Determinants of vitamin D status in pregnant fair-skinned women in Sweden

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Abstract

Low maternal vitamin D status during pregnancy may have negative consequences for both mother and child. There are few studies of vitamin D status and its determinants in pregnant women living at northern latitudes. Thus, the present study investigates vitamin D status and its determinants during the third trimester of women living in Sweden (latitudes 57–58°N). A total of ninety-five fair-skinned pregnant women had blood taken between gestational weeks 35 and 37. The study included a 4 d food diary and questionnaires on dietary intake, supplement use, sun exposure, skin type, travels to southern latitudes and measure of BMI. Serum 25-hydroxyvitamin D (25(OH)D) was analysed using the chemiluminescence immunoassay. In the third trimester of pregnancy, mean serum concentration of 25(OH)D was 47·4 (SD 18·1) nmol/l (range 10–93 nmol/l). In total, 65 % of women had serum 25(OH)D $<$ 50 nmol/l and 17 % $<$ 30 nmol/l. During the winter, 85 % of the pregnant women had serum 25(OH)D $<$ 50 nmol/l and 28 % $<$ 30 nmol/l. The main determinants of vitamin D status were as follows: season; use of vitamin D supplements; travels to southern latitudes. Together, these explained 51 % of the variation in 25(OH)D. In conclusion, during the winter, the majority of fair-skinned pregnant women had serum 25(OH)D $<$ 50 nmol/l in their third trimester and more than every fourth woman $<$ 30 nmol/l. Higher vitamin D intake may therefore be needed during the winter for fair-skinned pregnant women at northern latitudes to avoid vitamin D deficiency.

Key words: 25-Hydroxyvitamin D: Pregnancy: Dietary intake: Sun exposure

Low vitamin D status has been associated not only with sub-optimal bone health, but also with higher frequencies of CVD[1], type 1 diabetes[2], cancers[3,4], infectious diseases[3], multiple sclerosis[3,5] and psychological conditions such as depression and schizophrenia[6]. During pregnancy, vitamin D deficiency also has been associated with maternal health outcomes, e.g. hypertensive disorders[7], gestational diabetes[5,8] and risk of caesarean section[9]. Further, low maternal vitamin D status during pregnancy may have an impact on fetal imprinting[10], increase the risk of low birth weight[11–13] and small-for-gestational age[7,14], and may also effect the child's bone health[15]. Exposure of pregnant women and fetuses to high doses of vitamin D also requires careful attention due to risks of hypercalcaemia and hypercalciuria and other possible adverse effects[16].

A major function for vitamin D is its role in Ca and bone metabolism. During pregnancy, the need for Ca is increased due to the requirement of Ca to form the fetal skeleton. This may lead to the mobilisation of Ca from the maternal skeleton[17]. We have shown that there is a decrease in whole-body bone mineral content of about 2 % during pregnancy[18]. It is not known whether these pregnancy-induced skeletal changes are vitamin D dependent. However, women who are pregnant during the winter, when UVB exposure is low, have higher ultrasound indices of maternal bone loss[19]. This may indicate a role for vitamin D. Overall, longitudinal studies have suggested that decreases in maternal bone mineral density during reproduction are transient, with replenishment of skeletal minerals in the later stages of lactation and after lactation has ceased[20]. The role for vitamin D in the mineral replenishment process remains to be clarified. Normally, parathyroid hormone (PTH) plays a major role in maintaining Ca balance, by increasing Ca release from the skeleton and by increasing Ca reabsorption from the kidneys[21]. During pregnancy, however, the role of PTH is unclear and the inverse relationship between serum PTH and serum Ca may not be the same as in non-pregnant adults[22]. Also, the inverse relationship between PTH and 25-hydroxyvitamin D (25(OH)D) may be weaker in pregnant women[22,23].

