DOI: 10.4274/gulhane.galenos.2021.46036
Gulhane Med J 2022;64:152-8

Evaluation of 25-hydroxyvitamin D levels in obese individuals

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Date submitted:
05.04.2021
Date accepted:
12.06.2021
Online publication date:
15.06.2022

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Keywords: Adult obesity, vitamin D deficiency, 25(OH)D, parathyroid hormone, body composition

ABSTRACT

Aims: Although 25-hydroxyvitamin D [25(OH)D] deficiency is common, only a few studies have focused on the 25(OH)D level in obese people in Turkey. Therefore, this study assessed the relationship between 25(OH)D status and obesity in the Kayseri region in Turkey.

Methods: This cross-sectional study included otherwise healthy individuals aged 20 to 49 years. Obesity was defined as body mass index (BMI) ≥30. A bioelectrical impedance analyzer was used to measure the body composition. The short form of the "International Physical Activity Questionnaire" was used to assess the physical activity level. Blood 25(OH)D and parathyroid hormone (PTH) levels, dietary intake, body composition, and potential factors associated with the 25(OH)D status were determined.

Results: The study included 268 adults (mean age: 31.4±9.5 years, 69.5% were women). The proportion of vitamin D deficiency (<20 ng/mL) was 84.3% and 83.5% in the obese and normal-weight individuals, respectively. Obese individuals had lower 25(OH)D level [11.6 (8.1-17.9) vs. 13.5 (9.7-18.3) ng/mL, p=0.069] and higher PTH level [44 (35.0-63.0) vs. 36 (28.0-47.0) pg/mL, p<0.001] compared with their normal-weight counterparts. Dietary vitamin D and calcium intake were similar, whereas phosphorous intake was higher in the obese subjects [1052.7 (754.9-1118.4) vs. 945.7 (754.9-1118.4) mg/day p=0.015]. 25(OH)D levels correlated with BMI (p<0.001, r=-0.170), body fat mass (p<0.01, r=-0.179), and muscle mass (p<0.001, r=0.251).

Conclusions: 25(OH)D deficiency is common among obese and normal-weight individuals in this large middle Anatolian city of Turkey. This study found a relationship between serum 25(OH)D levels and BMI, body fat, and muscle mass in the study sample.

Introduction

Vitamin D and parathyroid hormone (PTH) are indispensable elements of calcium homeostasis and bone metabolism (1). Cumulative evidence indicates that vitamin D has a critical function in extra-skeletal health and diseases such as diabetes mellitus, cancers, cardiovascular diseases, and autoimmune disorders (2-4). The best indicator of vitamin D status is the serum 25-hydroxyvitamin D [25(OH)D] levels. Unfortunately, as is known today, most populations worldwide suffer from vitamin D insufficiency (1). Obesity potentially increases the incidence of vitamin D insufficiency since obese people have lower levels of 25(OH)D than the non-obese (5,6).

It has been found that throughout one's life, opposite connections exist among body weight, body mass index (BMI), or body fatness measures with vitamin D status (3,5,7).

Age, race, geography, skin color, habitual dressing style, and sun exposure are listed as some factors leading 25(OH)D levels to be different (8). Additionally, there is an inverse relationship between vitamin D status and BMI, body weight, and body fat. Although Turkey is located in a sun-soaked climate zone, sunlight is not exploited enough for several reasons: dietary habits, skin color, cultural factors, and clothing style. Apart from the study by Aypak et al. (9) and Tosunbayraktar et al. (10), most of the studies in Turkey analyze the 25(OH)D levels of postmenopausal women and the elderly.

Based on the previous studies (3,5,7), it was hypothesized that higher adiposity would lead to lower vitamin D levels. We particularly aimed to (a) evaluate the correlations of serum 25(OH)D with body composition measurements, (b) determine the link between vitamin D and PTH levels with obesity, and (c) compare serum 25(OH)D and PTH levels related to obesity in Kayseri/Turkey.

