Factors associated with the concentration of serum retinol in infants

**Fatores associados à concentração de retinol sérico em lactentes**

**Factores asociados a la concentración de retinol sérico en lactantes**

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**ABSTRACT**

**Objective:** To verify the factors associated with the concentration of retinol in infants assisted in a health public system.

**Methods:** Cross-sectional study carried out with 101 infants aged 18-24 months assisted at the health public system in the urban area of Viçosa city, Southeastern Brazil. The retinol concentration was analyzed by high performance liquid chromatography. In order to identify the dietary practices the 24 hours recall information was obtained. The interviews were accomplished with infant’s parents or with their caretakers at home. Evaluation of the variables associated to serum retinol levels was done by multiple linear regression analysis.

**Results:** The prevalence of vitamin A deficiency was 39.6%. The factors positively associated with serum retinol levels were paternal years of education and the diet protein content. On the other hand, the number of household inhabitants and the infant’s age were negatively associated with the levels of retinol.

**Conclusions:** Vitamin A deficiency is a public health problem among infants in Viçosa city. Socioeconomic, dietetic and biological variables are associated to serum concentration of retinol in infants.

**Key-words:** vitamin A; infant nutrition; infant; nutritional status.

**RESUMO**

**Objetivo:** Verificar os fatores associados à concentração de retinol em lactentes atendidos em serviço público de saúde.

**Métodos:** Estudo de corte transversal realizado com 101 crianças de 18 a 24 meses, atendidas no serviço público de saúde da área urbana da cidade de Viçosa. A análise da concentração de retinol no sangue venoso foi realizada por cromatografia líquida de alta eficiência. Para avaliação da prática alimentar, utilizou-se o recordatório de 24 horas. As entrevistas foram realizadas com os pais ou responsáveis pelas crianças em seus domicílios. Para avaliar as variáveis associadas ao retinol sérico, foi utilizada análise de regressão linear múltipla.

**Resultados:** Identificou-se 39,6% de prevalência da deficiência de vitamina A. Os fatores associados positivamente ao nível sérico de retinol foram tempo de escolaridade paterna e ingestão de proteína. Por outro lado, o número de moradores no domicílio e a idade da criança estiveram negativamente associados.

**Conclusões:** A hipovitaminose A se apresenta como um problema de Saúde Pública entre os lactentes do município de Viçosa. Fatores socioeconômicos, dietéticos e biológicos se associam à deficiência da vitamina A na população infantil.

**Palavras-chave:** vitamina A; nutrição do lactente; lactente; estado nutricional.
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Introduction

Vitamin A is of fundamental importance for vision, for cell differentiation and proliferation and for maintenance of epithelial integrity. Vitamin A deficiency is the most important cause of childhood blindness in developing countries. Furthermore, over recent decades evidence has been growing that vitamin A deficiency makes a significant contribution to morbidity and mortality due to infectious diseases even before their clinical signs emerge\(^{1,2}\).

It is estimated that subclinical vitamin A deficiency affects 213 million children worldwide and could be responsible for 250 to 500 thousand new cases of irreversible blindness every year\(^{3}\). In Latin America, prevalence rates of subclinical deficiency among children under 3 range from 4.6 to 75.9\(^{4}\). With relation to Brazil, there is little data on which to establish the prevalence and severity of this deficiency. Localized studies have shown that this deficiency is a public health problem, with prevalence rates ranging from 16 to 74\(^{4}\) among children under 6 and also that, contrary to expectations, this problem is not limited to the poorest regions of the country\(^{5}\). World Health Organization (WHO) estimates define vitamin A deficiency as a moderate public health problem in Brazil\(^{6}\).

According to Miller et al\(^{7}\), there are two main reasons why children under 2 living in developing countries can suffer from vitamin A deficiency. The first is if their mothers are themselves deficient and produce breastmilk with insufficient vitamin A levels. The second is that in general complementary feeding is a poor source of the vitamin. In addition to these two reasons, they also point out that some of these children have illnesses that can cause anorexia, malabsorption and increased catabolism, increasing the risk of nutritional deficiencies.

Few studies have attempted to demonstrate which factors are associated with serum retinol concentration in infants. The objective of this study was therefore to investigate factors associated with serum retinol levels in infants aged 18 to 24 months seen at public healthcare services in the urban area of the town of Viçosa, MG, Brazil.