Vitamin D is partly obtained from the diet and dietary supplements. In Sweden, the major dietary sources of vitamin D are fish, fish-containing foods and dairy products[24]. Vitamin D is also obtained by cutaneous synthesis induced after UVB light exposure[25]. Below latitude 35°N, cutaneous vitamin D synthesis is possible all year[25]. In the circulation, 25(OH)D is the metabolite, which is measured as a proxy for vitamin D status[26–29]. Among non-pregnant women, season[30], skin pigmentation[31], BMI[25,30] and dietary intake, e.g. fatty fish[32] and vitamin D supplement use[33], have been shown to be associated with serum 25(OH)D. For pregnant women, determinants of 25(OH)D are less understood, but are known to include season and ethnicity[12,34–37]. Lifestyle factors such as sun exposure, supplement use and dietary intake of vitamin D have not been well studied. However, a recent Belgian study reported significant determinants of serum 25(OH)D during pregnancy to include sunscreen use, preference for shadow and holidays to sunny climates, as well as vitamin D supplement use, ethnicity, alcohol use, smoking and education[38].

Studies measuring the vitamin D status of pregnant women in Western societies have found mean concentrations of 25(OH)D between 26 and 98 nmol/l[12,14,23,34,38–43]. Many of these studies were conducted at latitudes where cutaneous production of vitamin D is possible for most of the year. Low serum 25(OH)D in pregnant dark-skinned women has been reported in several studies[23,41,44,45]. Less is known about vitamin D status and its determinants in pregnant fair-skinned women living in Sweden or at similar northern latitudes. To our knowledge, only a few studies have been published about vitamin D status of pregnant fair-skinned women living at these latitudes[38,42,43,46,47], of which only one has reported the determinants of vitamin D status thoroughly[38]. The aims of the present study were to assess vitamin D status and to evaluate its determining factors during the third trimester of pregnancy in fair-skinned women living in Sweden.

Subjects and methods

Subjects

Women were recruited from July 2008 to July 2011 using posters in maternity health care clinics and in public places in the vicinity of Gothenburg, Sweden, and through advertisement on a web page addressing pregnant women in western Sweden. In total, ninety-five pregnant women were recruited. Inclusion criteria were age 25–40 years, pregnancy in gestational weeks 35–37 when starting the study and to declare oneself as healthy. Exclusion criteria were prescribed medicine intake known to affect Ca and bone metabolism, pregnancy during the last 1·5 years before the start of the present pregnancy, miscarriage after week 12 of pregnancy during the last 1·5 years, breast-feeding during the last year before the start of the present pregnancy, twin pregnancy and development of gestational diabetes or pre-eclampsia. The present study was conducted according to the guidelines laid down in the Declaration of Helsinki, and all procedures involving human subjects were approved by the Regional Ethics Committee in Gothenburg. Written informed consent was obtained from all women.

Study design

All women visited the Department of Internal Medicine and Clinical Nutrition, University of Gothenburg, Sweden when they were between 35 and 37 weeks pregnant. Venous blood was drawn in the morning after an overnight fast. Body weight in underwear (Tanita, BWB-800MA; Rex Frederiksbergs Vaegtfabrik) and height (standardised wall stadiometer) were measured. Women were asked what their body weight was before they became pregnant. Hence, pre-pregnancy BMI is based on self-reported pre-pregnancy body weight and the height measured at the study visit. Women were also asked questions about their medical history, sun exposure, skin type, dietary intake and physical activity. After birth, women were asked to report day of birth, birth weight and birth length.

Methods

Sun exposure was estimated using questions compiled by Burgaz et al.[32]. These included use of sunscreen (always, sometimes or never) and preference for sun or shade when outdoors in summer (always in the sun, both sun and shade or always in the shade). Women were asked whether they had used a sunbed during the previous 6 months. Skin types were defined using the Fitzpatrick scale (I = always burns, never tans, II = usually burns, tans with difficulty, III = sometimes burns mildly, tans gradually, IV = rarely burns, tans easily)[48]. Women were asked to estimate the number of hours spent outdoors between 09.00 and 18.00 hours on weekdays, weekends, summer and winter, respectively. Weekdays corresponded to working days and weekends to non-working days. Summer was defined as May–October and winter as November–April. Women were also asked to report travels to southern latitudes during the previous 6 months. Southern latitude was defined as a location below latitude 35°N where cutaneous synthesis of vitamin D is possible all year round[25].

