

# Vitamin E deficiency and associated factors among Brazilian school children

Deficiência de vitamina E e fatores associados entre crianças escolares brasileiras

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## ABSTRACT

**Objective:** Brazilian national data show a significant deficiency in pediatric vitamin E consumption, but there are very few studies evaluating laboratory-proven nutritional deficiency. The present study aimed to settle the prevalence of vitamin E deficiency (VED) and factors associated among school-aged children attended at a primary health unit in Ribeirão Preto (SP). **Methods:** A cross-sectional study that included 94 children between 6 and 11 years old. All subjects were submitted to vitamin E status analysis. To investigate the presence of factors associated with VED, socio-economic and anthropometric evaluation, determination of serum hemoglobin and zinc levels, and parasitological stool exam were performed. The associations were performed using Fisher's exact test. **Results:** VED ( $\alpha$ -tocopherol concentrations  $<7 \mu\text{mol/L}$ ) was observed in seven subjects (7.4%). None of them had zinc deficiency. Of the total of children, three (3.2%) were malnourished, 12 (12.7%) were anemic, and 11 (13.5%) presented some pathogenic intestinal parasite. These possible risk factors, in addition to maternal-work, maternal educational level, and monthly income, were not associated with VED. **Conclusions:** The prevalence of VED among school-aged children attended at a primary health unit was low. Zinc deficiency, malnutrition, anemia, pathogenic intestinal parasite, maternal-work, maternal educational level, and monthly income were not a risk factor for VED.

**Keywords:** Child; Vitamin E Deficiency; Risk Factors.

## RESUMO

**Objetivo:** Determinar a prevalência da deficiência de vitamina E (DVE) e os fatores associados a essa deficiência em escolares atendidos em uma unidade básica de saúde de Ribeirão Preto (SP). **Métodos:** Estudo transversal que incluiu 94 crianças entre 6 e 11 anos de idade, atendidas em uma unidade básica de saúde. Todos os indivíduos foram submetidos à análise do *status* de vitamina E. Para investigar a presença de fatores associados à DVE, foi realizada avaliação socioeconómica e antropométrica, determinação dos níveis séricos de hemoglobina e zinco, e exame parasitológico de fezes. As associações foram realizadas por meio do teste exato de Fisher. **Resultados:** A DVE (concentrações de  $\alpha$ -tocoferol  $<7 \mu\text{mol/l}$ ) foi observada em sete indivíduos (7,4%). Nenhum sujeito apresentou deficiência sérica de zinco. Do total de crianças, três (3,2%) eram desnutridas, 12 (12,7%) anêmicas e 11 (13,5%) apresentavam algum parasita intestinal patogênico. Estes possíveis fatores de risco, além do trabalho materno, escolaridade materna e renda mensal, não foram associados à DVE ( $p>0,05$ ). **Conclusão:** A prevalência de DVE em escolares atendidos em uma unidade básica de saúde foi baixa. Desnutrição, anemia, parasitose intestinal, renda mensal e trabalho e nível educacional maternos não se apresentaram como fatores de risco para a DVE.

**Palavras-chave:** Criança; Deficiência de Vitamina E; Fatores de Risco.

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## INTRODUCTION

Vitamin E (VE) is an essential lipid-soluble antioxidant nutrient required in small quantities for important biological processes. The VE role in oxidative stress has been widely studied, in addition to its participation in immunomodulation and several metabolic processes<sup>1</sup>. VE (tocopherol) is a generic term for several compounds. Among them,  $\alpha$ -tocopherol is the major VE form and represents over 90% of all plasmatic tocopherols.

In adult men, not only higher serum  $\alpha$ -tocopherol concentrations were related to lower general mortality, but also cancer and cardiovascular diseases were lower<sup>1,2</sup>. VE suboptimal serum levels are accepted as a risk factor for degenerative diseases; furthermore, the coronary artery disease severity was related to decreased VE<sup>2</sup>. In childhood, the role of VE was clearly shown in premature infants that had received VE deficient formula; those children developed hemolytic anemia and edema<sup>3</sup>. Moreover, VE, probably, is required for a normal neurological function<sup>4</sup>.

