Significance of postprandial blood concentrations of retinol, retinol-binding protein, and carotenoids when assessing the vitamin A status of children[1-5]

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ABSTRACT The effect of ingesting a breakfast rich in vitamin A on postprandial blood serum concentration of retinol, retinol-binding protein, and carotenoids has been investigated in children between 5 and 8 yr of age. They were divided by age in two groups (5 to 6 and 7 to 8 yr) and then randomly assigned in three groups to be studied cross-sectionally immediately before and at 2 and 4 h after the ingestion of a meal containing 337 μg of retinol equivalents (48% as retinol and 52% as carotenoids). The ingestion of breakfast did not alter significantly ($p > 0.05$) the postprandial serum concentrations of retinol, retinol-binding protein; or carotenoids in any of the age groups. These results indicate that up to 4 h the postprandial blood serum concentrations of these parameters are representative of their corresponding basal concentrations. Therefore, in practice and particularly under field survey conditions, the blood samples required to assess the vitamin A status of children can be obtained either fasting or within 4 h after breakfast without altering the results. *Am J Clin Nutr* 1984;39:62–65.

KEY WORDS Vitamin A, retinol, retinol-binding protein, carotenoids, preprandial and postprandial vitamin A levels, vitamin A status of children, assessment of vitamin A status, vitamin A field studies, time of blood collection

Introduction

In population and clinical studies, a common approach to assess the vitamin A nutritional status is to measure the blood serum or plasma concentrations of retinol and carotenoids. Since single vitamin A doses given orally are known to increase the blood concentration of this vitamin (1–4), there is always the question regarding the best time of sample collection so that the vitamin A concentration in blood is not affected by the vitamin A content of a previous meal. Therefore, it has been suggested that blood samples should be collected only under fasting conditions. In practice, particularly in field surveys, this requirement is impossible to fulfill, and more so in children. Blood samples are generally obtained throughout the morning, regardless of breakfast.

In a recent publication, Mejía and Arroyave (5) showed that adults ingesting a meal rich in vitamin A did not have significant postprandial changes in serum retinol, retinol-binding protein (RBP), and carotenoids concentrations up to a period of 4 h.

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Based on this information, it was inferred that the same phenomenon occurred in children.

Since children are more vulnerable than adults to be deficient in vitamin A, it was considered important to investigate specifically in this age group whether the ingestion of a breakfast rich in vitamin A had any effect on postprandial blood concentrations of retinol, RBP, or carotenoids.

Methods

Sixty-eight children (40 boys and 28 girls) 5 to 8 yr old were studied. They were deaf-mute students of a special education school in Guatemala City (School "Fray Pedro Ponce de León," of the Guatemalan National Committee for the Blind and Deaf) but otherwise normal and with apparent adequate nutritional status. Informed consent from the parents of the children and approval from the above mentioned Committee were obtained before conducting the experiment.

Knowing that in children the serum retinol levels are directly related to age (6), they were first divided in two groups: between 5 and 6 (n = 32) and between 7 and 8 yr of age (n = 36). Using a generalized randomized block design (7), they were then assigned into three cross-sectional groups to be studied, respectively, before (basal, n = 23), at 2 (n = 22), and 4 (n = 23) h after the ingestion of a breakfast meal designed to contain a relatively high amount of vitamin A. The data obtained were analyzed accordingly by a two-way analysis of variance. This experimental design was used to avoid repeating venipuncture in each child and also to isolate any possible effect of age.

After an overnight fast, the meal was fed around 8:00 AM and its composition was as follows: two scrambled eggs fried with margarine, a portion of fresh cheese (45 g), a glass of whole milk (180 ml), a glass of papaya juice (180 ml), and French bread spread with margarine (22 g). The margarine used was commercially fortified with β-carotene at a level determined in our laboratory of 17 μg/g. These food components were carefully weighed and according to food composition tables (8), the meal contained 337 μg of retinol equivalents, 48% as retinol and 52% as carotenoids. Retinol equivalents were calculated as described by Flores et al (9), using the conversion factors proposed by the FAO/WHO group of experts (10) of 1, 1/6, and 1/12 for retinol, β-carotene, and other carotenoids, respectively. According to the US National Research Council (11), this amount of vitamin A in the meal corresponds, respectively, to 67 and 48% of the daily dietary recommendations for children aged 4 to 6 and 7 to 10 yr.

A 2-ml venous blood sample was obtained from each child by arm venipuncture according to the experimental design. The blood serum was separated by centrifugation for the analyses of retinol, RBP, and total carotenoids. Retinol and total carotenoids were analyzed by the method of Bessey et al (12); by this method, both retinol and retinyl esters are measured together. RBP was determined by radial immunodiffusion (13), using commercial immunodiffusion plates (LC-Partigen, Behringwerke AG, Marburg, West Germany).

Results

Table 1 shows the blood serum concentrations of total retinol, RBP, and carotenoids during fasting (basal) and at 2 and 4 h postprandially according to the age of the children. There was no significant effect ($p > 0.05$) of the time of blood collection on the serum concentrations of these biochemical parameters in either of the two age groups. The corresponding F values were 0.2336, 1.7664, and 0.1166 for retinol, RBP, and carotenoids, respectively, none of which reached a statistically significant level.