Method

This was a cross-sectional study of children aged 18 to 24 months seen at public healthcare services in the urban area of the town of Viçosa, MG, Brazil.

All children registered with the town’s pediatric service who were within the age range of interest were visited at home and their parents or guardians were invited to take part in the study. A total of 124 children were registered at the service with valid, locatable addresses. Twenty-three (15\%) of these children did not take part because their parents did not agree to participate. The sample was therefore 101 children, 51 (50.5\%) females and 50 (49.5\%) males.

Data were obtained during interviews with the children’s mothers or guardians. The variables analyzed were as follows: related to the child (sex, duration of gestation, birth weight and length); socioeconomic conditions (educational level of parents, family income, number of inhabitants and number of rooms in family home); maternal variables (number of gestations and prenatal care); recent history of illness, intake of supplements and feeding practices.

Birth weight and length and variables related to gestation were checked against the children's vaccination card and prenatal care card, when these were available. Supplements given to the infants were also confirmed by the investigator. Family income per capita was calculated as total monthly wage divided by the number of people dependent on that income.

RESUMEN

Objetivo: Verificar los factores asociados a la concentración de retinol en lactantes atendidos en el servicio público de salud del área urbana de la ciudad de Viçosa (Minas Gerais, Brasil).

Métodos: Estudio de corte transversal realizado con 101 niños, de 18 a 24 meses, atendidos en el servicio de salud del área urbana de la ciudad de Viçosa. El análisis de la concentración de retinol en la sangre venosa fue realizado por cromatografía líquida de alta eficiencia. Para evaluar de la práctica alimentar, se utilizó el recordatorio de 24 horas. Las entrevistas fueron realizadas con los padres o responsables de los niños en sus domicilios. Para evaluar las variables asociadas al retinol sérico, se utilizó análisis de regresión lineal múltiple.

Resultados: Se identificó 39,6\% de prevalencia de la deficiencia de vitamina A. Los factores asociados positiva- mente al nivel sérico de retinol fueron tiempo de escolaridad paterna e ingestión de proteína. Por otra parte, el número de moradores en el domicilio y la edad del niño estuvieron negativamente asociados.

Conclusiones: La hipovitaminosis A se presenta como un problema de Salud Pública entre los lactantes del municipio de Viçosa. Factores socioeconómicos, dietéticos y biológicos se asocian a la deficiencia de la vitamina A en la población infantil.

Palabras clave: vitamina A; nutrición del lactante; lactante; estado nutricional.
Children's weight and length were also measured during the home visits, using standardized equipment and guidelines, using the procedures recommended by the WHO. They were weighed using an electronic digital pediatric scale with 15 kg capacity and 10 g intervals and measured with a wooden anthropometer with an amplitude of 120 cm and a 0.1 cm scale. Nutritional status was assessed on the basis of weight/age, length/age and weight/length, converted to Z-scores and taking -2 and +2 as cutoff points for nutritional problems. The WHO growth curves were adopted as the anthropometric reference standard and the WHO Anthro software package was used to analyze the data.

Serum retinol levels were assayed in 3mL blood samples taken by venipuncture after a minimum of 8 hours' fasting. Blood was collected and centrifuged and the serum separated in an area protected from sunlight and with indirect lighting. Serum samples were frozen at -18°C until analysis.

Retinol assays were performed by High Performance Liquid Chromatography in a Lichrospher 100, RP-18, 5μm, 150x4mm column using a methanol and water mobile phase (concentration 95:5), flow of 1.5mL/min and fluorescence detection. The external and internal standards were all-trans retinol acetate and all-trans retinyl acetate, diluted in the mobile phase. The internal standard is recommended because it allows losses during analysis to be controlled.

Vitamin A nutritional status deficiency was defined as a serum concentration below 20μg/dL. World Health Organization recommendations were followed in assessing whether the levels of deficiency observed constitute a public health problem.

Retinol assays were conducted in the Vitamin Analysis Laboratory at the Nutrition and Health department of the Universidade Federal de Viçosa (UFV). Parasitology tests were conducted and blood samples taken at a reputable clinical analysis laboratory.

Fecal parasitology was conducted using the Hoffman, Faust and Ritchie methods. Tests were classified as positive if at least one species of parasite was detected in the sample.

