Serum vitamin D level may be associated with body weight and body composition in male adolescents – a longitudinal study
Stężenie witaminy D w surowicy może mieć związek z masą i składem ciała u nastolatków płci męskiej – długotrwałe badanie

1,Saeid Doaei, 2,Seyed Alireza Mosavi Jarrahi, 3,Saheb Abbas Torki, 4,Rouhollah Haghsenas,
5,Zahra Jamshidi, 6,7,Shahla Rezaei, 8,Alireza Moslem, 9,Fereshteh Ghorat, 10,11,Adeleh Khodabakhshi,
12,Maryam Gholamalizadeh

1,Cancer Research Center, Shahid Beheshti University of Medical Sciences, Tehran, Iran
2,Department of Health Education, Research Center of Health and Environment, School of Health, Guilan
University of Medical Sciences, Rasht, Iran
3,Center for Cancer Epidemiology, West Asia Organization for Cancer Prevention, Sabzevar University
of Medical Sciences, Iran
4,Department of Nutrition, Faculty of Nutrition Sciences, Shiraz University of Medical Sciences, Shiraz, Iran
5,Department of Physical Education, Semnan University, Semnan, Iran
6,Student Research Committee, Shiraz University of Medical Sciences, Shiraz, Iran
7,Department of Clinical Nutrition, School of Health & Nutrition, Shiraz University of Medical Sciences,
Shiraz, Iran
8,Iranian Research Center on Healthy Aging, Sabzevar University of Medical Sciences, Sabzevar, Iran
9,Non-Communicable Diseases Research Center, Sabzevar University of Medical Sciences, Sabzevar, Iran
10,Department of Nutrition, School of Public Health, Kerman University of Medical Sciences, Kerman, Iran
11,Physiology Research Center, Kerman University of Medical Sciences, Kerman, Iran
12,Student Research Committee, Cancer Research Center, Shahid Beheshti University of Medical Sciences,
Tehran, Iran

Abstract
Introduction: The prevalence of both obesity and vitamin D deficiency has been dramatically increased worldwide.
Aim of the study: This study aimed to investigate the association between vitamin D serum level and anthropometric indices of
overweight and obese male adolescents at baseline and after 18 weeks of a weight reduction intervention.
Methods: This study was carried out on 90 male students aged 12 to 16 years who were randomly selected from two schools in
Tehran, Iran. The participants were assigned to two groups with high and low vitamin D level based on their serum vitamin D levels
at baseline. Five ml blood samples were collected at the baseline and after the 18 weeks of a weight reduction intervention. Height,
weight, body mass index (BMI), body fat percent and body muscle percent were measured using a bio impedance analysis (BIA) scale.
Results: Vitamin D level in non-obese adolescents was significantly higher than the obese participants (44.01 vs 37.67 ng/dl,
$p < 0.04$). However, there was no significant correlation between changes of vitamin D level and anthropometric measurements
after 18 weeks. Adjusting the effect of age did not alter the association. Further adjustments for physical activity, dietary intake of
vitamin D, and fat and muscle percentage had no effect on the results.
Conclusion: The serum level of vitamin D was negatively associated with obesity, but not with short-term changes of anthropometric
measurements in male adolescents.
Key words: vitamin D, obesity, adolescents, body composition, BMI.
Introduction

Obesity is one of the most serious health problems in both developing and developed countries [1]. Obesity and overweight has dramatically increased among children and adolescents over the past 3 decades [2]. The prevalence of obesity in Iranian adolescents is 11.9% [3]. The worldwide rise in the obesity rate of the school-aged children [4] is a risk factor for childhood obesity and is also associated with certain diseases including type 2 diabetes mellitus, heart diseases, hypertension, dyslipidemia, and other chronic conditions [4–6].

Vitamin D deficiency is an important public health problem and may be associated with obesity [5]. Some studies have reported that obesity might be related to deficiency of some micronutrients such as vitamin D [7–10]. Some studies found a link between vitamin D deficiency and obesity [7–10]. Vitamin D deficiency is apparently linked with unhealthy diet and the lack of sun exposure [8–10]. The sedentary lifestyle and less outdoor activity have important roles in both obesity risk and vitamin D deficiency [11]. Extra body adipose tissue may reduce the bioavailability of vitamin D as a result of trapping vitamin D in adipocytes [12, 13].

