Association of Serum Vitamin D Status with gestational diabetes mellitus and other laboratory parameters in early pregnant women

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Research Article

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Abstract

Background: Pregnant women develop certain metabolic alterations and substantial nutrient insufficiency, which may impact fetus development. Reports on the association of serum 25-hydroxy vitamin D (25(OH)D, VD) status with gestational diabetes mellitus (GDM) is still controversial. This study aimed to investigate the association of serum VD status with GDM and other laboratory parameters in early pregnant women.

Methods: A total of 1516 pregnant women were enrolled in this study. Maternal serum VD and other laboratory parameters—including hematology, coagulation, chemistry, and bone density—were measured utilizing various analytical methods in clinical laboratory at gestational weeks 11 to 14. Serum VD level was subsequently correlated with blood glucose and other laboratory parameters.

Results: The average VD concentration was 59.12 ± 12.5 nmol/L. None of the study subjects had VD <25 nmol/L; 434 (28.6%) women had VD <50 nmol/L, and 200 women (13.2%) had VD ≥ 75 nmol/L. There were 264 (17.4%) women diagnosed with GDM. There was not, however, a correlation between serum VD and GDM in early pregnancy. Interestingly, women with more parity and high serum alkaline phosphatase levels had higher serum VD levels. There was a positive correlation between serum VD and pre-albumin, activated partial thromboplastin time, and a negative correlation between serum VD, creatinine, and thrombin time.

Conclusions: There was no correlation between maternal serum VD and GDM in early pregnant women. There were, however, associations between serum VD and parity, serum alkaline phosphatase, creatinine, pre-albumin, and coagulation factors, which need further study to explain their pathophysiology and clinical significance.

Background

Vitamin D (VD) is a fat-soluble vitamin, which plays an important role in bone mineralization, calcium and phosphorus absorption, parathyroid status and immune system functionality [1]. In the human body, VD from sunlight and food is hydroxylated to 25-hydroxyvitamin D (25(OH)D) in the liver and is subsequently activated with a second hydroxylation to 1,25-dihydroxy vitamin D in the kidney [2].

Study found that during pregnancy, calcitriol increases in early gestation to meet a mother's needs, and maternal serum 25(OH)D crosses into the placenta for use in fetus development [3]. Abnormal VD levels may cause some adverse pregnancy complications, including polycystic ovary syndrome, gestational diabetes mellitus (GDM), pre-eclampsia, infertility, endometriosis, and cancers [4-7]. Studies on the association between VD status and pregnancy complications are still controversial. On the one hand, some studies indicated that the alteration of VD was associated with a variety of adverse pregnancy outcomes, especially GDM [8-10]. On the other hand, some studies failed to correlate VD status with the above-mentioned consequences [11,12].

In this study, the association between maternal serum VD levels and GDM and other laboratory parameters in early pregnancy were observed to help better understand metabolic events and their potential effects on fetus development, and the potential diagnostic and prognostic applications.

Patients And Methods

Patient enrollment and ethics approval

A total of 1516 women with ages from 18 to 46 were recruited from August 1, 2018 to August 31, 2019 at Foshan Women and Children's Hospital of Southern Medical University, which is a specialized tier-3, grade-A hospital in South China. Pregnant women with singleton pregnancies and conceiving naturally were included. The exclusion criteria included pre-existing diabetes, hypertension or other chronic diseases, abnormal child-bearing history, a history of drug use, and vitamin supplements—including vitamin D and calcium—that may affect metabolism or unavailable data. All pregnant women exerted a normal daily life without special sunlight exposure or quarantines. All the study subjects were living in the in the Foshan, Guangdong Province of South China. Ethics approval was obtained by the Institutional Review Board (IRB) and Ethics Committee of Foshan Women and Children's Hospital of Southern Medical University. All methods were carried out in accordance with relevant guidelines and regulations. Methods and laboratory testing are routine and standard operating required for medical practice in Guangdong Province including this hospital. All participants provided written informed consent.

