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Original Article

Association between the Maternal and Umbilical Cord Blood Vitamin D: A Cross-sectional Study

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ABSTRACT

Background: Vitamin D has multifaceted effects on the mother's health and fetus. Since its deficiency can cause numerous maternal and fetal complications, this study aimed to investigate the relationship between the maternal and umbilical cord blood vitamin D.

Methods: This cross-sectional study was conducted on 48 pregnant mothers and their 48 babies in Hafez Hospital, Shiraz, in 2021. First, 5cc the blood clot was taken from the mother and the baby's umbilical cord after delivery. Also, a checklist included the mother's age, pregnancy conditions, gender, weight, height, head circumference, age of baby teeth eruption, and the first and fifth minute Apgar score of their babies was filled. Data were analyzed using SPSS-26.

Results: The mean of vitamin D from the umbilical cord blood (35.95±11.97), and from mothers' serum (21.85±7.3); were correlated significant (r=0.80, P<0.001). The mean vitamin D serum level of mothers who received supplements during pregnancy (23.72±6.04) was higher than mothers who did not use them during pregnancy (8.29±19.45) (P<0.05). There was a significant relationship between the mother's vitamin D and umbilical cord vitamin D with the sprouting of the first milk tooth.

Conclusion: In this study, most mothers reported vitamin D deficiency, and their vitamin D levels were directly correlated to the umbilical cord vitamin D. These findings show the importance of monitoring the mother's vitamin D to indicate the vitamin D status of the fetus and encourage mothers to make effective use of sunlight and the enrichment food with vitamin D.

Keywords: Pregnancy, Infant, Mothers, Umbilical cord blood, Vitamin D deficiency

Introduction

Vitamin D, as an important micronutrient, has a vital role in homeostasis of calcium and bone health (1). In various studies, many diseases and disorders are related to vitamin D deficiency, including food allergies and asthma (2), cardiovascular diseases, metabolic syndrome and blood pressure (3), tooth decay (4), rheumatic diseases such as RA (5), high blood sugar and type-1 diabetic mellitus (6), mood disorders, and premenstrual syndrome (7). Also, some malignancies such as breast (8), colon (9), and prostate cancers (7) have been directly related to vitamin D deficiency.

The most common reason of vitamin D deficiency is poor intake through nutrition. Limited food sources such as salmon, tuna, fish oil, caviar, cheese, milk, mushrooms, and enriched foods contain vitamin D (10). Due to the dependence of vitamin D metabolism in the body on ultraviolet rays, the storage of vitamin D in the human body be influenced by on the amount of exposure to sunlight and receiving supplements containing vitamin D (10-12).

Many studies consider a serum level ≥30 ng/ml as an adequate vitamin D level (1,2,13). Vitamin D has important and multifaceted effects

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on pregnancy and the mother and fetus health. Vitamin D deficiency in pregnancy can cause numerous maternal and fetal complications (14).

Unfortunately, vitamin D deficiency is increasing worldwide (10, 15). In Iran, some studies have reported a prevalence of 40%, and vitamin D deficiency was reported up to 80% in different parts of Iran (16). In these studies, it has also been shown that women suffer from more deficiency than men (15-17).

Considering the importance of the role of vitamin D on the health of the mother and fetus, the present study aimed to investigate the relationship between the amount of maternal vitamin D and vitamin D in the umbilical cord blood of babies born in Hafez Hospital in Shiraz.

Methods

Study design

This descriptive-analytical cross-sectional study was conducted on 48 pregnant mothers and their 48 babies in Hafez Shiraz Hospital in 2021. In each of the four seasons of the year, 12 babies and 12 pregnant mothers were sampled; six women with normal delivery and six women with cesarean section were randomly selected.

Pregnancies associated with stillbirth, molar pregnancy, gestational age less than 35 weeks, and severe medical diseases of the mother were the exclusion criteria. Moreover, mothers with epilepsy who were taking antiepileptic drugs interfering with metabolism of vitamin D, including phenytoin, or mothers with organ failure such as kidney (under dialysis) or liver were excluded. Babies who had asphyxia or problems requiring NICU admission or had major congenital anomalies were also excluded.

