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Efficacy of micellized vs. fat-soluble vitamin D3 supplementation in healthy school children from Northern India

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Abstract

Background: Vitamin D deficiency is a widely recognized public health problem. Efficacy of a recently developed micellized form of vitamin D3 has not been studied. Hence, we undertook this study to compare its efficacy with the conventionally used fat-soluble vitamin D3.

Methods: In this open-labeled nonrandomized pilot study, we recruited 180 healthy children, aged 13–14 years in two groups and supplemented Group A (60 children) with 60,000 IU of fat-soluble vitamin D3/month with milk and Group B (120 children) with 60,000 IU/month of water miscible vitamin D3 under supervision for 6 months. Serum 25(OD)D, parathyroid hormone (PTH), calcium, phosphate, and alkaline phosphatase (ALP) levels were evaluated before and after supplementation in 156 children (54 in Group A and 102 in Group B) who completed the study.

Results: We observed a significantly greater increase in the serum 25(OH)D levels in group B as compared to group A (31.8 ± 9.1 ng/mL vs. 23.7 ± 10.4 ng/mL; $p < 0.001$). All children in group B achieved adequate levels of serum 25(OH)D (> 20 ng/mL) as against 83.3% children in group A. Serum PTH and ALP levels declined considerably in both the groups following supplementation.

Conclusions: Vitamin D supplementation significantly increased the serum 25(OH)D levels in both groups. Miscible form of vitamin D3 appears to be better in achieving higher levels of serum 25(OH)D than that observed with a similar dose of fat-soluble vitamin D3. Further studies with different dose regimens are required to establish its efficacy over the conventionally used fat-soluble vitamin D3.

Keywords: children; fat-soluble vitamin D3; micellized vitamin D3; supplementation.

Introduction

Vitamin D has been shown to be associated with several beneficial effects for humans of all ages. Until recently, vitamin D deficiency (VDD) was believed to be associated mostly with a risk of developing rickets and osteomalacia. Current research, however, shows VDD as a potential risk factor for several chronic diseases across all age and gender groups from India and other parts of the world [1, 2]. An increasing body of evidence prompted the development of nationwide recommendations to prevent VDD in Poland (2009), Hungary (2012), and Germany–Austria–Switzerland (2012) [3–5].

Recently, the Institute of Medicine (IOM) revised its guidelines for vitamin D intake in children and adults in the United States and Canada. A Recommended Dietary Allowance (RDA) of 600 IU/d was set for children older than 12 months with the goal of achieving a serum 25-hydroxyvitamin D [25(OH)D] concentration of > 20 ng/mL [6]. Endocrine Society (ES) treatment guidelines targeted higher levels of vitamin D and suggested treatment with 2000 IU/day of vitamin D for atleast 6 weeks or 50,000 IU once a week for at least 6 weeks to achieve a serum level of 25(OH)D > 30 ng/mL,

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followed by maintainence dose of 600–1000 IU/day in children who are 1–18 years of age [7].

Achieving serum 25(OH)D concentrations of around 20 ng/mL in all children would clearly be difficult without either a comprehensive food fortification strategy or widespread use of vitamin D supplements. Studies indicate that only 50% of a typical dosage of vitamin D is absorbed from the intestinal lumen owing to complex absorption process [8]. Micellization is a new delivery system for fat-soluble nutrients that disperses fatty substances into microscopic, water-soluble and micellar spheres enabling them to reach the absorptive surface of the intestinal tract, facilitating maximum absorption [9]. In view of the absence of data in humans with regards to efficacy of micellized vitamin D3 and limited supplementation studies in children both from India and abroad, we undertook this study to compare the efficacy of micellized vitamin D3 to the conventionally used fat-soluble vitamin D3.

Materials and methods

Study participants

The present open-labeled nonrandomized study was undertaken from January through July 2015 in two schools located in New Delhi based on the permission granted by the school management. Parents of children from all sections of class 8 from both the schools were provided with a detailed information sheet about the study protocol and informed written consent was obtained. Children with any systemic illnesses or those taking medications or supplements that interfere with the evaluation of bone mineral metabolic parameters like anticonvulsants, anti tubercular drugs, barbiturates, vitamin D, and steroids were excluded from the study. A total of 180 apparently healthy school children in the age group of 13–14 years (35 boys and 145 girls) were recruited as study subjects. All the girls had attained menarche and the boys were in pubertal stage 2/3. None of the children had received any calcium or vitamin D supplements prior to the study. The study was approved by the Institute Ethics Committee and was carried out in accordance with the principles of the Declaration of Helsinki.

Intervention

The study participants were divided into two groups based on their school for convenience. Group A (School A) received fat-soluble vitamin D3 (Calcirol 60,000 IU, Cadila Pharmaceuticals, India) once a month with unfortified milk containing 4% fat while Group B (School B) received water-miscible vitamin D3 (DePura 60,000 IU, Sanofi Pharmaceuticals, India) once a month for 6 months under supervision in the respective schools. Group A comprised 35 boys and 25 girls while Group B had 120 girls. In the absence of any significant differences in the pre- and post supplementation hormonal and biochemical values, between boys and girls, we grouped them together for analysis.

