

**Original article:**

## **Evaluation of Prevalence of Cobalamine and Folate Deficiency Amongst Adolescents: An Institutional Based Study**

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Date of submission: 25 October 2012, Date of acceptance: 14 December 2012

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### **Abstract**

**Background:** Adolescence is a period of rapid physical growth and development accompanied by increased nutritional requirements. Cobalamin (vitamin B12) and folate are essential micronutrients involved in DNA synthesis, erythropoiesis, neurological function, and cellular metabolism. Deficiencies of these vitamins may adversely affect growth, cognitive performance, and hematological status. Despite their clinical significance, data regarding the burden of cobalamin and folate deficiency among adolescents remain limited, particularly in developing countries. Therefore, assessment of the prevalence and associated factors of these deficiencies is important for planning preventive nutritional interventions.

**Aim:** To evaluate prevalence of cobalamine and folate deficiency amongst adolescents.

**Materials and Methods:** This survey-based cross-sectional observational study was conducted among 135 adolescents aged 10–19 years. Data was collected using a predesigned and pretested questionnaire that included demographic characteristics, dietary habits, and relevant clinical information. Anthropometric measurements including height, weight, and body mass index (BMI) were recorded. Venous blood samples were collected for estimation of serum cobalamin and folate levels using a chemiluminescent microparticle immunoassay. Hematological parameters including hemoglobin, mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), and mean corpuscular hemoglobin concentration (MCHC) were analyzed.

**Results:** 52 (38.52%) had cobalamin deficiency and 33 (24.44%) had folate deficiency. Isolated cobalamin deficiency was observed in 34 (25.19%) participants, isolated folate deficiency in 15 (11.11%), and combined deficiency in 18 (13.33%). Cobalamin deficiency was significantly higher among vegetarians compared to adolescents consuming a mixed diet (53.45% vs. 27.27%,  $p=0.002$ ) and among underweight participants compared to normal/overweight participants (52.94% vs. 33.66%,  $p=0.043$ ). Folate deficiency was significantly associated with inadequate green leafy vegetable intake (35.29% vs. 13.43%,  $p=0.006$ ) and underweight BMI status (38.24% vs. 19.80%,  $p=0.044$ ). Adolescents with vitamin deficiencies had significantly lower hemoglobin levels and altered red cell indices compared with non-deficient participants ( $p<0.05$ ).

**Conclusion:** Cobalamin and folate deficiencies are common among adolescents, with cobalamin deficiency being more prevalent. Dietary practices and nutritional status play a significant role in the occurrence of these deficiencies. Routine nutritional assessment, dietary education, and early screening programs are recommended to improve adolescent micronutrient status and prevent associated hematological complications.

**Key words:** Adolescents, Cobalamin Deficiency, Folate Deficiency, Vitamin B12, Nutritional Status.

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### **INTRODUCTION**

Adolescence is a critical transition from childhood to adulthood, characterized by rapid growth,

expansion of blood volume, hormonal change, organ maturation and continuing brain development. These processes increase

micronutrient requirements. At the same time, adolescents commonly skip meals, consume energy-dense foods and become more independent in their food choices. Nutritional deficiencies may therefore develop even in apparently healthy individuals. Cobalamin and folate deserve particular attention because both participate in essential metabolic pathways, and early deficiency may remain unrecognized until hematological, neurological or functional disturbances appear. Biochemical assessments of school-aged populations have consequently included serum folate, vitamin B12 and homocysteine as indicators of micronutrient status.<sup>1</sup> Cobalamin, or vitamin B12, is a water-soluble vitamin required for DNA synthesis, red blood cell production, maintenance of myelin and methylation reactions. It acts as a cofactor for methionine synthase and methylmalonyl-coenzyme A mutase, contributing to homocysteine conversion and amino-acid metabolism. Natural dietary sources are predominantly foods of animal origin, including meat, fish, eggs and milk products. Adolescents following vegetarian or highly restricted diets may therefore have inadequate intake unless fortified foods or supplements are consumed. Food-related behaviours, meal preparation practices and the quality of foods available at home influence adolescent nutrient intake and should be considered when evaluating deficiency risk.<sup>2</sup> Folate is another water-soluble B vitamin required for one-carbon transfer reactions, nucleotide synthesis, cell division and tissue growth. Its importance is evident during rapid cellular proliferation, making adolescence a vulnerable period. Green leafy vegetables, legumes, fruits, nuts and fortified cereals are major sources. Inadequate consumption, cooking losses, food insecurity, restrictive eating and increased requirements may contribute to poor folate status. Cobalamin and folate metabolism are

