Applied Medical Research, 2018
VOL 6, NO. 1, PAGE 1 – 8
DOI: doi.org/10.47363/AMR/2019(6)160

ORIGINAL RESEARCH

Open Access

Metabolic effects of vitamin D supplementation on electrolyte balance and pregnancy outcome of preeclamptic women in South West, Nigeria

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ABSTRACT

Objective: Preeclampsia (PE) remains a major cause of complications worldwide, especially in sub-Saharan African countries like Nigeria. Vitamin D seems to be a viable agent in the management of PE through its influence on electrolyte balance. This study aimed at assessing the effects of vitamin D supplementation on electrolytes of preeclamptic women and investigates the role of vitamin D in the improvement of pregnancy outcome.
Methods: Nearly, 150 women were recruited at 22 weeks gestation from antenatal clinics in Ibadan, Nigeria between year 2016 and 2018 and grouped into 50 normotensive pregnant women (Group A), 50 non supplemented preeclamptic women (Group B). Fifty preeclamptic women were given 1,000 IU/day of vitamin D3 for 8 weeks after recruitment at 22 weeks (Group C). Electrolytes, creatinine (Cr), and urea were quantified by the standard methods.
Results: The results obtained after supplementation with vitamin D showed a significant decrease ($p < 0.05$) in sodium, bicarbonate, and urea levels and a significant increase ($p < 0.05$) occurred in potassium, calcium, and magnesium in Group C at third trimester and postpartum when compared with the group B. No significant difference ($p > 0.05$) occurred in levels of chloride, phosphate, and creatinine in group C when compared with group B. There was no significant difference ($p > 0.05$) in gestational age at delivery, weight of fetus between the three groups, while a significant reduction ($p < 0.05$) occurred in the systolic and diastolic blood pressure and percentage of cesarean section in group C, when compared with group B.
Conclusion: Vitamin D supplementation is an important in correcting electrolyte imbalance associated with PE, thereby improving the pregnancy outcome.

ARTICLE HISTORY

Received January 08, 2019
Accepted January 23, 2019
Published February 02, 2019

KEYWORDS

Preeclampsia; vitamin D; supplementation; blood pressure; electrolytes

Introduction

Pre-eclampsia accounts for major cause of complications in pregnant women resulting in maternal morbidity and mortality, and affecting 3%–10% of all pregnancies worldwide [1]. It is usually characterized by susceptibility to pulmonary edema, coagulation defects, and renal failure [2] presence of systemic endothelial dysfunction and microangiopathy, in which the target organ may be the brain (seizures or eclampsia), the liver [the hemolysis, elevated liver function tests, and low platelet count (HELLP) syndrome], or the kidney (glomerular endotheliosis and proteinuria). Preeclampsia (PE) is also associated with increased frequency of caesarian section, preterm delivery, and abruptio placentae [3].

Electrolytes including calcium, sodium, and potassium may play an important role in the pathogenesis of hypertension because they contribute significantly to the normal functioning of the vascular smooth muscles, and therefore might have influence on the regulation of blood pressure.

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Magnesium has been known as an essential cofactor for many enzymes. It also plays an important role in nerve transmission and peripheral vasodilatation [4]. Several studies have associated hypocalemia to PE [5]. Calcium plays an important role in the function of the cardiac and vascular smooth muscles. It is known that the deficiency of calcium may lead to irritable nervous muscular symptoms, even tetanic convulsions, bleeding diathesis, capillary hemorrhages, tissue exudation and osteomalacia. Impaired renal function is associated with PE, and elevated blood urea, serum creatinine levels have been reported in some studies [6,7], while other few studies found no significant difference in creatinine in preeclamptic and normotensive pregnant women [8]. The cause of PE remains unknown but several studies have associated vitamin D insufficiency or deficiency to the risk of developing PE [9]. Vitamin D an important prohormone, possesses cardio-protective ability, and might influence endothelial and vascular smooth muscle cell function via the regulation of electrolytes and as well as influence the regulation of blood pressure [10,11].

Vitamin D supplementation can be very beneficial in prevention and management of PE. Systematic reviews [12,13] of controlled trials observed no association between vitamin D and prevention of PE, whereas other systematic reviews of observational studies reported an association [14,15]. These discrepancies in results could be due to confounding bias, and studies are few on this topic in Nigeria. Thus, a well-designed, prospective, and randomized trial of supplementation will be necessary to determine the effects of vitamin D supplementation on electrolyte balance and investigate the potential role of vitamin D in the reduction of blood pressure and improvement of pregnancy outcome.

