Intermittent high-dose vitamin D prophylaxis during infancy: effect on vitamin D metabolites, calcium, and phosphorus

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ABSTRACT In infants receiving intermittent high dose vitamin D prophylaxis (600,000 IU ergocalciferol per dose orally) every 3-5 mo, the serum concentrations of vitamin D metabolites, calcium (Ca), and phosphorus (P) were determined before and 2 wk after each dose. The 25-hydroxyvitamin D (25-OHD) concentrations increased to well above normal but the values returned to the normal range before each subsequent dose. The 24,25- and 25,26-dihydroxyvitamin D ([24,25-OH]D) levels followed a pattern similar to that of 25-OHD, and both were closely related to the latter (r = 0.85, p < 0.005, and r = 0.84, p < 0.005, respectively). The 1,25-(OH)2D concentrations did not vary in a consistent pattern and remained largely within the normal range. All infants had normal Ca levels before the first dose but 14 infants (34%) later had one or both Ca values above the upper normal limit of 2.80 mmol/L (2.81-3.32 mmol/L), indicating that the vitamin D doses were excessive despite the lack of accumulative increases in serum vitamin D concentrations.

Material and methods Venous blood was collected from 43 infants immediately before and 2 wk after the routine oral dose of 600,000 IU (15 mg) ergocalciferol at the ages of 1 (n = 9), 4 (n = 4), 7 (n = 8), 11 (n = 13), 15 (n = 6), and 20 (n = 3) mo. Infants studied after age 1 mo had received all the previous ergocalciferol doses according to this intermittent high-dose vitamin D program. All were born at term and were clinically healthy. They received the vitamin D doses according to the official national rickets prevention program in GDR. Informed consent for blood sampling was obtained from the parents, and the study was approved by the medical faculty of the University of Jena.

The samples were collected in late autumn to early winter (October-December). None of the children received other oral vitamin D supplements and no food items, including formula milk, were fortified with this vitamin. The young infants received

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The safety of this regimen has been questioned, however, because toxic effects of very high doses of vitamin D now are well recognized and because a high incidence of hypercalcemia was noted following even smaller doses of cholecalciferol (3). It has been suggested that high doses of vitamin D may damage the vascular system and thus predispose for atherosclerosis (5, 6).

For decades all infants in the German Democratic Republic (GDR) have received six doses of 600,000 IU (15 mg) ergocalciferol orally divided over the first 1.5 yr of life. The purpose of this investigation is to study the effect of this program on the serum levels of vitamin D metabolites, calcium (Ca), and phosphorus (P).
formula milk alone or as a supplement to breast milk. None were solely breast-fed.

The vitamin D metabolites 25-hydroxyvitamin D (25-OHD), 1,25-dihydroxyvitamin D (1,25-(OH)\(_2\)D), 24,25-dihydroxyvitamin D (24,25-(OH)\(_2\)D), and 25,26-dihydroxyvitamin D (25,26-(OH)\(_2\)D) were determined in 1 mL of serum according to previously described methods (7, 8). 25-hydroxyergocalciferol and 25-hydroxycholecalciferol were separated before quantitative analysis but the dihydroxy metabolites of ergocalciferol and cholecalciferol origin were assessed together after the purification processes. The binding assays are equipotent with regard to 25-hydroxyergocalciferol and 25-hydroxycholecalciferol (8) while 1,25-dihydroxyergocalciferol has a displacement potency which is lower than that of 1,25-dihydroxycholecalciferol by a factor of 1.3 (9). However, we found previously that this variance tends to be abolished by apparent differences in the regulation of circulating 1,25-dihydroxyergocalciferol vs 1,25-dihydroxycholecalciferol levels (9) and no adjustments for this factor were made. The potencies of the other dihydroxyergocalciferol vs cholecalciferol metabolites in the competitive protein binding assays are not known. Serum concentrations of Ca and P were determined on a DuPont Clinical Analyzer (DuPont, Wilmington, DE). For each infant, predose and postdose samples were analyzed in the same assay to avoid interassay variability.

