Vitamin D in pregnancy at high latitude in Scotland†

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Abstract
The aims of the present study were to determine compliance with current advice on vitamin D and to assess the influence of season, dietary intake, supplement use and deprivation on vitamin D status in pregnant mothers and newborns in the north of Scotland where sunlight exposure is low. Pregnant women (n 1205) and their singleton newborns were studied in the Aberdeen Maternity Hospital (latitude 57°8'N) between 2000 and 2006. Plasma 25-hydroxyvitamin D2 and 25-hydroxyvitamin D3 were measured at 19 weeks of gestation in mothers and at delivery in newborns. During pregnancy, 21·0 (95 % CI 18·5, 23·5) % of women took vitamin D supplements. The median intake was 5 mg/d and only 0·6 (95 % CI 0·1, 1·0) % took the recommended 10 mg/d. Supplement use, adjusted for season, dietary intake and deprivation, significantly increased maternal 25-hydroxyvitamin D (25(OH)D) by 10·5 (95 % CI 5·7, 15·2) nmol/l (P,0·001); however, there was no significant effect on cord 25(OH)D (1·4 (95 % CI 2·1, 4·5) nmol/l). The biggest influence on both maternal and cord 25(OH)D was season of birth (P<0·001). Compared with the least deprived women (top three deciles), the most deprived pregnancies (bottom three deciles) were characterised by a significantly lower seasonally adjusted 25(OH)D (211·6 (95 % CI 27·5, 215·7) nmol/l in the mother and 25·8 (95 % CI 22·3, 29·4) nmol/l in the cord), and a lower level of supplement use (10 (95 % CI 4, 17) v. 23 (95 % CI 20, 26) %). More should be done to promote vitamin D supplement use in pregnancy but the critical importance of endogenous vitamin D synthesis, and known adaptations of fat metabolism specific to pregnancy, suggest that safe sun advice may be a useful additional strategy, even at high latitude.

Key words: Pregnancy; Supplements; Latitude; Vitamin D

Pregnant and breast-feeding women in the UK are currently advised to take 10 μg of vitamin D daily in supplement form in order to protect against vitamin D deficiency(1–3). Some have argued that higher intakes may be necessary(4,5), while others such as the National Institute for Clinical Excellence have questioned the evidence base for these recommendations(5,6). The Institute of Medicine recently set a RDA for pregnancy and lactation of 15 μg/d for the USA and Canada(7). The UK Scientific Advisory Committee on Nutrition is currently undertaking a review of vitamin D requirements and recommendations. Concerns have been raised that even the current recommendations may be being overlooked by health professionals and the general public(8).

Vitamin D can only be considered as conditionally essential in the diet, as it can be synthesised by skin exposed to solar radiation of the appropriate wavelength. Indeed, it is an assumption in current UK recommendations that most people obtain the majority of their vitamin D by exposure of skin to sunlight(1–3). During winter months in the UK, UV sunlight exposure of the required wavelength is not sufficient to support vitamin D synthesis and the duration of this fallow period increases with increasing latitude(3,7–9). The critical importance of latitude and climate on sunlight exposure and vitamin D synthesis has led to concern in Scotland over the vitamin D status of the population in general and pregnant women in particular. A further concern for Scotland is the

Abbreviation: 25(OH)D, 25-hydroxyvitamin D.

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†The paper is dedicated to Professor A. Mike Wallace who sadly died during the preparation of the manuscript.
higher level of socio-economic deprivation compared with the rest of the UK, as deprivation is itself associated with poor vitamin D status\(^{10,11}\).

There is currently no national information on the vitamin D status of pregnant women\(^{3}\) and a notable absence of data at higher latitudes in the UK where the dietary need is likely to be greatest. The aims of the present study were to determine vitamin D status in mothers and newborns (cord blood) in a large population in the north-east of Scotland (latitude 57°N). We evaluated the influence of season, vitamin D intake from the diet and supplements, compliance with current advice on supplement use, and the modulating effect of deprivation.