Data collection

At the time of hospitalization, 5cc of blood clot was taken from the mother. After delivery, 5cc of clot blood was taken from the baby's umbilical cord. Also, along with the sampling, a checklist included information such as birth date, mother's age, phone number, and delivery method, number of pregnancies, and mother's education and addiction history, which was filled out with a face-to-face interview. Also, information about the baby, such as gender, weight, height, head circumference, and the first and fifth-minute Apgar scores were included in the checklist. Gestational age was determined based on an ultrasound performed in the first trimester.

Vitamin D was measured by HPLC in terms of ng/ml. Based on the amount of vitamin D, the participants were divided into three groups:

1. Vitamin D deficiency: Vitamin D serum level ≤ 20 ng/ml
2. Vitamin D insufficiency: 20.01 ng/ml < Vitamin D < 29/90
3. Vitamin D sufficiency: Vitamin D level ≥ 30 ng/ml

It is necessary to mention the results of the vitamin D test were given to the mothers by telephone contact. In vitamin D deficiency cases, they were recommended to use supplements of vitamin D supplements regularly, according to the national protocol and to use sunlight, and follow-up was done. Also, at the end of six months of each baby, the mothers were contacted and the presence or time of eruption of the baby's first milk tooth were asked.

Statistical Analysis

To data analyze, SPSS-26 version were used. To describe the analysis, we used the frequency and percentage frequency for qualitative variables, and the mean and standard deviation (SD) were used for quantitative variables. The Kolmogorov-Smirnov test was used to determine the normal distribution of the data. Then, ANOVA and T-student statistical tests were used to associate demographic variables with the mother and baby's vitamin D, depending on the normality of the data. Moreover, Pearson's correlation coefficient was used to investigate the correlation between mother's and baby's vitamin D and with the first and fifth minute Apgar scores. Fisher's Exact Test was also used for relationship of categorized variables. The significance level of P-value less than 0.05 was considered in all tests.

Ethical approval

The protocol of this study was approved by the Local Ethics Committee of Shiraz University of Medical Sciences (Code: IR.SUMS.REC.1400.126). Informed consent was also took from all parents.

Results

In the present study, 48 mothers and 48 babies participated; 45.8% of the babies were boys and 54.2% were girls. Table 1 shows the other demographic information of the study participants. In this study, 7% of mothers and 35% of infants had sufficient vitamin D, and the rest suffered from vitamin D deficiency (Table 2). Also, the mean of vitamin D of the umbilical cord of the studied babies was 35.95±11.97, and that of the mothers' serum was 21.85±7.3; there was a direct and significant relationship between these

Table 1. Demographic information of the studied population

Variables		Frequency	Percentage
Gender	Male	22	45.8%
	Girl	26	54.2%
Gravid	1	18	37.5%
	2	13	27.1%
	≥3	17	35.4%
Maternal education	> Diploma	23	47.9%
	Diploma	13	27.1%
	Postgraduate diploma	3	6.3%
	Bachelor's degree	9	18.8%
Mother's age (years)	15-20	5	10.4%
	21-25	14	29.2%
	26-30	10	20.8%
	> 30	19	39.6%
Place of residence	City	26	54.2 %
	Town	14	29.2%
	Village	8	19.7%
Birth weight (gr)	≤2500	6	12.5%
	>2500	42	87.5%
	<37	7	14/6%
Gestational age (weeks)	37-40	37	77.1%
	>40	4	8.3%
First minute Apgar score	≤7	3	6.3%
	>7	45	93.8%
5th minute Apgar score	≤7	2	4.2%
	>7	46	95.8%

two variables (r=0.80, P<0.001). Table 3 shows the relationship between three categories of vitamin D deficiency between mothers and neonates.

