**Determinants of the maternal 25-hydroxyvitamin D response to vitamin D supplementation during pregnancy**

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**Abstract**

*Context*

Current approaches to antenatal vitamin D supplementation do not account for inter-individual differences in 25-hydroxyvitamin D [25(OH)D] response.

*Objective*

We assessed which maternal and environmental characteristics were associated with 25-hydroxyvitamin D [25(OH)D] following supplementation with cholecalciferol.

*Design*

Within-randomisation-group analysis of participants in the MAVIDOS trial of vitamin D supplementation in pregnancy.

*Setting*

Hospital antenatal clinics

*Participants*

829 pregnant women (422 placebo, 407 cholecalciferol). At 14 and 34 weeks’ gestation, maternal anthropometry, health and lifestyle were assessed and 25(OH)D measured. Compliance was determined using pill counts at 19 and 34 weeks.

*Interventions*

1000IU/day cholecalciferol or matched placebo from 14 weeks’ gestation until delivery.

*Main outcome measure*

25-(OH)D at 34 weeks, measured in a single batch (Diasorin Liaison).

*Results*

25(OH)D at 34 weeks’ gestation was higher in the women randomised to vitamin D [mean (SD): 67.7 (21.3) nmol/l] compared with placebo [43.1 (22.5) nmol/l, p<0.001]. In women randomised to cholecalciferol, higher pregnancy weight gain from 14 to 34 weeks’ gestation [kg] (β=-0.81 (95%CI -1.39, -0.22)), lower compliance with study medication [%] (β=-0.28 (-0.072, -0.48)), lower early pregnancy 25(OH)D [nmol/l] (β=0.28 (0.16, 0.40)) and delivery in the winter vs the summer (β=-10.5 (-6.4, -14.6)) were independently associated with lower 25(OH)D at 34 weeks’ gestation.

*Conclusions*

Women who gained more weight during pregnancy, had lower 25(OH)D in early pregnancy and delivered in winter achieved a lower 25(OH)D in late pregnancy when supplemented with 1000 IU/day cholecalciferol. Future studies should aim to determine appropriate doses to enable consistent repletion of 25-hydroxyvitamin D during pregnancy.

**Keywords:** Vitamin D; pregnancy; osteoporosis; epidemiology; supplementation

**Background**

Maternal vitamin D insufficiency during pregnancy is common ([1](#_ENREF_1), [2](#_ENREF_2)), and there is evidence that this might have detrimental effects on maternal health, fetal development ([3](#_ENREF_3), [4](#_ENREF_4)) and the long-term skeletal health of children ([1](#_ENREF_1), [3](#_ENREF_3)). Severe maternal vitamin D deficiency during pregnancy can result in symptomatic hypocalcaemia in the neonate ([3](#_ENREF_3)). Associations have been reported between maternal 25-hydroxyvitamin D [25(OH)D] and obstetric complications, including pre-eclampsia, gestational diabetes, preterm birth and offspring anthropometry, although the findings are inconsistent ([3](#_ENREF_3), [4](#_ENREF_4)), and require confirmation in randomised controlled trials. Nonetheless, the Institute of Medicine (IOM) has suggested that risk of vitamin D insufficiency, defined as a 25(OH)D<50nmol/l, should be avoided during pregnancy ([5](#_ENREF_5)), and this is supported by the recent Global Consensus on the Prevention of Rickets ([6](#_ENREF_6)). Indeed many national guidelines recommend universal antenatal vitamin D supplementation to prevent vitamin D insufficiency ([7-9](#_ENREF_7)).

