

Insulin resistance, obesity and hypovitaminosis D adversely affect pregnancy outcome: Can Supplemental vitamin D cut this risk?

Original
Article

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ABSTRACT

Aim of work: To estimate serum 25-hydroxy vitamin D (25OH-VD) and insulin, and fasting blood glucose (FBG) early in pregnancy, determine the frequency and severity of insulin resistance (IR), gestational diabetes mellitus (GDM) and pre-eclampsia (PE) during pregnancy and the impact of VD supplemental therapy (VD-ST) on these effects.

Patients and Methods: 494 pregnant women fulfilling the inclusion criteria were randomly divided into two equal groups: Study group received VD-ST as a daily oral dose of 1000 IU soft gels with meal since 6th week till delivery, while control group did not receive VD-ST. All women gave blood samples for colorimetric estimation of FBG and ELISA estimation of serum insulin and 25-OH-VD levels. Evaluated parameters included body mass index (BMI), VD sufficiency status and homeostasis model assessment IR (HOMA-IR) score. Oral glucose tolerance test for diagnosis of GDM was performed at the 20th, 28th and 36th week GA and blood pressure was measured regularly at follow-up visits for diagnosis of PE.

Results: At time of enrolment, 405 women (81.9%) were overweight-obese, 86 women (17.4%) had IR and only 63 women (12.8%) had sufficient serum 25OH-VD level. At 3rd trimester, 68 women (13.8%) developed GDM, 71 women (14.4%) developed PE and 23 women (4.7%) developed both with significantly lower incidence in women who received VD-ST. Frequency of GDM and PE showed positive significant correlation with BMI and HOMA-IR score, while showed negative significant correlation with serum 25OH-VD. ROC curve analysis defined low 25OH-VD level and high HOMA-IR score as significant sensitive predictors for development of both GDM and PE ; while receiving VD-ST was the significant specific predictor for possibility of amelioration of such event. Kaplan-Meier regression curve defined a cumulative hazard for developing both GDM and PE of <20% with and 60% without VD-ST.

Conclusion: VD deficiency-insufficiency is a problem that requires national evaluation for predisposition and progress. The triad of maternal hypovitaminosis D, obesity and IR is associated with development of GDM and/or PE. The proposed VD supplementation regimen effectively reduced the frequencies of pregnancy-associated or induced complications; so it is effective to break that triad.

Key Words: Gestational diabetes mellitus, hypovitaminosis D, insulin resistance, pre-eclampsia, pregnancy, vitamin D supplemental therapy.

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INTRODUCTION

Obesity is a worldwide epidemic with annually increasing number of overweight and obese people up to overtaking the number of malnourished^[1]. Dietary energy density was directly associated with higher weight change and risk of excess adiposity^[2]. Visceral adipose tissue contributed beyond overall adiposity, particularly in women (3) and is associated with multiple comorbidities^[4].

Vitamin D (VD) is an important secosteroid hormone which after intestinal absorption was carried in blood stream bound to a binding protein to the liver to undergo hydroxylation to 25-OH VD (calcidiol)^[5]. Then, calcidiol

undergoes another hydroxylation in the kidneys to 1, 25-dihydroxy-VD (calcitriol); the biologically active form of VD^[6]. Biologically active VD acts through VD receptors that are widely distributed in skeletal and many non-skeletal tissues^[7] to produce a number of biological effects and therefore, contributes to human health improvement^[8].

Insulin resistance (IR) is typically defined as decreased responsiveness to insulin metabolic actions^[9], as insulin-mediated glucose disposal in skeletal muscle and adipose tissue^[10], through lower expression of glucose transporters in liver and skeletal muscle^[11] and/or disturbance in coupling of stimulation of insulin receptor by insulin^[12] ending at compensatory increase in insulin concentration despite of increased blood glucose levels^[11]. IR plays a major role in pathogenesis of type 2 diabetes

mellitus^[13], hypertension, coronary artery disease, and is tightly associated with development of obesity and its related problems^[14].

