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Vitamin D in Term Newborns: Relation with Maternal Concentrations and Birth Weight

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Vitamin D · Newborn · Pregnant women · Postpartum period

Abstract
Objective: To evaluate vitamin D serum levels of term newborns and relate them to maternal concentrations and birth weight. ***Methods:*** Cross-sectional study carried out with 225 mothers and their term newborns. Data collected were maternal health, prenatal care, gestational, and anthropometric data of the newborns. The following laboratory tests were performed: serum levels of 25(OH)D, calcium, phosphorus, magnesium, and alkaline phosphatase. ***Results:*** Of the 225 newborns included in the study, 119 (52.9%) were males, the mean birth weight was 3,198 ± 421.4 g, and the gestational age was 39.1 ± 1.1 weeks. Of these, 20 (8.9%) were small and 12 (5.3%) were large for gestational age. A 25(OH)D sufficiency was found in 25.8% of mothers and 92% of newborns. The mean 25(OH)D concentrations of newborns was higher than that of the mothers 48.7 ± 15.2 ng/mL vs. 26.0 ± 6.7 ng/dL ($p < 0.001$), correlating inversely with birth weight ($r = -0.249$; $p < 0.001$). Small for gestational age (SGA) newborns had higher concentrations of 25(OH)D compared to adequate and large for age ($p < 0.001$). ***Conclusion:*** In conclusion, this study showed strong positive correlation between maternal and neonatal 25(OH)D concentrations, with higher values in newborns. The highest 25(OH)D concentrations were found in SGA term infants. We speculated these findings could be influenced by newborn body composition.

Introduction

Vitamin D is identified as a micronutrient and is now also recognized as a prohormone involved in bone metabolism and the functioning of the immune, respiratory, endocrine, and cardiovascular systems [1, 2].

The maintenance of its blood concentrations depends predominantly on the endogenous production from skin sun exposure and, to a lesser extent, dietary intake. Ethnicity (skin pigmentation), latitude, season, use of sunscreen, and body mass index also influence vitamin D serum levels (25[OH]D) [2].

Appropriate concentrations of 25(OH)D during pregnancy influence the development and maturation of fetal tissues and systems through their action on gene expres-

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sion. Vitamin D deficiency at this stage can lead to complications for women, such as gestational diabetes and preeclampsia; and fetuses, such as intrauterine growth restriction, prematurity, insulin resistance, and regulatory T-cell impairment [3, 4].

The transfer of 25(OH)D to the fetus occurs primarily through the placenta and is more intense in the last trimester of gestation, since the placenta's receptors produce the enzyme that converts 25(OH)D into $1,25(OH)_2D$, its active form. There is a strong correlation between maternal and cord blood 25(OH)D concentrations [4].

In a recent meta-analysis, the authors found association of daily medical supplementation of around 2,000 IU vitamin D during pregnancy with a lower risk to small infants for gestational age and better postnatal growth of these children [5].

Vitamin D deficiency has reportedly had a high global prevalence among pregnant women and their newborns. A systematic review by Saraf et al. [6] showed an overall prevalence of 25(OH)D concentrations <20 ng/mL of 54 and 75% for mothers and their newborns, respectively.

The World Health Organization [7] and Brazil do not recommend universal supplementation of vitamin D during pregnancy. Nevertheless, studies in our country suggest a high percentage of deficiency/insufficiency [8–10].

Given the importance of vitamin D during pregnancy, the repercussions of its deficiency on maternal and infant health, and because Brazil is a sunny country that does not recommend universal supplementation, we decided to conduct this study which aims to describe 25(OH)D serum levels of term newborns and relate them to maternal concentrations and birth weight.

Materials and Methods

A cross-sectional study was performed between October and December 2016 in a maternity hospital of Crato, State of Ceará (latitude 07° 14 '03 "S). The Human Research Ethics Committee of the ABC Faculty of Medicine, Santo André approved the study under number 1.813.560. A recently published study with the same sample described maternal 25 (OH)D concentrations and associated nutritional status with the presence of diseases during gestation [11].

The sample included, consecutively, 225 term newborns and their mothers. Newborns who were sent to the neonatal intensive care unit, with severe malformations, genetic syndromes, and neonatal hypoxia were excluded.

Mothers were submitted to a standardized questionnaire with information on socioeconomic conditions, skin color (self-reported), sun exposure, photoprotection, diseases during pregnancy, and use of vitamin/mineral supplements. The prenatal card was checked for complete information about laboratory tests and anthropometric measurements.

