**Pregnancy vitamin D supplementation and bone mineral density of the mother: A post-hoc analysis of the MAVIDOS randomised placebo-controlled trial**

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**Declaration of interests**

RJM has received travel bursaries from Kyowa Kirin unrelated to this work. EMC has received travel bursaries, consultancy or lecture fees from Celltrion, Eli Lilly, Pfizer, Theramex, Thornton and Ross and UCB, unrelated to this work. KMG has received reimbursement for speaking at conferences sponsored by companies selling nutritional products and is part of an academic consortium that has received research funding from Bayer, Nestec, BenevolentAI Bio Ltd., Boehringer-Ingelheim and Danone, outside the submitted work. KAW received Honoraria from Abbott Nutrition and Nestle Nutrition unrelated to this work. CC reports personal fees from ABBH, Amgen, Eli Lilly, GSK, Medtronic, Merck, Novartis, Pfizer, Roche, Servier and Takeda, outside the submitted work. NCH reports personal fees, consultancy, lecture fees and honoraria from Alliance for Better Bone Health, AMGEN, MSD, Eli Lilly, Servier, Theramex, Shire, Consilient Healthcare, Kyowa Kirin and Internis Pharma, outside the submitted work.

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**Abstract (250 words)**

**Purpose**

Vitamin D supplementation during pregnancy has positive effects on the offspring’s bone development and bone mineral density (BMD) in childhood, but there are limited data on the effects on the mother’s skeleton. We assessed this in post-hoc follow-up of a randomised trial.

**Methods**

MAVIDOS was a randomised placebo-controlled trial of 1000 IU/day cholecalciferol from 14-17 weeks’ gestation until delivery. Participants were invited to have a Dual-energy X-ray Absorptiometry (DXA) scan of the whole-body, lumbar spine (LS) and left hip within 2 weeks after delivery and again 4 years after delivery.

BMD was compared between the cholecalciferol and placebo groups; in women with DXA at both time points, the effect of cholecalciferol was assessed using mixed effects modelling to account for repeated measures.

**Results**

401 and 443 participants had a DXA within 2 weeks after delivery and at mean 4.1 years after delivery, respectively. Cholecalciferol increased serum 25-hydroxyvitamin D status in late pregnancy. BMD at all sites was similar between the two randomisation groups at both time points (p>0.05 for all).

263 participants had a DXA at both birth and 4 years. BMD increased between 1 and 6% depending on site from within 2 weeks to 4 years after delivery. This was similar between randomisation groups. The pregnancy cholecalciferol had no effect on BMD at 4 years in repeated measures modelling.

**Conclusions**

Despite previously demonstrated benefits for the offspring skeleton in this trial, maternal BMD did not differ in the short or medium term in women randomised to 1000 IU/day vitamin D or placebo.

**Mini Abstract** (42 words)

The effect of pregnancy vitamin D supplementation on bone mineral density (BMD) in the mother immediately after delivery and at 4 years after delivery was assessed in a randomised placebo-controlled trial. The mothers’ BMD did not differ between the two intervention groups.

**Key words**

Vitamin D, cholecalciferol, bone mineral density, pregnancy; epidemiology, osteoporosis

**Background**

During pregnancy and lactation, mobilisation of calcium from the maternal skeleton [1], in addition to increased maternal intestinal absorption of calcium [2], ensures sufficient calcium is available for fetal mineral accretion. As a result, maternal bone mineral density (BMD) typically reduces during pregnancy [3, 4], and to a greater extent during lactation [5], before recovery by 1-2 years postpartum [6]. During pregnancy, changes in maternal calciotropic hormones including increased parathyroid hormone related peptide (PTHrP) and 1,25-dihydroxyvitamin D may play a role in these processes [2, 7].

Randomised controlled trials during pregnancy have demonstrated positive effects of pregnancy vitamin D supplementation on offspring BMD in childhood [8-11]. However, there are few studies that have examined the effect of vitamin D or calcium supplementation in pregnancy on the woman’s skeleton. We have previously demonstrated that supplementation with 1000 IU/day cholecalciferol during pregnancy reduced C-terminal telopeptide of type I collagen (CTX) in maternal urine samples collected during late pregnancy, suggesting reduced skeletal mobilisation [12]. In contrast, a study in The Gambia has suggested that calcium supplementation during pregnancy in women with habitually low calcium intake may have a detrimental long-term effect on the maternal skeleton [13, 14], whereas a more recent study in the United States of America suggested calcium supplementation during the second and third trimesters prevented maternal bone loss from early pregnancy to 12 months postpartum [3]. To our knowledge, there have been no studies that have reported on the long-term effects of vitamin D supplementation on the maternal skeleton, yet the findings reported in the study of women in The Gambia, albeit a calcium supplementation trial, highlights the need to ensure there are no detrimental effects of this. We hypothesized that pregnancy vitamin D supplementation would have a positive effect on maternal BMD in the short and medium term. We therefore undertook post-hoc DXA assessment of BMD in women who participated in a randomised controlled trial of vitamin D supplementation within two weeks after delivery and at 4 years after delivery to assess for differences in BMD between the two intervention groups.