Materials and Methods

Study design

This is a prospective, interventional study in which subjects were recruited from Antenatal Clinic in Obstetrics and Gynecology Department of University College Hospital and Ekiti State Teaching Hospital between the years 2016 and 2018. One hundred and fifty subjects were enrolled, 50 healthy pregnant women (Group A) and 50 nulliparous women with singleton pregnancies with PE (Group B) and 50 preeclamptic women were chosen randomly into Group C to be given vitamin D supplements for 8 weeks. They were between the ages of 18 and 35 years. Blood pressure and weight were measured and body mass index was calculated in each of the subjects at 22 weeks of pregnancy, using a standard analog sphygmomanometer, weighing balance, and meter rule, respectively. Participants were followed up till the end of their pregnancy, obstetric data were collected, which include: type of labor initiation, Apgar score, route of delivery, gestational age of baby, and weight.

Sample collection

Participants considered to have PE were subjects with elevated blood pressure, BP ≥ 140/90 mmHg twice and significant proteinuria of at least 300 mg in a 24-hour collection of urine, or a urine dipstick result of 1+ or greater, and physical manifestation of edema, according to the International Society for the Study of PE. Informed consent form was duly signed and questionnaires on sociodemographic information, medical history, health behavior, diets, and lifestyle were issued to the subjects. Blood and urine specimens were taken from each subjects starting from the 22nd week for the analysis of vitamin D and proteinuria, respectively, using urinalysis. Group C was placed on 1,000 IU of vitamin D3 for 8 weeks and blood samples were collected after supplementation and postpartumly. Blood samples were also collected from Group A and B at baseline (22 weeks), after 8 weeks and postpartumly. Nearly, 10 ml of venous blood were collected from each participant at 22 weeks, 30 weeks, and 3–7 days after delivery. Nearly, 5 ml of venous blood was dispensed into Plain bottle, blood was allowed to clot, centrifuged, and serum separated for analysis of vitamin D. Nearly, 5 ml were dispensed into Ethylenediaminetetraacetic acid bottle, centrifuged, and plasma separated for analysis of electrolytes, urea, and creatinine. Spot urine was collected for the assessment of protein using urine dipstick.

Biochemical assessment

Vitamin D was quantified by Enzyme Linked Immunosorbent Assay as described in Gran [16].

Serum vitamin D status was defined as (in ng/ml): deficient (<10 ng/ml), insufficient (10–30 ng/ml), sufficient (30 —100 ng/ml), according to Calbiotech Inc.25 (OH) vitamin D Elisa kit used for this analysis.

Ca^{2+} was done by colorimetric method on automated chemistry platform (LW C100 plus) as described in Burtis and Ashwood [17].

PO_4 was done by Molybdate method on automated chemistry platform (LW C100 plus) as described in Robert et al. [18].

Na^+, K^+, and **Cl^-,** was assessed using Ion Selective Electrode technique as described in Whelton [19].

HCO_3^- was quantified by using back titration technique as described in Faulkner [20].

Mg^{2+} was assessed by using Xylidyl blue method as described in Henry et al. [21].

Creatinine by using alkaline picrate method as described in Faulkner [20] on automated chemistry platform (LW C100 plus).

Urea by enzymatic colorimetric UV method as described in Tunstall-Pedoe [22] on automated chemistry platform (LW C100 plus).

Statistical analysis

The data obtained were grouped and expressed as mean ± standard deviation, and analyzed using the Statistical Package for Social Science version 16.0 by using one-way analysis of variance and Students *t*-test.

Results

In Table 1, the mean age of the normotensive and preeclamptic women were not statistically different ($p > 0.05$), while the systolic blood pressure, diastolic blood pressure, and BMI of the preeclamptic groups (Group B and C) were significantly higher than the normotensive group ($p < 0.05$).

In Table 2, there was no significant difference ($p > 0.05$) in the fetal weight among the three groups.

The % of women who undertook cesarean section was significantly higher in preeclamptic women who were not on supplementation when compared with control and preclamptic women on supplementation ($p < 0.05$).

The % of vagina delivery was significantly higher in control and in preeclamptic women on supplements when compared with preeclamptic women without supplements ($p < 0.05$).