Methods

Study design

This cross-sectional study was conducted in the Endocrinology Outpatients Clinic of Erciyes University Faculty of Medicine. Individuals were classified according to their BMI as normal-weight and obese. The assignment was made using several factors: wearing a headscarf, sociodemographic features, and physical activity level. A general questionnaire was administered through face-to-face interviews. Biochemical parameters [25(OH)D, PTH], 24-hour dietary intake for three consecutive days (two weekdays and one weekend day), and physical activity levels (PAL) were identified, anthropometric measurements were taken, and bioelectrical impedance analysis was performed.

Written consent was obtained from all participants at the beginning of the study, which was approved by the Ethics Committee of the Faculty of Medicine, Erciyes University, Kayseri, Turkey (approval number: 2013/659, date: 22.11.2013).

Inclusion criteria

Inclusion criteria were as follows: (1) Being between 20 and 49 years of age, (2) applying to the endocrine outpatient clinic at Erciyes University Faculty of Medicine, (3) having a low physical activity level (4) volunteering to give written consent, (4) agreeing to give blood samples.

Exclusion criteria

Individuals with any of the following conditions that could affect vitamin D metabolism were excluded from the study: being under 20 and more than 49 years of age, pregnancy, lactation, postmenopausal status, being underweight (BMI<18.5 kg/m^2), taking dietary vitamin D supplements, alcohol consumption above 50 g/day, more than 3% change in body weight or taking drugs, which could affect body weight in the previous three months, and having comorbid diseases.

Anthropometric measurements

A dietitian performed anthropometric measurements according to the criteria suggested by the World Health Organization (WHO) (11). A bioelectrical impedance analyzer (TANITA MC 780 MA, Tanita Corp., Tokyo, Japan) was used to measure body composition. Participants were instructed to avoid food or liquid intake and vigorous exercise 4 h before the measurement and not wear any metallic objects. Height was measured using a stadiometer with subjects standing barefoot, keeping their shoulders relaxed, arms hanging freely, and the head in Frankfort horizontal plane (12).

Waist and hip circumferences were measured while the individuals were standing, arms were open on both sides, and feet were closed. Waist circumference was measured with a tape measure between the iliac crest and the lowest ribs (mid-point crossing circumference) while the individual was exhaling. The tape measure was positioned horizontally, parallel to the floor, and the measurement was carried out, paying attention not to apply pressure on the skin. Hip circumference was determined from the highest point of the side of the hip (12).

Based on participants' BMI values calculated, individuals were classified as normal-weight (BMI=18.5-24.9 kg/m^2), and obese (BMI≥30 kg/m^2), according to WHO criteria (13).

Assessment of biochemical parameters

After 8-hour fasting overnight, blood samples were collected between 08.30-10.30 am. Then the serum was separated by centrifugation and stored at -80 °C until it was shipped on dry ice to Düzen Laboratory (accredited) in Ankara, where 25(OH) D and PTH analyses were performed. 25(OH)D was measured using Liquid Chromatography and Mass Spectrometry (LC-MS/MS), and PTH (Intact) was measured using a Test Roche Cobas e601 equipment by ECLIA method. Vitamin D status was considered deficient if the serum 25(OH)D level was 20 ng/mL or lower (≤50 nmol/L); inadequate if it was between 21 and 29 ng/mL (52-72 nmol/L); and adequate if it was 30 ng/mL and higher (≥75 nmol/L) (14).

Dietary assessment

The dietary intake was evaluated based on their food consumption frequencies and 24-hour dietary assessment (filled in three consecutive days, including two weekdays and one weekend). The participants were asked to record all the foods and beverages they consumed before the study. A Nutrient Database (BeBiS, Ebispro for Windows, Germany; Turkish version/BeBiS 7) was used to determine participants' energy and nutrient intakes (15,16). Portion sizes were estimated with 2-dimensional food models and a food atlas, including 3 to 5 portion size images of 120 foods (17).

Determination of physical activity levels

The short form of the "International Physical Activity Questionnaire" was used to determine physical activity levels. This form consisted of seven questions and provided information about the time spent sitting, walking, and moderate to vigorous activities. The total score included the duration (minutes) and frequency (days) of the activities performed. Physical activity levels were classified as physically inactive (<600 MET -min/week), low physical activity (600-3000 MET -min/week), and adequate physical activity (with health benefits) (>3000 MET -min/week) (18). Individuals with low activity levels were included in the current study.

