

Original Research Article

Vitamin D status in apparently healthy students of Maharishi Markandeshwar Medical College & Hospital, Kumarhatti, Solan

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Abstract: Vitamin D was primarily acknowledged for its importance in bone formation, however increasing evidence point to its interference with the proper function of nearly every tissue in our bodies. Thereby its deficiency has been incriminated in a long panel of diseases including cancers, autoimmune diseases, cardiovascular and neurological disorders. This cross-sectional study was carried out to assess vitamin D status and determinants of vitamin D deficiency in 150 apparently healthy MBBS first year students of MMMC&H, Solan. Vitamin D deficiency (52.6%) and insufficiency (42.6%) were highly prevalent in our students, and more common in females. Serum vitamin D was not significantly correlated with serum phosphorus, alkaline phosphatase. But a significant positive correlation was elucidated between serum calcium, sunlight exposure with vitamin D levels. Vitamin D deficiency is an important public health problem and in addition to its effect on the growing skeleton, it may affect other organ systems adversely. Our findings support a recommendation for adequate sunlight exposure, fortification of food and vitamin D supplementation for healthy students. Vitamin D supplementation is an efficient and feasible way to maintain serum vitamin D levels.

Keywords: Vitamin D; vitamin D deficiency; sunlight exposure; fortification; vitamin D supplementation; healthy students

INTRODUCTION:

Vitamin D commonly called sunshine vitamin, plays an important role in optimizing health and preventing disease. It has long been associated with calcium homeostasis and its deficiency is a known precursor to different bone disorders. There is mounting evidence that it has many other biological roles, such as cardiovascular, endocrinal, and immune system homeostasis [1]. It is obtained through either dietary sources or synthesis in the human skin by exposure to ultraviolet B (UVB) radiation. There are two main forms of vitamin D: vitamin D₂ (ergocalciferol) and vitamin D₃ (cholecalciferol). Vitamin D₂ is a plant sterol obtained from the consumption of yeasts and plants and vitamin D₃ is mainly formed in the skin after exposure to ultraviolet B radiation, but it is also present in foods such as fish and cod liver oil. In both children and adults, the primary source of vitamin D is exposure

to the sun while the content of vitamin D in food is low [2].

Vitamin D in its hydroxylated form, 25-hydroxyvitamin D (25(OH) D), is the major storage and circulating form of vitamin D and is considered a reliable marker of vitamin D status [3]. 25-(OH)D is substrate for the dihydroxylated form 1,25-dihydroxyvitamin D (1,25-(OH)D), which is the hormonally active form and exerts its action by binding to vitamin D receptor (VDR) which is a nuclear hormonal receptor that is expressed in most of the tissues [4]. The widespread distribution of VDR signifies important role of vitamin D in humans. It is estimated that one billion people worldwide have vitamin D insufficiency or deficiency, and hypovitaminosis D has been reported in both females and males and in all age groups and even found in the healthy individuals also [5]. It has a bearing not only on

skeletal but also on extra skeletal diseases. Owing to its multifarious implications on health, the epidemic of vitamin D deficiency in India is likely to significantly contribute to the enormous burden on the healthcare system of India. Low dietary vitamin D intake and poor exposure to sunlight are common causes of vitamin D deficiency in the general population. Sunlight is an important source of vitamin D. Several studies have shown a high prevalence of vitamin D deficiency in tropical and subtropical regions of India and other South Asian countries, despite abundant sunlight [6].

Solan, the mushroom city of India, is located at an altitude of 4928 feet above the sea level at latitudes 30.905°N and longitude 77.097°E in the Northern mountainous regions of India. However, the exposure to sunlight here is good and consumption of dairy products is adequate. Though vitamin D deficiency has assumed pandemic proportions all over the world, studies concerning vitamin D deficiency are very rare from this part of the country. Because of the health risks associated with low levels of vitamin D, the primary objective of this study was to determine serum vitamin D levels in otherwise healthy MBBS first year students of MMMC&H, Solan. Such regional data will be of immense use for the healthcare providers to plan health policy for the general population. Very few studies have investigated the determinants of circulating vitamin D in young adults using a set of variables that include lifestyle, dietary habits and anthropometric data. The secondary objective aimed to identify factors that represent determinants of vitamin D deficiency in the study participants.

MATERIAL & METHODS:

Study design and Participants:

This cross-sectional study was conducted among 150 apparently healthy MBBS 1st year students of MMMC&H, Solan. Informed consent was obtained from all the subjects and Institutional ethical committee has approved the study. Exclusion criteria included any malabsorption syndrome, renal disease, and liver disease, and musculoskeletal disorder, intake of any medication known to affect calcium and vitamin D metabolism and any other metabolic disease. Demographic data such as height, weight, BMI and other details like diet, sunlight exposure, and skin colour were collected using a preformed questionnaire.