Dietary intake of vitamin D was estimated using 4 d food diaries. Women were asked to record all food and drink consumed as precisely as possible on four consecutive days with at least one non-working day and a preferred start no later than 1 week after the study visit. Both oral and written information on how to fill in the food diary were given. Women were asked to report the amounts of consumed food items using household measures, weight in grams or using photographs of different portion sizes used in the Swedish portion guide 'Matmallen'[49]. Women were also asked not to change their diet. Women were contacted if any ambiguities were noted in their food diaries. Dietary intake was calculated using DietistXP, version 3.1 (The National Food Agency food database version 2009-11-10; Kost och näringsdata). In addition, a short FFQ was also used to study the frequency and quantity of foods rich in vitamin D consumed, e.g. fatty

fish and dairy products. Specifically, information about the intake of reduced-fat dairy milk and yogurt/sour milk was requested. Details of use, frequency, amount and brand of supplements containing vitamin D were also requested.

Each woman rated her physical activity on a scale between 1 and 10[50]. Women were informed that 1 indicated a sedentary lifestyle, 5 a few long walks each week and 10 exercise several times per week. The answer was converted to a physical activity level (PAL), where 1 corresponded to PAL 1·3 and 10 to PAL 2·2, respectively. Each step between them represented a 0·1 increase. In a validation study, PAL assessed using this scale was correlated (r 0·54; $P = 0.008$) with corresponding estimates obtained using criterion methods (i.e. the doubly labelled water method in combination with indirect calorimetry) in twenty-two healthy Swedish pregnant women (M Löf, personal communication). Here, the individual self-estimated PAL was used for validating energy intake from the 4 d diary food records and thus for identifying possible under-reporters according to Goldberg *et al.*[51] and Black[52]. No correction for under-reporting was made. BMR was calculated using the Food and Agriculture Organization/WHO/United Nations University's equation for non-pregnant women[53], and a general increase in BMR of 24 % for pregnant women in their third trimester was added as suggested by Butte & King[54].

Laboratory analyses

Blood samples were protected from UVB light and centrifuged no later than 45 min after sampling at 5°C, 3800 g, for 9 min (Centrifuge CR3i; Jouan Quality System). Serum was then aliquoted and stored at −70°C until analysed. The analyses of serum concentrations of 25(OH)D and PTH were performed by Central Laboratory, Sahlgrenska University Hospital, Gothenburg, Sweden. All samples were analysed at one time. Analyses of total 25(OH)D and 25-hydroxyvitamin D_3 were performed in serum with the LIAISON® 25(OH)D chemiluminescence immunoassay (DiaSorin). Intact PTH was analysed with an immunochemical two-step analysis of sandwich type, using chemiluminescent microparticle immunoassay technology (Abbott Laboratory Diagnostics Division). Intra-assay CV were 7·3, 5·7 and 5·3 % for 25(OH)D serum concentrations of 22, 50 and 150 nmol/l, respectively. For PTH, CV were 3·7, 4·5 and 3·5 % for PTH serum concentrations of 10, 40 and 730 ng/l, respectively.

Statistical analyses

Independent sample t tests and ANOVA were used to evaluate the difference in the mean concentration of 25(OH)D depending on lifestyle and other factors, such as parity, estimates of sun exposure, estimates of vitamin D intake and PTH. Estimates of sun exposure included season, time spent outdoors, recent travels to southern latitudes, skin type, sun preference and sunscreen use, and estimates of vitamin D intake included total intake of vitamin D (from diet and supplements), dietary intake of vitamin D, intake of different food items rich in vitamin D and vitamin D supplement use. The coefficients of determination for 25(OH)D serum concentrations were calculated using bivariate regression analyses for the following variables: estimates of sun exposure; estimates of vitamin D intake; PAL; body weight; BMI. The variables significant in the bivariate regression analyses were included in the multivariate regression analysis. The effects of interactions between factors on 25(OH)D concentrations were modelled by the inclusion of combinations of sun exposure estimates and vitamin D intake estimates. In the multivariate regression analysis, a variable was considered a confounder if its inclusion in the model caused a >10 % change in the coefficient of the slope. However, no potential confounders were found. The significance level was set to $P < 0.05$ (two-sided). All analyses were conducted using SPSS Statistics software (version 19.0; IBM).

Results

Descriptive characteristics for the ninety-five women in their third trimester of pregnancy are shown in Table 1. All women were fair-skinned and lived in western Sweden at latitudes 57–58°N. Their mean age was 32·2 years and 14 % of the women had BMI ≥ 25 kg/m² before becoming pregnant. The mean self-reported pre-pregnancy body weight was 64 kg and the mean body-weight gain until the third trimester was 13 kg. Parity ranged from 0 to 2 and half of the women were nulliparous. All women gave birth to full-term healthy babies with a mean birth weight of 3581 (SD 477) g and a mean birth length of 50 (SD 2) cm. Of these, 80 % had studied for three or more years at university. None of the women was current smokers and only one was using snuff.