A reverse association was found between body mass index (BMI) and vitamin D level [14, 15] in some studies, while the others found no association between BMI and vitamin D [16, 17].

One study found that each unit increase on BMI lead to decrease vitamin D level as 1.15% [18]. It is possible that the interaction between vitamin D and anthropometric indices can be mutual and serum vitamin D may also have some effects on body fat percentage. Controversial results were reported on the effect of vitamin D supplementation on anthropometric indices [19, 20]. In some studies, vitamin D supplementation reduced body fat mass, but had no effect on weight and waist circumferences [21, 22]. A possible underlying mechanism is that vitamin D deficiency can cause secondary hyperparathyroidism leading to rising intracellular free calcium in adipocytes which can blunt lipolytic response to catecholamines and sharpen lipogenesis that contribute to fat accumulation in body [23]. Improvement of vitamin D status to the normal levels also can reverse the process of fat accumulation and may lead to fat reduction [24]. So, this study aimed to investigate the association between vitamin D and anthropometric indices in overweight and obese male adolescents after 18 weeks of a comprehensive lifestyle intervention.

Material and methods

Study population

This study was carried out as an ancillary study within a randomized, controlled, school-based trial on 100 overweight and obese 12 to 16 years male adolescents who were randomly selected from January to May 2016 from two male high schools of Tehran, Iran. All participants were resident in the same metropolitan area (urban area) (the latitude: 35.715298, and the longitude: 51.404343). Thus, they were expected to be similar regarding the level of sun exposure. Five ml blood samples were collected at the baseline and after 18 weeks of the study. The participants were divided into two groups of high and low vitamin D level based on their serum vitamin D levels at baseline.

The inclusion criteria were as follows: adolescents with overweight according to BMI chart of World Health Organization (WHO), 12 to 16 years old, not being suffered from weight-related disease, not using weight affecting drugs, anti-inflammatory drugs, and calcium/vitamin D supplements, and having the willingness to participate in the study. Ten students were excluded due to fear of blood sampling and the final analysis was performed on the remaining 90 subjects. Subjects were assigned to two groups: high serum vitamin D level (≥ 40 ng/dl, n = 41) and low serum vitamin D level (< 40 ng/dl, n = 49), based on data of a previous study suggested that maintenance of 25(OH)D level between 40 and 60 ng/ml is ideal, safe, and optimal for multiple health outcomes [25].

Anthropometric measurements

The height of students was measured with a wall-mounted stadiometer to the nearest 0.1 cm (Seca 711; Seca, Hamburg, Germany). A bio impedance analysis scale (BIA) (Omron BF511, Kyoto, Japan) was used to measure weight (nearest 0.1 kg), BMI, body fat percentage (BF), and skeletal muscle percentage (SM) after entering the subjects’ age, gender, and height. This device is a digital, mobile, and non-invasive device that has eight electrodes that sends an extremely weak electrical current of 50 kHz and less than 500 μA through the body to determine the amount of muscle tissue. The validity of this device has been confirmed in previous studies [26]. All data were classified according to the z-score guidelines defined by WHO recommendations (for weight and BMI).

Serum vitamin D measurements

Blood samples (5 ml) were collected of all participants in the study at baseline and after 18 weeks. A direct competitive enzyme-linked immunoassortent assay (ELISA) method and vitamin D VIDAS Kit (Marory-l’Étoile, bioMérieux, France) were used for measuring 25-hydroxy vitamin D level. The VIDAS 25-OH vitamin D total assay is considered suitable measuring D2 and D3 serum and serum level with high accuracy. Correlation between the results from VIDAS Kit with the reference methods of chromatography and volume spectrometry was r = 0.93 which is indicative of the efficiency of this method.

Dietary intake of vitamin D

Intakes of vitamin D were assessed by a validated 168-item semi-quantitative FFQ [27]. The FFQ consisted of 168 food items with standard portion sizes commonly consumed by Iranian people. Face-to-face interviews were administered by a trained dietitian. Dietary vitamin D intake (μg/day) was calculated by using the analyzed in Modified Nutritionist-4 software program which was modified for Iranian foods.

