Original Research Article

Estimation of hemoglobin for screening of anemia in undergraduate students in Hyderabad city, India

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ABSTRACT

Background: Anemia is a condition that is marked by low levels of hemoglobin in the blood. Iron is a key component of hemoglobin, and iron deficiency is estimated to be responsible for half of all anemia globally. The present study was conducted to find out the prevalence of anemia among undergraduate students of (17-21 years) in Hyderabad city, India.
Methods: A cross-sectional study was conducted in different UG colleges in urban areas of Hyderabad for 3 months and a total 57 women participants were selected to volunteer study. All participants underwent screening using anthropometry with haemoglobin estimation using Sahli's method. The 24-hour dietary recall was also recorded and analysed to assess the nutritional adequacy of the diet.
Results: Prevalence rate of iron deficiency anemia was found to be 57.89 per cent, of the total 54.38 per cent were mild anemic and 3.51 percent were moderately anemic.
Conclusions: Nutritional anaemia is one of the most preventable forms of anaemia if resources available individualize therapy focusing on the confluence of various causative factors should be implemented.

Keywords: Haemoglobin, Nutritional deficiency, Nutritional anaemia, Sahli's method

INTRODUCTION

WHO defined anemia as a condition where the number of RBC is less.[1] Anaemia may occur due to nutritional inadequacies or any other clinical conditions like malabsorption and hookworm infestation where intestinal absorption of iron altered due to gut environment. Nutritional anaemia can also be defined as the condition resulting from the inability of the erythropoietic tissue to maintain a normal hemoglobin concentration due to inadequacy of one or more nutrients.[2] Adolescence is the period of the growth spurt in all aspects hence their nutritional status plays a vital role in the progress.[3] Anaemia is widely prevalent among all age groups, but since the requirement of iron is more and loss of around 12.5-15mg iron each month through menstruation,

adolescent girls are more vulnerable to anaemia.[4] The prevalence of anemia in adolescent girls is 56% in India and the prevalence of anemia in the state of Telangana among women aged 15-49 years is 56.7%.[5,6] A report by Kasturba Bora studied the trends and prevalence of severe anemia in India showing severe anemia (HB < 7g/dl) in Telangana among the other states. The trends also varied between rural and urban areas.[7]

The rate of incidence in urban area women 15-49 years of Telangana is 55% and in the rural area is 58.1%. The National Family Health Survey (NFHS)-2 data 2015-2016, adolescent girls aged between 15-19 years shows iron deficiency anemia contributing to 59.4%.[8] Although there is a fall in the prevalence of anemia as per the NFHS-2 from 67.4% (2005-06) to 59.4% due to the

various intervention taken up such as oral supplementation under national nutritional anaemia prophylaxis programme, WIFS programme launched under the Ministry of Health and Family welfare- Government of India, fortification, dietary diversification and through health communications, the impact of anemia among adolescent girls is still substantive.[9,10]

Anaemia affects the overall nutritional status of adolescents and causes adverse consequences as it progresses.[11,5] It affects the growth, onset of menarche, academic performance, physical performance, cognitive development etc., The most common cause of anemia is iron-deficiency anemia. Other conditions like parasitic infections, excessive blood loss during menstruation and growth spurts with suboptimal hematopoietic contents also contribute to anemia. Many nutrients play a vital role in the process of erythropoiesis- a process of formation of Red blood cells in the bone marrow, such as proteins (glycine, histidine, proline and hydroxyl proline), iron, copper, vitamin E, riboflavin, pyridoxine, folate and cyanocobalamin and vitamin C.[2] Deficiency in any of these nutrients may retard the process thus contributing to anemia.

This study is aimed at reviewing the impact of dietary inadequacies and prevalence of anemia among the adolescent girls of urban area from a good socio-economic background.

METHODS

The study was conducted in degree colleges of Hyderabad city, TS, India for a period of 3 months i.e.., from August to October 2019. The sample size was calculated using Slovin's formula (n= N/(1+Ne2).

n= 70 participants, e = 5% error tolerance (i.e., 0.05)

$$n= 65/1+ 65 \times (0.05)^2$$

$$n= 59.5 (60)$$

Out of the 60 participants, 3 were excluded due to their unwillingness to participate in the study. Criteria of exclusion also included the presence of non-nutritional anemia due to infection, genetic disorders and certain cancers. So total 57 non-pregnant, non- lactating (NPNL) women of 17-20 years belonging to various colleges of the city participated in the study. Written consent from each participant was taken, they were explained about the procedure involved in the drawing the blood sample for hemoglobin estimation.

Estimation of hemoglobin by Sahli's haemoglobinometer

The incidence of anemia among adolescents was known by estimating their hemoglobin levels using Sahli's Haemoglobinometer.

The Sahli's method is one of the most accepted visual method, less expensive, convenient and it is as reliable as automated methods. It is a colorimetric method where hemoglobin content is estimated by measuring the intensity of the colour compound formed from the sample and compared with the colour of standard hemoglobin.