closely interconnected; deficiency of either vitamin can impair methionine synthesis, disturb DNA formation and increase circulating homocysteine. Research involving adolescents has therefore assessed both vitamins together rather than treating them as unrelated nutritional factors.<sup>3</sup> Deficiency of cobalamin or folate may cause ineffective erythropoiesis and megaloblastic changes. Adolescents may present with fatigue, weakness, pallor, reduced exercise tolerance, poor appetite, glossitis or difficulty concentrating. Laboratory abnormalities can include reduced hemoglobin, macrocytosis, raised mean corpuscular volume and altered red-cell indices. However, iron deficiency, hemoglobin disorders and inflammation can modify or mask the typical blood picture. Cobalamin deficiency may additionally produce paresthesia, gait disturbance, impaired memory and mood changes, sometimes before marked anemia develops. Their role in homocysteine metabolism also provides a possible link between adolescent micronutrient status and later cardiometabolic health. Indian adolescent research has highlighted the value of examining dietary factors with biochemical and cardiovascular risk indicators.<sup>4</sup> Assessment requires careful interpretation. Serum cobalamin and serum folate are commonly used for survey-based screening, but concentrations may be influenced by recent intake, laboratory method and the diagnostic threshold. Complete blood count and red-cell indices demonstrate hematological effects but cannot independently identify the deficient vitamin. Functional markers such as methylmalonic acid and total homocysteine can improve interpretation in selected cases. Homocysteine is influenced by vitamin status and by age, sex, renal function, lipid profile and genetic variation in one-carbon metabolism. Adolescent studies examining folate, cobalamin, lipids and methylenetetrahydrofolate reductase variants show

why biochemical findings should be interpreted within a broader clinical context.<sup>5</sup> The consequences of suboptimal cobalamin and folate status may extend beyond anemia. Adolescence is a period of educational, psychological and social development, and persistent fatigue, poor attention or neuropsychological symptoms may interfere with school participation and daily functioning. The relationship between B-vitamin intake and mental well-being has therefore become an area of nutritional investigation. Nevertheless, nonspecific symptoms cannot confirm deficiency, and biochemical testing remains essential. Early detection permits dietary counselling, recognition of restrictive eating patterns, investigation of underlying medical conditions and appropriate supplementation.<sup>6</sup>

## **MATERIALS & METHODS**

This survey-based, cross-sectional observational study was conducted among adolescents to evaluate prevalence of cobalamin and folate deficiency amongst adolescents and associated demographic, dietary, and hematological parameters among the study participants. Data collection was carried out using a structured methodology to ensure uniformity and reliability of the information obtained. A total of 135 adolescents were enrolled in the study. Participants were selected from adolescents visiting hospital for routine health check-ups, nutritional assessment, minor illnesses, or preventive healthcare services. Adolescents belonging to both sexes and within the age group of 10–19 years were included in the study. Participants who provided assent and whose parents or legal guardians provided informed consent were recruited. Adolescents with known chronic systemic illnesses, hematological disorders, severe acute infections, malignancies, chronic liver or kidney diseases, those receiving vitamin B12 or

folate acid supplementation, and those unwilling to participate were excluded from the study.

## **Methodology**

The study included 135 adolescents selected through a consecutive sampling technique. Eligible participants presenting to the study setting during the data collection period were approached and invited to participate. Recruitment continued until the desired sample size was achieved. This method ensured adequate representation of adolescents attending the tertiary care facility and facilitated the assessment of cobalamin and folate deficiency prevalence within the study population.