Reference values for the biochemical indices were obtained from Norwegian infants who were on no vitamin D supplementation (studied during summer) or on 200–400 IU/d (studied during winter). The normal range for the various substances were 25-OHD: 20–130 nmol/L, 1,25-(OH)\(_2\)D: 60–215 pmol/L, 24,25-(OH)\(_2\)D: < 0.4–6.0 nmol/L, 25,26-(OH)\(_2\)D: < 0.3–2.0 nmol/L, Ca: 2.35–2.80 mmol/L, and P: 1.60–2.40 mmol/L (10, 11).

The data were expressed as medians with a range, and differences were tested for statistical significance with Wilcoxon’s test for unpaired (comparing results from different age groups) and paired data (comparing pretreatment and posttreatment results for each age group). Correlations were assessed by linear regression analysis.

Results

Before the initiation of ergocalciferol prophylaxis at age 1 mo, five of the nine infants were vitamin D deficient (25-OHD < 20 nmol/L) and three infants had extremely low levels (25-OHD < 10 nmol/L, Fig 1). No infant had low Ca or P levels or clinical signs of rickets. Before each subsequent dose all infants had 25-OHD values within the normal range and the median level did not vary (Fig 1). The median level at 4 wk (17 nmol/L) was, however, significantly lower than later predose values \((p = 0.01)\).

25-hydroxyergocalciferol was nondetectable or present in trace concentrations before the first dose but accounted for the median of 70–81% of the total 25-OHD level before each subsequent dose (Fig 2).

The 25-OHD levels increased markedly within 2 wk of each dose of ergocalciferol \((p < 0.001)\) and the values were invariably above normal limits. The median postdose
The 1,25-(OH)₂D concentration increased in all the infants after the first dose (medians 80 vs 163, p < 0.005, Fig 3). There were no consistent patterns following later doses, and the values remained largely within normal limits (Fig 3).

There was a close positive relationship between 25-OHD and 1,25-(OH)₂D levels before the first vitamin D dose at 4 wk (r = 0.78, p < 0.0005) and an inverse association between 25-OHD before treatment and the magnitude of increase in 1,25-(OH)₂D concentration after the first dose (r = -0.61, p < 0.01). Later there were no significant relationships between the two metabolites.

The serum concentrations of 24,25-(OH)₂D and 25,26-(OH)₂D followed a pattern similar to that of 25-OHD and both dihydroxy metabolites were closely related to 25-OHD (Figs 4 and 5).

None of the infants were hypercalcemic before the first dose but 14 of the treated infants (34%) later had one or both Ca values above the high normal limit (Fig 6). The infants with high Ca levels did not differ from the others with respect to the vitamin D metabolites.

The serum concentration of P increased significantly after the first dose of vitamin D (p = 0.02, Fig 7). Five infants (12%) had elevated P levels after the start of treatment; four (all three with high P values after the dose at 1 mo and the infant at 7 mo) also had hypercalcemia. There were, however, no significant correlations between serum P and Ca or between serum P and vitamin D metabolite levels at any age.

Discussion

The serum concentration of 25-OHD is an accepted index of vitamin D nutritional status since it is directly related to the oral intake of vitamin D and to exposure to sunshine (12). 1,25-(OH)₂D is the hormonal form of vitamin D and its serum concentration is normally regulated independently of the 25-OHD level to maintain Ca and P homeostasis (12). Low levels of 25-OHD may limit the rate of 1,25-(OH)₂D synthesis on the basis of substrate deficiency (11). The physiological significance of 24,25-(OH)₂D and 25,26-(OH)₂D has not been established but both may represent degradation products of 25-OHD (13).