Feeding practices were assessed on the basis of the results of a 24-hour recall. Certain specific questions were also asked with relation to feeding practices: whether the child consumed milk soon after main meals and the ages at which water, teas fruit and/or fruit juices, non-breastmilk milks and savory mashes were introduced to the child's diet.

The chemical composition of breastmilk used for nutritional calculations was as recommended by the Institute of Medicine. Breastmilk intake was estimated according to the WHO/UNICEF suggestion that the volume of milk consumed by this age group is 61.16mL/kg of body weight per day. For children who were only breastfed once or twice a day, breastmilk intake was estimated at 27mL/kg per feed. The nutritional composition of the children's diets was calculated using Diet-Pro version 4.0. Dietary vitamin A intake was then classified as healthy or deficient on the basis of Estimated Average Requirements.

A database was constructed using Epi-Info, version 6.04, which is designed for epidemiological analyses. Regression analysis was performed in SPSS for Windows, version 10.

Univariate analysis was conducted using the Mann-Whitney test to compare median serum retinol results on the basis of categorical variables. Pearson's correlation was used to investigate associations between serum retinol and numerical variables. Variables that proved to be significantly associated with retinol concentration (p<0.05) were then included in a multivariate linear regression model. Linear regression was adopted because the sample size meant that categorizing some of the variables would have been problematic.

The children's mothers were given nutritional guidance and children who had parasites or abnormal biochemical test results were referred for treatment. This research was approved by the Human Research Ethics Committee at the UFV.

**Results**

The mean age of the children assessed was 21.15 (SD=1.89) months. They came from families with a mean per capita income of US$ 217.36 (SD=114.4) and their mothers were 25.3 (SD=5.52) years of age and had been educated for 7.2 (SD=2.98) years. The length/age, weight/age and weight/length Z-scores were below -2 for 4, 3 and 3% of the children respectively. At the other end of the scale, 2% of the children had weight/length and 3% had weight/age Z-scores greater than +2.

There was a high prevalence of vitamin A deficiency among the infants assessed here (39.6%, n=40). Median serum retinol in the study population was 21.8μg/dL, with a range of 9 to 59.9μg/dL. Median retinol levels were not statistically different when broken down by sex, birth weight, use of ferrous compound during gestation or after birth, use of multivitamins after birth, history of admission, illness during the previous 15 days or use of ferrous compound or multivitamins by the child (Table 1).

With relation to parasite infections, 21% of the children assessed had at least one type of parasite and this was associated with reduced serum retinol concentration. The most common parasite was *Giardia lamblia* (66.7%) followed by *Ascaris lumbricoides* (28.6%). In common with parasite infections, the
Factors associated with the concentration of serum retinol in infants

Table 1 - Concentration of serum retinol broken down by biological characteristics and maternal variables for children aged 18 to 24 months seen at public healthcare services in the town of Viçosa (MG), Brazil, 2005