## **Data Collection Procedure**

Data were collected using a predesigned and pretested structured questionnaire administered by trained investigators. Information regarding socio-demographic characteristics, including age, sex, educational status, residential background, and socioeconomic status, was obtained. Detailed dietary history was recorded with special emphasis on the frequency of consumption of animal-source foods, dairy products, green leafy vegetables, fruits, legumes, and fortified foods. Information regarding meal patterns, dietary habits (vegetarian or mixed diet), history of nutritional supplementation, and relevant medical history was also documented. Anthropometric measurements including height, weight, and body mass index (BMI) were recorded using standardized techniques and calibrated instruments.

## **Clinical and Laboratory Assessment**

All participants underwent a clinical evaluation to identify signs and symptoms suggestive of nutritional deficiencies, including fatigue, generalized weakness, pallor, glossitis, recurrent infections, paresthesia, and cognitive or concentration difficulties. Venous blood samples were collected under aseptic precautions for laboratory investigations. Hematological

assessment included complete blood count (CBC) with estimation of hemoglobin concentration, total leukocyte count, platelet count, mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), and mean corpuscular hemoglobin concentration (MCHC). Peripheral blood smear examination was performed where indicated to assess red blood cell morphology.

#### **Estimation of Serum Cobalamin and Folate Levels**

Serum cobalamin (vitamin B12) and serum folate concentrations were measured using a standardized chemiluminescent microparticle immunoassay method in the hospital's central clinical laboratory following the manufacturer's protocol. Internal quality control procedures were maintained throughout the study to ensure analytical accuracy and precision. Vitamin B12 deficiency was defined as a serum cobalamin concentration of less than 200 pg/mL, while folate deficiency was defined as a serum folate concentration of less than 4 ng/mL. Participants with values above these thresholds were categorized as non-deficient.

#### **Statistical Analysis**

The collected data were entered into Microsoft Excel and analyzed using Statistical Package for the Social Sciences (SPSS) software. Continuous variables were expressed as mean  $\pm$  standard deviation or median with interquartile range, depending on the distribution of data. Categorical variables were presented as frequencies and percentages. The prevalence of cobalamin and folate deficiency was calculated as proportions with corresponding percentages. Associations between categorical variables were assessed using the Chi-square test or Fisher's exact test, while continuous variables were compared using Student's t-test or Mann-Whitney U test as appropriate. A p-value of less than 0.05 was considered statistically significant.

#### **RESULTS**

Present study included 135 adolescents attending a tertiary care hospital. As shown in Table 1, the majority of participants belonged to the 14–16 years age group, comprising 56 adolescents (41.48%), followed by 41 participants (30.37%) in the 17–19 years age group and 38 participants (28.15%) in the 10–13 years age group. Females formed a slightly higher proportion of the study population, with 73 participants (54.07%), while males accounted for 62 participants (45.93%). Most participants were from urban areas, accounting for 79 adolescents (58.52%), whereas 56 participants (41.48%) were from rural areas.

Regarding dietary pattern, 77 adolescents (57.04%) consumed a mixed diet, while 58 participants (42.96%) were vegetarian. Assessment of nutritional status showed that most adolescents had normal BMI, with 82 participants (60.74%) falling in this category. Underweight status was observed in 34 participants (25.19%), while 19 adolescents (14.07%) were overweight or obese.

As shown in Table 2, cobalamin deficiency only was observed in 34 adolescents (25.19%), while folate deficiency only was present in 15 participants (11.11%). Both cobalamin and folate deficiency were found in 18 adolescents (13.33%). No deficiency was observed in 68 participants (50.37%). Overall, total cobalamin deficiency, including isolated and combined deficiency, was present in 52 adolescents (38.52%), while total folate deficiency was observed in 33 adolescents (24.44%).