Obesity, VD deficiency and IR alone or together may play key role in the development of multiple gynecological/obstetric diseases^[15] where the maternal-fetal unit is under the influence of vitamin D, so breakdown in its homeostasis may underlie infertility, pre-eclampsia (PE), and gestational diabetes mellitus (GDM)^[16]. Women who developed insufficient β -cell compensation for pregnancy-induced IR were mostly liable to develop GDM with its related maternal and fetal complications^[17]. Also, obesity and or gestational weight gain is associated with fetal large for gestational age (GA)^[18], increased body fat mass especially abdominal fat^[19] and is associated with neonatal cardiometabolic makers independent of neonatal adiposity^[20].

Hypothesis

The current study hypothesized the presence of interplay between maternal obesity, IR and hypovitaminosis D (Hypo-VD) early in pregnancy for development of pregnancy-induced GDM and/or PE and early correction of Hypo-VD could break this vicious circle if present.

Objectives

Estimation of serum 25OH-VD and insulin, and fasting blood glucose (FBG) early after assurance of diagnosis of pregnancy, to determine the frequency of IR, GDM and PE throughout pregnancy and the role of VD supplemental therapy (VD-ST) for lessening or ameliorating these effects.

Design

Prospective observational comparative clinical trial

Setting

Benha University Hospitals

PATIENTS AND METHODS

The study protocol was approved by the Local Ethical Committee and only women signed written fully informed consent were included in the study. All pregnant women who attended the Antenatal Outpatient Clinics for assurance of being pregnant were eligible for evaluation for study inclusion. After assuring pregnancy diagnosis, all women were asked to attend the clinic fasting on the next morning for clinical evaluation and giving fasting blood sample for laboratory investigations. Women with current DM, history of previous GDM or obesity-inducing endocrinopathy, evident manifestations of hypoparathyroidism, thyrotoxicosis, renal or hepatic diseases, essential hypertension and women lost during the course of pregnancy were excluded from the study.

Enrolled women were randomly allocated, using sealed

envelops containing cards labeled by group title, prepared by a blinded assistant and chosen by pregnant woman herself, into two groups:

- Control group included pregnant women who will complete their pregnancy without VD-ST.

- Study group included pregnant women assigned to receive VD-ST since 6th week GA till delivery according to Grant *et al.*^[21] in a daily oral dose of 1000 IU soft gels to be taken with meal (Sunvite High Potency Vitamin D3 1000 IU; Puritan's Pride, Inc., Oakdale, NY, USA).

Laboratory investigation

A) Colorimetric estimation of FBG by glucose oxidase method^[22] at time of enrollment and during each follow-up visit.

B) ELISA estimation of serum 25OH-VD levels (Cayman Chemical, Ann Arbor, MI, USA)^[23] and insulin (Mercodia ELISA; ALPCO Diagnostics, Uppsala, Sweden)^[24].

Clinical Evaluation

1. Body mass index (BMI) was calculated using the formula: weight (kg)/ height (m²) and patients were classified according to BMI using the World Health Organization ranges as underweight: BMI<18.5 kg/m², normal weight: BMI=18.5-24.9 kg/m², overweight: BMI=25-29.9 kg/m² and obese: BMI \geq 30 kg/m²^[25]. Obesity grades were defined after the WHO expert consultation^[26] as BMI <24.9 as average, 25-<30 kg/m² as overweight, BMI \geq 30-<35 kg/m² as obese and BMI \geq 35 kg/m² as morbid obese

2. Vitamin D sufficiency status was defined according to 25OH-VD concentration as follows: \geq 75 nmol/L sufficient level, 50-75 nmol/L insufficient level and <50 nmol/L deficient level. Vitamin D deficiency was categorized as mild, moderate and severe if 25OH-VD concentration was 25-50 nmol/L, 12.5-25 nmol/L and <12.5 nmol/L, respectively^[27].

3. Insulin resistance (IR) was measured by homeostasis model assessment IR (HOMA-IR) score. The HOMA-IR score was calculated as fasting serum insulin (μ U/ml) x [FBG (mg/ml)/18]/22.5^[28] was considered abnormal at HOMA-IR score of \geq 2^[29].

