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Original Research Article

A Study of Association between Maternal Vitamin D Levels with Neonatal Anthropometry and Clinical Outcome in a Rural Tertiary Care Center

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Conflict of interest: Nil

Abstract

Introduction: Maternal vitamin D plays an important role in fetal growth and neonatal health. Its association with neonatal anthropometry and clinical outcome remains uncertain. This study evaluated maternal vitamin D status and its relationship with neonatal vitamin D level, anthropometric measurements, and neonatal outcome.

Methods: This hospital-based prospective observational study included 90 pregnant women and their newborns delivered at a tertiary care hospital in Rajasthan between April 2021 and March 2022. Maternal and neonatal serum vitamin D levels were measured using chemiluminescence immunoassay. Maternal background characteristics, gestational age, birth weight, neonatal anthropometry, NICU admission, and neonatal outcome were recorded. Statistical analysis included Chi-square test, Pearson correlation, and receiver operating characteristic (ROC) analysis.

Results: The mean maternal and neonatal vitamin D levels were 36.60 ± 13.96 ng/mL and 39.07 ± 13.70 ng/mL, respectively. Maternal vitamin D was sufficient in 74.44% of mothers, 85.56% of newborns had normal vitamin D levels. Maternal vitamin D status was not significantly associated with maternal background characteristics, gestational age, and birth weight, weight for gestational age, neonatal length, head circumference, NICU admission, or neonatal outcome (all $p > 0.05$). Very strong positive correlation was observed between maternal and neonatal vitamin D levels ($r = 0.95$, $p < 0.001$). Maternal vitamin D cut-off of >17.5 ng/mL predicted normal neonatal vitamin D status with 98.75% sensitivity, 100.00% specificity, and 98.89% diagnostic accuracy.

Conclusion: Maternal vitamin D level was strongly associated with neonatal vitamin D level and accurately predicted neonatal vitamin D status. No significant association was found between maternal vitamin D status and neonatal anthropometry or short-term neonatal outcome in this study.

Keywords: 25-Hydroxyvitamin D, Newborn, Pregnancy, Vitamin D Deficiency.

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Introduction

Secosteroids, also known as Vitamin D is a fat-soluble vitamin that is plays a major role in maintaining the homeostasis of calcium and phosphorus and required for growth.[1]

Vitamin D is produced from 7-dehydrocholesterol in the epithelial cells during exposure to ultraviolet B radiation from the sun and this is the major source of vitamin D in most of the people. Naturally vitamin D is present in fish liver oil, egg yolk, fatty salmon, but it is deficient in breast milk (12-60 IU/L). In developed countries, children get vitamin D mostly from formula milk and fortified foods. [2] Maternal vitamin D deficiency affects the neonate and may cause gestational age (SGA), premature birth, impaired bone and tooth development, and an increased risk of infectious illnesses.[3] Severe vitamin D deficiency is a very common problem and its prevalence varies from 5.9% in US [4], 7.4% in Canada [5], and 13% in Europe [6]. Age, ethnicity, geographical region affects this variation.[7] It is estimated that 490 million people in India are vitamin D deficient.[6] Various previous Indian studies done on apparently health people from the community reported a wide range of prevalence from 34.5% to 90% which included people from different age groups.[8–11]

A school-based study on premenarchal girls (n = 214) in Pune was conducted by Kadam et al. in

2011. It showed a prevalence of 34.2% of Vitamin D.[12] Another school-based study done by Kapil et al. in 2017 on 1222 school children aged 6–18 years in Kangra and Kullu districts of Himachal Pradesh, showed a prevalence of 81% and 80% respectively.[13]

Hence, Fulfilling the requirements for normal vitamin D levels during pregnancy is important and recognizing the risk factors of vitamin D deficiency and ensuring primary diagnosis of vitamin D status during pregnancy is essential to prevent LBW & other complications in a newborn, Therefore The target population of this study was pregnant mothers from tribal area of south Rajasthan with a focus on finding out to the association between maternal vitamin D levels and low birth weight or clinically unwell offspring.

Materials and Methods

The current hospital-based observational prospective study was conducted in Obstetrics wards & NICU of Pacific Institute of Medical Sciences, Umarda, Udaipur, (Rajasthan) between April 2021 and March 2022. All pregnant mothers admitted to Obstetrics wards with history of vitamin D and calcium supplementation during antenatal visit and their respective newborns delivered during our study period were included in this study. Mothers with pre-eclampsia or eclampsia and the mothers who were being treated with phenobarbitone, phenytoin, or any other anti-epileptics were also excluded from this study.