**Methods**

MAVIDOS was a randomised placebo-controlled trial of vitamin D supplementation in pregnancy. Full details of the study methodology and the primary outcome (offspring bone mass at birth) have been previously published [15, 16]. The trial was approved by the Southampton and South-West Hampshire Research Ethics Committee and with approval from UK Medicines and Healthcare products Regulatory Agency (MHRA). All women gave written informed consent to participate in each phase of the study. MAVIDOS was registered on the International Standard Randomized Controlled Trial registry (ISRCTN 82927713) and the European Clinical Trials Database (EudraCT 2007–001716–23).

The trial originally recruited women from three centres in the United Kingdom (University Hospital Southampton NHS Foundation Trust, Oxford University Hospitals NHS Foundation Trust and Sheffield Hospitals NHS Trust); follow-up of the women and their children after birth has been limited to those recruited in Southampton.

Women aged > 18 years with a singleton pregnancy and between 11-14 weeks’ gestation were invited to participate in the study between 10th October 2008 and 11th February 2014 when attending for their dating scan. Owing to an ethical stipulation, only women with a baseline 25(OH)D measured on the local hospital platform [all three laboratories participate in the Vitamin D External Quality Assessment Scheme (DEQAS) vitamin D quality assurance system ([http://www.deqas.org/](https://protect.checkpoint.com/v2/___http%3A//www.deqas.org/___.bXQtcHJvZC1jcC1ldXcyLTE6dW5pdmVyc2l0eWhvc3BpdGFsc291dGhhbXB0b246YzpvOjMyNmRiNTU5MzhiYjE0ZmE2OGZlMGExNjJmOTYwYzY2OjY6ZDE5YTpkOTZkYzAwNmQxN2Q0ODVlMzY2YWQyZjM4ODVjZWJiZmU4OGE2OTZkZDRjZDU4OTZmYzExYTdmYmY1NzExNjk1OnA6VA))] between 25 and 100 nmol/l were eligible to participate. Women were randomised in a 1:1 ratio to either oral cholecalciferol 1000 IU/day or matched placebo started from 14 to 17 weeks' gestation and continued until delivery. All women received standard antenatal and intrapartum care delivered by health professionals blinded to the study allocation and were able to continue taking up to 400 IU/day vitamin D supplementation.

*Pregnancy assessments*

Lifestyle, health and nutrition were assessed by interviewer-led questionnaire at randomisation (14-17 weeks’ gestation) and again at 34 weeks’ gestation. Anthropometry and non-fasted venous blood samples were also taken at these visits. Serum was stored at −80°C. 25(OH)D concentration was assessed by chemiluminescence immunoassay (Liaison automated platform, Diasorin, Minnesota, USA). All samples were analysed in a single batch at Medical Research Council (MRC) Human Nutrition Research, Cambridge, UK. Within- and between-assay coefficients of variation were 4.1 and 6.1%.

*Assessments after delivery*

Height and weight were measured and body mass index (BMI) calculated from these measurements. Whole body, lumbar spine and left hip dual-energy X-ray absorptiometry (DXA) scans were obtained using a Hologic Discovery instrument (Hologic Inc., Bedford, MA) within two weeks of and 4 years after delivery. The same instrument was used for both assessments. Outcomes of interest were bone area (BA), bone mineral content (BMC) and BMD for whole body, whole body less head (WBLH), lumbar spine and total left hip. Two researchers blinded to randomisation reviewed the scans; images with substantial movement artefact affecting the whole body and/or both legs/both arms were excluded. In scans with movement artefact in one limb, the region of interest (ROI) of the unaffected limb was transposed into the limb with movement artefact. The DXA instrument underwent daily calibration using a spine phantom. The experimental coefficient of variation for this instrument when a spine phantom was repeatedly scanned in the same position 16 times, in a single session with no repositioning, was 0.68%.

Duration of breastfeeding of the child born from the study pregnancy was established in an interviewer-led questionnaire at 1 and 2 years after delivery. At the DXA scan 4 years after delivery, other pregnancy and duration of breast feeding of any subsequent children was additionally determined by interviewer-led questionnaire.

**Statistical methods**

Between group comparisons, as well as comparisons of the mothers’ characteristics between those included versus not included in this follow-up, were performed using t-tests, Mann-Whitney U tests and Chi-squared tests for normally distributed continuous, non-normally distributed continuous and categorical variables, respectively. Results are presented as mean (standard deviation [SD]), median (interquartile range [IQR]) and n (%), respectively. The differences in DXA outcomes between randomisation groups within 2 weeks of delivery were examined using linear regression to allow for adjustment for height and weight (to increase the precision of effect size estimates) and their age. Additionally at 4 years after delivery, further completed pregnancies since the study pregnancy (yes/no) and total duration of lactation since the study pregnancy were included in the models as these factors are recognised to have short-term effect on BMD [5, 6]. We assessed for a statistical interaction of the intervention with baseline 25(OH)D insufficiency using a threshold of 50 nmol/l.

