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Vitamin D Status and Obstetric Outcome

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Abstract

Background: Vitamin D deficiency in pregnancy is associated with wide ranging clinical outcomes, including obstetric complications, preterm birth and adverse offspring outcomes affecting the skeletal, immune and respiratory systems.Vitamin D may have a role in immune tolerance in a pregnant woman, to prevent fetal rejection. Vitamin D travels to the fetus by passive transfer and the fetus is entirely dependent on maternal stores. Therefore, it is essential to know vitamin D level during antenatal period to prevent adverse outcomes. The association of maternal vitamin D deficiency with asthma and impaired lung function in offspring were described by some authors.

Aim of Study: The aim of this work is to assess maternal vitamin D status and its association with pregnancy outcomes.

Patients and Methods: Prospective observational cohort study was designed on 80 pregnant women 18-38 years with singleton pregnancy attended Obstetrics and Gynecology Department at Mansoura University Hospital from May 2022 to May 2023, with no medical disorders or history of vitamin D intake during pregnancy. Chronic diseases as liver diseases and severe infections or take corticosteroids were excluded.

Results: There was statistically significant association between maternal vitamin D level and gestational DM (p= 0.023), while, no statistically significant association between maternal vitamin D and neonatal outcome was found. The majority of the pregnant women, 73.8% were vitamin D insufficient, 11.2% were vitamin D deficient and 15% had sufficient vitamin D level. The mean value of the maternal vitamin D level was 19.17±8.78ng/ml and the mean value of fetal vitamin D level was 15.51±7.80ng/ml. Infants were divided into: insufficient group 58 (72.5%), deficient 16 (20%) and sufficient groups 6 (7.5%). There was highly statistically significant correlation between maternal and fetal vitamin D level.

Conclusion: Our study suggests that vitamin D deficiency in pregnant women and their infants is still a serious health problem. However, there were no statistically significant associations between maternal vitamin D and both maternal & neonatal outcome except maternal gestational diabetes mellitus.

Recommendations: Routine measurement of maternal serum vitamin D can be advised to become an integral part of the antenatal care protocol. VD supplementation can be subsequently added to carefully selected groups to decrease the risk of multiple adverse maternal and neonatal health outcomes including gestational DM, preeclampsia, primary cesarean section, abortion, fetal intrauterine growth restriction, risk of preterm birth, low birth weight, and neonatal hypocalcemia.

Key Words: *Vitamin D – Maternal outcome – Neonatal outcome – Pregnancy.*

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Introduction

VITAMIN D plays a major role in human reproduction. The effect of vitamin D on pregnancy is a subject of major interest in current medicine. Vitamin D influences folliculogenesis, modulates endometrial receptivity, and regulates embryogenesis **[1]**. It plays an important role in trophoblast invasion **[2]**. There are plenty of studies that prove its effects on placental implantation, angiogenesis and endothelial function, immune function, inflammatory response during pregnancy, oxidative stress, and glucose homeostasis **[3]**.

It is hypothesized that during the first half of pregnancy vitamin D may contribute to fetal growth and to the 'programming' of some fetal functions and organs, including among others, the central nervous system, bone and dental enamel. Therefore, it seems reasonable to maintain sufficient maternal vitamin D and calcium levels that will cross the placenta to the fetus **[4]**.

A balanced, nutritious diet is an important aspect of a healthy pregnancy and its outcome. Vitamin D plays an important role in regular bone growth and adequate function of innate immune system, including barrier function of mucous membrane **[5]**.

Low maternal vitamin D levels during pregnancy are associated with various adverse obstetric outcomes, such as gestational diabetes mellitus (GDM), preeclampsia, and primary cesarean sec-

tion. Additionally, gestational vitamin D deficiency has been linked to multiple adverse fetal and neonatal health outcomes including fetal intrauterine growth restriction, a higher risk of preterm birth, abortion, low birth weight and neonatal hypocalcemia **[6]**.

Furthermore, due to the decisive effects of vitamin D deficiency on pregnancy outcome and maternal and neonatal health, coherent planning for supplementing mothers with vitamin D deficiency and a suitable follow-up during pregnancy should be highlighted more, for instance, by introducing and strictly implementing comprehensive policies and measures **[7]**.