Low serum concentrations of 25-OHD and the close relationship between 25-OHD and 1,25-(OH)₂D levels before treatment at 4 wk and the rise in initial 1,25-(OH)₂D concentrations following treatment indicate clearly that the majority of the infants were vitamin D deficient although they had not yet developed rickets (11). Routine vitamin D supplementation during winter therefore seems warranted for infants in GDR as in other countries with similar climatic conditions (14, 15).

Although one oral dose of 600,000 IU ergocalciferol resulted in 25-OHD levels far above normal limits and often in concentrations considered to be in the toxic range (16, 17), there was no evidence of cumulative increases in serum levels of 25-OHD or any of its metabolites during the prophylactic program. It is also unlikely that the serum concentration of unmetabolized vitamin D increased progressively since the level of this precursor generally decreases before that of 25-OHD (18).
FIG 3. Serum concentrations of 1,25-(OH)2D before (B) and after (A) each dose. Median values and normal range are plotted.

FIG 4. Relationship between simultaneously obtained 25-OHD and 24,25-(OH)2D levels. The detection limit for 24,25-(OH)2D is plotted as a broken line.
FIG 5. Relationship between simultaneously obtained 25-OHD and 25,26-(OH)₂D levels. The detection limit for 25,26-(OH)₂D is plotted as a broken line.

Calcium
(mmol/l)

FIG 6. Serum concentrations of Ca before (B) and 2 wk after (A) each oral dose of ergocalciferol. Median values and normal range are plotted.
The parallel increase in the levels of 25-OHD and the dihydroxy metabolites 24,25-(OH)₂D and 25,26-(OH)₂D, which probably have a much shorter half life than 25-OHD (19), supports previous suggestions that elevated 25-OHD concentrations lead to an increased metabolism of 25-OHD through these dihydroxy metabolites (20) as well as through other pathways (21). The increased metabolic rate was apparently adequate to inhibit accumulations of 25-OHD. On the other hand the 25-hydroxyergocalciferol concentrations were still high 3–5 mo after the last dose, demonstrating a long term effect of each dose of ergocalciferol. Because 25-OHD concentrations seem to decline at a slower rate with lower 25-OHD levels (18, 20), it is likely that smaller or less frequent doses of ergocalciferol would have given adequate protection against vitamin D deficiency.

The 1,25-(OH)₂D concentrations were maintained within the normal range and were unaffected by the tremendous variations in 25-OHD levels after vitamin D deficiency had been corrected. This observation contradicts the suggestion by Stern et al (22) that the circulating 1,25-(OH)₂D concentration is loosely regulated in young children.

Nevertheless, one third of the infants aged > 4 wk had hypercalcemia at least on one occasion and as often before as after subsequent doses. However, vitamin D metabolites other than 1,25-(OH)₂D and especially 25-OHD may stimulate intestinal absorption and bone release of Ca and P when present in high concentrations and thus be responsible for the toxic effects of vitamin D (16, 23, 24).

From this study it is also possible that a failure of induced hypercalcemia to suppress markedly 1,25-(OH)₂D synthesis may be partly responsible for persistently high Ca levels even after the concentrations of the various vitamin D metabolites have returned to the normal range.

There is historical evidence for variations in individual susceptibility to the toxic effects of vitamin D (25). In patients with Williams syndrome, increased sensitivity to vitamin D may be due to increased conversion of the parent vitamin to 25-OHD (26) or to increased 1,25-(OH)₂D levels (27). This study suggests that the susceptibility to hypercalcemia for any given serum concentration of vitamin D metabolites may vary considerably among infants.

Although all the infants appeared healthy and repeated inquiries in GDR have failed to identify clinical vitamin D toxicity as a result of the prophylactic program, hypervitaminosis D and hypercalcemia may have untoward long term effects on cerebral, renal, and cardiovascular function (28). A renewed interest has arisen in intermittent high-dose vitamin D prophylaxis in defined risk groups (29). Intermittent ergocalciferol prophylaxis providing one oral dose of 600 000 IU every 3–5 mo during the first 1.5 y of life is clearly excessive and must be considered unsafe.

References