In the subset of participants who had a DXA within 2 weeks of delivery and at 4 years, BMD from the two scans was compared using paired t-tests while between randomisation group comparisons were performed using unpaired t-tests. Percentage change in BMD from birth to 4 years was calculated as change in BMD at 4 years divided by BMD within 2 weeks of delivery (x 100), and compared between groups using unpaired t-tests. Mixed effects modelling was used to assess the differences between randomisation groups in BMD at 4 years to account for the repeated DXA measurements. Adjustment for maternal age, further completed pregnancies since the study pregnancy (yes/no), total duration of lactation since the study pregnancy and the MAVIDOS child’s age (as a measure of time since study pregnancy completion) were included in the models. All analysis was performed using Stata V17.0 (StataCorp LP, College Station, TX, USA).

**Results**

1134 participants were randomised to placebo or cholecalciferol, of whom 965 participants remained in the study until delivery. 767 of these participants were recruited in Southampton, of whom 401 (52%) had a DXA within 2 weeks of delivery and 443 (58%) had a DXA scan at 4 years after delivery (Figure 1). In comparison to all participants who remained in the study until delivery, those who had a DXA were, on average, older and tended to be more highly educated and less likely to smoke in early pregnancy (Supplementary tables 1 and 2). For the mothers who had a DXA within two weeks of delivery, BMI was also higher and these participants were more likely to be of White ethnicity that the mothers who did not participate in this DXA scan (Supplementary Table 1); these differences were not evident for the group of mothers who participated in the DXA at 4 years after delivery (Supplementary Table 2).

The characteristics of the mothers included in this analysis at randomisation are shown by treatment group for each DXA scan time point in Table 1. Gestation at delivery was similar for each randomisation group (Tables 2 and 3), but as would be expected, 25(OH)D status in late pregnancy was greater in the mothers randomised to cholecalciferol (Tables 2 and 3).

*BMD of the mothers within two weeks of delivery*

There was no difference in BA, BMC nor BMD for whole body, WBLH, lumbar spine or total hip between the two randomisation groups when measured within two weeks of birth (Table 2). Adjustment for age, height and weight did not alter these findings (Supplementary Table 3). There was no evidence of a statistical interaction between the intervention and baseline maternal 25(OH)D insufficiency for DXA outcomes.

*BMD of the mothers 4 years after delivery*

At 4 years after delivery, the two groups of participants were of similar weight and BMI (Table 3). The proportion of participants in each group who reported one or more further completed pregnancies since the study pregnancy did not differ (placebo 31.8 %, cholecalciferol 37.7%, p=0.20), but duration of lactation was longer in the cholecalciferol group for both the infant born from the study pregnancy and total duration of lactation for any infant since the study pregnancy (Table 3).

BA, BMC and BMD for whole body, WBLH, lumbar spine and total hip were not different between the two randomisation groups at 4 years after delivery (Table 3), with no evidence of a statistical interaction with baseline 25(OH)D. Using linear regression to adjust for the mother’s age, height, weight, subsequent pregnancy and total duration of lactation, there was no evidence of a difference between pregnancy vitamin D supplementation versus placebo on the mothers’ DXA measures at 4 years after delivery (Supplementary Table 4).

*Longitudinal BMD measurements within 2 weeks of and 4 years after delivery*

263 of the participants had a DXA scan within 2 weeks of delivery and again at 4 years after delivery. In both the placebo and cholecalciferol groups, BMD increased from the immediate post-partum scan to 4 years later, ranging from a 1% increase at the lumbar spine to 6% increase for WBLH (Figure 2). Using mixed effects modelling with repeated measures, there was no difference between randomisation groups on maternal BMD (Table 4) and no interaction between the intervention and phase of follow-up (p>0.05 for all).

**Discussion**

In this randomised placebo-controlled trial, moderate dose vitamin D supplementation during pregnancy did not result in a difference in the mothers’ BMD within 2 weeks of delivery nor at 4 years after delivery compared to placebo. This is despite a positive effect of the intervention on the offspring BMD persisting into mid childhood being previously demonstrated in the offspring born to the mothers participating in this trial [11, 17].

During pregnancy and lactation, mobilization of the maternal skeleton is one of a number of adaptations that ensures sufficient calcium is available to the fetus. We therefore hypothesized that vitamin D supplementation would reduce maternal skeletal resorption by enabling increased dietary calcium absorption. Despite a reduction in a marker of bone resorption (urinary CTX) during late pregnancy in this trial [12] and at delivery in a trial of pregnancy vitamin D supplementation in Bangladesh [18], moderate dose vitamin D supplementation in pregnancy did not have a detectable positive effect on the mothers’ BMD either at delivery in this work or that of Wei et al [19] or in the medium term, as shown in this analysis. These findings are consistent with trials in non-pregnant women of child-bearing age that have not demonstrated a positive effect of vitamin D supplementation on BMD [20].

