

Fat-Soluble Vitamin Supplementation and Serum Levels in Infants and Young Children: A Cross-Sectional Study in China

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Abstract: *Objective:* To investigate the current status of fat-soluble vitamin supplementation in infants and young children in Huai'an, China the impact of different supplementation doses on their serum levels, and the nutritional status of fat-soluble vitamins in different age groups. *Methods:* Full-term infants aged 0-3 years who underwent health examinations at the Department of Child Health Care, Huai'an Maternal and Child Health Care Hospital from November 2017 to March 2018 were included. A self-designed questionnaire was used to investigate the supplementation of vitamins A and D preparations in infants, and high-performance liquid chromatography-mass spectrometry (HPLC/MS/MS) was applied to detect serum levels of fat-soluble vitamins A, D, and E. *Results:* 1. Supplementation of vitamins A, D, and E preparations in infants within the month before the survey: 35.7% (75/210) of infants did not receive vitamin A supplementation, 57.6% (121/210) received intermittent supplementation, and 6.7% (14/210) received daily supplementation; 25.7% (54/210) of infants did not receive vitamin D supplementation, 29.5% (62/210) received intermittent supplementation, and 44.8% (94/210) received daily supplementation; none of the infants received vitamin E supplementation. 2. Serum vitamin A levels in infants who did not receive vitamin A supplementation within the past month were (1.04 ± 0.30) $\mu\text{mol/L}$, those who received intermittent supplementation were (1.01 ± 0.28) $\mu\text{mol/L}$, and those who received daily supplementation were (1.00 ± 0.39) $\mu\text{mol/L}$ (all $p > 0.05$); serum vitamin D levels in infants who did not receive vitamin D supplementation within the past month were (71.33 ± 23.92) nmol/L, those who received intermittent supplementation were (75.40 ± 20.52) nmol/L, and those who received daily supplementation were (81.43 ± 19.29) nmol/L, with the daily supplementation group showing higher levels than the non-supplementation group ($p < 0.05$). 3. Serum vitamin A levels in infants were (1.02 ± 0.29) $\mu\text{mol/L}$, with 39.5% being normal, 55.9% marginally deficient, and 8.6% clinically deficient; serum vitamin D levels were (77.05 ± 21.25) nmol/L, with 90.5% being normal and 9.5% deficient; serum vitamin E levels were 10.24 ± 2.79 mg/L, with 98.6% being normal and 1.4% deficient. 4. Serum vitamin A levels in infants aged < 1 year were (0.89 ± 0.22) $\mu\text{mol/L}$, lower than those in children aged 1–3 years ($p < 0.05$); clinical deficiency of serum vitamin A in infants aged < 1 year was 13.7%, and marginal deficiency was 68.4%, both higher than those in children aged 1–3 years (2.2% and 31.2%, respectively); normal serum vitamin A levels in infants aged < 1 year were 17.9%, lower than those in children aged 1–3 years (66.7%); differences between age groups were statistically significant (all $p < 0.05$). There were no statistically significant differences in serum vitamin D and E

levels and their deficiency rates different age groups of infants (all $p > 0.05$). *Conclusion:* The daily supplementation of vitamins A and D preparations, especially vitamin A, in healthy infants in Huai'an, China during winter remains unsatisfactory; vitamin A deficiency is severe in healthy infants, especially infants, and the nutritional status of vitamin D still needs improvement, while vitamin E deficiency is rare. Pediatric workers should emphasize the importance of vitamin AD supplementation, especially vitamin A supplementation, in infants through publicity and education, and continuously improve the nutritional status of vitamins AD in infants.