Gestational age at labor was significantly reduced in the PE groups ($p < 0.05$). The % of preterm delivery was statistically higher ($p < 0.05$) in the PE groups when compared with control, while there was no statistical difference in the % of preterm delivery in preeclamptic women on supplements when compared with preeclamptic women without supplements ($p > 0.05$).

In Table 3, there was a significant increase in levels of Na, Cl, HCO_3, urea, and Cr in PE with and without supplementation (Group B and C) in second trimester when compared with control ($p < 0.05$), while the levels of K, Ca, Mg, and PO_4 reduced significantly in preeclamptic women with and without supplementation in second trimester when compared with control ($p < 0.05$).

In the non-supplemented PE group, there was no statistical difference in the increased levels of Na, Cl, PO_4, and Mg in second, third trimester, and postpartum ($p > 0.05$), while there was an increase in the level of HCO_3, Ca, and urea in third trimester and postpartum which was statistically different from second trimester($p < 0.05$). A significant reduction occurred in Cr and a decrease in K levels at postpartum when compared with second trimester in the non-supplemented PE group ($p < 0.05$).

After supplementation, there was a significant decrease in Na, HCO_3, and urea levels and a significant increase occurred in K, Ca, and Mg levels in Group C at third trimester and postpartum ($p < 0.05$). No significant difference occurred in levels of Cl, PO_4, and Cr in the supplemented group when compared with the non-supplemented PE group ($p > 0.05$).

In Figure 1, the frequency of cesarean section was higher in preeclamptic women when compared with control, while vagina delivery was more frequent in normotensive pregnant women. After vitamin D supplementation, the percentage of cesarean section decreased in preeclamptic women, while vaginal delivery increased significantly.

Table 1. Baseline characteristics of normotensive pregnant women (Group A), preeclamptic women without supplement (Group B), preeclamptic women on supplement (Group C).

Variables	Control ($n = 50$)	PE without Supplement ($n = 50$)	PE with Supplement ($n = 50$)
Age	33.1±2.4[a]	32.4±2.3[a]	32.9±3.1[a]
Systolic blood pressure	118±6.5[b]	150.8±12.5[c]	149.7±8.4[c]
Diastolic blood pressure	80.3±4.6[a]	91.9±13.2[b]	90.9±9.9[b]
Body mass index (BMI)	24.1±1.4[a]	31.2±3.2[b]	30.7±3.8[b]

Values of the same subscript within the same column are not statistically different at ($p > 0.05$) between the control and case group, while values with different subscripts are significantly different at ($p < 0.05$).

Table 2. Effects of vitamin D supplementation on pregnancy outcome.

Variables	Control (*n* = 50)	PE without Supplement (*n* = 50)	PE with Supplement (*n* = 50)
Fetal weight	2.85 ± 0.30[a]	2.88 ± 0.62[a]	2.91 ± 0.3[a]
Apgar score (1 minute)	8.47 ± 1.13[a]	7.8 ± 1.37[a]	7.7 ± 0.88[a]
Apgar score (5 minutes)	9.06 ± 0.70[a]	9.0 ± 1.25[a]	8.8 ± 0.94[a]
Gestation at labor (Wks)	38.13 ± 1.45[a]	36.2±1.93[b]	36.4±1.72[b]
Preterm labor % (*n*)	4(2)[a]	20 (10)[b]	16 (8)[b]
*Systolic blood pressure	120 ± 3.4[a]	152.0 ± 8.2[b]	135.3 ± 2.5[c]
*Diastolic blood pressure	78.2 ± 4.2[a]	92.5 ± 4.4[b]	87.9 ± 4.7[c]

*After supplementation at third trimester, blood pressure reading was taken in the three groups.

Table 3. Blood levels of vitamin D, electrolytes, urea, creatinine of normotensive pregnant women, preeclamptic women without supplement, preeclamptic women on supplement.