Serum vitamin D analysis

Serum VD was analyzed at gestational weeks 11 to 14 using a Mokosensor-A300 immune colloidal gold analyzer (MedicalSystems, Ningbo, China) following the manufacturer's instructions.

Chemistry analysis

Fasting and 1h, 2h oral glucose tolerance test (OGTT) blood glucose, alanine aminotransferase, alkaline phosphatase, pre-albumin, creatinine, cystatin C, calcium, and magnesium were analyzed on a Beckman-Coulter Automatic Chemistry Analyzer platform (Beckman-Coulter, Brea, CA, USA) following the manufacturer's instructions and laboratory operating procedures. The collection of blood samples, including the selection of a collecting tube, was followed by routine clinical laboratory procedures.

Glucose tolerance test

A 75g oral glucose tolerance test (75g OGTT) was performed between 24 and 28 weeks of gestation. Either of the following criteria met was diagnosed as GDM: fasting serum glucose ≥ 5.1 mmol/L, 1h of OGTT serum glucose ≥ 10 mmol/L, or 2h of OGTT serum glucose ≥ 8.5 mmol/L.

Hematology analysis

Hematology analysis was performed on a Sysmex XN2000 hematology analyzer platform (Sysmex, Kobe, Japan), and hematological parameters, including the indices, were obtained.

Coagulation test

The blood coagulation test was performed on a Stago STA-R Evolution (Diagnostica Stago, Asnières sur Seine Cedex, France) platform following the manufacturer's instructions using a heparin anti-coagulant blood collection tube.

Bone density examination

Bone density was measured using a Hong Yang BMD-1000 Ultrasound Bone Sonometer (Baoding, Hebei, China) following the manufacturer's instructions.

Statistical Analysis

Statistical analysis was performed using the SPSS (version 24). Independent-samples t-test, One-Way ANOVA, and Pearson's correlation were used in the analysis. A $P < 0.05$ were considered statistically significant.

Results

Basic information of study subjects

Table 1 lists the basic information of study subjects in this study.

Association between maternal serum vitamin D level and gestational diabetes mellitus

Serum VD level was correlated with maternal fasting, OGTT 1h, 2h blood (serum) glucose levels. VD was also correlated with GDM and non-GDM status of pregnant women. As shown in Table 2, there is no association between VD and glucose levels in early gestation women ($p > 0.05$ for all).

Correlation of serum vitamin D level with age, blood chemistry, coagulation, and trace elements parameters

While correlating chemistry parameters with VD level in serum, results showed that there was no association between serum VD and age, alanine aminotransferase (ALT), cystatin C, calcium, and magnesium, as shown in Table 3 ($p > 0.05$ for all).

Correlation of serum vitamin D with hematologic parameters

Serum VD level was also correlated with hematologic parameters of early pregnant women. Table 4 shows that there is no association between serum VD level and red blood cells (RBC), hemoglobin (HGB), white blood cells (WBC) and neutrophils (Neu, %), and platelet (PLT) ($p > 0.05$ for all).

Correlation of serum vitamin D with parity

Serum VD level in pregnant women was further correlated with parity number. Table 5a and 5b shows that women going into birth 3 times had a higher VD level than those with 2 or 1 parities ($p=0.016$ and $p=0.039$, respectively).

Association between serum vitamin D level and alkaline phosphatase activities

There was a positive correlation between serum VD level and alkaline phosphatase activities, of which a higher serum VD level tended to have higher alkaline phosphatase activities (Table 6a and 6b, $p=0.005$ and 0.004 , respectively).

Correlation of serum vitamin D with bone density, body weight of newborns, coagulation, and other chemistry parameters (Pearson's correlation)

Furthermore, maternal serum VD was correlated with bone density, the body weight of newborns, coagulation, and other chemistry parameters besides those mentioned above. Table 7 shows that serum VD level had a positive correlation with activated partial thromboplastin time (aPTT) ($p=0.047$), pre-albumin ($p=0.001$), and a negative correlation with thrombin time (TT) ($p=0.000$), creatinine ($p=0.006$). There was no association between serum VD level and bone density, body weight of newborns, D-Dimer, and fibrinogen ($p>0.05$ for all) (Table 7).