Keywords: Infants and young children; Vitamin AD supplementation; Vitamin A; Vitamin D; Vitamin E

Online publication: Aug 31, 2026

1. Introduction

As essential fat-soluble nutrients, vitamins A, D, and E are widely involved in metabolic regulation in the body and play a significant role in the growth and development of infants and young children. They promote bone and visual system development, maintain the integrity of mucosal tissues, and support normal reproductive system function. With the development of the national economy and the widespread promotion of vitamin supplementation, nutritional supplements for infants and young children have become increasingly abundant, providing more avenues for infants to obtain fat-soluble vitamins. However, there are currently few studies on the supplementation of vitamins A and D in infants and young children, the impact of different supplementation doses on their serum levels, and the serum levels of fat-soluble vitamins in different age groups. This study investigated the supplementation of vitamins A and D preparations in infants and young children through questionnaires, measured their serum concentrations of fat-soluble vitamins to understand their nutritional status, and observed the impact of vitamin supplementation interventions on serum levels of vitamins A, D, and E in infants and young children, aiming to provide scientific references for the standardized supplementation of fat-soluble vitamins and clinical nutritional management in infants and young children.

2. Subjects and methods

2.1. Subjects

Full-term infants aged 0–3 years who underwent health examinations at the Department of Child Health Care, Huai'an Maternal and Child Health Care Hospital from November 2017 to March 2018 were enrolled in this study. Infants with infectious diseases, severe congenital heart disease, genetic metabolic diseases, and growth retardation were excluded. This study was approved by the hospital's medical ethics committee, and guardians provided informed consent.

2.2. Methods

2.2.1. Questionnaire

A self-designed questionnaire was used to investigate the supplementation of vitamins A, D, and E preparations in infants within the past month from the parents of the included infants.

2.2.2. Grouping by age

The included infants were divided into two groups: those aged < 1 year and those aged 1–3 years. Grouping by vitamin AD supplementation dose: Non-supplementation group for vitamin A preparations, intermittent supplementation group (daily supplementation of 1–1499 IU of vitamin A preparations), and daily supplementation group (daily supplementation of ≥ 1500 IU of vitamin A preparations); non-supplementation group for vitamin D preparations, intermittent supplementation group (daily supplementation of 1–399 IU of vitamin D preparations), and daily supplementation group (daily supplementation of ≥ 400 IU of vitamin D preparations).

2.2.3. Laboratory testing

Approximately 120 μL of capillary blood was collected as the test sample, and the serum concentrations of vitamins A, D, and E were measured by Beijing Hehe Medical Diagnostics Co., Ltd.'s central laboratory using high-performance liquid chromatography-tandem mass spectrometry (HPLC-MS/MS).

2.4. Vitamin grading criteria

2.4.1. Vitamin A grading criteria

Serum vitamin A < 0.7 $\mu\text{mol/L}$ is considered clinical deficiency, serum vitamin A between 0.7–1.05 $\mu\text{mol/L}$ is considered marginal deficiency, and serum vitamin A ≥ 1.05 $\mu\text{mol/L}$ is considered normal ^[1, 2].

2.4.2. Vitamin D grading criteria

This study refers to the evaluation criteria in China's 2015 "Recommendations for the Prevention and Treatment of Vitamin D Deficiency Rickets", using serum 25-(OH)D levels to assess vitamin D nutritional status: < 50 nmol/L is considered deficiency, 50–250 nmol/L is considered the appropriate range, and > 250 nmol/L is considered vitamin D excess ^[3].

2.4.3. Vitamin E grading criteria

Serum vitamin E < 5 mg/L is considered deficiency, serum vitamin E between 5–20 mg/L is considered normal, and > 20 mg/L is considered excess ^[4].

2.5. Statistical analysis

SPSS 21.0 software was used for statistical analysis. Continuous variables were described using mean $\pm$ standard deviation, and count variables were expressed as the number of cases and percentage. Comparison of means between different groups was performed using analysis of variance, and comparison of rates were compared using the chi-square test. $p < 0.05$ was considered statistically significant.

3. Results

3.1. General information

A total of 210 healthy infants were included, with 117 cases (55.7%) aged < 1 year and 93 cases (44.3%) aged 1–3 years. There were 102 female infants (48.6%) and 108 male infants (51.4%).

3.2. Vitamin A and D supplementation status of infants within the past month

For vitamin A supplementation: 35.7% (75/210) of infants did not receive any vitamin A supplements, 57.6% (121/210) received intermittent supplementation, and 6.7% (14/210) received daily supplementation.