The severe VE deficiency (VED) may cause various degrees of neurologic deficits, hemolytic anemia, impaired lipid absorption, and could lead to a progressive neurological syndrome including cerebellar ataxia and posterior cord injury; nevertheless, a severe deficiency is rare and may occur related to a specific anomaly of hepatic VE or intestinal secretion of VE, excessive endogenous consumption, abetalipoproteinemia, cystic fibrosis, chronic cholestatic liver disease, short-bowel syndrome, and other fat malabsorption syndromes<sup>4-6</sup>.

Like other nutritional deficiencies, in addition to the VE low ingestion, such as protein-calorie undernourishment and low consumption of fruits and vegetables<sup>7</sup>, other factors like low serum zinc levels, iron deficiency anemia, parasitic infestation, and low socioeconomic profile may be associated with VED and can aggravate and/or contribute to this deficiency<sup>8</sup>.

Studies regarding healthy schoolchildren VE status in developing countries are lacking, but some authors showed insufficient ingestion<sup>9</sup>; thus, this study aimed to evaluate the VE inappropriate concentrations prevalence in schoolchildren enrolled

in a primary health care service in Ribeirão Preto (SP, Brazil) and verify the association between some risk factors and this condition.

## METHODS

### Study design and sampling

This is a cross-sectional descriptive study addressed also by analytic designs. All 104 children aged 6 to 11 years old attending a public primary pediatric clinic at Ribeirão Preto (São Paulo, Brazil) between September 2013 and March 2014, who after the routine pediatric appointment were not receiving VE supplements, were invited to take part in the study. Of these, ten were excluded because they had a fever, diarrhea, or hemorrhagic disease within 15 days prior to the appointment.

The purpose of the study was explained to the parents or legal guardians who also signed the written informed consent to the procedure. The study protocol was approved by the Research Ethics Committee of the University Hospital, Ribeirão Preto Medical School, University of São Paulo.

Demographic and socioeconomic data (maternal-work, maternal educational level, and monthly income) were obtained using a questionnaire-based interview with parents or legal guardians. The height and weight of children were also measured on the same day of blood sampling according to the procedure detailed<sup>10-12</sup>, and the body mass index (BMI) was calculated.

After at least 6 hours of fasting, blood samples were obtained to measure  $\alpha$ -tocopherol, hemoglobin, and zinc concentrations. Plasma  $\alpha$ -tocopherol concentrations were measured by HPLC<sup>13</sup>; hemoglobin levels were determined by the Coulter STKS method (Coulter Corporation, Hialeah, FL, USA); and serum zinc was obtained by flame atomic absorption spectrophotometry (SpectrAA 55B, Varian, Techtron, Victoria, Australia). Plasma  $\alpha$ -tocopherol concentrations lower than 7  $\mu$ mol/L were the cutoff for pediatric populations, and children with concentrations lower than that were considered as VED in this study<sup>13</sup>. Anemia was defined as hemoglobin concentration lower than 11.5 g/dL<sup>14</sup>.

The normal plasma zinc range is 50-150 µg/dL. In this study, children with plasma zinc concentrations lower than 50 µg/dL were zinc deficient<sup>15</sup>.

In order to evaluate intestinal parasitic diseases of the children, three stool samples were obtained by the parents on nonconsecutive days during the month of the study. Direct exam and spontaneous sedimentation method were used to determine the protozoan, nematodes presence<sup>16</sup>, and zinc sulfate flotation<sup>17</sup>.

In this study, the possible factors associated with VED that were investigated were: sex, maternal-work, maternal educational level (<8 years of maternal education), monthly income, BMI classification (z-scores less than -2 for the weight for age), anemia, zinc levels (< 50 µg/dL), and intestinal parasitosis.

## Statistical analysis

Data were expressed as the number and percentage of subjects. Statistical analysis was performed by non-parametric methods. Fisher's test was used to determine whether there were nonrandom associations between VED and one of the following variables: sex, maternal-work, maternal educational level, BMI classification, anemia, and parasitic infestation. Whenever the data distribution was not normal (age, zinc serum values, monthly income), the Mann-Whitney test was used to compare the medians of the children with and without VED, with a 95% confidence level.