Physical activity assessment

For adjusting the effect of confounding factors, data on physical activity level were collected using physical activity tracker smart bands (MI 2, Xiaomi, China) which was validated in a previous study [28].
Intervention

The intervention was implemented for 12 weeks in two levels. In the first level, the environmental and lifestyle changes were applied to the school level and all of the students in the intervention school were covered. The five dimensions of Ottawa Charter [29] were used for systemic implementation of the interventions in school level. Our intervention objects were:

- modify the health policy at the school level to influence weight and BMI,
- creating supportive environment to weight reduction,
- strengthen community action to achieve a healthy weight,
- developing personal skills to adopt a healthy lifestyle, and
- reorienting health services to prevent and treatment of obesity.

We had some strategies for every object and many operational activities for every strategy. In the second level, the personalized diet and physical activity intervention were implemented for each participant. In addition, parents were provided an educational session regarding healthy meals and creating a supportive environment at home for healthy diet and physical activity. The personalized diet was adopted with free snacks offered in school days by researchers. Furthermore, a high-intensity interval training was carried out for improving the physical activity at the schools. In this method, students warmed up for 10 min under supervision of an exercise physiologist and they were involved in high-intensity exercise for a minimum of 30 min. The details of intervention program have been explained elsewhere [30].

Statistical analysis

To ensure normality of data distribution, Kolmogorov-Smirnov test was applied. Binominal logistic regression was used to evaluate the association between vitamin D and obesity and the correlation of vitamin D with weight changes. Confounders such as age, food intake, and physical activity were adjusted in different regression models. Also, we applied generalized linear model (GLM) repeated measurements to evaluate the correlation between changes of serum vitamin D and anthropometric indices. SPSS version 21 was used for data analysis and p-value < 0.05 was considered as the significant level.

Ethics statement

Written consent forms were obtained before the study. The study was approved by the Ethics Committee of the National Nutrition and Food Technology Research Institute, Tehran, Iran (reference number: Ir.sbmnu.nntftri.rec.1394.22).

Results

Eight percent of the participants had vitamin D deficiency (< 20 ng/ml), 13% had insufficient (20–30 ng/ml), and 79% of them had normal levels of this vitamin (> 30 ng/ml). The level of vitamin D in 46% of them was in the optimal area for health related outcomes (> 40 ng/ml, n = 41). Anthropometrics measurements and serum vitamin D levels of the two groups were summarized in Table I. The mean of serum vitamin D levels in

Table I. Characteristics of the study participants

<table>
<thead>
<tr>
<th></th>
<th>Overweight (48) Mean (SD)</th>
<th>Obese (42) Mean (SD)</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (year)</td>
<td>13.92 (0.91)</td>
<td>13.95 (0.92)</td>
<td>0.86</td>
</tr>
<tr>
<td>25(OH) D (ng/ml)</td>
<td>44.012 (14.69)</td>
<td>37.6667 (14.02)</td>
<td>0.04</td>
</tr>
<tr>
<td>Height (cm)</td>
<td>167 (8.25)</td>
<td>168 (8.07)</td>
<td>0.62</td>
</tr>
<tr>
<td>Weight (kg)</td>
<td>65.1582 (12.79)</td>
<td>81.9690 (13.31)</td>
<td>0.001</td>
</tr>
<tr>
<td>BMI (kg/m²)</td>
<td>23.2521 (1.62)</td>
<td>29.0381 (2.94)</td>
<td>0.001</td>
</tr>
<tr>
<td>BMI z-score</td>
<td>1.04 (0.44)</td>
<td>2.02 (0.38)</td>
<td>0.001</td>
</tr>
<tr>
<td>BF%</td>
<td>22.9876 (5.66)</td>
<td>30.8738 (5.02)</td>
<td>0.001</td>
</tr>
<tr>
<td>SM%</td>
<td>36.7874 (2.38)</td>
<td>33.8262 (2.09)</td>
<td>0.001</td>
</tr>
<tr>
<td>Vitamin D intake (μg/day)</td>
<td>7.4795 (1.34)</td>
<td>5.7150 (1.14)</td>
<td>0.245</td>
</tr>
<tr>
<td>Physical activity (min/day)</td>
<td>45 (15.36)</td>
<td>37 (12.67)</td>
<td>0.133</td>
</tr>
</tbody>
</table>

SM% – skeletal muscle percentage; BF% – body fat percentage; BMI – body mass index
Subjects with high level vitamin D had lower weight ($p < 0.01$) than low level of vitamin D. No significant differences were found in age, BMI, BF, SM, physical activity and dietary intake between two groups (Table II).

