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Hb level in relation to vitamin D status in healthy infants and toddlers

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Abstract

Objective: To assess the presence of an association between vitamin D deficiency and anaemia in Jordanian infants and toddlers, in whom both vitamin D deficiency and anaemia have previously been proved to be common separately.
Design: Cross-sectional prospective study.
Setting: Department of Paediatrics, Princess Rahma Teaching Hospital, Jordan.
Subjects: Healthy infants and children aged 6–36 months who were seen for primary care.
Results: Out of 203 infants and toddlers included in the study, the anaemia prevalence was 40·4 %. The prevalence of anaemia among infants (*n* 110) was 51·8 %, whereas it was 26·9 % among toddlers (*n* 93). No association between vitamin D status and anaemia was found.
Conclusions: Vitamin D deficiency is not a risk for anaemia in infants and toddlers.

Keywords
Infants
Toddlers
Anaemia
Vitamin D deficiency

Vitamin D, a-fat soluble vitamin, represents vitamin D_2 (ergocalciferol) and vitamin D_3 (cholecalciferol). Vitamin D_3 is found in some natural foods including fish, fish oil and egg yolk, or may be fortified in certain foods such as milk, margarine, butter and infant formula. On the other hand, vitamin D can also be made in skin through solar UV-B (wavelengths of 290–315 nm) irradiation of 7-dehydrocholesterol[1]. The classical function of vitamin D is to maintain Ca and phosphate homeostasis, thereby promoting bone mineralization[2]. In addition, vitamin D plays a role in cellular growth and regulation in many organs[3].

Vitamin D deficiency causes rickets in children and osteomalacia in adults. It also precipitates and exacerbates osteopenia, osteoporosis and fractures in adults.

Long-term vitamin D insufficiency is associated with increased risks of several chronic diseases including cancers, hypertension, heart diseases, diabetes and multiple sclerosis[4,5]. A high prevalence of vitamin D deficiency has been reported in infants, children and adolescents from different countries around the world[6–9]. Vitamin D deficiency might be attributed to low sunlight exposure, dark skin and breast-feeding without vitamin D supplementation[10].

A high degree of association between Fe-deficiency anaemia and vitamin D deficiency in Asian children has been reported, with Fe-deficiency anaemia found to be a significant risk factor for low vitamin D levels in children[11,12]. In another study, thirteen out of seventeen infants with Fe-deficiency anaemia had serum 25-hydroxyvitamin D (25(OH)D) concentration below the normal range despite the fact that these infants received 10 μg vitamin D/d from the age of 1 month. Infants' Hb level, serum Fe as well as 25(OH)D concentrations were increased after they were treated with intramuscular iron dextran[13]. Studies on patients with chronic kidney disease have also supported this association[14,15].

Several mechanisms for such an association have been proposed. Vitamin D may influence Hb through a direct effect on erythropoiesis where it has a synergistic action with erythropoietin; it also increases the storage and retention of Fe and reduces pro-inflammatory cytokines[14–18]. Thus, vitamin D deficiency reduces the ability of red blood cells to become active. On the other hand, it is known that Fe deficiency impairs fat and vitamin A intestinal absorption. Therefore, it is suggested that absorption of vitamin D may also be impaired[12]. Accordingly, it is still controversial which deficiency causes the other; however it is important to be aware of such association especially when proposing treatment.

There are numerous natural sources of vitamin D for nurslings. These include primarily the stores they developed parentally (for newborns), the vitamin D they produce with exposure of their skin to sunlight, and to lesser extent from human milk[19,20].

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Although human breast milk has only a small amount of Fe, anaemia is uncommon in the breast-fed baby. Healthy, full-term babies have sufficient Fe stores in their bodies to last for at least the first 6 months[21,22]. The Fe in breast milk is better absorbed than that from other sources; vitamin C and high lactose levels in breast milk aid in Fe absorption. Fe in mothers' milk is absorbed five times more than that from cows' milk or Fe-fortified formula[21–24].

Vitamin D deficiency and insufficiency are common among children who live in northern Jordan (28·0 % and 28·4 %, respectively)[25]. A significant association between infant feeding practices and vitamin D status was found. Infants who were exclusively breast-fed had higher risk for vitamin D deficiency and insufficiency than those who were bottle-fed[25]. On the other hand, WHO estimates that an average of 52·7 % of Jordanian infants (aged 0·50–0·99 years) and 40·76 % of toddlers (aged 1·00–2·99 years) are anaemic[26].

The present study investigated the existence of a correlation between vitamin D status and anaemia in healthy infants and toddlers living in northern Jordan. The findings of the study are intended to improve treatment protocols for both conditions, and also to attain best understanding of the ideal feeding pattern for children in an attempt to prevent such nutrition-related problems, because inadequate vitamin D status and Fe-deficiency anaemia are considered as major public health problems in Jordan[27].