Regarding vitamin D supplementation: 25.7% (54/210) did not receive any vitamin D supplements, 29.5% (62/210) received intermittent supplementation, and 44.8% (94/210) received daily supplementation. None of the infants received vitamin E supplementation.

3.3. Comparison of serum vitamin A and D levels among infants with different vitamin A and D supplementation doses in the past month

For vitamin A, the serum vitamin A levels were (1.04 ± 0.30) $\mu\text{mol/L}$ in the non-supplementation group, (1.01 ± 0.28) $\mu\text{mol/L}$ in the intermittent supplementation group, and (1.00 ± 0.39) $\mu\text{mol/L}$ in the daily supplementation group. There were no statistically significant differences among the groups (all $p > 0.05$). For vitamin D, the serum vitamin D levels were (71.33 ± 23.92) nmol/L in the non-supplementation group, (75.40 ± 20.52) nmol/L in the intermittent supplementation group, and (81.43 ± 19.29) nmol/L in the daily supplementation group. The daily supplementation group had significantly higher serum vitamin D levels than the non-supplementation group ($p < 0.05$).

Table 1. Comparison of serum vitamin A and D levels among infants with different vitamin A and D supplementation doses in the past month

Vitamin A supplementation dose (IU/day)	Vitamin A n(%)	Vitamin A ($\mu\text{mol/L}$)	Vitamin D n(%)	Vitamin D (nmol/L)
None	75 (35.7)	1.04 ± 0.30	54 (25.7)	71.33 ± 23.92^a
Intermittent	121 (57.6)	1.01 ± 0.28	62 (29.5)	75.40 ± 20.52
Daily	14 (6.7)	1.00 ± 0.39	94 (44.8)	81.43 ± 19.29
<i>F</i>		0.255		4.269
<i>P</i>		0.775		0.015

$p < 0.05$ compared with the non-supplementation group.

3.4. Serum Vitamin A, D, and E levels and distribution in infants and young children

The serum vitamin A level in infants and young children was 1.02 ± 0.29 $\mu\text{mol/L}$ (maximum: 2.17 $\mu\text{mol/L}$, minimum: 0.35 $\mu\text{mol/L}$); 83 cases (39.5%, 83/210) had normal levels; 109 cases (55.9%, 109/210) had marginal deficiency; and 18 cases (8.6%, 18/210) had clinical deficiency. The serum vitamin D level was 77.05 ± 21.25 nmol/L (maximum: 141.25 nmol/L , minimum: 27.50 nmol/L); 20 cases (9.5%, 20/210) had deficiency; and 190 cases (90.5%, 190/210) had normal levels. The serum vitamin E level was 10.24 ± 2.79 mg/L (maximum: 25.50 mg/L , minimum: 3.80 mg/L); 3 cases (1.4%, 3/210) had deficiency; and 207 cases (98.6%, 207/210) had normal levels.

3.5. Serum vitamin A, D, and E levels in infants and young children of different age groups

The serum vitamin A level in infants aged < 1 year was 0.89 ± 0.22 $\mu\text{mol/L}$, which was lower than that in the 1–3 year age group (1.18 ± 0.29 $\mu\text{mol/L}$) ($p < 0.05$); the serum vitamin D level in infants aged < 1 year was 78.51 ± 21.60 nmol/L , and that in the 1–3 year age group was 75.22 ± 29.76 nmol/L ($p > 0.05$); the

serum vitamin E level in infants aged < 1 year was 10.15 ± 2.93 mg/L, and that in the 1–3 year age group was 10.35 ± 2.60 mg/L ($p > 0.05$).

