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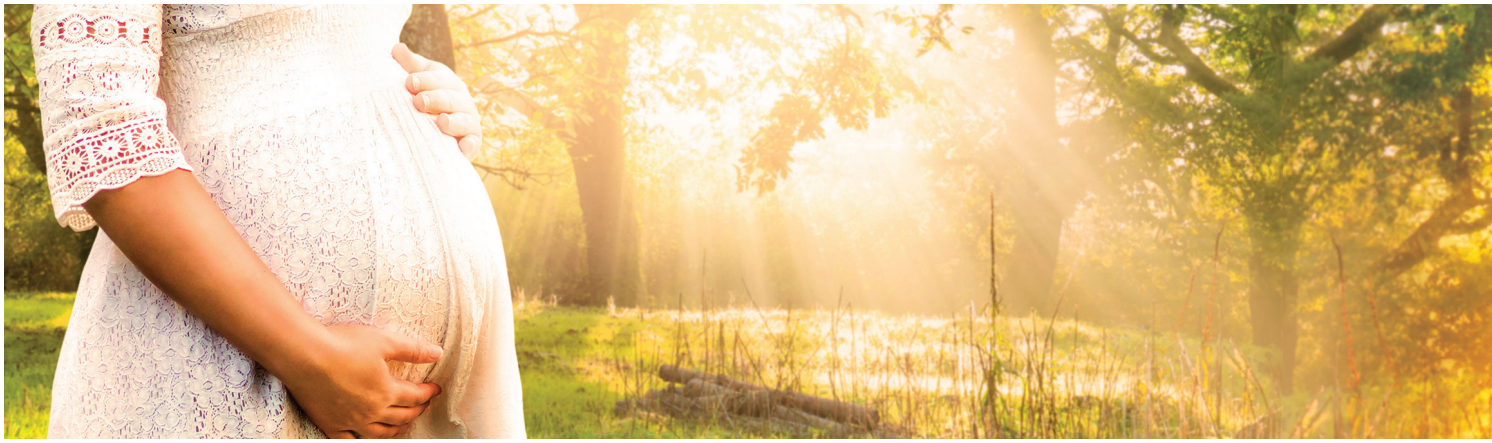

The Influence of Prenatal Vitamin D Supplementation on Dental Caries in Infants

Robert J. Schroth, DMD, PhD; **Jodie Christensen**, DMD; **Margaret Morris**, MD, MEd; **Patricia Gregory** RN, MN, PhD; **Betty-Anne Mittermuller**; **Cheryl Rockman-Greenberg**, MD, CM, FRCP(C), FCCMG

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ABSTRACT

Objectives: Early childhood caries (ECC) originates prenatally. This study investigated whether a relation exists between levels of vitamin D in the umbilical cord and caries in offspring.

Methods: A prospective cohort of expectant mothers was selected from a high-risk urban population receiving prenatal care in Winnipeg, Canada. Participants self-selected into 1 of 2 groups. The intervention group received 2 oral prenatal doses of 50 000 international units (IU) of vitamin D in addition to routine prenatal care. The control group received routine prenatal care. A prenatal questionnaire was completed at the first visit. Umbilical cord blood was analyzed for 25 hydroxyvitamin D (25(OH)D). At the time of their infant's first birthday, participants returned for a follow-up questionnaire and a dental examination of the infant. A p value ≤ 0.05 was significant.

Results: In all, 283 women were recruited (mean age 23.4 ± 5.6 years), 141 in the intervention group and 142 in the control group. The mean cord 25(OH)D level was 49.6 ± 24.3 nmol/L and did not differ between the groups. For the follow-up visit, 175 women returned. Overall, 26.3% of infants had ECC, and the mean decayed tooth (dt) score was 0.94 ± 2.16 teeth (range 0–16). There was no significant difference in prevalence of ECC between the intervention and control groups (p = 0.21). Poisson regression determined an inverse relation between 25(OH)D levels and dt scores (p = 0.001). Socioeconomic factor index (SEFI), age and enamel hypoplasia, but not vitamin D supplementation were significantly and independently associated with dt. Multiple logistic regression models also revealed that higher SEFI score, age and enamel hypoplasia were associated with ECC.

Conclusion: No relation was found between the 2 groups and prevalence of ECC. However, significance was seen in an inverse relation between 25(OH)D levels and the number of decayed primary teeth. Further studies with higher levels of vitamin D supplementation are needed.

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Early childhood caries (ECC) is a multifactorial disease, influenced by environmental factors, such as dietary intake, oral microbiome and social determinants of health.[1,2] Many children with ECC require rehabilitative surgery in hospital under general anesthesia.[3] ECC is known to impact childhood health and well-being.[4]

The role of diet in the development and prevention of caries was the focus of much research during the 1920s and 1930s.[5–7] Lady May Mellanby's pioneering studies[5] explored the impact of vitamin D rich diets on caries and tooth resistance. Her 1928 paper concluded that diets rich in vitamin D helped prevent caries initiation and limited or arrested the spread of caries. Conversely, diets low in vitamin D showed no such suppression.[5]

