

Full Length Research Paper

Nutritional status in school children: Deficiencies in iron, folic acid and Vitamin B₁₂

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This study was conducted to assess deficiencies in iron, folic acid and Vitamin B₁₂ as well as nutritional status in a sample of school-age children in Turkey. The study was conducted on 172 children (boys: 80; girls: 92) aged 6 to 13 years from two different primary schools in the capital city of Turkey, Ankara. Data were obtained on children anthropometry, 24 dietary recalls, hematological and biochemical parameters of vitamin and iron bioavailability. The prevalence of deficiencies in iron, folic acid and Vitamin B₁₂ were 15.7, 0.6 and 3.5%, respectively. In this study 18% of children were determined underweight or risk of underweight and 15.2% of children were determined overweight or obese. Body weight measurements for age were significantly low in children who had iron, folic acid or Vitamin B₁₂ deficiency ($p < 0.01$). Intake of energy was inadequate in 38% of the subjects, while iron and Vitamin B₁₂ intake were inadequate in 29 and 58%, respectively. As a result of this study many dietary deficiencies especially for iron, are still common and vitamin B₁₂ deficiency rather than folic acid deficiency was seemed to be the more prominent in Turkish grade school children.

Key words: School-aged children, folic acid, vitamin B₁₂, iron, anemia.

INTRODUCTION

Dietary deficiencies of vitamins and minerals are commonly seen in developed countries and are also an important public health concern in developing countries. These deficiencies negatively affect human health and economic progress of the country, lead to important public health concerns, and result in manpower and economic losses. Despite the fact that vitamin and mineral deficiencies are found in all age groups, young children and women of reproductive age are at higher risk (UNICEF and Micronutrient Initiative, 2004; Micronutrient Initiative, 2009). Children in the age group of 6 to 14 years are specified as school-aged in Turkey, and comprise a large part of the total population (17.2%) according to the Turkish- 2010- Address-Based Population Registration System (Turkey Statistical

Institute, 2011). This age is a period of rapid growth and development and is a time during which lifelong behaviors are formed (Feigelman, 2007; Pekcan et al., 2004). The primary reasons for deficiencies of iron, Vitamin B₁₂ and folic acid in childhood are that they are not sufficiently present in the diet. In addition, the prevention of absorption of vitamins and minerals in foods by various reasons may also lead to deficiencies of these substances (Rasmussen et al., 2001; Zimmermann et al., 2007).

Iron deficiency is a common nutritional deficiency in Turkey and in other countries and is an important health concern for infants, adolescents, pregnant women, and persons living at a low socioeconomic level, especially in developing countries (Zimmermann et al., 2007; Merovitch et al., 2006; Schneider et al., 2005). According to the World Health Organization (WHO), iron deficiency anemia (IDA) is seen in 36% of the population in developing countries and in 8% of the population in developed countries (Mamiro et al., 2005; Siegel et al.,

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2006; Khusun et al., 1999). Iron deficiency (ID) and IDA in Turkey are more prevalent than in developed countries. This rate is reported to be between 15.2 and 62.5% in studies performed in Turkey. Additionally, IDA comprises 48 to 75% of anemia's in infants (Yalcin et al., 2008). Anaemia decreases the ability to study and learn and negatively impacts on labor capacity. It especially impairs the health of mothers and children, increases the mortality rate and decreases resistance to diseases (UNICEF and Micronutrient Initiative, 2004; Micronutrient Initiative, 2009; Zimmermann et al., 2007). The most frequent causes of megaloblastic anemia in children are deficiencies in vitamin B₁₂ and folic acid. Folic acid and vitamin B₁₂ play a role in the synthesis of DNA in cells of the gastrointestinal, urogenital, and nervous system as well as in hematopoietic cells (Rasmussen et al., 2001). It has been suggested that iron deficiency is an important problem in school-aged children in various studies performed in Turkey (Açkurt et al., 1995; Keskin et al., 2005; Aydınok et al., 1998; Koçak et al., 1995; Koç et al., 2000; Kılınç et al., 2002). However, there is insufficient knowledge regarding the frequency of vitamin B₁₂ and folic acid deficiencies (Koç et al., 2005). The purpose of this study was to assess the nutritional status of school-aged children with low socioeconomic status in regards to dietary deficiencies in iron, folic acid, and vitamin B₁₂.