Statistical Analysis

Statistical Package for the Social Sciences (SPSS) Statistics for Windows, version 22.0 (IBM Corp., Armonk, NY) was used for statistical analysis. A minimum of 240 samples was evaluated with a 95% confidence interval in NCSS-PASS software, and vitamin D deficiency was estimated to be 32% in obese adults (19). The normality of the data distribution was determined via the histogram, Shapiro-Wilk test, and q-q graphics. Descriptive analysis [mean±standard deviation, median (25 percentile-75 percentile), and frequencies (%)] was performed. Independent Sample t-test and Mann-Whitney U test were used to compare continuous variables, whereas chi-square analysis was used for categorical variables. The Spearman rank correlation test was performed, and scatter plot graphs were drawn to investigate the relationship between quantitative data. p<0.05 was noted to be statistically significant.

Results

Two hundred sixty-eight adults (83 men and 185 women) aged 20-49 years were recruited and classified as normal weight and obese. The measurements of anthropometric and bioelectrical impedance analysis and daily energy and some nutrient intake, and biochemical findings of participants, classified based on BMI values, are presented in Table 1. According to Table 1, 40.6% (BMI=18.5-24.9 kg/m^2, n=109) of the individuals were normal-weight, whereas 59.4% of them were obese (BMI ≥30 kg/m^2 n=159). The mean ages of the normal-weight and obese individuals were 26.2±7.3 and 35.0±9.2 years, respectively (p<0.001). Mean body weight and height for the normal-weight individuals were 63.0±10.6 kg and 166.6±10.2 cm, and 93.7±16.5 kg and 164.4±10.5 cm for the obese individuals. Additionally, the median waist circumference for normal weight and obese subjects was 79.0 cm (71.0-88.0) and 106 cm (99.0-114.0), respectively (p<0.001, Table 1). However, basal metabolic rate, body fat percentage, body fat mass, lean body mass and total body water of the obese participants [1734.0 (1547.0-2071.0) kcal/day, 36.6±6.3%, 56.3±10.6 kg, 55.8 (50.5-68.8) kg, 40.1 (36.1-49.1) kg, respectively] were statistically higher than their normal-weight counterparts [1379.0 (1284.0-1673.0) kcal/day, 23.7±5.4%, 45.7±9.3 kg, 45.2 (41.1-56.4) kg, 32.6 (29.5-40.3) kg, respectively].

Energy intakes of the normal-weight and obese individuals were 1911.7 (1420.3-2233.8) kcal/day and 1639.9 (1320.7-1879.0) kcal/day, respectively (p<0.05). Carbohydrates, protein, and fat percentages were not statistically different between the

Table 1. Body composition, daily energy intake, nutrient intake, and biochemical parameters of the participants