<table>
<thead>
<tr>
<th>Variables</th>
<th>n (101)</th>
<th>Median (µg/dL)</th>
<th>Range (µg/dL)</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sex</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Female</td>
<td>51</td>
<td>21.02</td>
<td>9.00-39.48</td>
<td>0.21</td>
</tr>
<tr>
<td>Male</td>
<td>50</td>
<td>22.63</td>
<td>11.45-39.99</td>
<td></td>
</tr>
<tr>
<td>Stool testa</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Negative</td>
<td>79</td>
<td>22.39</td>
<td>9.00-39.48</td>
<td>0.01*</td>
</tr>
<tr>
<td>Positive</td>
<td>21</td>
<td>18.95</td>
<td>9.87-26.55</td>
<td></td>
</tr>
<tr>
<td>Took ferrous compound during gestationb</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Yes</td>
<td>91</td>
<td>21.79</td>
<td>9.00-39.99</td>
<td>0.44</td>
</tr>
<tr>
<td>No</td>
<td>8</td>
<td>23.69</td>
<td>12.02-28.47</td>
<td></td>
</tr>
<tr>
<td>Took multivitamins during gestationb</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Yes</td>
<td>14</td>
<td>24.00</td>
<td>18.05-35.58</td>
<td>0.015*</td>
</tr>
<tr>
<td>No</td>
<td>85</td>
<td>21.19</td>
<td>9.00-39.99</td>
<td></td>
</tr>
<tr>
<td>Took ferrous compound after deliveryb</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Yes</td>
<td>20</td>
<td>24.24</td>
<td>9.87-34.57</td>
<td>0.08</td>
</tr>
<tr>
<td>No</td>
<td>79</td>
<td>21.75</td>
<td>9.00-39.99</td>
<td></td>
</tr>
<tr>
<td>Took multivitamins after deliveryb</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Yes</td>
<td>3</td>
<td>31.44</td>
<td>18.05-32.07</td>
<td>0.16</td>
</tr>
<tr>
<td>No</td>
<td>96</td>
<td>21.83</td>
<td>9.00-39.99</td>
<td></td>
</tr>
<tr>
<td>Low birth weight</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Yes</td>
<td>12</td>
<td>17.74</td>
<td>9.87-29.92</td>
<td>0.21</td>
</tr>
<tr>
<td>Illness during previous 15 days</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>No</td>
<td>60</td>
<td>22.15</td>
<td>9.00-39.99</td>
<td>0.43</td>
</tr>
<tr>
<td>Yes</td>
<td>41</td>
<td>20.66</td>
<td>11.45-35.58</td>
<td>0.34</td>
</tr>
<tr>
<td>Admission prior to study</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>No</td>
<td>71</td>
<td>21.28</td>
<td>9.00-39.99</td>
<td>0.34</td>
</tr>
<tr>
<td>Yes</td>
<td>30</td>
<td>22.69</td>
<td>9.87-34.57</td>
<td></td>
</tr>
<tr>
<td>Currently takes ferrous compound</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Yes</td>
<td>9</td>
<td>20.46</td>
<td>15.23-39.99</td>
<td>0.92</td>
</tr>
<tr>
<td>No</td>
<td>92</td>
<td>21.83</td>
<td>9.00-39.48</td>
<td></td>
</tr>
<tr>
<td>Currently takes multivitamins</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Yes</td>
<td>6</td>
<td>21.89</td>
<td>18.05-39.99</td>
<td>0.64</td>
</tr>
<tr>
<td>No</td>
<td>95</td>
<td>21.78</td>
<td>9.00-39.48</td>
<td></td>
</tr>
</tbody>
</table>

Mann-Whitney. *variables selected for the model; *the test was not performed for one child; **two mothers did not know the answer to these questions

mother not taking multivitamins during gestation was also significantly associated with lower retinol levels (Table 1).

Table 2 lists the correlations between retinol concentration and the numerical variables investigated in this study. It will be observed that there was a weak negative correlation between both age of child and number of residents in home and serum retinol. In contrast, father’s educational level and calorie, protein and calcium content of the diet were positively correlated with retinol.

The infants’ median vitamin A intake was 410.45µg (range: 32.76 to 3,587.98µg). Eighteen percent of the children assessed had insufficient vitamin A intake, but median vitamin A intake was not correlated with serum retinol concentration.

Variables that were significantly correlated with serum retinol concentration were chosen for the multivariate linear regression model (Table 3). In this model, only father’s educational level, protein intake, number of residents in home
Table 2 - Pearson's correlation coefficient for serum retinol against variables relating to children aged 18 to 24 months seen at public healthcare services in the town of Viçosa (MG), Brazil, 2005