Discussion

The association between maternal serum VD and diabetes in early pregnancy is still controversial [13,14]. GDM was the most common complication of pregnancy with a prevalence of 1% to >30% globally [15]. With excessive peripheral insulin resistance and insufficient insulin production, elevation of blood glucose increases the risk of short-term and long-term consequences in both mother and infant, including pre-eclampsia, macrosomia, cardiovascular disease and obesity [16]. Previous studies found that VD deficient rats had lower insulin secretion than VD deficient rats supplemented with VD [17]. A meta-analysis of 29 observational studies indicated that VD insufficiency was related to increased risk of GDM [18]. Similarly, high serum 25(OH)D in early pregnancy protects from the development of GDM [19].

The results from this study indicated that there was no association between maternal serum VD level and GDM (Table 2). This result is similar with the report from a previous meta analysis [20]. A randomized controlled trial also showed a limitation of VD supplementation for GDM prevention in VD sufficient populations [21]. Since VD level is affected by several factors, such as seasons, geographical location, race, diet/lifestyle, medical conditions, etc., it is difficult to perform quality control and draw comparisons among studies. In addition, the lack of standardized measurement methods is another concern. Thus, a consensus on VD study is needed for guiding reliable VD related studies in the future.

Growing evidence has indicated that serum VD deficiency may lead to other pregnancy adverse outcomes, such as macrosomia, pre-eclampsia, and other health issues [22,23]. A previous study,

however, demonstrated that VD supplementation hardly reduced the risk of pre-eclampsia, preterm birth, and low body weight [24]. This study's results suggested that there was no association between maternal VD level and age (≤ 35 vs >35), serum alanine aminotransferase (ALT), cystatin C, calcium, and magnesium (Table 3). Serum VD level was then further correlated with maternal hematological parameters red blood cells (RBC), hemoglobin (HGB), white blood cells (WBC) and the percentage of neutrophils (Neu, %), and platelets (PLT). However, the statistical results were all insignificant (Table 4).

In this study, we found that there was a positive correlation between maternal serum VD level and the number of parity; women who had given birth 3 times had higher serum VD than those giving birth 2 or 1 time (s) ($p=0.016$ and 0.039 , respectively, Table 5a and 5b). One study mentioned that there was no significant difference between the serum VD level and age, parity, or gestational age [25]. The mechanism underlying this phenomenon is therefore unclear; there could be certain compensation in metabolism function during pregnancy that requires further investigation in the future.

Our results also indicated that there was a positive correlation between serum VD and alkaline phosphatase (ALP) activity; women who had high serum VD also had higher ALP activity (Table 6a and 6b). A study observed that there was an association between ALP and moderate VD deficiency group; however, the correlation was not significant [26]. Another study also found that there was no correlation between ALP and vitamin status in school children [27].

Serum ALP is widely used in clinical practice as a marker of bone turnover [28]. Medical conditions with osteolysis had elevated serum ALP levels [29]. Based on our results, we hypothesize that there is a relationship between VD and ALP in maintaining the bone and mineral metabolic homeostasis, a hypothesis that merits further investigation.

Lastly, our study also found that there was a positive correlation between serum VD level and thrombin time (TT) measurement in pregnant women, which means that VD had an effect on blood coagulation. A previous study showed that treatment with VD supplementation had a prothrombotic effect in patients with VD insufficiency, an issue that requires further study [30]. We also observed, however, that there was a positive correlation between maternal serum VD level and activated partial thrombin time (aPTT) (Table 7). The mechanism underlying this phenomenon is unclear. We hypothesize that VD plays a role in serum calcium turnover, leading to the postpone of aPTT which requires further investigation. Our results also indicated that there was a negative correlation between serum VD and creatinine level, a positive correlation between serum VD and pre-albumin levels in pregnant women, which suggests that VD plays a role in creatinine clearance and enhancing albumin production (Table 7).

