging blood samples at 4,000 rpm for 7-10 minutes. Serum was separated and stored at 10 ℃ in Eppendorf tubes and analyzed for the following parameters.

5. Serum Total Protein

Estimated using biuret method.[10]

6. Serum Albumin

Determined using BCG (Bromocresol green) method.[11]

7. Serum Iron

Ferene-S method[12] was employed for this determination using a diagnostic kit (M/S Clonitol, Italy).

8. Serum Ferritin

Ferritin was determined using a quantitative test kit based on a solid phase ELISA. The assay system used one anti-ferritin antibody for the solid phase (microtiter wells) immobilization and a mouse monoclonal anti-ferritin antibody in the antibody enzyme (horseradish peroxidase) conjugate solution. The test sample was allowed to react simultaneously with the antibodies, resulting in the sandwiching of ferritin molecules between the solid phase and the enzyme-linked antibodies. After a 60 minute incubation at room temperature, the wells were washed with water to remove unbound labeled antibodies. A solution of tetra methyl benzidine was added and incubated for 20 minutes, which turned the solution blue. The addition of 2N HCl changed the color to yellow. The color intensity was measured spectrophotometrically at 450 nm. The concentration of analyte was directly proportional to the color intensity of the test sample.[13]

The following parameters were worked out from hemoglobin and RBC count:

9. Mean Corpuscular Volume (MCV)

MCV is a measure of the average size of the red blood cells.

MCV = hematocrit (volume fraction) / RBC count (M/mm^3)

10. Mean Corpuscular Hemoglobin (MCH)

MCH refers to the hemoglobin content of the individual red blood cells. It is derived from the ratio of hemoglobin to red blood cell count.

MCH (pg) = Hemoglobin (g/dl) / RBC count (M/mm^3)

11. Statistical Analysis

Descriptive statistic and analysis of variance[14] was worked out using SPSS version 9.0 for Windows. Fischer PLSD test was used for multiple comparison of means.

RESULTS AND DISCUSSION

1. Hemoglobin and Related Parameters

Hemoglobin is one of the definite and reliable measures to assess the existence of iron deficiency anemia. As shown in Table 1, girls belonging to the low socioeconomic group had significantly ($P < 0.05$) lower hemoglobin than those belonging to the other two income groups. The difference in the hemoglobin value between those in the high and medium groups was statistically insignificant.

The results obtained in the present study are in line with those of Reddy,[15] who conducted a hemoglobin survey among urban residents in Chennai, India. The survey showed that 87.5 percent of the subjects under study were anemic. These results were further substantiated by the study of Godwin *et al.*.[16] A nutritional

Table 1. Effect of socioeconomic status on some hematological parameters of day scholar university girls in Pakistan

Parameters	Socioeconomic status			Overall values	Normal Values[1)]
	High	Medium	Low		
Hemoglobin (g/dL)	$13.8^{a}\pm0.4$	$12.9^{a}\pm0.3$	$10.6^{b}\pm0.2$	12.4±0.3	12-14
Hematocrit (%)	$41.7^{a}\pm2.4$	$35.5^{b}\pm2.2$	$28.0^{c}\pm2.0$	35.0±2.2	36-47
RBC count (M/mm^3)	$4.6^{a}\pm0.2$	$3.9^{b}\pm0.2$	$3.4^{b}\pm0.2$	3.8±0.2	4.2-5.4
MCV (fL)	$93.2^{a}\pm0.9$	$91.0^{b}\pm0.8$	$88.1^{b}\pm1.1$	90.7±0.9	78-98
MCH (pg)	$30.6^{a}\pm0.8$	$29.4^{a}\pm0.2$	$28.0^{b}\pm0.4$	29.3±0.5	27-32
Serum proteins (g/dL)	$8.5^{a}\pm0.4$	$7.2^{b}\pm0.4$	$5.7^{c}\pm0.4$	7.2±0.4	6.0-8.3
Serum albumin (g/dL)	$5.1^{a}\pm0.4$	$4.2^{a}\pm0.3$	$3.4^{b}\pm0.1$	4.2±0.23	3.5-5.0
Serum iron (g/dL)	$95.2^{a}\pm8.8$	$64.8^{b}\pm7.3$	$42.6^{c}\pm6.6$	67.5±7.5	37-145
Serum ferritin (ng/mL)	30^{a}	27^{a}	20^{b}	29	10-150
Daily iron intake (mg)	13^{a}	13^{a}	10^{b}	12	18

Means in a row followed by different letters are significantly different at $P<0.05$
1) Gowenlock AH. Varley's Practical Clinical Biochemistry, 6th ed. 1988
2) MCV: mean corpuscular volume 3) MCH: mean corpuscular hemoglobin

survey of Pakistan[17] also indicated the existence of anemia in all age groups. No difference has been found in its prevalence between pregnant and non-pregnant women, among those having different levels of education or between those living in urban and rural areas. Low hemoglobin can be attributed to low intake of iron, poor absorption and parasitic infestation. It is evident from Table 1 that iron intake is about 33 percent lower than the desired level and the difference is significantly ($P<0.05$) higher in girls belonging to the low-income group.