Risk factors for vitamin D insufficiency are well described, and include ethnicity, extensive skin covering and liberal use of sun protection, overweight/obesity, low dietary vitamin D intake and smoking ([1](#_ENREF_1), [10](#_ENREF_10), [11](#_ENREF_11)), in addition to the seasonal variation that is observed at temperate latitudes ([11](#_ENREF_11), [12](#_ENREF_12)). Although vitamin D supplementation can improve maternal 25(OH)D status ([10](#_ENREF_10)), little is known about how maternal characteristics might influence the 25(OH)D achieved following supplementation. In non-pregnant adults, baseline 25(OH)D concentration, body weight/adiposity and age are important determinants of the incremental rise in 25(OH)D following vitamin D supplementation ([13](#_ENREF_13), [14](#_ENREF_14)). During pregnancy, maternal haemodilution is accompanied by a number of physiological changes to both vitamin D metabolism ([15](#_ENREF_15)) and maternal body composition ([16](#_ENREF_16)); such adaptations might lead to differences in the determinants of response to vitamin D supplementation between pregnant and non-pregnant women. Clinically, understanding how individuals respond could lead to individualised antenatal counselling regarding vitamin D supplementation to ensure vitamin D repletion is achieved without increasing the risk of vitamin D toxicity. We therefore undertook this study to determine maternal characteristics associated with achieved 25(OH)D following antenatal vitamin D supplementation in the context of a double-blind, randomised, placebo-controlled trial.

**Methods**

*The Maternal Vitamin D Osteoporosis Study (MAVIDOS)*

The MAVIDOS study is a multicentre, double-blind, randomised, placebo-controlled trial of vitamin D supplementation in pregnancy. The primary outcome was neonatal bone mass. A detailed description of the study methods ([17](#_ENREF_17)) and primary findings have been published previously ([18](#_ENREF_18)). The study was approved by the Southampton and South West Hampshire Research Ethics Committee. MAVIDOS was registered prospectively (ISRCTN:82927713; EUDRACT:2007-001716-23); full approval from UK MHRA was granted, and written, informed consent was obtained from all participants.

Briefly, women attending one of three United Kingdom (UK) hospitals [University Hospital Southampton NHS Foundation Trust, Southampton, UK (latitude 50.9° North); Oxford University Hospitals NHS Foundation Trust, Oxford, UK (latitude 51.8° North); Sheffield Hospitals NHS Trust (University of Sheffield), Sheffield, UK (latitude 53.4° North)] for early pregnancy ultrasound screening (11-14 weeks’ gestation) were invited to participate in the study. Inclusion criteria were: age over 18 years, singleton pregnancy, and gestation less than 17 weeks based on last menstrual period (LMP) and ultrasound measurements. Women with known metabolic bone disease, renal stones, hyperparathyroidism or hypercalciuria, those taking medication known to interfere with fetal growth, fetal anomalies on ultrasonography and women already using >400IU/day vitamin D supplementation were excluded. A screening blood sample was obtained and analysed on the local NHS platform [all three laboratories (Southampton, Oxford and Sheffield) participate in DEQAS vitamin D quality assurance system (http://www.deqas.org/)]. Women with 25(OH)D between 25 and 100nmol/l and serum calcium <2.75mmol/l were eligible to enroll fully in the study.

Participants were randomised to either cholecalciferol 1000 IU/day or matched placebo [Merck KGaA, (Darmstadt, Germany)/ Sharp Clinical Services (Crickhowell, UK; previously DHP-Bilcare)], which was commenced before 17 weeks’ gestation. The study medication was provided in a blister pack in a single box containing all medication for the whole pregnancy. All participants received standard antenatal care, and could continue self-administration of dietary supplements containing up to 400IU/day vitamin D.

*Maternal assessments during pregnancy*

Prior to commencing the study medication, and again at 34 weeks’ gestation, the women attended the research centre for a detailed assessment of diet (including supplement use), lifestyle (smoking, physical activity participation, employment) and health (past medical history, current medication use) using interviewer-led questionnaires. Ethnicity was determined by participant self-report, and subsequently categorised as white or non-white.

Anthropometric measurements included height, measured to the nearest 0.1cm using a stadiometer, and weight, assessed to the nearest 0.1kg using calibrated electronic scales. Four site (triceps, biceps, subscapular and suprailiac) skinfold thicknesses were measured to the nearest 0.2mm using a Harpenden skinfold calliper. Pregnancy weight gain was calculated as the difference between the weights at commencing study medication and 34 weeks’ gestation.

