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Original Article

Obstetrics and Gynaecology Section

Effects of Maternal Vitamin D Deficiency on the Newborn: A Cohort Study

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ABSTRACT

Introduction: Vitamin D deficiency occurs during pregnancy all over the world because of lifestyle modifications, less sun exposure, sunscreen application and reduced amount of vitamin D in diet. At present, there are very limited number of studies regarding the prevalence of vitamin D deficiency during pregnancy and its effects in the newborns in the North Eastern region of India.

Aim: To estimate serum vitamin D levels in pregnant women and newborn and correlation between maternal and newborn hypovitaminosis D. The study also assess the association of maternal vitamin D levels with the outcomes of early neonatal period.

Materials and Methods: A prospective cohort study was conducted in Assam Medical College and Hospital (AMCH), Dibrugarh, Assam, India, in the Obstetrics and Gynaecology Department over a period of one year (1st July 2019 to 30th June 2020). Seventy pregnant women attending antenatal ward, labour room of AMCH were recruited. The 25-hydroxy vitamin D level was assessed in the maternal and cord blood. Delivery details and foetal parameters were recorded. Primary outcome was to assess the correlation between maternal and cord blood vitamin D level and secondary outcome was to assess the association of maternal vitamin D level with demographic characteristics, economic and obstetrical characteristics, newborn characteristics and perinatal outcome. Analysis of Variance (ANOVA), chi-square test and Fischer's-exact test, Pearson's correlation coefficient (r) were used for statistical analysis.

Results: Out of 70 participants, majority of the pregnant women (61.43%) were vitamin D deficient, 22.86% were vitamin D insufficient and 15.71% had sufficient vitamin D levels. Maternal vitamin D level was significantly associated with maternal age ($p=0.0043$) and socio-economic status ($p=0.0417$). There was a significant positive association between the vitamin D level of the mother with the birth weight ($p<0.001$), birth length ($p=0.0001$), head circumference ($p=0.0003$) and chest circumference ($p=0.001$) of the newborns and also a positive correlation was found between the vitamin D level of the mother and the cord blood ($r=0.984$). There was a significant association between the vitamin D level of the mother and perinatal outcomes such as Neonatal Intensive Care Unit (NICU) admission ($p=0.0169$) and Respiratory Distress Syndrome (RDS) ($p=0.0029$).

Conclusion: The study shows that there is a high prevalence of vitamin D deficiency during pregnancy in North Eastern part of India and it has a significant role in foetal development and perinatal outcomes.

Keywords: Hypovitaminosis D, Infantile rickets, Newborn outcomes, Nutritional deficiency, Pregnant women

INTRODUCTION

Vitamin D is a fat-soluble vitamin and can also act as a steroid hormone. It is synthesised in the skin with the help of Ultraviolet (UV) rays or can be ingested through food. Vitamin D helps in calcium absorption in the intestine and thus it is helpful for bone and other body tissues. Pregnant women should get adequate vitamin D and calcium. Vitamin D deficiency in the pregnant women increases the risk for infantile rickets, neonatal low birth weight, neonatal hypocalcaemia, neonatal seizure, and impaired lung and immune development. Vitamin D deficiency during pregnancy is also a risk factor for diabetes mellitus, pre-eclampsia and increase caesarean section rate [1].

Cholecalciferol or vitamin D3 is the natural form of vitamin D which is derived from diet (animal products). It can also be synthesized in the skin via the interaction of UV light with cholesterol. Plant sterols are a good source of vitamin D2 or ergocalciferol. Both vitamin D2 and D3 binds to vitamin D binding protein and are activated by hydroxylation. Vitamin D is first hydroxylated in the liver. The second hydroxylation occurs mostly in the kidneys and other body tissues regulated by calcium, phosphorus and parathyroid hormone and is activated to1,25-Dihydroxyvitamin D {1,25 $(OH)_2$ D}. Then 1,25 $(OH)_2$D interacts with the Vitamin D Receptor (VDR). VDR is a transcription factor which is involved in bone metabolism and cellular growth. The 25-hydroxy vitamin D can cross the placenta and thus, the foetus depends mainly on the maternal vitamin D level [2].

During pregnancy, maternal physiology changes to maintain all the needs for the growth of the foetus. These changes affects the vitamin D homeostasis. As the fetal vitamin D level is dependent on the maternal vitamin D level, its deficiency during pregnancy causes deficient level in the foetus. Vitamin D deficiency is present all over the globe during pregnancy because of lifestyle modifications, reduced sun exposure, sunscreen use and less intake of vitamin D in diet [3].