Sample size was calculated taking 85% as the combined prevalence of maternal vitamin D severe deficiency, deficiency and insufficiency as reported by Sathish P et al. [14] with a 95% confidence level and 7.5% absolute precision. The calculated sample size was 87 and after adjusting for 10% attrition, the final required sample size was 96. Out of the 96 pregnant mother included initially, blood from 6 new-born could not be collected as either they were shifted elsewhere or withdrew consent, so the final calculations were done on data of 90 participants.

First, the purpose of the study was explained to the mother or the father/primary caregiver of study subjects in the local language, who fulfilled the inclusion criteria and whose parents gave written informed consent, who were enrolled for the study. A detailed antenatal history was taken including age, parity, socio-economic status, diet, drug usage etc was recorded. Gestational age was calculated by LMP & EDD and ultrasonography. Mother's samples were collected bedside within 24 hours of admission to obstetrics wards of our hospital in plain vials and were sent to the biochemistry lab for evaluation of serum vitamin D levels by Total Vitamin D (Vit-D) Chemiluminescence Immuno Assay (CLIA) technique. BMI was classified according to WHO Asian population[15] and Socio-economic status according to Modified B.G. Prasad classification[16]. Vitamin D deficiency is defined as vitamin D level less than 50 nmol/L (20 ng/mL) of 25-hydroxyvitamin D, 25(OH)D, and insufficiency level is between 51 and 74 nmol/L (21-29 ng/mL).[17]

After the baby was born, a sample of serum vitamin D of the newborn was also collected bedside and sent for analysis by the same technique, and anthropometry and the clinical status of the newborn were also recorded. Classification of neonatal vitamin D status was based on serum 25-hydroxyvitamin D [25(OH)D] concentration: deficient(<10 ng/mL), insufficient(10-19 ng/mL), normal (20-60 ng/mL), and hypervitaminosis D/toxicity (>60 ng/mL)[18].Gestation age was assessed by the new Ballard score. Neonates were divided based on their gestational age into full-term neonates (≥37 weeks Gestational Age) and preterm neonates (<37 weeks' gestational age). Also, neonates were divided based on their birth weight into normal birth weight neonates (full-term between 2,500-3,500 g or preterm >10th percentile) and LBW neonates (full-term <2,500 g or preterm <10th percentile of the population).[19]

This study was approved by the Ethical and Research Committee, Pacific Institute of Medical Sciences, Udaipur, Rajasthan. For the patients who fulfilled the selection criteria, their parents were informed in detail about the study and written informed consent was obtained before enrolment.

Continuous variables were expressed as mean ± standard deviation or median (IQR) based on whether the data was parametric or non-parametric in nature. Ordinal and nominal variables were expressed as frequency and percentage. Association was assessed using chi-square test of association or Fischer exact test as applicable. Pearson/spearman correlation coefficient was used to assess the relationship between changes in maternal and neonatal vitamin D levels. ROC (Reciever Operating Curve) was used to assess the optimal maternal vitamin D cutoff for prediction of neonatal vitamin D level along with diagnostic accuracy. A p value <0.05 was considered statistically significant. Statistical analysis was performed using Jamovi.

Result

In the present study, total 90 pregnant mother and their new-borns were included with mean maternal age of 24.06 ± 6.24 years. Mean vitamin D level among the mothers and new-born was 36.60 ± 13.96 ng/mL and 39.07 ± 13.70 ng/mL respectively. Mean birth weight of the newborns was 2.64 ± 0.57 kg. 67 (74.44%) of the mothers had sufficient vitamin D levels (mean±SD:

43.09±8.53 ng/mL), 14 (15.56%) were deficient (mean±SD: 12.14±5.08 ng/mL), and 9 (10.00%) had insufficient (mean±SD: 26.33±3.04 ng/mL) vitamin D. Among newborns, vitamin D was normal in 77 (85.56%) (mean±SD: 41.55±9.83 ng/mL), insufficient among 7 (7.78%) (mean±SD:15.57±3.10 ng/mL), deficient in 3 (3.33%) (mean±SD: 6.33±3.06 ng/mL), and 3 (3.33%) had hypervitaminosis D/toxicity (mean±SD: 63.00±3.46ng/mL).

