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ORIGINAL RESEARCH article

Prevalence of vitamin D deficiency in medical students

Abdulghani A. Msalati [1], Abdulla Bashein [1], Ebtesame Aljaloh [1], Ghrew Murad [2, 3]
Khaled Sedaa [1] and **Ahmed Zaid [1 *]**

[1] Department of Biochemistry, Faculty of Medicine, University of Tripoli, Tripoli, Libya,
[2] Faculty of Biology, Medicine and Health, University of Manchester, Manchester, UK,
[3] Department of Medicine, Faculty of Medicine, University of Tripoli, Tripoli, Libya
[*]Author to whom correspondence should be addressed

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Abstract: The prevalence of vitamin D deficiency has recently been recognized in different parts of the world, even affecting healthy populations. The deficiency of vitamin D can lead to rickets in children and osteomalacia in adults. Few studies have been done to evaluate the status of vitamin D in the medical community around the world. No studies have been done in Libya to evaluate the status of vitamin D in medical students. This study aimed to evaluate the prevalence of low levels of vitamin D among healthy Libyan medical students in the first year at the University of Tripoli, Tripoli. A prospective study was conducted on 100 medical students in the first year in the Faculty of Medicine with a mean age of 19.5 years, 38 males and 62 females. Blood samples were taken and investigated for different biochemical parameters including serum calcium, serum vitamin D and serum parathyroid hormone levels. A questionnaire containing different data was completed for each student including personal data (age, address, nationality, and other data related to vitamin D deficiency. The prevalence of vitamin D deficiency in all the participating students was 74.0% (58.7% in males and 83.8% in females), while 21.0% had vitamin D insufficiency (28.0% in males and 16.12% in females). Only 05.0% of the students who had normal vitamin D were males. The mean 25-hydroxy vitamin D level was 19.49±8.56 ng/ml in males and 12.08±6.70 ng/ml in female students. The statistical analysis revealed that there is a highly significant difference among the students in vitamin D deficiency. In conclusion, low concentration of vitamin D is highly prevalent among the medical students included. Urgent action has to be taken to prevent adverse consequences of low vitamin D in the young population.

Introduction

Globally, vitamin D deficiency is one of the most common nutritional deficiencies, affecting nearly half the world's population [1, 2]. Vitamin D is a critical nutrient, fundamentally needed by the human body to function properly; and suboptimal status is detrimental for an array of health outcomes. In Arabic countries, female undergraduate students are usually a particularly vulnerable population to vitamin D deficiency, due to several factors such as cultural reasons, dress code, limited sun exposure, extensive use of sunscreen and limited dietary intake of vitamin D-rich food [3]. Vitamin D deficiency can be manifested as fatigue, general muscle pain and weakness, muscle cramps, joint pain, chronic pain, weight gain, high blood pressure,

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restless sleep, poor concentration, headaches, bladder problems, constipation or diarrhea [4]. Vitamin D deficiency has been shown to play a role in almost every major disease. This includes osteoporosis and osteopenia, varieties of cancer (including breast, prostate and colon), heart disease, high blood pressure, obesity, metabolic syndrome and diabetes, autoimmune diseases, multiple sclerosis, rheumatoid arthritis, osteoarthritis, bursitis, gout, infertility and PMS, Parkinson's disease, depression and personal affective disorder, Alzheimer's disease, chronic fatigue syndrome, fibromyalgia, chronic pain, periodontal disease, psoriasis, dementia, erectile dysfunction and schizophrenia [2, 4-6].