Weight and height data were used to calculate the pregestational body mass index (BMI, kg/m^2), which was classified as underweight (<18.5 kg/m^2), normal (18.5–24.9 kg/m^2), overweight (25–29.9 kg/m^2), and obesity (≥30 kg/m^2). Weight gain during pregnancy was calculated and classified as low, adequate, and high [12].

Information on weight, length, and head circumference of the newborns were collected within the first 24 h after delivery. The gestational age was calculated according to the date of the last menstruation; alternatively, we employed the first-trimester ultrasonography data and, finally, the newborn's clinical evaluation [13]. The newborns were classified by gestational age, using INTERGROWTH-21st [14].

Blood samples were taken within 24 h after delivery. We collected 5 mL of blood through peripheral venipuncture of the mothers and their newborns, which were photoprotected, refrigerated, and send to the local clinical laboratory, where they were centrifuged and serum levels of 25(OH)D (25[OH]D2 and 25[OH]D3; Electrochemiluminescence, with UniCel DXI 800 Immunoassay System, Beckman Coulter®), Calcium, Phosphorus, Magnesium, and Alkaline Phosphatase (Reflectance Spectrophotometry, Vitros 5600 Integrated System, Ortho-Clinical Diagnostics, Raritan, NJ, USA) were dosed. The 25(OH)D serum concentrations <20 ng/mL were defined as deficiency; 20–30 ng/mL, as insufficiency; and >30 ng/mL, as sufficiency [13]. All calculations followed the best clinical practice. The mean of 19.9 ng/mL (SD 0.948 ng/mL and an intra-control CV of 4.8%) was the referred reproducible value to measure the 25(OH)D. For the same parameter, the intermediate precision was 19.9 ng/mL (SD 1.23 ng/mL and an intra-control CV of 6.2%). The lowest detectable value (sensitivity) of the method was 1.25 ng/dL.

Statistical Analysis

A spreadsheet was created in Excel®, containing information on identification, general characteristics, data from the questionnaires on mother and newborns, anthropometric data, and laboratory test results. The spreadsheet was revised, consolidated, and imported into the Statistical Package for Social Science, version 25.0 (IBM®) for statistical analyses. The categorical variables were shown as absolute number and percentage, compared using the chi-square test; the continuums were tested for their normality with Shapiro-Wilk test, showed as mean (SE) and compared by Student *t* test. Pearson's correlation was used to assess the correlation between maternal, newborn, and birth weight 25(OH)D serum levels. Statistical significance was set at $p < 0.05$.

Based on data from a previous study similar to this [15], the number of included newborns has the power to detect a difference of 3 ng/mL of 25(OH)D, with a CI of 95%.

Results

Of the 225 newborns included in the study, 119 (52.9%) were males, with a mean birth weight of 3,198 ± 421.4 g (variation: 2,030–4,115 g) and gestational age of 39.1 ± 1.1

Table 1. General characteristics and serum levels of 25(OH)D, calcium, phosphorus, magnesium, and alkaline phosphatase of newborns (n = 225)

Variable	n (%) or mean ± SD
Gender	
Female	106 (47.1)
Male	119 (52.9)
Birth weight, g	3,198±421.4
Length at birth, cm	48.1±1.9
Classification	
Low weight	11 (4.9)
Adequate weight	208 (92.4)
High weight	6 (2.7)
Head circumference, cm	34.4±1.3
Gestational age, weeks	39.1±1.1
Classification	
Small	20 (8.9)
Adequate	193 (85.8)
Large	12 (5.3)
Vitamin D metabolism	
25(OH)D	48.7±15.2
<20 ng/mL	3 (1.3)
20–30 ng/mL	15 (6.7)
>30 ng/mL	207 (92.0)
Calcium, mg/dL	9.2±0.8
Phosphorus, mg/dL	7.7±1.8
Magnesium, mg/dL	1.9±0.5
Alkaline phosphatase, U/L	155.2±44.7

week (37–41 weeks). Of these, 20 (8.9%) were classified as small (small for gestational age [SGA]) and 12 (5.3%) as large (large for gestational age [LGA]) for gestational age (Table 1).

The mean age of the mothers was 25.6 ± 6.6 years (variation: 15.3–44.3 years). There was a predominance of dark skin color 178 (79.1%), low education (<4 years) 197 (87.6%), and work in the urban region 127 (56.4%). Most of the women underwent prenatal examination, of which 108 (48.0%) were primiparous. The main complication observed during gestation was urinary tract infection (72, 32%), followed by specific gestational hypertension and gestational diabetes mellitus (Table 2), with smaller rates. There was a predominance of surgical deliveries (164, 72.9%).