Biochemical analyses:

Venous blood taken under aseptic conditions was allowed to clot. Serum was separated by centrifugation and analysis was performed on the clear serum. The serum levels of calcium, phosphorus and

alkaline phosphatase activity were measured using routine laboratory methods to get additional information about vitamin D status.

Serum vitamin D was measured as 25-OH D on CLIA Autoplex based on Chemiluminescence method. Vitamin D status was defined as deficient, insufficient and sufficient by serum 25-OH D concentrations <10, 11-29 and > 30ng/ml, respectively.

STATISTICAL ANALYSIS:

Continuous variables were expressed as mean \pm SD whereas discrete variables were expressed as percentage. Pearson's correlation tests were used to find out the significant correlation between the variables. A p value of <0.05 was considered as significant. Significant difference between gender was analyzed using independent t-test and results were considered statistically significant if the p value is <0.05.

RESULTS:

Of the total subjects enrolled (n=150), 63(42%) were males and 87 (55.3%) were females. 64(42.6%) subjects were found to have vitamin D levels 10-29ng/ml which are considered as insufficient to carry out all the physiological functions of vitamin D appropriately. A large proportion, 79 (52.6%) were found to have levels < 10ng/ml and thus, are prone to bone disorders. Only 07 (4.6%) subjects had the optimum levels of > 30ng/ml (Table 1). Deficiency was more in females (55.1%) than in males (47.6%). Insufficiency was more in males (46%) when compared to females (40.2%). Optimum levels of vitamin D were seen in 6.3% and 3.4% of male and female subjects respectively. 95.3 % subjects had less than optimum level of vitamin D.

Descriptive statistics and laboratory biochemical parameters are shown in Table 2. The students studied had a mean age of 19.46 \pm 1.01 years (17-23 years). The mean sunlight exposure was found to be 26.67 \pm 10.53 minutes per day. The mean serum vitamin D, calcium, phosphorus and alkaline phosphatase (ALP) levels were 12.39 \pm 8.55ng/dl, 9.01 \pm 0.34mg/dl, 3.35 \pm 0.49mg/dl and 82.74 \pm 15.89 IU/L respectively. On comparing the genders, mean values of serum vitamin D, calcium, phosphorus and alkaline phosphatase were found to be higher in males. Table 3 compared serum vitamin D levels with study variables. Serum vitamin D was not significantly correlated with serum phosphorus, alkaline phosphatase, and consumption of egg, nonvegetarian food and milk. A significant positive correlation was found between serum vitamin D & serum calcium & sunlight exposure

($r=0.295$, $r=0.221$). Independent t-test was used to find the significant difference between male and female subjects as depicted in Table 4. Significant difference exists between male and female in serum calcium levels ($p=0.021$). Mean sunlight exposure was more in males (29.60 ± 10.48) when compared to females (24.54 ± 10.10) which makes p value significant. Table 5 shows the correlation of vitamin D with study variables

with respect to genders. Among males, serum vitamin D was significantly positively correlated with serum calcium and sunlight exposure. Among females, serum vitamin D was significantly positively correlated with serum calcium. Among both males and females, vitamin D was negatively correlated with serum phosphorus, alkaline phosphatase and nonvegetarian food but p value was not significant for all these four parameters.