Patients and Methods

We conducted a prospective observational cohort study on 80 pregnant women 18-38 years with singleton pregnancy attended Obstetrics and Gynecology Dept., at Mansoura University Hospital.

Ethical considerations:

All procedures were carried out in accordance with the ethical standards of the institutional and/or national research committee. The study received the approval by Institutional Review Board (IRB), Faculty of Medicine, Mansoura University(Code No. MS.20.03.1231). nformed written consent was obtained from the participants before inclusion in the study.

Inclusion criteria:

Include: 1- Singleton pregnancy, 2- Age: 18-38 years, 3- No known medical disorders (hypertension, diabetes, kidney, etc.), 4- No history of vitamin D intake during pregnancy.

Exclusion criteria:

Include: Chronic diseases as liver diseases and Severe infections or take corticosteroids.

We classified participants into 3 groups; 1) Deficient if the vitamin D level is <10ng/ml, 2) insufficient between 10 and 30ng/ml, and if it is >30ng/ml, it was considered within the normal range (sufficient). All patients participated in the study were subjected to the following: 1) Full history taking, 2) Complete general and obstetric examination, 3) Abdominal ultrasonography examination, 4) obstetric ultrasound and 5) Basic investigations as blood pressure and random blood glucose, CBC and urine analysis. In addition, estimation of serum 25-OH-D in both maternal blood and fetal blood from umbilical cord at delivery as following:

- 5ml of maternal blood was collected in the non-fasting condition. Serum was stored at −20°C until they analyzed for 25(OH) D.
- 5ml of umbilical cord blood was collected at delivery and stored at −20°C until they analyzed for 25(OH) D.
- Maternal non-fasting blood samples taken at first and last month of gestation and were collected and stored initially at −20°C, and then at −80°C, with no further freeze-thaw cycles until 25(OH) D measurement.
- Serum 25(OH)D was measured by RIA using a kit from Diasorin Inc, following the manufacturer's instructions **[8]**. The serum 25(OH)D level is expressed as nanograms per milliliter. The quality and reproducibility were determined using quality controls provided with the kits. Two controls (low control, 14.1ng/mL; high control, 54.1ng/mL) were provided, and the coefficient of variation for all kits used was 12.54% for the low control and 12.95% for the high control. The controls fell within the acceptable range given by the manufacturer.

Statistical analysis:

Data were analyzed using the Statistical Package of Social Science (SPSS) program for Windows (Standard version 26). The normality of data was first tested with one-sample Kolmogorov-Smirnov test. Qualitative data were described using number and percent. Association between categorical variables was tested using Chi-square test while Monte carlo test were used when expected cell count less than 5. Continuous variables were presented as mean ± SD (standard deviation) for normally distributed data were compared with ANOVA test. The results were considered significant when the $p \leq 0.05$. The smaller the *p*-value obtained, the more significant are the results.

Results

The mean age of the studied cases was 26.51± 4.67 years. gravida 3 (30%) was the most common among them, more than two third cases were para 1 and para 2 (33.8% and 31.2%, respectively) (Table 1). There was no statistically significant association found between maternal vitamin D (deficiency, insufficiency or sufficiency) with age, gravidity or parity among studied cases (Table 3).

The mean value of the maternal vitamin D level was 19.17±8.78ng/ml (Table 2). The majority of the pregnant women (73.8%) were vitamin D insufficient, 11.2% were vitamin D deficient and 15% had sufficient vitamin D level (Fig. 1).

Among the studied group, 6 (7.5%) had gestational DM, while, gestational HTN, PET and primary cesarean section were reported in 4 (5%) of cases for each (Table 4).

Women diagnosed as vitamin D deficiency had a higher incidence rate of gestational DM compared with vitamin D insufficiency or sufficiency (50% vs 33.3% & 16.7). There was statistically significant association between maternal vitamin D level and gestational DM. In addition, women diagnosed as

vitamin D insufficiency had higher incidence rates of gestational HTN (100%), PET (50%), preterm labor (83.3%) and primary CS (75%) (with no statistically significant associations) (Table 5).

Table (1): Demographic data and obstetric history among the studied group.