*Compliance with study medication*

Participants were asked to bring any remaining study medication to each assessment. The pills were counted and compliance calculated as number consumed/expected consumption based on number of days since medication was dispensed, and expressed as a percentage. The 34 week visit was used for the calculation of compliance, and the count at 19 weeks was used if a 34 week count was not available.

*Assessment of 25(OH)D status*

On the day that the study medication was dispensed and at 34 weeks’ gestation, a non-fasted venous blood sample was obtained, and serum stored at -80°C. 25(OH)D was assessed by radioimmunoassay (Liaison RIA automated platform, Diasorin, Minnesota, USA). All samples were analysed in a single batch at the end of the study at MRC Human Nutrition Research, Cambridge, UK. Details of assay performance and quality control through participation in DEQAS, NIST and NEQAS are given elsewhere ([19](#_ENREF_19), [20](#_ENREF_20)).

*Statistical Analysis*

Women who had a measurement of 25(OH)D at both 14 and 34 weeks’ gestation, and delivered a liveborn infant were included in the analysis (since pathology associated with fetal death might influence 25(OH)D concentrations). Maternal characteristics were compared between the women who did and did not complete the study using t-tests, Mann-Whitney U tests and χ2 tests for normally distributed, non-normally distributed and categorical variables, respectively. Linear regression was used to assess the association between maternal characteristics and 25(OH)D at 34 weeks’ gestation for each treatment group separately. Multivariate linear regression was subsequently performed including all variables with a p<0.2 from the linear regression. Additionally, maternal factors associated with achieving a vitamin D replete status (>50nmol/l) were determined using Poisson regression with robust standard errors ([21](#_ENREF_21)). The cut-point of 50nmol/l as the definition for vitamin D replete status was chosen to reflect the IOM guidelines ([5](#_ENREF_5)). Additionally, we considered a 25(OH)D>125nmol/l as indicating risk of toxicity, as suggested by the IOM ([5](#_ENREF_5)). In the primary trial analysis, we classified season of birth according to the UK Meteorological office recommendations (www.metoffice.gov.uk) with winter (December-February); spring (March-May); summer (June-August); and autumn (September-November). Since 25(OH)D concentrations are non-linearly associated with season, to facilitate ready comparison, we collapsed this classification into 2 groups with a notional “winter” (the months in which 25(OH)D concentrations tended to be lowest: December-May) and a “summer” (the months in which 25(OH)D concentrations tended to be highest: June-November). Finally, in sensitivity analysis, we excluded women who reported having taken any additional vitamin D-containing supplements within 90 days of the late pregnancy blood sampling. All analyses were performed in Stata v14 (Statacorp, College Station, Texas, USA). A p value of <0.05 was considered statistically significant.

**Results**

829 women, who delivered a live born infant and had measurements of 25(OH)D at both 14 and 34 weeks’ gestation, were included in the analysis (Figure 1). Women with missing 25(OH)D measurements at 34 weeks, who delivered a live born infant (n=136) were of similar age, parity, height, ethnicity, educational achievement, early pregnancy BMI and smoking status to those included in this analysis (p>0.05 for all). There were no significant differences in baseline characteristics between women randomised to placebo and vitamin D supplementation (Table 1). Compliance with study medication was high in both treatment groups [placebo median 95% (IQR 88-98%), cholecalciferol median 96% (IQR 89-99%), p=0.11].

*Maternal 25(OH)D status at 34 weeks’ gestation by randomisation group*

Maternal 25(OH)D at 34 weeks’ gestation was greater in the women randomised to cholecalciferol [mean 67.7nmol/l (SD 21.3nmol/l)] compared with the placebo group (mean 43.1nmol/l (SD 22.5 nmol/l), p<0.0001]. 83.3% of women randomised to cholecalciferol achieved vitamin D replete status at 34 weeks’ gestation (>50nmol/l) compared with 35.6% in the placebo group (p<0.001). Of the women who were not vitamin D replete at baseline (n=509), 78.8% in the cholecalciferol group were replete at 34 weeks’ gestation, compared with only 28.3% of the placebo group (p<0.001). Similarly, only 48.4% of women who were vitamin D replete at baseline and received placebo remained vitamin D replete at 34 weeks’ gestation, compared with 89.8% in the cholecalciferol group (p<0.001). In both treatment groups, the proportion of women who were vitamin D replete at 34 weeks’ gestation was lower in those who delivered in winter (Table 2). No participant reported symptoms suggestive of vitamin D toxicity. Two participants (0.5%) randomised to placebo and one to cholecalciferol (0.3%) (p difference=0.58) had a 25(OH)D≥125nmol/l at 34 weeks’ gestation, with the maximum value being 139nmol/l.

