25-Hydroxyvitamin D: functional outcomes in infants and young children

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ABSTRACT
Vitamin D deficiency occurs in the United States in exclusively breastfed infants who have high levels of skin pigmentation, inadequate vitamin D supplementation, and insufficient sunlight exposure. I review serum 25-hydroxyvitamin D [25(OH)D] concentrations and functional outcomes of vitamin deficiency in young children and breastfed and nonbreastfed infants. These outcomes include the presence or absence of vitamin D deficiency rickets, bone mineral content, and serum parathyroid hormone concentration. Daily vitamin D supplements of 400 IU/L keep serum 25(OH)D concentrations higher than 50 nmol/L and prevent rickets in infants and young children. The available evidence is not sufficient to support the use of bone mineral content or parathyroid hormone concentrations in infants and young children as functional outcomes to define deficient or sufficient levels of 25(OH)D. I therefore propose a research agenda to establish the functional definitions of vitamin D sufficiency or deficiency in infants and young children. Am J Clin Nutr 2008;88(suppl):529S–33S.

INTRODUCTION
The typical US diet contains few natural sources of vitamin D other than fatty fish and liver, which are foods not common in the diets of infants younger than 12 mo of age or young children aged 1–5 y. All US formulas for infants contain ≥400 IU of vitamin D/L. Thus, infants who consume ≥500 mL of formula per day receive 200 IU/d. The Institute of Medicine (1) considers this level adequate, but this is less than the 400 IU/d that the American Academy of Pediatrics (AAP; 2) and the Canadian Pediatric Society (3) currently recommend for infants and young children. For infants exclusively fed human milk, which has very little vitamin D (≈22 IU/L; 4), the AAP recommends a vitamin D supplement of ≥400 IU/d, but many breastfed infants in the United States do not take vitamin D supplements (2).

The skin naturally produces vitamin D from the reaction of sunlight (ultraviolet B irradiation) with 7-dehydrocholesterol; however, the multitude of variables that affect skin synthesis of vitamin D make it difficult to recommend a specific amount of sunlight exposure for all infants and young children (2). Furthermore, because of the relation of sun exposure to skin cancer, the Centers for Disease Control and Prevention, with the support of many organizations (including the AAP, the American Academy of Dermatology, and the American Cancer Society), has launched a campaign to increase public awareness about sunlight exposure and the risks of various skin cancers (5). Moreover, the AAP and the American Academy of Dermatology recommend that infants younger than 6 mo not be exposed to the sun and that all children and adolescents use sunscreens and clothing to protect them from ultraviolet B radiation exposure and to prevent skin cancer (6).

Newborn infants have a unique source of vitamin D: the mother, who transfers vitamin D to the fetus across the placenta (Figure 1; 7). Although cholecalciferol (the parent compound of vitamin D) does cross the placenta, maternal and fetal cholecalciferol blood concentrations are low and placental transfer contributes minimally to the vitamin D status of the fetus or newborn infant. Maternal 25-hydroxyvitamin D [25(OH)D] concentrations are higher than fetal concentrations, and this metabolite crosses the placenta in relatively large quantities. 25(OH)D from the mother is the major source of vitamin D for the fetus as well as newborn until the infant receives vitamin D from other dietary sources (such as infant formula) or supplements. 1,25-Dihydroxyvitamin D [1,25(OH)2D], the physiologically active metabolite, does not cross the placenta. However, the placenta can synthesize 1,25(OH)2D directly, and this might contribute to the infant’s circulating level of 1,25(OH)2D (Figure 1; 7).

It is important to note in the following review of the literature that the references cited cover a span of ≥3 decades. Most of these reports did not distinguish between measures of 25(OH)D3 from 25(OH)D2 (ie, most studies measured total 25(OH)D), whereas later studies may have reported values for both forms of 25(OH)D. As would be expected, changes to the method of measurement of 25(OH)D were made over this time period. This adds to the difficulty of interpreting and comparing the results of these studies done over a relatively long time period.