4. Gestational diabetes mellitus (GDM): All enrolled women were asked to fast and attend the antenatal care unit at the 20th, 28th and 36th week GA and underwent the 75g Oral Glucose Tolerance Test for diagnosis of GDM according to the criteria of the International Association of Diabetes and Pregnancy Groups^[30] for abnormal OGTT, as follows: FBG \geq 92 mg/dl, 1-h BG \geq 180 mg/dl and 2-h BG \geq 153 mg/dl.

5. Blood pressure (BP) was measured and recorded regularly at follow-up visits for diagnosis of PE which was defined as the development of hypertension after

the 12th week GA in women who were normotensive at time of 1st antenatal visit with systolic BP (SBP) ≥ 140 mmHg and/or diastolic BP (DBP) ≥ 90 mmHg on at least two occasions, 4 hours apart, and proteinuria (one dipstick measurement $\geq 2+$ on a voided random urine sample)^[31,32].

Study outcomes

1. Frequency of development of GDM and PE throughout pregnancy.
2. Relation between preliminary data and frequency of GDM and PE.
3. Effect of VD-ST on frequency and severity of GDM and PE.

Statistical analysis

Obtained data were presented as mean \pm SD, ranges, numbers and ratios. Results were analyzed using paired t-test. Possible correlations were evaluated using Spearman's correlation coefficient and these correlations were verified as sensitivity and specificity for prediction of

development of GDM and PE using the receiver operating characteristic (ROC) curve analysis judged by the area under the curve (AUC). Statistical analysis was conducted using the SPSS (Version 15, 2006) for Windows statistical package. *P* value < 0.05 was considered statistically significant.

RESULTS

Throughout the study duration, 526 women were eligible for evaluation; 32 were excluded for not fulfilling the study inclusion criteria and 494 women were enrolled and randomly divided into two equal groups. At time of enrolment, calculated BMI defined 97 obese (19.6%), 308 overweight (62.3%) and 89 average (18.1%) women. Estimation of FBG and serum insulin and calculation of HOMA-IR score defined 86 women (17.4%) had IR. Estimation of serum VD defined 63 women (12.8%) had sufficient, 142 women (28.7%) had insufficient and 289 women (58.5%) had deficient VD levels. There was non-significant ($p > 0.05$) difference between both groups as regards enrolment data as shown in table 1.

Table 1: Baseline data of women enrolled in both groups

| Data | | Control (n=247) | Study (n=247) | P value | |
|--------------------------------|---------------|-----------------|-----------------|------------------|-------|
| Age (years) | | 33.9 \pm 2.4 | 33.6 \pm 2.1 | 0.978 | |
| BMI data | Weight (kg) | 80 \pm 9.2 | 81.1 \pm 6.4 | 0.098 | |
| | Height (cm) | 169.6 \pm 3.7 | 169.4 \pm 3.7 | 0.338 | |
| Blood pressure measures (mmHg) | Average | 62 (25.1%) | 47 (19%) | 0.229 | |
| | BMI | Overweight | 144 (58.3%) | 151 (61.1%) | 0.059 |
| | Obese | 41 (16.6%) | 49(19.9%) | | |
| | BMI (kg/m2) | | 27.8 \pm 3.1 | 28.3 \pm 2.5 | |
| Insulin resistance (IR) data | Systolic | | 118.3 \pm 3.8 | 118 \pm 4.6 | 0.863 |
| | Diastolic | | 82.5 \pm 4.3 | 83.6 \pm 3.1 | 0.627 |
| | FBG (mg/dl) | | 118.9 \pm 11 | 120.9 \pm 12.2 | 0.068 |
| | Serum insulin | | 4.85 \pm 1.3 | 4.7 \pm 1.2 | 0.597 |
| | IR | Yes | 45 (18.2%) | 41 (16.6%) | 0.635 |
| No | 202 (81.8%) | 206 (83.4%) | | | |
| VD sufficiency data | HOMA-IR score | | 1.44 \pm 0.41 | 1.45 \pm 0.46 | 0.843 |
| | Sufficient | | 31 (12.7%) | 32 (13%) | 0.712 |
| | Insufficient | | 68 (27.5%) | 74 (30%) | |
| | Mild | | 92 (37.2%) | 85 (34.4%) | |
| Deficient | Moderate | | 49 (19.8%) | 44 (17.8%) | 0.798 |
| | Severe | | 7 (2.8%) | 12 (4.8%) | |
| | Level | | 45.8 \pm 20.3 | 46.3 \pm 20.8 | |