The research questions were:

1. What is prevalence of iron, vitamin B₁₂ and folic acid deficiency and iron deficiency anemia in a sample of school-aged children in Ankara, Turkey?
2. Do gender differences occur in the mean of the hematological analysis?
3. What is the prevalence of underweight, overweight and obesity in this age group of children?
4. Do BMI for age classification results differ according to iron, folic acid or B₁₂ deficiencies?
5. Do dietary intake of energy and nutrients correlate with deficiencies?

METHODS

Sample and setting

This cross-sectional study was conducted in two different elementary schools in Ankara. Ankara is the capital city of Turkey and the country's second largest city after Istanbul. It is centrally located in Anatolia. From the data provided by the Ministry of National Education and the National Statistical Centre of Turkey one district within the metropolitan area of Ankara were identified. This data was also confirmed by the Municipality of Ankara. This district is inhabited by citizens with distinguished low SES and consequently this is reflected on the SES of the pupils attending the schools within the respected school zones. From the low SES district two public schools was selected randomly among all the public schools located in that zone.

A sample of 250 students between the ages of 7 and 11 years old receiving education in the primary section of the elementary school was selected by simple random sampling. The parents of all 250 students in these schools were invited to participate in the study through letters that were sent home with students, and only 1

child per family was eligible. One hundred seventy-two students (68%; 80 boys and 92 girls) returned permission slips from parents indicating a willingness to participate in the study. Human subjects' approval was obtained from the local ethics committee of the Medical Faculty of Gazi University (Ankara, Turkey).

Measures

Procedures

The children's gender, age, parental education, professional and marital status were assessed from a questionnaire administered during the initial interview with parents. The children's food consumption was evaluated by the 24 h dietary recall using the BeBiS 4 (Nutrition Information System) programme (BeBiS Nutrition Data Base, 2004). Measurements of body weight and height from anthropometric parameters were determined.

The hematological and biochemical parameters of bioavailability of vitamins and iron were measured in the laboratories of the Diskapi Yildirim Beyazit Education and Research Hospital and the Turkey Yuksek Ihtisas Research and Practice Hospital. A blood sample was taken from each participant after an overnight fast.

Dietary assessment

Dietary intake was evaluated as the percent of dietary recommended intake (DRI) for age and gender for specific nutrients and energy. The total daily energy intake of each child was assessed at the initial interview and was compared with the DRI for age and gender (Food and Nutrition Board, 2002). Children were considered to consume an inadequate-energy or nutrient diet if their total daily energy or nutrient intake was > 67% of the DRI and excessive energy or nutrient diet as an intake percentage of 133% greater than DRI (Serra-Majem et al., 2002).

Anthropometric measurements

Measurements of body weight and height from anthropometric parameters were determined with a height meter (SECA 767-220), and body mass index (BMI) was calculated by the formula:

$$\text{BMI} = \text{kg/m}^2 \text{ (Lohman et al., 1988)}$$

The BMI measurements were calculated based on WHO growth references (WHO, 2007), for children between 5 and 19 years of age.

Hematological analysis

Hemoglobin was estimated by Drabkin's cyanmethemoglobin test (Cook, 1985). Serum ferritin, vitamin B₁₂ and folate levels were measured in all patients with a retinal vein occlusion (RVO) (IMX Analyzer; Abbott Laboratories Diagnostics Division, Abbott Park, IL, USA). The IMX ferritin and B₁₂ assays are based on microparticle enzyme immunoassay (MEIA) technology, whereas the IMX folate assay uses an ion-capture assay technique. For these immunologic assays, inter- and intra-assay coefficients of variation were <10%.