Table 1: Basic characteristics

Variables		Total (n=150) N (%)	Male (63) N (%)	Female (87) N (%)
Serum Vitamin D	Deficiency	79 (52.6)	30 (47.6)	49 (56.3)
	Insufficiency	64 (42.6)	29 (46)	35 (40.2)
	Optimum	07 (4.6)	4 (6.3)	03 (3.4)
Diet	Non-veg	54 (36)	31 (49.2)	23 (26.4)
	Veg	78 (52)	23 (36.5)	55 (63.2)
	Veg and egg	18 (12)	9 (14.2)	9 (10.3)
Egg consumption/week	1	21 (14)	11 (17.4)	10 (11.4)
	2	21 (14)	11 (17.4)	10 (11.4)
	3	14 (9.3)	10 (15.8)	4 (4.5)
	4	16 (10.6)	8 (12.6)	8 (9.1)
	No egg	78(52)	23 (36.5)	55 (63.2)
Non veg consumption / week	1	19 (12.6)	12 (19)	7 (8)
	2	10 (6.6)	7 (11.1)	3 (3.4)
	3	8 (5.3)	8 (12.7)	0 (0)
	Once per month	17 (11.3)	4 (6.3)	13 (14.9)
	Vegetarian	78 (52)	23 (36.5)	55 (63.2)
No. of cups of milk consumption/ day	0	26 (17.3)	9 (14.2)	17 (19.5)
	0.5	18 (12)	4 (6.3)	14 (16)
	1	64 (42.6)	30 (47.6)	34 (39)
	2	40 (26.6)	18 (28.5)	22 (25.2)
	3	2 (1.3)	2 (3.1)	0 (0)
Skin colour	Dark	5 (3.3)	2 (3.1)	3 (3.4)
	Fair	34 (22.6)	11(17.4)	23 (26.4)
	Wheatish	111 (74)	48 (76.1)	63 (72.4)
Sunscreen users		46 (30.6)	14 (22.2)	32 (36.7)
BMI Status	Normal	104(69.3)	45 (71.4)	59 (67.8)
	Overweight	21 (14)	10 (15.8)	11 (12.6)
	Underweight	22 (14.6)	6 (9.5)	16 (18.3)
	Obese	3 (2)	2 (3.1)	1 (1.1)

Table 2: Descriptive statistics and laboratory biochemical parameters

	Total	Males	Females
Age	19.46±1.01	19.52±1.02	19.42±1.00
Sunlight exposure in minutes	26.67±10.53	29.60±10.48	24.54±10.10
Serum calcium	9.01±0.34	9.09±0.30	8.96±0.36
Serum phosphorus	3.35±0.49	3.37±0.52	3.33±0.47
Serum alkaline phosphatase	82.74±15.89	85.18±15.74	80.97±15.84
Serum vitamin D	12.39±8.55	13.73±10.57	11.43±6.63

Table 3: Correlation of Serum Vitamin D with study variables

Variable	Pearson correlation	P value
Serum calcium	0.295	0.0002
Serum phosphorus	-0.090	0.273
Serum alkaline phosphatase	-0.0004	0.961
Egg consumption/ week	0.086	0.290
Milk consumption / day	0.036	0.655
Non vegetarian consumption / week	-0.139	0.089
Sunlight exposure in minutes	0.221	0.006

Table 4: Male and female differences

Variable	Independent t test	P value
Serum Vitamin D	1.63	0.105
Serum Calcium	2.326	0.021
Serum Phosphorus	0.451	0.653
Serum Alkaline phosphatase	-1.605	0.110
Sunlight exposure in minutes	2.981	0.003

Table 5: Correlation of Vitamin D with study variables with respect to gender

Variable	Male		Female	
	Pearson correlation	P value	Pearson correlation	P value
Vitamin D				
Serum calcium	0.347	0.005	0.235	0.028
Serum phosphorus	-0.036	0.779	-0.174	0.107
Serum alkaline phosphatase	-0.026	0.839	-0.127	0.241
Sunlight exposure	0.390	0.001	0.078	0.467
Egg consumption / week	0.087	0.496	0.024	0.823
Milk consumption / day	0.025	0.841	0.011	0.913
Non veg consumption/ week	-0.240	0.058	- 0.082	0.450

DISCUSSION:

Vitamin D deficiency continues to be an unrecognized epidemic in many populations around the world. It has been reported in healthy children, young adults, middle-aged adults and the elderly, and is common among both males and females. In this study, we found a high prevalence of vitamin D deficiency (52.6%) among otherwise healthy MBBS first year students of MMMC&H, Solan. In fact, only 4.6% of the students evaluated had sufficient levels of vitamin D, defined as > 30ng/dl. This finding is surprising for young, healthy students living at latitude where endogenous vitamin D synthesis is possible throughout the year.

A similar high prevalence of low vitamin D (96% deficiency and 4% insufficiency) Was reported in medical students in preclerkship years of a Saudi medical school[2]. Also Arya et al[7] reported an approximately 78.3% prevalence of vitamin D deficiency in urban adults of Lucknow, while Kiran *et*

al. [8] found 73.91% prevalence of vitamin D deficiency in healthy subjects of Kancheepuram district of Tamil Nadu. In another study from Jammu (North India), vitamin D deficiency was found to be prevalent in 76.39% of the total population [9]. Recently, Gonzalez-Padilla *et al.*[10] reported a high prevalence of hypovitaminosis D in medical students from Spain and Multani *et al.*[11] found that 87.5% of resident doctors from India had low vitamin D levels. However, in a study that was conducted in a hospital in the Boston area of USA, only 36% of the healthy students , residents and physicians aged 18-29 years were found to be vitamin D deficient [12]. The above reported variation in prevalence can be partially explained by the difference in the populations studied, dietary habits and food fortification, the time of the year when the blood samples were collected and possibly, the different laboratory methodologies used in addition to racial and genetic factors. This study was conducted during summer using the chemiluminescence immunoassay methodology.