Table 2. Comparison of serum vitamin A, D, and E levels in infants and young children of different age groups ($\bar{x} \pm s$)

Age group	Vitamin A ($\mu\text{mol/L}$)	Vitamin D (nmol/L)	Vitamin E (mg/L)
< 1 year (117)	0.89 ± 0.22	78.51 ± 21.60	10.15 ± 2.93
1–3 years (93)	1.18 ± 0.29	75.22 ± 29.76	10.35 ± 2.60
<i>F</i>	68.917	1.242	0.267
<i>p</i>	< 0.000	0.266	0.606

$p < 0.05$ indicates a statistically significant difference between groups.

3.6. Distribution of serum vitamin A, D, and E levels by age group

In the < 1 year infant group, the prevalence of serum vitamin A deficiency was 13.7% (16/117) and marginal deficiency was 68.4% (80/117), both higher than those in the 1–3 year toddler group (2.2% [2/93] and 31.2% [29/93], respectively). The proportion of infants with normal serum vitamin A levels in the < 1 year group was 17.9% (21/117), lower than that in the 1–3 year group (66.7% [62/93]). The differences between the two age groups were all statistically significant (all $p < 0.05$).

For serum vitamin D, in the < 1 year infant group, 10.3% (12/117) had vitamin D deficiency, 89.7% (105/117) had normal vitamin D levels, and no cases of vitamin D excess were observed. In the 1–3 year toddler group, 8.6% (8/93) had vitamin D deficiency, 91.4% (85/93) had normal vitamin D levels, and no cases of vitamin D excess were observed. The difference in vitamin D deficiency rate between the two age groups was not statistically significant ($p > 0.05$).

For serum vitamin E, in the < 1 year infant group, 1.7% (2/117) had vitamin E deficiency, 98.3% (115/117) had normal vitamin E levels, and no cases of vitamin E excess were observed. In the 1–3 year toddler group, 1.1% (1/93) had vitamin E deficiency, 98.9% (92/93) had normal vitamin E levels, and no cases of vitamin E excess were observed. The difference in vitamin E deficiency rate between the two age groups was not statistically significant ($p > 0.05$). Details are shown in **Table 3**.

Table 3. Composition ratios of serum vitamin A, D, and E in different age groups of infants [n(%)]

Age group	Clinical deficiency	Vitamin A			Vitamin D		Vitamin E	
		Marginal deficiency	Normal	Insufficient	Adequate	Excessive	Insufficient	Adequate
<1 year	16 (13.7)	80 (68.4)	21 (17.9)	12 (10.3)	105 (89.7)	0	2 (1.7)	115 (98.3)
1–3 years	2 (2.2)	29 (31.2)	62 (66.7)	8 (8.6)	85 (91.4)	0	1 (1.1)	92 (98.9)
χ^2	8.782	29.689	51.450	0.165	0.436	-	0.148	0.613
<i>p</i>	0.003	<0.000	<0.000	0.814	0.814	-	1	0.631

$p < 0.05$ indicates a statistically significant difference between groups.

4. Discussion

Fat-soluble vitamins are essential nutrients for children's growth and development, playing vital physiological roles in bone development, visual function formation, maintenance of mucosal barriers, and

improvement of reproductive function. Monitoring serum levels of vitamins A, D, and E in children not only evaluates their nutritional status regarding fat-soluble vitamins but also provides a scientific basis for clinical implementation of individualized nutritional supplementation.