Data are presented as mean \pm SD and numbers; percentages are in parenthesis; BMI: Body mass

At the start of 3rd trimester, all women had higher FBG and serum insulin levels with higher HOMA-IR score compared to their respective baseline measures. The 3rd trimester measures were significantly ($p<0.05$) higher in control women compared to their baseline measures and to 3rd trimester measures of study women. On the other hand, FBG and calculated HOMA-IR score were significantly higher, while serum insulin levels were non-significantly higher in study women compared to their

baseline measures. As regards IR, at the 3rd trimester, 166 women (33.6%) were IR; 113 in control and 53 women in study group with significantly ($p<0.05$) higher difference in frequency of IR between both groups and in control group versus its 12th wk frequency. Moreover, at the start of 3rd trimester, 68 women (13.8%) developed GDM; 45 (9.3%) and 23 (9.3%) women in control and study groups, respectively with significantly ($p=0.004$) lower difference in favor of study group (Table 2).

Table 2: Glucogenic data detected at 3rd trimester compared to at the 12th wk GA in both groups

| Group | | Control | | | Study | | |
|---------------|-----------|-------------|---------------|---------|-------------|---------------|-------|
| Parameter | Time | 12th wk GA | 3rd trimester | P value | 12th wk GA | 3rd trimester | P= |
| FBG | | 118.9±11 | 136.7±9.8 | 0.0001 | 120.9±12.2 | 124.3±23* | 0.001 |
| Insulin | | 4.85±1.3 | 5.85±1.32 | <0.001 | 4.7±1.2 | 4.98±1.19* | 0.248 |
| HOMA-IR score | Frequency | 45 (18.2%) | 113 (45.7%) | <0.001 | 41 (16.6%) | 53 * (23.9%) | 0.169 |
| | Score | 202 (81.8%) | 134 (54.3%) | | 206 (83.4%) | 194 (76.1%) | |
| GDM | Y | 0 | 45 (18.2%) | | 0 | 23 (9.3%) | |
| | N | 0 | 202 (81.8%) | | 0 | 224 (90.7%) | |

Data are presented as mean±SD and numbers; percentages are in parenthesis; FBG: Fasting blood glucose; HOMA-IR: Homeostasis model assessment of insulin resistance; IR; GDM: Gestational diabetes mellitus

Furthermore, at the start of 3rd trimester, all women got increased blood pressure measures. SBP measures were significantly higher, while DBP measures were non-significantly higher in both groups compared to their baseline measures with significantly higher measures in control versus study women. Seventy-one women developed high blood pressure measures up to levels

diagnosed as PE; 46 in control and 25 in study group with significantly higher frequency among women of study women compared to their baseline measures (Table 3). Twenty-three women (4.7%) developed both GDM and PE; 16 in control and 7 in study groups with significantly lower incidence of both GDM and PE among study versus control women.

Table 3: Blood pressure data estimated at 3rd trimester compared to at the 12th wk GA in both groups

| Group | | Control | | | Study | | |
|------------|------|------------|---------------|---------|------------|---------------|---------|
| Parameter | Time | 12th wk GA | 3rd trimester | P value | 12th wk GA | 3rd trimester | P value |
| SBP (mmHg) | | 118.3±3.8 | 123.2±12.5* | 0.009 | 118±4.6 | 121.6±13.9 | 0.039 |
| DBP (mmHg) | | 82.5±4.3 | 85.6±6.9 | 0.078 | 83.6±3.1 | 84.1±8.5 | 0.078 |

Data are presented as mean±SD and numbers; percentages are in parenthesis; SBP: Systolic blood pressure; DBP: Diastolic blood pressure

Development of GDM showed positive significant correlation with BMI and HOMA-IR score, while showed negative significant correlation with serum 25-OHD. Similarly, development of PE were positively correlated with BMI and FBG at 3rd trimester and

negatively correlated with serum 25-OHD. Moreover, development of both GDM and PE positively correlated with BMI, FBG at 3rd trimester, HOMA-IR score and negatively correlated with serum 25-OHD (Table 4).