In this study, there was no statistically significant relationship between the mother's vitamin D level and the season of delivery (P=0.84), type of delivery (P=0.93), gender of the baby (P=0.15), gestational age (P=0.61), birth weight of the baby (P=0.07), mother's age, education, and place of residence, respectively (P=0.64), (P=0.26), and (P=0.77).

Moreover, there was no statistically significant relationship between the mean of vitamin D of the baby's umbilical cord and the delivery season (P=0.40), type of delivery (P=0.40), the baby's gender (P=0.47), gestational age (P=0.98), and regular use of vitamin D supplement in mothers (P=0.11), mother's age, education, and place of residence, respectively (P=0.38), (P=0.78), and (P=0.37). However, the relationship between the mean of vitamin D of the baby's umbilical cord and baby's birth weight was significant (P<0.05) (Table 4).

Pearson's correlation coefficient showed no statistically significant correlate between the mother's vitamin D level and the first minute Apgar score (r=0.11, P=0.44) and fifth minute Apgar score (r=-0.02, P=0.88).

Furthermore, there was no statistically significant correlation between the mean of vitamin D of the baby's umbilical cord and the first minute Apgar score (r=-0.03, P=0.79) and fifth minute Apgar score (r=-0.22, P=0.13).

Of the studied participants, 27 mothers (56.3%) were taking supplement of vitamin D according to the national guidelines of Iran (one jelly tablet of 1000 units of vitamin D per day from the beginning of pregnancy) and 21 mothers (43.8%) were not taking supplement of vitamin D during pregnancy. The mean of vitamin D serum level of mothers who received this supplement

Table 2. Analysis of vitamin D in the mother's serum and the baby's umbilical cord

Vitamin D level	Infant's first milk tooth		
	Category	Frequency	percent
In the mothers (ng/ml)	≤20	24	50%
	20.01-29.9	17	35.4%
	≥30	7	14.6%
In the babies' umbilical cord (ng/ml)	≤20	6	12.5%
	20.01-29.9	7	14.6%
	≥30	35	72.9%

Table 3. The relationship between vitamin D in the mother's serum and the baby's umbilical cord

Vitamin D in mothers' serum (ng/ml)	Vitamin D in the umbilical cord of newborns (ng/ml)			
	≤20	20.01-29.9	≥30	P value*
≤20	6	5	13	
20.01-29.9	0	2	15	0.047**
≥30	0	0	7	

* Fisher's Exact Test

** P-value is considered significant < 0.05.

Table 4. The relationship between the maternal and umbilical cords vitamin D with demographic characteristic of the studied population

Variables		Maternal vitamin D Mean ± SD	P Value	Umbilical cords vitamin D Mean ± SD	P Value
Season delivery	Spring	21.04±7.74	0.84	32.07±13.46	0.40*
	Summer	23.53±9.89		34.25±14.10	
	Fall	21.44±6.52		37.64±11.86	
	Winter	21.39±5.16		39.70±7.30	
Gender	Male	23.26±6.48	0.15	37.06±11.29	0.47**
	Girl	20.19±8.10		34.56±12.86	
Type of delivery	Normal	21.94±8.15	0.93	34.44±13.02	0.40**
	Cesarean section	21.76±6.64		37.38±10.90	
Maternal education	< Diploma	23.46±6.86	0.26	37.57±11.87	0.78*
	Diploma	22.21±8.42		34.82±12.21	
	Postgraduate diploma	16.73±3.76		30.90±12.33	
	Bachelor's degree	18.93±7.16		34.93±13.10	
Mother's age (years)	15-20	17.80±5.28	0.64	27.72±8.72	0.38*
	21-25	22.28±8.55		35.34±13.67	
	26-30	22.67±7.39		36.39±11.73	
	> 30	22.17±7.05		38.25±11.34	
Place of residence	City	21.14±7.34	0.77	35.35±12.09	0.92*
	Town	22.64±8.17		36.90±13.18	
	Village	22.75±6.52		36.02±10.65	
Birth weight (gr)	<2000	17.40±0.61	0.07	34.38±2.62	<0.05*
	2000-2500	20.83±7.81		41.02±11.94	
	2500-3000	25.21±7.67		41.78±11.31	
	>3000	19.59±6.42		29.83±10.35	
Gestational age (weeks)	<37	21.04±6.28	0.61	35.97±5.03	0.98*
	37-40	22.35±7.70		36.00±12.63	
	>40	18.65±6.24		35.02±16.70	