Demographic data and obstetric history	The study group (n=80)
Age (years):	
Mean ± SD	26.51±4.67
Min-Max	18.00-37.00
Gravidity:	
PG	15 (18.8%)
G2	20 (25.0%)
G3	24 (30.0%)
G4	13 (16.2%)
G5	6 (7.5%)
G6	2 (2.5%)
Parity:	
P0	15 (18.8%)
P1	27 (33.8%)
P2	25 (31.2%)
P3	9 (11.2%)
P4	4 (5.0%)

SD: Standard deviation.

Table (2): Vitamin D level and gestational age.

	Mean ± SD	Min-Max
Maternal vitamin D (ng/ml)	19.17±8.78	1.90-39.89
Fetal vitamin D (ng/ml)	15.51±7.80	1.20-38.33
Gestational age (weeks)	37.80±1.64	30-40

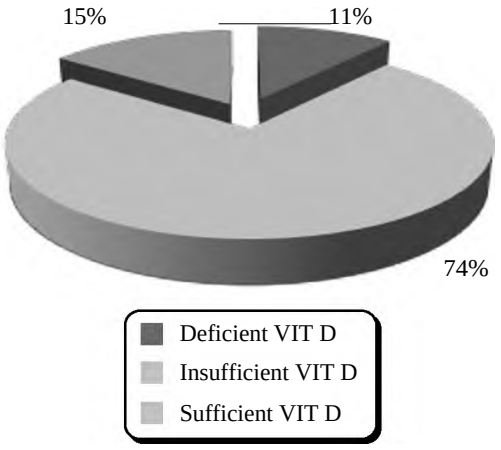


Fig. (1): Maternal Vitamin D level.

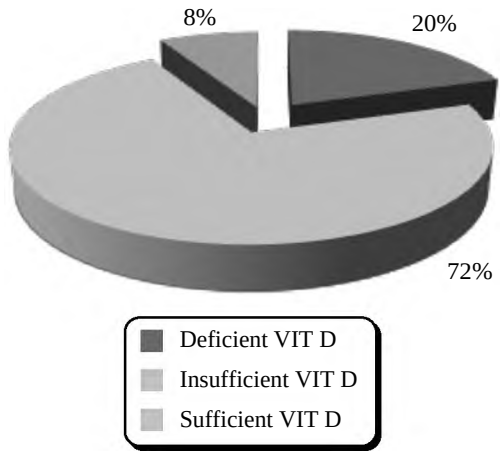


Fig. (2): Fetal Vitamin D level.

Table (3): Association between maternal vitamin D and obstetric history.

Obstetric history	Total	Deficient (n=9)	Insufficient (n=59)	Sufficient (n=12)	*p*-value
Age (years):					
Mean ± SD		28.1±3.9	26.4±4.7	25.5±4.7	0.450
Gravidity:					
PG	15	0 (0.0%)	12 (80.0%)	3 (20.0%)	0.642
G2	20	2 (10.0%)	16 (80.0%)	2 (10.0%)	
G3	24	3 (12.5%)	17 (70.8%)	4 (16.7%)	
G4	13	2 (15.4%)	8 (61.5%)	3 (23.1%)	
G5	6	1 (16.7%)	5 (83.3%)	0 (0%)	
G6	2	1 (50.0%)	1 (50.0%)	0 (0%)	
Parity:					
P0	15	12 (80%)	0 (0%)	3 (20.0%)	0.747
P1	27	3 (11.1%)	21 (77.8%)	3 (11.1%)	
P2	25	3 (12.0%)	17 (68.0%)	5 (20.0%)	
P3	9	2 (22.2%)	6 (66.7%)	1 (11.1%)	
P4	4	1 (25.0%)	3 (75.0%)	0 (0%)	

Table (4): Maternal outcome among the studied group.

Maternal outcome	The study group (n=80) No (%)
Gestational DM	6 (7.5%)
Gestational HTN	4 (5%)
PET	4 (5%)
Primary CS	4 (5%)

CS : Cesarean section. HTN: Hypertension.
DM: Diabetes mellitus. PET : Preeclampsia.

Table (5): Association between maternal vitamin D and maternal outcome.