Regular sun exposure, use of photoprotection and multivitamin supplement containing vitamin D since of beginning of gestation (cholecalciferol, 250–400 UL/day) were found in 144 (64.0%), 44 (19.6%), and 20 (8.9%) of mothers evaluated, respectively.

Pregestational BMI was 19 (8.5%) for underweight, 49 (21.9%) for overweight, and 28 (12.0%) for obesity. Weight gain during pregnancy was lower, adequate and higher than recommended in 83 (36.9%), 70 (31.1%), and 71 (31.6%) of the women, respectively (Table 2).

A sufficiency of 25(OH)D (>30 ng/mL) was observed in 58 (25.8%) and 207 (92.0%) of mothers and newborns, respectively (Table 1). The mean of the 25(OH)D serum levels in newborns was higher than that of the mothers: 48.7 ± 15.2 ng/mL (16.3–100.4 ng/mL) vs. 26.0 ± 6.7 ng/dL (6.8–52.5 ng/mL), and the mean difference was –22.7 ± 11.5 ng/mL ($p < 0.001$).

A positive and statistically significant correlation was found between the 25(OH)D serum levels of the newborns and their mothers ($r = 0.697$; $p < 0.001$; Fig. 1). Calcium ($r = 0.093$, $p = 0.164$), phosphorus ($r = –0.017$; $p = 0.796$), magnesium ($r = 0.108$; $p = 0.107$), and alkaline phosphatase ($r = 0.068$; $p = 0.308$) concentrations were not associated with those of 25(OH)D serum levels in newborns.

The 25(OH)D serum levels of the newborns were inversely correlated with birth weight ($r = –0.249$; $p < 0.001$; Fig. 2). Newborns SGA showed higher concentrations of 25(OH)D and alkaline phosphatase compared to the adequate for gestational ages and LGAs (Fig. 3).

Discussion

In this study, we observed that serum levels of 25(OH)D in term newborns were higher and correlated directly with maternal ones. We also observed that SGA newborns have higher levels of 25(OH)D compared to adequate and LGA.

Sun exposure and intake of supplements containing vitamin D are important for 25(OH)D concentrations maintenance in pregnant women and infants. Cutaneous vitamin D synthesis can be impaired depending on small areas of exposed skin in the presence of air pollution and during winter [16] even in tropical countries that are predominantly sunny throughout the year.

Scientific Societies consider concentrations of 25(OH)D higher than 30 ng/mL as baseline sufficiency values during pregnancy [15]. Despite increasing evidence that low concentrations of 25(OH)D in pregnant women are associated with maternal and neonatal complications, such as specific gestational hypertension, gestational diabetes, bacterial vaginosis, prematurity, and restricted intrauterine growth. The World Health Organization does not recommend vitamin D supplementation for all pregnant women. This is justified because vitamin D metabolism has different and that are not yet wholly clarified and

Table 2. Characterization of puerperae included in the study (n = 225)

Variable	n (%) or mean ± SD
Maternal age, years	25.6±6.6
Paternal age, years	28.9±7.9
Ethnicity	
White skin	47 (20.9)
Dark skin	178 (79.1)
Maternal work	
Housewife	55 (24.4)
Urban	127 (56.4)
Rural	43 (19.1)
Maternal schooling, full years of study	
<4 years	197 (87.6)
4–8 years	13 (5.8)
>8 years	15 (6.7)
People living in the household	4.6±2.2
Number of gestations	2.1±1.6
Primiparous, n (%)	108 (48.0)
Prenatal care visits	7.8±2.1
Pregestational BMI, kg/m^2	
<18.5	19 (8.5)
18.5–25	128 (57.1)
25–30	49 (21.9)
≥30	28 (12.5)
Weight gain during pregnancy	
Adequate	83 (36.9)
Low	70 (31.1)
High	71 (31.6)
Tobacco use	9 (4.0)
Alcohol use	11 (4.9)
Multivitamin with vitamin D (cholecalciferol)	20 (8.9)
Folic acid	198 (88.0)
Iron	204 (90.7)
Regular sun protection	44 (19.6)
Regular sun exposure	144 (64.0)
Hours/days regular sun exposure	1.9±1.0
Diseases during pregnancy	
Urinary infection	72 (32)
Gestational-specific hypertensive disease	20 (8.9)
Gestational diabetes mellitus	2 (0.9)
Bleeding	17 (7.6)
Type of delivery	
Vaginal	61 (27.1)
Surgical (caesarean)	164 (72.9)
25(OH)D concentrations, ng/mL	26.0±6.8
25(OH)D concentrations, ng/mL	
<20	43 (19.1)
20–30 ng/mL	124 (55.1
>30 ng/mL	58 (25.8)

BMI, body mass index; PIH, pregnancy-induced hypertension; GDM, gestational diabetes mellitus.

because further studies are required on the potential adverse events of supplementation, depending on the recommended dose, for both mother and child in the long term [17].