The active form of vitamin D, 1,25-dihydroxyvitamin D [1,25(OH)2D] regulates blood calcium by affecting the absorption of calcium from the intestines.[8] Its role in calcium and phosphorus homeostasis is key in the proper formation and maintenance of hard tissues,[8] including mineralization of teeth.[9,10] Vitamin D is also required in several cellular pathways including the immune response, cellular differentiation, proliferation and apoptosis.[11] Therefore, vitamin D may offer some protection against cariogenic microorganisms.[12-14]

Vitamin D is obtained exogenously through diet and supplements or endogenously via solar radiation. Populations residing in northern latitudes are disadvantaged regarding endogenous production, especially during the winter months (i.e., November to March) when sun-induced vitamin D synthesis in the skin is severely limited by less exposure to sunlight as well as latitude, altitude and other environmental factors.[15]

In Canada, milk, margarine and milk alternatives are fortified with vitamin D and are the major sources of dietary vitamin D.[16] Health Canada lists the dietary reference intake (DRI) of vitamin D as 600 international units (IU) for all age groups from 1 to 70 years and for pregnant and lactating women.[17] Most people can achieve this DRI through daily supplements, including prenatal vitamins, which supply 400 IU vitamin D. The Canadian Paediatric Society recommends that infants receive 400 IU daily in the summer and up to 800 IU during the winter months.[18]

Clinically, measurement of vitamin D status is based on the dominant form of plasma vitamin D, 25(OH)D, which accounts for both cutaneous and dietary sources.[19] Achieving optimal levels of 25(OH)D is especially important for pregnant women as fetal concentrations rely mainly on maternal concentrations. Optimal concentrations are ≥ 75 nmol/L.[20] However, the Institute of Medicine (IOM) lists adequate concentration as ≥ 50 nmol/L.[21] Vitamin D inadequacy has been found to be associated with many disease outcomes.[11,22] Prenatal vitamin D deficiency has also been identified as a possible risk factor for ECC.[12]

Cockburn et al.[23] reported that 400 IU of vitamin D daily during pregnancy was significantly associated with a lower prevalence of enamel defects in offspring. Furthermore, a recent study by Schroth et al.[12] reported, for the first time, that maternal prenatal vitamin D levels may influence the development of ECC: mothers of infants with ECC demonstrated lower prenatal 25(OH)D levels than mothers of caries-free infants. It is during the second trimester, when primary tooth calcification begins in utero, that maternal vitamin D status affects the teeth.

The purpose of this study was to investigate whether prenatal vitamin D supplementation of pregnant women during pregnancy increases 25(OH)D levels in the umbilical cord and whether that affects caries development in their infants.

Methods

This study was undertaken because of a government priority in response to recommendations made to Manitoba's Minister of Health by the Maternal and Child Healthcare Services Taskforce to improve prenatal 25(OH)D status and potentially improve infant oral health.[24] The taskforce recommended prenatal supplementation with 100 000 IU of vitamin D.[24]

Participants were recruited from the Women's Hospital Outpatient Department of the Health Sciences Centre, Winnipeg, Canada (latitude 49°53′ N) during 1 of their prenatal appointments. This clinic primarily serves an inner-city clientele, including Indigenous women, those with limited socioeconomic status (SES) and newcomers to Canada. Participants self-selected to 1 of 2 groups. Those in the intervention (i.e., supplementation) group were recruited during their first or second trimester and consented to take 2 oral doses of 50 000 IU of vitamin D in their second and third trimesters. Women not willing to take the vitamin D supplements and those recruited past the window of opportunity to be in the intervention group served as controls.

All participants received standard prenatal care, including prenatal vitamins. Those with known hypercalcemia, kidney disease, inborn errors of metabolism, chronic illness (excluding diabetes) and current involvement in a related clinical study were excluded.

All participants completed a questionnaire administered via interview; it collected information on demographics, nutrition, intake of foods containing vitamin D and sunlight exposure. Postal codes were used to examine socioeconomic status (SES) by calculating socioeconomic factor index (SEFI), an area-based measure. SEFI

is derived from Canadian census data for unemployment rate at age ≥ 15, average household income at age ≥ 15, proportion of single-parent households and proportion of the population age ≥ 15 years not graduating from high school. Those in the intervention group received 2 oral doses of 50 000 IU vitamin D, administered by a nurse in the outpatient clinic during scheduled prenatal visits. The first dose was given during the second trimester, the second during the third trimester. This supplementation was in addition to regular prenatal vitamins.

At delivery, cord blood was collected and assayed for 25(OH) D. Samples were analyzed by Hospitals in Common Laboratory (HICL), Mount Sinai Hospital, in Toronto, Canada, using chemiluminescence immunoassay. In this study, thresholds used to quantify 25(OH)D levels were ≥ 75 nmol/L (optimal based on HICL and Winnipeg's Health Sciences Centre), ≥ 50 nmol/L (adequate based on IOM) and < 35 nmol/L (common threshold used to denote deficiency).[14,21,25]