4.1. Current status of vitamin A and D supplementation and its impact on serum vitamin A and D levels

According to the “Chinese Dietary Reference Intakes 2013 Edition”, the recommended daily intake of vitamin A for infants aged 0–6 months is 300 µg RAE/day (1000 IU/day), for those aged 7–12 months is 350 µg RAE/day (approximately 1167 IU/day), and for children aged 1–3 years is 310 µg RAE/day (approximately 1033 IU/day)^[2]. Currently, commercially available regular vitamin AD preparations mostly contain vitamin A 1500 IU/capsule and vitamin D 500 IU/capsule (for infants < 1 year old) and vitamin A 2000 IU/capsule and vitamin D 700 IU/capsule (for children > 1 year old). Vitamin D preparations contain vitamin D 400 IU/capsule. This study investigated the recent one-month vitamin AD supplementation status in healthy infants and young children and found that only 6.7% of infants supplemented with 1500 IU of vitamin A daily, while 35.7% did not supplement with vitamin A preparations. For vitamin D, 44.8% of infants supplemented with 400 IU daily, while 25.7% did not supplement with vitamin D preparations. Thus, the supplementation status of vitamins AD in infants and young children in the Huai’an region is not ideal, especially for vitamin A, with a lower supplementation rate. This study also found that the serum vitamin D level in infants and young children supplemented with 400 IU of vitamin D daily was 81.43 ± 19.29 nmol/L, significantly higher than that in the non-supplemented group (71.33 ± 23.92 nmol/L). This indicates that regular supplementation with preventive doses of vitamin D preparations can significantly improve serum vitamin D levels in infants and young children. However, this study found no significant correlation between the daily dose of vitamin A preparations supplemented in infants and young children and their serum vitamin A levels, possibly due to the low proportion (14/210) of infants and young children regularly supplemented with vitamin A preparations included in the study. Further research is planned to expand the sample size to verify the correlation between the daily vitamin A supplementation dose and serum vitamin A levels in infants and young children. Therefore, pediatric workers need to continue to increase publicity efforts and popularize knowledge about vitamin AD supplementation to prevent various clinical problems caused by vitamin AD deficiency in children.

4.2. Serum status of fat-soluble vitamins A, D, and E in infants

Vitamin A is one of the essential nutrients during the growth and development of the body, participating in various physiological processes. Vitamin A deficiency can lead to xerophthalmia, anemia, growth restriction, immune dysfunction, and an increased incidence of infectious diseases^[5]. As one of the four major nutritional deficiency diseases highlighted by the World Health Organization, vitamin A deficiency (VAD) has a relatively high prevalence among children in developing countries. Epidemiological surveys indicate that approximately 250 million children aged 0–4 years worldwide have inadequate vitamin A status^[6].

The clinical manifestations of vitamin A deficiency vary depending on the duration and severity of the deficiency. When body stores of vitamin A decline, the risk of infection increases; when serum levels drop to subclinical deficiency, the risks of recurrent infections and anemia increase, along with growth retardation; when serum concentrations further decrease to clinical deficiency, follicular hyperkeratosis appears on the skin, and in severe cases, secondary infections may occur^[7].

This study found that the normal serum vitamin A level in healthy infants and young children in the Huai'an region was 39.5%, with marginal deficiency at 55.9% and clinical deficiency at 8.6%. Among them, infants < 1 year old had a normal serum vitamin A level of 17.9%, clinical deficiency at 13.7%, and marginal deficiency at 68.4%; children aged 1–3 years had a normal serum vitamin A level of 66.7%, higher than that in infants < 1 year old, with clinical deficiency at 2.2% and marginal deficiency at 31.2%, both lower than those in infants < 1 year old. The above study results show that the vitamin A deficiency rate in infants and young children in the Huai'an region is relatively high, mostly in a subclinical deficiency state, especially in infants < 1 year old, whose vitamin A deficiency rate is significantly higher than that in children aged 1–3 years. The possible reason is that infants' diets are mainly milk-based, and breast milk has a relatively low vitamin A content, and inconsistent standards for vitamin A content in infant formula ^[8,9]. Infants generally start adding green leafy vegetables and animal-based foods rich in vitamin A from 6 months old, but the intake is not much, and their gastrointestinal function is not fully developed, resulting in poor absorption. Additionally, infants grow rapidly and have a high demand for vitamin A, thus increasing the risk of vitamin A deficiency. Approximately two-thirds of children aged 1–3 years have normal serum vitamin A levels, mainly related to their dietary structure. After 1 year old, children's food varieties diversify, and their daily intake of vegetables and animal-based foods rich in vitamin A significantly increases compared to that in infants < 1 year old, and their gastrointestinal function becomes more mature; thus, the vitamin A deficiency rate significantly decreases compared to that in infants < 1 year old.

