RESEARCH PAPER

Determinants of Nutritional Anemia in Adolescents

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Objectives: To associate the severity of nutritional anaemia with serum levels of ferritin, vitamin B12 and folate; and to determine demographic, socio-economic and nutritional correlates for nutritional anemia in adolescents.

Methods: Cross-sectional hospital-based study among 200 adolescents (10-18 y) with anemia. Dietary intake (24-h recall), and serum levels of folate, vitamin B12 and ferritin were estimated.

Results: Iron, folate and vitamin B12 deficiency was present in 30.5% 79.5% and 50% of adolescents, respectively. Statistically significant association was observed between severity of anemia and serum vitamin B12 levels, iron intake, folate intake, Vitamin B12 intake, vegetarian diet, attainment of menarche and history of worm infestation.

Conclusions: Folate and vitamin B12 deficiencies are more common than iron deficiency in anemic adolescents. Low dietary intake of these nutrients seems to be a significant determinant of their deficiencies.

Keywords: *Ferritin, Folic acid, Iron-deficiency, Vitamin B12 deficiency*

Adolescence is a vulnerable period in the human life cycle for the development of nutritional anemia. Anemia has a negative effect on cognitive performance in adolescents [1]. Choudhary, *et al*. [2] reported two-thirds of the anemic adolescents in community were suffering from iron deficiency anemia (IDA). In a study by Patra, *et al*. [3] on severely anemic adolescents admitted in a tertiary care hospital, megaloblastic anemia was most common type of anemia (42.5%) and iron-deficiency accounted for 15% cases [3]. Compared to the vast amount of work done in pregnant mothers and young children, there are relatively few published studies in India evaluating deficiencies of Iron, Vitamin B12 and Folate in adolescents having nutritional anemia and its association with severity of anemia. This study was planned with an objective of finding association of the levels of micronutrients with the severity of nutritional anemia in adolescents.

Methods

The present study was a hospital-based cross-sectional observational study conducted in the Department of Pediatrics, Lady Hardinge Medical College, New Delhi, India, during November 2011 to April 2013. Adolescents (age 10-18 years) attending the outpatient department or admitted in the hospital, and having hemoglobin values below the cut-offs (Hb <12g/dL in 10-18 y girls and 10-14 y boys and Hb<13g/dL in 15-18 y boys) were included in the study after written informed consent from parent/guardian. Those who had received blood transfusion or hematinics in past 4 weeks or having apparent infection (fever, diarrhea, cough or burning micturition) or any chronic disease were excluded from the study. A detailed history and physical examination of the study population was carried out. The premorbid dietary intake of the child was assessed by 24-hour recall method. This data was entered in 'Diet soft' software from which the daily intake of iron, folate and vitamin B12 was calculated.

Complete blood count with peripheral smear examination was done. Serum ferritin was estimated by a Microplate Immunoenzymometric assay using Calbiotech Ferritin ELISA Kit, California, US (FR06F) while serum Folate and Vitamin B12 were estimated through automated immunoassay system using Beckman Coulter Access-2 (Beckman Coulter, Inc. Access Folate Reagent- A14208 and Access Vitamin B12 Reagent-33000), California, US.

The severity of anemia was graded as mild (>10 g/dL but below age related cut-off for defining anemia), moderate (7-9.9 g/dL) and severe (<7g/dL). Serum vitamin B12 level of <200 pg/mL, folate level <5 ng/mL and ferritin level < 30ng/mL were considered as deficient.

Normocytic anemia was defined as MCV 78-98 fL in males and 78-102 fL in females; microcytic as MCV <78 fL; and macrocytic as MCV >98 fl in males and >102 fL in females. The SPSS 21.0 software was used for data analysis. Fischer's exact test and analysis of variance were performed; $P<0.05$ was considered as significant.

RESULTS

A total of 350 adolescents (155 inpatients) with pallor were screened of which 200 (99 males) satisfied the inclusion criteria. Two-thirds (69.5%) of participants belonged to early adolescence age (10-13.9 y), -Almost half (54%) of the adolescents had normal BMI (+1 to -2 SD), 27.5% were very underweight (<-3 SD), 17.5% were underweight (-2 to -3 SD), and only 1 % were overweight (+1 to +2 SD). Dietary evaluation revealed that energy intake was deficient in 94.5% of adolescents, iron intake was deficient in 99.5%, vitamin B12 intake was deficient in 14.5% and folate intake was deficient in 62.5% of anemic adolescents.