Table 1: Distribution of the newborns according to their neonatal vitamin D level and maternal background characteristics (N = 90)

Maternal background characteristics		Neonatal vitamin D level		Total (N=90) [n (%)]	χ^2 (df)	P* value
		Less than normal (n=10) [n (%)]	Normal (n=80) [n (%)]			
Age	>30 years	3 (16.67)	15 (83.33)	18 (100.00)	0.70 (1)	0.40
	≤30 years	7 (9.72)	65 (90.28)	72 (100.00)		
Parity	>2	2 (20.00)	8 (80.00)	10 (100.00)	0.90 (1)	0.34
	≤2	8 (10.00)	72 (90.00)	80 (100.00)		
Residence	Rural	0 (0.00)	75 (100.00)	75 (100.00)	-	-
	Urban	10 (66.67)	5 (33.33)	15 (100.00)		
Socioeconomic status	Lower-middle & Lower	6 (10.71)	50 (89.29)	56 (100.00)	0.02 (1)	0.88
	Middle and above	4 (11.76)	30 (88.24)	34 (100.00)		
Diet	Non-vegetarian	3 (7.32)	38 (92.68)	41 (100.00)	1.10 (1)	0.29
	Vegetarian	7 (14.29)	42 (85.71)	49 (100.00)		
BMI	Normal/Obese	3 (11.54)	23 (88.46)	26 (100.00)	0.01 (1)	0.93
	Underweight	7 (10.94)	57 (89.06)	64 (100.00)		

***Chi-square test of association or Fischer exact test as applicable**

Majority of the mothers were aged ≤30 years (80.00%), residing in rural area (83.33%), belonged to Lower-middle & Lower Socio-economic class ((62.22%), vegetarian (54.44%) and underweight (71.11%). Less than normal neonatal vitamin D was found in mothers aged >30 years, having parity >2, residing in urban area, belonging to Lower-middle & Lower Socio-economic class, consuming vegetarian diet and of normal or obese BMI category.

Although none of the background maternal characteristics were found to be statistically significant with neonatal vitamin D level.[Table-1]

Table 2: Association of maternal vitamin D status with gestational and neonatal anthropometric characteristics (N = 90)

Gestational and neonatal anthropometric characteristics		Maternal vitamin D		Total (N = 90)	Test statistics (df)	p value
		Less than normal (n = 23)	Normal (n = 67)			
Gestational age [n (%)]	Preterm	4 (36.36)	7 (63.64)	11 (100.00)	0.77 (1)	0.38
	Term/Post-term	19 (24.05)	60 (75.95)	79 (100.00)		
Weight for gestational age [n (%)]	AGA/LGA	14 (23.73)	45 (76.27)	59 (100.00)	0.30 (1)	0.58
	SGA	9 (29.03)	22 (70.97)	31 (100.00)		
Birth weight [n (%)]	LBW	9 (28.13)	23 (71.88)	32 (100.00)	0.17 (1)	0.68
	Normal Birth Weight	14 (24.14)	44 (75.86)	58 (100.00)		
Length (cm)[mean±SD]		48.13±4.71	47.76±4.21	47.86±1.76	0.35 (88)	0.73
Head circumference (cm) [mean±SD]		33.09±1.81	33.24±1.76	33.22±1.77	0.36 (88)	0.72

Preterm birth was noted in 4 (36.36%) mothers with low serum vitamin D as compared to 7 (63.64%) mothers with normal vitamin D levels, term/post-term delivery was reported in 19 (24.05%) and 60 (75.95%) mothers, respectively, but this difference was not statistically significant (χ^2 = 0.77, df = 1; p = 0.38). 9 (29.03%) of the SGA (Small for Gestational Age) babies were born to mothers having less than normal vitamin D levels and 22 (70.97%) in mothers with normal vitamin D levels (χ^2 = 0.30, df = 1; p = 0.58). Mothers of 9 (28.13%) low birth weight newborns had low vitamin D levels and 23 (71.88%) had with normal vitamin D levels, with no significant association (χ^2 = 0.17, df = 1; p = 0.68). The mean neonatal length was 48.13 ± 4.71 cm in mothers having low vitamin D and 47.76 ± 4.21 cm in normal group (p = 0.73). The corresponding mean head circumference was 33.09 ± 1.81 cm and 33.24 ± 1.76 cm, respectively (p = 0.72). Maternal vitamin