Importantly, women supplemented with vitamin D during pregnancy did not have a lower BMD than the placebo group. Jarjou et al reported an unexpected finding of lower size-adjusted BMC in women in The Gambia randomised to calcium supplementation during pregnancy at long-term follow-up. The authors of that work suggested that this may have resulted from an alteration in the women’s ability to adapt to a habitually low calcium intake after supplementation was discontinued [13, 14]. Differences in the outcomes of these two studies could also reflect the differences in habitual calcium intake. Calcium intake was not formally assessed in women in the MAVIDOS study, but assessment of calcium intake in pregnant women in the UK by other authors suggest this to average 1345 mg/day (SD 459 mg/day) [21] compared with approximately 350 mg/day in women in the Gambia [14]. Calcium supplementation is not routinely recommended during pregnancy in the UK. Although undertaken in a cohort who did not have severe vitamin D deficiency, our findings are therefore important to provide reassurance that a negative effect of pregnancy vitamin D supplementation on BMD is not suggested in the short and medium term. Indeed, in the outcomes examined in this trial including maternal obstetric health [22, 23] and offspring anthropometry, bone health [11, 15, 17] and eczema [24], we have not identified any negative effects of the intervention in this UK study population.

Interestingly, duration of lactation was longer in the women randomised to cholecalciferol. An effect of pregnancy vitamin D supplementation on duration of lactation has not previously been reported. More of the mothers randomised to cholecalciferol attempted breast feeding suggesting this could be a chance finding. However, it is recognized that vitamin D supplementation increases breast milk vitamin D content [25]. We have not been able to identify any existing literature on whether vitamin D supplementation promotes breast development during pregnancy and/or human milk supply, but this should be explored in future research.

MAVIDOS is one of the largest RCTs of pregnancy vitamin D supplementation completed to date, and whilst this analysis was not part of the pre-specified analysis plan for MAVIDOS [17], extending the outcomes of the trial can extend scientific knowledge, and in this case, demonstrate safety of the intervention. However, this analysis has limitations. First, women with very low levels (<25nmol/l) of 25(OH)D at recruitment were excluded from participation due to an ethical stipulation and governance issues. Very deficient women may have benefitted the most from vitamin D supplementation, although there was no evidence of a statistical interaction with baseline 25(OH)D. Second, most participants were of White ethnicity and, those that attended for DXA scan follow-up either soon after birth or 4 years postpartum tended to be more highly educated and less likely to smoke. Therefore, this group of more highly educated women are likely to have a more prudent diet and level of exercise which could partially compensate for detrimental effects of lower vitamin D status in pregnancy on the maternal skeleton [26]. The group of women with DXA at birth and at 4 years after delivery did differ with just under 60% of the women who had a DXA at 4 years also having had a scan shortly after delivery. Together, these characteristics may limit the generalizability of our findings to other populations. Finally, as study participants were recruited in early pregnancy, a baseline DXA could not be performed prior to randomization. Therefore, we cannot be certain that despite randomisation there was not a difference in baseline BMD between the two groups and that change in BMD from early pregnancy to delivery was not affected by the intervention. Furthermore, DXA was undertaken soon after delivery and then again 4 years later, but not several months after breast feeding commencement, so we cannot exclude a transient protective effect on the mother’s skeleton during breastfeeding.

**Conclusion**

In conclusion, supplementation with 1000 IU/day during mid and late pregnancy did not lead to a difference in maternal BMD either immediately after delivery or at 4 years after delivery compared to placebo. Whilst these findings do not support a beneficial effect of vitamin D supplementation during pregnancy for the maternal skeleton, the lack of a negative difference would provide further reassurance to women and support increasing the currently recommended supplementation in pregnancy to improve offspring bone health and obstetric outcomes.

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**FIGURE LEGENDS**

**Fig 1**: Consort diagram

**Fig 2**: Bone mineral density within 2 weeks (circles, “birth”) and 4 years (squares) after delivery in women randomised to placebo (black) and cholecalciferol 1000 IU/day (white) during pregnancy. Markers represent mean, bars represent 95% CI. (\* different from birth measurement for same randomization group p<0.01; \*\* different from birth measurement for same randomization group p<0.001)