Participants and their infants were then invited to return for a follow-up examination around the child's first birthday. A questionnaire was administered to collect information on demographics, birthweight, prematurity and current or past health problems. Information regarding diet, oral hygiene, timing of the eruption of the first tooth and dental home status was also obtained. Primary dentition was assessed by the principal investigator (RJS) who was blinded to groups and cord 25(OH) D levels. ECC and severe ECC (S-ECC) were defined according to current standards.[1] Caries scores using the dmft index (i.e., combined total for decayed, missing due to caries and filled primary teeth) and dmfs index (i.e., combined total for decayed, missing due to caries and filled primary tooth surfaces) were also recorded. Developmental defects of enamel, such as enamel hypoplasia, were assessed.[26]

Data were entered into a database (Excel, Microsoft, Redmond, Wash.) and analyzed using statistical software (v. 9, NCSS, Kaysville, Utah) and R. Analysis involved descriptive statistics (frequencies, means ± standard deviation [SD]) and bivariate tests, X^2 tests, t tests and analysis of variance (ANOVA). Linear regression, logistic regression and negative binomial analyses were performed. A p value ≤ 0.05 was considered significant.

This prospective cohort study was approved by the University of Manitoba's Biomedical Research Ethics Board. Participants provided written informed consent.

Results

Overall, 283 women were enrolled in the study (mean age 23.4 ± 5.6 years): 141 in the intervention group and 142 in the control group. No significant difference in age was found between groups (22.9 ± 5.3 years intervention v. 24.0 ± 5.9 control, p = 0.11). There was also no significant difference between participants in the intervention and control groups in terms of number of children (2.0 ± 1.6 v. 2.2 ± 1.8, p = 0.38) or socioeconomic characteristics, such as household income (p = 0.99), receiving social assistance (p = 0.26), education level (p = 0.85) and employment level of the mother (p = 0.72) or mother's partner (p = 0.46). The groups were relatively well matched for prenatal vitamin use (p = 0.21), skin colour as rated by the participant (p = 0.34) and milk consumption (p = 0.064). Other characteristics of participants appear in **Table 1**.

Cord 25(OH)D concentrations were available for 216 participants (76.3%); the mean 25(OH)D level was 49.6 ± 24.3 nmol/L (**Table 1**). There were no significant differences between the intervention and control groups in terms of mean 25(OH)D cord level or the proportion attaining concentrations ≥ 50 nmol/L and ≥ 75 nmol/L (**Table 1**). Multiple regression analysis revealed that season of delivery and skin colour were significantly and independently associated with cord 25(OH)D levels (**Table 2**). Those who delivered in winter and those with darker skin colour had significantly lower 25(OH)D levels. Meanwhile, supplementation was not associated with higher mean 25(OH)D levels.

For follow up, 175 participants (61.8%) returned with their infants. Reasons for loss to follow up included moving (n = 4; 1 intervention and 3 control), fetal demise (n = 9; 6 intervention and 3 control) and infant placed in foster care (n = 11, 7 intervention and 4 control).

The proportion of study participants returning for follow up was consistent between groups (63.8% intervention v. 59.9% controls, p = 0.49). However, returning participants had significantly higher cord 25(OH)D levels at delivery than those lost to follow up (52.6 ± 23.1 nmol/L v. 44.0 ± 25.7 nmol/L, p = 0.013) and were significantly older (24.1 ± 5.6 years v. 22.3 ± 5.5 years, p = 0.01). Likewise, women who attended the follow-up study appointment were more likely to have completed high school or beyond than those lost to follow up (50.9% v. 37.0%, p = 0.024). They were also more likely to have taken prenatal vitamins than those lost to follow up (86.3% v. 75.9%, p = 0.028). Of interest, the proportion of women receiving government assistance was similar among those returning and those lost to follow up (42.8% v. 50.0%, p = 0.24) and among those who were regular milk drinkers (88.0% v. 86.1%, p = 0.45). However, women returning for follow up were less likely to be of in a lower income level than those lost (64.0% v. 80.0%, p = 0.042). There was also no difference in the prevalence of participants who identified as Indigenous between those returning and those lost to follow up (p = 0.086).

The mean age of returning infants was 19.7 ± 8.1 months, and this did not differ between groups (19.4 ± 7.8 months v. 20.0 ± 8.5 months, $p = 0.60$). Overall, 52% were male.

Table 1 shows results from the follow-up visit. There were no differences between groups in how much milk mothers consumed during pregnancy ($p = 0.26$), prenatal vitamins taken during pregnancy ($p = 0.71$) or the child's current health ($p = 0.67$).

Overall, 26.3% of the infants had both ECC and met the criteria for S-ECC; mean decayed tooth (dt) score was 0.94 ± 2.16 teeth (range 0–16). Other dental findings appear in **Table 3**. There was no apparent difference in the proportion of infants with S-ECC between the two groups (22.2% intervention v. 30.6% control, $p = 0.21$). Caries tooth and tooth surface scores did not appear to differ statistically between infants born to mothers in the intervention and control groups (**Table 3**). Poisson regression revealed a statistically significant inverse relation between cord 25(OH)D levels and dt scores ($p = 0.001$).

Two negative binomial model analyses were undertaken for dt scores (**Table 4**). Model 1 included which group they were in, while model 2 included 25(OH)D levels. Overall, SEFI, child age and enamel hypoplasia were significantly and independently associated with higher dt scores. In model 1, the dt incidence rate ratio (IRR) was significantly higher among infants with less ideal socioeconomic conditions (i.e., higher SEFI score) (IRR = 1.92), of older age (IRR = 1.10) and with enamel hypoplasia (IRR = 5.30). In model 2, the dt IRR was significantly higher among infants who were older, had higher SEFI score, and enamel hypoplasia (IRR= 1.06, IRR = 3.13 and IRR = 4.18, respectively).