Serum concentration of 25(OH)D is typically used to determine vitamin D status. It reflects vitamin D produced in the skin, as well as that acquired from the diet and has a fairly long circulating half-life of 15 days. It does not, however, reveal the amount of vitamin D stored in other body tissues. The level of serum 1,25(OH)D is not usually used to determine vitamin D status because it has a short half-life of four hours and is tightly regulated by parathyroid hormone, calcium and phosphate, such that it does not decrease significantly until vitamin D deficiency is already well advanced. Although not always required for the diagnosis of vitamin D insufficiency, measurement of serum parathyroid hormone (PTH) level may help establish the diagnosis of vitamin D insufficiency. PTH level is often elevated in patients with vitamin D insufficiency, indicating secondary hyperparathyroidism [7]. Inadequate circulating 25(OH) D is associated with elevated PTH; this condition is called secondary hyperparathyroidism. The secondary hyperparathyroidism maintains serum calcium in the normal range at the expense of mobilizing calcium from the skeleton and increasing phosphorus wasting in the kidney. PTH-mediated increases in osteoclastic activity creates local foci of the bone weakness and cause of generalized decrease in bone mineral density (BMD) resulting in osteopenia and osteoporosis [8, 9]. Vitamin D deficiency is clinically silent. Manifestations are as follows: In young children who have little mineral in their skeleton [8, 9], this defect results in a variety of skeletal deformities classically known as rickets [10]. In adults, the epiphyseal plates are closed, and there is enough mineral in the skeleton to prevent skeleton deformities so that this mineralization defect is known as osteomalacia [11, 12]. The replacement of vitamin D needed for treating vitamin D deficiency depends on the severity of the deficiency. Treatment involves an initial high-dosage treatment phase until the required serum levels are reached, followed by the maintenance of the acquired levels. The lower the 25(OH)D serum concentration is before treatment, the higher the dosage that is needed to quickly reach an acceptable serum level. The present study aimed to examine the prevalence of vitamin D deficiency among young a medical college student and to look at the probable cause of deficiency from medical and cultural point of view.

Materials and methods

Study design and subject recruitment: A prospective study was conducted on 100 male and female students in the first year in the Faculty of Medicine, University of Tripoli. Blood samples were taken and investigated for different biochemical parameters such as serum calcium, serum vitamin D and serum parathyroid hormone levels. A questionnaire containing different data was completed for each subject including personal data (age, address and nationality). Other data such as vitamin D consumption and health questions, food consumption and sun exposure were collected. Data were personally collected from each subject. The informed consent form was signed by every participant. The study was ethically approved by the Scientific Committee of the Libyan Academy, Tripoli, Libya (20/2020).

Samples selection: A sample size of one hundred samples was collected randomly from first-year medical students in the University of Tripoli, according to inclusion and exclusion criteria.

Inclusion criteria: Age group of 18-21 years from both sexes of medical students.

Exclusion criteria: Age <18 and >21 years old, diabetes mellitus, renal disorders and liver pathology.

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Collection of blood sample: Three ml of venous blood sample was collected in white tubes for the estimation of biochemical parameters. The blood drawn was allowed to coagulate and the serum was separated by centrifugation and stored at -20°C until assayed. Each tube was labeled with a sticker containing the patient data number and time of collection.

Measurement of vitamin D: 25[OH] D was measured by direct ELISA kit method using vitamin D ELISA Kit (ORGENTEC Diagnostika GmbH Company, Germany). The reference value of the used kit: vitamin D deficiency: <20 ng/ml, vitamin D insufficiency: 20.0 ng/ml-30.0 ng/ml, vitamin D sufficiency: >30 ng/ml-100 ng/ml, and vitamin D toxic: >100.0 ng/ml.

Measurement of calcium: The colorimetric test using O-cresolphtaleine was used for calcium measurement the kit was obtained from a calcium assay kit (Biomagreb Company - TUNISIA). The reference value of the used kit: adult's serum calcium: 9.0 mg per dl-10.6 mg per dl or 2.25 mmol/l-2.65 mmol/l.

Measurement of parathyroid hormone: Electro-chemiluminescence assay was used parathyroid hormone assay kit (Roche Diagnostics-USA). The reference value of the used kit is 8.0 pg/ml-79.6 pg/ml.

Statistical analysis: Data was entered in the SPSS V. 20 program. Percentage, frequency, mean, standard deviation and t-test were used. A correlation between different parameters was calculated by correlation coefficient.

Results

Characteristics of participating students: **Table 1** shows the characteristics of the study sample including 62.0% (n=62) female students and 38.0% (n=38) male students. The age of the students involved in this study ranged from 18 years to 21 years old with a mean value of 19.7 years and a standard deviation of 1.03. Thus, 14.0% (n=14) of the sample lived in flats while 28.0% (n=28) lived in villas and most of the participants 57.0% (n=57) lived in houses (**Table 1**).