Hematocrit is the measurement of the percentage of red blood cells in whole blood. It is an important determinant of anemia resulting from an increased red blood cell breakdown in the spleen. In early cases of moderate iron deficiency, a marginally low hemoglobin value may be associated with a near normal hematocrit. It appears as if the degree of anemia is inversely proportional to the hematocrit value and has a positive correlation with hemoglobin concentration. The results of the present study revealed that hematocrit decreased with decreasing hemoglobin levels. A reduction in hematocrit is indicative of a low intake of iron.[18]

RBC count determines the total number of red cells in a blood sample. It has been observed that at any stage, the red cell number will not be proportionately as low as that of the hemoglobin and hematocrit. Girls belonging to high socioeconomic groups had normal and significantly ($P>0.05$) higher RBC counts than girls in the medium- and low-income groups. The difference in the figures for the medium- and low socioeconomic groups was insignificant and was at the lower end of the normal range. RBC count appeared to vary with socioeconomic status. The results obtained in the present study are in line with those of Saxena and Wong[19] who determined hematological indices among young females and observed that females had significantly lower RBC counts. The decrease in RBC count may be due to the poor bio-availability of iron as studied by Samuelson *et al.*[20] who estimated the iron intake and iron status of females (16-24 years). Red cell indices, i.e. MCV and MCH, are derived from measurements of hemoglobin, hematocrit, and/or RBC count. A decreased value of MCV may indicate iron deficiency anemia. As shown in Table 1, all of the subjects belonging to the different socioeconomic groups had values within the normal range. These results are in line with those of Armstrong[21] and Rietjens *et al.*[22] who estimated the values of MCV for young females and found that the majority of the subjects had values within the normal range, despite low Hb values. Females belonging to different socioeconomic backgrounds had normal values for MCH. However, the values varied with their socioeconomic status.[23] The results are further supported by the studies of Karazawa *et al.*,[24] Castro *et al.*,[25] and Mangwendeza *et al.*,[26] who carried out hematological studies of young women and found that there were significant differences in MCH values due to social and nutritional factors.

2. Serum Total Protein and Albumin

Formation of hemoglobin is as dependent upon good protein intake as the iron itself. Albumin is the major serum protein that is synthesized by the liver from amino acids derived from the diet. Although the values for the test subjects fell within the normal range (Table 1), they appeared to vary with socioeconomic stratification. The results obtained in the present study for serum albumin showed a variation trend similar to that of serum total protein. These results are in line with those of Yegin and Soysal[27] who reported that subjects belonging to low socioeconomic strata had lower total protein and albumin values. It was interesting to note that the serum total protein values were related to the degree of anemia.

3. Serum Iron

Serum iron is particularly useful for differentiating between nutritional deficiencies of iron and iron deficits arising from chronic infections or inflammation. Iron

deficiency results in a fall in serum iron levels. All of the test subjects (Table 1), irrespective of socioeconomic background, had normal serum iron. However, it varied with socioeconomic status and higher income groups had significantly ($P<0.05$) higher serum iron.[28] Low serum iron in low-income groups has been attributed to poor bioavailability and absorption of iron.[29] It may be postulated from the foregoing discussion that family income influences the state of iron nutrition and hence susceptibility to iron deficiency anemia.

4. Serum Ferritin

Intake, loss and storage determine the total amount of iron in the body. Iron is stored primarily as ferritin.[30] Under normal conditions, a direct relationship exists between serum ferritin concentration and the amount of iron stored in the body.[31] Serum ferritin was also seen to vary significantly ($P<0.05$) with socioeconomic stratification (Table 1). All the volunteers had ferritin values within the normal range and close to the lower margin.

The results of the present study indicate that the iron status of day scholar university girls was directly governed by their socioeconomic status. When compared with the reference values of hemoglobin and related parameters, about one-third of the participants appeared to suffer from IDA of varying intensities. However, none of the participants experienced any sort of problem related to IDA.