The mean serum concentration of 25(OH)D was 47·4 (SD 18·1) nmol/l (range 10–93 nmol/l) (Fig. 1). Concentrations of serum 25(OH)D <30, <50 and <75 nmol/l were found in 17, 65 and 92 % of the women, respectively. During the winter, 85 % of women had serum 25(OH)D <50 nmol/l and 28 % had concentrations <30 nmol/l. However, during the summer, 41 % of women had 25(OH)D concentrations <50 nmol/l and only 2 % had <30 nmol/l. The mean serum concentration of 25(OH)D was >50 nmol/l only from June to September (Fig. 2). Serum concentrations of 25(OH)D were highest during the summer, with the highest mean in August

Table 1. Characteristics of the ninety-five pregnant women living in Sweden

(Mean values and 95 % confidence intervals)

| Characteristics	Mean	95 % CI
Age (years)	32·8	32·1, 33·5
Gestational week in the third trimester	35·7	35·5, 35·9
Height (cm)	168·9	167·6, 170·2
Weight before pregnancy (kg)	64·3	62·7, 65·9
Weight in the third trimester (kg)	77·3	75·3, 79·3
Weight gain at the third trimester (kg)	13	12·1, 13·9
BMI before pregnancy (kg/m²)	22·5	22·0, 23·0
Infant birth weight (g)	3581	3485, 3677
Infant birth length (cm)*	50	49, 52
Gestational length (d)†	281	276, 286

* n 89.
† n 90.
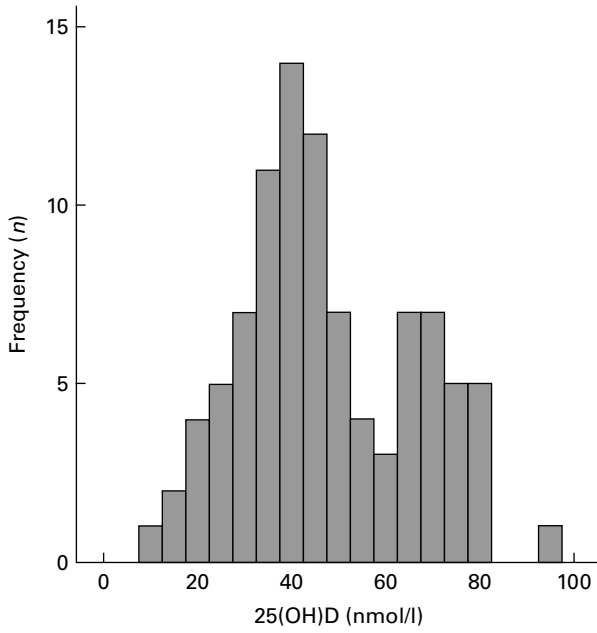
Fig. 1. Distribution of serum concentrations of 25-hydroxyvitamin D (25(OH)D) in the third trimester of pregnancy (n 95). Mean 47·4 (SD 18·1) nmol/l.

(69 nmol/l). The lowest concentrations were seen in late winter and April had the lowest mean (33 nmol/l). The mean serum concentration was 53 % higher in the summer compared with winter. The difference in serum 25(OH)D between summer and winter was highly significant ($P<0.001$).

The mean dietary intake of vitamin D was 6·1 µg/d and the mean total intake of vitamin D (from diet and supplements)

was 9·3 µg/d. Data from the FFQ on the dietary intake of vitamin D-rich foods are shown in Table 2. No association was seen between serum concentrations of 25(OH)D and dietary vitamin D intake or the intake of food rich in vitamin D, such as fatty fish or dairy products. A trend was seen between the intake of low-fat yogurt and sour milk and serum concentrations of 25(OH)D ($P=0.08$). More than half of the women (56 %) were taking supplements containing vitamin D, and for these women, the mean vitamin D supplement intake was 5·8 µg/d and the total vitamin D intake was 12·0 µg/d. The mean serum concentration of 25(OH)D was 46 % higher among women taking supplements containing vitamin D, compared with those who did not ($P<0.001$). A significant association was found between the total vitamin D intake (from diet and supplements) and serum concentrations of 25(OH)D ($P=0.008$). Body weight and BMI were not associated with serum 25(OH)D concentrations.