Materials and methods

We carried out a prospective cohort study of pregnancies in Aberdeen, Scotland (latitude 57°N) between 2000 and 2006. The present study was conducted according to the guidelines laid down in the Declaration of Helsinki and all procedures involving human subjects/patients were approved by the Grampian Research Ethics Committee. Written informed consent was obtained from all subjects. Eligible women were enrolled sequentially at the Aberdeen Maternity Hospital. Information on each pregnancy was abstracted from the Aberdeen Maternity and Neonatal Databank and the data anonymised before linking to the dietary and laboratory results obtained from all subjects. Eligible women were enrolled sequentially at the Aberdeen Maternity Hospital. Information on each pregnancy was abstracted from the Aberdeen Maternity and Neonatal Databank and the data anonymised before linking to the dietary and laboratory results obtained from all subjects. Eligible women were enrolled sequentially at the Aberdeen Maternity Hospital. Information on each pregnancy was abstracted from the Aberdeen Maternity and Neonatal Databank and the data anonymised before linking to the dietary and laboratory results obtained from all subjects.

Statistical analysis

Statistical analysis was carried out using STATA/SE version 11 (Stata Corporation) and Genstat (VSN International). The primary response variable was the log-transformed plasma 25(OH)D concentration in the mother and cord blood (to ensure approximate normal distributions). We were interested in whether the time-of-year effect followed the natural approximately sinusoidal pattern in the amount and strength of solar radiation, so sinusoidal variation was assessed by taking the cosine and sine of the month (1–12) the maternal sample was taken or the month of birth, scaled to the range (0, 2\(\pi\)). The sine term (phase) was not significant in any of the models and was dropped from the analysis. For results grouped by season, the following commonly accepted categories were used: winter (December, January, February); spring (March, April, May); summer (June, July, August); autumn (September, October, November). Vitamin D supplement use was coded as 1 (consumption recorded at any stage) or 0 (no consumption recorded at any stage). Separate analysis of supplement use (any \(v\). none) and the reported intake (\(\mu g/d\)) was carried out by the stage of pregnancy. Deprivation was assessed using the Scottish Index of Multiple Deprivation 2004\(^{16}\).

This metric provides a comprehensive picture of multiple deprivations by area (UK postcode) based on thirty-one indicators which describe various aspects of deprivation including current income, employment, housing, health, education, skills and training, and geographic access to services and telecommunications. The deciles are based on the whole population. Because the proportion of women in the most deprived deciles was relatively small, pregnancies were grouped into three categories (deciles 1–3, 4–7 and 8–10). Smoking habits were also recorded, but these had no independent effect on 25(OH)D concentrations. Factors potentially affecting vitamin D status were first examined individually by ANOVA if discrete or by regression if continuous, thereby
ignoring the potential confounding effect of other factors. They were then examined in a full, single-stratum linear model incorporating the effects of all factors, so that the effect uniquely attributable to each factor could also be estimated and tested. Multiple linear regression was used for continuous dependent variables and logistic regression for binary outcomes.

**Results**

We studied 1205 pregnant women and their singleton newborns. The number of non-Caucasian (African, Asian and Indian) mothers recruited to the study was small (n 42, 3% of the study population). Plasma 25(OH)D was compared with non-Caucasian pregnancies but no further analysis was undertaken as low power leading to non-significant effects could be misleading. For non-Caucasian mothers, the characteristics were age at delivery (31·5 (SD 5·0) years), height (160·7 (SD 6·2) cm), gestational age at delivery (38·8 (SD 1·8) weeks) and 43% primiparous. The birth weight was 3183 (SD 563) g in non-Caucasian pregnancies and 57% of babies were female. The characteristics of Caucasian pregnancies are shown in Table 1.