	Obese (n=159, 59.4%)	Normal-weight (n=109, 40.6%)	p
Age (year), mean±SD	35.0±9.2	26.2±7.3	**<0.001***
Body weight (kg), mean±SD	93.7±16.5	63.0±10.6	**<0.001***
Height (cm), mean±SD	164.4±10.5	166.6±10.2	0.085*
BMI (kg/m^2), median (25 percentile-75 percentile)	33.4 (30.8-36.8)	23.0 (20.9-24.1)	**<0.001****
WC (cm), median (25 percentile-75 percentile)	106 (99.0-114.0)	79 (71.0-88.0)	**<0.001****
HC (cm), mean±SD	119.9±10.5	97.3±6.1	**<0.001***
BMR (kcal/day), median (25 percentile-75 percentile)	1734.0 (1547.0-2071.0)	1379.0 (1284.0-1673.0)	**<0.001****
Body fat (%), mean±SD	36.6±6.3	23.7±5.4	**<0.001***
Body fat mass (kg), mean±SD	56.3±10.6	45.7±9.3	**<0.001***
LBM (kg), median (25 percentile-75 percentile)	55.8 (50.5-68.8)	45.2 (41.1-56.4)	**<0.001****
TBW (kg), median (25 percentile-75 percentile)	40.1 (36.1-49.1)	32.6 (29.5-40.3)	**<0.001****
Energy (kcal/day), median (25 percentile-75 percentile)	1911.7 (1420.3-2233.8)	1639.9 (1320.7-1879.0)	**0.001****
Protein (%), median (25 percentile-75 percentile)	15.0 (13.0-16.0)	15.0 (13.0-18.0)	0.120**
Fat (%), mean±SD	35.8±6.3	35.9±5.7	0.868*
Carbohydrate (%), mean±SD	49.3±7.1	48.7±6.6	0.518*
Calcium (mg/day), median (25 percentile-75 percentile)	627.7 (467.5-833.8)	603.3 (468.9-777.3)	0.395**
Phosphorus (mg/day), median (25 percentile-75 percentile)	1052.7 (754.9-1118.4)	945.7 (754.9-1118.4)	**0.015****
Vitamin D (mcg/day), median (25 percentile-75 percentile)	1.3 (0.9-2.1)	1.2 (0.7-1.9)	0.179**
25(OH)D (ng/mL), median (25 percentile-75 percentile)	11.6 (8.1-17.9)	13.5 (9.7-18.3)	0.069**
PTH (pg/mL), median (25 percentile-75 percentile)	44 (35.0-63.0)	36 (28.0-47.0)	**<0.001****

*Student's t-test, **Mann-Whitney U test.
WC: Waist circumference, HC: Hip circumference, BMR: Basal metabolic rate, LBM: Lean body mass, TBW: Total body water, SD: Standard deviation, PTH: Parathyroid hormone, BMI: Body mass index

normal-weight and obese subjects (p>0.05). Moreover, dietary intake of vitamin D and calcium was similar between the normal-weight and obese counterparts (p>0.05). However, the dietary phosphorous intake of the obese participants [1052.7 (754.9-1118.4)] mg/day was higher than that of those with normal-weight participants [945.7 (754.9-1118.4)] mg/day (p<0.05) (Table 1).

Although it was statistically insignificant, 25(OH)D levels of the normal-weight individuals were higher than those of the obese [13.5 (9.7-18.3) ng/mL and 11.6 (8.1-17.9) ng/mL, respectively]. The PTH levels of the obese subjects [44 (35.0-63.0) pg/mL] were statistically higher than those of the normal weight [36 (28.0-47.0) pg/mL, p<0.001] (Table 1).

Vitamin D levels of the participants according to BMI classification and gender are shown in Table 2. Vitamin D deficiency was 84.3% in the obese individuals and 83.5% in their normal-weight subjects (<20 ng/mL). Additionally, vitamin D insufficiency (20-29.9 ng/mL) was detected in 14.4% of the obese participants and 14.7% of the normal-weight participants.

Correlations between 25(OH)D and BMI, body fat mass, and body muscle mass are shown in Figures 1, 2, and 3. A negative, weak, and statistically significant relationship was found between 25(OH)D, BMI (p<0.001, r=-0.170), and body fat mass (p<0.01, r=-0.179). Additionally, a positive, weak, and statistically significant relationship was found between muscle mass (p<0.001, r=0.251) and 25(OH)D (Figures 1, 2, 3).

Discussion

The most notable finding in the current study was that serum 25(OH)D levels appeared to be associated with BMI, body fat, and muscle mass. Although no statistically significant difference was observed, median 25(OH)D levels of the obese were lower than those of the normal-weight in the current study. Accordingly, 84.3% of obese individuals were found to have vitamin D deficiency. Although they had higher levels of vitamin D deficiency than those with normal body weight (83.5%), no statistically significant difference was found between the groups. Previous studies have stated that vitamin D deficiency may be a risk factor for obesity. However, the mechanisms behind lower levels of 25(OH)D and higher PTH have not been fully understood. Several hypotheses have been proposed about this relationship (6,9,20,21). Aypak et al. (9) stated that an increase in BMI causes 25(OH)D levels to decrease and that obese individuals are at risk of 25(OH)D deficiency. In Parikh et al.'s (20) study, that serum 25(OH)D levels of the obese (23.5±12.2 ng/mL) were significantly lower than that of the normal weight individuals (31±14.4 ng/mL). However, Karlsson et al. (21)