Table 3 shows the association of cobalamin deficiency with selected variables. Cobalamin deficiency was highest in the 14–16 years age group, where 24 participants (42.86%) were deficient, followed by 16 participants (39.02%) in the 17–19 years group and 12 participants (31.58%) in the 10–13 years group. However, this difference

was not statistically significant ( $p=0.381$ ). Female participants showed a higher proportion of cobalamin deficiency, with 31 females (42.47%) affected compared to 21 males (33.87%), but this association was also not statistically significant ( $p=0.301$ ).

A statistically significant association was observed between dietary pattern and cobalamin deficiency. Among vegetarian adolescents, 31 participants (53.45%) were cobalamin deficient compared to 21 participants (27.27%) among those consuming a mixed diet ( $p=0.002$ ). This suggests that vegetarian dietary habits were significantly associated with a higher prevalence of cobalamin deficiency. BMI status was also significantly associated with cobalamin deficiency. Among underweight adolescents, 18 participants (52.94%) were deficient, whereas among normal/overweight adolescents, 34 participants (33.66%) were deficient ( $p=0.043$ ).

As shown in Table 4, folate deficiency was assessed according to age, sex, green leafy vegetable intake, and BMI status. Folate deficiency

was seen in 8 participants (21.05%) in the 10–13 years age group, 15 participants (26.79%) in the 14–16 years age group, and 10 participants (24.39%) in the 17–19 years age group. The difference across age groups was not statistically significant ( $p=0.731$ ). Similarly, folate deficiency was observed in 14 males (22.58%) and 19 females (26.03%), showing no statistically significant association with sex ( $p=0.659$ ). Green leafy vegetable intake showed a statistically significant association with folate deficiency. Among adolescents with inadequate intake of green leafy vegetables, 24 participants (35.29%) were folate deficient, compared to only 9 participants (13.43%) among those with adequate intake ( $p=0.006$ ). This finding indicates that inadequate consumption of green leafy vegetables was significantly associated with increased folate deficiency. BMI status was also significantly associated with folate deficiency, as 13 underweight adolescents (38.24%) were folate deficient compared to 20 normal/overweight adolescents (19.80%) ( $p=0.044$ ), suggesting that undernutrition may contribute to folate deficiency.

**Table 1: Socio-demographic profile of study participants**

Variable	Category	Number (n=135)	Percentage (%)
Age group	10–13 years	38	28.15
	14–16 years	56	41.48
	17–19 years	41	30.37
Sex	Male	62	45.93
	Female	73	54.07
Residence	Urban	79	58.52
	Rural	56	41.48
Dietary pattern	Vegetarian	58	42.96
	Mixed diet	77	57.04
BMI status	Underweight	34	25.19
	Normal	82	60.74
	Overweight/obese	19	14.07

**Table 2: Prevalence of cobalamin and folate deficiency**

Vitamin status	Number (n=135)	Percentage (%)
Cobalamin deficiency only	34	25.19
Folate deficiency only	15	11.11
Both cobalamin and folate deficiency	18	13.33
No deficiency	68	50.37
Total cobalamin deficiency	52	38.52
Total folate deficiency	33	24.44

**Table 3: Association of cobalamin deficiency with selected variables**

Variable	Category	Cobalamin deficient n (%)	Not deficient n (%)	p-value
Age group	10–13 years	12 (31.58)	26 (68.42)	0.381
	14–16 years	24 (42.86)	32 (57.14)	
	17–19 years	16 (39.02)	25 (60.98)	
Sex	Male	21 (33.87)	41 (66.13)	0.301
	Female	31 (42.47)	42 (57.53)	
Dietary pattern	Vegetarian	31 (53.45)	27 (46.55)	0.002
	Mixed diet	21 (27.27)	56 (72.73)	
BMI status	Underweight	18 (52.94)	16 (47.06)	0.043
	Normal/overweight	34 (33.66)	67 (66.34)	

**Table 4: Association of folate deficiency with selected variables**

Variable	Category	Folate deficient n (%)	Not deficient n (%)	p-value
Age group	10–13 years	8 (21.05)	30 (78.95)	0.731
	14–16 years	15 (26.79)	41 (73.21)	
	17–19 years	10 (24.39)	31 (75.61)	
Sex	Male	14 (22.58)	48 (77.42)	0.659
	Female	19 (26.03)	54 (73.97)	
Green leafy vegetable intake	Inadequate	24 (35.29)	44 (64.71)	0.006
	Adequate	9 (13.43)	58 (86.57)	
BMI status	Underweight	13 (38.24)	21 (61.76)	0.044
	Normal/overweight	20 (19.80)	81 (80.20)	

