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Maternal 25-Hydroxyvitamin D and Preterm Birth in Twin Gestations

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Abstract

OBJECTIVE—To assess whether there was an independent association between maternal 25-hydroxyvitamin D concentrations at 24–28 weeks of gestation and preterm birth in a multicenter U.S. cohort of twin pregnancies.

METHODS—Serum samples from mothers who participated in a clinical trial of 17 α-hydroxyprogesterone caproate for the prevention of preterm birth in twin gestations (2004–2006) were assayed for 25-hydroxyvitamin D using liquid-chromatography-tandem mass spectrometry (n=211). Gestational age was determined early in pregnancy using a rigorous algorithm. Preterm birth was defined as delivery of the first twin or death of either twin at less than 35 weeks of gestation.

RESULTS—The mean (standard deviation) serum 25-hydroxyvitamin D was 82.7(31.5) nmol/L; 40.3% of women had 25-hydroxyvitamin Dless than 75 nmol/L. Preterm birth less than 35 weeks occurred in 49.4% of mothers with 25-hydroxyvitamin D less than 75 nmol/L compared with

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26.2% among those with 25-hydroxyvitamin D 75 nmol/L or more (P<.001). After adjustment for maternal race and ethnicity, study site, parity, prepregnancy body mass index, season, marital status, education, gestational age at blood sampling, smoking status and 17 α-hydroxyprogesterone caproate treatment, maternal 25-hydroxyvitamin D 75 nmol/L or more was associated with a 60% reduction in the odds of preterm birth compared with less than 75 nmol/L (adjusted odds ratio[OR] 0.4, 95% confidence interval [CI] 0.2–0.8). A similar protective association was observed when studying preterm birth less than 32 weeks (OR 0.2, 95% CI 0.1–0.6) and after confounder adjustment.

CONCLUSIONS—Late second trimester maternal 25-hydroxyvitamin D less than 75 nmol/L is associated with an increase in the risk of preterm birth in this cohort of twin pregnancies.

Introduction

The health-promoting role of vitamin D during pregnancy is contentiously debated (1-4). Optimal maternal 25-hydroxyvitamin D concentrations are not known. Nearly one in three pregnant women in the United States has serum 25-hydroxyvitamin D less than 50 nmol/L (5), which experts agree increases the risks of bone-related disease for mothers and fetuses (3,6). Approximately two in three pregnant women have serum 25-hydroxyvitamin D less than 75 nmol/L (5), concentrations associated with cancer, diabetes, cardiovascular disease, autoimmune disorders, and other adverse health outcomes in nonpregnant adults (7,8).

It remains unclear whether vitamin D deficiency poses risks for nonskeletal outcomes during pregnancy (9,10). The ability of maternal decidual cells to convert 25-hydroxyvitamin D to 1,25-dihydroxyvitamin D, the hormonally active form of the vitamin, as well as the presence of vitamin D receptors on the placenta (11) highlight a potentially important role for vitamin D in pregnancy outcomes, including spontaneous preterm birth. 1,25-dihydroxyvitamin D has immunomodulatory and antiinflammatory properties and regulates key genes for successful implantation (12), yet few research studies have explored the relation between 25-hydroxyvitamin D and preterm birth (13-15), and none have examined this association in twin pregnancies. Mothers carrying twins have greater nutrient demands (16) and dramatically higher rates of preterm birth than singleton gestations (17). Research into the role of maternal vitamin D deficiency, a common, modifiable factor, in poor outcomes in twin gestations may help improve maternal and child health in these vulnerable pregnancies.

Our objective was to assess the association between maternal 25-hydroxyvitamin D concentrations at 24–28 weeks of gestation and preterm birth in a multicenter U.S. cohort of twin pregnancies.