D status was not significantly associated with gestational age, birth weight, weight for gestational age, neonatal length, or head circumference. [Table 2]

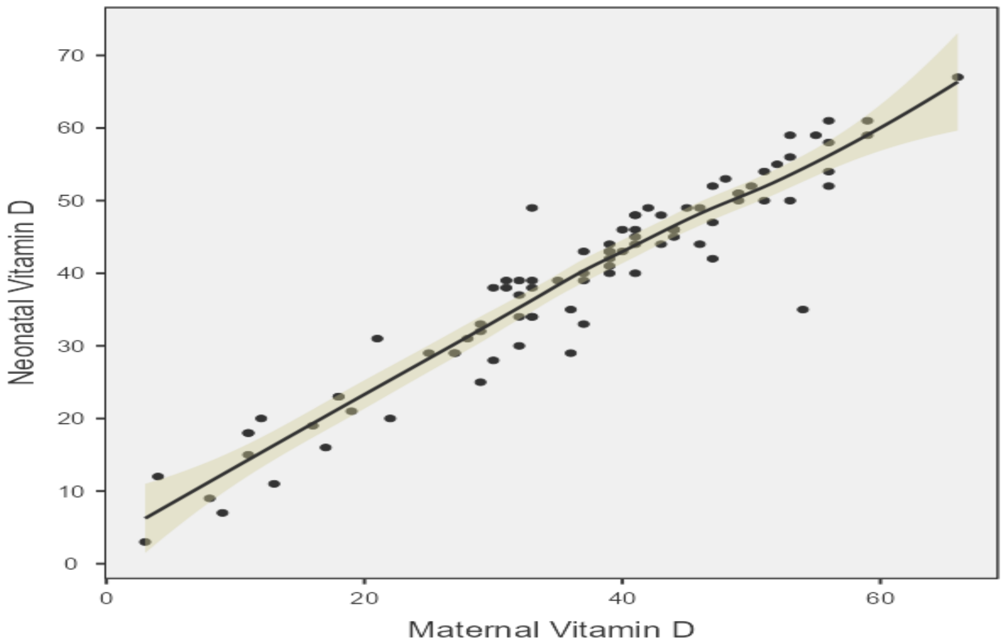

Figure 1: Scatter plot showing the relationship between maternal and neonatal vitamin D levels (N=90).

Figure 1. Shows the relationship between maternal and neonatal vitamin D levels. There was a very strong positive correlation between the two variables (Pearson's r = 0.95, p < 0.001). As maternal vitamin D levels increased, neonatal vitamin D levels also increased. The points on the scatter plot were closely clustered around the fitted regression line, indicating a strong linear relationship.

The correlation was statistically significant, with a 95% confidence interval of 0.93 to 0.97.