**TABLES**

**Table 1**: Maternal characteristics at randomisation for women who had DXA within 2 weeks of delivery (N=401), at 4 years after delivery (N=443), and those with DXA at both time points (N=263)

|  |  |  |  |
| --- | --- | --- | --- |
|  | DXA within 2 weeks of delivery | DXA 4 years after delivery | DXA at both 2 weeks and 4 years after delivery |
|  | Placebo | Cholecalciferol | Placebo | Cholecalciferol | Placebo | Cholecalciferol |
| n | 197 | 204 | 225 | 218 | 129 | 134 |
| Age (years), mean (SD) | 31.2 (4.9) | 31.5 (4.8) | 31.6 (4.7) | 32.0 (4.5) | 31.7 (4.8) | 32.4 (4.4) |
| Nulliparous, (%) | 43.3 | 40.6 | 37.9 | 42.5 | 37.5 | 40.3 |
| Height (cm), mean (SD) | 165.7 (6.7) | 165.5 (5.8) | 166.5 (6.3) | 165.7 (6.1) | 166.5 (6.5) | 165.7 (6.1) |
| Weight (kg), median (IQR) | 68.0 (61.7, 77.2) | 68.2 (59.9, 88.4) | 71.1 (64.3, 81.2) | 69.9 (62.1, 79.5) | 68.0 (62.7, 77.0) | 70.3 (62.0, 77.7) |
| BMI (kg/m2), median (IQR) | 24.7 (22.5, 28.1) | 24.5 (22.1, 28.2) | 25.5 (22.9, 29.5) | 24.9 (22.4, 28.6) | 24.6 (22.5, 27.9) | 25.1 (22.5, 28.6) |
| Smoking, (%) | 6.9 | 4.6 | 5.3 | 2.4 | 3.3 | 1.6 |
| White ethnicity, (%) | 95.8 | 98.0 | 96.6 | 95.2 | 96.7 | 98.5 |
| Educated to degree level or higher, (%) | 55.4 | 53.3 | 51.2 | 55.6 | 58.8 | 59.4 |
| 25(OH)D (nmol/l), mean (SD) | 45.4 (15.8) | 44.9 (15.7) | 45.2 (15.7) | 46.3 (16.5) | 45.3 (15.3) | 45.9 (16.4) |

**Table 2:** Maternal characteristics, anthropometry and DXA measures within 2 weeks of delivery by randomisation group (N=401)

|  |  |  |  |
| --- | --- | --- | --- |
|  | **Placebo** | **Cholecalciferol** | **p\*** |
| n | 197 | 204 |  |
| Age (years), mean (SD) | 197 | 31.7 (4.9) | 204 | 31.9 (4.8) | 0.74 |
| Gestation at delivery (weeks), median (SD) | 197 | 40.3 (39.3, 41.0) | 204 | 40.4 (39.4, 41.0) | 0.85 |
| Time since delivery of study pregnancy (days), median (IQR) | 197 | 7 (1, 10) | 204 | 7 (2, 11) | 0.41 |
| 25(OH)D in late pregnancy (nmol/l), mean (SD) | 178 | 42.7 (20.3) | 188 | 68.8 (19.2) | <0.001 |
| Height (cm), mean (SD) | 187 | 165.7 (6.7) | 198 | 165.5 (5.8) | 0.72 |
| Weight (kg), median (IQR) | 197 | 75.4 (12.5) | 203 | 74.8 (13.0) | 0.68 |
| BMI (kg/m2), median (IQR) | 187 | 26.7 (24.4, 30.3) | 197 | 26.7 (23.7, 30.2) | 0.63 |
| **Whole body** |  |  |  |  |  |
| BMC (g) | 197 | 2132 (256) | 203 | 2130 (262) | 0.94 |
| BA (cm2) | 197 | 1976 (158) | 203 | 1971 (147) | 0.71 |
| BMD (g/cm2) | 197 | 1.076 (0.069) | 203 | 1.078 (0.073) | 0.83 |
| **Whole body less head** |  |  |  |  |  |
| BMC (g) | 197 | 1621 (215) | 203 | 1618 (219) | 0.88 |
| BA (cm2) | 197 | 1752 (153) | 203 | 1745 (143) | 0.65 |
| BMD (g/cm2) | 197 | 0.922 (0.060) | 203 | 0.924 (0.063) | 0.85 |
| **Lumbar spine**  |  |  |  |  |  |
| BMC (g) | 197 | 60.7 (9.5) | 204 | 60.5 (9.7) | 0.83 |
| BA (cm2) | 197 | 58.5 (5.5) | 204 | 58.3 (5.1) | 0.70 |
| BMD (g/cm2) | 197 | 1.035 (0.105) | 204 | 1.034 (0.112) | 0.97 |
| **Total left hip** |  |  |  |  |  |
| BMC (g) | 197 | 31.2 (4.7) | 204 | 30.7 (4.8) | 0.33 |
| BA (cm2) | 197 | 33.1 (3.5) | 204 | 32.5 (3.1) | 0.09 |
| BMD (g/cm2) | 197 | 0.942 (0.097) | 204 | 0.943 (0.103) | 0.93 |