Biochemical analyses

Blood samples were collected at baseline and 1 week following the last supplementation dose for analysis of serum 25(OH)D, parathyroid hormone (PTH), calcium, phosphate, and alkaline phosphatase (ALP). Serum 25(OH)D was measured by chemiluminiscence (Diasorin, Stillwater, MN, USA) and PTH was measured using electrochemiluminiscence method (Roche Diagnostics, GmbH). Calcium, phosphate and ALP were estimated by auto analyzer. Severity of vitamin D deficiency was graded as severe (25(OH)D < 5 ng/mL); moderate (25(OH)D 5–10 ng/mL) and mild (25(OH)D 10–20 ng/mL) as per Lips classification [10]. Serum levels 25(OH)D > 20 ng/mL was considered adequate. Secondary hyperparathyroidism was defined as serum PTH values > 65 pg/mL. Spot urine analysis for calcium-creatinine ratio was carried out only in postsupplementation samples to observe for any hypercalciuria. Urinary Ca-Cr ratio > 0.29 was considered abnormal [11].

Statistical analysis

All continuous variables are expressed in terms of mean ± standard deviation (SD). Paired t-test was used to compare the mean 25(OH) D, PTH, calcium, phosphate and ALP levels, before and after vitamin D3 supplementation. For parameters where the SD was large relative to the mean, values were log transformed before the analysis. A p-value of < 0.05 was considered as significant. All statistical analysis was implemented on Stata 12.1 (Stata Corp, College Station, TX, USA).

Results

Out of the 180 children that were recruited, 156 children [(32 boys and 22 girls girls from Group A) and (102 girls from Group B)] completed the study as per the protocol. Twenty-four children [16 subjects (three boys and three girls) from Group A and 18 girls from group B] were excluded from further analysis due to noncompliance.

Baseline characteristics of hormonal and biochemical parameters

The overall prevalence of vitamin D deficiency was observed in 98.7% subjects with secondary hyperparathyroidism in 50%. Furthermore, mild, moderate, and severe vitamin D deficiency was noted in 10.3%, 7.7%, and 80.8%, respectively. No significant difference in the prevalence of mild (11.1% vs. 9.8%), moderate (13.0% vs. 5.0%), and severe (74.1% vs. 84.3%) vitamin D deficiency was observed between the two Groups A and B. In addition,

the mean baseline serum 25(OH)D, PTH, calcium, and PO_4 levels between the two study groups were no different. The mean baseline serum ALP levels, however, were significantly higher in Group A (Table 1). All subjects were asymptomatic and did not reveal any bony abnormalities such as genu varum or genu valgum on clinical examination.

Impact of vitamin D3 supplemenation on serum 25(OH)D, PTH, calcium, phosphate, and ALP

Following supplementation, a significant increase was observed in the overall mean serum 25(OH)D levels (5.6 ± 3.7 ng/mL to 34.5 ± 9.6 ng/mL ($p < 0.001$) with the rise in group B being higher as compared to that in Group A (31.8 ± 9.1 ng/mL vs. 23.7 ± 10.4 ng/mL; $p < 0.001$). The number of subjects who achieved adequate serum 25(OH)D levels (> 20 ng/mL) postsupplementation in group A and B were 45 (83.3%) and 102 (100%), respectively ($p < 0.001$). Nine (16.7%) children from Group A continued to remain vitamin D deficient (25(OH)D < 20 ng/mL).

The mean baseline serum PTH decreased significantly from 91.8 ± 131.0 pg/mL to 41.2 ± 25.0 pg/mL ($p < 0.001$) with the prevalence of secondary hyperparathyroidism declining from 50% to 7.1% following supplementation. Though no significant difference was observed in serum calcium in either group pre- and postsupplementation, there was a marked overall decline in the serum ALP levels, primarily because of significant decline in group B. Table 1 summarizes the mean values of biochemical parameters in pre- and postsupplementation in both the groups.

Despite achieving adequate serum 25(OH)D levels following supplementation (4.0 ± 0.04 ng/mL vs. 30.9 ± 9.5 ng/mL; $p < 0.001$), 11 subjects (six from Group A and five from group B) continued to have serum PTH levels above the normal range. However, these subjects did show a significant decrease in the baseline serum PTH levels (222.0 ± 154.9 pg/mL to 100.0 ± 53.6 pg/mL; $p = 0.04$) and an appreciable decrease in the serum ALP levels (311.9 ± 200.8 IU to 251.9 ± 142.1 IU, $p = 0.16$) following supplementation.

None of the children except one in the fat-soluble group developed hypercalciuria (Urinary Ca/Cr ratio > 0.29).

Discussion

In view of poor sun exposure, negligible intake of vitamin D through the diet and absence of government policy

Table 1: Mean hormonal and biochemical Parameters Pre and Post Vitamin D supplementation.