It is already well known fact that major source of vitamin D is the sunlight. 30 minutes of exposure of the skin over the arms and face to sunlight, without application of sunscreen, preferably between 10am to 2pm (as maximum ultraviolet B rays are transmitted during this time) daily is adequate to avoid vitamin D deficiency [13]. Many studies have established that vitamin D deficiency is prevalent in Indians despite the sunny climate throughout the year, irrespective of their age, group and gender. We found a positive correlation between sunlight exposure and vitamin D status. Our findings are supported by a study conducted on healthy adults above 20 years age living in Potheri village of Kancheepuram district of Tamil Nadu [8]. In our study, mostly students are leading sedentary indoor lifestyle with limited sunlight exposure (26.67 ± 10.53 minutes per day). The students are free from their classes only in the early mornings and late afternoons when the sun's rays are less efficient for vitamin D synthesis.

The effects of sunlight exposure on vitamin D synthesis are decreased in individuals with darker skin pigmentation and in sunscreen users. Majority of the students (74%) were Wheatish, 22.6% had fair complexion and only 3.3% were dark-skinned. Sunscreen use is another potential confounder influencing cutaneous vitamin D synthesis. It has been shown that when used properly, a sunscreen with a sun protection factor (SPF) of 8 reduces the production of provitamin D₃ by 95%, and 99% by a SPF of 15 [14]. 30.6% of the students were using sunscreen in our study. Serum calcium was positively correlated with serum vitamin D in our study indicating that vitamin D is involved in the absorption of calcium from the gut. Serum phosphorus and alkaline phosphatase levels were within the normal limits but showed no correlation with vitamin D status in our study, was similar to the study conducted in Karachi [15]. Vitamin D deficiency prevents efficient absorption of dietary calcium and phosphorus causing a decrease in serum ionized calcium, resulting in secondary hyperparathyroidism. This engenders loss of phosphorus in the urine and decreases intestinal absorption of phosphorus causing low or low normal phosphorus concentration. Low normal calcium and low normal phosphorus both will cause insufficient calcium phosphorus product leading to a defect in the mineralization of the collagen matrix. The consequence is rickets in children and osteomalacia in adults [16]. During deficiency of vitamin D, there may be low levels of calcium and phosphorus but alkaline phosphatase levels may rise. But in our study, calcium was reduced but phosphorus and alkaline phosphatase levels were within normal limits indicating

that bone mineralization was not yet affected. Hypovitaminosis D can be found despite seemingly sufficient sun exposure, suggesting that a concomitant proper dietary intake is required [17]. Diet such as milk, egg or non-vegetarian food consumption also had no influence on vitamin D levels in this study. Most dietary sources of vitamin D are very low in vitamin D content. Most of the food items rich in vitamin D are of animal origin. But 52% students were found to be vegetarian in our study. Indians do not usually consume oily fish (sardines, tuna, and mackerel) which are rich in D content. Egg yolk is also having only 20IU of vitamin D. Dietary source of vitamin D for vegetarians is unfortified milk which contain only 2IU of vitamin D/100ml. Milk is rarely fortified with vitamin D in India. Dilution and adulteration of milk and milk products further affect the vitamin D content of milk. In India, vitamin D fortified food products are not commonly sold as in case of Western countries, which made our study subjects vulnerable to vitamin D deficiency.

In general, hypovitaminosis D has been reported in the literature as being more prevalent in females [2]. This study proclaimed serum vitamin D levels lower in females than in males. This could be explained by poor dietary habits, decreased exposure to sunlight and widespread use of sunscreens in females compared with male subjects. Differences in the hormonal milieu between genders may also affect transport proteins and enzymes involved in vitamin D metabolism. The results of this study, which showed that 95.3% of young educated students in the medical field have low vitamin D, raise a great concern about the future health consequences of these young students.

CONCLUSIONS:

The present study is one of the few studies carried out to evaluate the status of vitamin D in the medical community. In 95.3% of the young medical students studied, there was low vitamin D levels found despite living in sun-rich environment. An urgent action through the modification of lifestyle and dietary habits, along with improved food fortification and treatment of this deficiency with supplemental doses of vitamin D, must be undertaken in order to prevent any future untoward consequences of hypovitaminosis D in this young healthy population.

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