Conclusion

This study did not show an association between maternal serum VD level and GDM. There was a positive correlation between serum VD and parity, serum alkaline phosphatase, activated partial thrombin time, pre-albumin levels, and a negative correlation between serum VD and thrombin time and serum creatinine levels, which could have potential clinical significance and requires further investigation.

Declarations

Acknowledgments: None.

Author contributions: CL, MD: principle investigator and conception; ZL, MD and LJ, MS: statistical analysis; YL, MD, FW, MD, DS, MD, SL, MD, and AC, BS: patients' enrolment and clinical information collection; ZR, BS: data collection and integration; RJ, BS: laboratory analysis and data collection; FH, BS: Bone Densitometry; HH, MD, PhD: manuscript writing; XL, MD: project organization and conception.

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Availability of data and materials

Data is available from the corresponding author upon inquiry.

Ethics approval and consent to participate

Ethics approval was obtained by the Institutional Review Board (IRB) and Ethics Committee of Foshan Women's and Children's Hospital of Southern Medical University. The approval document number is FSFY-MEC-2018-014, the file will be provided upon inquiring. All experimental protocols were approved by the IRB of the hospital. All methods were carried out in accordance with relevant guidelines and regulations. Methods and laboratory testing are routine and standard operating required for medical practice in Guangdong Province including this hospital. All participants provided written informed consent.

Consent for publication

Not applicable.

Competing interests

The authors declare no conflict of interest.

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Tables

Table 1 Basic information of study subjects

Parameters	n	Min	Max	Mean	STDEV
Age	1516	18	46	29.42	4.58
BMI	1516	13.80	40.90	26.76	3.25
BWON	1516	940	4640	3182.32	452.04
Vitamin D	1516	27.35	102.40	59.12	12.57
Glucose	1516	3.40	6.30	4.31	0.34
ALT	1489	2.00	481.00	13.16	16.07
ALP	972	23.00	290.00	83.93	31.74
Pre-albumin	951	63.00	349.00	222.83	32.85
Creatinine	1466	27.00	96.00	47.14	7.21
Cystatin C	323	0.47	1.84	0.96	0.22
Calcium	1478	0.94	1.84	1.37	0.13
Magnesium	1477	0.97	1.93	1.33	0.15
Bone density Z value	924	-3.06	1.34	-0.51	0.49
D-Dimer	1431	0.10	9.06	0.47	0.39
Fibrinogen	1437	0.78	6.74	4.02	0.66
aPTT	1437	2.00	48.40	36.28	3.07
TT	1436	13.00	27.20	14.80	0.87

BMI: body mass index; BWON: Body weight of newborn; aPTT: activated partial thromboplastin time; TT: thrombin time; Min: minimum; Max: maximum; STDEV: standard deviation

Table 2 Correlation of diabetic status with serum vitamin D levels (independent-samples t-test)

	Glucose/ GDM	n	Average VD	STDEV	t	<i>p</i>
Fasting glucose	<5.1	1478	59.09	12.59	-0.663	0.507
	≥5.1	38	60.46	11.48		
Glucose of 1h GTT	<10	1358	59.27	12.59	1.350	0.178
	≥10	158	57.86	12.36		
Glucose of 2h GTT	<8.5	1342	59.30	12.56	1.491	0.136
	≥8.5	174	57.79	12.54		
GDM	No	1252	59.27	12.58	0.940	0.347
	Yes	264	58.46	12.52		

GDM: gestational diabetes mellitus; glucose (mmol/L); GTT: glucose tolerance test

Table 3 Correlation of serum vitamin D level with age and blood chemistry parameters (independent-samples t-test)