Materials and methods

Study participants

The final sample consisted of 203 infants and toddlers between the age of 6 and 36 months who were seen between October 2008 and January 2009 for primary care at Princess Rahma Teaching Hospital. This hospital is the main public paediatrics hospital in the city of Irbid, which is located about 85 km north of the capital Amman. The study was approved by the Institutional Review Board at Jordan University of Science and Technology. All parents approved their infants' and toddlers' participation in the study and signed a consent form. Infants with disease that affects the level of vitamin D, such as gastrointestinal, liver or renal diseases, or who consumed vitamin D or Fe supplements were excluded from the study.

One of the parents completed a self-guided questionnaire which included questions on the type of feeding, the duration of breast-feeding, use of supplements and whether the infant has chronic diseases. A detailed description of field data collection can be found elsewhere[25].

Laboratory measurements

About 5 ml of venous blood was collected in a heparinized test-tube from each infant/toddler who participated in the study. Plasma was assayed for 25(OH)D using the enzyme immunoassay method (Immunodiagnostic Systems, UK).

Plasma vitamin D levels were defined as follows: vitamin D deficiency, <50 nmol/l (<20 ng/ml); vitamin D insufficiency, <75 nmol/l (<30 ng/ml); and vitamin D sufficiency, ≥75 nmol/l (≥30 ng/ml). The reference range was provided by the manufacturer of the assay as previously published[28]. Anaemia was defined as Hb < 11 g/100 ml blood using WHO criteria for age 0·50–4·99 years[29]. The participants were divided into two groups according to age: infants aged 6–12 months and toddlers aged >12–36 months.

Statistical analyses

Data were analysed using the SPSS statistical software package version 16·0 (SPSS Inc., Chicago, IL, USA). The χ^2 test was performed to test for difference in anaemia prevalence between participants who had vitamin D deficiency, insufficiency and sufficiency, as well as by feeding pattern. One-way ANOVA followed by the Duncan *post hoc* test was performed to compare the means of continuous variables according to vitamin D status. Findings with $P < 0·05$ were considered to be statistically significant.

Results

Characteristics of the study population

The majority of the study population were males (60·6 %); more than half of them were infants (54·2 %). The majority of the study population had vitamin D sufficiency (normal level), followed by vitamin D deficiency then vitamin D insufficiency. The prevalence of anaemia among the study population was 40·4 %. No statistically significant association between vitamin D status and Hb level was found (Table 1).

Hb level among the study population (total) and stratified by age group

Mean Hb level was 10·81 (SD 1·52) g/100 ml blood for infants and 11·47 (SD 0·98) g/100 ml blood for toddlers. About half of the infants were anaemic, while the vast majority of toddlers were not anaemic; most of the anaemic infants had sufficient vitamin D levels while most of the anaemic toddlers had sufficient and deficient vitamin D levels (Table 2). No statistically significant association between vitamin D status and Hb level was found in either age category (Table 3).

Hb level stratified by type of feeding

Although the average Hb level in the present study tended to be higher in bottle-fed than in breast-fed participants, this increment in Hb level was not statistically significant (Table 4). Similarly, the prevalence of anaemia was higher in breast-fed participants but the difference was also not statistically significant (Table 4).

Table 1 Characteristics of the study population: healthy infants and toddlers aged 6–36 months, Irbid, Jordan, October 2008–January 2009

Characteristic	%	*n*	Mean	SD	*P* value
Gender					
Male	60·6	123			
Female	39·4	80			
Age					
6–12 months	54·2	110			
>12–36 months	45·8	93			
Vitamin D status					
Sufficient	45·8	93			
Insufficient	24·6	50			
Deficient	29·6	60			
Hb category*					
Anaemic	40·4	82			
Non-anaemic	59·6	121			
Hb level (g/100 ml) according to vitamin D status					
Sufficient			11·15	1·3	0·797
Insufficient			11·08	1·4	
Deficient			11·10	1·1	

ANOVA was performed to test the differences between variables; $P < 0.05$ was considered as statistically significant.
*Anaemia was defined as Hb < 11 g/100 ml blood.

Table 2 The prevalence of anaemia stratified by vitamin D status among healthy infants (6–12 months) and toddlers (>12–36 months), Irbid, Jordan, October 2008–January 2009

				Vitamin D status						
				Sufficient		Insufficient		Deficient		
Age group	Hb category*	%	*n*	%	*n*	%	*n*	%	*n*	*P* value
6–12 months (*n* 110)	Anaemic	51·8	57	47·4	27	28·1	16	24·6	14	0·665
	Non-anaemic	48·2	53	50·9	27	20·8	11	28·3	15	
>12–36 months (*n* 93)	Anaemic	26·9	25	40·0	10	20·0	5	40·0	10	0·672
	Non-anaemic	73·1	68	42·6	29	26·5	18	30·9	21	

The χ^2 test was performed to test the differences between variables; $P < 0.05$ was considered as statistically significant.
*Anaemia was defined as Hb < 11 g/100 ml blood.