<table>
<thead>
<tr>
<th>Variables related to gestation</th>
<th>r</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number of prenatal consultations</td>
<td>-0.020</td>
<td>0.84</td>
</tr>
<tr>
<td>Start of prenatal care (months)</td>
<td>0.002</td>
<td>0.98</td>
</tr>
<tr>
<td>Dose of ferrous compound during gestation (capsules)</td>
<td>0.010</td>
<td>0.92</td>
</tr>
<tr>
<td>Duration of supplementation with ferrous compound during gestation (months)</td>
<td>-0.005</td>
<td>0.96</td>
</tr>
<tr>
<td>Dose of multivitamins during gestation (capsules)</td>
<td>-0.010</td>
<td>0.98</td>
</tr>
<tr>
<td>Duration of supplementation with multivitamins during gestation (months)</td>
<td>-0.010</td>
<td>0.97</td>
</tr>
<tr>
<td>Duration of gestation (months)</td>
<td>-0.060</td>
<td>0.52</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Biological variables</th>
<th>r</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (months)</td>
<td>-0.220</td>
<td>0.03*</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Variables related to birth</th>
<th>r</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Birth weight (g)</td>
<td>0.080</td>
<td>0.41</td>
</tr>
<tr>
<td>Birth length (cm)</td>
<td>0.040</td>
<td>0.66</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Socioeconomic variables</th>
<th>r</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age of mother (years)</td>
<td>-0.008</td>
<td>0.93</td>
</tr>
<tr>
<td>Mother's educational level (years in education)</td>
<td>-0.110</td>
<td>0.28</td>
</tr>
<tr>
<td>Father's educational level (years in education)</td>
<td>0.270</td>
<td>0.01*</td>
</tr>
<tr>
<td>Number of siblings</td>
<td>-0.190</td>
<td>0.06</td>
</tr>
<tr>
<td>Number of residents in home</td>
<td>-0.220</td>
<td>0.03*</td>
</tr>
<tr>
<td>Number of rooms in home</td>
<td>-0.110</td>
<td>0.28</td>
</tr>
<tr>
<td>Number of bedrooms in home</td>
<td>-0.070</td>
<td>0.50</td>
</tr>
<tr>
<td>Number of residents/bedroom</td>
<td>-0.050</td>
<td>0.58</td>
</tr>
<tr>
<td>Income (expressed in multiples of minimum monthly wage)</td>
<td>0.010</td>
<td>0.91</td>
</tr>
<tr>
<td>Per capita income in multiples of minimum monthly wage</td>
<td>0.050</td>
<td>0.61</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Dietary variables</th>
<th>r</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Breastfeeding duration (days)</td>
<td>0.100</td>
<td>0.36</td>
</tr>
<tr>
<td>Age at introduction of water and tea (months)</td>
<td>-0.030</td>
<td>0.75</td>
</tr>
<tr>
<td>Age at introduction of juice and/or fruit (months)</td>
<td>0.110</td>
<td>0.27</td>
</tr>
<tr>
<td>Age at introduction of non-breastmilk milks (months)</td>
<td>0.060</td>
<td>0.51</td>
</tr>
<tr>
<td>Age at introduction of savory mashes (months)</td>
<td>0.070</td>
<td>0.46</td>
</tr>
<tr>
<td>Breastmilk intake (mL)</td>
<td>0.030</td>
<td>0.71</td>
</tr>
<tr>
<td>Cow's milk intake (mL)</td>
<td>0.160</td>
<td>0.11</td>
</tr>
<tr>
<td>Calories</td>
<td>0.200</td>
<td>0.04*</td>
</tr>
<tr>
<td>Carbohydrates (g)</td>
<td>0.170</td>
<td>0.08</td>
</tr>
<tr>
<td>Lipids (g)</td>
<td>0.080</td>
<td>0.38</td>
</tr>
<tr>
<td>Protein (g)</td>
<td>0.230</td>
<td>0.01*</td>
</tr>
<tr>
<td>Calcium (mg)</td>
<td>0.210</td>
<td>0.03*</td>
</tr>
<tr>
<td>Vitamin C (mg)</td>
<td>0.111</td>
<td>0.27</td>
</tr>
<tr>
<td>Vitamin A (µg)</td>
<td>0.160</td>
<td>0.12</td>
</tr>
<tr>
<td>Iron (g)</td>
<td>0.130</td>
<td>0.18</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Nutritional status</th>
<th>r</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Current weight (kg)</td>
<td>0.060</td>
<td>0.51</td>
</tr>
<tr>
<td>Current length (cm)</td>
<td>0.040</td>
<td>0.65</td>
</tr>
<tr>
<td>L/A z score</td>
<td>0.140</td>
<td>0.18</td>
</tr>
<tr>
<td>W/A z score</td>
<td>0.110</td>
<td>0.28</td>
</tr>
<tr>
<td>W/L z score</td>
<td>0.050</td>
<td>0.62</td>
</tr>
</tbody>
</table>

L/A: length/age; W/A: weight/age; L/A: weight/length; *Variables included in the multivariate model.
Factors associated with the concentration of serum retinol in infants

Factors associated with the concentration of serum retinol in infants and child’s age remained significantly associated with serum retinol levels. This analysis showed that each extra year the father had spent in education was associated with an increase of 0.052µg/dL (p=0.013) in serum retinol concentration; each gram of protein intake was related to an increase of 0.097µg/dL (p=0.026) of retinol; each extra person living in the same household as the child reduced serum retinol by 0.819µg/dL (p=0.012); and for each month increase in age serum retinol reduced by 0.725µg/dL (p=0.020). Taken together, the variables in the model explain 14.9% of the variation in retinol levels among the infants assessed.