Biochemical parameter	Overall (n = 156)		p-Value	Group A (n = 54) (fat soluble)		p-Value	Group B (n = 102) (water miscible)		p-Value
	Pre	Post		Pre	Post		Pre	Post	
25(OH)D	5.6 ± 3.7	34.5 ± 9.6	< 0.001	6.1 ± 4.3	29.8 ± 10.2	< 0.001	5.3 ± 3.4	37.1 ± 8.3	< 0.001
PTH	91.8 ± 131.0	41.2 ± 25.0	< 0.001	118.8 ± 205.2	45.0 ± 20.8	0.009	77.3 ± 58.0	39.1 ± 26.9	< 0.001
Calcium	9.8 ± 0.4	9.8 ± 0.5	0.60	9.7 ± 0.4	9.7 ± 0.5	0.70	9.9 ± 0.4	9.9 ± 0.5	0.70
Phosphate	4.1 ± 0.5	4.3 ± 0.5	< 0.001	4.1 ± 0.5	4.4 ± 0.5	< 0.001	4.1 ± 0.4	4.2 ± 0.6	0.07
Alkaline phosphatase	210.5 ± 123.7	170.1 ± 90.9	< 0.001	282.7 ± 162.2	251.2 ± 101.0	0.13	171.9 ± 72.8	126.7 ± 43.2	< 0.001

with regards to fortification of foods with vitamin D, supplementation would play an important role in the management of widespread vitamin D deficiency, especially in India [12, 13]. Till date, most supplementation studies in children and adults were carried out using fat-soluble vitamin D3. The first Indian study documenting effectiveness of monthly and bimonthly supplementation of 60,000 IU of cholecalciferol (Calcirol) with water in school girls given for a period of 1 year under supervision showed only a rise of 7.6 ng/mL [14]. However, a recent study by Kuchay et al. with a similar dose of vitamin D supplementation with milk for 1 year showed a much higher rise in serum 25(OH)D levels (21.1 ng/mL). This profound difference could probably be due to the fact that fat-soluble vitamin D3 is better absorbed when given with milk than when supplemented with water [15].

Recently, micellization of fat-soluble nutrients has been shown to markedly enhance their absorption from the gut. Studies have revealed 3.6 times greater mucosal absorption of vitamin A in subjects provided with micellized form than those who received emulsions [16]. Similarly, in a randomized cross-over trial, 12 healthy subjects when administered 500 IU of vitamin E in standard oil, emulsified, or micellized form showed plasma level of vitamin E after 4 h of supplementation to be five times greater in micellized group than in the oil group [17].

Using micellized form of vitamin D3 (Group B), we observed a significantly higher rise of mean serum 25(OH) D levels (31.8 ng/mL) as against those who were supplemented fat-soluble vitamin D3 (Group A) with milk (23.7 ng/mL) in the present study and also in a report by Kuchay et al. [15] (21.1 ng/mL). Moreover, all study subjects (100%) in group B achieved serum 25 (OH) D levels of >20 ng/mL as against 83.3% in group A. Significantly higher number of subjects from group B (78.4%) achieved more than >30 ng/mL as against 48.2% in group A and 60.9% in another recent study [15]. These observations are suggestive of better absorption of micellized form of vitamin D3.

Secondary hyperparathyroidism noted in vitamin D deficient subjects (50%) in this study is consistent with two other earlier studies which showed secondary hyperparathyroidism in 30–40% of vitamin D deficient subjects [18, 19]. Eleven subjects (six from Group A and five from group B) who continued to have serum PTH levels above the normal range could possibly be attributed to the fact that some parathyroid glands take longer duration to revert back to normal physiological functioning after prolonged state of secondary hyperparathyroidism due to chronic vitamin D deficiency. This is similar to the phenomenon occuring in hyperthyroidism where pituitary TSH remains suppressed for a very long time despite the patient being in remission with normal serum T3 and T4 levels [20].

Decline in serum ALP levels in both the groups following 6 months of supplementation has also been reported earlier by us and others [14, 15, 19, 21, 22]. We observed no significant change in serum calcium values following supplementaion as also reported by Maalouf et al. while addressing short and long-term safety of weekly high dose of vitamin D3 supplementation [23]. However, others did show statistically significant increase in the serum calcium values [14, 15, 19, 21]. Whether such small changes in serum calcium levels are of any clinical relevance is debatable.

Conclusions

Monthly supplementation with 60,000 IU of both fat-soluble and miscible vitamin D3 is effective and is not associated with any adverse effects. However, miscible vitamin D3 appears to be more efficacious in achieving higher levels of serum 25(OH)D than that observed with a similar dose of fat-soluble vitamin D3 following supplementation. Further large-scale randomized clinical trials in different age groups are required to establish its efficacy over fat-soluble vitamin D.

Weaknesses of our study include nonrandomization of the study subjects, small sample size in Group A and inability to evaluate the baseline urinary calcium-creatinine ratio, ionized calcium, and bone markers due to financial constraints.

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