Iron deficiency and iron deficiency anemia were defined using age- and sex-specific thresholds proposed by UNICEF (UNICEF Micronutrient Initiative, 2004) and WHO (WHO, 2001). Iron deficiency was defined as serum ferritin concentration <15 µg/L, indicating depleted iron stores, and iron deficiency anemia was defined as iron deficiency with hemoglobin concentration <11.5

Table 1. Mean and standard error of the mean (SEM) for age, body weight, height and BMI by gender of children.

	Boys (n:80)		Girls (n:92)		Total (n:172)	
	Mean	SEM	Mean	SEM	Mean	SEM
Age (year)	9.1	0.17	9.4	0.16	9.26	0.12
Height (cm)	133.4	1.04	133.5	1.08	133.5	0.75
Body weight (kg)	28.7	0.74	28.6	0.74	28.6	0.52
BMI (kg/m ²)	15.9	0.25	15.8	0.22	15.9	0.16

g/dL, which is the cutoff point for anemia for children aged 7 to 11 years old. Plasma vitamin concentrations were compared with published reference values to determine the proportion of children with a deficiency. Below-normal concentration of folic acid was defined as 3 ng/ml, and 100 pg/ml for Vitamin B₁₂ (Herbert et al., 1994).

Statistical analysis

All values are reported as the mean \pm standard error of mean (SEM). The data were analysed using the Statistical Package for Social Sciences Software for Windows 10.0 (SPSS Inc. Chicago, IL, USA). The Kolmogorov-Smirnov test was used to determine whether outcome variables were normally distributed. Tests for the difference between two means were used to compare age, anthropometric measurements, hematologic and biochemical values, and intake of energy and nutrients by gender. Pearson correlations coefficients were used to determine relationships between hematologic and biochemical parameters and the intake of energy and nutrients. In all analyses, 5% and 1% significance levels were used.

RESULTS

Regarding the education background of the parents of the study participants, 82.6% of the mothers and 56.4% of the fathers had an education level of primary school or less. In examining the professional status, 95.3% of the mothers and 55.2% of the fathers were housewives and independent businessmen, respectively.

Mean values for age, body weight, height, and BMI by gender are shown in Table 1. The mean age for boys and girls in the study (\pm SEM) was 9.1 ± 0.17 and 9.4 ± 0.16 years, respectively. There were no significant differences among values for age, body weight, height or BMI according to gender ($p > 0.05$).

The prevalence of iron deficiency (ID) (serum ferritin $< 15 \mu\text{g/L}$) was found to be 13.7% for boys and 17.4% for girls. The mean level of serum ferritin (\pm SEM) for boys and girls was found to be 33.3 ± 2.04 and $26.3 \pm 1.52 \mu\text{g/L}$, respectively. The mean hemoglobin levels for boys and girls were found to be 13.7 ± 0.11 and $13.5 \pm 0.11 \text{ g/dL}$, respectively, and the mean hematocrit levels for males and females were found to be 38.7 ± 0.01 and $38.4 \pm 0.28\%$, respectively. The prevalence of iron deficiency anemia (serum ferritin $< 15 \mu\text{g/L}$, concentration of hemoglobin $< 11.5 \text{ g/dL}$) was found to be 3.3% and the

prevalence of folic acid deficiency (serum folic acid $< 3 \text{ ng/ml}$) was found to be 1.1% in girls. Iron deficiency anemia and folic acid deficiency were not determined in boys. The prevalence of Vitamin B₁₂ deficiency (Serum vitamin B₁₂ $< 100 \text{ pg/ml}$) for boys and girls was found to be 3.8 and 3.3%, respectively (Table 2).

Mean serum folic acid concentration was found to be 10.4 ± 0.30 and $10.8 \pm 0.30 \text{ ng/ml}$ for boys and girls, respectively. Mean serum vitamin B₁₂ concentrations were found to be 357.1 ± 19.40 and $346.7 \pm 17.33 \text{ pg/ml}$ for boys and girls, respectively. The only statistically significant biochemical difference between boys and girls was the mean value for serum ferritin ($p < 0.05$).