In regards of investigation of the correlation between changes of vitamin D and anthropometric indices after 18 weeks in both groups and after adjustments for age, vitamin D and physical activity, no significant association was found (F within: 0.01 ($p = 0.9$), F between: 3.3 ($p = 0.07$) (Table III). Using logistic regression for evaluating the effect of serum D level (before and after the study) on weight changes, we found that serum vitamin level had no significant correlation with changes of body weight. The result remained meaningful after more adjustment on age (model 1), physical activity (model 2), dietary vitamin D intake (model 3), BF, and SM (model 4) (Table IV).

### Discussion

The results identified that serum vitamin D level had an inverse association with BMI in male adolescents and obese students had lower vitamin D level compared with non-obese students. However, no significant association was found between changes of vitamin D levels with changes of anthropometric indices after 18 weeks.

In line with our study, Rodrigues et al. [31] have found the reverse association between vitamin D and weight in school-aged

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**Table II.** Comparison of the participants with high and low levels of vitamin D

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Low vitamin D level ($n = 49$) at baseline</th>
<th>High vitamin D level ($n = 41$) at baseline</th>
<th>$P$-value</th>
<th>Low vitamin D level ($n = 49$) after intervention</th>
<th>High vitamin D level ($n = 41$) after intervention</th>
<th>$P$-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (year)</td>
<td>13</td>
<td>13</td>
<td>0.69</td>
<td>14 (0.93)</td>
<td>14 (0.75)</td>
<td>0.73</td>
</tr>
<tr>
<td>25(OH)D (ng/ml)</td>
<td>27.9878</td>
<td>51.9816</td>
<td>0.000</td>
<td>19.67 (7.4)</td>
<td>41.31 (16.2)</td>
<td>0.001</td>
</tr>
<tr>
<td>Height (cm)</td>
<td>169</td>
<td>165</td>
<td>0.02</td>
<td>171 (8.25)</td>
<td>165 (16.44)</td>
<td>0.92</td>
</tr>
<tr>
<td>Weight (kg)</td>
<td>76.8683</td>
<td>69.7693</td>
<td>0.01</td>
<td>78.4 (13)</td>
<td>71.8 (14.7)</td>
<td>0.62</td>
</tr>
<tr>
<td>BMI (kg/m²)</td>
<td>26.4976</td>
<td>25.4959</td>
<td>0.22</td>
<td>26.7 (3.8)</td>
<td>25.6 (3.8)</td>
<td>0.37</td>
</tr>
<tr>
<td>BMI z-score</td>
<td>1.64</td>
<td>1.42</td>
<td>0.14</td>
<td>1.61 (0.69)</td>
<td>1.51 (0.59)</td>
<td>0.52</td>
</tr>
<tr>
<td>BF%</td>
<td>26.9951</td>
<td>26.3940</td>
<td>0.68</td>
<td>27.6 (6.96)</td>
<td>25.15 (7.69)</td>
<td>0.35</td>
</tr>
<tr>
<td>SM%</td>
<td>35.3220</td>
<td>35.4754</td>
<td>0.79</td>
<td>35.6 (2.9)</td>
<td>36 (2.75)</td>
<td>0.85</td>
</tr>
<tr>
<td>Vitamin D intake (µg/day)</td>
<td>6.19 (0.53)</td>
<td>7.04 (1.6)</td>
<td>0.58</td>
<td>6.58 (0.69)</td>
<td>7.83 (1.9)</td>
<td>0.78</td>
</tr>
<tr>
<td>Physical activity (min/day)</td>
<td>44 (17)</td>
<td>39 (14)</td>
<td>0.42</td>
<td>41 (12)</td>
<td>37 (11)</td>
<td>0.69</td>
</tr>
</tbody>
</table>

SM% – skeletal muscle percentage; BF% – body fat percentage; BMI – body mass index

*Adjusted for age, dietary intake of vitamin D, changes in vitamin levels and physical activity.
children. The results can be caused by this fact that storing vitamin D in obese adipose tissue may have a negative effect on vitamin D bioavailability [12]. Moreover, some other studies have reported reverse correlation between vitamin D and BMI [32–36]. One study on 3528 male and female adolescents aged 12–19 with the aim of investigating vitamin D and cardiovascular risk factors, a significant reverse association was reported between vitamin D level and BMI [35].