Maternal outcome	Total	Maternal Vitamin D			*p*-value
		Deficient (n=16)	Insufficient (n=58)	Sufficient (n=6)	
Gestational DM:					
Yes	6	3 (50.0%)	2 (33.3%)	1 (16.7%)	0.023*
No	74	6 (8.1%)	57 (77.0%)	11 (14.9)	
Gestational HTN:					
Yes	4	0 (0%)	4 (100%)	0 (0%)	0.443
No	76	9 (11.8%)	55 (72.4%)	12 (15.8)	
PET:					
Yes	4	1 (25%)	2 (50%)	1 (25%)	0.752
No	76	8 (10.5%)	57 (75.0%)	11 (14.5)	
Preterm labor:					
Yes	6	0 (0%)	5 (83.3%)	1 (16.7%)	0.835
No	74	9 (12.2%)	54 (73.0%)	11 (14.9)	
Primary CS:					
Yes	4	0 (0%)	3 (75%)	1 (25%)	0.685
No	76	9 (11.8%)	56 (73.7%)	11 (14.5)	

CS: Cesarean section, DM: Diabetes mellitus, HTN: Hypertension, PET:Preeclampsia, *: Significant.

Eighteen infants were included, mean fetal vitamin D level was 15.51±7.80ng/ml for the total group of infants. The mean gestational age of infants was 37.80±1.64 weeks (Table 2). Infants were divided into three groups according to their vitamin D levels. The number of infants in the insufficient group 58 (72.5%) was higher than that in deficient 16 (20%) and sufficient groups 6 (7.5%) which was expected due to the majority of mothers (73.8%) had insufficient vitamin D level (Fig. 2). In addition, impaired lung function respiratory distress, low birth weight and preterm labor were 24 (30%), 7 (8.8%) and 6 (7.5%), respectively. With no bone deformity in neonates (Table 6).

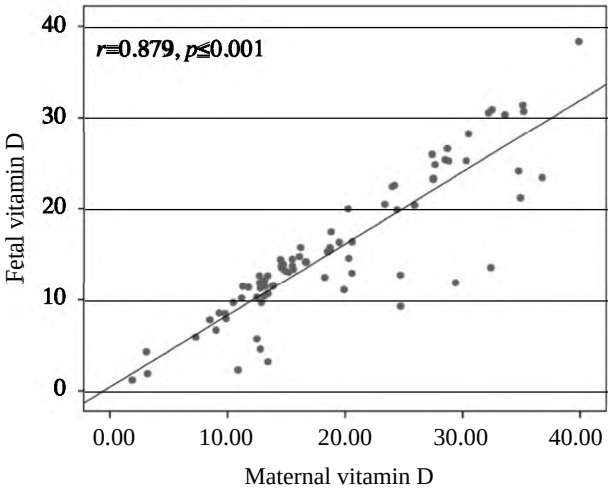
Both low birth weight and respiratory distress had higher incidence rate in vitamin D insufficient group (71.4% and 70.8%, respectively), followed by vitamin D deficient group (14.3% and 25%, respectively). The mean gestational weeks was higher among vitamin D sufficient group (37.9±1.4), while the lowest value was reported in vitamin D deficient group (37±1.3). But no statistically significant association between maternal vitamin D and neonatal outcome was found (Table 7). There was highly statistically significant correlation between maternal and fetal vitamin D level (Fig. 3).

Table (6): Neonatal outcome among the studied group.

Neonatal outcome	The study group (n=80) No (%)	Gestational Age Mean ± SD
Preterm labor:		
Yes	6 (7.5%)	23.17±3.81
No	74 (92.5%)	26.78±4.65
Low birth weight:		
Yes	7 (8.8%)	34.28±2.98
No	73 (91.2%)	38.13±0.96
Impaired lung function respiratory distress:		
Yes	24 (30%)	36.67±2.42
No	56 (70%)	38.28±0.80

Table (7): Association between neonatal vitamin D and neonatal outcome.

Neonatal outcome	Total	Maternal Vitamin D			*p*-value
		Deficient (n=16)	Insufficient (n=58)	Sufficient (n=6)	
Low birth weight:					
Yes	7	1 (14.3%)	5 (71.4%)	1 (14.3%)	0.742
No	73	15 (20.5)	53 (72.6%)	5 (6.8%)	
Respiratory distress:					
Yes	24	6 (25.0%)	17 (70.8%)	1 (4.2%)	0.651
No	56	10 (17.9)	41 (73.2%)	5 (8.9%)	
Gestational weeks		37±1.3	37.6±3.4	37.9±1.4	0.380



r: Pearson's correlation coefficient

Fig. (3): Scatter diagram for positive correlation between maternal and fetal vitamin D level.