One billion people worldwide have vitamin D deficiency or insufficiency. In India, data are very limited regarding the vitamin D deficiency prevalance during pregnancy. At the time of the study, there were limited studies which showed the effects of vitamin D deficiency on the pregnant woman and the newborn [4]. Nalini S et al., conducted a study regarding vitamin D status in pregnant women visiting a tertiary care centre of North Eastern India [5]. Another study was conducted by Talukdar RK et al., regarding vitamin D Status in pregnant women of North East India and impact of vitamin D deficiency in pregnancy on foeto-maternal outcome [6]. Arora S et al., also conducted a study regarding the same [7].

In the lights of above facts, we proposed to conduct the study to estimate serum vitamin D levels in pregnant women and newborns and to find out the correlation between maternal and newborn hypovitaminosis D. We also assessed the association of maternal vitamin D levels with the outcomes in early neonatal period.

MATERIALS AND METHODS

A prospective cohort study was conducted in the Obstetrics and Gynaecology Department, Assam Medical College and Hospital, Dibrugarh, Assam, India, over a period of one year (1st July 2019 to 30th June 2020). A total of 70 pregnant women attending antenatal ward, Labour room were recruited for the study. Approval from the Institutional Ethics Committee (H) was taken prior to the commencement of the study (AMC/EC/PG/1991) dated 22/05/2019. Informed consent was taken from each study subject after explaining the nature of the study in their own understandable language.

Inclusion and Exclusion criteria: Inclusion criteria were women aged between 18 to 45 years with singleton pregnancy at 37 to 40 completed weeks. Exclusion criteria included unwilling patients and patients with thyroid disorder, parathyroid disorder, adrenal diseases, hepatic diseases, renal disorders, metabolic bone disease, diabetes mellitus, malabsorption, prolong draining per vagina, gross oligohydramnios, proved sexually transmitted infections, overt infection and neonates with cardiac disease, lung disease, congenital anomalies like oesophageal atresia, renal agenesis, preterm babies and instrumental delivery.

Sample size calculation: Considering the prevalence of maternal hypovitaminosis D to be 86% [7] and taking 95% confidence interval with relative recession of 10%, sample size for the present study was calculated and rounded off to be 70.

Study Procedure

Detailed history was taken from the pregnant women including demographic details, dietary history (vegetarian or non vegetarian), past medical history (no significant illness was found in any of the pregnant women), previous obstetric history (parity). Dose and duration of vitamin D tablets taken during pregnancy were also recorded. Socio-economic status was classified according to modified Kuppuswamy socio-economic scale updated for the year 2019 [8].

Samples (5 mL each) were taken from maternal blood and cord blood during delivery. Labour and delivery details were also recorded. Newborn anthropometric measurements like birth weight, length, head and chest circumference were measured at birth. The APGAR (Appearance, Pulse, Grimace, Activity, and Respiration) score at birth, any neonatal complications such as RDS, Intraventricular Haemorrhage (IVH), Necrotising Enterocolitis, admission to NICU, phototherapy were also recorded.

Vitamin D was analysed by the DIAMETRA 25-OH Vitamin D quantitative solid phase Enzyme Linked Immunosorbent Assay (ELISA) technique. Thereafter, the mother and their newborns were classified into four groups according to the United States (US) Endocrine Society Classification depending on the vitamin D levels [9].

Group 1: Vitamin D Deficient group (≤20 ng/mL)

Group 2: Vitamin D Insufficient group (21-29 ng/mL)

Group 3: Vitamin D Sufficient group (≥30-150 ng/mL)

Group 4: Vitamin D intoxication (>150 ng/mL)

These groups were followed-up and studied for the outcomes of pregnancy such as mode of delivery, newborn anthropometric measurements, APGAR score, NICU admission, RDS and IVH.

STATISTICAL ANALYSIS

The data analysis was done using the program, Statistical Package for Social Sciences (SPSS for Windows, version 21.0, Chicago, SPSS Inc.) and Microsoft Excel 2010. Mean±standard deviation was used to present results on continuous measurements. Results were compared using Analysis of Variance (ANOVA). Among the three groups, significance between two individual groups was found out by post hoc analysis. Discrete data were expressed as number (%). Chi square test and Fischer's exact test (where the cell counts were <5 or 0) were used to analyse discrete data. The associations among the continuous variables was assessed by Pearson's correlation coefficient (r). The p-value <0.05 was considered statistically significant.