Variables		Control (*n* = 50)	PE without Supplement (*n* = 50)	PE With Supplement (*n* = 50)
Na (mmol/l)	Second trimester	130.3 ± 12.0[a]	143.3 ± 14.2[b]	144.1 ± 3.02[b]
	Third trimester	133.8 ± 7.0[a]	146.9 ± 14.4[b]	140.5 ± 8.9[c]
	Postpartum	135.3 ± 6.3[a]	146.8 ± 11.1[b]	139.3 ± 7.8[c]
K (mmol/l)	Second trimester	3.73 ± 0.53[a]	2.87 ± 0.49[b]	2.73 ± 0.51[b]
	Third trimester	4.02 ± 0.47[c]	2.83 ± 0.65[b]	3.35 ± 0.30[a]
	Postpartum	4.44 ± 0.49[c]	2.91 ± 0.73[d]	3.37 ± 0.24[a]
Cl (mmol/l)	Second trimester	102.3 ± 10.8[a]	106.2 ± 10.4[b]	114.0 ± 4.02[b]
	Third trimester	102.1 ± 6.8[a]	109.6 ± 7.3[b]	105.7 ± 5.79[b]
	Postpartum	105.4 ± 3.4[a]	111.8 ± 5.3[b]	106.1 ± 3.19[b]
HCO_3 (mmol/l)	Second trimester	18.6 ± 4.03[a]	25.15 ± 3.3[b]	27.8 ± 1.0[b]
	Third trimester	17.2 ± 2.85[a]	29.55 ± 3.11[c]	16.55 ± 1.76[a]
	Postpartum	17.3 ± 3.01[a]	31.3 ± 3.36[c]	15.8 ± 2.33[a]
Ca (mmol/l)	Second trimester	2.82 ± 0.63[a]	1.41 ± 0.37[b]	1.85 ± 0.19[b]
	Third trimester	2.35 ± 0.59[b]	1.43 ± 0.34[a]	2.15 ± 0.15[c]
	Postpartum	2.33 ± 0.43[b]	1.43 ± 0.35[a]	2.16 ± 0.16[c]
PO_4 (mmol/l)	Second trimester	0.89 ± 0.71[a]	0.63 ± 0.60[b]	0.67 ± 0.09[b]
	Third trimester	0.72 ± 0.12[b]	0.69 ± 0.09[b]	0.71 ± 0.24[b]
	Postpartum	0.73 ± 0.17[b]	0.63 ± 0.13[b]	0.82 ± 0.47[b]
Urea (mmol/l)	Second trimester	1.76 ± 0.54[a]	2.69 ± 0.30[b]	2.75 ± 0.62[b]
	Third trimester	1.63 ± 0.48[a]	2.95 ± 0.65[c]	1.97 ± 0.84[d]
	Postpartum	1.85 ± 0.17[a]	2.72 ± 0.83[c]	1.89 ± 0.87[d]
Cr (µmol/l)	Second trimester	43.05 ± 11.1[a]	64.15 ± 28.1[b]	69.8 ± 27.2[b]
	Third trimester	42.8 ± 8.87[a]	61.35 ± 31.16[b]	68.0 ± 21.26[b]
	Postpartum	46.15 ± 12.06[a]	54.8 ± 32.35[c]	57.6 ± 15.07[b]
Mg (mmol/l)	Second trimester	0.84 ± 0.11[a]	0.62 ± 0.23[b]	0.65 ± 0.08[b]
	Third trimester	0.78 ± 0.27[a]	0.64 ± 0.25[b]	0.75±0.05[c]
	Postpartum	0.80 ± 0.49[a]	0.70 ± 0.08[b]	0.79±0.32[c]

Values of the same subscript within the same column are not statistically different at ($p > 0.05$) between the control and case group, while values with different subscripts are significantly different at ($p < 0.05$). Na: Sodium, K: Potassium, Cl: Chloride, HCO_3: Bicarbonate, Cr: Creatinine, PO_4: Phosphate, MDA: Malonaldehyde, SOD: Superoxide Dismutase.