Malnutrition was defined as a weight-for-age, height-for-age, and weight/height z-scores < -2. The Epi Info program version 2002 software (Centers for Disease Control and Prevention) was used to calculate the height for age, weight for height, and weight for age z-scores, comparing them with the National Center for Health Statistics standard reference population<sup>18</sup>.

The research project was approved by the Research Ethics Committee on 08/04/2003, according to the HCRP process no. 6007/2003.

## RESULTS

A total of 94 children participated in the study, of which 48 (51.0%) were male. Plasma α-tocopherol concentrations <12 µmol/L were observed in 31 (33.0%) children. These values were <7 µmol/L in seven (7.4%) subjects. Among the children with VED, 28.6% (2 out of 7) were boys, and 71.4% (5 out of 7) were girls. According to the gender, there was no significant difference (2 out of 48 boys; and 5 out of 46 girls) regarding the prevalence of VED (p=0.26) (Table 1).

**Table 1**  
Data of the children with and without VED

Variable	VED <sup>(a)</sup> present	VED <sup>(a)</sup> absent
Boys	28.6%	52.9%
Girls	71.4%	47.1%
Hb <sup>(b)</sup> < 11.5g/100ml	0.0%	14.0%
Hb <sup>(b)</sup> ≥ 11.5g/100ml	100.0%	86.0%
Maternal-work	71.4%	56.3%
Unemployed or housekeeper	28.6%	43.8%
≤ 8 years of maternal education	85.7%	67.8%
>8 years of maternal education	14.3%	32.2%
NMSPC <sup>(c)</sup> median	0.94	0.71

<sup>(a)</sup> VED: vitamin E deficiency; <sup>(b)</sup> Hb: hemoglobin; <sup>(c)</sup> NMSPC: national minimum salary per capita.

The age medians of children with and without VED were 7.08 and 7.92 years old, respectively. No significant difference was observed when they were compared (p=0.35). Among all children analyzed, two patients had z-scores less than -2 for the weight for age and one patient had weight for height z-score less than -2. None of them were with VED.

Serum zinc was measured in 87 children, and no child showed serum zinc levels <50 µg/dL. Two children without VED had the lowest zinc concentration (60 µg/dL). The groups with and without VED had similar zinc concentrations medians: 93 µg/dL and 92 µg/dL, respectively (p=0.50). Furthermore, 12.7% (12 out of 94) children were anemic, and

there was no association between anemia and VED in this group ( $p=0.59$ ) (Table 1).

Fecal samples were obtained from 81 subjects (median of three samples), and 11 (13.5%) children had some pathogenic intestinal parasite. *Giardia lamblia* and *Enterobius vermicularis* were the most frequently found pathogens (8.6 and 7.4%, respectively). The simultaneous presence of *G. lamblia* and *E. vermicularis* was found in two children. There was no relation between helminths and protozoan intestinal infections and VED in this group ( $p=0.53$ ).

All children studied were registered at public schools. Maternal work, maternal educational level, and monthly income (analyzed by the national minimum salary per capita - NMSPC) were not associated with VED ( $p=0.69$ , 0.43, 0.35, respectively) (Table 1).

## DISCUSSION

Studies regarding the VE status in healthy schoolchildren in developing countries are scarce, although some authors believe that it is a prevalent deficiency, with important consequences, like anemia<sup>19</sup>. Part of this situation could be attributed to the fact that the VE normal range criterion for serum levels among children has been a cause of concern<sup>20</sup>. Numerous studies have assumed serum levels lower than 12  $\mu\text{mol/L}$  as a criterion for inadequate  $\alpha$ -tocopherol concentrations in pediatric populations<sup>21,22</sup>. Notwithstanding, in other studies, VED in children has been defined as plasmatic concentrations of either total tocopherol or  $\alpha$ -tocopherol lower than 7  $\mu\text{mol/L}$ .