In the evaluation of age-adjusted BMI, 18% were determined to be underweight or at risk of being underweight ($< 3\text{rd}$ or 3 to 15th percentile, respectively) and 15.2% were considered to be overweight or obese (85 to 97th or $\geq 97\text{th}$ percentile, respectively). The measurements of body weight were lower in the children with any anemia ($p < 0.05$). No difference was found between the percentile values of height and BMI according to age in children with any anemia when compared to healthy children ($p > 0.05$).

The mean intake of energy and nutrients according to gender of the study participants is shown in Table 3. No significant difference was found in nutrient intake between boys and girls ($p > 0.05$).

Daily energy and nutrient intake levels considered as inadequate ($< 67\%$) or over the recommended level ($< 133\%$) were evaluated by gender (Table 4). No significant differences were found between boys and girls and for inadequate and excessive intake levels of energy and nutrients when comparing children with or without anemia for these parameters ($p > 0.05$).

There was a significant positive correlation ($p < 0.05$) between levels of hemoglobin and hematocrit and the intake of energy, protein, and carbohydrates. Correlations were also found between serum vitamin B₁₂ levels and the intake of protein, animal protein and calcium, ferritin levels and the intake of animal protein and Vitamin B₁₂ as well as for serum folic acid concentration and the intake of folate. A statistically significant negative correlation ($p < 0.05$) was determined between the values of hemoglobin and hematocrit and dietary fat as well as for Serum vitamin B₁₂ levels and the intake of carbohydrate and dietary fibre (Table 5).

Table 2. The values of hematologic and biochemical parameters compared to the bioavailability of vitamins and iron in children.

Indicators	Boys		Girls		Total	
	n	%	n	%	n	%
Iron deficiency						
Ferritin (>15 µg/L)	69	86.3	76	82.6	145	84.3
Ferritin (<15 µg/L)	11	13.7	16	17.4	27	15.7
Iron deficiency anemia						
Ferritin(>15 µg/L), Hgb(>11.5 g/dl)	80	100.0	89	96.7	169	98.3
Ferritin(<15 µg/L), Hgb(<11.5 g/dl)	-	-	3	3.3	3	1.7
Folic acid deficiency						
Folic Acid >3 ng/ml	80	100.0	91	98.9	171	99.4
Folic Acid <3 ng/ml	-	-	1	1.1	1	0.6
Vitamin B₁₂ deficiency						
Vitamin B ₁₂ >100 pg/ml	77	96.2	89	96.7	166	96.5
Vitamin B ₁₂ <100 pg/ml	3	3.8	3	3.3	6	3.5

Table 3. The intake levels of energy and nutrients according to gender of the children.

	Boys (n:80)		Girls(n:92)	
	Mean	SEM	Mean	SEM
Energy (kcal)	1515.9	48.5	1577.7	50.83
Protein (g)	51.7	2.01	52.4	1.74
Plant protein (g)	28.5	1.20	28.8	1.02
Animal protein (g)	23.4	1.65	23.8	1.33
Fat (g)	58.1	2.06	61.1	2.21
Carbohydrate (g)	192.0	7.77	199.8	8.27
Dietary fiber (g)	18.2	0.70	17.6	0.63
Vitamin A (µg)	763.1	76.36	775.9	60.67
Vitamin B ₆ (mg)	1.04	0.005	1.0	0.004
Folate (µg)	269.5	12.4	261.0	10.06
Vitamin B ₁₂ (µg)	2.4	0.23	2.06	0.15
Vitamin C (mg)	82.6	7.43	76.7	5.68
Calcium (mg)	463.6	25.76	515.8	28.31
Magnesium (mg)	181.2	7.13	191.2	8.10
Iron (mg)	9.5	0.43	9.0	0.35
Zinc (mg)	7.3	0.33	6.9	0.25