Discussion

On the basis of the prevalence observed here, vitamin A deficiency can be considered a public health problem among infants living in the urban area of Viçosa. A study conducted in the city of São Paulo with children aged 6 to 24 months found a 21.4% prevalence of vitamin A deficiency(15), with Martins, Santos and Assis(16), investigating preschool children in the state of Sergipe and found that 32.1% had retinol levels below 20µd/dL. The prevalence of vitamin A deficiency varied from 26 to 46% among Argentinean children aged from 6 months to 2 years(17). The prevalence of vitamin A deficiency among children aged 12 to 71 months in Honduras was 14%(18). Among Brazilian children under 5, assessed as part of the National Survey on Demography and Health of Women and Children (Pesquisa Nacional de Demografia e Saúde da Criança e da Mulher), 17.4% had low serum vitamin A levels(19). Therefore, the prevalence of vitamin A deficiency observed in the present study is comparable with the observations of other studies.

The importance of acquiring information on the prevalence of this deficiency merits mention, since little is currently known about the situation of vitamin A deficiency in Brazil. Notwithstanding, diagnosis on the basis of serum retinol has its limitations, since retinol is controlled homeostatically and serum concentrations may not reduce until the body’s reserves are compromised, and as such the marker is only a reliable indicator for diagnosing the deficiency when reserves are low or depleted(1). This aspect of serum retinol assays means that they do not provide a basis for drawing conclusions on the children’s vitamin A reserves. Furthermore, there have been suggestions that the cutoff points and criteria used to interpret serum retinol levels and diagnose deficiency should be reviewed(20).

The univariate analysis of categorical variables in this study found no differences when retinol results were broken down by sex, birth weight, taking ferrous compound during gestation or after delivery, taking multivitamins after delivery, illness during the previous 15 days, previous history of admissions or current ferrous compound and multivitamin use. However, there were significant differences in retinol concentration for children with parasite infections and the children of women who had taken multivitamins during gestation. Among the numerical variables, there were correlations between serum retinol levels and age, number of residents in household, father’s educational level and calorie, protein and calcium intakes. However, only father’s educational level, protein intake, number of residents in household and child’s age remained significant in the linear regression model.

Table 3 - Multivariate linear regression model for retinol levels of children aged 18 to 24 months seen at public healthcare services in the town of Viçosa (MG), Brazil, 2005

<table>
<thead>
<tr>
<th>Variables</th>
<th>β</th>
<th>Standard error</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Constant</td>
<td>36.203</td>
<td>7.279</td>
<td>0.000</td>
</tr>
<tr>
<td>Father’s educational level (years)</td>
<td>0.052</td>
<td>0.021</td>
<td>0.013</td>
</tr>
<tr>
<td>Protein (g)</td>
<td>0.097</td>
<td>0.043</td>
<td>0.026</td>
</tr>
<tr>
<td>Number of residents in home</td>
<td>-0.819</td>
<td>0.318</td>
<td>0.012</td>
</tr>
<tr>
<td>Age of child (months)</td>
<td>-0.725</td>
<td>0.306</td>
<td>0.020</td>
</tr>
</tbody>
</table>