Materials and Methods

This is a secondary analysis of data and samples from a randomized, double-blinded, placebo-controlled trial of 17 α-hydroxyprogesterone caproate for the prevention of preterm birth in twin gestations (2004–2006). Details of the study have been published previously (18). Briefly, women carrying twin pregnancies who were 16 weeks to 20 weeks 3 days of gestation were recruited from 14 study centers in the United States after providing informed, written consent. They were randomized to weekly injections of either 17 α-hydroxyprogesterone caproate or placebo until 34 weeks and 6 days of gestation or delivery, whichever occurred first. Medical charts were abstracted by trained, certified personnel to ascertain data on antepartum and intrapartum events, obstetrical interventions, and neonatal outcomes. The study was approved by the institutional review boards at each clinical site and at the data coordinating center.

An ancillary study to the clinical trial to investigate the pharmacokinetics and pharmacodynamics of 17 α-hydroxyprogesterone caproate began with approximately 1 year left in recruitment. The participants of the ancillary study provided a nonfasting blood sample at 24–28 weeks of gestation. These samples were centrifuged and frozen at −80°C for future analysis. Serum samples were analyzed in a single batch for 25-hydroxyvitamin D [25-hydroxyvitamin D_2 + 25-hydroxyvitamin D_3] using liquid-chromatography-tandem mass spectrometry (19). The assay had a lower detection limit of 1 ng/mL and no upper limit. The intra-assay coefficient of variation was 8.2% and 5.9% for 25-hydroxyvitamin D_2 and 25-hydroxyvitamin D_3, respectively. There is no universally acceptable definition of vitamin D deficiency. Therefore, we explored 25-hydroxyvitamin D as a continuous variable, as a binary variable (less than 75nmol/L compared with 75 nmol/L or more) (6), and categorized based on distribution quartiles.

Gestational age was determined according to an algorithm on the basis of the last menstrual period and the results of ultrasonography of the larger fetus for women who conceived spontaneously (20). For women who conceived by in vitro fertilization, the duration of gestation was calculated on the basis of the date of embryo transfer and the age of the embryos when transferred (18). The primary outcome for the clinical trial was delivery of the first twin or death of either twin at less than 35 weeks gestation. For the purposes of this analysis, we have termed that outcome “preterm birth” and analyzed by pregnancy, not individual infant. Spontaneous preterm birth was defined as a preterm birth at less than 35 weeks occurring after preterm labor with intact membranes or preterm prelabor rupture of the fetal membranes. Prespecified criteria encoded in the study manual of operations were used to define whether the birth was indicated or spontaneous, and this distinction was made prior to the measurement of 25-hydroxyvitamin D.

Because 17 α-hydroxyprogesterone caproate had no significant effect on the occurrence of preterm delivery (18), all women were included in this secondary analysis. Continuous variables were compared using the Wilcoxon rank sum test. Categorical variables were analyzed using chi-square or Fisher exact test, where appropriate. Multivariable logistic regression models were used to quantify the association between maternal 25-hydroxyvitamin D and preterm birth after adjusting for prepregnancy body mass index (BMI), race and ethnicity, maternal age, parity, smoking, marital status, education, study site, gestational age at blood draw, 17 α-hydroxyprogesterone caproate treatment and season at blood draw (confounders identified using theory-based causal diagrams (21)). Nominal two-tailed *P* values were reported and no adjustments were made for multiple comparisons. $P<.05$ was considered significant. Statistical analysis was performed using SAS software (SAS Institute, Cary, NC).

Results

Of the 661 women enrolled in the randomized trial, 211 (31.9%) had a serum sample drawn at 24–28 weeks for serum 25-hydroxyvitamin D measurement. A total of 20 mothers in the parent study who were eligible for the ancillary study delivered before 24 weeks. Participants who had serum available for 25-hydroxyvitamin D measurement were slightly more likely than women without stored serum to be lean and better educated, but there were no significant differences in any other characteristic (Table 1). In the sample with 25-hydroxyvitamin D assayed, no total 25-hydroxyvitamin D concentrations fell below the detectable range. There were 75 cases of preterm birth (one of which qualified because of a stillbirth) at less than 35 weeks (35.6%), including 50 cases of spontaneous preterm birth at less than 35 weeks (50/186 deliveries that were not indicated preterm birth less than 35 weeks, 26.9%) and 25 patients were indicated deliveries prior to 35 weeks. Twenty-five women (11.8%) delivered before 32 weeks, including 10 women who delivered before 30

weeks. Of the 117 parous patients in this analysis, 16 had a previous delivery less than 35 weeks of gestation (13.7%).