The mean self-estimated PAL was 1.6 (SD 0.2) (range 1·3–2·2). The mean food intake level (energy intake:BMR) was 1.3 (SD 0.2) (range 0.8–1.8). No association was seen between PAL and serum 25(OH)D. When validating energy intake v. PAL, 24 % of the subjects were identified as under-reporters.

Sun exposure and other lifestyle variables potentially related to 25(OH)D concentrations are shown in Table 3. Women who preferred to stay in the sun when outdoors during the summer had 21 % higher mean serum concentrations of 25(OH)D compared with women who preferred to stay in the shade or who preferred a mix of sun and shade ($P=0.03$). The median times spent outdoors during the summer were 2 h during weekdays and 4 h during week-
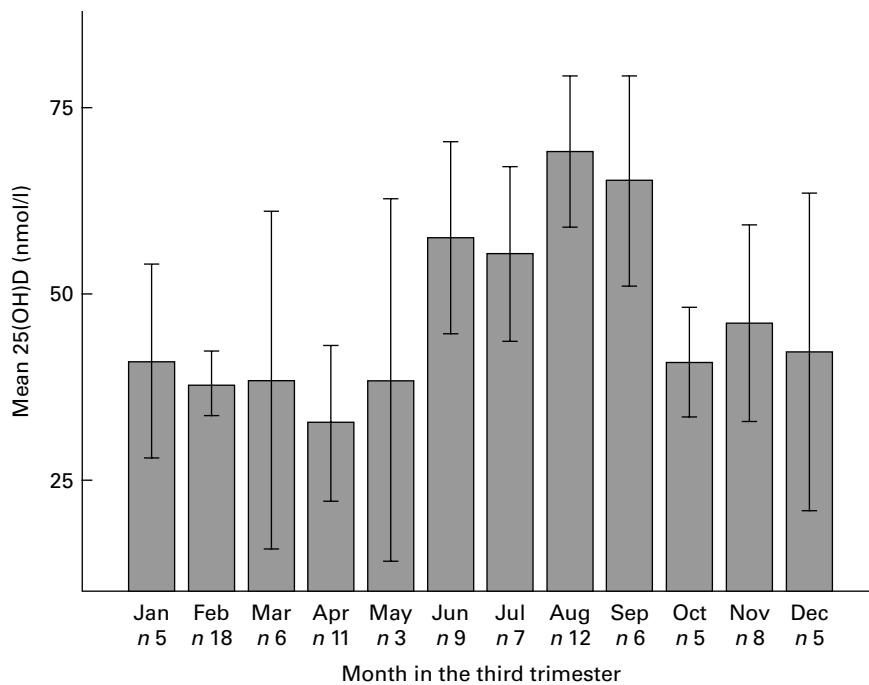

Fig. 2. Monthly mean 25-hydroxyvitamin D (25(OH)D) in the third trimester of pregnancy (n 95). Values are means, with 95 % CI represented by vertical bars. A significant difference was seen between the winter and summer months ($P<0.001$).

Table 2. Dietary vitamin D intake from the 4 d food record and the intake of vitamin D-rich foods and supplements from the FFQ

(Mean values and 95 % confidence intervals, n 95)

Dietary intake	Mean	95 % CI
Dietary vitamin D intake (μg/d)*	6·1	5·5, 6·7
Fatty fish (g/week)†	110	93, 127
Total milk intake (ml/d)†	220	181, 259
Low-fat milk (ml/d)†	197	161, 233
Total yogurt/sour milk intake (ml/d)†	168	145, 190
Low-fat yogurt/sour milk (ml/d)†	82	58, 106
Spread (g/d)†	18	14, 22
Vitamin D supplement intake (μg/d)†	3·3	2·6, 4·0
Vitamin D supplement intake (μg/d)†‡	5·8	5·0, 6·6

* From the 4 d food record.
† From the FFQ.
‡ Supplement users only (n 53).

ends. During the winter, the median time spent outdoors was 1 and 2 h, respectively. Serum 25(OH)D was not associated with the time spent outdoors, either during the summer or winter. In addition, no association was seen between serum 25(OH)D and skin type. However, subjects who more frequently used sunscreen tended to have higher serum 25(OH)D ($P=0.07$). Also, a positive relationship was found between time spent outdoors during the summer and the use of sunscreen ($P=0.013$ for non-working days and $P=0.031$ for working days). None of the women had used a sunbed in the last 6 months.