\* p is obtained with a t-test, Chi-squared test or Mann-Whitney U test

**Table 3:** Maternal characteristics, anthropometry and DXA measures at 4 years after delivery by randomisation group (N=443)

|  |  |  |  |
| --- | --- | --- | --- |
|  | **Placebo** | **Cholecalciferol** | **p\*** |
| n | 225 | 218 |  |
| Age (years), mean (SD) | 225 | 36.2 (4.8) | 218 | 36.3 (4.6) | 0.67 |
| Time since delivery of study pregnancy (years), median (IQR) | 225 | 4.1 (4.0, 4.2) | 218 | 4.1 (4.0, 4.1) | 0.32 |
| 25(OH)D in late pregnancy (nmol/l), mean (SD) | 200 | 42.0 (21.5) | 194 | 70.4 (19.0) | <0.001 |
| Height (cm), mean (SD) | 225 | 166.5 (6.3) | 218 | 165.8 (6.2) | 0.24 |
| Weight (kg), median (IQR) | 224 | 69.9 (63.7, 82.8) | 218 | 68.2 (60.4, 79.4) | 0.09 |
| BMI (kg/m2), median (IQR) | 224 | 24.9 (22.8, 29.5) | 218 | 24.9 (22.2, 29.1) | 0.24 |
| Other pregnancy since MAVIDOS pregnancy (yes), % | 223 | 31.8 | 215 | 37.7 | 0.20 |
| Attempted lactation for infant born from MAVIDOS pregnancy (yes), % | 223 | 58.7 | 218 | 69.3 | 0.02 |
| Duration of lactation for infant born from MAVIDOS pregnancy (completed months), median (IQR) | 223 | 0 (0, 7) | 218 | 2 (0, 10) | 0.01 |
| Duration of any lactation since MAVIDOS birth (completed months), median (IQR) | 224 | 1 (0, 11) | 218 | 3 (0, 13) | 0.01 |
| **Whole body** |  |  |  |  |  |
| BMC (g) | 183 | 2232 (268) | 193 | 2194 (285) | 0.19 |
| BA (cm2) | 183 | 1982 (153) | 193 | 1965 (155) | 0.27 |
| BMD (g/cm2) | 183 | 1.124 (0.078) | 193 | 1.114 (0.080) | 0.22 |
| **Whole body less head** |  |  |  |  |  |
| BMC (g) | 194 | 1731 (226) | 195 | 1707 (236) | 0.32 |
| BA (cm2) | 194 | 1751 (148) | 195 | 1736 (148) | 0.32 |
| BMD (g/cm2) | 194 | 0.986 (0.067) | 195 | 0.980 (0.066) | 0.38 |
| **Lumbar spine**  |  |  |  |  |  |
| BMC (g) | 217 | 63.2 (9.4) | 215 | 62.8 (9.8) | 0.67 |
| BA (cm2) | 217 | 60.3 (5.8) | 215 | 59.9 (5.7) | 0.50 |
| BMD (g/cm2) | 217 | 1.047 (0.104) | 215 | 1.046 (0.105) | 0.91 |
| **Total left hip** |  |  |  |  |  |
| BMC (g) | 224 | 32.5 (4.6) | 218 | 32.2 (4.7) | 0.46 |
| BA (cm2) | 224 | 33.5 (3.2) | 218 | 33.4 (3.0) | 0.69 |
| BMD (g/cm2) | 224 | 0.969 (0.103) | 218 | 0.962 (0.109) | 0.52 |

\* p is obtained with a t-test, Chi-squared test or Mann-Whitney U test

**Table 4:** Bone mineral density (BMD) within 2 weeks of delivery and at 4 years after delivery in women with DXA data at both time points, by randomisation to placebo or cholecalciferol in pregnancy(N=263)

|  |  |  |  |
| --- | --- | --- | --- |
|  | **Placebo** | **Cholecalciferol** | **Effect of cholecalciferol compared with placebo on maternal BMD at 4 years, accounting for BMD within 2 weeks of delivery**  |
| Site of BMD measure-ment | N | Within 2 weeks of delivery, mean (SD) | 4 years, mean (SD) | % change from birth to 4 years, mean (SD) | N | Within 2 weeks of delivery, mean (SD) | 4 years, mean (SD) | % change from birth to 4 years, mean (SD) | Unadjusted | Adjusted^ |
| β(g/cm2) | 95% CI | β (g/cm2) | 95% CI |
| Whole body | 111 | 1.079 (0.063) | 1.120 (0.073) a | 3.8 (3.2) | 120 | 1.078 (0.076) | 1.114 (0.080) a | 3.4 (3.5) | -0.002  | -0.019, 0.015 | -0.000 | -0.017, 0.017 |
| Whole-body-less-head | 117 | 0.922 (0.056) | 0.979 (0.063) a | 6.3 (2.7) | 121 | 0.924 (0.065) | 0.981 (0.068) a | 6.2 (2.9) | 0.001 | -0.014, 0.015 | 0.002 | -0.013, 0.016 |
| Lumbar spine | 126 | 1.033 (0.101) | 1.046 (0.103) b | 1.2 (4.6) | 132 | 1.038 (0.116) | 1.046 (0.107) | 1.0 (4.8) | -0.000 | -0.025, 0.025 | 0.003 | -0.022, 0.029 |
| Hip | 128 | 0.939 (0.085) | 0.960 (0.095) a | 2.2 (5.9) | 134 | 0.938 (0.104) | 0.960 (0.108) a | 2.4 (4.9) | -0.002 | -0.025, 0.021 | 0.002 | -0.021, 0.026 |