Table 5 shows prenatal and post-natal data for infants with S-ECC compared with those who were caries free. Prenatal variables, such as mother's age ($p = 0.12$), taking vitamin D during pregnancy ($p = 0.95$) and mother's education level ($p = 0.063$) had no significant effect. However, mothers of infants with S-ECC were significantly more likely to be receiving government assistance ($p = 0.011$), have a household income below $28 000 ($p = 0.002$), be unemployed ($p < 0.001$) and rate their own dental health as poor ($p = 0.024$). Of note, infants of mothers receiving the Healthy Baby Prenatal Benefit were more likely to have S-ECC ($p <0.001$).

Although cord levels of 25(OH)D in infants with S-ECC were lower than those of caries-free infants (50.6 ± 21.6 nmol/L v. 53.3 ± 23.7 nmol/L), the difference was not statistically significant (**Table 5**). Post-natal characteristics, such as sex ($p = 0.71$), birthweight ($p = 0.55$) and prematurity ($p = 0.88$), were not associated with S-ECC. S-ECC was not significantly associated with breastfeeding ($p = 0.66$) or bottle feeding ($p = 0.088$). However, infants with S-ECC were bottle fed for longer than caries-free infants (18.4 ± 9.0 months v. 14.3 ± 4.9 months; $p = 0.008$). Infants with S-ECC were significantly older than infants without S-ECC (23.8 ± 8.8 months v. 18.2 ± 7.3 months; $p < 0.001$). Those with S-ECC were also more likely to have enamel hypoplasia ($p = 0.006$) and developmental defects of enamel ($p = 0.009$) than caries-free infants.

Two multiple logistic regression models for S-ECC were performed (**Table 6**). The first included study group and the second included 25(OH)D level. Overall, higher SEFI score (odds ratio [OR] = 1.64), each additional year of age (OR = 0.013) and the presence of enamel hypoplasia (OR = 12.25) were associated with increased odds of S-ECC. In addition, first pregnancy was associated with lower odds of S-ECC (OR = 0.29) in model 1.

Table 1: Characteristics of mothers and infants in our study group ($n = 283$).

Variable	Total population, *n* (%)	Intervention group, *n* (%)	Control group, *n* (%)	*p*
Maternal characteristics				
First pregnancy				
Yes	128 (45.2)	66 (46.8)	62 (43.7)	0.63
No	154 (54.6)	75 (53.2)	79 (55.6)	
Prenatal vitamins				
Yes	232 (82.3)	120 (85.1)	112 (79.4)	0.21
No	50 (17.7)	21 (14.9)	29 (20.6)	
Drink milk				
Often/sometimes	249 (88.0)	119 (84.4)	130 (91.5)	0.064
Rarely/never	34 (12.0)	22 (15.6)	12 (8.5)	
Skin colour				
Dark	103 (8.3)	8 (5.8)	15 (10.7)	0.34
Mid	151 (54.5)	77 (56.2)	74 (52.9)	
Light	103 (37.2)	52 (38.0)	51 (36.4)	

Table 1 continued ▶

Table 1: Characteristics of mothers and infants in our study group (*n* = 283).

Variable	Total population, *n* (%)	Intervention group, *n* (%)	Control group, *n* (%)	*p*
Healthy Baby Prenatal Benefit				
Yes	142 (50.4)	68 (48.6)	74 (52.1)	0.55
No/just applied	140 (49.6)	72 (51.4)	68 (47.9)	
Education level				
< Grade 12	153 (54.5)	77 (55.0)	76 (53.9)	0.85
≥ Grade 12	128 (45.5)	63 (45.0)	65 (46.1)	
Government assistance				
Yes	127 (45.5)	68 (48.9)	59 (42.1)	0.26
No	152 (54.5)	71 (51.1)	81 (57.9)	
Mother's employment				
Full or part time	74 (26.1)	36 (25.5)	38 (26.8)	0.81
Unemployed/other	209 (73.9)	105 (74.5)	104 (73.2)	
Partner's employment				
Full or part time	142 (63.9)	79 (68.1)	63 (59.4)	0.18
Unemployed/other	80 (36.1)	37 (31.9)	43 (40.6)	
Household income				
< $28 000	113 (40.1)	53 (37.6)	60 (42.6)	0.48 (0.99 when "not sure" excluded)
> $28 000	51 (18.1)	24 (17.0)	27 (19.1)	
Not sure	118 (41.8)	64 (45.4)	54 (38.3)	
Ethnic background				
Caucasian	60 (21.4)	29 (20.7)	31 (22.0)	0.34
Aboriginal	183 (65.1)	95 (67.9)	88 (62.4)	
Black	10 (3.6)	6 (4.3)	4 (2.8)	
Asian	18 (6.4)	7 (5.0)	11 (7.8)	
Other	4 (1.4)	0	4 (2.8)	
Prefer not to answer	6 (2.1)	3 (2.1)	3 (2.1)	
Indigenous heritage				
Yes	183 (65.1)	95 (67.9)	88 (62.4)	0.34
No	98 (34.9)	45 (32.1)	53 (37.6)	
Infant characteristics				
Premature				
Yes	18 (10.3)	8 (8.9)	10 (11.8)	0.53
No	157 (89.7)	82 (91.1)	75 (88.2)	
Infant had serious health problems at birth				
Yes	49(30.3)	29 (32.1)	24 (28.2)	0.57
No	112(69.7)	61 (67.8)	61 (71.8)	
Prenatal vitamins				
Yes	146 (83.4)	76 (84.4)	70 (82.3)	0.71
No	29 (16.6)	14 (15.6)	15 (17.7)	
Vitamin D supplements				
Yes	8 (4.6)	4 (4.4)	4 (4.7)	0.93
No	167 (95.4)	86 (95.6)	81 (95.3)	