RESULTS

The mean age of the study population was 31.03±8.92 years. Majority 24 (34.29%) of the participants were ≥40 years. Majority of the pregnant women 23 (32.86%) were obese (Body Mass Index (BMI) ≥30). Mean BMI in the study population was 24.43±5 [Table/Fig-1].

Characteristics		Maternal vitamin D level			p-value**
		Deficiency n=43	Insufficiency n=16	Sufficiency n=11	
Maternal age (years)	<40	23 (53.49%)	13 (81.25%)	10 (90.91%)	0.0043*
	≥40	20 (46.51%)	3 (18.75%)	1 (9.09%)	
Mean age (in years)		33.47±8.96	29.19±8.09	24.18±5.62	
Mean body mass index (Kg/m^2)		25.05±5.32	24.06±4.85	22.54±3.59	0.3186

[Table/Fig-1]: Maternal vitamin D level association with demographic characteristics.
*Statistically significant at p<0.05 level. **Among three groups, significance between two individual groups was found out by post hoc analysis

Majority of the pregnant women {n=43, (61.43%)} were vitamin D deficient (vitamin D level ≤20 ng/mL) and 16 (22.86%) were vitamin D insufficient (vitamin D level 21-29 ng/mL). Only 11 (15.71%) had sufficient vitamin D level (vitamin D level ≥30-150 ng/mL). Among the vitamin D deficient mothers, majority {n=18, (41.86%)} were obese, 13 (30.23%) were underweight, 7 (16.28%) had a normal BMI and 5 (11.63%) were overweight. Among the vitamin D insufficient mothers, majority {n=7, (43.75%)} had a normal BMI, 4 (25%) were underweight, 4 (25%) were obese and 1 (6.25%) were overweight. Among the vitamin D sufficient mothers, majority 7 (63.64%) were normal BMI, 3 (27.27%) were underweight and 1 (9.09%) were obese.

In cord blood, vitamin D deficiency was seen in 44 (62.86%), insufficiency was seen in 13 (18.57%), and sufficiency was seen in 13 (18.57%). In our study, maternal vitamin D level was significantly associated with maternal age (p=0.0043) [Table/Fig-1]. In our study, maternal vitamin D level was not significantly associated with maternal diet (p=0.349) [Table/Fig-2]. In our study, maternal vitamin D level was significantly associated with maternal socio-economic status (p=0.0417) [Table/Fig-3]. In our study, maternal vitamin D level was significantly associated with maternal vitamin D supplementation (p=0.001) [Table/Fig-4].

Dietary history	Maternal vitamin D level			p-value
	Deficiency n=43 n (%)	Insufficiency n=16 n (%)	Sufficiency n=11 n (%)	
Vegetarian	19 (44.19)	8 (50)	6 (54.55)	0.349
Non-vegetarian	24 (55.81)	8 (50)	5 (45.45)	

[Table/Fig-2]: Maternal vitamin D level association with dietary history.
p<0.05 level- statistically significant; Chi-square test was used

Characteristics	Maternal vitamin D level			p-value
	Deficiency (n=43) n (%)	Insufficiency (n=16) n (%)	Sufficiency (n=11) n (%)	
Socio-economic class				
Upper	1 (2.33)	2 (12.50)	0	0.0417*
Upper middle	8 (18.60)	4 (25)	3 (27.27)	
Lowe middle	8 (18.60)	5 (31.25)	5 (45.45)	
Upper lower	4 (9.30)	0	0	
Lower	22 (51.16)	5 (31.25)	3 (27.27)	
Gravida				
Primi	25 (58.14)	10 (62.50)	4 (36.36)	0.3551
Multi	18 (41.86)	6 (37.50)	7 (63.64)	
Mode of delivery				
Spontaneous vaginal delivery	18 (41.86)	11 (68.75)	3 (27.27)	0.0747
Caesarean	25 (58.14)	5 (31.25)	8 (72.73)	

[Table/Fig-3]: Maternal vitamin D level association with economic and obstetrical characteristics.
*Statistically significant at p<0.05 level; Data presented as number (percentage); Chi-square test was used

Vitamin D supplement (1000 mg tablet)	Maternal vitamin D level			p-value
	Deficiency n=43 n (%)	Insufficiency n=16 n (%)	Sufficiency n=11 n (%)	
No	35 (81.39)	11 (68.75)	1 (9.1)	0.001*
Yes	8 (18.60)	5 (31.25)	10 (90.90)	

[Table/Fig-4]: Maternal vitamin D level association with vitamin D supplementation.
*p<0.05 level statistically significant; Chi-square test was used

There was a significant positive association between the vitamin D level of the mother with the birth weight, birth length, head circumference and chest circumference of the newborns. No significant association was found between the vitamin D level of the mother and the APGAR score of the newborn at 1 and 5 minute [Table/Fig-5].