Of the subjects, 18 % had travelled to southern latitudes during the past 6 months. Mean serum concentration of 25(OH)D was 35 % higher in this group compared with women who had not travelled to southern latitudes ($P=0.001$). During the winter, 25 % of the women had neither recently travelled to southern latitudes nor taken vitamin D supplements. These women had a mean serum 25(OH)D of 30 nmol/l, which was significantly lower than for those who used vitamin D supplements and/or had travelled to southern latitudes ($P<0.001$).

The mean concentration of PTH was 43·8 (sd 15·6) ng/l. A significant inverse association was seen between serum concentrations of 25(OH)D and PTH ($P=0.008$, r −0·271; Table 4). Women with serum 25(OH)D <50 nmol/l had significantly higher serum PTH (47·1 (sd 16·1) ng/l), compared with women with serum 25(OH)D >50 nmol/l (37·7 (sd 12·9) ng/l) ($P=0.005$). The mean serum PTH was significantly higher during the winter than during the summer ($P=0.011$).

No association was seen between the maternal serum concentration of 25(OH)D and infant birth weight or birth length. However, a negative relationship was seen between PTH and birth weight ($P=0.03$; $\beta = -6.06$ (sd 2·72)). A significant positive association was found between serum 25(OH)D and maternal height ($P=0.010$), parity ($P=0.050$) and sex of the baby, where mothers giving birth to boys had higher serum 25(OH)D ($P=0.006$).

Multivariate regression analyses showed that the major factors determining the concentration of 25(OH)D were season, use of vitamin D supplements and travels to southern latitudes. Together, these factors explained 51 % of the variation in serum 25(OH)D (Table 5).

Discussion

The strengths of the present study are the wide range of possible determinants of vitamin D status studied, including detailed measures of vitamin D intake from diet and supplements separately, several estimates of sun exposure and investigations of seasonal variation in 25(OH)D. Previous studies in pregnant women have found an association between serum concentrations of 25(OH)D and season[34–36,38,43], ethnicity or skin type[34–36,38,44], total vitamin D intake[34], supplement use[36,38,41], education[38], smoking and alcohol use[38], and sun exposure[34,36,38,43]. None of these studies included, however, measurements of vitamin D intake from diet and supplements separately, and only a Belgian national survey included different estimates of sun exposure[38,43].

The mean 25(OH)D in the present study was lower than means reported previously in fair-skinned pregnant women at similar latitudes, e.g. Sweden[47] and Denmark[42], and similar to those reported in Caucasian Belgian pregnant women[38,43]. Despite similar latitudes and ethnicity, differences in vitamin D status between studies may depend on trimester, season of blood sampling and the method used for 25(OH)D analysis. It must be remembered that there is no 'gold standard' for measuring 25(OH)D. The chemiluminescence immunoassay used in the present study has been shown to give lower serum concentrations of 25(OH)D, compared with high-pressure liquid chromatography–atmospheric pressure chemical ionisation-MS[55].

Table 3. Lifestyle factors and 25-hydroxyvitamin D (25(OH)D) concentrations of the ninety-five pregnant women during the third trimester, living in Sweden

(Mean values, number of subjects and percentages)

Lifestyle factors	n	%	Mean 25(OH)D (nmol/l)
Parity			
0	49	52	50·8
1	35	37	44·5
2	11	12	41·2
Sunscreen use			
Never	4	4	30·7
Sometimes	58	62	46·9
Always	32	33	50·8
Preference of sun or shade			
Sun	22	23	54·8*
Shade or sun and shade	72	76	45·3
Skin type†			
I	3	3	45·0
II	18	19	43·6
III	64	67	48·1
IV	10	11	50·6
Travels to southern latitudes‡			
Yes	17	18	60·2**
No	78	82	44·6
Use of vitamin D supplements			
Yes	53	56	55·1***
No	42	44	37·7

Mean values were significantly different from the rest of the group: * $P<0.05$, ** $P<0.01$, *** $P<0.001$.
† I, always burns, never tans; II, usually burns, tans with difficulty; III, sometimes burns mildly, tans gradually; IV, rarely burns, tans easily.
‡ Travels to latitude 35°N or below, during the last 6 months before the measurements.