Table 3 The association of vitamin D status and Hb level stratified by age group among healthy infants (6–12 months) and toddlers (>12–36 months), Irbid, Jordan, October 2008–January 2009

				Hb level (g/100 ml)		
Age group	Vitamin D status	%	*n*	Mean	SD	*P* value
6–12 months (*n* 110)	Sufficient	49·1	54	10·93	1·07	0·503
	Insufficient	24·5	27	10·74	1·71	
	Deficient	26·4	29	10·65	1·42	
>12–36 months (*n* 93)	Sufficient	41·9	39	11·44	1·11	0·816
	Insufficient	24·7	23	11·51	1·13	
	Deficient	33·3	31	11·49	1·03	

ANOVA was performed to test the differences between variables; $P < 0.05$ was considered as statistically significant.

Discussion

The study findings indicate the presence of high anaemia prevalence among our Jordanian infants and toddlers, particularly infants, but there was no statistically significant association between vitamin D status and Hb level. The prevalence of vitamin D deficiency was lower than that of anaemia (29·6 % *v.* 40·4 %).

Our findings are in agreement with Özsoylu and Aytekin, who could not find a relationship between anaemia and rickets due to vitamin D deficiency(30), but disagree with Yoon *et al.*(31) and Grindulis *et al.*(12) who found a significant association of Fe-deficiency anaemia and low vitamin D concentration among children <2 years of age. However no mechanism for this association has been proposed. An association between vitamin D deficiency and a greater risk of anaemia and lower mean Hb was demonstrated in adults(16).

In contrast to Yoon *et al.*(31) and Grindulis *et al.*(12), Heldenberg *et al.*(13) found that severe Fe-deficiency

Table 4 Anaemia prevalence stratified by type of feeding among healthy infants (6–12 months) and toddlers (>12–36 months), Irbid, Jordan, October 2008–January 2009

	Anaemic* (*n* 82)		Non-anaemic (*n* 121)			Hb level (g/100 ml)		
Type of feeding	%	*n*	%	*n*	*P* value	Mean	SD	*P* value
Exclusive breast-feeding (*n* 83, 40·9 %)	42·2	35	57·8	48	0·687	10·98	1·42	0·152
Bottle-feeding (*n* 38, 18·2 %)	34·2	13	65·8	25		11·49	1·16	
Breast-/bottle-feeding (*n* 82, 40·4 %)	41·5	34	58·5	48		11·09	1·31	

The χ^2 test was performed to test the differences between variables; $P<0·05$ was considered as statistically significant.
*Anaemia was defined as Hb < 11 g/100 ml blood.

anaemia in infants aged 1 to 12 months decreased plasma vitamin D concentrations; a finding which was attributed to reduced vitamin D intestinal absorption caused by Fe deficiency. However in a study in rats, Katsumata *et al.*[32] found that severe Fe-deficiency anaemia can affect both bone formation and bone resorption by affecting the synthesis and metabolism of Fe-dependent enzymes, prolyl and lysyl hydroxylases in collagen synthesis and renal 25-hydroxyvitamin D 1-hydroxylase, resulting in the synthesis of abnormal collagen and reduced plasma 1,25-dihydroxycholecalciferol concentration, thus reduced bone mineral density.

In our study we found no correlation between anaemia and early feeding practices; however infants who were exclusively breast-fed had a higher risk for anaemia. This finding is in agreement with Yoon *et al.*[31] who found most of the anaemic children <2 years of age were breast-fed.

The prevalence of anaemia among our Jordanian infants and toddlers aged 6–36 months was found to be 40·4 %; this is lower than the previously published worldwide anaemia prevalence, which covered 48·8 % of the global population of children of pre-school age, of 47·4 (95 % CI 45·7, 49·1) %[33]. In our study the anaemia prevalence in infants 1 year and younger, and among the remaining children older than 1 year, was 51·8 % and 26·9 %, respectively. Previously published WHO results showed relatively similar prevalence of anaemia among Jordanian infants (2003 *v.* 2008) but higher prevalence of anaemia among Jordanian toddlers[26].

The decrease in anaemia prevalence after the age of 1 year may be attributed to several factors. First is the introduction of solid foods and the decreased dependence on milk as the main diet. This is supported by an observational cohort study which demonstrated that infants weaned at <6 months of life had, regardless of milk feeding, higher Hb and ferritin levels than those weaned at >6 months[34]. Second is the high prevalence of anaemia among Jordanian pregnant women who live in the north[35]. Significantly high incidence of anaemia was found among infants born to anaemic mothers in Jordan[36]. This explains the lower prevalence and risk of anaemia in bottle-fed infants compared with those exclusively breast-fed and breast-/bottle-fed. This finding is supported by many previous studies which have demonstrated that breast-feeding is not by itself a causal factor for anaemia[21,23,24]. Therefore it would be important to explore the relationship between anaemia during pregnancy and the development of anaemia among Jordanian infants.

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