Although our study was carried out with healthy pregnant women from a sunny country, only a quarter of the mothers had sufficient levels of 25(OH)D. In turn, <10% of newborns showed values lower than 30 ng/mL. Serum levels of 25(OH)D of the newborns were higher than those of their mothers. These results are similar to those found by Pena et al. [8] in a study performed with pregnant women with preeclampsia in a Brazilian city with characteristics similar to the one we studied (Recife, Brazil), and in a previous study on the same group, with preterm infants (São Bernardo do Campo, Brazil) [18].

Study conducted in India [19] also found a high percentage of 25(OH)D insufficiency/deficiency in pregnant women and direct correlation between maternal and newborn serum levels. However, the authors observed a higher percentage of insufficiency/deficiency in the newborn compared to the mothers. The high percentage of pregnant women with associated diseases (26% with preeclampsia) and umbilical cord blood collection are some of the factors that may justify the divergent results of the 25(OH)D levels of our study compared to the Indian study.

Vitamin D metabolism in pregnant and lactating women is dynamic and influenced by several factors, among which we highlight the single nucleotide polymorphisms of vitamin D receptor gene (VDR) [20] and vitamin D epimers [21]. Italian study showed association between some maternal VDR polymorphisms with neonatal anthropometric measures and the risk of prematurity [22]. The authors did not evaluate the combined effect of 25(OH)D serum levels with VDR polymorphisms.

Vitamin D epimers such as 3-epi 25(OH)D3 and 3-epi-1,25α(OH)2D3 have increased production during gestation and in infants in the first 3 months of life [23, 24]. While the biological functions of these epimers are not precisely known, recent studies have described that they have affinity for 25(OH)D and 1,25 (OH) D receptors and may inhibit PTH production [24]. The 3-epi-25-hydroxycholecalciferol represents 14 and 25% of total concentrations of 25 (OH)D in the umbilical cord and newborns, respectively [25]. Commercial 25(OH)D assays laboratory methods lack the sensitivity to detect these epimers, which may interfere with the interpretation of the results in pregnant women and infants.

Other vitamin D metabolism markers such as calcium, phosphorus, magnesium, and alkaline phosphatase were

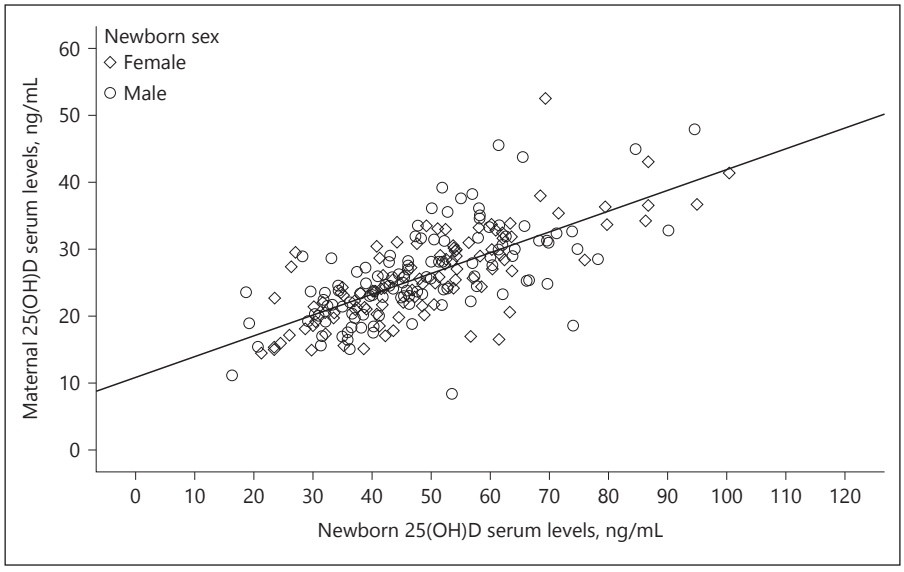

Fig. 1. Correlation of maternal and neonatal 25(OH)D (ng/mL) serum levels. Pearson's correlation significance level: all (r = 0.697; $p < 0.001$); males (r = 0.653; $p < 0.001$); females (r = 0.746; $p < 0.001$).