*Determinants of maternal 25(OH)D at 34 weeks’ gestation*

In univariate analysis, maternal age, baseline 25(OH)D, season of delivery and compliance with study medication were significantly associated with 34 week 25(OH)D in both the placebo and vitamin D supplementation groups (Table 3). Additionally, women who reported smoking in late pregnancy had significantly lower 25(OH)D in the placebo group, but this association was not observed amongst women randomised to cholecalciferol. Conversely, markers of maternal weight and adiposity were significantly inversely associated with maternal 25(OH)D in the cholecalciferol group, but not the women randomised to placebo.

In multiple linear regression analysis, maternal factors significantly associated with greater 25(OH)D at 34 weeks’ gestation in the vitamin D supplementation group were lower pregnancy weight gain [kg] (β=-0.81; 95%CI: -1.39, -0.22; p=0.007), higher compliance [%] (β=0.28; 95%CI: 0.072, 0.48; p=0.008), higher early pregnancy 25(OH)D [nmol/l] (β=0.28; 95%CI: 0.16, 0.40; p<0.001) and summer delivery [summer vs winter] (β=10.51; 95%CI: 6.40, 14.63; p<0.001) (Figure 2a). In the placebo group (Figure 2b), higher early pregnancy 25(OH)D [nmol/l] (β=0.59; 95%CI: 0.49, 0.68; p<0.001), summer delivery [summer vs winter] (β=24.97; 95%CI: 21.77, 28.17; p<0.001), and greater maternal age (years) (β=0.32; 95%CI: 0.022, 0.62; p=0.04 ) remained significantly associated with greater 25(OH)D at 34 weeks’ gestation. When achievement of vitamin D replete status at 34 weeks’ gestation was considered instead of absolute 25(OH)D concentration, in multivariate analyses, delivery in summer (RR=1.20: 95%CI: 1.09, 1.33; p<0.001), white ethnicity (RR=1.267; 95%CI: 1.17, 1.37; p<0.001), greater compliance with medication [%] (RR=1.01; 95%CI: 1.00, 1.02; p=0.03), and greater early pregnancy 25(OH)D concentration [nmol/l] (RR=1.003; 95%CI: 1.001, 1.006; p=0.007) were significantly associated with achieving 25(OH)D>50nmol/l in the women randomised to cholecalciferol.

*Interaction between baseline 25(OH)D and randomisation group*

When comparing achieved 25(OH)D at 34 weeks’ gestation between placebo and cholecalciferol groups, it was apparent that there was a statistically significant interaction between baseline 25(OH)D and randomisation group (p<0.001). Thus there was a smaller difference in 25(OH)D concentrations at 34 weeks’ gestation between the placebo and treatment arms with increasing 25(OH)D at 14 weeks’ gestation.

*Sensitivity analyses*

As participants were permitted to continue taking daily vitamin D supplements containing up to 400IU, in the sensitivity analysis we excluded 229 women (n=117 randomised to cholecalciferol) who reported taking other vitamin D containing dietary supplements at the late pregnancy interview. Similarly, 81.0% of women randomised to cholecalciferol were vitamin D replete at 34 weeks’ gestation, compared with 29.4% of women randomised to placebo (p<0.001). The maternal characteristics associated with 25(OH)D at 34 weeks’ gestation and achieving vitamin D replete status were similar to those observed in the whole cohort.