DISCUSSION

The results of studies conducted to determine the iron deficiency shows that deficiency levels differ according to regions in Turkey. Turkey has seven regions, 4 side regions (Marmara, Aegean, Mediterranean and Black Sea) and 3 inner regions (Central Anatolia, Eastern Anatolia, Southeast Anatolia). In these regions nutritional status and habits vary significantly according to their climate, location, human habitat, agricultural diversities,

transportation etc. A study conducted on school-aged children between the ages of 12 and 13 living in three biggest city of Turkey determined a prevalence of 19.1% for iron deficiency and 3.9% for iron deficiency anemia (Keskin et al., 2005). Another study conducted on children between 6 to 12 age in one province of the Southeast Anatolia region the prevalence of ID and IDA was found 24.7 and 12.5%, respectively while in another province of the same region the prevalence of ID was found to be 7.8% (Kılınç et al., 2002, Koç et al., 2000). In

Table 4. The evaluation of inadequate and excessive daily energy and nutrient intake levels in the children (%).

	Boys (n:80)		Girls (n:92)		Total (n:172)	
	Inadequate Intake (%)	Excessive Intake (%)	Inadequate Intake (%)	Excessive Intake (%)	Inadequate Intake (%)	Excessive Intake (%)
Energy	42.5	-	34.8	3.3	38.4	1.7
Protein	3.8	71.8	2.2	71.7	2.9	71.9
Dietary fiber	56.3	-	51.1	1.1	53.5	0.6
Vitamin A	27.5	36.3	16.3	39.1	21.5	37.5
Vitamin B ₆	18.8	37.5	9.8	34.8	14.0	36.0
Folate	8.8	61.3	9.8	63.0	9.3	62.2
Vitamin B ₁₂	61.3	20.0	54.3	22.8	57.6	21.5
Vitamin C	21.3	60.0	14.1	60.9	17.4	60.5
Calcium	81.3	1.3	77.2	-	79.1	0.6
Magnesium	28.3	21.3	26.1	20.7	28.3	20.9
Iron	28.0	20.0	31.3	15.2	28.5	17.4
Zinc	22.5	30.0	20.7	19.6	21.5	24.4

Table 5. Correlation of dietary energy and nutrients intake with serum hematological analysis parameters.

	Hemoglobin	Hematocrit	Serum ferritin	Serum folic acid	Serum vitamin B ₁₂
	r	r	r	r	r
Energy	0.165 [*]	0.152 [*]	-0.020	0.076	-0.027
Protein	0.176 [*]	0.193 [*]	0.108	-0.023	0.152 [*]
Plant protein	0.145	0.101	-0.076	0.103	-0.112
Animal protein	0.018	0.062	0.194 [*]	-0.176 [*]	0.194 [*]
Fat	-0.008 [*]	0.042	0.035	0.039	0.099
Carbohydrate	0.224 [*]	0.173 [*]	-0.070	0.099	-0.103 [*]
Dietary fiber	0.138	0.157 [*]	-0.085	0.095	-0.175 [*]
Vitamin A	0.013	0.112	0.021	-0.136	-0.002
Vitamin B ₆	0.052	0.048	-0.035	0.111	-0.008
Folate	0.113	0.077	-0.044	0.158 [*]	-0.041
Vitamin B ₁₂	0.028	0.015	0.151 [*]	-0.076	0.054
Vitamin C	0.013	0.002	-0.089	0.094	-0.121
Calcium	0.139	0.211 [*]	0.093	-0.058	0.161 [*]
Magnesium	0.119	0.124	-0.069	0.115	-0.074
Iron	0.138	0.107	-0.004	0.060	-0.073
Zinc	0.129	0.152 [*]	0.111	-0.021	0.058

*p<0.05

Eagean region the prevalence of ID was determined as 6.5% in children between 6 and 11 years of age (Aydinok et al., 1998). Iron deficiency was determined to be at a prevalence of 6.5% among adolescents living in cities and 33% among those living in rural regions in Turkey (Kilingç et al., 2002, Koç et al., 2000). According to the results of the present study, the prevalence of iron deficiency was found to be 13.7% for boys and 17.4% for girls. The prevalence of iron deficiency anemia was found to be 3.3% in girls and these results were related to nutritional status of them.