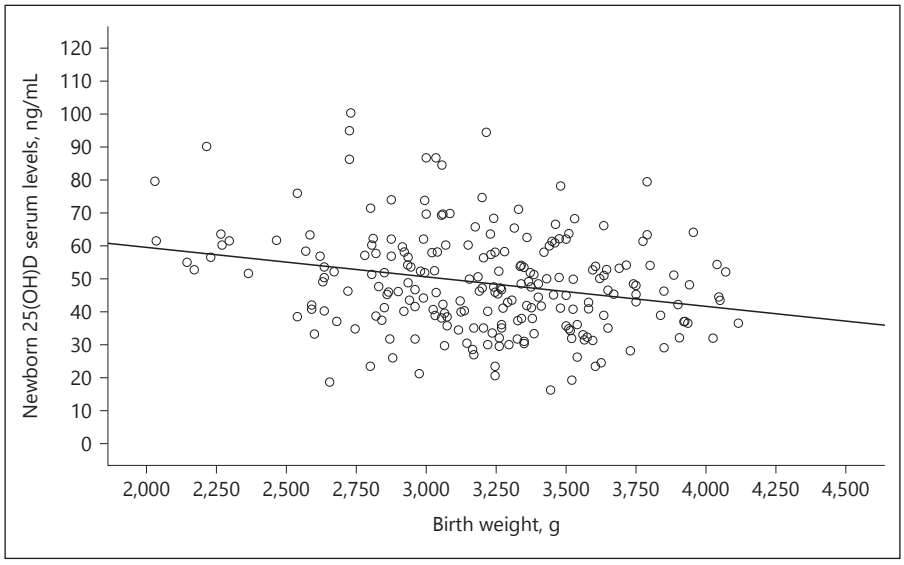

Fig. 2. Correlation of newborn 25(OH)D (ng/mL) serum levels with birth weight. Pearson correlation; $r = -0.249$; $p < 0.001$.

not associated with 25(OH)D concentrations in newborns in this study. Hypocalcemia is the first change observed in newborns with inadequate levels of 25(OH)D; however, they usually occur when concentrations are below 10 ng/mL [15]. Only 2 mothers had values smaller than this and the calcium concentrations in their newborns were normal [26].

Previous studies have shown that lower concentrations of 25(OH)D at the onset and middle of gestation are inversely associated with birth weight and that insufficiency/deficiency increases the risk of restricted intrauterine growth and newborns SGA [5, 27, 28]. Sarma et al. [29] found that maternal hypovitaminosis D is associated with shorter femur length at 34 weeks of gestation and lower birth length. However, no significant association was noted between maternal Vitamin D serum levels with birth weight and head circumference of newborns [29].

In this study, 25(OH)D concentrations of the newborns were inversely correlated with birth weight. In turn, the SGA had higher values of 25(OH)D compared to the others. One of the plausible explanations for this seemingly paradoxical finding may be attributed to the percentage of body fat influencing circulating 25(OH)D