The mean (standard deviation, SD) serum 25-hydroxyvitamin D was 82.7(± 31.5) nmol/L, and the median was 85.7 nmol/L; 40.3% of women had 25-hydroxyvitamin D less than 75 nmol/L, and 18.0% less than 50 nmol/L. Mothers with serum 25-hydroxyvitamin D less than 75 nmol/L were more likely than mothers with 25-hydroxyvitamin D 75 nmol/L or more to be non-Hispanic black or Hispanic, young, unmarried, obese before pregnancy, and to have less than a high school education and delivered earlier (Table 2). The median 25-hydroxyvitamin D concentrations among mothers identifying their race and ethnicity as non-Hispanic white, non-Hispanic black, and Hispanic or other were 96.6 nmol/L, 50.1 nmol/L, and 73.9 nmol/L, respectively.

Preterm birth less than 35 weeks occurred in 49.4% of mothers with 25-hydroxyvitamin D less tthan 75 nmol/L compared with 26.2% among those with 25-hydroxyvitamin D 75 nmol/L or more (Table 3, $P<.001$). Maternal serum 25-hydroxyvitamin D 75 nmol/L or more was associated with a 60% reduction in the odds of preterm birth compared with less than 75 nmol/L. This association remained after adjustment for maternal race and ethnicity, study site, parity, prepregnancy BMI, smoking status, marital status, education, 17 α-hydroxyprogesterone caproate treatment and season at blood sampling. When 25-hydroxyvitamin D was studied as a continuous variable, every 31.5 nmol/L (1-SD) increase in 25-hydroxyvitamin D was associated with a 50% decrease in the odds of preterm birth at less than 35 weeks. In quartile analysis, women with 25-hydroxyvitamin D in the highest fourth of the distribution had a reduced odds of preterm birth less than 35 weeks compared with those in the bottom fourth (Table 3).

Results were similar when we limited analysis to spontaneous preterm birth less than 35 weeks. Gravida with serum 25-hydroxyvitamin D less than 75 nmol/L had more spontaneous preterm birth (29/72; 40.3%) than women with 25-hydroxyvitamin D 75 nmol/L or more (21/114; 18.4%, $P<.01$). The adjusted OR (95% CI) for spontaneous preterm birth associated with a 1-SD increase in 25-hydroxyvitamin D was 0.5 (0.3–0.9). There were too few cases of indicated preterm birth less than 35 weeks (n=25) to perform statistical modeling. However, in bivariate analyses women with serum 25-hydroxyvitamin D less than 75 nmol/L were more likely to have indicated preterm birth less than 35 weeks than mothers with 25-hydroxyvitamin D 75 nmol/Lor more (23.2% compared with 11.4%, $P<.05$).

Maternal 25-hydroxyvitamin D concentration was also negatively associated with preterm birth less than 32 weeks (Table 4). Compared with 25-hydroxyvitamin D less than 75 nmol/L, serum 25-hydroxyvitamin D 75 nmol/L or more was associated with an 80% reduction in risk of preterm birth less than 32 weeks, after adjustment for confounders. A 1-SD increase in 25-hydroxyvitamin D was associated with a 60% decrease in the likelihood of preterm birth less than 32 weeks. None of these associations differed by maternal race and ethnicity.

Discussion

In this multicenter U.S. cohort of women carrying twins, we observed that serum vitamin D measured at 24–28 weeks was inversely associated with the risk of preterm birth at less than 35 weeks and less than 32 weeks and spontaneous preterm birth less than 35 weeks, even after adjusting for covariates such as prepregnancy BMI, race and ethnicity, and season.