*One-way ANOVA
**Independent T-Test

during pregnancy (23.72±6.04) was higher than mothers who did not use the supplement or did not receive it regularly during pregnancy (8.29±19.45); the difference was statistically significant (P<0.05).

In the present study, all 48 examined infants were monitored at the end of 6 months for the eruption of the first milk tooth. Twenty-seven babies (56.3%) had their first milk tooth sprout by the end of six months, and 21 babies (43.8%) did not have a milk tooth yet. There was a statistically significant relationship between the mother's vitamin D level and umbilical cord vitamin D with the sprouting of the first milk tooth (Table 5).

Table 5. The relationship between mother's vitamin D level and the sprouting of the infant's first milk tooth in the first 6 months of life

Vitamin D level	Infant's first milk tooth			
	Category	Yes	No	P value*
In the mothers (ng/ml)	≤20	4	20	<0.001
	20.01-29.9	16	1	
	≥30	7	0	
In the umbilical cord of babies	≤20	0	6	<0.001
	20.01-29.9	1	6	
	≥30	26	9	

* Fisher's Exact Test

Discussion

In this study, the mean vitamin D levels of mothers and infants and their relationship with each other and with other demographic variables were investigated. The results of the study

showed that vitamin D deficiency is highly prevalent among the studied mothers. Half of the mothers had severe vitamin D deficiency (<20 ng/ml). During pregnancy, given the mother's need for more calcium and vitamin D to maintain the natural homeostasis of the mother's body and the placenta function, many researchers consider vitamin D levels more than 30 ng/ml to be ideal during pregnancy for the development and growth of the fetus and the needs of the mother (1, 2, 13). According to the results, the prevalence rate of 85.4% of maternal serum vitamin D less than 30 ng/ml was worrying.

Unfortunately, vitamin D deficiency is a health problem worldwide (10, 15). In a meta-analysis study, Palasires et al. reported that vitamin D deficiency was very common in pregnancy (12). A systematic review and meta-analysis study in Iran reported the mean concentration of vitamin D in women with pregnancy as 15.02 and in babies as 14.59 (9). Our study is in line with the results of this meta-analysis.

A study by Moradzadeh et al. revealed that 47.85% of women in Kurdistan were suffering from vitamin D deficiency and indicated that the prevalence of vitamin D deficiency in Iran was similar to the Middle East (16).

In the present study, there was a significant direct relationship between the serum vitamin D levels of mothers and vitamin D in the newborn's umbilical cord. In this regard, other studies, including a Turkey cross-sectional study (18), a prospective study by Cetinkaya et al. (19), a case-control study on Asian infants by OG Brooke et al. (20), and a meta-analysis study on pregnant Iranian women (9) also showed this direct relationship.

In the present study, the mean of vitamin D in the mothers' serum was higher in the summer season than in other seasons. However, there was no relationship between the serum vitamin D of mothers and the season. In a case study in Tabriz, the vitamin D deficiency in infants born to mothers who spent the third trimester of pregnancy in winter was higher, so the researchers emphasized the need to use supplement of vitamin D, especially during cold seasons to maintain status of the mother's vitamin D and prevent infantile rickets (21). Holick et al. also showed that seasonal changes, and body coverage were effective in the prevalence of vitamin D deficiency (11). Among the reasons for the lack of correlation between vitamin D and season in this study, we can mention the possibility of more sampling at a point in the season or the moderate climate of Shiraz and Fars province, where there are many sunny days even in the cold seasons.