Working principle

The hydrochloric acid used in Sahli's method converts the hemoglobin to acid haematin which forms a brown colour.[12] This colour formed is diluted to the point until it matches with permanent standard hemoglobin comparator in the haemometer. The Haemoglobinometer tube has calibrations and the values are expressed as gram% and in per cent values. The corresponding values can be noted.

The participants were well informed about the test. Hemoglobin values in gram per cent are noted and are compared with the standard reference ranges given by WHO cut off values for assessing anemia in adolescents Table 1.

Table 1: WHO cut off values for assessing anaemia.

Cut off values	Inference
>12 g/dl	Normal
<11.9 g/dl - >11 g/dl	Mild anaemia
<10.9 g/dl - > 8 g/dl	Moderate anaemia
<8 g/dl	Severe anaemia

Demographic characteristics

Demographic details, socio-economic status were also included in a separate form. The anthropometric measurements were taken to find out any macronutrient deficiencies among the adolescent's girls.

Height, weight was measured and compared with standard reference height and weights and body mass index was calculated using formula weight (kg)/ height (m^2), to know the degree of nourishment using the reference BMI for Asians.

Diet survey

In order to assess dietary inadequacies, a diet survey was done. A pre-set questionnaire including 24hr dietary recall and semi-quantitative food frequency questionnaire was used. The participants were explained about the questionnaire and asked to seek our help whenever necessary during the survey.

Statistical analysis

Mean and standard deviation of the data was known using Microsoft Excel 2013. Student's one sample T-test was used with a p-value significant at 5% for the mean dietary intakes.

RESULTS

Anthropometric assessments were taken and analyzed using ICMR standards for Indians. Mean height and weight were compared with the standards indicating that there were no indications of long term nutritional deprivation during the growth years. Table 2 represents the mean height and weight of participants.

Body mass index was calculated to know adiposity, participants BMI was calculated and further they were classified according to BMI Table 3.

The 24-hour dietary recall was used to find out the deviation of dietary intake from the RDA for nutrients like proteins, iron, folic acid, vitamin B12 and vitamin C. The food frequency questionnaire was used to know the frequency of consumption and deduce a prolong deficiencies of nutrients in them.

The dietary habits of the participants were also noted. Dietary nutrient intake and food preferences were summarized in Table 4. Classification of participants as per the degree of anemia using WHO cutoff values represented in Table 5.

Table 2: Mean anthropometric measurements of participants.

Age in years (N)	Height (cm)	SD	P value	Weight (kg)	SD	P-value
17 (9)	156.33	4.95	3.17*	56.44	14.2	2.78*
18 (29)	154.4	7.27	2.29*	52.76	14.28	0.09
19 (11)	157.18	5.64	3.64*	57.18	10.28	0.7
20 (08)	150.25	4.68	0.45	52	13.14	0.65

n=57, *significant difference at 5% level of significance.

Table 3: Distribution of participants according to BMI.

Age in years	Underweight <18.5 (kg/m^2)*	Normal 18.5-22.9 (kg/m^2)*	Over weight 23-26.9 (kg/m^2)*	Obese ≥27 (kg/m^2)*	Total (n=57)
17	8	1	0	0	9
18	20	5	4	0	29
19	9	1	1	0	11
20	7	1	0	0	8

* BMI cut off for Asians.

Table 4: Mean dietary intake of food in g/ml.

Type of meal	Mean intake	SD	*Recommended portions	P values
Cereals and millets	127.2	46.65	270	18.08
Pulses and legumes	44.65	28.24	60	3.29
Dairy products	131.5	10.65	300	9.26
Green leafy vegetables	38.53	47.55	100	7.65
Roots and tubers	58.82	55.19	200	15.13
Other vegetables	56.15	67.88	100	3.82
Oil	27.43	20.41	20	[NS]2.15
Sugar	8.43	10.34	20	6.62

* RDA for sedentary worker healthy adult women.; Source: Dietary Guidelines for Indians- A Manual, NIN-ICMR, Hyderabad, 2010.
NS- No significant difference at 5% level of significance.

Table 5: Percentage of women participants having a different degree of anemia (n=57).