Table 1 continued ▶

Table 1: Characteristics of mothers and infants in our study group (n = 283).

Variable	Total population, *n* (%)	Intervention group, *n* (%)	Control group, *n* (%)	*p*
Drink milk during pregnancy				
Often/Sometimes	149 (85.1)	74 (82.2)	75 (88.2)	0.26
Rarely/Never	26 (14.9)	16 (17.8)	10 (11.8)	
Infant's current health				
Very good	125 (71.4)	63 (70.0)	62 (72.9)	0.67
Good	50 (28.6)	27 (30.0)	23 (27.1)	
Current condition of infant's mouth				
Very good	84 (48.6)	46 (48.6)	38 (45.2)	0.69
Good	71 (41.0)	34 (41.0)	37 (44.1)	
Poor	18 (10.4)	9 (10.4)	9 (10.7)	
Government assistance				
Yes	101 (58.1)	50 (55.6)	51 (60.7)	0.49
No	73 (41.9)	40 (44.4)	33 (39.3)	
Received Healthy Baby Prenatal Benefit				
Yes	123 (70.3)	60 (66.7)	63 (74.1)	0.28
No	52 (29.7)	30 (33.3)	22 (25.9)	
Season of birth				
May–Oct.	62(46.6)	31(44.9)	31(48.4)	0.48
Nov.–Apr.	71(53.4)	38(55.1)	33(51.6)	
Serum analysis				
Available 25(OH)D results	216 (76.3)	107 (75.9)	109 (76.8)	
Mean 25(OH)D, nmol/L ± SD	49.6 ± 24.3	51.6 ± 22.1	47.4 ± 26.2	0.087*
Optimal 25(OH)D threshold				
< 75 nmol/L	177 (81.9)	85 (79.4)	92 (84.4)	0.34
≥ 75 nmol/L	39 (18.1)	22 (20.6)	17 (15.6)	
Adequate 25(OH)D threshold				
< 50 nmol/L	121 (56.0)	53 (49.5)	68 (62.4)	0.057
≥ 50 nmol/L	95 (44.0)	54 (50.5)	41 (37.6)	

Note: SD = standard deviation.
**1-tailed.*

Table 2: Linear regression for cord 25(OH)D level.

Variable	Estimate	Standard error	95% confidence interval	*p*
Intercept	66.92	7.08	—	—
First pregnancy	–3.70	4.10	–11.82, 4.41	0.37
(reference = no)	–6.16	4.33	–14.72, 2.41	0.16
Government assistance	–10.94	3.95	–18.76, –3.13	0.007
(reference = no)	–13.06	4.08	–21.14, –4.98	0.002
Season of delivery winter	–2.90	2.20	–7.26, 1.47	0.19
(reference = summer)	0.11	3.86	–7.54, 7.76	0.98
Medium/dark skin colour (reference = Light)	7.78	5.05	–2.2, 17.78	0.13

Note: SEFI = socioeconomic factor index .

Table 3: Dental outcomes for infants (n = 175).

Dental outcome	All infants, n (%)	Intervention group, n (%)	Control group, n (%)	*p*
S-ECC				
Yes	46 (26.3)	20 (22.2)	26 (30.6)	0.21
No	129 (73.7)	70 (77.8)	59 (69.4)	
dt score ± SD	0.94 ± 2.16 (range 0–16)	0.87 ± 2.39	1.01 ± 1.89	0.66
dmft score ± SD	1.03 ± 2.28 (range 0–16)	0.92 ± 2.42	1.14 ± 2.11	0.52
dmfs score ± SD	1.65 ± 4.69 (range 0-37)	1.57 ± 4.69	1.73 ± 4.71	0.81
Enamel hypoplasia				
Yes	12 (7.1)	4 (33.3)	8 (66.6)	0.16
No	158 (92.9)	86 (54.4)	72 (45.6)	
Enamel opacity				
Yes	39 (24.4)	23 (59.0)	16 (41.0)	0.35
No	121 (75.6)	61 (50.4)	60 (49.6)	
Development defects of enamel				
Yes	47 (29.4)	26 (55.3)	21 (44.7)	0.65
No	113 (70.6)	58 (51.3)	55 (48.7)	

Note: dmfs = decayed, missing, filled surfaces, dmft = decayed, missing, filled primary teeth, dt = decayed tooth, SD = standard deviation, S-ECC = severe early childhood caries.