Little is known about maternal vitamin D status in relation to risk of preterm birth in twin pregnancies. In a trial of 504 mothers carrying singleton pregnancies who were randomized to receive 400, 2000 or 4000 international unit vitamin D_3 per day from 12–16 weeks to delivery, there was no difference in rates of preterm birth without preeclampsia by treatment

group (22). These authors reported that mothers with 25-hydroxyvitamin D less than 80 nmol/L before delivery were more likely to deliver preterm without preeclampsia than mothers with higher concentrations, but this finding may be explained by reverse causality. Using 131 cases of preterm birth less than 35 weeks and 134 term controls, all of whom were singleton pregnancies with a history of preterm birth, from a U.S. multicenter randomized trial of omega-3 fatty acid supplementation, investigators observed no relationship between 25-hydroxyvitamin D at 12–16 weeks or 25–28 weeks and recurrent preterm birth (13). Our findings may differ because all women in this population had a history of preterm birth and received weekly injections of 17 α-hydroxyprogesterone caproate throughout pregnancy. Researchers using a Canadian cohort of 221 singleton pregnancies at high risk of preeclampsia observed no association between 25-hydroxyvitamin D at 19 weeks and preterm birth less than 37 weeks (14). In a sample of 884 human immunodeficiency virus-infected African mothers carrying singletons, there was no association between 25-hydroxyvitamin D at a mean (SD) of 20 (4) weeks and risk of preterm birth at less than 37 weeks or less than 34 weeks after controlling for multivitamin supplementation, age, enrollment CD4 cell count, and human immunodeficiency virusstage (15). Similarly, gestational age (rather than preterm birth) was studied in three other singleton gestation cohorts, and results were mixed (17,23,24).

Our finding that poor vitamin D status is associated with early preterm birth suggests that the anti-inflammatory and immunomodulating roles of vitamin D may be relevant(12). Vitamin D regulates uterine-natural killer cells and other immune cells in vitro by suppressing inflammatory cytokine production (25). A similar impact of vitamin D on cultured trophoblastic cells has been observed (26), which implies a general antiinflammatory role in pregnancy. Vitamin D promotes innate immune responses in monocytes by stimulating antimicrobial activity (27). In addition, vitamin D regulates genes critical for successful implantation, including *calbindin-D9K* and *HOXA* through intracrine or paracrine pathways (12).

The demands for micronutrients including vitamin D in twin pregnancies are believed to be higher than those for singleton pregnancies (16,28), yet a lack of data to inform nutrient needs in twin pregnancy has resulted in the same recommended dietary allowance (600 international unit vitamin D per day) for singleton and multiple pregnancies (3). We therefore did not expect to observe a higher median 25-hydroxyvitamin D in this multicenter U.S. cohort of twin pregnancies than what has been reported in other U.S. singleton samples (29-31), including in a nationally representative group of pregnant mothers (74.4 nmol/L for non-Hispanic whites; 33.2 nmol/L for non-Hispanic blacks, and 54.0 nmol/L for Mexican-Americans (5). The higher median 25-hydroxyvitamin D concentrations may be due to differences in intake of vitamin D through supplements or diet, or to our inclusion of women from the southern United States, who may be exposed to greater amounts of solar radiation. Unfortunately, we lacked data on vitamin D intake and sunlight exposure to evaluate the contributing factors. However, characteristics such as race and ethnicity and obesity, which are typically associated with vitamin D during pregnancy, were also related in our cohort.

Limitations of our study warrant comment. We used a convenience sample of mothers who provided a blood sample at 24–28 weeks as part of the ancillary study. Our results may be biased if vitamin D concentrations affected selection into the ancillary study. For instance, if vitamin D deficiency caused preterm birth, then excluding the 20 mothers in the parent study who delivered before 24 weeks (and therefore could not participate in the ancillary study) would lead to an underestimation of effects. Although we adjusted for many known factors to be associated with both 25-hydroxyvitamin D and preterm birth, unmeasured confounding by intake of other micronutrients, physical activity, and maternal genotype may have biased our findings, and imperfect measurement of prepregnancy obesity using self-reported data

and socioeconomic position using education and marital status may have led to residual confounding. Additionally, only one blood sample before the third trimester was obtained in the parent study, so we could not evaluate other windows of vitamin D exposure. Strengths of our study include measurement of 25-hydroxyvitamin D before the clinical onset of preterm delivery using liquid chromatography-tandem mass spectrometry (the gold standard method), and estimation of gestational age in pregnancy based on a well-established algorithm (18).