$r^2$ for this model=0.149
but they did detect differences on the basis of weight/age Z-score and per capita family income. Escobar et al\textsuperscript{(17)} did not observe differences in retinol concentration related to age, sex, birth weight, vitamin A intake, illness in previous 15 days, anthropometric indices or vaccination history. In a study published by Nestel et al\textsuperscript{(18)}, prevalence of vitamin A deficiency was not related to mother’s educational level, presence of coughing or diarrhea in the previous 15 days or length/age or weight/age Z-scores, but they did observe differences associated with age. In preschool children in Congo, an association was observed between retinol levels and vitamin A intake, but not with anthropometric indices\textsuperscript{(21)}. Muniz-Junqueira and Queiroz\textsuperscript{(22)} did not detect a relationship between the deficiency and intestinal parasitosis or weight/age. In Recife\textsuperscript{(23)} and the Distrito Federal\textsuperscript{(24)}, no association was detected between hypovitaminosis and malnutrition. In Teresina, children’s retinol results were not associated with their ages, their mothers’ or fathers’ educational level or income or with their nutritional status\textsuperscript{(25)}. Among Indian children\textsuperscript{(26)}, an association was detected between nutritional status and intake of foods rich in vitamin A, in that children with low intakes also had poor anthropometric figures. Additionally, maternal age and education were also associated with intake of foods rich in vitamin A.

With relation to the association between intestinal parasite infection and vitamin A deficiency, it is thought that this is possibly because the parasites cause diarrhea, which in turn affects absorption of fats and vitamin A.

Linear regression analysis illustrates the influence that each variable in the model has on the dependent variable. In the model constructed here, father’s educational level and protein intake were positively correlated with retinol and number of people living in the home and age of child were negatively correlated with serum retinol.

Family income is an indicator of structural processes within society and is a factor that determines the health and nutrition conditions in which children live\textsuperscript{(16,18)}. In this study, family income was not associated with retinol concentration, possibly because a large number of respondents did not provide this information, reducing the sample size available for analysis. Additionally, the children investigated were from low-income families and so the distribution of family income was homogeneous. Notwithstanding, it is possible that the socioeconomic variables “father’s educational level” and “number of people in household” reflect family income. In general, a higher educational level increases the likelihood of employment and, consequently, leads to higher income. In contrast, large numbers of residents in the home is a finding often observed in low-income families. One review has suggested that nutritional factors and socioeconomic characteristics could influence vitamin A concentrations in the breastmilk of Brazilian women\textsuperscript{(27)}, which would have an indirect effect on retinol levels in their infants. In contrast, Oliveira, Oliveira and Bergamaschi\textsuperscript{(28)} do not believe that consensus has yet been reached on the relationship between these variables and vitamin A concentrations in breastmilk. It is noteworthy that the greater the number of children in a household, the lower the likelihood they will be eating foods that are rich in vitamin A\textsuperscript{(26)} which could constitute a risk factor for low retinol levels.

Vitamin A intake on the day before the interview was not correlated with retinol. This result, however, is to be expected since 24-hour recalls are of limited utility for assessing habitual intakes because they only reflect 1 day’s intake. A population’s vitamin A intake has a heterogeneous distribution, with a high coefficient of variation, which imposes limitations on the results of intake estimates\textsuperscript{(17)}. With relation to the finding that protein intake was positively correlated with retinol, it is believed that if an infant’s diet includes sufficient protein it should also have an adequate vitamin A content, since a large proportion of protein sources are also sources of vitamin A. Furthermore, children in the age group studied consume large quantities of milk, which is their primary source of both protein and vitamin A.

The negative correlation between retinol and age was to be expected, since preschool children are at greatest risk of deficiency. Their vulnerability is the result of a high demand for vitamin A for growth and development, compounded by illnesses that reduce the efficiency of vitamin A uptake\textsuperscript{(29)}. Confirmation of vitamin A deficiency before 2 years of age could help to explain a part of the elevated prevalence rates of vitamin A deficiency observed in the preschool age group\textsuperscript{(16,18)}.

The factors father’s educational level, number of residents in home and protein intake all reflect socioeconomic characteristics, demonstrating the vulnerability of children from low-income families to nutritional deficiencies and the need for early intervention in this subset to prevent and combat them.

It should be borne in mind that this was a cross-sectional study design which is not the most appropriate for tracing cause and effect relationships. Notwithstanding, it is a study design that is widely used to investigate health status and requires fewer resources making it feasible in a larger number.
of situations. The sample was selected by convenience, which makes extrapolation of the results to the whole population problematic, but it did achieve widespread coverage of the whole of the town’s urban area and reflects the nutritional status of the children registered with the health service. Therefore, these results support the conclusions that in a sample of children from the town of Viçosa, MG, Brazil, vitamin A deficiency proved to be a public health problem and that the factors associated with retinol levels in this group of infants are their fathers’ educational level, the number of residents in their homes, their protein intake and their ages.

References