Discussion

The present results confirm our previous observations in adults which showed that the blood serum concentrations of retinol, RBP, and carotenoids are not altered by the ingestion of a meal containing relatively high amounts of vitamin A from natural food sources at least over a period of 4 h (5). Our data indicate that the same phenomenon occurs in children from 5 to 8 yr of age.

Several studies in humans have shown that single oral doses of vitamin A can increase the serum concentration of retinol (1–3). However, the amounts of vitamin A given have been larger and, furthermore, were not as an intrinsic part of a meal as tested in the present experiment. After ingesting a meal, the rate of gastric emptying may not be fast enough to provide at one point in time an amount of vitamin A for intestinal absorption sufficient to cause detectable changes in the serum concentration of the vitamin.

Loerch et al (4) have shown that when rats are given a single vitamin A dose by gastric intubation, they undergo changes in the blood concentration of the vitamin that are proportional to their vitamin A status. Furthermore, the peak change in plasma retinol after the dose occurred at different time periods, depending on their vitamin A nutriture, ie, at about 3 h in vitamin A-sufficient rats (basal plasma retinol >30 μg/dl) and not before 5 h in marginally deficient animals (basal plasma retinol <30 μg/dl).

TABLE 1
Basal and postprandial blood serum concentrations of total retinol, RBP, and carotenoids after the ingestion of the vitamin A-rich breakfast meal

H after the meal	Retinol (μg/dl)		RBP (μg/ml)		Carotenoids (μg/dl)	
	5–6 yr	7–8 yr	5–6 yr	7–8 yr	5–6 yr	7–8 yr
0 (basal)	36.5 ± 6.5* (n = 10)	44.11 ± 11.1 (n = 13)	34.3 ± 5.7 (n = 10)	39.5 ± 9.7 (n = 13)	78.8 ± 32.7 (n = 10)	99.6 ± 43.2 (n = 13)
2	38.9 ± 7.7 (n = 11)	45.8 ± 8.4 (n = 11)	36.7 ± 5.8 (n = 11)	41.2 ± 6.6 (n = 11)	83.1 ± 35.1 (n = 11)	93.6 ± 44.8 (n = 11)
4	41.8 ± 8.6 (n = 11)	41.6 ± 7.3 (n = 12)	36.4 ± 7.5 (n = 11)	38.7 ± 7.9 (n = 12)	90.7 ± 16.0 (n = 11)	76.2 ± 23.6 (n = 11)

* $\bar{x}$ ± SD. There are no significant differences in any of the vertical columns ($p > 0.05$).

Based on these observations, we were concerned to know if in our study the children would have responded differently depending on their different basal serum retinol concentration. The retinol values in our study ranged from 23.6 to 68.3 μg/dl. There were only five children with retinol levels below 30 μg/dl: two in the basal group (29.5 and 26.9 μg/dl), two in the 2-h group (28.0 and 23.6 μg/dl), and one in the 4-h group (27.4 μg/dl). Therefore, in order to examine the above question, we distributed the children in two groups, those below and those above the overall median retinol value of 40.6 μg/dl. Their retinol levels were then analyzed cross-sectionally by one-way analysis of variance at 0, 2, and 4 h after the ingestion of the breakfast meal. No significant differences were found in any of the two groups in relation to the postprandial times of blood collection ($p > 0.05$). The retinol values for the group below the median were, respectively, 35.0 ± 3.6, 35.4 ± 5.7, and 34.9 ± 3.4 μg/dl for 0, 2, and 4 h postprandially ($F = 0.0504$). Those above the median were 51 ± 9.0, 47.1 ± 6.9, and 46.9 ± 5.8 μg/dl ($F = 1.3111$). This observation indicates that within the range of values found in the present investigation, the basal retinol levels did not have any influence on the postprandial serum concentration of retinol.

Although there was not a significant number of children with low serum retinol levels (<30 μg/dl), based on the animal studies already cited (4), it is likely that a similar situation occurs in the general population. It has been described that when an oral dose of vitamin A is given to marginally vitamin A-deficient rats, the retinol absorption peak does not appear in plasma before 5 h, in contrast to the response in vitamin A-sufficient animals, in which this peak appears at an earlier time (between 3 and 4 h). Therefore, if within 4 h there was not a postprandial change in serum retinol in the children of our study, the same phenomenon can be expected in children with initially low serum retinol (<30 μg/dl), in which the maximum rise in plasma retinol would occur beyond this time period.

In conclusion, our study indicates that up to 4 h after a vitamin A-rich meal, there is not a significant increase in children's blood serum concentration of retinol, RBP, and carotenoids. These results provide some assurance that, in practice, particularly under field conditions, the blood samples to be obtained for assessing the vitamin A status of children can be safely collected throughout the morning, regardless of breakfast. Caution should be taken, however, not to extrapolate these results beyond the 4-h period evaluated in the present investigation.

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