25-HYDROXYVITAMIN D CONCENTRATIONS IN NEWBORNS
According to numerous investigators, 25(OH)D concentrations in umbilical cord blood at the time of delivery range from 68% to 108% of maternal levels (8–12). Those authors reported a strong, positive correlation between infant and maternal serum 25(OH)D concentrations and bone mineral content in both infants and young children. A study of the newborn infant’s blood from the time of delivery found very similar levels of 25(OH)D to the levels in maternal blood, except for infants born to mothers with lower vitamin D levels (13). Infants born to African American mothers have lower vitamin D levels than those born to white mothers (14).enser 0.1% to 0.2% of maternal vitamin D levels (15).

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concentrations. We know that cord blood 25(OH)D concentrations in infants, as in maternal blood, vary seasonally; the seasonal impact on cord blood is greater in white than in African American infants (Table 1; 13–16). These ethnic differences are not surprising because mothers with more skin pigmentation or little exposure to sunlight tend to deliver infants with lower 25(OH)D concentrations, even if they take vitamin D supplements during pregnancy (11, 16). In one study from the Netherlands, >50% of pregnant women with dark skin pigmentation had serum 25(OH)D concentrations <25 nmol/L (17).

Maternal vitamin D supplements can have a positive effect on 25(OH)D concentrations in newborn infants. In one study, 1000–IU supplements of vitamin D in pregnant women raised serum 25(OH)D concentrations in the cord blood of their infants by 12–15 nmol/L compared with concentrations in unsupplemented controls (18–20). However, a maternal supplement of 400 IU/d (the amount typically present in prenatal vitamins) had little effect on infant cord blood concentrations (21).

**25-HYDROXYVITAMIN D CONCENTRATIONS IN INFANTS AND YOUNG CHILDREN**

Researchers have increasingly attempted to categorize 25(OH)D concentrations in adults as sufficient, insufficient, or deficient on the basis of functional outcomes related to bone health. These outcomes include osteomalacia, osteoporosis, bone mineral content, fractures, and falls. However, the field has not reached consensus on this issue. Researchers have also used the inverse correlation between 25(OH)D and parathyroid hormone (PTH) concentrations to define a normal 25(OH)D concentration in adults, because PTH concentrations do not depend directly on season, diet, or vitamin D intake.

One could potentially assess vitamin D deficiency rickets, bone mineral content, and the relation between 25(OH)D and serum PTH concentration as functional outcomes for determining bone health in relation to a normal 25(OH)D concentration in infants and young children. However, investigators have not studied sufficiently the relations between 25(OH)D concentrations and optimal intestinal calcium absorption or fracture rates in children to use these as functional outcomes for determining normal 25(OH)D concentrations.

Although 25(OH)D concentrations have been measured in infants younger than 1 y of age (discussed below), few studies have measured 25(OH)D concentrations in healthy US children aged 1 to 5 y. The National Health and Nutrition Examination Study (NHANES) study measured 25(OH)D concentrations in children aged 1–5 y for the first time in 2003–2004; a preliminary study found that mean concentrations were >55 nmol/L, which was the highest mean value for any age group measured in NHANES between 2000 and 2004 (22). The percentage of children aged 1–5 y in NHANES with 25(OH)D concentrations <25 nmol/L was very small (22). In a recent survey of 133 healthy children aged 6 and 22 mo from Alaskan Women, Infants, and Children clinics, 11% had a serum 25(OH)D concentration <37 nmol/L, and 20% had a concentration of 37–62 nmol/L (23). At the time of the survey, 30% of the children (n = 41) were still breastfeeding; they were more likely to have a 25(OH)D concentration <37 nmol/L (relative risk: 12; 95% CI: 3.6, 39).

**25-HYDROXYVITAMIN D CONCENTRATIONS IN VITAMIN D DEFICIENCY RICKETS**

Vitamin D deficiency rickets occurs in growing children who are typically vitamin D deficient for many months before a clinical diagnosis is made. In the United States, rickets occurs most commonly between the ages of 6 and 18 mo and is rarely reported.
in children older than 5 y (24). Clinicians use X-rays to confirm the diagnosis. The United States does not have a mandatory national reporting system for this diagnosis. Data on hospitalization rates are available, but less than one-half of all children with vitamin D deficiency rickets are hospitalized (24).