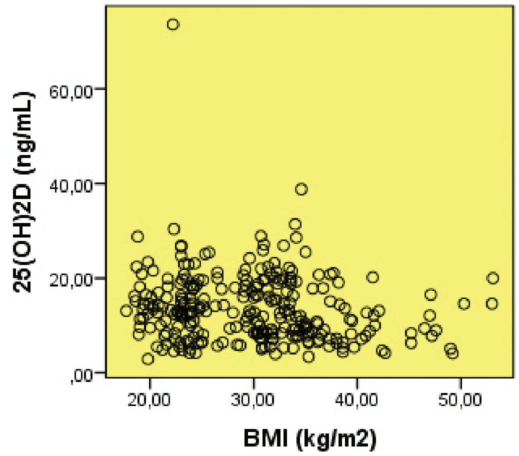

Figure 1. Correlation between 25(OH)D level and BMI. A negative, weak and statistically significant relationship was found between 25(OH)D, and BMI (p<0.001, r=-0.170)

BMI: Body mass index

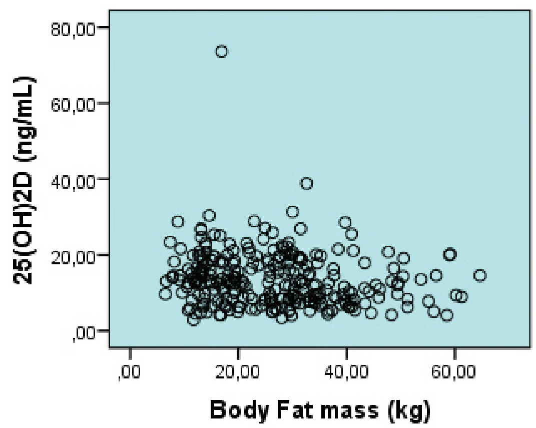

Figure 2. Correlation between 25(OH)D level and body fat mass. A negative, weak and statistically significant relationship was found between 25(OH)D, and body fat mass (p<0.01, r=-0.179)

Table 2. Vitamin D levels of the participants according to BMI categories and gender

	Obese (n=159, 59.4%)			Normal weight (n=109, 40.6%)		
	Men	**Women**	**Total**	**Men**	**Women**	**Total**
Deficiency (<20 ng/mL), n (%)	38 (70.3)	96 (91.4)	134 (84.3)	17 (58.6)	74 (92.5)	91 (83.5)
Insufficiency (20-29.9 ng/mL), n (%)	15 (27.8)	8 (7.6)	23 (14.4)	12 (41.4)	4 (5.0)	16 (14.7)
Normal (≥30 ng/mL), n (%)	1 (1.9)	1 (1.0)	2 (1.3)	-	2 (2.5)	2 (1.8)
	p=0.001*			p<0.001*		

*Pearson chi-square test; BMI: Body mass index

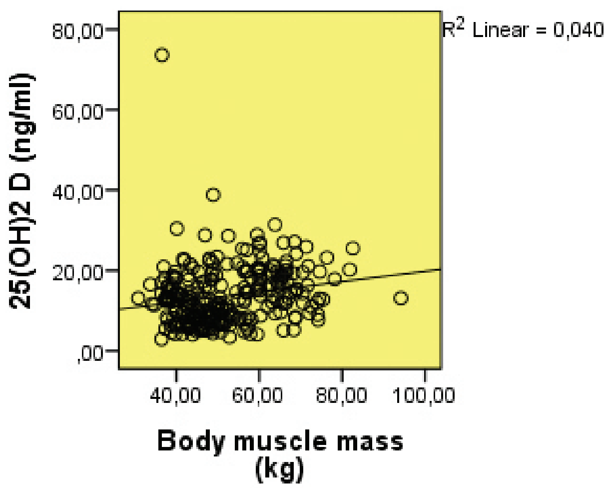

Figure 3. Correlation between 25(OH)D level and body muscle mass. A positive, weak and statistically significant relationship was found between 25(OH)D, and body muscle mass ($p<0.001$, $r=0.251$)

determined that although obese pregnant women received a greater amount of dietary vitamin D than normal-weight women, the former had lower levels of serum 25(OH)D. Likewise, Mai et al. (6) identified a negative relationship between low serum 25(OH)D levels and BMI in the Trøndelag health study (HUNT) on 26.616 adults between the ages of 19-55. In contrast, Karlsson et al. (21) found similar results on the dietary vitamin D levels of the obese and normal-weight individuals.