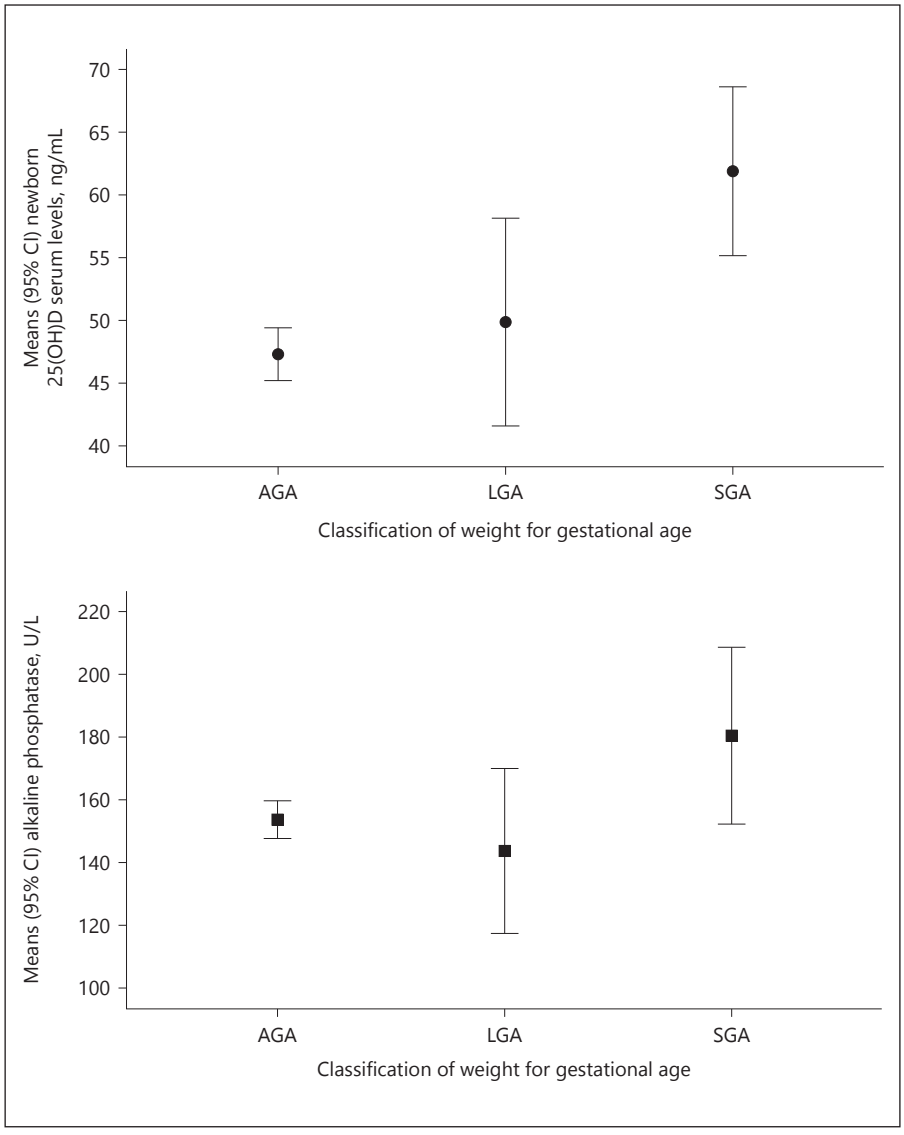

Fig. 3. Concentrations of 25(OH)D and alkaline phosphatase in newborns, according to the classification of weight for gestational age. Analysis of variance for 25 (OH)D ($p < 0.001$); SGA vs. AGA ($p < 0.001$); SGA vs. LGA ($p = 0.066$); AGA vs. LGA ($p = 0.830$). Analysis of variance for alkaline phosphatase ($p < 0.001$); SGA vs. AGA ($p < 0.001$); SGA vs. LGA ($p = 0.063$); AGA vs. LGA ($p = 0.730$). AGA, adequate for gestational age; SGA, small for gestational age; LGA, large for gestational age.

levels or vice versa [30, 31]. Newborns large and SGA have, respectively, a higher and lower percentage of body fat, which may interfere with the levels of 25(OH)D.

Recent studies suggest that vitamin D deficiency in early pregnancy influences the body composition of newborns and postnatal growth [32, 33]. On the other hand, Czech-Kowalska et al. [34] did not find relationship between lower maternal 25(OH)D serum levels with higher fat mass in their newborns. Depending on gestational age that the serum levels of 25(OH)D are evaluated, the results regarding anthropometry and body composition of the newborns could be different.

This study has some limitations, such as the cross-sectional design with a single 25(OH)D serum levels evaluation, a risk of memory bias related to maternal habits, photoprotection, sun exposure, and vitamin supplementation.

In conclusion, this study showed strong positive correlation between maternal and neonatal 25(OH)D concentrations with higher values in newborns. The highest 25(OH)D concentrations were found in SGA term infants. We speculated these findings could be influenced by newborn body composition.

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Statement of Ethics

The study protocol was approved by The Human Research Ethics Committee of the ABC Faculty of Medicine, Santo André under opinion Nº 1.813.560. Subjects (parents or guardians) have given their written informed consent.

Disclosure Statement

The authors declare no conflicts of interest.

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Author Contributions

C.U.P.E., M.E.P.M., F.I.S.-S., and R.O.S.S.: participated in the study design; C.U.P.E., M.E.P.M., E.E.M., J.L.A.L., J.L.S.R., J.G., and C.M.N.: participated in the collection and analysis of samples and the interpretation of outcomes. All authors participated in the construction of the manuscript and approved the final version to be submitted for publication.

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