**Discussion**

We have assessed anthropometric and demographic factors associated with the response to antenatal supplementation with 1000 IU/day cholecalciferol. This dose achieved vitamin D repletion in over 80% of women, without leading to 25(OH)D levels potentially associated with vitamin D toxicity (at least within the included baseline of 25-100nmol/l 25(OH)D). However, gaining less weight during pregnancy, having a higher 25(OH)D in early pregnancy, delivering in summer and having higher compliance with supplementation were independently associated with achieving a greater 25(OH)D concentration in late pregnancy amongst women randomised to vitamin D supplementation. Thus, those women who are at risk of vitamin D insufficiency in early pregnancy, gain greater weight, and deliver in winter might need supplementation with a higher dose of cholecalciferol to achieve similar 25(OH)D concentrations. However, when vitamin D replete status was considered as the outcome, only non-white maternal ethnicity and delivery in winter were significant predictors of vitamin D non-replete status following supplementation.

To our knowledge, the factors which determine the response to vitamin D supplementation in pregnancy have not previously been assessed. However, our findings are consistent with those in non-pregnant adults ([13](#_ENREF_13), [14](#_ENREF_14)). It is well recognised that individuals who are overweight or obese are at higher risk of vitamin D insufficiency, and this is similarly observed in pregnancy ([2](#_ENREF_2), [22](#_ENREF_22)). Studies in non-pregnant adults have also shown that obese individuals achieve a lower 25(OH)D with the same dose of supplementation as non-obese individuals ([14](#_ENREF_14)). Meta-analysis of vitamin D supplementation studies has suggested that over 50% of the variance in 25(OH)D increment in response to supplementation is explained by body weight ([13](#_ENREF_13)). Although the relationship between body weight and 25(OH)D increment following supplementation could reflect sequestration in adipose tissue, we found that in multivariate analysis, pre-pregnancy BMI and late pregnancy triceps skinfold thickness (as a marker of adiposity), were not associated with 25(OH)D after supplementation, but that pregnancy weight gain was negatively associated. Similarly, we have previously demonstrated that greater gestational weight gain is associated with a decline in 25(OH)D status during pregnancy, independent of supplement use ([12](#_ENREF_12)). Weight gain in pregnancy represents not only increased fat mass but also feto-placental tissues and haemodilution ([16](#_ENREF_16)); our data, therefore, suggest that overall volume of dilution, and not just adiposity, may be important for response to vitamin D supplementation. However, importantly, when using a 25(OH)D>50nmol/l as a cut-point for repletion, pregnancy weight gain was not an independent predictor of achieving vitamin D repletion.

Despite receiving 1000IU cholecalciferol per day, 25% of mothers delivering in winter had a 25(OH)D less than 50 nmol/l. This is a higher non-repletion rate than that reported in other recent pregnancy supplement studies ([23-25](#_ENREF_23)). However, it is notable that there were marked differences in baseline 25(OH)D concentrations between these investigations, and we observed that initial 25(OH)D status was positively associated with both the likelihood of achieving vitamin D replete status and absolute 25(OH)D status at 34 weeks’ gestation. Importantly, the difference between the 25(OH)D achieved at 34 weeks’ gestation in women randomised to placebo compared with cholecalciferol decreased with increasing baseline 25(OH)D. This is consistent with previous studies in adults, which have shown that the incremental response to vitamin D supplementation is higher in vitamin D insufficient than replete subjects ([13](#_ENREF_13), [14](#_ENREF_14)), and that the increase in 25(OH)D relative to supplementation dose is negatively associated with dose of vitamin D supplement ([26](#_ENREF_26)). This suggests that physiological processes such as saturation of the hepatic 25-hydroxylase involved in the conversion of cholecalciferol to 25(OH)D or conversion to 24- or 4-OH metabolites, together with renal catabolism, limit attainment of very high 25(OH)D concentrations ([27](#_ENREF_27)). This mechanism might be important in preventing hypervitaminosis D. However, studies comparing the effectiveness of differing doses of vitamin D in pregnancy have shown that 4000IU/day can achieve a higher 25(OH)D than 400IU/day ([10](#_ENREF_10), [28](#_ENREF_28)), but whether these higher doses are of clinical benefit is yet to be demonstrated ([4](#_ENREF_4), [29](#_ENREF_29)) and at the general population level, lower doses would be compatible with keeping 25(OH)D below a concentration which might be concerning.