Table 4. Parathyroid hormone (PTH) concentrations related to the different levels of 25-hydroxyvitamin D (25(OH)D) of the ninety-five pregnant women during the third trimester, living in Sweden (Number of subjects, percentages and 95 % confidence intervals)

25(OH)D (nmol/l)	n	%	PTH (ng/l)	95 % CI
<30	16	17	49·4	41·1, 57·7
30−49·9	46	48	46·3	41·6, 51·0
≥50	33	35	37·7	33·3, 42·1
Total	95	100	43·8	40·7, 46·9

Sunscreen use tended to be positively associated with serum 25(OH)D. This is probably explained by the positive association also found between sun preference and serum 25(OH)D and the finding that women who spent more time outdoors during the summer were significantly more frequent sunscreen users. Sunscreen use, therefore, seems to rather reflect time spent outdoors in the sun than its inhibiting effect on endogenous vitamin D production. Possibly, a higher number of subjects would give more power to find a significant positive relationship between sunscreen use and serum 25(OH)D, especially since the Belgian national survey also reported that women using sunscreen lotion had a significant lower risk of severe vitamin D deficiency[38].

During the winter, UVB-mediated production of vitamin D is absent at northern latitudes[25]. Accordingly, a majority of the women in the present study had serum 25(OH)D <50 nmol/l during this period. Additionally, women who had travelled to southern latitudes below latitude 35°N within the previous 6 months or preferred to stay in the sun in the summertime had significantly higher concentrations of 25(OH)D. This confirms similar findings in the Belgian survey in pregnant women[38]. A study of Swedish elderly women also found similar relationships[32].

Both the mean dietary intake of vitamin D (6·1 µg/d) and the mean total intake of vitamin D (9·3 µg/d; from diet and supplements) in the present study were lower than the recommended daily intake from the Swedish National Food Agency and the Nordic Nutrition Recommendations (10 µg/d for pregnant women)[56,57] and the Institute of Medicine from 2010 (15 µg/d for adults, including pregnant women)[58]. However, the mean dietary intake in the present study was similar to that reported for women of the same age group of 6·2 µg/d in the new Swedish national survey from 2011[24]. The mean total intake of vitamin D in the present study was somewhat higher than that reported for adult women at all ages (mean 7·0 µg/d including supplements)[59]. This may partly be explained by the fact that the women in the present study were in the third trimester of pregnancy and may have a higher energy intake than non-pregnant and non-lactating women. When adding vitamin D intake from diet and supplements, only 39 % of the women in the present study had a total vitamin D intake of ≥10 µg/d and only 12 % ≥15 µg/d. However, when considering vitamin D intake from diet alone, only 9 % had a vitamin D intake of ≥10 µg/d and only 1 % ≥15 µg/d.

The significant association found between the total intake of vitamin D and serum 25(OH)D is in agreement with previous studies in pregnancy[34,36,60]. This association was mainly explained by the strong association between vitamin D supplement intake and 25(OH)D concentrations. A trend was, however, seen between the intake of reduced-fat yogurt and sour milk and concentrations of 25(OH)D.

Of the total subjects, 24 % under-reported energy intake, and may potentially have led to under-reporting of dietary vitamin D intake as well. No attempt was made to correct under-reporting. However, removal of under-reporters from data analyses did not change the significance of the results. Also, when under-reporters were removed from data analyses, no significant association was seen between the dietary intake of vitamin D, or food items rich in vitamin D, and serum 25(OH)D. The percentage of women under-reporting energy intake is consistent with the results from earlier studies of pregnant women. For example, we have previously shown the percentage of under-reporting among Indonesian women during the third trimester to be 17·6 %[61], whereas an Irish study found the proportion of under-reporting in gestational week 14 to be up to 45 %[62]. Both studies found high BMI and low education to be important predictors for under-reporting[61,62].