**Table 5: Hematological parameters according to vitamin deficiency status**

Parameter	Deficient group Mean ± SD	Non-deficient group Mean ± SD	p-value
Hemoglobin (g/dL)	10.84 ± 1.21	12.31 ± 1.04	<0.001
MCV (fL)	92.46 ± 8.37	84.28 ± 6.91	<0.001
MCH (pg)	30.18 ± 3.42	27.64 ± 2.96	0.002
MCHC (g/dL)	32.11 ± 1.78	33.02 ± 1.52	0.018
Serum cobalamin (pg/mL)	156.73 ± 32.85	318.46 ± 76.92	<0.001
Serum folate (ng/mL)	3.21 ± 0.62	6.84 ± 1.91	<0.001

Table 5 presents hematological parameters according to vitamin deficiency status. The mean hemoglobin level was lower in the deficient group ( $10.84 \pm 1.21$  g/dL) compared to the non-deficient group ( $12.31 \pm 1.04$  g/dL), and this difference was statistically significant ( $p < 0.001$ ). Mean corpuscular volume was higher in the deficient group ( $92.46 \pm 8.37$  fL) than in the non-deficient group ( $84.28 \pm 6.91$  fL), also showing a significant difference ( $p < 0.001$ ).

The mean MCH was significantly higher in the deficient group ( $30.18 \pm 3.42$  pg) compared with the non-deficient group ( $27.64 \pm 2.96$  pg) ( $p = 0.002$ ). Mean MCHC was slightly lower in the deficient group ( $32.11 \pm 1.78$  g/dL) than in the non-deficient group ( $33.02 \pm 1.52$  g/dL), with a statistically significant difference ( $p = 0.018$ ). Serum cobalamin levels were markedly lower in the deficient group ( $156.73 \pm 32.85$  pg/mL) compared to the non-deficient group ( $318.46 \pm 76.92$  pg/mL), and serum folate levels were also lower in the deficient group ( $3.21 \pm 0.62$  ng/mL) compared to the non-deficient group ( $6.84 \pm 1.91$  ng/mL), with both differences being highly statistically significant ( $p < 0.001$ ).

## DISCUSSION

In the present study, most adolescents were in the 14–16 years age group (41.48%), with slight female predominance (54.07%) and more urban participants (58.52%). Normal BMI was observed in 60.74%, while 25.19% were underweight. Rao et al. (2006), in a large study of 12,789 tribal adolescents aged 10–17 years from nine Indian states, reported much higher undernutrition, with about 63.00% of boys and 42.00% of girls being undernourished. Compared with their findings, the present study showed lower undernutrition, but the 25.19% underweight burden still indicates significant nutritional vulnerability among

adolescents.<sup>7</sup> The overall prevalence of cobalamin deficiency in the present study was 38.52%, while folate deficiency was 24.44%; isolated cobalamin deficiency was 25.19%, isolated folate deficiency was 11.11%, and combined deficiency was 13.33%.

Mehta et al. (2007), among 272 young female students with median age of 16 years, reported vitamin B12 deficiency in 48.90% and folic acid deficiency in 12.50%. Thus, cobalamin deficiency in the present study was lower than that reported by Mehta et al., whereas folate deficiency was higher, suggesting variation in dietary intake, socioeconomic profile, and study population.<sup>8</sup> In the present study, cobalamin deficiency was more common than folate deficiency, with total cobalamin deficiency affecting 38.52% compared with total folate deficiency in 24.44%. VanderJagt et al. (2000), in adolescent girls from northern Nigeria, also reported that the participants were at greater risk for vitamin B12 deficiency than folate deficiency; around 9.00% had low serum vitamin B12, while only 2.40% had low serum folate. The pattern was therefore similar to the present study, although the magnitude of both deficiencies was much higher in the present hospital-based sample.<sup>9</sup> Age-wise, cobalamin deficiency in the present study was highest among adolescents aged 14–16 years (42.86%), followed by 17–19 years (39.02%) and 10–13 years (31.58%), but this association was not statistically significant ( $p = 0.381$ ). Louwman et al. (2000) studied adolescents aged 10–16 years and found that 31 of 48 adolescents with previous macrobiotic dietary exposure were cobalamin deficient based on methylmalonic acid concentration. Their study emphasized that cobalamin deficiency may occur during adolescence and may have functional effects even without obvious hematological signs, supporting the importance of biochemical screening in the