In our study, there was no correlation between the method of delivery and the amount of vitamin D in maternal serum and in the umbilical cord. Similarly, Abbasian et al. also revealed that the mean of vitamin D of the mother's serum and umbilical cord had no significant difference with the type of delivery (22). Meanwhile, Barrett et al. in their study found that maternal vitamin D levels to be lower in babies who were born through cesarean section compared to babies who were born via normal vaginal delivery (14).

There was no significant difference between the levels of vitamin D and the gender of the baby, and this result is similar to the researches by Abbasian et al. (22) and Salek et al. (23). Despite the results of this study, in the meta-analysis study by Agahjafari et al., revealed that there was a significant relationship between maternal vitamin D and a newborn's umbilical cord with gestational age (24). In a meta-analysis study, researchers concluded that high levels of vitamin D in the mother can be a protective factor to prevent prematurity (25). Since vitamin D is mainly transferred to the fetus in the third trimester of pregnancy (26) and in the present study, infants with very low gestational age who needed hospitalization in the NICU were not included, the lack of association between vitamin D and gestational age could be due to the absence of infants with low gestational age.

Similar to this research, in the studies of Abbasian et al. (22) and Salek et al. (23) there was no significant relationship between vitamin D in the mother's serum or newborn's umbilical cord with the mother's age, place of residence, and education. Similar to this study, a meta-analysis study by Palacios et al, indicated that taking of vitamin D supplements during pregnancy was related to the vitamin D levels of mothers and their babies. These researchers noted that the adverse risk of pregnancy and neonatal consequences including preeclampsia was reduced with vitamin D supplementation (12).

A randomized clinical trial, by Mojibian et al., found no significant relationship between low levels of vitamin D in women with pregnancy and the 5-minute Apgar score of infants (27); this result is consistent with our findings.

In the results of the study by Salek et al. (23) and Akhlaghi et al. (28) as well as the results of this study, there was no correlation between the number of pregnancies and vitamin D deficiency

of the mother. This result can be because most of the women in his study were primiparous (28).

The essential role of vitamin D and calcium in the development of teeth is a clear and proven fact (26). Misra et al. showed in a study that premature babies who had lower levels of vitamin D were more exposed to enamel tissue defects, both in primary and permanent teeth (29).

The present study revealed a significant relationship between the mean serum vitamin D of the mother and the age at which the first milk tooth erupted. Similarly, a study conducted in India in 2020 by Jairam et al. showed a strong connection between maternal serum vitamin D during pregnancy and delayed primary teeth development in infants (30). Milk teeth grow in most children between 4 and 7 months. The growth of milk teeth is a factor in evaluating the optimal growth of the child and the sufficiency of vitamin D and calcium in the infant's body (26). Given that all the participated infants, according to their mothers, received vitamin A+D drops according to the national protocol, it can be stated that the provision of sufficient vitamin D in mothers during pregnancy and after that. An adequate reserve of vitamin D in the baby can be effective in the process of optimal growth and health of the teeth.

Conclusion

In this study, mothers had a high level of vitamin D deficiency and level of their vitamin D was directly associated to vitamin D in the umbilical cord. These findings show the importance of regulating the mother's vitamin D, especially in the third trimester of pregnancy, when vitamin D is most transferred to the placenta and fetus. It can be said that the mother's vitamin D monitoring can indicate the status of fetus's vitamin D.

Also, poor correlation between the mother's serum vitamin D and the mother's age, place of residence (city or village), and education showed that vitamin D deficiency was a health problem at all social, cultural, and economic levels. These results recommend increasing society's awareness of the importance of vitamin D and the complications of its deficiency, use of vitamin D supplements, especially in high-risk groups including women with pregnancy and infants, and encouraging mothers to make effective use of sunlight and the enrichment of food with vitamin D.

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Conflicts of interest

The authors declare that they have no competing interests.

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