^ adjusted for maternal age at DXA, completed pregnancy since study pregnancy (yes/no), total duration of lactation since study pregnancy (completed months), age of study child. (a) Different compared to birth p<0.001 (b) Different compared to birth p<0.01

**References**

1. More C, Bhattoa HP, Bettembuk P, Balogh A. The effects of pregnancy and lactation on hormonal status and biochemical markers of bone turnover. Eur J Obstet Gynecol Reprod Biol. 2003;106(2):209-13.

2. Cross NA, Hillman LS, Allen SH, Krause GF, Vieira NE. Calcium homeostasis and bone metabolism during pregnancy, lactation, and postweaning: a longitudinal study. Am J Clin Nutr. 1995;61(3):514-23.

3. Cullers A, King JC, Van Loan M, Gildengorin G, Fung EB. Effect of prenatal calcium supplementation on bone during pregnancy and 1 y postpartum. Am J Clin Nutr. 2019;109(1):197-206. doi: 10.1093/ajcn/nqy233.

4. To WW, Wong MW, Leung TW. Relationship between bone mineral density changes in pregnancy and maternal and pregnancy characteristics: a longitudinal study. Acta Obstet Gynecol Scand. 2003;82(9):820-7. doi: 10.1034/j.1600-0412.2003.00227.x.

5. Ferreira LB, Tirapeli KG, Silva CC, Goldberg TBL. Lumbar spine bone mineral density in women breastfeeding for a period of 4 to 6 months: systematic review and meta-analysis. Int Breastfeed J. 2023;18(1):68. doi: 10.1186/s13006-023-00607-8.

6. To WW, Wong MW. Changes in bone mineral density of the os calcis as measured by quantitative ultrasound during pregnancy and 24 months after delivery. Aust N Z J Obstet Gynaecol. 2011;51(2):166-71. doi: 10.1111/j.1479-828X.2010.01266.x.

7. Ardawi MS, Nasrat HA, HS BAA. Calcium-regulating hormones and parathyroid hormone-related peptide in normal human pregnancy and postpartum: a longitudinal study. Eur J Endocrinol. 1997;137(4):402-9.

8. Curtis EM, Moon RJ, D'Angelo S, Crozier SR, Bishop N, Gopal-Kothandapani JS, et al. Maternal pregnancy vitamin D supplementation is associated with greater offspring bone mineral density at 4 years: findings from the MAVIDOS trial. Journal of Bone Mineral Research. 2019;34:68.

9. Brustad N, Garland J, Thorsen J, Sevelsted A, Krakauer M, Vinding RK, et al. Effect of High-Dose vs Standard-Dose Vitamin D Supplementation in Pregnancy on Bone Mineralization in Offspring Until Age 6 Years: A Prespecified Secondary Analysis of a Double-Blinded, Randomized Clinical Trial. JAMA Pediatr. 2020;174(5):419-27. doi: 10.1001/jamapediatrics.2019.6083.

10. Moon RJ, Green HD, D'Angelo S, Godfrey KM, Davies JH, Curtis EM, et al. The effect of pregnancy vitamin D supplementation on offspring bone mineral density in childhood: a systematic review and meta-analysis. Osteoporos Int. 2023;34(7):1269-79. doi: 10.1007/s00198-023-06751-5.

11. Moon RJ, S DA, Curtis EM, Ward KA, Crozier SR, Schoenmakers I, et al. Pregnancy vitamin D supplementation and offspring bone mineral density in childhood follow-up of a randomized controlled trial. Am J Clin Nutr. 2024;120(5):1134-42. doi: 10.1016/j.ajcnut.2024.09.014.

12. Curtis EM, Parsons C, Maslin K, D'Angelo S, Moon RJ, Crozier SR, et al. Bone turnover in pregnancy, measured by urinary CTX, is influenced by vitamin D supplementation and is associated with maternal bone health: findings from the Maternal Vitamin D Osteoporosis Study (MAVIDOS) trial. Am J Clin Nutr. 2021;114(5):1600-11. doi: 10.1093/ajcn/nqab264.