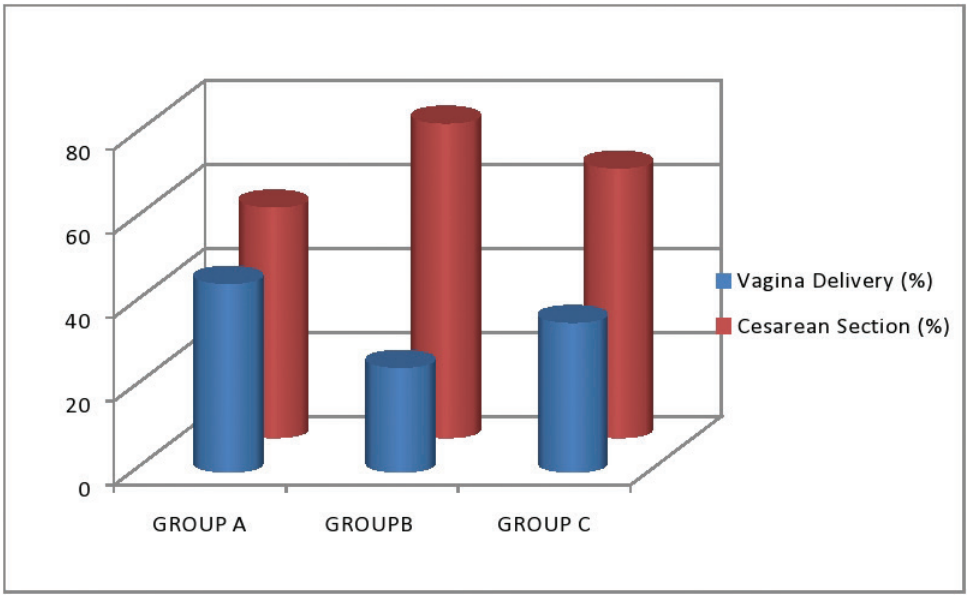

Figure 1. Graphical representation of the percentage of women who had vaginal delivery and cesarean section in normotensive pregnant women (Group A), preeclamptic women without supplement (Group B), preeclamptic women on supplement (Group C).

Discussion

The significant increase in levels of Na, Cl, HCO_3, Urea, and Cr and reduction in the levels of K, Ca, and PO_4 in the PE group observed in this present study shows that PE is associated with electrolyte imbalance.

The decrease in potassium levels in the PE group reported in this study might be due to its influence on BP. Though mechanism explaining the relationship between reduced serum potassium levels and low blood pressure control is poorly understood, studies have shown that reduced levels of serum potassium enhance vascular responsiveness to vasopressors such as norepinephrine which might indirectly lead to vasoconstriction and increased platelet aggregation which increases blood pressure [23].

Infact, observational studies indicate that increasing potassium by 750–1,000 mg/day can lower BP by 2 or 3 mm Hg [24]. Vitamin D supplementation in this study was able to increase potassium levels which may be an important yardstick for reducing elevated blood pressure in PE. Proposed mechanisms by which potassium can influence BP include natriuresis, reduced vasoconstrictive sensitivity to norepinephrine and angiotensin II, increased sodium/potassium AT Pase activity, and proliferation in vascular smooth muscle, and sympathetic nervous system cells [23].

A significant increase in sodium levels was observed in the PE group in this study, similar to the result of Abdellah and Abdrabo [25]. This increase in sodium levels might be due to the decrease in intravascular circulating volume found in preeclamptics. Increased blood sodium levels lead to sodium and water retention, the expansion of ECF and intravascular volume, increased venous return, and an elevated cardiac index. Whole body autoregulation takes place as blood continues to flow to the tissue, thereby increasing the total peripheral resistance resulting in high blood pressure [25]. The "peripheral arterial vasodilation hypothesis" of sodium and water retention in the pathogenesis of PE states that with increased endothelial damage, sodium retention, and increased sensitivity to angiotensin lead to hypertension, edema, and proteinuria, the diagnostic triad of PE [26]. However, after supplementation with vitamin D in this present study, a marked reduction in Na levels in the PE group was observed.

Hypocalcemia was observed in PE group in this study which is in accordance with Li et al. [27]. Calcium works in combination with other ions such as sodium, potassium, and magnesium to provide an ionic balance to the vascular membrane, vasodilatation, and consequently resulting in reduced BP. Low calcium levels may lead to hypertension by stimulating either the parathyroid hormone (PTH) or renin release, thereby increasing intracellular calcium which triggers vascular smooth muscle constriction and increase blood pressure [27]. Calcium absorption has been found to be positively

associated with serum 1,25(OH)2D concentrations in late pregnancy. However, there was a significant increase in calcium levels after oral intake of 25OH vitamin D in preeclamptic women in this study, which means that, there might be a possible reversal of vascular constriction which might ultimately result in decrease in blood pressure. Increased levels of chloride seen in the PE group may result in increased osmolality leading to suppressed dilatation of vessels [28].