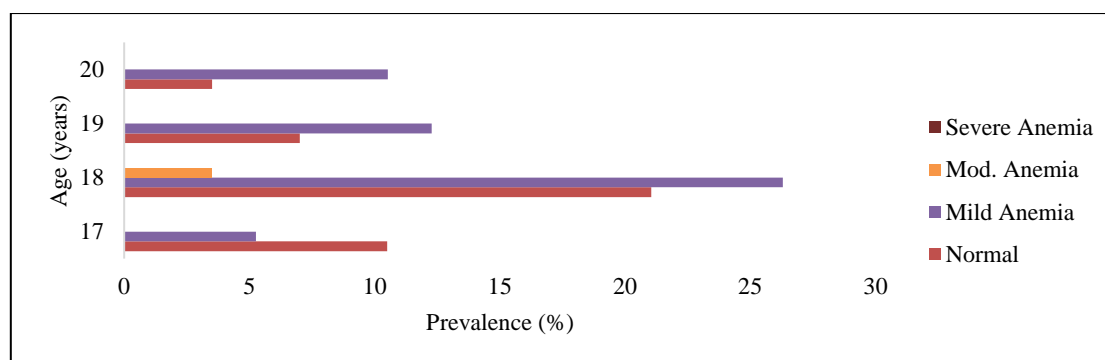| Age in years	Normal N (%)	Mild N (%)	Moderate N (%)	Severe N (%)
17	6 (10.52)	3 (5.26)	0	0
18	12 (21.05)	15 (26.31)	2 (3.51)	0
19	4 (7.02)	7 (12.28)	0	0
20	2 (3.51)	6 (10.53)	0	0
Total	24 (42.1)	31 (54.38)	2 (3.51)	0

Figure 1: Comparison of status of anaemia at different age groups.

DISCUSSION

The present study confirms that the rate of malnutrition was found to be very high among young women belonging to well to do families in Hyderabad city, TS, India. The data obtained revealed that 2.8% of participants were having antacids occasionally, 4.9% were having hormonal disturbances like hypothyroidism and PCOD. 11.2% were taking calcium supplements and 3.9% were using iron supplements as prophylactic measures prescribed by the physician. 26.8% of participants were having a history of irregular menses or oligomenorrhoea.

Anthropometric measurements data revealed that there is a significant difference in height of 17, 18 and 19-year-old women at 5% level of significance. Whereas only 17-year-old girls were found to be a deficit in weight when compared to ICMR reference values given in NNMB report, 2002(data pooled from 16 strata). BMI measurements results indicated that 87.7% of participants were maintaining desirable body weight, whereas only 12.28% of participants were categorized as underweight, none of the participant's falls under the category of overweight and obese.

Evaluation of hemoglobin levels indicated that 54.38 and 3.51 per cent women were having mild anemia and moderate anemia respectively, remaining 42.1 per cent women were having normal hemoglobin levels. None of the participants had hemoglobin levels <8 g/dl to be considered as severe anemic.

Dietary data procured from questionnaire revealed that 97% participants were non-vegetarian, of total 17% and 80% participants were consuming at least one serving of non-veg food daily and weekly respectively in the form of red meat, chicken and egg. Consumption of fish is found to be less i.e. only 5.2% and 42% of participants reported to have fish weekly and monthly respectively. Consumption of caffeinated beverages was reported to be >150 ml daily in 91.1% and about 200 ml daily in 8.8% participants. Around 40% of participants were reported to consume 20-30 g medium-dark chocolates daily, whereas only 11.4% reported consuming more than 50g chocolate daily. 56.1% of participants were incorporating >100 g cereal grain daily in their diet whereas 43.9 per cent were able to include <100 g grains in their diet.

Analysis of 24-hour diet recall also concluded that mean dietary intake of greens and other vegetables were far below than the recommendations. The consumption of pulses and the dairy product was found to be low and there was a significant difference at p>0.005 between the intake and recommendations made by ICMR (Table 4), also the protein in terms of quantity and quality is insufficient in the diet of most of the women. India being agricultural country accounts 11.2 % of total global vegetable production and are available in the local market in all seasons and vegetables are considered as an integral part of almost all preparation in all Indian cuisines across the country yet the mean intake of green leafy vegetables which is considered to be an important source of iron especially is also to found to be less i.e 38.53±47.55 g than the ICMR recommendations of 100g/day.[13]

Community nutrition approaches may help to overcome malnutrition at various levels and decrease overall disease burden among women of different section. Awareness and education related to nutrition adequacy will produce a positive approach in given communities. Skill-based education at high school level addressing health issues in the curriculum may help to promote positive behaviour amongst students.

Limitations

The present study is limited in terms of the small sample size thus for better representation of prevalence of anemia sample size need to be vast enough. There is also a need to develop easy, reliable and less expensive method by which iron deficiency can be diagnosed at early stages as hemoglobin count reduces at later stages only. Methods like serum ferritin test should be encouraged at community level for early case findings.

CONCLUSION

It was concluded that young women in their reproductive years are susceptible to anaemia, nutritional anaemia predisposes not only because of dietary inadequacy of iron and other nutrients but gut acidity or pH and food toxicants like soy proteins, tannins, phytic acid and inclusion of excessive fiber and calcium in the diet. These inhibitors bound to non-heame iron and reduce its uptake by the body.[14] More research and understanding of the regulation of iron and its absorption in the gut will help to come with newer strategies to eliminate Anaemia globally. Anemia can be prevented by food based strategies and by supplementation, fortification and education.

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