13. Jarjou LM, Sawo Y, Goldberg GR, Laskey MA, Cole TJ, Prentice A. Unexpected long-term effects of calcium supplementation in pregnancy on maternal bone outcomes in women with a low calcium intake: a follow-up study. Am J Clin Nutr. 2013;98(3):723-30. doi: 10.3945/ajcn.113.061630.

14. Jarjou LM, Laskey MA, Sawo Y, Goldberg GR, Cole TJ, Prentice A. Effect of calcium supplementation in pregnancy on maternal bone outcomes in women with a low calcium intake. Am J Clin Nutr. 2010;92(2):450-7. doi: 10.3945/ajcn.2010.29217.

15. Cooper C, Harvey NC, Bishop NJ, Kennedy S, Papageorghiou AT, Schoenmakers I, et al. Maternal gestational vitamin D supplementation and offspring bone health (MAVIDOS): a multicentre, double-blind, randomised placebo-controlled trial. Lancet Diabetes Endocrinol. 2016;4(5):393-402. doi: 10.1016/s2213-8587(16)00044-9.

16. Harvey NC, Javaid K, Bishop N, Kennedy S, Papageorghiou AT, Fraser R, et al. MAVIDOS Maternal Vitamin D Osteoporosis Study: study protocol for a randomized controlled trial. The MAVIDOS Study Group. Trials. 2012;13:13. doi: 1745-6215-13-13 [pii];10.1186/1745-6215-13-13 [doi].

17. Curtis EM, Moon RJ, D'Angelo S, Crozier SR, Bishop NJ, Gopal-Kothandapani JS, et al. Pregnancy Vitamin D Supplementation and Childhood Bone Mass at Age 4 Years: Findings From the Maternal Vitamin D Osteoporosis Study (MAVIDOS) Randomized Controlled Trial. JBMR Plus. 2022;n/a(n/a):e10651. doi: <https://doi.org/10.1002/jbm4.10651>.

18. Krupa C, Qamar H, O'Callaghan KM, Onoyovwi A, Al Mahmud A, Ahmed T, et al. Prenatal but not continued postpartum vitamin D supplementation reduces maternal bone resorption as measured by C-terminal telopeptide of type 1 collagen without effects on other biomarkers of bone metabolism. Endocr Metab Sci. 2024;14:None. doi: 10.1016/j.endmts.2023.100154.

19. Wei W, Shary JR, Garrett-Mayer E, Anderson B, Forestieri NE, Hollis BW, Wagner CL. Bone mineral density during pregnancy in women participating in a randomized controlled trial of vitamin D supplementation. Am J Clin Nutr. 2017;106(6):1422-30. doi: 10.3945/ajcn.116.140459.

20. Méndez-Sánchez L, Clark P, Winzenberg TM, Tugwell P, Correa-Burrows P, Costello R. Calcium and vitamin D for increasing bone mineral density in premenopausal women. Cochrane Database Syst Rev. 2023;1(1):Cd012664. doi: 10.1002/14651858.CD012664.pub2.

21. Olausson H, Laskey MA, Goldberg GR, Prentice A. Changes in bone mineral status and bone size during pregnancy and the influences of body weight and calcium intake1. The American Journal of Clinical Nutrition. 2008;88(4):1032-9. doi: <https://doi.org/10.1093/ajcn/88.4.1032>.

22. Moon RJ, D'Angelo S, Crozier SR, Curtis EM, Fernandes M, Kermack AJ, et al. Does antenatal cholecalciferol supplementation affect the mode or timing of delivery? Post hoc analyses of the MAVIDOS randomized controlled trial. J Public Health (Oxf). 2023;45(3):738-47. doi: 10.1093/pubmed/fdac160.

23. Citeroni-Clark NL, D'Angelo S, Crozier SR, Kermack A, Godfrey KM, Cooper C, et al. The effect of pregnancy vitamin D supplementation on maternal blood pressure: real-world data analysis within the MAVIDOS randomised placebo-controlled trial. Arch Gynecol Obstet. 2025. doi: 10.1007/s00404-025-07958-z.

24. El-Heis S, D'Angelo S, Curtis EM, Healy E, Moon RJ, Crozier SR, et al. Maternal antenatal vitamin D supplementation and offspring risk of atopic eczema in the first 4 years of life: evidence from a randomized controlled trial. Br J Dermatol. 2022;187(5):659-66. doi: 10.1111/bjd.21721.

25. Han SM, Huang F, Derraik JGB, Vickers MH, Devaraj S, Redeuil K, et al. A nutritional supplement during preconception and pregnancy increases human milk vitamin D but not B-vitamin concentrations. Clin Nutr. 2023;42(12):2443-56. doi: 10.1016/j.clnu.2023.09.009.

26. Barker M, Lawrence W, Crozier S, Robinson S, Baird J, Margetts B, Cooper C. Educational attainment, perceived control and the quality of women's diets. Appetite. 2009;52(3):631-6. doi: 10.1016/j.appet.2009.02.011.