Low levels of magnesium as seen in PE in this study, is similar to the work of O'Brien et al. [29]. This could be due to decreased dietary intake, increased clearance by the kidneys, hemodilution due to the expansion of the extracellular space and increased consumption of minerals by the growing foetus [29]. Magnesium affects the cardiac and smooth muscle cells by altering the transport of calcium and its binding to the membrane and organ cells. Magnesium can also act peripherally to produce peripheral vasodilatation and a fall in blood pressure. Low levels predispose to an increase in the arterial pressure [26]. However, a slight increase in magnesium was observed after vitamin D supplementation in this present study. Magnesium helps in inhibiting phosphatidyl inositol-4, 5-bisphosphate specific phospholipase C activity and subsequent calcium release in the cells, thereby decreasing intracellular calcium levels which causes partial membrane depolarization and decreased repolarization along with opening of Ca^{2+} membrane channels. This action is important in the reduction of blood pressure [30].

Increased levels of urea and creatinine observed in this study, is associated with renal dysfunction which occurs in PE as a result of hemodynamic changes, glomerular lesions, and podocyte damage [31,32]. However, vitamin D supplementation only decreased the levels of urea while creatinine was not affected.

Mechanism by which vitamin D correct the electrolyte imbalance in PE as observed in the results of this present study is unclear, but suggested mechanism is via the renin angiotensin aldosterone system (RAAS). RAAS is a main regulator of blood pressure and plays a critical role in the regulation of volume and electrolyte homeostasis. Increased activation of RAAS is associated with hypertension, and vitamin D is a potent inhibitor of renin synthesis [33]. Vitamin D helps to maintain extracellular calcium ion levels in the body by controlling the amount of calcium that is absorbed from the small intestine [34]. Increased vitamin D levels suppress parathyroid hormone (PTH) which mobilizes the calcium from stores, specifically the bones, to increase the amount of calcium in blood [35]. Even though there are compensation mechanisms throughout the body, adequate amounts of vitamin D are necessary for the proper absorption and regulation of calcium. Vitamin D can stimulate intestinal magnesium absorption and also play a key role in the intestinal absorption of phosphate and magnesium [29].

A slight reduction in systolic and diastolic blood pressure of the PE group observed after supplementation at the third trimester in this present study might be due to the cumulative effect of vitamin D on apolipoprotein gene expression [36], PTH suppression, electrolytes, RAS, and decreased production of lipid peroxidation products [37]. Insufficient vitamin D levels has been shown to be associated with higher cesarean section delivery in PE [38], however, after supplementation in this present study, there was a reduction in the number of cesarean section in the preeclamptic group. The reduction in number of cesarean section might be as a result of the effect of vitamin D supplementation on pregnancy hormones (Prostagladins) which are very important in contraction of the muscle of the uterus during labor. Reports are conflicting on the role of vitamin D and the risk of preterm birth. Premature amniotic membrane rupture and preterm delivery have been associated with vitamin D deficiency and inflammatory response [39]. This present study showed no significant effect of vitamin D supplementation on birth weight and Apgar score of fetus. In any case, fetal weight is a complex process dependent on many factors, including genetic background, birth interval, trophoblast implantation, placental development, nutrition, and physical activity [40]. In our study, birth weight was not significantly different in the two PE groups and this result is similar to Kovacs *et al*. study that found no significant difference in offspring birth length in UK Asian women that supplemented with 1,000 IU ergocalciferol daily at third trimester compared with the control group [41]. In this study, 1- and 5-minute Apgar scores showed no significant differences between the two PE groups, but Hossain *et al*. demonstrated 1 -and 5-minute Apgar scores were significantly higher in the supplemented group compared to control [42]. The difference of results may be due to heterogeneity of method of studies, population, time and dose of supplementation during pregnancy.

Vitamin D is very important in the reduction of high blood pressure. It also help in regulating electrolyte homeostasis thereby improving pregnancy outcome in PE. Vitamin D should be added to antenatal supplements in pregnant women and taken routinely because of its potential role in the prevention of PE. In the treatment and management of PE, vitamin D should be administered routinely.

Further Study

Further research with larger sample sizes that will establish the relationship between maternal vitamin D levels and pregnancy outcomes is necessary, and also studies that would establish the relationship between vitamin D and angiogenic proteins in PE is important.

Acknowledgments

The authors acknowledge the contribution of the staff of the Department of Biochemistry and Science Laboratory Technology of Ekiti State University. Ado Ekiti, Ekiti State, Nigeria.

Competing interests

The authors declare no competing interests.

Ethical approval

All authors hereby declare that ethical clearance was given by Joint Ethical Committee of the College of Medicine and the University College Hospital Ibadan, and Oyo State Ethical committee, Nigeria. All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards.

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