Mean (SD) hemoglobin was 9.4 (2.5) g/dL with 50.5% having mild anemia, 29% having moderate anemia while 20.5% having severe anemia; 55% had normocytic anemia, 27.5% had microcytic, 8.5% had macrocytic, and 9% had dimorphic anemia. Median levels of micronutrients across different groups of anemia severity are presented in ***Table* I**. Iron deficiency was present in 30.5% subjects, vitamin B12 deficiency in 50% of subjects and folate deficiency was present in 79.5% of subjects. Isolated iron, vitamin B12 and folate deficiency was seen in 5%, 4% and 25%, respectively while combined folate and vitamin B12 deficiency was seen in 32%. Deficiency of all the three micronutrients was documented in 12% while 9% had no deficiency.

Severe anemia was significantly associated with history of worm infestation, attainment of menarche, vegetarian diet and low serum B12 levels (***Table* II**). Low intake of iron, vitamin B12 and folate were also significantly associated with severe anemia.

DISCUSSION

Our study showed that deficiency of folate, vitamin B12 and iron are common in anemic adolescents. Higher proportion of severely anemic individuals (20.5%) in our study can be attributed to hospital-based nature of the study. Prevalence of iron deficiency, vitamin-B12 deficiency and folate deficiency in our study is comparable to earlier reports from India [3-6]. Under the

TABLE I MICRONUTRIENT INTAKE AND LEVELS ACROSS DIFFERENT GROUPS OF ANEMIA SEVERITY

	Mild Anemia	*Moderate Anemia*	*Severe Anemia*	*Total*
Iron (mg/d), intake, mean (SD)	11.4 (3.4)	9.9 (3.2)	9.4 (3.0)	10.5 (3.4)
Vit.B12 (µg/d) intake, mean (SD)	1.0 (0.8)	0.8 (0.7)	0.5 (0.7)	0.9 (0.8)
Folate (mg/d) intake, mean (SD)	144.0 (59.4)	135.3 (50.4)	110.2 (47.435)	135.1 (55.9)
Serum ferritin (ng/mL), median (IQR)	45.3 (12.3-150.8)	45.1(5.8-163.8)	35.4 (2.2-132.7)	43.6 (6.3-144.0)
Serum vitamin B12 (pg/mL), median (IQR)	206.0 (160.0-302.5)	241.0 (166.5-313.0)	119.0 (160.0-241.0)	198.5 (145.3-305.5)
Serum folate (ng/mL), median (IQR)	4.0 (2.9-4.6)	3.7 (3.1-4.9)	3.27 (2.30-4.94)	3.7 (2.9-4.7)

TABLE II ASSOCIATION OF SOCIODEMOGRAPHIC PARAMETERS WITH SEVERITY OF ANEMIA

Variables	*Severe anaemia (n=41) n (%)*	*Mild- moderate anemia (n=159) n (%)*	*OR (95% CI)*	*P value*
Female gender	25 (60.9)	76 (47.7)	1.70 (0.84-3.43)	0.16
Vegetarian diet	17 (41.4)	22 (13.8)	4.41 (2.04-9.51)	<0.001
Upper lower socioeconomic status	32 (78.0)	105 (66.0)	1.82 (0.81-4.10)	0.18
History of worm infestation	13 (31.7)	29 (18.2)	2.08 (0.96-4.50)	0.04
Post menarchal	15 (36.5)	46 (28.9)	1.02 (0.40-2.57)	0.02
BMI < –3SD	12 (29.2)	43 (27.0)	1.11 (0.52-2.83)	0.84
Serum ferritin <30 ng/mL	18 (43.9)	43 (27.0)	2.11 (1.03-4.29)	0.055
Serum vitamin B12 <200 pg/mL	28 (68.2)	72 (45.2)	2.60 (1.25-5.39)	0.01
Serum folate <5 ng/mL	31 (75.6)	128 (80.5)	0.75 (0.33-1.69)	0.51

WHAT THIS STUDY ADDS?

- Deficiency of folic acid, vitamin B12 and iron are common among adolescents with anemia.

National programs (Iron plus initiative and weekly iron folate supplementation), the beneficiary receives supplemental iron and folic acid. Deficiency of B12 is currently not being addressed through these programs [7,8].

Vegetarianism was significantly associated with severe anemia which was similar to the findings by Verma, *et al*. [9]. Attainment of menarche was also significantly associated with of anemia which was in agreement with the findings of Heath, *et al*. [10] who reported that high menstrual blood loss was associated with increased risk of anemia. A significant association between history of worm infestation and severity of anemia in the present study was in agreement to findings of Shield, *et al*. [11] who demonstrated a statistically significant inverse correlation between hookworm egg count and hemoglobin level.

Our study findings are limited by the hospital-based design of the study. Another limitation is that presence of infections which were not picked up on detailed history and examination could have erroneously elevated the serum ferritin above the cut-off used for defining iron deficiency anemia.

We conclude that low intake of iron folate and vitamin B12 is a significant determinant towards causing nutritional anemia in adolescents. Supplementation with not only iron and folic acid but also vitamin B12, besides deworming, is required through national programs.

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