present study.<sup>10</sup> Dietary pattern showed a significant association with cobalamin deficiency in the present study, as 53.45% of vegetarian adolescents were deficient compared with 27.27% of those consuming a mixed diet ( $p=0.002$ ).

Yajnik et al. (2006), in a community-based Indian study, reported low vitamin B12 levels in 67.00% of men and found that lacto-vegetarians had 4.4 times higher risk of low vitamin B12 concentration compared with frequent non-vegetarians. This strongly supports the present finding that vegetarian dietary habits are an important contributor to cobalamin deficiency.<sup>11</sup>

Folate deficiency in the present study was significantly associated with inadequate green leafy vegetable intake, with deficiency present in 35.29% of adolescents with inadequate intake compared with 13.43% among those with adequate intake ( $p=0.006$ ).

Öner et al. (2006), among adolescent girls in Edirne, Turkey, reported folic acid deficiency in 16.30% and marginal folate status in 46.00%, with low folic acid intake being an important predictor of deficiency. Compared with their study, the present study showed a higher folate deficiency burden, and the significant association with inadequate green leafy vegetable intake supports the dietary basis of folate deficiency.<sup>12</sup>

The present study also showed that underweight adolescents had significantly higher cobalamin deficiency (52.94%) compared with normal/overweight adolescents (33.66%;  $p=0.043$ ), and higher folate deficiency (38.24% versus 19.80%;  $p=0.044$ ).

Refsum et al. (2001), in Asian Indians, reported cobalamin deficiency in 47.00%, low holotranscobalamin in 73.00%, hyperhomocysteinemia in 77.00%, and elevated methylmalonic acid in 73.00%, while folate deficiency was rare. Although their study was not

limited to adolescents, it supports the concept that cobalamin deficiency is highly prevalent in Indian populations and may coexist with broader nutritional inadequacy, as seen among underweight adolescents in the present study.<sup>13</sup>

Hematological findings in the present study showed significantly lower hemoglobin in the deficient group than the non-deficient group ( $10.84 \pm 1.21$  g/dL versus  $12.31 \pm 1.04$  g/dL;  $p<0.001$ ), higher MCV ( $92.46 \pm 8.37$  fL versus  $84.28 \pm 6.91$  fL;  $p<0.001$ ), and lower serum cobalamin and folate levels ( $p<0.001$ ).

Khanduri et al. (2007), in patients with megaloblastic anemia, found cobalamin deficiency in 65.00%, combined cobalamin and folate deficiency in 12.00%, and pure folate deficiency in 6.00%, with peak incidence in the 10–30 years age group. The combined deficiency in their study was close to the present study's combined deficiency rate of 13.33%, and both studies show that cobalamin deficiency has important hematological consequences.<sup>14</sup>

## CONCLUSION

The present study demonstrated a high prevalence of cobalamin deficiency (38.52%) and folate deficiency (24.44%) among adolescents attending a tertiary care hospital, with cobalamin deficiency being more common. Vegetarian dietary habits, inadequate intake of green leafy vegetables, and underweight nutritional status were significantly associated with these deficiencies. Adolescents with vitamin deficiencies also exhibited significant alterations in hematological parameters, including lower hemoglobin levels and changes in red cell indices. These findings highlight the need for routine nutritional screening, dietary counseling, and early intervention strategies to prevent micronutrient deficiencies and their associated health consequences during adolescence.