Discussion

Vitamin D inadequacy is highly prevalent among the pregnant population worldwide [9]. There is relationship between low vitamin D and adverse maternal outcomes such as recurrent pregnancy loss, primary caesarean section and postpartum depression, pre-eclampsia, eclampsia and gestational diabetes [10,11]. Vitamin D travels to the fetus by passive transfer and the fetus is entirely dependent on maternal stores. Therefore, it is essential to know vitamin D level during antenatal period to prevent adverse outcomes [12].

In the same line with our findings, in Iraq, Ibrahim et al. [13] demonstrated that the mean age of studied pregnant women was 29.13 years, 31.5% of women were primiparous and 68.5% multiparous. In addition, there was no relationship between mother's age, parity or gravidity and maternal vit D deficiency [14,15]. On the other hand, a significant association was found between the maternal vitamin D levels and maternal age [16,17]. This difference between studies may attribute to variation in sample size, time of the study and older ages of the participants.

In addition, our findings go parallel with Pahuja et al. [5] andVatandost et al. [18], where they found that 69 % &68 %of study participants were vitamin D insufficient, respectively (using <20ng/ml cut-off). Such widespread insufficiency of vitamin D could be linked to the high prevalence of some of the health problems, pregnancy-related complications and communicable & non-communicable diseases in South Asian countries [19]. This variance between our findings and other previous results may be attributed to that the sample size was older than ours, variability in the degree of sunlight exposure due to differences in geographical locations and wearing of covered clothes. Throughout the year, people living in tropical areas get more sunlight exposure than those living in subtropical regions.

Our study supports the hypothesis that low vitamin D level can play a role in the development of pre-eclampsia according to Osman et al. [20] in Egypt.

In agreement with our findings, mean vitamin D level was 13.16±7.16 ng/mL in infants. The mean gestational age was 38.45±1.10 weeks. But the number of infants in the deficient group wassignificantly higher than that in insufficient and sufficient groups [21]. Regarding respiratory distress syndrome, several clinical studies have described low vitamin D levels in preterm infants who suffered from respiratory distress syndrome [22,23]. Additionally, it was concluded that vitamin D deficiency or lower levels of vitamin D within 24h of birth were always associated with respiratory distress developmentin a recent meta-analysis [24]. However, no association was found between vitamin D deficiency & respiratory distress syndrome and the umbilical cord vitamin D level does not lead to a higher or lower risk of respiratory distress [25].

In the same line, in a previous meta-analysis that included 16,515 individuals from 20 observa-

tional studies on the correlation between vitamin D status and GDM in a broad range of populations revealed that vitamin D deficiency significantly increased the risk of GDM by 45% **[26]**. In Saudi Arabia, the analysis of the association between vitamin D status and incidence of GDM indicated a 2.87-fold increased risk of development of GDM among vitamin D deficient pregnant women compared to vitamin D non-deficient women and this risk was sustained or substantially increased when the data was analyzed after adjusting for sunlight exposure, physical activity, calcium and vitamin D intake and other factors that are known to influence vitamin D levels **[27].**

Furthermore, in a case-control study, GDM pregnant women (24–28 weeks of gestation) with vitamin D deficiency (<50nmol/L) treated with vitamin D3 1200 IU/d had a significant increase in vitamin D serum levels and a significant decrease in fasting plasma glucose, postprandial 2h plasma glucose, and glycosylated hemoglobin at 36 weeks of gestation, which supports the positive glucose metabolic effects of vitamin D3 supplementation on mothers **[28]**. In contrast, concluded that no statistically significant association was found between vitamin D levels and gestational DM **[29]**.