There was some variation in the numbers sampled in each season: 25, 34, 25 and 16% for maternal blood and 18, 19, 28 and 35% for cord blood in winter, spring, summer and autumn, respectively. However, deviation from equal sampling was sufficiently small that adjustment for this made no significant difference to any of the summary statistics.

The mean dietary intake of vitamin D in pregnancy was 3·7 (95% CI 3·6, 3·8) μg/d in all women. The mean intake of vitamin D in women taking supplements was 10·7 (95% CI 8·8, 12·5), 16·4 (95% CI 14·1, 18·6) and 13·7 (95% CI 11·6, 15·8) %, respectively. The corresponding percentage of taking supplements in these three periods was female. The characteristics of Caucasian pregnancies were age at delivery (30·7 (SD 5·3) years), height (164 (SD 7) cm), gestational age at delivery (39·3 (SD 5·3) weeks) and 50 (95% CI 46, 54) % of women exhibiting vitamin D levels below 25 nmol/l, commonly used as the threshold for deficiency. The geometric mean concentration in cord blood at delivery was 46% lower than that in the mother at 19 weeks of gestation and 50 (95% CI 46, 54) % of cord blood was below the 25 nmol/l cut-off. In 3% of the maternal samples and 13% of the fetal samples, plasma 25(OH)D was below the detection limit of the assay (4 nmol/l). The highest 25(OH)D values were seen in summer (53 nmol/l). The lowest 25(OH)D concentrations (34 nmol/l), and the greatest proportion of cases below the 25 nmol/l cut-off, were observed in autumn (27%) and winter (28%) when sunlight exposure is lowest.

Univariate regression analysis of the effect of the factors thought to influence vitamin D status showed significant effects on maternal 25(OH)D of time of year (cosine coefficient −0·252; 95% CI −0·308, −0·197; P<0·001), intake from the diet (coefficient 0·022; 95% CI 0·002, 0·041; P=0·030), supplement use (coefficient 0·268; 95% CI 0·166, 0·371; P<0·001) and deprivation (coefficient −0·151; 95% CI −0·213, −0·088; P<0·001). There was no significant effect of body weight or BMI at booking. The corresponding univariate analysis in cord plasma 25(OH)D also showed a significant effect of time of year (cosine coefficient −0·519; 95% CI −0·611, −0·427; P<0·001) and deprivation (coefficient −0·149 95% CI −0·254, −0·044; P=0·005). Again, there was no significant effect of body weight or BMI at booking. In contrast to the maternal plasma results, neither the dietary intake of vitamin D or supplement use by the mother had a significant effect on the newborn plasma 25(OH)D. Maternal plasma 25(OH)D was an important determinant of the 25(OH)D of the newborn (coefficient 0·376; 95% CI 0·284, 0·468; P<0·001) despite being sampled in different seasons.

Multiple regression analysis of the factors influencing plasma 25(OH)D in the mother and newborn is summarised in Table 3. The maternal model included the influence of time of year (cosine fit), vitamin D intake from the diet, the use of vitamin D-containing supplements, and the level of deprivation. The same parameters were fitted to cord 25(OH)D, but this model also included maternal plasma 25(OH)D.

Most of the variation in cord plasma 25(OH)D was accounted for by time of year, following a smooth sinusoidal path (Fig. 1). The newborn plasma 25(OH)D curve minima, adjusted for all the parameters in the regression model, corresponded to the solar nadir in December to January (31·7; 95% CI 30·4, 33·1 nmol/l). The curve maxima corresponded to the zenith in June to July (38·5; 95% CI 37·4, 39·8 nmol/l). The maximum-to-minimum difference in the newborn was 26·8 (95% CI 22·4, 31·3) nmol/l. There was also a strong annual variation in maternal 25(OH)D status (Fig. 1). The fully adjusted maternal plasma 25(OH)D curve minima in December to January was 31·3 (95% CI 29·1, 33·6) nmol/l and the maxima in June to July was 51·7 (95% CI 48·3, 55·2) nmol/l. The maximum-to-minimum difference in maternal plasma was 20·4 (95% CI 15·7, 25·1) nmol/l. The cosine fit was again highly significant, though it can be seen from the