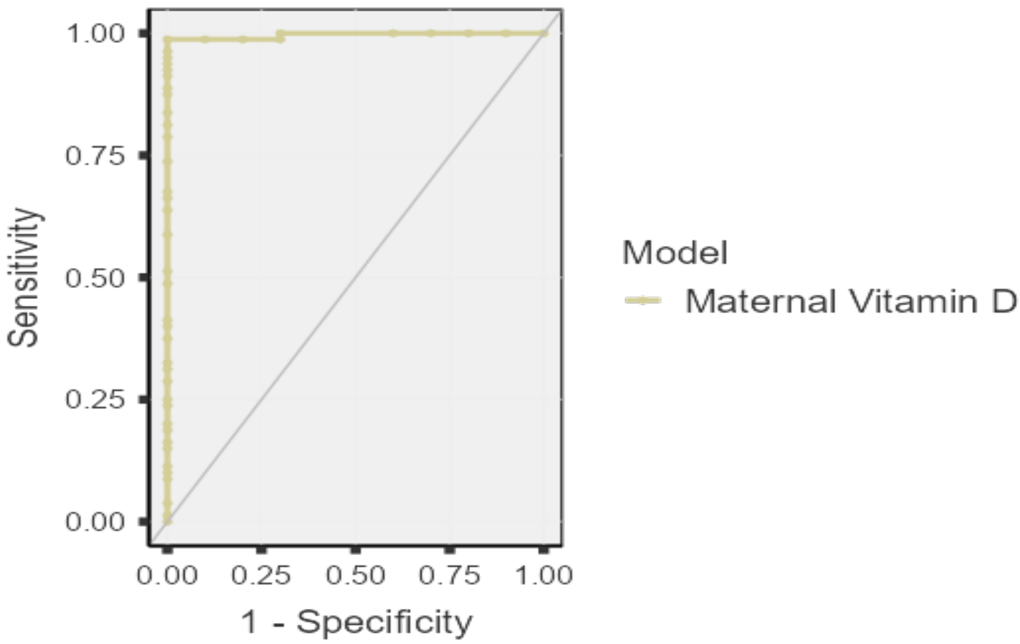

Figure 2: Receiver operating characteristic (ROC) curve of maternal vitamin D level for predicting normal neonatal vitamin D status (N=90).

Using an optimal maternal vitamin D cut-off value of 17.5 ng/mL, the sensitivity for identifying newborns with normal vitamin D levels was 98.75% (95% CI: 93.23–99.97%) and the specificity was 100.00% (95% CI: 69.15–100.00%). The positive predictive value was 100.00%, and the negative predictive value was 90.91%. The overall diagnostic accuracy was 98.89%.

A maternal vitamin D level of 17.5 ng/mL showed excellent ability to identify newborns with normal vitamin D status. [Figure 2]

Table 3: Distribution of NICU admission and neonatal outcome according to maternal vitamin D status (N = 90).

Neonatal outcome		Maternal vitamin D		Total (N = 90) [n (%)]
		Less than normal (n = 23) n (%)	Normal (n = 67) n (%)	
Need for NICU admission	No	17 (28.33)	43 (71.67)	60 (100.00)
	Yes	6 (20.00)	24 (80.00)	30 (100.00)
Outcome	Death	0 (0.00)	2 (100.00)	2 (100.00)
	Discharged	23 (26.14)	65 (73.86)	88 (100.00)

Among the 60 newborns who did not require NICU admission, 17 (28.33%) were born to mothers with low vitamin D levels and 43 (71.67%) to mothers with normal vitamin D levels. Of the 30 newborns requiring NICU admission, 6 (20.00%) were born to mothers having low vitamin D and 24 (80.00%) to the normal vitamin D. 88 newborns were discharged and two neonatal deaths occurred among mothers with normal vitamin D levels. [Table 3]

Discussion

During the study period, a total of 90 mothers admitted in obstetric wards & their neonates were assessed and mong the total admission, 30 neonates required admission to NICU, which were also included in the study.

In the present study, mean maternal age was 24.06 ± 6.24 years. Similar findings were reported by Nupur Rajoria et al.[20], mean maternal age of 24 years among pregnant women. Mean maternal vitamin D in this study was 36.60 ± 13.96 ng/mL and mean neonatal vitamin D level was 39.07 ± 13.70 ng/mL. These values are higher than most Indian study findings. Sachan et al.[21] Reported mean maternal and cord blood vitamin D levels of 14.0 ± 9.3 ng/mL and 8.4 ± 5.7 ng/mL, respectively. Arora et al.[22] In their study also observed mean maternal and cord blood vitamin D levels of 12.5 ± 6.8 ng/mL and 12.3 ± 7.1 ng/mL, respectively, which was much lower than present study. In this study, 74.44% of mothers had sufficient vitamin D, only 15.56% were deficient and 10.00% had insufficient vitamin D. Arora et al.[22] Found vitamin D deficiency in 86% and insufficiency in 9.5% of pregnant women. Sachan et al.[21] found maternal hypovitaminosis D in 84% of pregnant women, and Jani et al.[23] Reported serum 25(OH)D levels below 20 ng/mL in 91% of pregnant women.