The main physiological function of vitamin D is to maintain calcium and phosphorus balance in the body, as well as normal neuromuscular function and bone health ^[10]. Vitamin D is one of the essential fat-soluble vitamins for the human body, playing an important role in maintaining health, promoting cell proliferation and differentiation, and participating in immune regulation, nervous system development, reproductive function, endocrine regulation, and growth of epithelial tissues and hair ^[11]. In recent years, a large number of studies have shown that abnormal vitamin D levels are closely related to the occurrence and development of various diseases, such as cardiovascular diseases, autoimmune diseases, and infectious diseases ^[11]. Currently, approximately 1 billion people worldwide have insufficient or deficient vitamin D status, with rapidly growing children and adolescents being a high-risk group. The main sources of vitamin D for the human body are skin synthesis after sunlight exposure and food intake. For infants and young children, milk is the main nutritional source, but breast milk has relatively low vitamin D content, approximately 20 IU/L; if the mother herself has vitamin D deficiency, the vitamin D content provided by breast milk will further decrease ^[12]. Although infant formula usually contains added vitamin D, its content is easily affected by factors such as light, air, temperature, and packaging materials during production, storage, and transportation, leading to a decrease. Some studies have pointed out that even if infants consume 500 mL or more of formula daily, they may still need additional vitamin D supplementation to meet their growth and development needs ^[13]. Moreover, natural foods generally have low vitamin D content. According to the Chinese Dietary Reference Intakes (2013 Edition), the recommended daily intake of vitamin D for infants and young children is 400 IU/day, and it is difficult to meet the body's daily physiological needs solely through dietary supplementation ^[2]. With changes in people's lifestyles, infants and young children have gradually reduced opportunities and time for sunlight exposure. Thus, vitamin D supplementation is the main way for infants and young children to obtain vitamin D ^[14].

This study found that the serum vitamin D deficiency rate in infants and young children in the Huai'an

region was 9.5%, with no significant difference between infants < 1 year old (10.3%) and children aged 1–3 years (8.6%). The above results show that the serum vitamin D nutritional status in infants and young children in the Huai'an region is better than that of vitamin A, but nearly 10% of infants and young children still have vitamin D deficiency. In recent years, with the continuous progress of vitamin D research, its biological functions have been further elucidated, and relevant clinical guidelines have been continuously updated and improved. Vitamin D supplementation strategies have gradually evolved from traditional measures to promote bone development and prevent rickets to comprehensive health interventions that also consider immune regulation, maintenance of metabolic homeostasis, and prevention and treatment of cardiovascular diseases^[15]. Therefore, pediatric workers still need to continue to strengthen publicity efforts, improve parents' awareness and recognition of vitamin D, improve the vitamin D nutritional status of infants and young children, and promote their healthy growth.

Vitamin E has a powerful antioxidant function and also participates in protecting cell membranes and intracellular nucleic acids from free radical attacks, protecting neuromuscular tissue from oxygen free radical damage, and maintaining normal development and function. Vitamin E deficiency can cause adverse symptoms such as reproductive disorders, hemolytic anemia, and neuromuscular movement disorders. Due to the relatively rich vitamin E content in the diet, vitamin E deficiency rarely occurs in individuals with a balanced diet. In this study, only 3 cases (1.4%) of vitamin E deficiency were found in infants and young children, indicating good vitamin E nutritional status in healthy infants and young children in the Huai'an region.

5. Conclusion

In conclusion, the current status of routine vitamin A and D supplementation, especially vitamin A, remains unsatisfactory among healthy infants in Huai'an area during winter. A substantial prevalence of vitamin A deficiency was observed in healthy infants, particularly among those of younger age, whereas vitamin D nutritional status warrants further improvement, and vitamin E deficiency was uncommon. Our findings offer clinical evidence to support the supplementation of fat-soluble vitamins in infants. Pediatric practitioners should reinforce health education and counseling regarding the importance of vitamin A and D supplementation—with particular emphasis on vitamin A, and to make continuous efforts to optimize the vitamin A and D nutritional status of infants.

Disclosure statement

The authors declare no conflict of interest.

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