Table 4: Correlation of the development of GDM or PE in the 3rd trimester with BMI, HOMA-IR score and serum 25-OHD

| | BMI | | FBG at 3rd trimester | | HOMA-IR score | | Serum 25-OHD | |
|------|-------|--------|----------------------|--------|---------------|--------|--------------|--------|
| | Rho | P | Rho | P | Rho | P | Rho | P |
| GDM | 0.171 | <0.001 | 0.533 | <0.004 | 0.149 | <0.001 | -0.211 | <0.001 |
| PE | 0.190 | <0.001 | 0.107 | =0.017 | 0.061 | 0.177 | -0.219 | <0.001 |
| Both | 0.219 | <0.001 | 0.388 | <0.001 | 0.131 | 0.004 | -0.248 | <0.001 |

Rho: Spearman's correlation coefficient; BMI: Body mass index; FBG: Fasting blood glucose; HOMA-IR: Homeostasis model assessment of insulin resistance; IR; 25-OHD: 25-hydroxy vitamin D; GDM: Gestational diabetes mellitus

ROC curve analysis of clinical and laboratory data of enrolled women as predictors for development of both GDM and PE defined Hypo-VD as the significant sensitive predictor for such event followed by high HOMA-IR score and FBG at the 3rd trimester, while receiving VD-

ST was the significant specific predictor for possibility of amelioration or prevention of such event (Fig. 1). Kaplan-Meier Regression curve defined a cumulative hazard for developing both GDM and PE of <20% with and 60% without VD-ST (Fig. 2).