On the basis of the number of rickets case reports in the US literature in the past 15 y, some people believe that the diagnosis is becoming more common. However, the vast majority of these cases occur in children from ethnic minority groups, especially African American infants whose mothers breastfeed them exclusively without giving them vitamin D supplements. In North Carolina, clinicians reported 30 cases of rickets in breastfed African American infants between 1990 and 1999, including 17 cases reported between 1998 and 1999 (25). This increasing number of rickets cases from North Carolina corresponded with an increase in breastfeeding initiation rates from 5.2% to 34.7% between 1988 and 1998 in the North Carolina Women, Infants, and Children Clinics (25). Similarly, a recent article reported 166 cases of vitamin D deficiency rickets in US children younger than 5 y between 1986 and 2003 (24). Of these children, 83% were African American, and 96% were breastfed (24).

Does a threshold of serum 25(OH)D concentration exist below which rickets is likely to occur? The 1997 Institute of Medicine committee that issued recommendations for dietary reference intakes based primarily on data from the United States, Norway, and China determined that an intake of ≥200 IU/d of vitamin D prevented physical signs of vitamin D deficiency (rickets) and would maintain serum 25(OH)D concentrations >27.5 nmol/L in infants and young children (1). Most reports of 25(OH)D concentrations in children with vitamin D deficiency rickets come from developing countries around the world; a recent review summarized these reports (26). In 5 of the studies, the mean serum 25(OH)D concentration in patients with rickets was <27.5 nmol/L (27–31). However, 6 studies found a mean serum 25(OH)D concentration of 30–50 nmol/L in children with rickets (32–37). In many of these reports, calcium intake was a potentially confounding factor.

Only one of these reports was from North America. The authors of this report measured 25(OH)D concentrations in 9 children aged 2–42 mo with rickets in Canada and the northern part of the midwestern United States (32). In 3 infants with mild rickets, serum 25(OH)D concentrations averaged 46.7 ± 17.5 nmol/L, whereas in 5 infants with moderate rickets, 25(OH)D concentrations averaged (±SD) 29.9 ± 12.5 nmol/L. The remaining infant had severe rickets and a 25(OH)D concentration of 20.0 nmol/L. Not surprisingly, the authors found a negative correlation (r = −0.7) between serum 25(OH)D and PTH concentrations (32).

Because the available literature shows that vitamin D deficiency rickets occurs in infants and young children with a serum 25(OH)D concentration that is higher than 27.5 nmol/L, the evidence does not support using this serum concentration as a target for preventing clinical rickets. However, the studies used different assay methods, and future research should examine cases of rickets in infants around the world using a standardized 25(OH)D assay.

### BONE MINERAL CONTENT

Another functional outcome that some have proposed for identifying vitamin D deficiency in infants and young children is bone mineral content (BMC). Several BMC measurement techniques are potentially available: single-photon X-ray absorptiometry, dual-energy X-ray photon absorptiometry, quantitative ultrasound, and computerized tomography of bone.

Only 2 randomized controlled studies have examined the association between 25(OH)D concentration and BMC in North America (14, 38). Both used single-photon X-ray absorptiometry to measure BMC of the one-third distal radius in breastfeeding, non-Hispanic white infants. In the first study, the researchers randomly assigned breastfeeding infants to receive 400 IU vitamin D/d or a placebo and followed them for the first 6 mo of life (14, 38). In this study (Table 2), BMC increased significantly at 3 mo but not at 6 mo in the vitamin D group, but 25(OH)D concentrations at both 3 and 6 mo in this group were well above 50 nmol/L and significantly higher than in the placebo group. In the second and larger study (Tables 3 and 4), 25(OH)D concentrations were also well above 50 nmol/L (400 IU/d) in the vitamin D group and nearly twice as high as the concentration in the placebo group at 6 wk, 3 mo, and 6 mo of age (14). Significant differences noted in BMC at 6 mo were inconsistent with higher vitamin D intakes, because BMC was higher in the placebo group than in the vitamin D group at this time point (Table 4; 14).