It is evident from our findings that 1000 IU/day cholecalciferol in pregnancy does not eliminate the seasonal variation in 25(OH)D status observed in pregnant women in the UK ([11](#_ENREF_11), [12](#_ENREF_12)). Similarly, non-white ethnicity was associated with a higher risk of not achieving vitamin D replete status in the supplemented women. Hollis et al. similarly found that even with 4000IU/day vitamin D during pregnancy, African-American women had lower 25(OH)D in late pregnancy than Caucasian or Hispanic women ([10](#_ENREF_10)). Thus, future studies should aim to determine the dose required to achieve optimal 25(OH)D status amongst women of non-white ethnicity and amongst those who deliver in winter months. Maternal age was also positively associated in univariate analyses with 25(OH)D achieved at 34 weeks’ gestation in women who received vitamin D supplementation. It has previously been shown in pregnant women that age is positively associated with 25(OH)D status ([30](#_ENREF_30), [31](#_ENREF_31)). Whilst lower uptake of supplementation in younger women ([12](#_ENREF_12)) could partly explain this observation, our finding would additionally suggest that even in younger women who do use supplements, the achieved 25(OH)D is lower. Data from healthy and hospitalised adults have similarly shown that older individuals achieve a higher 25(OH)D following vitamin D supplementation ([13](#_ENREF_13), [32](#_ENREF_32)). As such, young pregnant women might particularly require advice on the need for, and compliance with, vitamin D supplementation.

Although our findings are novel, and may enable the development of individualised advice for antenatal vitamin D supplementation, there are a number of limitations which should be considered in the interpretation of this study. Firstly, we could not, as a result of stipulations made during the ethics approval process, include participants with 25(OH)D concentrations<25nmol/l or >100nmol/l. As baseline 25(OH)D was associated with the likelihood of achieving vitamin D replete status, it is likely that women with very low levels of 25(OH)D at baseline will require a higher supplementation dose to achieve vitamin D repletion. However, this needs to be confirmed in future studies. Secondly, only a small proportion of the women included in this study were of non-white ethnicity. This reflects the local populations and care should be taken in translating these findings to a more ethnically diverse population. Thirdly, we did not examine genetic determinants of the response to vitamin D supplementation. It has been demonstrated previously that the incremental rise in 25(OH)D following supplementation differs by single nucleotide polymorphisms in vitamin D binding protein ([33](#_ENREF_33), [34](#_ENREF_34)) and 25-hydroxylase genes ([33](#_ENREF_33)). Although this genetic information can enable a more comprehensive understanding of the biochemical response to vitamin D supplementation, the current inability to undertake genotyping on a widespread population basis means this additional information would not allow for alterations to current clinical practice regarding vitamin D supplementation in pregnancy. Finally, during pregnancy, a number of physiological changes occur to vitamin D metabolism, including an increase in vitamin D binding protein and 1,25-dihydroxyvitamin D ([15](#_ENREF_15)), indices that we were not able to include in our analysis.

In conclusion, we have demonstrated that women who gain more weight during pregnancy, have lower 25(OH)D in early pregnancy or deliver in winter tend to achieve a lower 25(OH)D in late pregnancy when supplemented with 1000IU/day cholecalciferol than do women with the converse attributes. Future studies should aim to determine appropriate doses to enable consistent repletion of 25-hydroxyvitamin D during pregnancy, and our findings support the notion that clinical approaches to vitamin D repletion may be informed by individual characteristics. As such, personalised vitamin D supplementation advice could become part of future antenatal care.