This difference among studies may be due to presence of obesity prior to and during pregnancy, family history of diabetes and previous history of GDM in studied population which leads to high prevalence of GDM. In addition, lower vitamin D concentrations have been demonstrated to be associated with insulin resistance, maternal glycemia, and high risk of GDM. However, the relationship of vitamin D with risk of GDM has not been well defined **[30].**

Heterogeneity in study findings could be attributable to differences in ethnicity, geographic setting, stage of gestation, endpoints, level and duration of vitamin D supplementation **[31].**

Interestingly, in a large prenatal cohort in America, it was found that higher 25OHD concentrations were associated with higher odds of pregnancy induced hypertension (PIH), as with every 25nmol/L increase in plasma 25OHD concentration, the risk of developing PIH increased 1.32-fold **[32]**. Other studies have concluded that there is no significant correlation between VD status and preeclampsia,pregnancy induced hypertension and cesarean delivery **[33,34]**. However, many observational studies did report that vitamin D levels were associated with adverse maternal, fetal, and neonatal outcomes, including increased risk of developing preeclampsia, preterm labor, gestational diabetes, being small for gestational age, low birth weight, an increased rate of cesarean delivery and infertility **[35]**. Inconsistent results regarding the association between maternal VD status during pregnancy and PIH may be related to multiple confounding factors, such as race, season, diet (including VD intake and patterns), and the method of measuring 25OHD levels **[6].**

In accordance regarding association between maternal vitamin D and neonatal outcome found that vitamin D was found to be correlated with respiratory distress syndrome (p=0.004) **[17,36]**, However, Rabbani et al. **[15]** did not reveal a significant association between maternal vitamin D levels and any of newborn anthropometric measurements (birth weight, length and head circumference). On the other hand, they found that there was a strong positive association between maternal and newborn vitamin D levels which agree with our results.

The discrepancies observed in the literatures could be in part explained by inconsistencies across published studies in definition of cut-offs for 25(OH)D deficiency, time of sampling (early or late pregnancy), specimens used to assess 25(OH) D concentrations (most of them in maternal blood and some of them in cord blood), heterogeneous populations involved (i.e. African, Caucasian, Indian or Vietnamese women), seasonal differences (summer vs winter), the number of subjects and significant heterogeneity found in previous meta-analysis **[37].**

Conclusion: Our study suggests that vitamin D deficiency in pregnant women and their infants is still a serious health problem. However, our results concluded that there were no statistically significant associations between maternal vitamin D and both maternal & neonatal outcome except maternal gestational DM.

Recommendations: 1- Routine measurement of maternal serum vitamin D can be advised to become an integral part of the antenatal care protocol. 2-Vitamin D supplementation should be considered for vitamin D deficient pregnant mothers and their newborns and the antenatal visits should include education on the safety and importance of sufficient sunlight exposure during pregnancy.

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علاقة مستوى فيتامين (د) فى دم الأم والمولود بنتائج الحمل والولادة

يؤدى نقص فيتامين (د) فى النساء الحوامل إلى مشاكل صحية مختلفة لدى الأمهات وأطفالهن بما في ذلك داء سكرى الحمل، تسمم الحمل، القيصرية الأولية، الإجهاض، اضطرابات الهيكل العظمى والجهاز التنفسى لحديثى الولادة، وتقييد نمو الجنين داخل الرحم، خطر الولادة المبكرة، انخفاض الوزن عند الولادة، ونقص الكالسيوم فى الدم عند الأطفال حديثى الولادة.

الهدف من الدراسة هو تقييم حالة فيتامين د للأم وارتباطها بنتائج الحمل.

تم إجراء هذه الدراسة على ثمانين امرأة حامل فى الفئة العمرية من ١٨–٣٨ سنة، مع عدم وجود اضطرابات طبية أو تناول فيتامين (د) خلال فترة الحمل. تم استبعاد الحالات التى تعانى من الأمراض المزمنة مثل أمراض الكبد والالتهابات الشديدة أو تناول الكورتيكوستيرويدات. الحالات المشاركة فى الدراسة تم اختيارها من قسم التوليد وأمراض النساء، مستشفى المنصورة الجامعى من مايو ٢٠٢٢ إلى مايو ٢٠٢٣ بعد موافقة اللجنة الأخلاقية بكلية الطب جامعة المنصورة وبعد الحصول على الموافقة المكتوبة من المرضى على إجراء خطوات الدراسة.

قد خلصت الدراسة إلى أنه لا يزال نقص فيتامين (د) لدى النساء الحوامل وأطفالهن مشكلة صحية خطيرة. ومع ذلك، خلصت نتائجنا إلى أنه لا توجد ارتباطات ذات دلالة إحصائية بين فيتامين د فى الأمهات والنتائج فى كل من الأمهات وحديثى الولادة باستثناء سكرى الحمل.