**Table 1. Subject characteristics**

<table>
<thead>
<tr>
<th>Mean (SD)</th>
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<tbody>
<tr>
<td>Age at delivery (years)</td>
</tr>
<tr>
<td>Height (cm)</td>
</tr>
<tr>
<td>Primiparous (%)</td>
</tr>
<tr>
<td>Baby sex (% female)</td>
</tr>
<tr>
<td>Gestational age at delivery (weeks)</td>
</tr>
<tr>
<td>Birth weight (g)</td>
</tr>
</tbody>
</table>
### Table 2. Vitamin D intake from the diet, supplement use and maternal* and cord 25-hydroxyvitamin D (25(OH)D) concentrations by season† (Mean values and 95 % confidence intervals)

<table>
<thead>
<tr>
<th>Season</th>
<th>Vitamin D intake</th>
<th>Maternal plasma 25(OH)D</th>
<th>Cord plasma 25(OH)D</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Dietary intake (µg/d)</td>
<td>Supplement use (%)</td>
<td>nmol/l</td>
</tr>
<tr>
<td></td>
<td>Arithmetic mean</td>
<td>95 % CI</td>
<td>Arithmetic mean</td>
</tr>
<tr>
<td>All seasons</td>
<td>3.74</td>
<td>3.63, 3.86</td>
<td>21.0</td>
</tr>
<tr>
<td>Winter</td>
<td>3.69&lt;sup&gt;a&lt;/sup&gt;</td>
<td>3.46, 3.92</td>
<td>19.4&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>Spring</td>
<td>3.81&lt;sup&gt;b&lt;/sup&gt;</td>
<td>3.57, 4.05</td>
<td>22.3&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>Summer</td>
<td>3.91&lt;sup&gt;c&lt;/sup&gt;</td>
<td>3.64, 4.17</td>
<td>20.6&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>Autumn</td>
<td>4.03&lt;sup&gt;d&lt;/sup&gt;</td>
<td>3.65, 4.40</td>
<td>21.6&lt;sup&gt;d&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

<sup>a,b,c,d</sup> Mean values within a column with unlike superscript letters were significantly different (P<0.05).

* Maternal blood was sampled at 19 weeks of gestation.
† Excluding non-Caucasian mothers.
‡ Winter (December, January, February); spring (March, April, May); summer (June, July, August); autumn (September, October, November).
Women in the UK are advised to take 10 μg of vitamin D daily in supplement form throughout pregnancy in order to avoid deficiency. Only 21% of women in the present study population in the north of Scotland reported taking vitamin D supplements compared with 15% in pregnant women in the south of England (Southampton). However, the true picture is worse than even this low percentage suggests. Only about 1% of women took the recommended 10 μg/d or more at any stage in pregnancy (the value was 3% in Southampton), with the great majority taking 5 μg/d (the most commonly available dosage). The use of supplements was associated with a higher maternal circulating concentration of 25(OH)D, though the effect was small. Supplement use is confounded by a number of factors such as deprivation, and full adjustment showed that supplement use increased maternal plasma 25(OH)D by 10.5 nmol/l. However, there was no detectable effect (adjusted or unadjusted) on the more important cord plasma 25(OH)D at birth. This reinforces concerns over the effectiveness of public health policy in promoting supplement use, though there was no evidence that compliance in the north of Scotland was significantly worse than elsewhere in the UK. The correlation between the intake of vitamin D from the diet and plasma 25(OH)D was weak in the mother and absent in the newborn, and there was no evidence that the dietary intake of vitamin D was low compared with non-pregnant women in the rest of the UK. The fact that the D2 form was undetectable in any of the maternal or cord blood analysed reinforces the conclusion that supplements and the diet made a minor contribution to vitamin D status in these pregnancies.