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**Figure Legends**

**Figure 1:** Consort diagram

**Figure 2:** Independent determinants of maternal 25(OH)D at 34 weeks gestation a) following supplementation with 1000IU cholecalciferol per day from 14 weeks’ gestation until delivery; and b) receiving placebo from 14 weeks’ gestation until delivery. Shown as standard deviation change in 25(OH)D per unit predictor. \*p<0.05, \*\*p<0.01

**Table 1**: Maternal characteristics at baseline according to randomisation group.

|  |  |  |
| --- | --- | --- |
|  | Placebo | 1000IU/day cholecalciferol |
| N | 422 | 407 |
| Gestation (weeks), mean (SD) | 15.9 (1.5) | 15.9 (1.5) |
| Maternal age (years), mean (SD) | 30.7 (5.4) | 30.7 (5.0) |
| Nulliparous (%) | 44.8 | 42.7 |
| Current smoker (%) | 7.7 | 7.7 |
| BMI (kg/m2), median (IQR) | 25.4 (22.7-29.7) | 24.6 (22.2-28.6) |
| Height (cm), mean (SD) | 165.6 (6.6) | 165.5 (6.3) |
| White ethnicity (%) | 94.8 | 95.6 |
| 25(OH)D (nmol/l), median (IQR) | 44.4 (33.2-57.0) | 45.7 (34.3-57.8) |

**Table 2**: Percentage of women achieving vitamin D replete status (>50nmol/l) according to randomisation group and season of delivery.

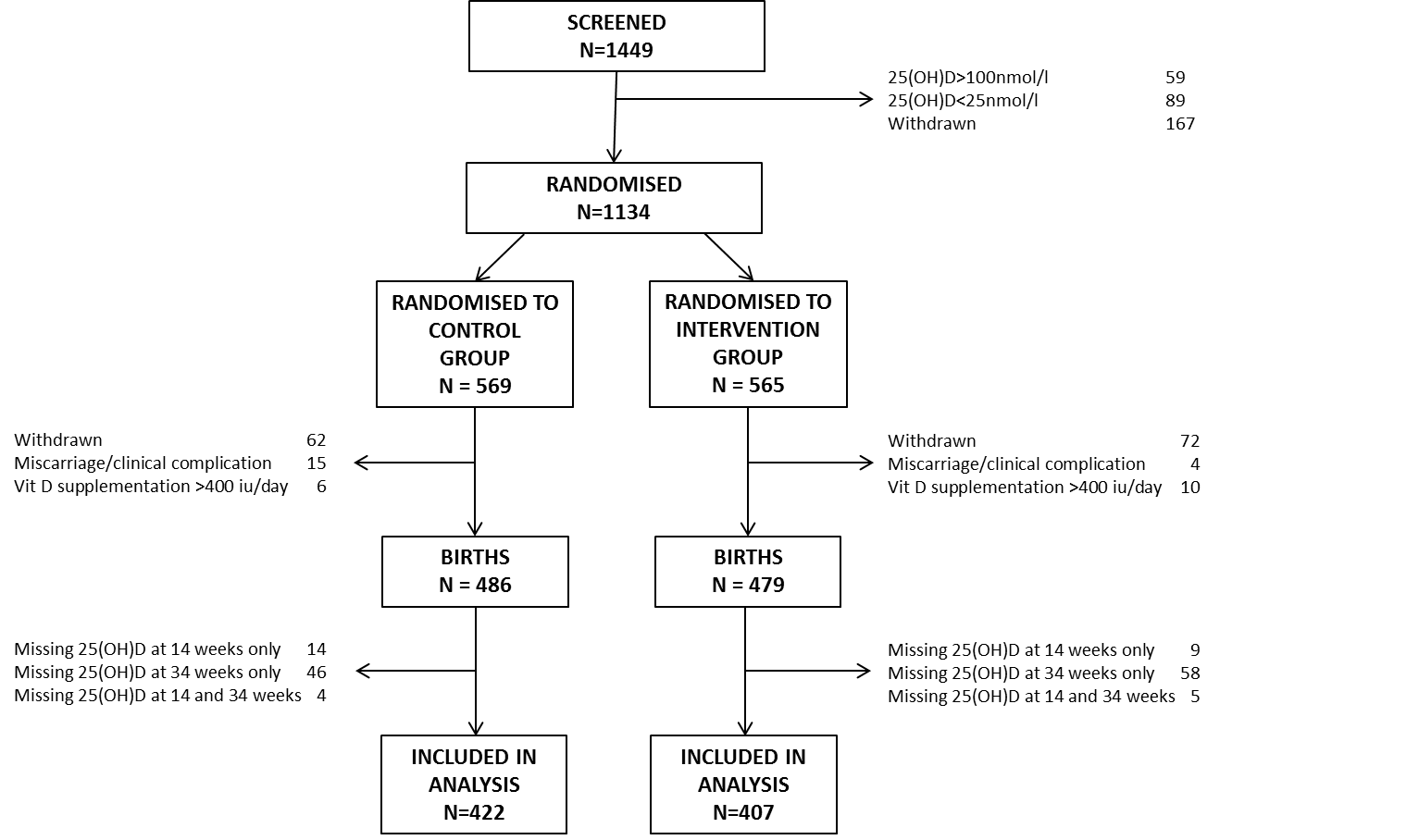
|  |  |  |  |
| --- | --- | --- | --- |
| Season of delivery | Placebo | 1000 IU/day cholecalciferol | p comparing randomisation groups |
| Winter (December-May) | 13.9 | 75.0 | <0.001 |
| Summer (June-November) | 54.2 | 90.1 | <0.001 |
| p comparing seasons | <0.001 | <0.001 |  |