Table 4: Negative binomial model for decayed teeth (dt) index.

Variable	Estimate	Standard error	Odds ratio	95% confidence interval	*p*
Model 1					
Intercept	–3.65	0.66	—	—	—
Group 2 (control group) (reference = intervention group)	0.57	0.39	1.76	0.81, 3.94	0.14
Area based SEFI	0.65	0.21	1.92	1.22, 3.14	0.002
First pregnancy (reference = no)	–0.04	0.40	0.96	0.43, 2.22	0.93
Infant's age (months)	0.09	0.02	1.10	1.048, 1.16	< 0.001
Enamel hypoplasia (reference = no)	1.67	0.69	5.30	1.43, 26.27	0.02
Bottle duration (months)	0.02	0.03	1.02	0.96, 1.078	0.51
Model 2					
Intercept	–3.51	0.89	—	—	—
25(OH)D level (nmol/L)	0.01	0.009	0.03	0.99, 1.03	0.22
Area based SEFI	0.75	0.25	3.13	1.24, 3.85	0.002
First pregnancy (reference = no)	–0.02	0.44	0.98	0.38, 2.51	0.96
Infant's age (months)	0.06	0.03	1.06	1.004, 1.13	0.03
Enamel hypoplasia (reference = no)	1.43	0.91	4.18	0.83, 34.24	0.03
Bottle duration (months)	0.03	0.03	1.03	0.96, 1.11	0.35

Note: SEFI = socioeconomic factor index.

Table 5: Association between prenatal and post-natal factors and severe early childhood caries (S-ECC; *n* = 175).

Variable	S-ECC, *n* (%)	Caries free, *n* (%)	*p*
Prenatal characteristics			
Mean age of mother, years ± SD	23.0 ± 5.5	24.5 ± 5.6	0.12
Vitamin D during pregnancy			
Yes	38 (82.6)	106 (82.2)	0.95
No	8 (7.4)	23 (7.8)	
Prenatal vitamins			
Yes	36 (80.0)	114 (88.4)	0.16
No	9 (20.0)	15 (11.6)	
Mother's education level			
< Grade 12	28 (60.9)	57 (44.9)	0.063
≥ Grade 12	18 (39.1)	70 (55.1)	
Government assistance			
Yes	26 (59.1)	48 (37.2)	0.011
No	18 (40.9)	81 (62.8)	
Rating of own dental health			
Good	13 (28.3)	54 (41.8)	0.024
Fair	23 (50.0)	65 (50.4)	
Poor	10 (21.7)	10 (7.8)	
Household income			
< $28 000	24 (88.9)	49 (56.3)	0.002*
≥ $28 000	3 (11.1)	38 (43.7)	
Mother's employment			
Full or part time	5 (10.9)	53 (41.1)	< 0.001
Unemployed/other	41 (89.1)	76 (58.9)	
First pregnancy			
Yes	15 (32.6)	70 (54.7)	0.010
No	31 (67.4)	58 (45.3)	
Healthy Baby Prenatal Benefit			
Yes	36 (78.3)	60 (46.9)	< 0.001
No	10 (21.7)	68 (53.1)	
Partner's employment			
Full or part time	19 (52.8)	78 (72.9)	0.025
Unemployed/other	17 (47.2)	29 (27.1)	
Indigenous heritage			
Yes	32 (69.6)	74 (58.3)	0.18
No	14 (30.4)	53 (41.7)	
Post-natal characteristics			
Mean age of infant at follow-up, months ± SD	23.8 ± 8.8	18.2 ± 7.3	< 0.001
Mean 25(OH)D, nmol/L ± SD	50.6 ± 21.6	53.3 ± 23.7	0.54
Optimal 25(OH)D threshold			
≥ 75 nmol/L	7 (17.9)	21 (20.6)	0.73
< 75 nmol/L	32 (82.1)	81 (79.4)	

Table 5 continued ▶

Table 5: Association between prenatal and post-natal factors and severe early childhood caries (S-ECC; *n* = 175).

Variable	S-ECC, *n* (%)	Caries free, *n* (%)	*p*
Adequate 25(OH)D threshold			
≥ 50 nmol/L	19 (48.7)	51 (50.0)	0.89
< 50 nmol/L	20 (51.3)	51 (50.0)	
Sex of infant			
Male	25 (54.3)	66 (51.2)	0.71
Female	21 (45.7)	63 (48.8)	
Premature			
Yes	5 (10.9)	13 (10.1)	
No	41 (89.1)	116 (89.9)	0.88
Mother's rating of infant's dental health			
Very good	10 (22.7)	74 (57.2)	< 0.001
Good	22 (50.0)	49 (38.0)	
Poor	12 (27.3)	6 (4.7)	
Enamel hypoplasia			
Yes	7 (16.3)	5 (3.9)	0.006
No	36 (83.7)	122 (96.1)	
Developmental defects of enamel			
Yes	19 (45.2)	28 (23.7)	0.009
No	23 (54.8)	90 (76.3)	
Started brushing/cleaning teeth			
Yes	43 (93.5)	113 (87.6)	0.41*
No	3 (6.5)	16 (12.4)	
Breastfed			
Yes	29 (63.0)	86 (66.7)	0.66
No	17 (37.0)	43 (33.3)	
Duration, months ± SD	7.6 ± 6.9	7.2 ± 5.5	0.73
Bottle fed			
Yes	39 (86.7)	120 (94.5)	0.088
No	6 (13.3)	7 (5.5)	
Duration, months ± SD	18.4 ± 9.0	14.3 ± 4.9	0.008
Bottle at bedtime			
Yes	31 (68.9)	70 (54.3)	0.087
No	14 (31.1)	59 (45.7)	
Milk drinker			
Regular	44 (95.6)	116 (89.9)	0.36*
Other	2 (4.4)	13 (10.1)	