Preterm delivery is a major contributor to the excess infant morbidity and mortality in twin pregnancies (32), and identifying modifiable risk factors for preterm birth in twins is a major public health priority (33). Observational studies beginning early in pregnancy of large, representative cohorts of mothers carrying multiple fetuses are the logical next step to assess whether maternal 25-hydroxyvitamin D concentrations are consistently associated with preterm birth. If these results are replicated, randomized trials of vitamin D supplementation may be warranted.

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Appendix: List of Centers in the NICHD MFMU Network

In addition to the authors, other members of the *Eunice Kennedy Shriver* National Institute of Child

Health and Human Development Maternal–Fetal Medicine Units Network are as follows:

University of Pittsburgh — S. Caritis, E. Daugherty, M. Cotroneo, H. Simhan

University of Alabama at Birmingham — W. Andrews, J. Sheppard, A. Northen

Northwestern University — M. Dinsmoor (NorthShore University HealthSystem), G. Mallett, P. Simon, M. Huntley, M. Ramos

Drexel University — M. Hoffman, S. Wilson, C. Tocci, M. Lake, M. Talucci

University of Utah — K. Anderson, F. Porter (LDS Hospital), A. Guzman (McKay-Dee Hospital Center), K. Jolley (Utah Valley Regional Medical Center), S. Quinn (LDS Hospital)

Columbia University — R. Berkowitz, S. South, L. Paley, S. Bousleiman, V. Carmona

The Ohio State University —F. Johnson, C. Latimer

Case Western Reserve University — C. Milluzzi, C. Heggie, H. Ehrenberg, B. Stetzer, A. Merlino

University of North Carolina at Chapel Hill — K. Boggess, K. Dorman, S. Timlin

Wayne State University — G. Norman, C. Sudz, S. Blackwell

Brown University — D. Allard

University of Texas Southwestern Medical Center, Dallas — K. Leveno, L. Moseley

University of Texas Health Science Center at Houston — D. Soebbing-Cross, J. Martinez, B. Glenn-Cole, L. Gilstrap

Wake Forest University Health Sciences — P. Meis, M. Swain, K. Johnson, K. Lanier, C. Leftwich

The George Washington University Biostatistics Center — E. Thom, A. Braga, E. Cardenas, L. Leuchtenburg

Eunice Kennedy Shriver National Institute of Child Health and Human Development — S. Tolivaisa

MFMU Network Steering Committee Chair (*University of Texas Medical Center, Galveston, TX*) — G. Anderson, M.D

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Table 1

Characteristics of Participants From the Parent Clinical TrialWith and Without Available Serum For Assay of 25-Hydroxyvitamin D.

Characteristic	Women With Available Serumfor 25-Hydroxyvitamin D n= 211	WomenWithout Available Serum for 25-Hydroxyvitamin D n= 450*	*P*
Maternal race and ethnicity			0.36
Non-Hispanic white	131(62.1)	255 (56.7)	
Non-Hispanic black	47 (22.3)	107 (23.8)	
Hispanic or other	33 (15.6)	88 (19.5)	
Maternal age, y			0.62
Younger than 20	11 (5.5)	32 (7.5)	
20–29	84 (42.0)	180 (42.4)	
30 or older	105 (52.5)	213 (50.1)	
Marital status			0.96
Married or living with partner	157 (74.4)	334 (74.2)	
Not married	54 (25.6)	116 (25.8)	
Maternal education			0.04
Less than high school	32 (15.2)	72 (16.0)	
High school or equivalent	32 (15.2)	105 (23.3)	
Greater than high school	147 (69.7)	273 (60.7)	
Parity			0.93
0	94 (44.5)	202 (44.9)	
1 or more	117 (55.5)	248 (55.1)	
Smoking status			0.27
Smoker	18 (8.5)	51 (11.3)	
Nonsmoker	193 (91.5)	399 (88.7)	
Prepregnancy BMI, kg/m^2			0.02
Less than 25	95 (45.5)	215 (49.2)	
25–29.9	64 (30.6)	92 (21.1)	
30 or more	50 (23.9)	130 (29.7)	
Gestational age at blood draw, wk	26.2 (± 1.1)	-	-
Season of blood draw		-	-
Winter	56 (26.7)		
Spring	67 (31.9)		
Summer	37 (17.6)		
Fall	50 (23.8)		
Latitude of study site			0.79
40°N to 42°N	156 (73.9)	337 (74.9)	
30°N to 36°N	55 (26.1)	113 (26.1)	
17α-hydroxyprogesterone caproatetreatment group	93 (44.1)	234 (52.0)	0.06