However, some other studies found contradictory results. In a study on students (aged 14 to 17) 96% of subjects had vitamin D deficiency but no significant association was found between vitamin D and BMI [37]. More Studies on teenage girls in Malaysia and South Korea [39, 40] and one recent study in Bulgaria [38], have reported no relationship between BMI and vitamin D. In the other study on 11–19-year-old students, Cizmecioglu et al. did not find any association between weight and vitamin D status but they observed a negative correlation between vitamin D and BMI in overweight and obese individuals who suffered from vitamin D deficiency [41]. On the other hand, sex may influence on the level of vitamin D. Muscogiuri et al. found that 25OHD concentration was higher in males compared with females in all BMI groups. Males with vitamin D deficiency had lower fat mass compared with females [42].

The differences in the obtained results of the studies can be due to the difference in the characteristics of the studied populations, race, sex, and the season of the investigation [43, 44]. Nessvi et al. reported that Asians have the lowest level of vitamin D among the world. Also, vitamin D is in the lowest level in winter compared to the other seasons [45]. Levis et al. assessed vitamin D serum in men and women living in south Florida in late summer and winter. The result of seasonal variation identified a significant increase in vitamin D serum level during summer (14% in men and 13% in women) [46]. Turer et al. have investigated prevalence of vitamin D deficiency among 6–18 American children and found a higher prevalence of deficiency among the children with severe obesity (49% and 21% respectively). The lowest prevalence was reported between Caucasians. Also, it was more common in winter/spring than the summer and autumn [47].

No significant association was found between vitamin D serum levels with changes of anthropometric indices after 18 weeks. This may be due to reduced exposure to sunlight due to school exam season [48]. The interactions of obesity and serum level of vitamin D with genotype and gene expression is also frequently reported [49, 50]. The role of genetic profile in serum vitamin D level was reported by Brouwer-Brolsma et al. This study revealed carrier of minor allele of CYP2R1, CYP24A1, and DHCR7 may have higher vitamin D level [51]. Exposure to sunlight, vitamin D intake, and the genotype can explain about 35% of the variations of vitamin D levels [51].

However, this study had some limitations. The level of exposure to sunlight and the participants’ genotype were not exactly examined in our study. We did not assess sun exposure, but all participants were collected of same area that have similar life style and sun exposure. For reducing the bias of seasonal variations, we enrolled all the adolescents in the same season. Another limitation of this study was using BIA for the assessment of body composition which has lower accuracy compared with gold standard methods for anthropometric measurements such as dual X-ray absorptiometry [52].

### Conclusions

Vitamin D serum levels had a reverse association with body weight in male adolescents. However, no significant association was observed between vitamin D serum levels and changes of anthropometric indices after 18 weeks. Future perspective studies with special focus on mediating factors can be helpful in identifying the association of vitamin D with anthropometric measurements.

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**Table IV. Effect of vitamin D serum level on the weight change of participant before and after study**

<table>
<thead>
<tr>
<th>Model</th>
<th>P-value</th>
<th>B (95% CI)</th>
<th>P-value</th>
<th>B (95% CI)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Model 1</td>
<td>0.713</td>
<td>0.987 (0.923–1.056)</td>
<td>0.89</td>
<td>1.004 (0.940–1.074)</td>
</tr>
<tr>
<td>Model 2</td>
<td>0.661</td>
<td>0.896 (0.920–1.054)</td>
<td>0.85</td>
<td>1.006 (0.920–1.054)</td>
</tr>
<tr>
<td>Model 3</td>
<td>0.398</td>
<td>0.960 (0.874–1.055)</td>
<td>0.926</td>
<td>1.004 (0.920–1.097)</td>
</tr>
<tr>
<td>Model 4</td>
<td>0.387</td>
<td>0.957 (0.868–1.057)</td>
<td>0.954</td>
<td>1.003 (0.919–1.096)</td>
</tr>
</tbody>
</table>

Model 1: Adjusted for AGE; model 2: Additional adjustments for physical activity; model 3: Further adjustment for dietary vitamin D intake; model 4: Further adjustment for fat and muscle percentage.
References


31. Rodríguez-Rodríguez E, Navia-Lombán B, López-Sobaler AM, Ortega RM. Associations between abdominal fat and body mass...


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