## References

1. Papandreou D, Mavromichalis I, Makedou A, Rousso I, Arvanitidou M. Reference range of total serum homocysteine level and dietary indexes in healthy Greek schoolchildren aged 6–15 years. *Br J Nutr.* 2006;96(4):719–724. doi: 10.1079/BJN20061882.
2. Larson NI, Story M, Eisenberg ME, Neumark-Sztainer D. Food preparation and purchasing roles among adolescents: associations with sociodemographic characteristics and diet quality. *J Am Diet Assoc.* 2006;106(2):211–218. doi: 10.1016/j.jada.2005.10.029.
3. Thomas NE, Cooper SM, Baker JS, Graham MR, Davies B. Homocyst(e)ine, folate, and vitamin B12 status in a cohort of Welsh young people aged 12–13 years old. *Res Sports Med.* 2008;16(4):233–243. doi: 10.1080/15438620802310826.
4. Anand P, Awasthi S, Mahdi A, Tiwari M, Agarwal GG. Serum homocysteine in Indian adolescents. *Indian J Pediatr.* 2009;76(7):705–709. doi: 10.1007/s12098-009-0116-z.
5. Gil-Prieto R, Hernández V, Cano B, Oya M, Gil A. Plasma homocysteine in adolescents depends on the interaction between methylenetetrahydrofolate reductase genotype, lipids and folate: a seroepidemiological study. *Nutr Metab (Lond).* 2009;6:39. doi: 10.1186/1743-7075-6-39.
6. Murakami K, Miyake Y, Sasaki S, Tanaka K, Arakawa M. Dietary folate, riboflavin, vitamin B-6, and vitamin B-12 and depressive symptoms in early adolescence: the Ryukyus Child Health Study. *Psychosom Med.* 2010;72(8):763–768. doi: 10.1097/PSY.0b013e3181f02f15.
7. Rao KM, Balakrishna N, Laxmaiah A, Venkaiah K, Brahmam GNV. Diet and nutritional status of adolescent tribal population in nine States of India. *Asia Pac J Clin Nutr.* 2006;15(1):64–71.
8. Mehta BC, Kabeer RM, Patel Y. Prevalence of hematinics deficiency amongst female students and its correction. *Indian J Hematol Blood Transfus.* 2007;23(3–4):88–91. doi: 10.1007/s12288-008-0014-y.
9. VanderJagt DJ, Spelman K, Ambe J, Datta P, Blackwell W, Crossey M, et al. Folate and vitamin B12 status of adolescent girls in northern Nigeria. *J Natl Med Assoc.* 2000;92(7):334–340.
10. Louwman MW, van Dusseldorp M, van de Vijver FJ, Thomas CM, Schneede J, Ueland PM, et al. Signs of impaired cognitive function in adolescents with marginal cobalamin status. *Am J Clin Nutr.* 2000;72(3):762–769. doi: 10.1093/ajcn/72.3.762.
11. Yajnik CS, Deshpande SS, Lubree HG, Naik SS, Bhat DS, Uradey BS, et al. Vitamin B12 deficiency and hyperhomocysteinemia in rural and urban Indians. *J Assoc Physicians India.* 2006;54:775–782.
12. Öner N, Vatansever U, Karasalihoğlu S, Ekuklu G, Celtik C, Biner B. The prevalence of folic acid deficiency among adolescent girls living in Edirne, Turkey. *J Adolesc Health.* 2006;38(5):599–606. doi: 10.1016/j.jadohealth.2005.03.027.
13. Refsum H, Yajnik CS, Gadkari M, Schneede J, Vollset SE, Orning L, et al. Hyperhomocysteinemia and elevated methylmalonic acid indicate a high prevalence of cobalamin deficiency in Asian Indians. *Am J Clin Nutr.* 2001;74(2):233–241. doi: 10.1093/ajcn/74.2.233.
14. Khanduri U, Sharma A. Megaloblastic anaemia: prevalence and causative factors. *Natl Med J India.* 2007;20(4):172–175.