Table 1: Participants' characteristics of the participants

Gender	n, percentage
Male	38 (38.0%)
Female	62 (62.0%)
Age (years)	
Range	18-21
Mean	19.73
SD	1.03
Residence	
Flats	14 (14.0%)
Villas	28 (28.0%)
Houses	57 (57.0%)

Vitamin D consumption and health questions: **Table 2** summarizes the findings of the answers related to vitamin D and food consumption. Nine percent (n=09) of the participating students were on vitamin D supplements versus 91.0% not on any supplements. Six out of nine taking supplements were females. 48.0% (n=48) of the sample taught that it is not important to take any form of supplements (23 females versus 25 males). Most of the participants 88.0% (n=88) did not take any vitamin D supplement before this study while the remaining 12.0% took vitamin D supplements before this study. Most of these were females (10 females versus 02 males). 56 of the participants (56.0%) knew about the importance of vitamin D and how it helps the body absorption of calcium these 40 females versus 16 males while 44.0% did not know (22 females versus 22 males). 93.0% of the participants were not tested previously for vitamin D deficiency and

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07.0% (six females and one male) were tested for vitamin D before. One out of them was found to have normal vitamin D levels. 59.0% of the participating students knew the foods containing vitamin D (41 females versus 18 males). 87.0% of the participants like to consume dairy products. 59.0% of the participating drink less than four cups of milk weekly, 33.0% reported that they drink five to 10 cups weekly and 8.0% drink more than 10 cups weekly. 82.0% of the participants eat less than five cups of yogurt per week, 15.0% eat six to 10 cups weekly and 03.0% eat more than 10 cups per week. A high percentage (90.0%) of the participants eat from one to 10 slices of cheese per week and 10.0% eat more than 10 slices weekly. 77.0% of the participants like to eat fish but 30.0% eat fish once weekly (20 females versus 10 males) and 11.0% eat fish twice weekly (07 females versus 04 males) and the majority do not eat fish at all (59.0%). 66.0% of participants drink orange juice that is fortified with vitamin D and 34.0% do not. Of those who drink orange juice that is fortified with vitamin D, 65.2% drink less than four cups per week and 34.8% drink more than four cups per week.

Table 2: Parameters of vitamin D and food consumption

Vitamin D and food consumption			
Supplements	On supplement	Not on supplement	
	09 (09.0%)	91 (91.0%)	
Taking Vitamin D before the current study	Took vitamin D	Did not take vitamin D	
	12 (12.0%)	88 (88.0%)	
Knowledge about vitamin D importance	Know	Do not know	
	56 (56.0%)	44 (44.0%)	
Previous testing for vitamin D	Tested	Not tested	
	07 (07.0%)	93 (93.0%)	
Knowledge about foods containing vitamin D	Know	Do not know	
	59 (59.0%)	41 (41.0%)	
Dairy products consumption	Yes	No	
	87 (87.0%)	13 (13.0%)	
Milk cups taken per week	<4	5 - 10	>10
	59 (59.0%)	33 (33.0%)	8 (08.0%)
Yogurt cups taken per week	<5	6-10	>10
	82 (82.0%)	15 (15.0%)	03 (03.0%)
Cheese slices taken per week	1-10	>10	
	90 (90.0%)	10 (10.0%)	
Liking eating fish	Yes	No	
	77 (77.0%)	23 (23.0%)	
Eating fish (times/week)	00	01	02
	59 (59.0%)	30 (30.0%)	11 (11.0%)
Drinking orange juice fortified with vitamin D	Yes	No	
	66 (66.0%)	33 (33.0%)	
Orange juice cups fortified with vitamin D taken per week	00	<04	≥04
	34 (34.0%)	43 (43.0%)	23 (23.0%)

Vitamin D insufficiency was seen in a total of 21 cases (21.0%) of the study group. Of these, 11 (52.4%) were males and 10 (47.6%) were female students with mean±SD of vitamin D in males being 19.49±8.56 ng/ml and in females 12.08±6.70 ng/ml. The statistical analysis revealed that there is a very highly significant difference ($p<0.001$).