Parameters		n	Average VD	STDEV	t	<i>p</i>
Age	<=35	1303	59.23	12.628	0.813	0.416
	>35	211	58.47	12.245		
ALT	<50	1472	59.15	12.55	0.858	0.391
	>=50	17	56.53	12.19		
Cystatin C	<1.17	274	59.12	12.45	1.355	0.176
	>=1.17	49	56.49	12.97		
Calcium	<1.42	942	59.04	12.75	-0.857	0.392
	>=1.42	536	59.63	12.38		
Magnesium	<1.12	71	58.33	11.38	-0.634	0.526
	>=1.12	1406	59.31	12.68		

ALT: alanine aminotransferase (U/L); Cystatin C (mg/L); Calcium (mmol/L); Magnesium (mmol/L)

Table 4 Correlation of serum vitamin D with hematologic parameters (independent-samples t-test)

Indices	Values	n	Mean	STDEV	t	<i>p</i>
RBC	<3.5	58	57.30	10.33	-1.356	0.180
	>=3.5	1439	59.20	12.66		
HGB	<=110	232	58.91	12.28	-0.282	0.778
	>110	1265	59.16	12.63		
WBC	<10	1081	59.08	12.63	-0.239	0.811
	>=10	416	59.25	12.46		
Neu (%)	<70	483	58.80	12.69	-0.683	0.495
	>=70	1014	59.28	12.53		
PLT	100-300	1244	59.31	12.61	1.298	0.195
	>300	253	58.19	12.39		

RBC: red blood cell (*10¹²/L); HGB: hemoglobin (g/L); WBC: white blood cell (*10⁹/L); Neu: neutrophils (% of WBC); PLT: platelet (*10⁹/L)

Table 5a Correlation of serum vitamin D with parity (One-Way ANOVA)

Parity	n	Mean	STDEV	SEM	Min	Max	F	<i>p</i>
1	784	59.20	12.58	0.45	27.35	102.40	1.949	0.120
2	652	58.66	12.44	0.49	27.43	92.50		
3	74	62.35	13.34	1.55	39.83	94.55		

Table 5b Correlation of serum vitamin D with parity (One-Way ANOVA)

(I) Parity	(J) Parity	Diff. of the means (I-J)	SEM	Sig.
1	2	0.54289	0.66543	0.415
	3	-3.15422	1.52677	0.039
2	3	-3.69711	1.54004	0.016

SEM: standard error of the mean; Diff. of the means: difference of the means; Sig.: significance

Table 6a Correlation of serum vitamin D and alkaline phosphatase activities (One-Way ANOVA)

ALP	n	Mean	STDEV	SEM	Min	Max	F	<i>p</i>
<45	63	54.82	13.17	1.66	27.35	91.30	4.724	0.009
45-125	822	59.47	12.72	0.44	27.43	102.40		
>125	87	60.90	11.57	1.24	34.78	91.15		

ALP: alkaline phosphatase activities

Table 6b Correlation of serum vitamin D and alkaline phosphatase activities (One-Way ANOVA)

(I) ALP	(J) ALP	Diff. of the means (I-J)	SEM	Sig.
<45	45-125	-4.65378*	1.65429	0.005
	>125	-6.08375*	2.09344	0.004
45-125	>125	-1.42998	1.42670	0.316

Table 7 Correlation of serum vitamin D with bone density, body weight of newborns, coagulation, and other chemistry parameters (Pearson's correlation)

Parameters	n	Vitamin D	
		Pearson Correlation	Sig. (2-tailed)
Bone density Z value	924	0.019	0.555
Body weight of newborns	1516	-0.001	0.955
D-Dimer	1431	0.035	0.180
Fibrinogen	1437	0.043	0.107
TT	1436	-0.178	0.000
aPTT	1437	0.052	0.047
Creatinine	1466	-0.071	0.006
Pre-albumin	951	0.108	0.001

TT: thrombin time; aPTT: activated partial thromboplastin time