In this study, the median serum PTH levels of the obese were significantly higher than that of the individuals with normal body weight, which was in concordance with other studies (22,23). The current explanation for this phenomenon is the increased sequestration of 25(OH)D in excess subcutaneous fat, ultimately decreasing the bioavailability of vitamin D for calcium absorption. The diminished availability of serum 25(OH) D causes a compensatory increase in PTH secretion to maintain serum calcium concentrations (22,23). We think that our results confirm this statement.

Moy and Bulgiba (24) identified that women have lower levels of 25(OH)D and a higher frequency of 25(OH)D deficiency. This study also found, in compliance with the literature, that both obese (91.4%) and normal-weight (92.5%) women had significantly higher vitamin D deficiency (<20 ng/mL) than obese (70.3%) and normal-weight men (58.6%). It is speculated that the intersexual difference in 25(OH)D levels originates from the fact that women are less exposed to exposure sunlight exposure than men because of cultural reasons such as different dressing styles. Previous studies have demonstrated that low 25(OH)D levels of the obese were associated with lifestyle factors such as physical activity, alcohol consumption, and smoking status (25,26). Although individuals were assigned to groups based on several factors (such as wearing a headscarf, sociodemographic features, and physical activity level), there were no differences in vitamin D levels between obese and normal-weight individuals. Moreover, studies have stated that low 25(OH)D levels of the obese were associated with dietary factors such as low dietary vitamin D and calcium (25,26). In contrast to these study results, dietary vitamin D and calcium intake were similar between normal and obese subjects.

The finding of lower serum 25(OH)D concentration in higher fat mass in the current study agrees with several other observational studies (27-29). Prior studies largely support a positive association between higher serum 25(OH)D concentration and better muscle mass (30,31). Similarly, this study generated a positive and significant relationship between 25(OH)D levels and muscle mass.

There are some strengths and limitations of this study. First, only a few studies have investigated the link between vitamin D and PTH levels and obesity in Turkey. Second, the author conducted all the interviews, thus ensuring a consistent technique and interpretation of the answers given. However, bias may arise because of the study design and the type of sampling as the work was designed as a pilot study, and therefore, the sample chosen may not represent the general population, and the conclusions cannot be generalized. Future studies should consider multiple centers. Due to the differences in demographic features, geographic regions, assessment methods of 25(OH)D level, and threshold values, it is not easy to compare vitamin D status found in different studies.

Conclusion

This study showed that 25(OH)D deficiency is widespread in obese and normal-weight people and that the differences are not significant. Further studies are warranted to investigate the potential pathological mechanisms and establish optimal strategies to improve vitamin D status in obese and normal-weight populations.

Acknowledgments

We would like to thank the physicians involved in data collection and the women who participated in the study.

Ethics

Ethics Committee Approval: The study was approved by the Ethics Committee of the Faculty of Medicine, Erciyes University, Kayseri, Turkey (approval number: 2013/659, date: 22.11.2013).

Informed Consent: A consent form was filled out by all participants.

Peer-review: Externally peer-reviewed.

Authorship Contributions

Surgical and Medical Practices: K.Ü., Concept: G.K., N.İ., Design: G.K., N.İ., E.B., Data Collection or Processing: G.K., E.B., K.Ü., Analysis or Interpretation: G.K., N.İ., E.B., Literature Search: G.K., N.İ., E.B., K.Ü., Writing: G.K., N.İ.

Conflict of Interest: No conflict of interest was declared by the authors.

Financial Disclosure: This work was supported by the Scientific and Technological Research Council of Turkey (TÜBİTAK) 3001 Research & Development Projects Funding Program (approval number: 114S043).

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