Health questions: The answers to the questions related to health are summarized in **Table 3**, in terms of experiencing any malady such as back pain, body aches and bone fractures unrelated to a specific injury,

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55.0%, 62.0% and 99.0%, did not experience any back pain, body aches, bone fracture, respectively, as well as 45.0%, 38.0% and 01.0% reported suffering. **Table 3** showed that 44 of those suffering from back pain have vitamin D deficiency and only one has normal vitamin D levels. The back pain was more frequent in female students 36 (58.1%) versus nine males (23.6%). The body ache was more common in female 30 females (48.3%) versus seven males (18.3%). The statistical analysis revealed that there is no significant difference with the individuals not suffering from back pain. The only participant who had bone fractures was a female. In addition, 13.0% of the participants experienced abdominal pain, gas, bloating or diarrhea when taking dairy products (12 females versus one male).

Table 3: Relationship of vitamin D deficiency with back pain, body aches and bone fracture

			Vitamin D deficiency			
Symptoms			Yes	No	X^2	P value
Back pain	Yes	45 (45.0%)	44	1	0.67	0.4
	No	55 (55.0%)	52	3		
Body ache	Yes	38 (38.0%)	37	1	0.30	0.41
	No	62 (62.0%)	59	3		
Bone fracture	Yes	01 (01.0%)	01	0	Fisher's exact test	
	No	99 (99.0%)	95	4		

Cultural factors and vitamin D deficiency: When examining cultural factors such as religious beliefs, views of beauty and concern for health, 53.0% (39 females versus 14 males) of the participants indicated that they do not show their skin in public and view it as being immodest as a result of their culture. Specific factors such as views of beauty, health and religion, however, revealed variations in responses. When responding to the statement "I think most people think light colored skin is more attractive than darker-skin" 39.0% (27 females versus 12 males) agreed. Accordingly, the response of the participants to the next question was a feeling of unhappy if the sun made their skin darker. This resulted in an agreement of 49.0% (32 females versus 17 males). The response to the question "Being careful when going out in the sun so their skin complexion won't look bad had "an agreement response of 45.0% (37 females versus 08 males) (**Table 4**). The responses of participants to the question of whether sun exposure makes the skin wrinkle and makes them unhappiness 32.0% responded sometimes and 25.0% as never avoiding sun exposure. Health concerns with the statement that these individuals cover their skin and use sunscreen because of the risk of cancer resulted in a 10.0% response as always and 54.0% answered as never use sunscreen. Covering skin as a result of religious beliefs however was reported by 56.0% answered as always, while 22.0% answered as never (**Table 5**).

Table 4: Reported reasons for covering skin, cultural beliefs and religion

Reasons for covering skin			
Variables	Disagree	No opinion	Agree
Showing skin is not modest Because of culture/traditions	29.0%	18.0%	53.0%
I think most people think light colored skin is more attractive than darker skin	39.0%	22.0%	39.0%
I would be unhappy if the sun made my skin darker	36.0%	15.0%	49.0%
I'm careful going into the sun So I won't look Bad	40.0%	15.0%	45.0%

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Table 5: Variables for reasons for covering skin

Reasons for covering skin					
Variables	Never	Rarely	Sometimes	Frequently	Always
Covering skin is due to religious beliefs	22%	05%	13%	04%	56%
Avoid sun because skin will wrinkle	25%	22%	32%	04%	17%
I cover my skin and use sunscreen because of the risk of cancer.	54%	10%	25%	01%	10%
When I am outside of the house during the day my legs are covered	07%	02%	23%	07%	61%
When I am outside of the house during the day my hair/head are covered (hijab)	36%	03%	06%	01%	54%
When I am outside of the house during the day my face are covered (Nikab)	94%	00%	03%	00%	03%

Sun exposure: In **Table 6**, data revealed that 61.0% of the student participants always cover their legs when they are outdoor (52 females versus9 males), 54.0% always cover their hair (53 females versus only one male) only two females always cover their faces and another three females sometimes cover their face by Alnekab, the remaining 95.0% never cover their face.