Characteristic	Women With Available Serumfor 25-Hydroxyvitamin D n= 211	WomenWithout Available Serum for 25-Hydroxyvitamin D n= 450*	*P*
Pregnancy achieved using assisted reproductive technology	47 (22.3)	109 (24.2)	0.58
Infant birthweight, g	2182 (±510)	2084 (± 627)	0.16
Gestational age at delivery ,wk	35.2 (± 2.8)	34.4 (± 4.2)	0.32
Birth prior to 35 weeks	75 (±35.6)	183 (±41.2)	0.17
Spontaneous birth less than 35 weeks†	50 (26.9)	137 (34.4)	0.07

BMI, body mass index.

Data presented as n (%) or mean (± standard deviation) unless otherwise specified.

*Missing data on covariates were as follows: n=36missing maternal age; n=15 missing prepregnancy BMI; n=6 missing birth outcome data due to loss to follow-up.

†Out of those who were not indicated deliveries at less than 35 weeks.

Table 2

Maternal Characteristics in Association With Serum 25-Hydroxyvitamin D Concentrations at 24–28 Weeks.

Characteristic	Less Than 75 nmol/L n = 85	75 nmol/L or More n = 126	*P*
Maternal race and ethnicity			< 0.001
Non-Hispanic white	26 (30.6)	105 (83.3)	
Non-Hispanic black	40 (47.1)	7 (5.6)	
Hispanic or other	19 (22.3)	14 (11.1)	
Maternal age, y			< 0.001
Younger than 20	9 (11.4)	2 (1.7)	
20-29	43 (54.4)	41(33.9)	
30 or older	27 (34.2)	78 (64.5)	
Marital status			< 0.001
Married or living with partner	48 (56.5)	109 (86.5)	
Not married	37 (43.5)	17 (13.5)	
Maternal education			< 0.001
Less than high school	20 (23.5)	12 (9.5)	
High school or equivalent	18 (21.2)	14 (11.1)	
Greater than high school	47 (55.3)	100 (79.4)	
Parity			0.17
0	33 (38.8)	61 (48.4)	
1 or more	52 (61.2)	65 (51.6)	
Smoking status			0.06
Smoker	11 (12.9)	7 (5.6)	
Nonsmoker	74 (87.1)	119 (94.4)	
Pre-pregnancy BMI, kg/m^2			< 0.001
Less than 25	26 (31.0)	69 (55.2)	
25–29.9	27 (32.1)	37 (29.6)	
30 or more	31 (36.9)	19 (15.2)	
Season of blood draw			0.33
Winter	21 (25.0)	35 (27.8)	
Spring	32 (38.l)	35 (27.8)	
Summer	11 (13.1)	26 (20.6)	
Fall	20 (23.8)	30 (23.8)	
Latitude of study site			0.06
40°N to 42°N	57 (67.1)	99 (78.6)	
30°N to 36°N	28 (32.9)	27 (21.4)	
Treatment group			0.88
17α-hydroxyprogesterone caproate	38 (44.7)	55 (43.7)	
Inactive	47 (55.3)	71 (56.4)	
Gestational age at blood draw, wk	26.2 (± 1.1)	26.3 (± 1.2)	0.37
Gestational age at delivery, wk	34.5 (± 3.4)	35.7 (± 2.3)	<0.01

Characteristic	Less Than 75 nmol/L n = 85	75 nmol/L or More n = 126	*P*
Infant birthweight, g	2056 (± 535)	2263 (± 477)	<0.01

BMI, body mass index.