Note: SD = standard deviation.
**Fisher's exact test.*

Table 6: Logistic regression for severe early childhood caries versus caries free.

Variable	Estimate	Standard error	Odds ratio	95% confidence interval	*p*
Model 1					
Intercept	–0.66	0.64	—	—	—
Group 2 (control) (reference = intervention group)	0.16	0.41	1.18	0.52, 2.65	0.69
Area based SEFI	0.50	0.21	1.64	1.09, 2.53	0.020
First pregnancy (reference = no)	–1.25	0.45	0.29	0.11, 0.67	0.005
Infant's age (months)	0.069	0.024	1.07	1.02, 1.12	0.004
Enamel hypoplasia (reference = no)	2.51	0.76	12.25	2.91, 60.69	0.001
Model 1					
Intercept	–2.62	0.85	—	—	—
25(OH)D level (nmol/L)	–0.0033	0.0099	0.10	0.98,1.02	0.74
Area based SEFI	0.65	0.26	1.92	1.18, 3.25	0.011
First pregnancy (reference = no)	–0.93	0.48	0.39	0.15, 0.99	0.051
Infant's age (months)	0.068	0.027	1.07	1.02, 1.13	0.013
Enamel hypoplasia (reference = no)	3.29	0.95	26.92	4.82, 230.68	0.001

Note: SEFI = socioeconomic factor index.

Discussion

This prospective study attempted to explore whether supplementation with 100 000 IU of vitamin D during pregnancy improved umbilical cord 25(OH)D levels and infant oral health. It also allowed us to observe which infants developed S-ECC. Overall, 81.9% of participants had suboptimal cord levels of 25(OH)D (< 75 nmol/L), which is a concern as the developing fetus is dependent on the mother for vitamin D. It was recently reported that pregnant women with inadequate vitamin D status are at a higher risk of having offspring who develop ECC.[12]

Early research demonstrated an association between vitamin D status (via diet and sun exposure) and caries.[5,27] However, few studies have examined the link between cord 25(OH)D levels at birth and caries development in offspring.[12,28] Although our study did not find a direct link between supplementation and lower odds of S-ECC, we found that infants with higher cord 25(OH)D levels had significantly lower dt scores, which is consistent with an earlier study that was the first to link prenatal 25(OH)D levels and ECC.[12]

The mean cord concentration of 25(OH)D of all participants in this study failed to reach the IOM's threshold for adequacy. It was similar to the prenatal levels previously reported in our study linking maternal 25(OH)D levels and caries in offspring (48 nmol/L).[12] Surprisingly, we did not identify significant differences in 25(OH)D levels between the intervention and control groups (51.6 ± 22.1 nmol/L v. 47.4 ± 26.2). However, it was apparent that a higher proportion of infants in the intervention group met or exceeded the IOM adequacy threshold. It could be argued that the dose of vitamin D administered (2 doses of 50 000 IU) was modest and insufficient to raise cord levels to optimal or adequate levels. Tannous et al.[29] reported no toxicity from administering 100 000 IU of vitamin D weekly for 4 weeks among children 2–16 years. Thus, a higher dose or more frequent administration to participants in our study may have been needed. As well, 400 IU/day of vitamin D_3 for 8 weeks generally only leads to an increase in 25(OH)D of 11 nmol/L in healthy adults.[30] Therefore, intakes of vitamin D_3 in the range of 400 IU and fortified foods are unable to produce marked increases in 25(OH)D levels to optimal ranges if baseline levels are low.[8] A significant limitation of this study was the fact that supplementation was in the form of vitamin D_2, which is less effective than vitamin D_3 in sustaining higher 25(OH)D levels for a longer period.[31,32] Considering this, it is interesting that hospital pharmacies in Canada carry only plant-based high-dose vitamin D_2 supplements.

Despite our inability to demonstrate an association between vitamin D supplementation and ECC, the idea that vitamin D

supplementation could reduce the risk of caries is still plausible as evidenced by the meta-analysis of past studies on the topic of vitamin D and caries.[27] There is evidence that supplementation with D_2, D_3 and solar irradiation are all associated with a reduced risk of caries in children.[27] Our finding of an significant inverse relation between cord 25(OH)D concentrations and dt scores on Poisson regression would also suggest that higher prenatal 25(OH)D levels may be protective against caries. Concentrations above 50 nmol/L have been shown to be protective against caries in children.[33,34]

This study draws attention to the increased prevalence of caries in urban Canadian children of low socioeconomic status. Overall, 23.6% of the infants presented with S-ECC. Our findings suggest that this population is subject to increased risk factors for ECC. Infants with ECC were significantly more likely to have mothers who receive government assistance ($p = 0.011$), have a household income < $28 000 ($p = 0.002$) and are unemployed ($p < 0.001$). Limited parental education and low family income are known to increase the risk of dental problems during early childhood.[35-38] However, this study did not find an association between education level and ECC ($p = 0.063$). Surprisingly, mothers receiving the Healthy Baby Prenatal Benefit were more likely to have infants with ECC ($p < 0.001$), although this finding may be confounded by the involvement of other factors, such as income or employment. ECC is highly associated with the social determinants of health.[2] We used an area-based socioeconomic measure, SEFI, in our regression models and found that this covariate was significantly and independently associated with caries development in infants.