Drewel et al. (2006) verified that 68% of US healthy children aged 2-5 years had total tocopherol concentrations lower than 7  $\mu\text{mol/L}$ <sup>23</sup>. Although using a different criterion, other studies have reported 66 and 69% of preschool healthy children with serum  $\alpha$ -tocopherol lower than 12  $\mu\text{mol/L}$ <sup>21,24</sup>. Augusto et al., evaluating 702 Brazilian children aged 4 to 10 years, with low consumption of fruits and vegetables, showed that 9% had VED<sup>7</sup>.

Our data pointed few VED children. VE is found in vegetable oils, whole grains, sunflower seeds, wheat germ, and dark green leafy vegetables<sup>25</sup>.

Regarding dietary VE, most people consume less than the estimated average requirement (EAR) of VE even in developed countries; this fact can lead to an inadequate VE status that is usually clinically asymptomatic<sup>2</sup>, but, taking into account the antioxidant effects of VE, it is probably an important anti-inflammatory vitamin that could have relevant effect in the management of inflammatory diseases<sup>26</sup>. Children fed with milk fortified with vitamin E have low LDL-Cholesterol, showing that vitamin status is important for an adequate cholesterol status in childhood<sup>27</sup>. In the school-aged children, the current VE EAR is 6 mg of  $\alpha$ -tocopherol per day<sup>25</sup>.

The low prevalence of VED we found may be due to the fact that these children have access to good sources of VE like soy oil and bean, both largely consumed by Brazilian people<sup>28-30</sup>. Moreover, all children enrolled in this study were registered at public schools where balanced meals are prepared under the control of nutritionists and offered to the students daily. However, these hypotheses require further specific investigations.

Even though the majority of the 6- to 11-year-old children included in this study were considered without VED, it is important to analyze possible risk factors that may be correlated with this condition to contribute to the improvement of the suspicion of marginal VED in children.

Schoolchildren, in general, have lower rates of morbidity, mortality, and nutritional deficiencies compared to infants and preschoolers, a fact that may explain the relatively few studies on this age range<sup>8</sup>. In our study, the age of children did not influence the prevalence of VED.

Hoeft, Weber, and Eggersdorfer pointed out that vitamin deficiencies are the most common in male children<sup>8</sup>. Nevertheless, in this study, there was no difference between boys and girls regarding VED.

Although few malnourished children were observed, malnutrition was not related to VED. Subclinical VED in apparently well-nourished children represents part of one of the most important nutritional problems, hidden hunger. This phenomenon had been reported before<sup>31, 32</sup>.

The deficiency of multiple micronutrients is common. VE serum levels may be decreased in association with zinc deficiency<sup>33</sup>. Nevertheless, the lowest zinc values observed in our children were

not related to VED. There was also no association between VED and iron deficiency anemia.

The overall prevalence of intestinal parasite infections among children in this study was low (13.5%). Similar data were found by Muniz et al.<sup>34</sup> This can be mostly attributed to the urbanization process with the improvement of sanitation in Southwest Brazil, but is the note the case in poor regions of the country where hygienic problems persist<sup>35</sup>. Similar to other variables studied, in the present study, the intestinal-parasitic infections were not related to VED either.

In this study, we could not detect socioeconomic-demographic factors influences the VE status. Hoeft, Weber, and Eggersdorfer, in a different way, verified that malnutrition and micronutrient deficiencies are still highly prevalent and related to lower-income families and less educated mothers<sup>8</sup>.

The main limitation of the present study was the lack of food intake assessment using standardized and validated instruments. This would help to understand the prevalence of 7.4% of DVE. Another limitation of this study is the relatively small number of subjects, which may have restricted the power of the analyses performed. For this reason, the children of the present study cannot be considered representative of Brazilian schoolchildren. On the other hand, there is a lack of information about VED among healthy schoolchildren, and these data may contribute to this issue.

In conclusion, our data showed that 7.4% of school-aged children presented subclinical VED. Gender, anemia, serum zinc, malnutrition, intestinal parasitosis, and some socioeconomic-demographic factors were not correlated with VED. Despite this fact, VED should be better identified and evaluated to support appropriate actions in order to improve health status in this age group and, consequently, the elimination of VED repercussions on the future of these children.

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