Data presented as n (%) or mean (± standard deviation) unless otherwise specified.

Table 3

Association Between Maternal Serum 25-Hydroxyvitamin D at 24–28 Weeks and Preterm Birth at Less Than 35 Weeks

Maternal 25-Hydroxyvitamin D	Preterm Birth Less Than 35 Weeks	Birth 35 Weeks or More	Pearson *P*	Unadjusted OR (95% CI)	Adjusted* OR (95% CI)
Less than 75 nmol/L	42 (49.4)	43 (50.6)		1.0 (ref)	1.0 (ref)
75 nmol/L or more	33 (26.2)	93 (73.8)	<0.001	0.4 (0.2, 0.7)	0.4 (0.2, 0.8)
Per onestandard deviation increase† (continuous)	n/a	n/a		0.5 (0.4,0.7)	0.5 (0.3, 0.8)
Quartile 1 (median 43.6 nmol/L)	27 (51.9)	25 (48.1)		1.0 (ref)	1.0 (ref)
Quartile 2 (median 72.7 nmol/l)	24 (45.3)	29 (54.7)	0.50	0.8 (0.4, 1.7)	1.0(0.4, 2.5)
Quartile 3 (median 95.4 nmol/L)	15 (28.3)	38 (71.7)	0.01	0.4 (0.2, 0.8)	0.4(0.2, 1.1)
Quartile 4 (median 116 nmol/L)	9 (17.0)	44 (83.0)	<0.001	0.2 (0.1, 0.5)	0.2 (0.1, 0.7)

OR, odds ratio; CI, confidence interval.

Data are n (%) unless otherwise specified.

*Adjusted for maternal race-ethnicity, gestational age at blood draw, study site, parity, prepregnancy BMI, season of blood draw, smoking status, marital status, education, and 17 α-hydroxyprogesterone caproate treatment

†One standard deviation is 31.5nmol/L.

Table 4

Association Between Maternal Serum 25-Hydroxyvitamin D at 24–28 Weeks and Preterm Birth at Less Than 3 Weeks

Maternal 25-Hydroxyvitamin D	Preterm Birth Less Than 32 Weeks	Birth 32 Weeks or More	Pearson *P*	Unadjusted OR (95% CI)	Adjusted* OR (95% CI)
Less than 75 nmol/L	16 (18.8)	69 (81.2)		1.0 (ref)	1.0 (ref)
75 nmol/L or more	9 (7.1)	117 (92.9)	0.01	0.3 (0.1, 0.8)	0.2 (0.1, 0.6)
Per one standard deviation increase† (continuous)	n/a	n/a		0.6 (0.4,0.9)	0.4 (0.2, 0.8)
Quartile 1 (median 43.6 nmol/L)	10 (19.2)	42 (80.8)		1.0 (ref)	1.0 (ref)
Quartile 2 (median 72.7 nmol/L)	7 (13.2)	46 (86.8)	0.40	0.6 (0.2, 1.8)	0.5 (0.1, 1.7)
Quartile 3 (median 95.4 nmol/L)	6 (11.3)	47 (88.7)	0.26	0.5 (0.2, 1.6)	0.4 (0.1, 1.5)
Quartile 4 (median 116 nmol/L)	2 (3.8)	51 (96.2)	0.01	0.2 (0.03, 0.8)	0.1 (0.02, 0.7)

OR, odds ratio; CI, confidence interval.

Data are n (%) unless otherwise specified.

*Adjusted for maternal race-ethnicity, gestational age at blood draw, study site, parity, pre-pregnancy BMI, season of blood draw, smoking status, marital status, education, and 17 α-hydroxyprogesterone caproate treatment

†One standard deviation is 31.5 nmol/L