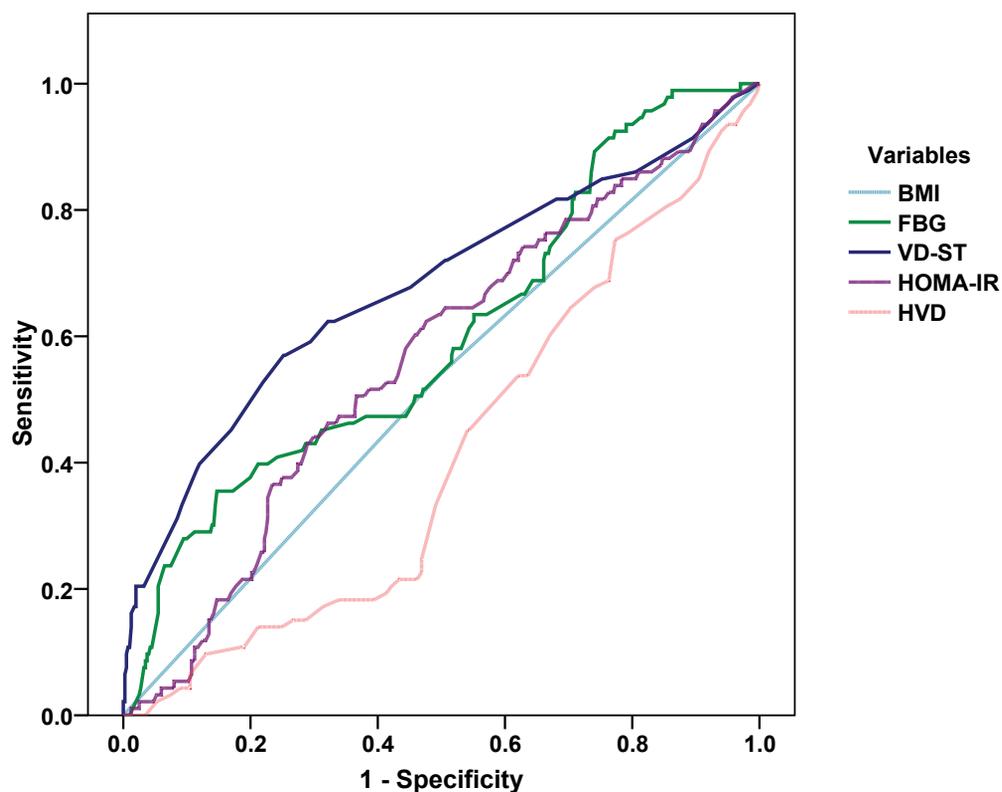


Fig. 1: ROC curve analysis for variable as predictors for development of both GDM and PE

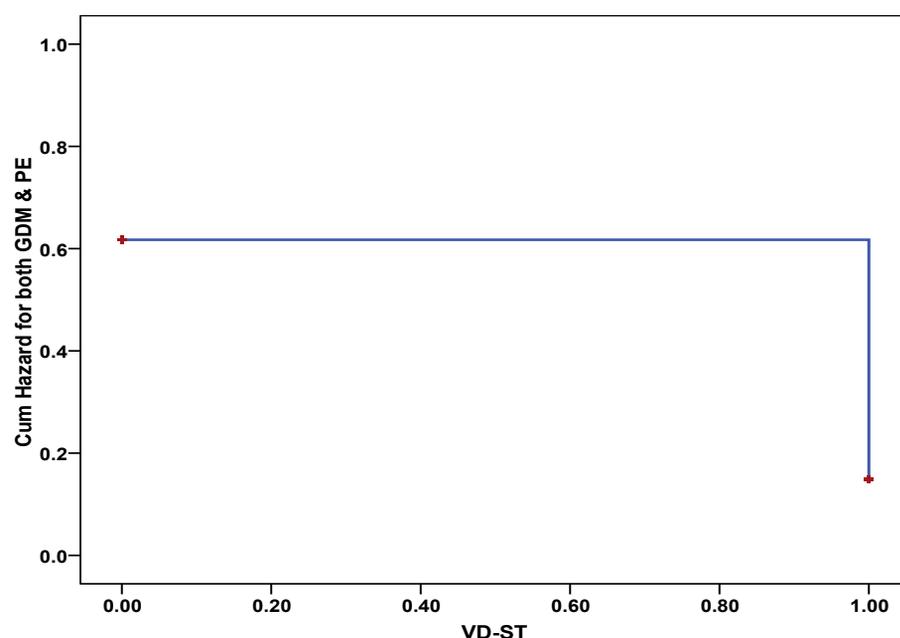


Fig. 2: Kaplan-Meier Regression curve for cumulative hazard for developing both GDM and PE with and without VD-ST

DISCUSSION

The current observational study provided multiple interesting findings; firstly out of the studied women, when getting pregnant, only 19.6% have average weight, 17.4% were insulin resistant and 58.5% had hypovitaminosis D (Hypo-VD). These figures point to the fact that high percentage of studied population had pre-pregnancy obesity and Hypo-VD and may be insulin resistant, so they could be mostly considered as having pre-pregnancy risk for committing pregnancy-induced or –associated morbidities that may affect pregnancy outcome.

The reported figure of Hypo-VD goes in hand with Loy *et al.*^[33] who reported VD inadequacy rate of 41.3%, and Al-Shaikh *et al.*^[34] who reported VD deficiency in 86.4% of pregnant females in Riyadh and GDM was the commonest complication detected in 11.1%. Moreover, Karras *et al.*^[35] out of literature systemic review found the prevalence of VD insufficiency among pregnant Greece women ranged from 9.3 to 41.4%, whereas that of VD deficiency from 22.7 to 90.3%.