Thus, the results of the limited studies of the relation between 25(OH)D concentrations and BMC in US infants are inconsistent and do not support using BMC as an indicator of vitamin D status.

### TABLE 2

<table>
<thead>
<tr>
<th>Time</th>
<th>400 IU Vitamin D (n = 9)</th>
<th>Placebo (n = 9)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>25(OH)D</td>
<td>BMC</td>
</tr>
<tr>
<td>3 mo</td>
<td>95</td>
<td>79 ± 3</td>
</tr>
<tr>
<td>6 mo</td>
<td>82</td>
<td>75 ± 5</td>
</tr>
</tbody>
</table>

1 Adapted from reference 38.
2 P < 0.01.
3 P < 0.003.
4 x ± SE (all such values).

### TABLE 3

<table>
<thead>
<tr>
<th>Time</th>
<th>400 IU Vitamin D</th>
<th>Placebo</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>25(OH)D</td>
<td>PTH</td>
</tr>
<tr>
<td>6 wk</td>
<td>76 ± 24</td>
<td>48 ± 36</td>
</tr>
<tr>
<td>3 mo</td>
<td>97 ± 26</td>
<td>52 ± 23</td>
</tr>
<tr>
<td>6 mo</td>
<td>92 ± 30</td>
<td>47 ± 26</td>
</tr>
</tbody>
</table>

1 All values are x ± SE. Adapted from reference 14.
2 For vitamin D group, n = 24. For placebo group, n = 22.
3 P < 0.01.
4 For vitamin D group, n = 24. For placebo group, n = 22.
5 P < 0.001.
6 For vitamin D group, n = 19. For placebo group, n = 19.
TABLE 4
Bone mineral content (BMC) in breastfed infants taking or not taking vitamin D supplements

<table>
<thead>
<tr>
<th>BMC</th>
<th>mg/cm</th>
</tr>
</thead>
<tbody>
<tr>
<td>400 IU Vitamin D/d</td>
<td>Placebo</td>
</tr>
<tr>
<td>1.5 mo</td>
<td>81 ± 12</td>
</tr>
<tr>
<td>3 mo</td>
<td>80 ± 13</td>
</tr>
<tr>
<td>6 mo</td>
<td>89 ± 12</td>
</tr>
<tr>
<td>Change in BMC between 1.5 and 6 mo</td>
<td>9 ± 13</td>
</tr>
</tbody>
</table>

All values are mean ± SE. Researchers measured BMC by single photon absorptiometry. Adapted from reference 14.

VITAMIN D AND BREASTFED INFANTS

At present, vitamin D deficiency rickets in the United States is almost exclusively a disease of breastfed infants who do not receive vitamin D supplements and who have limited sun exposure. The Institute of Medicine report stated that 200 IU vitamin D maintains 25(OH)D concentrations >27.5 nmol/L, the lowest point in the institute’s recommended normal range (1). However, many US breastfed infants do not receive supplemental vitamin D. In 1999–2002, NHANES found that only 8.7 ± 1.3% of infants (aged 0–12 mo) were receiving supplemental vitamin D at the time of the survey (39); most of the infants in the survey were receiving formula fortified with vitamin D, so they did not need vitamin D supplements. Only 308 of 1964 children aged 1–18 y in the survey (15.7%) had been breastfed at any time during the first 2 y of life (personal communication, Mary Frances Picciano, Office of Dietary Supplements, October 2007).

Another cross-sectional study measured 25(OH)D concentrations during the first 9 mo of life in 84 breastfed infants (83% white) born during the winter months (October–March) in Iowa (40). The infants were exclusively breastfed at enrollment but could receive supplemental formula and certain foods after 112 d of age. At age 112 d, 70% had a serum 25(OH)D concentration <27.5 nmol/L; the proportion declined steadily with age and reached 23% by age 280 d, presumably because these children were beginning to eat solid foods or drink formula, although the survey did not collect these data (Table 5). Only 13% of the infants in this study received vitamin D supplements. The researchers also measured serum PTH concentrations and found that their correlation with 25(OH)D concentrations was low (r² = 0.06; 40). This finding was consistent with previous data from Wisconsin showing similar 25(OH)D concentrations in exclusively breastfed infants who did not receive vitamin D supplements during the first 6 mo of life (Tables 3 and 4; 14, 38). PTH concentrations did not differ in the placebo and vitamin D supplemented groups in the Wisconsin study (Table 4; 14). These results imply that PTH is not a good indicator of vitamin D deficiency.