The plasma 25(OH)D value observed in these pregnant women in the north of Scotland (latitude 57°12′N) was about 10 nmol/l lower than that observed in pregnant women in the south of England (latitude 50°57′N). In the Southampton study, 18% of pregnant women were classed as deficient based on a cut-off of 27.5 nmol/l. Application of the same cut-off to Aberdeen pregnancies gave a deficiency prevalence of 29% (21% using the more commonly cited deficiency cut-off of 25 nmol/l). Sampling in the Southampton study was carried out later in pregnancy, but there is little evidence for a consistent change in plasma 25(OH)D levels across pregnancy, and a recent review suggested that plasma 25(OH)D levels do not change unless intake or synthesis changes. There are published values for 25(OH)D deficiency in pregnant women in Northern Ireland but comparison with that cohort is difficult as the sampling was confounded by season. There was a strong seasonal variation in vitamin D levels in the mother and newborn. Cord blood vitamin D followed a classic sinusoidal pattern which mirrors the annual variation in UVB exposure at this latitude. Maternal blood was not sampled at delivery but we would expect the seasonal variation in cord 25(OH)D to reflect maternal plasma at this stage of pregnancy. Maternal plasma, sampled in mid-gestation, also demonstrated a strong seasonal variation, but there was a significant deviation from the sinusoidal pattern at this stage of pregnancy. For the 8 months of the year when UVB exposure is lowest, there was little variation in maternal 25(OH)D but a step change to a higher level in May and down again in September. Winter and spring values are generally low in the UK, but the rapid post-summer drop in plasma 25(OH)D observed here at high latitude in mid-gestation would be consistent with a greater rate of vitamin D sequestration into adipose tissue resulting from the high rate of maternal fat deposition at this stage of pregnancy; approximately 160 g/week.

The absence of a significant response in cord 25(OH)D to vitamin D supplement use in these women contrasts with the findings of a large (n > 1000) intervention study in women from a similar ethnic background and latitude (Edinburgh, Scotland; latitude 55°57′N). Supplementation with vitamin D at the recommended level of 10 μg/d from week 12 of gestation to term increased maternal plasma 25(OH)D by about 10 nmol/l at delivery, cord plasma by 8 nmol/l, and neonate plasma at 6 d post-partum by 14 nmol/l. If all the women in the present Aberdeen study who did not consume supplements followed current advice, and achieved the effect...
observed in the Cockburn trial, the prevalence of deficiency in the north of Scotland would be reduced by two-thirds (from 23 to 8%) and the observed difference in plasma 25(OH)D between the north of Scotland and the south of England (17) would be abolished.

The Cockburn trial (24) shows that supplement use at the recommended levels works, albeit with relatively modest effects on status. Therefore efforts to improve the uptake of current vitamin D advice in pregnancy should continue but a number of factors are likely to work against the effectiveness of this strategy. The need to consume 10 µg of supplemental vitamin D every day throughout pregnancy is demanding and the way the supplements were actually consumed in practice here was insufficient to influence the newborn 25(OH)D in spite of there being a detectable effect on maternal status. Also, even if every pregnant woman in the present study followed the advice fully, about 8% would still be deficient in vitamin D. Furthermore, women most likely to benefit from vitamin D supplement use are typically the most disadvantaged in the society who are also less likely to follow the advice on supplement use in pregnancy (12). This is borne out by the data here for vitamin D supplement use. Non-pregnant women from deprived households have lower intakes of vitamin D (10) and lower concentrations of plasma 25(OH)D (11), and the data presented here suggest that this also holds true for pregnancy. However, they appear to have a further disadvantage with respect to vitamin D. Even after adjustment for dietary intake and supplement use, there remains a residual effect of deprivation on 25(OH)D concentrations in the mother and newborn reflecting cutaneous synthesis. The prevalence of deficiency in non-pregnant women in Aberdeen was much reduced if they reported a recent holiday abroad (25). Financial constraints clearly limit opportunities to holiday abroad in countries with more intense sunlight but, even within the UK, there is a strong social gradient in the frequency of use of green spaces (26), with likely knock-on effects on exposure to sunlight.