Table 6: Relations between sun exposure and vitamin D level

		Vitamin D level	
Variables	Normal	Insufficient	Deficient
Always covering leg 61%	00	10	51
Always covering hair 55%	00	07	48
Always covering faces 02%	00	00	02

Vitamin D concentration: The mean vitamin D (25(OH) D) level of the study participants is 14.90±8.24 ng/ml. Sufficient vitamin D (25(OH)D) status (≥30 ng/ml) is seen only in five males (05.0%) and not seen in any case of female students. Most of the participants (74.0%) were vitamin D deficient (≤20 ng/ml). Of these 52 are females (70.3%) and 22 are males (29.7%). Vitamin D insufficiency (≥20 ng/ml and ≤ 30 ng/ml) is seen in a total of 21 cases (21.0%) of the study group. Of these, 11 (52.4%) are males and 10 (47.6%) are females (**Table 7**). The mean±SD of vitamin D in males is 19.49±08.56 ng/ml and in females is 12.08±6.70 ng/ml. The statistical analysis revealed that there is a very highly significant difference ($p<0.001$).

Table 7: Vitamin D sufficient, insufficient and deficient in Libyan students

Vitamin D concentration	Male (n=38) (n, %), mean±SD	Female (n=62) (n, %), mean±SD
> 30-100 ng per ml (sufficient)	(05) 05.0% 34.76±4.18	(00) 00.0%
20-29 ng per ml (insufficient)	(11) 52.4% 24.42±1.83	(10) 47.6% 24.81±2.84
< 20 ng per ml (deficient)	(22) 29.7% 13.56±4.36	(52) 70.3% 09.91±4.27

Calcium concentration: Average calcium level was 8.52±0.51 mg per dl, hypocalcaemia (calcium level below 8.6) was noticed in 55 of the sample (55.0%), 40 females and 15 males and 45.0% of the participants had normal calcium levels. Mean±SD of calcium in males is 8.72±0.60 mg per dl and in females is 8.38±0.39 mg per dl. The statistical analysis showed that there is a very highly significant difference ($p<0.001$). The correlation between calcium and vitamin D is significant at $p<0.01$ level (**Table 8**).

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Table 8: Correlation between vitamin D and calcium concentration

		Calcium	Vitamin D
Calcium	Pearson Correlation	1	0.340**
	Sig. (2-tailed)		0.001
	n	100	100
Vitamin D	Pearson Correlation	0.340**	001
	Sig. (2-tailed)	0.001	
	n	100	100

The normal serum PTH ranged between 8.0 to 79.6 pg per ml. 31.0% of the participants were below normal levels (21 females versus 10 males). 63.0% were at normal level (36 females versus 27 males) and 06.0% had a high PTH level (five females versus one male). Mean±SD of PTH in male is 23.32±20.59 pg per ml and in female is 34.45±35.13 pg per ml. A statistical analysis showed that there is significant difference ($p<0.05$). The correlation between calcium and PTH is not a significant as shown in **Table 9**. Furthermore, the correlation between vitamin D and parathyroid hormone is not significant as shown in **Table 10**.

Table 9: Correlation between parathyroid hormone and calcium

		Calcium	**PTH**
Calcium	Pearson Correlation	1	-0.066
	Sig. (2-tailed)		0.512
	n	100	100
PTH	Pearson Correlation	-0.066	1
	Sig. (2-tailed)	0.512	
	n	100	100

Table 10: Correlation between parathyroid hormone and vitamin D

		PTH	**Vitamin D**
PTH	Pearson Correlation	1.00	-0.186
	Sig. (2-tailed)		0.063
	n	100	100
Vitamin D	Pearson Correlation	-0.186	1.000
	Sig. (2-tailed)	0.063	
	n	100	100

Discussion

Vitamin D is a crucial nutrient for bone health. Because maximum bone density is not achieved until about the age of 30 years, young adults are at risk of poor bone health related to vitamin D status [2]. Evidence has linked vitamin D deficiency to other health conditions such as cancer, diabetes and cardiovascular disease [4-6]. This study appears to be the first of its kind conducted among Libyan medical students to investigate the prevalence of vitamin D deficiency in this population. The purpose of the current project was to gain a baseline assessment of the real situation of vitamin D among university students to make information available for developers of health promotion programs aimed at decreasing the hazards associated with