**Table 3:** 25-hydroxyvitamin D status at 34 weeks gestation according to maternal characteristics in women randomised to placebo or vitamin D supplementation from 14 weeks gestation. Shown as nmol/l change in 25(OH)D per unit predictor.

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
|  | Placebo | | 1000 IU/day cholecalciferol | |
|  | Beta (95% CI) | p | Beta (95% CI) | p |
| Maternal age (years) | **0.67 (0.28, 1.07)** | **0.001** | **0.70 (0.28, 1.11)** | **0.001** |
| Parity (yes vs no) | -1.25 (-5.69, 3.20) | 0.581 | -0.29 (-4.60, 4.03) | 0.896 |
| Smoking at 34 weeks gestation (yes vs no) | **-13.45 (-22.12, -4.78)** | **0.002** | -1.49 (-9.50, 6.52) | 0.715 |
| Ethnicity (other vs white) | -8.69 (-18.59, 1.21) | 0.085 | 1.99 (-8.50, 12.48) | 0.709 |
| Height (cm) | 0.15 (-0.19, 0.48) | 0.389 | -0.072 (-0.41, 0.27) | 0.675 |
| BMI at 14 weeks gestation (kg/m2) | -0.24 (-0.69, 0.20) | 0.284 | **-0.47 (-0.90, -0.048)** | **0.029** |
| Weight at 34 weeks gestation (kg) | -0.056 (-0.22, 0.11) | 0.492 | **-0.23 (-0.38, -0.085)** | **0.002** |
| Weight gain early to late pregnancy (kg) | -0.23 (-0.85, 0.40) | 0.473 | **-0.65 (-1.26, -0.039)** | **0.037** |
| Triceps SFT at 34 weeks gestation (mm) | -0.059 (-0.38, 0.26) | 0.718 | **-0.42 (-0.74, -0.10)** | **0.01** |
| Moderate/strenuous exercise in late pregnancy (hrs/week) | 1.36 (-1.75, 4.47) | 0.389 | -0.76 (-3.63, 2.10) | 0.60 |
| 25(OH)D at 14 weeks gestation (nmol/l) | **0.52 (0.40, 0.64)** | **<0.001** | **0.21 (0.089, 0.33)** | **0.001** |
| Season of delivery (summer vs winter) | **22.77 (19.05, 26.50)** | **<0.001** | **10.09 (6.039, 14.15)** | **<0.001** |
| Compliance (%) | **0.23 (0.044, 0.42)** | **0.016** | **0.39 (0.19, 0.59)** | **<0.001** |

SFT: skinfold thickness

**Figure 1:** Consort diagram



**Figure 2:** Independent determinants of maternal 25(OH)D at 34 weeks gestation a) following supplementation with 1000IU cholecalciferol per day from 14 weeks’ gestation until delivery; and b) receiving placebo from 14 weeks’ gestation until delivery. Shown as standard deviation change in 25(OH)D per unit predictor. \*p<0.05, \*\*p<0.01

1. *Cholecalciferol 1000IU/day*

****

1. *Placebo*

**