Several studies highlight the relation between bottle use and caries incidence. Common bottle-feeding habits, such as prolonged bottle use, bedtime bottle use and the addition of sugar promote caries development in young children.[39,40] Similarly, our study found that infants with ECC were bottle fed for longer than caries-free infants ($p = 0.008$). However, the use of a bottle at bedtime was not significantly associated with ECC. Other notable predictors of ECC included child's age ($p < 0.001$) and presence of enamel hypoplasia ($p = 0.006$). This mirrors what was found in an earlier prospective study on the association between maternal 25(OH)D levels and caries in infants.[12] A recent systematic review and meta-analysis[41] concluded that enamel hypoplasia is the strongest risk factor for ECC in children from upper-middle-income countries, and it was significantly and independently associated with ECC in our study. Although enamel hypoplasia has been an underappreciated risk factor, there is increasing awareness of its significance in ECC development.[12,42,43] The numerous risk factors reported for these defects include inadequate vitamin D levels during pregnancy.[12,23,28]

There was no significant difference between the intervention and control groups in percentage of infants with ECC. Similarly no difference was seen in decayed tooth and tooth surface scores between infants born to mothers of either group. However, this observation may be explained by the non-significant difference in vitamin D status between the 2 groups.

There were several limitations to this study. First, 38.2% of participants did not complete the study, although the proportion of returning mothers did not differ between the groups. Those returning had significantly higher cord 25(OH)D levels than those lost to follow up. Had more mothers returned, we might have been able to determine whether there was an association between supplementation and reduced risk of caries. As previously suggested, the vitamin D dose may have been too low or given too infrequently to cause a significant difference in cord 25(OH)D levels between the 2 groups; the mean level among those in the intervention group was only 51.6 nmol/L. The form of vitamin D administered, vitamin D_2, is also contributes to lower 25(OH)D values than that of vitamin D3. Future studies may benefit by investigating different dose regimens and vitamin D subtypes. This study may also be limited by the choice of participants by convenience rather than randomly. However, a population at high-risk of low vitamin D status and ECC was intended for this study to make it generalizable to the urban inner-city population. Another limitation was that we measured 25(OH) D levels only via cord blood. Had we included a prenatal baseline level (before any high-dose supplementation), we would have been able to obtain a more accurate measure of the effect of vitamin D_2. Another limitation is the confounding that arises from the inability to adjust for participants' self-selection into the 2 groups.

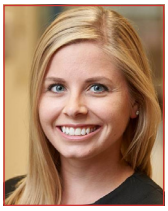
Strengths of the study include the prospective design and the moderately sized and deliberately chosen sample. Cohort studies also have the advantage of decreased bias. In addition, the dental assessments were completed by a single practitioner who was blinded to each participant's grouping. The design of the experiment allowed for the observation of natural caries development and for the assessment of multiple outcomes after altering a single factor, 25(OH)D level.

Conclusion

Vitamin D supplementation in a prospective cohort of pregnant women was not associated with higher cord 25(OH)D levels compared with a control group. The prevalence of ECC in the 2 groups also did not differ. However, there was a significant inverse relation between cord 25(OH)D levels and the number of decayed primary teeth.

THE AUTHORS

Dr. Schroth is professor and clinician scientist, department of preventive dental science, College of Dentistry and departments of pediatrics and child health and community health sciences, Max Rady College of Medicine, Rady Faculty of Health Sciences, University of Manitoba. He is also research scientist, Children's Hospital Research Institute of Manitoba.



Dr. Christensen is a dentist in private practice, Winnipeg, Manitoba.

Dr. Morris is professor, department of obstetrics, gynecology and reproductive sciences, Max Rady College of Medicine, Rady Faculty of Health Sciences, University of Manitoba.

Dr. Gregory is research, scholarship & quality improvement coordinator, department of nursing, School of Health Sciences and Community Services, Red River College, Winnipeg, Manitoba.

Ms. Mittermuller is research coordinator, Dr. Gerald Niznick College of Dentistry, Rady Faculty of Health Sciences, University of Manitoba and Children's Hospital Research Institute of Manitoba.

Dr. Rockman-Greenberg is distinguished professor, department of pediatrics and child health, Max Rady College of Medicine, University of Manitoba and research scientist, Children's Hospital Research Institute of Manitoba.

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Correspondence to: *Dr. Robert J. Schroth, 507 — 715 McDermot Ave., Winnipeg MB R3E 3P4. Email: robert.schroth@umanitoba.ca*

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