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vitamin D deficiency. The characteristics of the study sample included 62.0% females and 38.0% males. The age of students involved in this study ranged from 18 to 21 years of age with a mean of 19.73. Most of the participants were vitamin D deficient. Of these about 75.0% were females. What might be the reason for this highly significant decrease in vitamin D levels among young colleague students at the University of Tripoli? The present study indicates males and females share the same diet and a small group among them was taking vitamin supplements. The answer is probably in the extent of skin exposure to sunlight, because most of the study participant in the same extent cover their bodies. The present findings support the previous data from numerous studies that revealed a high prevalence of vitamin D deficiency and insufficiency among Middle Eastern populations. One of the studies supports the present finding which was conducted by AlQuaiz et al. [13] who studied the state of vitamin D in pre-clerkship years at King Faisal University in Dammam (Saudi Arabia). The study involved 95 male and 103 female students. The mean age of all students was 19.54 years. In 100% of the students, the vitamin D level was low. The prevalence of vitamin D deficiency in all students was 96.0% with 92.6% in males and 99.0% in females, while the remaining 04.0% had vitamin D insufficiency. The mean 25-hydroxy vitamin D concentration was 26.83±12.60 nmol/l in males and 16.03±8.28 nmol/l in females. Another study supported current findings carried out at Saudi Arabia by Hasanato [14] studied one hundred and seventy-eight female medical students including 70.8% were vitamin D deficient (<20 ng/ml), 16.3% had insufficient vitamin D (20-30 ng/ml) and 12.9% had normal vitamin D level (>30 ng/ml). Comparing our results with the Iranian study conducted on 254 students (128 males and 126 females) at the University in Shiraz reported that half of the female students were vitamin D insufficient and half of them had vitamin D deficiency [15]. The prevalence of vitamin D insufficiency and marginal status among male students was equally half. Serum vitamin D of female students was significantly less than the males, this finding also supports the current study. On the side of the Caribbean Sea, a study carried out on 103 medical students from the University of Las Palmas de Gran Canaria Spain [16], the results showed that vitamin D deficiency is observed in 30.0% of the students and vitamin D insufficiency in 30.0% which is in accord with present findings. Another study was done on medical students in Pakistan [17], this included 100 medical students and employees. It is observed that 70.0% of the subjects showed vitamin D deficiency. Further 15.0% were reported with insufficient levels of vitamin D and 15.0% were found with a normal range of vitamin D. However, in a study conducted in a hospital in Boston in the USA, 35.0% of the healthy students, residents and physicians aged 18-29 years were found to be vitamin D deficient. The markedly higher prevalence of low vitamin D in medical students in the University of Tripoli compared to the low prevalence of vitamin D deficiency in the individuals from Boston. Despite the abundance of sunlight found in the areas may be related to the difference in dietary habits and food fortification in addition to racial, cultural and genetic factors. A Jordanian study found that the prevalence of hypovitaminosis D was 60.0% in the study group as a whole. Dress styles covering the whole body, totally or nearly totally, have adverse effects on 25(OH)D level and may produce a state of secondary hyperparathyroidism in the long run. Although Jordan enjoys plenty of sunshine, these data are suggestive of widespread hypovitaminosis D in Jordan [18]. Compared to the present findings of systemic review and meta-analysis [19] which identified 1692 studies, of which 129 studies with 21 474 participants from 23 African countries were included in the systematic review and 119 studies were included. The pooled prevalence of low vitamin D status is 18.5% (CI, 10.66-27.78) with a cutoff of serum 25(OH)D concentration less than 30 nmol per l; 34.2% (CI, 26.22-43.68) for a cutoff of less than 50 nmol per l; and 59.5% (CI, 51.32-67.50) for a cutoff of less than 75 nmol per l. The overall mean 25(OH) D concentration was 67.78 nmol per l (CI, 64.50-71.06), mean serum 25(OH) vitamin D concentration was lower in populations living in Northern African countries or South Africa compared with sub-Saharan Africa, as well as well in urban areas compared with rural areas. In women compared with men and in newborn babies compared with their mothers which are in agreement with our results.

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Conclusion: The findings indicate that 75.0% of the young educated Libyan medical students have vitamin D deficiency, which raises a big concern about the skeletal health of young students in the future. A health action should be taken to avoid vitamin D deficiency and its complications.

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Conflict of interest: The authors declare the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.
Ethical issues: Including plagiarism, informed consent, data fabrication or falsification and double publication or submission were completely observed by the authors.
Data availability statement: The data that support the findings of this study are available from the corresponding author upon reasonable request.
Authors' declarations: The authors confirm that all relevant ethical guidelines have been followed and any necessary IRB and/or ethics committee approvals have been obtained.

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