In our study, there was a significant association between the maternal vitamin D level and perinatal outcomes such as baby admission to NICU and RDS [Table/Fig-6].

Characteristics	Maternal vitamin D level			p-value
	Deficiency	Insufficiency	Sufficiency	
Mean birth weight (kg)	2.42±0.14	2.61±0.07	2.94±0.27	<0.001*
Mean birth length (cm)	44.76±5.44	47.12±4.84	53.18±5.04	0.0001*
Mean head circumference (cm)	31.44±1.90	31.99±2.12	35.49±2.26	0.0003*
Mean chest circumference (cm)	28.82±1.53	29.34±2.82	33.48±2.43	0.001*
Mean APGAR score at 1 minute	8.56±0.98	7.81±2.26	8.36±1.29	0.2007
Mean APGAR score at 5 minute	9.63±0.62	8.94±2.43	9.55±0.52	0.1791

[Table/Fig-5]: Maternal vitamin D level association with newborn characteristics.
*Statistically significant at p<0.05 level; significance between 2 individual groups was find out by Post hoc analysis

Perinatal outcome	Total		Deficiency		Insufficiency		p-value
	n	%	n	%	n	%	
Admission to NICU	26	37.14	23	53.49	3	18.75	0.0169*
Neonatal jaundice	26	37.14	21	48.84	5	31.25	0.2264
Respiratory distress syndrome	17	24.29	17	39.53	0	0.00	0.0029*
Sepsis	12	17.14	9	20.93	3	18.75	0.8533
Necrotising enterocolitis	6	8.57	6	13.95	0	0	0.1149
Intraventricular haemorrhage	1	1.43	1	2.33	0	0	0.5384
Stillbirth/IUFD	1	1.43	0	0	1	6.25	0.2712

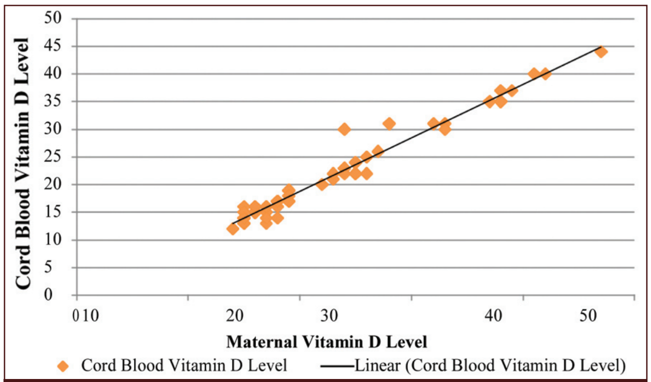
[Table/Fig-6]: Association between maternal vitamin D level with perinatal outcomes.
*p<0.05 level- Statistically significant; Chi-square test was used
IUFD: Intrauterine foetal death

In this study, cord blood vitamin D deficiency was seen in all the newborns of the vitamin D deficient mothers. We found a significant positive correlation between the maternal vitamin D level and cord blood vitamin D level [Table/Fig-7].

Scatter plot showed positive correlation between the maternal vitamin D level and cord blood vitamin D level (r=0.984) [Table/Fig-8].

Maternal vitamin D level	Mean cord blood vitamin D level (ng/mL)	p-value***	Correlation coefficient (r)
Deficiency	15.51±1.74	<0.001*	0.984**
Insufficiency	23.75±3.11		
Sufficiency	35.91±4.32		

[Table/Fig-7]: Correlation between maternal and cord blood vitamin D level.
*Statistically significant at p<0.05 level; **Positive correlation when correlation coefficient greater than zero; ***ANOVA followed by post hoc analysis was done to find out the significance between two individual groups



[Table/Fig-8]: Scatter plot depicting correlation between maternal and cord blood vitamin D level.