Fig. 1. Seasonal variation in log-transformed 25-hydroxyvitamin D (25(OH)D) in maternal blood at 19 weeks of gestation (a) and in cord blood at delivery (b). The maternal data are shown with the cosine fitted curve (—) together with the step change (-----) between two distinct concentrations of maternal 25(OH)D. The newborn log 25(OH)D concentration is shown with the cosine fitted curve (--). Values are means at each month, with 95% CI represented by vertical bars.

Fig. 2. Deprivation category and dietary intake of vitamin D, frequency of supplement use, and maternal and cord plasma 25-hydroxyvitamin D (25(OH)D). Values are means, with 95% CI represented by vertical bars. Deprivation category was based on the Scottish Index of Multiple Deprivation (SIMD), with population deciles grouped into deciles 1–3, 4–7 and 8–10. Maternal blood was sampled at 19 weeks of gestation.
Multiple lines of evidence point to the overwhelming importance of sunlight in determining vitamin D status in pregnancy, even in the north of Scotland. At high latitude, the period during which cutaneous synthesis is possible may be very limited; therefore, efforts to improve vitamin D status through sunlight exposure critically depend on the effectiveness of mechanisms to store vitamin D synthesised in some parts of the year and mobilise it in others. Storage of vitamin D in adipose tissue is thought to be more efficient for endogenously synthesised rather than ingested vitamin D(27,28). Body fatness increases the sequestration of vitamin D into adipose tissue and significant fat mobilisation is required to maximise the availability of stored vitamin D(27,29–31). Seasonal cycles of fat storage in summer, when vitamin D is being synthesised, and fat mobilisation in winter, when endogenous vitamin D stores are needed, have occurred throughout much of human evolutionary history(32,33). Modern humans in industrialised societies typically no longer experience such seasonal variation, or other sustained periods of fat mobilisation, but pregnancy is a notable exception. During the first two trimesters of pregnancy, the mother deposits about 3.5 kg of fat(23). The very high rate of mobilisation of maternal adipose tissue stores that occurs in the last trimester has the effect of making available to the placenta, and ultimately to the fetus; a significant proportion of the nutrients stored in maternal adipose tissue(25) during the developmental period when Ca deposition in the fetal skeleton is maximal and vitamin D needs are greatest(23). The critical importance of maternal stores extends through to lactation as the vitamin D status of the infant appears to be more influenced by the vitamin D status of the mother during pregnancy than by maternal vitamin D status during lactation when mobilisation continues(34). Such mechanisms have helped humans to reproduce successfully at high latitude while coping with the lottery of the particular sequence of seasons experienced in each pregnancy. Mobilisation in late pregnancy of maternal vitamin D stores accumulated over extended periods, and possibly more than one summer, could also ameliorate the effect of a diet chronically poor in vitamin D. It may also provide a biological basis for possible additional public health measures to augment the current strategy of supplementation.

Promoting safe sun exposure and access to green space in summer has the potential to have a significant impact on vitamin D status in pregnancy, even in the north of Scotland. Such a policy could also have wider health benefits as populations that are exposed to the greenest environments also experience the lowest levels of health inequality related to deprivation(35). We propose that a trial of safe sun advice in the critical summer months at high latitude be carried out to assess the impact on the vitamin D status of the mother during pregnancy and lactation, and the newborn and neonate in pregnancies commencing in different seasons.

We excluded non-white subjects from much of the analysis as the numbers were too small to draw any meaningful conclusions. However, it should be noted that plasma 25(OH)D in non-Caucasians was significantly lower in the maternal and cord blood and the likelihood is that all of the above considerations will be exacerbated in women with darker skin and poorer UV absorption, where the need for improved vitamin D status is greater.

Acknowledgements

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