Iron deficiency is a condition related to the inadequate

intake of the iron from food, primarily depending on nutrition or low bioavailability of iron (Zimmermann et al., 2007). In this study, inadequate iron intake was determined higher in girls than boys whereas no significant difference was found for iron intake in children with or without iron deficiency anemia ($p > 0.05$).

Anemias may also occur, infrequently, due to deficiencies in folic acid and Vitamin B₁₂. (Rasmussen et al., 2001; Zimmermann et al., 2007). However, the number of studies examining these vitamin deficiencies in school-aged children in Turkey is limited. In one study performed on school-aged children between the ages of

7 and 17 in three regions of Turkey, it was found that the prevalence of folic acid deficiency was 1.6% and while the prevalence of Vitamin B₁₂ was 1.3% in the Central Anatolia region (Açkurt et al., 1995). In a study performed in Southeast Anatolia region, it was determined that the prevalence of Vitamin B₁₂ deficiency was 10.8%, but no folic acid deficiency was found in children (Koç et al., 2005). Additionally, in a province of Southeast Anatolia region, the prevalence of deficiencies of folic acid and Vitamin B₁₂ were found to be 21.8 and 2.2%, respectively between the ages of 12 and 22 (Öncel et al., 2006). According to the results of our study, the prevalence of folic acid deficiency was determined 1.1% in girls that is similar to the results found in the study performed in Central Anatolia region. The prevalence of Vitamin B₁₂ deficiency was determined to be 3.8% in boys and 3.3% in girls, which was higher than the prevalence reported in other studies, except for the study performed in Southeast Anatolia region.

Although in the present study, 61% of children intake level of Vitamin C was determined over DRI, a loss of 90 to 95% may occur for the amount of this vitamin in food depending on the method of preparation. In addition, intake of folate also decreases when Vitamin C is lost in vegetables. This vitamin to be taken on margin may be mentioned when considered the losses from preparing and cooking. In one study conducted on adolescent girls in Turkey, it was determined that 18.4% of the individuals had inadequate intake of folate with food, so the folic acid deficiency was high (4.8 times higher in individuals with inadequate intake of folate with food, 2.4 times higher in low socioeconomic levels and 1.9 times higher in individuals with inadequate intake of Vitamin C with food) (Öner et al., 2006). In this study, the prevalence of inadequate intake of folate with food was found to be 9.3% in boys and 9.8% in girls. A positive correlation between serum folic acid and folate intake was also determined in both gender ($p < 0.05$).

In the present study, we found that 58% of the participating children had inadequate intake of vitamin B₁₂ and we observed a positive correlation between Serum vitamin B₁₂ and the intake of protein, animal protein, and calcium, and a negative correlation between serum vitamin B₁₂ and carbohydrate and dietary fiber intakes. In order to mitigate Vitamin B₁₂ deficiency, socioeconomic level, malabsorption and composition of the diet should also be considered.

Turkey has characteristics of both developing and developed countries. In addition to malnutrition in children, Turkey has also reported an increased prevalence of obesity in recent years, likely due to imbalanced nutrition (Pekcan et al., 2004; Yalcin et al., 2008). In the current study, 18% of the children were determined to be underweight or at risk of underweight while 15.2% of the children were overweight or obese. Several studies have shown that anemia has effects on cognitive performance, immune function, attention

capacity, ability to perform work and the physical growth of children (Nokes et al., 1998). In the current study, measurements of body weight were determined to be statistically low according to age only in the children with any deficiency ($p < 0.05$).

Conclusion

The results of this study show that iron deficiency is still common, in particular in school-aged children and that vitamin B₁₂ deficiency is more prevalent than folic acid deficiency in this age group. In addition, the results were supported that the dietary intakes of children correlated with vitamin deficiencies. However, this cross-sectional study could not establish causal relationships, but could only generate a hypothesis about the possible role of dietary intakes in the prevalence of iron, folic acid and vitamin B₁₂ deficiency among Turkish schoolchildren. Nonetheless, much more research is needed.

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