In the current study, majority of the mothers were aged ≤30 years (80.00%), residing in rural area (83.33%), belonged to Lower-middle & Lower Socio-economic class ((62.22%), vegetarian (54.44%) and underweight (71.11%). Less than normal neonatal vitamin D was found in mothers aged >30 years, having parity >2, residing in urban area, belonging to Lower-middle & Lower Socio-economic class, consuming vegetarian diet and of normal or obese BMI category. Although none of the background maternal characteristics were found to be statistically significant with neonatal vitamin D level. A similar study conducted by Arif sabta aji et al,[24] in Indonesia also concluded that there is no association between maternal age and neonatal vitamin D status (P>0.05). Similar to this results, Regina wierzejska et al,[25] found that primiparous and multiparous were 42.5% and 57.5% of the total study population, and no association between birth order of the child with their vitamin d status. Pramila kalra et al,[26] concluded similar results that there is no correlation between the maternal dietary preference and neonatal vitamin D status. Similar findings were reported by Sachan et al.[21] and Amulya V et al.[27] from India, Bowyer L et al.[28] from Australia and Javaid MK et al.[29], that maternal background, dietary or demographic characteristics had no association with neonatal vitamin D level. In contrast, Jaiswal S et al.[30] Reported that maternal nutritional factors and socioeconomic status had association with neonatal vitamin D deficiency.

In this study, Preterm birth was noted in 4 (36.36%) mothers with low serum vitamin D as compared to 7 (63.64%) mothers with normal vitamin D levels, term/post-term delivery was reported in 19 (24.05%) and 60 (75.95%) mothers, respectively, but this difference was not statistically significant ($\chi^2 = 0.77$, df = 1; p = 0.38). 9 (29.03%) of the SGA (Small for Gestational Age) babies were born to mothers having less than normal vitamin D levels and 22 (70.97%) in mothers with normal vitamin D levels ($\chi^2 = 0.30$, df = 1; p = 0.58). Mothers of 9 (28.13%) low birth weight newborns had low vitamin D levels and 23

(71.88%) had with normal vitamin D levels, with no significant association ($\chi^2 = 0.17$, df = 1; p = 0.68). Arij Sabta Aji et al.[24] Indicated that there is no association of maternal vitamin d status with the neonatal anthropometric outcome. Nasrin Khalessi et al.[31] in their study in Iran suggested that low maternal vitamin D status is associated with LBW newborns. The mean neonatal length was 48.13 ± 4.71 cm in mothers having low vitamin D and 47.76 ± 4.21 cm in normal group (p = 0.73). The corresponding mean head circumference was 33.09 ± 1.81 cm and 33.24 ± 1.76 cm, respectively (p = 0.72). Nasrin Khalessi et al.[31] concluded that mothers with vit D deficiency gave birth to neonates with head circumference (HC) <33cm. Hashemipour et al.[32] Also showed an independent correlation between neonatal head circumference with maternal vitamin D level. Maternal vitamin D status was not significantly associated with gestational age, birth weight, weight for gestational age, neonatal length, or head circumference. A similarly, Alam MS et al.,[33] found no significant association between maternal vitamin D status and low birth weight (p = 0.628), birth length (p = 0.817), head circumference (p = 0.834), or large-for-gestational-age babies (p = 0.120). In this study, there was a very strong positive correlation between maternal and neonatal vitamin D levels (Pearson's r = 0.95, p < 0.001). Similar strong positive correlation was found by Sachan et al.[21], Arora et al.[22], and Bowyer et al.[28] From India and Australia. In the present study, maternal vitamin D cut-off value of 17.5 ng/mL showed excellent ability to identify newborns with normal vitamin D status, with sensitivity of 98.75%, specificity of 100.00%, and an overall diagnostic accuracy of 98.89%. In the present study, among the 60 newborns who did not require NICU admission, 17 (28.33%) were born to mothers with low vitamin D levels and 43 (71.67%) to mothers with normal vitamin D levels. Of the 30 newborns requiring NICU admission, 6 (20.00%) were born to mothers having low vitamin D and 24 (80.00%) to the normal vitamin D. Similarly, Alam MS et al.[33], and Amulya V et al.[27] Found no significant association between maternal vitamin D deficiency and adverse neonatal outcomes, including NICU admission. In contrast, Arora S et al.[22] Observed that maternal vitamin D deficiency was associated with a higher frequency of adverse neonatal outcomes, including low birth weight and NICU admission.

Conclusion

Maternal vitamin D level had a strong positive correlation with neonatal vitamin D level and can predict neonatal vitamin D status. There was no statistical significant relation between maternal vitamin D status with maternal background characteristics, gestational age, birth weight, neonatal anthropometry, NICU admission, or neonatal outcome. Assessment of maternal vitamin D during pregnancy may help to identify newborns at risk of vitamin D deficiency.

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