Literature Cited

1) Quintas ME, Requejo AM, Ortega RM, Redondo MR, Lopez-Sobaler AM, Gasper MJ. The female Spanish population: a group at risk of nutritional iron deficiency. *Int J Food Sci Nutr* 48:271-9, 1997
2) WHO. Nutritional anemia: WHO technical report series No. 3. World Health Organization, Geneva. 1972
3) Siegenberg D, Baynes RD, Bothwell TH. Ascorbic acid prevents the dose-dependent inhibitory effects of polyphenols and phytates on nonheme-iron absorption. *Am J Clin Nutr* 53:537-41, 1994
4) National health survey of Pakistan. Pakistan Medical and Research Council, Islamabad, pp.50-51, 1998
5) Sabahat K. Nutrition status of females as affected by hostel diet. M.Sc. thesis, Institute of Chemistry, University of the Punjab. Lahore, 1994
6) Nagra SA, Gilani AH, Ahmad MD, Haq E. A longitudinal study in body weight of Pakistani infants as influenced by socioeconomic status. *J Trop Pediar* 30:217-221, 1984
7) Horwath CC. Dietary intake and nutrition status among university undergraduates. *Nutr Res* 11:395-404, 1991
8) Graitcer PL, Goldsby JB, Nichaman MZ. Hemoglobins and hematocrits: are they equally sensitive in detecting anemias? *Am J Clin Nutr* 34:61, 1981
9) Gibson RS. Principals of Nutritional assessment. Oxford University press. New York, 1990
10) Tietz NW. Clinical Guide to Laboratory Tests, 3rd Edition WB Saunders Company Philadelphia, PA pp.518-519, 1995
11) Doumas BT, Biggs HG. Determination of serum albumin. In: Standard Methods of Clinical Chemistry. GA Cooper, ed. Vol. 7, p.175, Academic Press, Inc.. New York, 1972
12) Artiss JD, Vinogradov S, Zak B. Spectophotometric study of several sensitive reagents for serum iron. *Clin Biochem* 14:311-15, 1981
13) Primus FJ, Kelley EA. "Sandwich"-Type Immunoassay of Carcinoembryonic Antigen in patients Receiving Murine Antibody for Diagnosis and Therapy. *Clin Chem* 34:261-265, 1988
14) Kennedy JJ, Bush AJ. An introduction to the design and analysis of experiment in behavioral research. Ohio State University press. 1985
15) Reddy V. Prevalence of anemia and iodine deficiency disorders In: nutrition trends in India, NIN, pp.37-38, 1993
16) Godwin ID, Jencks JA. Normal hematologic values obtained with a counter Model S. *South Med J* 7:47-9, 1980
17) National Nutrition Survey. Nutrition Division, National Institute of Health. Government of Pakistan, Islamabad. 1998
18) Kuvibidila S, Yu L, Ode D, Mbele V, Warrier, RJ. An epidemiological study of hemoglobin levels and prevalence of anemia in young women. *Ann Soc Belg Med Trop* 73:227-34, 1993
19) Saxena S, Wong ET. Heterogeneity of common hematologic parameters among racial, ethnic and gender subgroups. *Arch Pathol Lab Med* 114:715-9, 1990
20) Samuelson G, Bratteby LE, Berggren K, Elverby JE, Kempe B. Dietary iron intake and iron status in adolescents. *Acta Pediar* 85:1033-8, 1996
21) Armstrong P. Full blood count values in adolescents. *Ir Med J* 82:68-9, 1989
22) Rietjens GJ, Kuipers H, Hartgens F, Keizer HA. Red blood cell profile of elite Olympic distance triathletes, a three-year follow-up. *Int J Sports Med* 23:391-6, 2002
23) Shiga S, Koyanagi I, Kannagi R. Clinical reference values for laboratory hematology tests calculated using the interactive truncation method with correction. *Rinsho Byori* 38:93-103, 1990
24) Karazawa EH, Jamra M. Normal hematologic parameters. *Rev Saude Publica* 23:58-66, 1989
25) Castro OL, Haddy TB, Rana SR. Age and sex related blood cell values in healthy black Americans. *Public Health Rep* 102:232-7, 1987
26) Mangwendeza MP, Mandisodza A, Siziya S. Hematology-reference values for healthy elderly blacks residing in Harare, Zimbabwe. *Cent Afr J Med* 46:120-3, 2000
27) Yegin MM, Soysal T, Keha EE, Cil MY, Baken E. Estimation of serum total protein and albumin. *Clin J Med* 101:165-6, 1984

28) Sue-Houston RD, Susan L, Summers BS, Kay S, Soltesz RD. Lifestyle and dietary practices influencing iron status in university women. *Nutr Res* 17:9-22, 1997
29) Nicklas TA, Kuvibidila S, Gatewood LC, Metzinger AB, Frempong KO. Prevalence of anemia and iron deficiency in urban Haitian children two to five years of age. *J Trop Pediatr* 44:133-8, 1998
30) Bothwell TH. Overview and mechanisms of iron regulation. *Nutr Rev* 53:237-45, 1995
31) Lipschitz DA, Cook JD, Finch CA. A clinical evaluation of serum ferritin as an index of iron stores. *N Engl J Med* 290:1213-6, 1974