DISCUSSION

In the present study, majority of the pregnant women (61.43%) were vitamin D deficient, 22.86% were vitamin D insufficient and 15.71% had sufficient vitamin D level. Similar findings were seen in a study by Arora S et al., where majority (86%) of the subjects were vitamin D deficient, 9.5% were vitamin D insufficient and 4.5% were vitamin D sufficient [7]. A significant association was found between the maternal vitamin D levels and maternal age (p=0.0043) in our study. Kim I et al., had similar findings in their study done in Korea where they found that mean age was more in vitamin D deficient mothers and this difference was statistically significant (p=0.031) [10].

In our study, among the vitamin D deficient mothers, an association was observed between the maternal vitamin D and socio-economic status. Similarly, Sharma S et al., reported that among the vitamin D deficient, majority (52.7%) were from lower socio-economic class and maternal vitamin D level was associated with socio-economic status (p=0.0001) [11]. In this study, among the vitamin D deficient mothers, there was no association between the maternal vitamin D level and BMI. Sathish P et al., found similar results where among the vitamin D deficient mothers, majority {13 (41.94%)} were obese, 10 (32.26%) were underweight, 5 (16.13%) were normal BMI, 3 (9.67%) were overweight and p-value was 0.085, which suggested no association between the maternal vitamin D level and BMI [12].

No significant association was found between the maternal vitamin D level and gravida of the mothers (p-value 0.3551) in our study. Ciryam SS et al., in their study also found no statistical significant association between gravida of the mothers and vitamin D level (p-value 0.982) [13]. In the present study, there was no association between the maternal vitamin D level and mode of delivery (p=0.074792). In a study by Zamal A et al., no significant association was observed between maternal vitamin D levels with mode of delivery (p=0.282) [14]. In the present study, there was a significant association between the maternal vitamin D level and vitamin D supplementation (p=0.001). A similar study by Prasad D et al., found significant association between maternal vitamin D levels with vitamin D supplementation (p=0.000) [15].

In the present study, a significant positive correlation was found between the maternal and cord blood vitamin D level (p<0.001, r=0.984). Similarly Arora S et al., also found a significant positive correlation between the maternal and cord blood vitamin D level (p<0.001, r=0.84) [7].

In this study, a significant association was found between the maternal vitamin D level and the birth weight, birth length, head circumference and chest circumference of the newborns. Sathish P et al., also reported significant association between the maternal vitamin D level and the birth weight (p=0.001), birth length (p=0.016), head circumference (p=0.001) and chest circumference (p=0.001) of the newborns [12].

In our study, no correlation was found between the maternal vitamin D level and APGAR score of the newborns at 1 minute and 5 minutes. Similarly, Prasad D et al., found no correlation between the maternal vitamin D level and APGAR score of the newborns (p=0.683) [15].

A significant association was found between the maternal vitamin D level and baby admission to NICU in our study. A study by Gupta S et al., also found that neonatal ICU hospitalisation was more frequent in the vitamin deficiency group (p-value was 0.003) [16].

In the present study, we found no association between the maternal vitamin D levels and neonatal jaundice (p=0.2264). Similarly, Talukdar RK and Joshi SM, found no association between the maternal vitamin D levels and neonatal jaundice (p=0.854) [6].

We found significant association between the maternal vitamin D level and RDS of the babies. Similarly, in a study by Ardastani AG et al., found vitamin D correlated with RDS (p=0.004) [17].

In this study, there was no association between the maternal vitamin D level and neonatal sepsis. Similarly in a study by Thiele DK et al., found no correlation between maternal vitamin D status with neonatal sepsis (p=0.58) [18].

In our study, no association was found between necrotising enterocolitis, IVH of the babies and the maternal vitamin D level. Similarly Kim I et al., found no association between necrotising enterocolitis (p=0.1149), IVH (p=0.5384) of the babies and the maternal vitamin D level [10].

In the present study, there was no association between the maternal vitamin D level and stillbirth or intrauterine death. Similarly, Prasad D et al., found that low maternal vitamin D levels were not associated with stillbirth or intrauterine death (p=0.529) [15].

Limitation(s)

The study was limited by small sample size due to high cost of the test, conducted in a single center within a short span of time. There are several confounding factors in the study like ethnicity, occupation, factors related to sun exposure, environment, skin color and genetic background.

CONCLUSION(S)

It can be concluded that there is a high prevalence of vitamin D deficiency during pregnancy in this North Eastern part of India and it has significant role in foetal development and perinatal outcomes. However, this observation needs to be validated in more extensive, multi-centric and comprehensive studies.

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