Supplements of 400 IU vitamin D will maintain serum 25(OH)D concentrations well above 70 nmol/L in breastfed infants, who as noted are at the highest risk of vitamin D deficiency (2, 14). This will prevent rickets in this population. The 1999–2004 NHANES and the cases of rickets in breastfed infants reported in North Carolina show that most breastfed infants in the United States do not receive vitamin D supplements (25, 39, 41), even though 50 y of clinical experience in the United States has shown that 400 IU of vitamin D not only prevents but also treats rickets (42). About 50% of the 4 million infants born in the United States each year are eligible for the Special Supplementation nutrition Program for Women, Infants, and Children (WIC). Although WIC clinics have increased their efforts to promote breastfeeding (25), WIC does not provide breastfeeding infants with 400 IU vitamin D/d, as recommended by the AAP and others, which places this population of infants at unnecessary risk of rickets (2).

CONCLUSIONS

Good evidence exists that many breastfed infants not supplemented with vitamin D during the first 6 mo of life have serum vitamin D concentrations <50 nmol/L and are therefore at increased risk of rickets. This is especially true for infants who have high skin pigmentation and little sun exposure. Good evidence also shows that supplements of 400 IU vitamin D/d keep 25(OH)D concentrations higher than 70 nmol/L and prevent rickets in white infants. Data are insufficient to determine whether 400 IU/d is the optimal level of supplementation for infants with darker skin pigmentation. The evidence is currently insufficient to recommend using BMC or serum PTH levels as functional measures of vitamin D sufficiency or deficiency states in infants and young children.

I recommend the following areas of research to further define vitamin D sufficiency and deficiency states in infants and young children:

1) Vitamin D deficiency rickets occurs much more frequently in US children aged 1–5 y than in older children. In these younger children, we should measure functional outcomes in cross-sectional and longitudinal descriptive studies of 25(OH)D concentrations [measured by using a standardized assay (43)] in relation to rickets, BMC, and PTH levels. This should be done in various ethnic groups and at different times of the year.

2) We need randomized trials comparing the outcomes from supplementation with 200 IU [as recommended by the Institute of Medicine (1)] with outcomes from supplementation with 400 IU [as recommended by the AAP for infants and children (2, 42)] vitamin D per day in any ethnic group. The functional outcomes to measure include the presence or absence of rickets, BMC, serum 25(OH)D concentrations, and serum PTH concentrations.

TABLE 5
Plasma 25-hydroxyvitamin D (25(OH)D) concentrations in 35 unsupplemented breastfed infants in Iowa

<table>
<thead>
<tr>
<th>Age (d)</th>
<th>25(OH)D</th>
<th>No. of infants with 25(OH)D &lt; 27.5 nmol/L</th>
</tr>
</thead>
<tbody>
<tr>
<td>112 (n = 33)</td>
<td>21.5 ± 23.0</td>
<td>112 (23)</td>
</tr>
<tr>
<td>168 (n = 35)</td>
<td>32.2 ± 26.2</td>
<td>168 (57)</td>
</tr>
<tr>
<td>224 (n = 33)</td>
<td>43.2 ± 25.0</td>
<td>224 (33)</td>
</tr>
<tr>
<td>280 (n = 35)</td>
<td>54.4 ± 27.4</td>
<td>280 (23)</td>
</tr>
</tbody>
</table>

Adapted from reference 40.
3) To functionally define vitamin D deficiency, insufficiency, and sufficiency in infants and young children, we need additional randomized controlled trials with vitamin D intakes >400 IU/d.
4) To determine the optimal levels of 25(OH)D and BMC in newborn infants and fetuses, we need randomized controlled trials of maternal vitamin D supplements that exceed 400 IU/d.

The author had no conflicts of interest to report.

REFERENCES