One limitation of the present study is that the women included may not be representative for the whole Swedish population of pregnant women. Of these subjects in the present study, 80 % had three or more years of education at university level, which is a higher number than among women in the same age group in the general population

Table 5. Factors predicting the concentrations of 25-hydroxyvitamin D of the ninety-five pregnant women during the third trimester, living in Sweden

Factors	Bivariate model				Multivariate model			
	β	SEM	P	R²	β	SEM	P	R²
Season*	19·40	3·19	0·000	0·29	16·25	2·76	0·000	0·51
Vitamin D supplement†	17·34	3·31	0·000	0·23	14·06	2·76	0·000	
Travels to southern latitudes‡	15·57	4·61	0·001	0·11	10·17	3·55	0·005	
Sun preference§	9·50	4·34	0·031	0·05	4·26	3·23	0·190	

Coding for each variable is shown below within parentheses.
* (1) Winter (November−April) or (2) summer (May−October).
† (1) No or (2) yes.
‡ Travels to latitude 35°N or below, during the last 6 months; (1) no or (2) yes.
§ (1) Shade or sun and shade, or (2) sun.

(37 %)[63]. Additionally, only 14 % had a BMI $\geq 25 \text{ kg/m}^2$ before entering pregnancy, compared with 37 % of the pregnant women in the same region[64]. Thus, the women in the present study were leaner, highly educated and, possibly, more health conscious than pregnant women in general. The interpretation of these results needs therefore to be made with some caution. However, despite the high education and normal body weight, the majority of the subjects had concentrations of 25(OH)D < 50 nmol/l. Thus, it may be speculated that even lower concentrations of 25(OH)D are expected in the general population of pregnant women living in Sweden, especially in pregnant women with high pigmentation and those wearing concealing clothing. The Belgian national survey showed that vitamin D deficiency was three- to sixfold higher among women of Asian, African or Hispanic descent when compared with Caucasians[38]. More targeted screening surveys on vitamin D status in pregnant women in Sweden and in other countries are needed to confirm this.

At present, there are no general recommendations for optimal vitamin D levels. According to the latest guidelines from the Institute of Medicine, serum concentrations of 25(OH)D ≥ 50 nmol/l are recommended[58]. The recommendation is based on the importance of vitamin D for bone health[58]. Among non-pregnant adults, a negative relationship is seen between serum PTH and serum 25(OH)D[65–67], and that above 50 nmol/l of serum 25(OH)D, there is no further increase in serum PTH[66]. We also found that serum PTH was significantly higher at serum concentrations of 25(OH)D below 50 nmol/l. A weaker association between serum PTH and serum 25(OH)D has previously been found among pregnant women[22,23,40,68]. A seasonal variation in serum PTH, with highest serum concentrations of PTH during the winter, was also found. This is in agreement with a Danish study of non-pregnant adults[30]. However, whether PTH is a marker for determining adequate vitamin D status during pregnancy needs further evaluation. A specific cut-off for serum 25(OH)D may be needed for pregnant women, due to the plasma volume expansion during pregnancy[69] and possible health benefits of maternal vitamin D status for the mother, fetus and child[70]. However, more research is needed to evaluate the importance of vitamin D status during pregnancy for such outcomes[70] and to determine what serum levels of 25(OH)D should be considered as appropriate during pregnancy.

There is an ongoing discussion on whether or not to recommend supplementation of vitamin D to all pregnant women[70]. It is not clear what dose of vitamin D is required during pregnancy to have an effect on maternal or paediatric outcome. Supplementation with 25 µg/d may be needed to maintain maternal serum concentrations of at least 50 nmol/l[70]. This corresponds with the findings of Cashman *et al.*[33], where a daily intake of 28·0 µg for non-pregnant adults was needed to maintain serum 25(OH)D concentrations > 50 nmol/l during the winter.

In conclusion, during the winter, the majority of fair-skinned pregnant women had serum 25(OH)D < 50 nmol/l in their third trimester, at latitudes 57–58°N. In addition, more than every fourth women was vitamin D deficient during the winter. The main determinants for vitamin D status were season, use of vitamin D supplements and travels to southern latitudes. Thus, also fair-skinned pregnant women at northern latitudes are at risk of vitamin D deficiency, especially during the winter. Higher vitamin D intake may therefore be needed during the winter for pregnant women at these latitudes to maintain 25(OH)D serum concentrations > 50 nmol/l and to avoid maternal vitamin D deficiency.

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