Furthermore, Naseh *et al.*^[36] and Kilicaslan *et al.*^[37] detected VD deficiency rates of 27% and 53% and insufficiency rates of 73% and 47%, respectively, in their series of pregnant women. Moreover, Wierzejska *et al.*^[38] detected higher VD levels in maternal and umbilical cord blood in summer than in winter, but only 16% of studied pregnant women had the optimal VD level during summer. Recently, in 2018; Gustafsson *et al.*^[39] reported decreased 1,25(OH)₂D in 45% of pregnant women with significant difference than women with adequate vitamin D status and found no association of VD indices and parathyroid hormone changes during pregnancy.

Secondly, pregnancy itself is a diabetogenic process as evidenced by the increased levels of FBG in all patients, increased frequency of IR women and higher HOMA-IR score at the 3rd trimester compared to the 12th wk GA. Moreover, 68 women developed GDM throughout pregnancy course for a frequency of 13.8%. Moreover, throughout the duration of pregnancy, 71 women developed PE despite being normotensive at the 12th wk GA for a frequency of 14.4% and at the start of 3rd trimester, all patients showed increased blood pressure measures, irrespective of progressing to PE or not. Twenty-three women developed both GDM and PE with significantly lower incidence in women received VD-ST.

Thirdly, low 25OH-VD level showed positive significant correlation, while VD-ST showed negative significant correlation with a development of PE and GDM and with IR. These findings are in accordance with Lacroix *et al.*^[40] who detected higher risk of GDM in women had lower 1st trimester 25OHD levels and Lu *et al.*^[41] who documented that maternal vitamin D insufficiency is associated with increased risk of GDM. Moreover, Al-Shaikh *et al.*^[34] documented that GDM was the commonest complication encountered among pregnant women who had vitamin D deficiency for a frequency of 11.1% and De-Regil *et al.*^[42] found 7.5% and 6.5% of pregnant women developed PE and GDM, respectively. In line with detected correlations, Jafarzadeh *et al.*^[43], Dodds *et al.*^[44] and Triunfo *et al.*^[45] reported that in pregnant women, lower 25OHD concentrations showed an inverse association with maternal glycaemia and increased risk of GDM. Also, Naseh *et al.*^[46] found that maternal weight before delivery negatively correlated with maternal serum VD level.

The obtained data concerning VD-ST point to the role played by its deficiency or insufficiency in the

development of maternal pregnancy-induced or –associated complications and provide a guideline for its use as a pre-pregnancy prophylactic therapy. In line with the reported beneficial effect of VD-ST, in 2018; Roštami *et al.*^[47] found VD-ST improved serum 25(OH)D levels and decreased adverse pregnancy outcomes including PE, GDM and preterm delivery by 60, 50 and 40% than in women maintained without of VD-ST. Also, Shu and Huang^[48] detected a significant decrease in peripheral systolic and diastolic blood pressure in hypertensive patients after 8 weeks of VD supplementation. Moreover, Wilson *et al.* (2018) found that the risk of GDM was reduced by 53% with high (> 81 nmol/L) VD status when compared to moderate-high and Zhang *et al.*^[50] reported that that low blood VD level could increase the risk of GDM, and VD supplementation during pregnancy could ameliorate the condition of GDM.

VD-ST was provided as single daily dose of 1000 IU, this supplementation regimen is in accordance with Cooper *et al.*^[51] who found VD supplementation with 1000 IU/day during pregnancy is safe and sufficient to ensure replete VD in most of the pregnant women and with Yilmaz *et al.*^[52] who documented that VD supplementation by 1200 IU/day since the 12th gestational week can prevent neonatal VD deficiency.

CONCLUSION

The obtained results and review of literature allowed concluding that VD deficiency-insufficiency is a settled problem that requires national evaluation for predisposition and progress. Maternal hypovitaminosis D, obesity and insulin resistance constitute a triad that deleteriously affects pregnant women and is associated with development of GDM and/or PE. The proposed VD supplementation regimen effectively reduced the frequencies of pregnancy-associated or induced complications; so it is effective to break that triad. Wider scale studies are mandatory to evaluate the effect of pre-gestational VD-ST on oncoming pregnancy and the effect of injectable forms of VD that may be more convenient for patients.

CONFLICT OF INTEREST

There are no conflicts of interest.

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