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# *The Effect of Moderate Intensity CrossFit Training on Serum Vitamin D Levels and Anthropometric Measures in Overweight/Obese Adult Women: A Randomized Controlled Trial*

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***ABSTRACT***

**Background:** Vitamin D deficiency is common among women, particularly those who are overweight or obese. There is evidence that physical activity benefits on body vitamin D status. Therefore, this study assessed the effect of CrossFit training on serum 25(OH)$D_3$ levels and anthropometric measures in women. **Methods:** This randomized clinical trial enrolled 28 overweight/obese adult women. Participants were randomly allocated to a CrossFit (n=14) or a non-intervention group (n=14) for 8 weeks. Participants were then asked to complete CrossFit training for 1 hour, 3 times per week. Fasting levels of 25(OH)$D_3$ and anthropometric measurements, including weight, height, body mass index (BMI), waist-to-hip ratio (WHR), lean body mass (LBM), and body fat mass (BFM), were measured for each participant at the beginning and end of the study. **Results:** In total, 24 women (12 participants in the CrossFit and 12 participants in the non-intervention group) completed the trial. After 8 weeks of intervention, it was found a significant increase in serum 25(OH)D3 levels in the CrossFit group compared with other group (1.11±0.45 ng/ml in the CrossFit group vs. 0.19±0.44 ng/ml in the non-intervention group, *P*<0.01). Furthermore, in the CrossFit group, significant decreases were found in body weight, BMI, and WHR and a significant increase in LBM, compared with the non-intervention group (*P*< 0.05). **Conclusion:** Findings indicate that 8 weeks of CrossFit training can improve vitamin D levels and anthropometric measures in overweight/obese adult women.

## Introduction

The prevalence of obesity is increasing at an alarming rate (Engin, 2017). It has been estimated that 66% of adults in the US are overweight or obese (Elagizi *et al.*, 2020). In addition, obesity is prevalent among more than 50% of Iranian adults (Rahmani *et al.*, 2015). Obesity is associated with multiple adverse outcomes such as diabetes, cardiovascular diseases, cancer, and mortality (Piché *et al.*, 2020). Moreover, micronutrient deficiencies are other complications of obesity (Pellegrini *et al.*, 2021). Among the deficiencies, vitamin D deficiency has received significant attention (Fiamenghi and Mello, 2021). A meta-analysis showed that vitamin

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D deficiency was 35% higher in obese subjects compared to the normal-weight group (Pereira-Santos *et al.*, 2015). Hence, finding effective strategies to control vitamin D levels in obese individuals is of great importance.

Several approaches, including vitamin supplementation, food fortification, and high exposure to sunlight, have been suggested for controlling vitamin D deficiency (Holick, 2017, Holick *et al.*, 2011). However, these approaches may be costly and impose complications for people (Kennel *et al.*, 2010). For instance, exposure to sunlight may increase the risk of skin cancers (Holick, 2020). Recently, it has been suggested that physical activity may affect plasma nutrient levels, particularly serum 25(OH)$D_3$ (Fernandes and Barreto, 2017). In a review article, Maïmoun et al. concluded that physical activity can increase vitamin D metabolites through influencing calciotropic hormones (Maïmoun and Sultan, 2009). Also, it has been shown that different training programs can affect vitamin D levels and body composition (De-la *et al.*, 2022, Zhang and Cao, 2022).

CrossFit training is a form of cardiovascular exercise that alternates between short bursts of intense anaerobic activity and periods of lower-intensity recovery or rest (Claudino *et al.*, 2018). It is proposed that CrossFit training has a greater beneficial effect on body composition and other health parameters than regular exercise (Meyer *et al.*, 2017). Furthermore, because CrossFit training requires a shorter time commitment than regular exercise, the commitment to doing it increases (Schlegel, 2020).

Based on the literature search, the authors found no studies investigating the effect of CrossFit training on serum 25(OH)$D_3$ levels in adults. However, there is evidence for other types of exercises. In a randomized controlled trial (RCT) involving male adolescents aged 12 to 16 years, Haghshenas *et al*. reported that 8 weeks of high-intensity interval training (HIIT) could increase serum concentrations of 25(OH)$D_3$ (Haghshenas *et al.*, 2019). However, in a meta-analysis of RCTs, Khan *et al*. concluded that physical activity has no significant effect on serum 25(OH)$D_3$ (Khan *et al.*, 2024). Investigating the effect of CrossFit training on serum 25(OH)$D_3$ is particularly relevant among Iranian women with special coverage and lower exposure to sunlight (Tabrizi *et al.*, 2018). Therefore, vitamin D deficiency is more common among Iranian women compared to Western women (Lips, 2007). This study aimed to investigate the effects of 8 weeks CrossFit training on serum vitamin D levels and anthropometric measures in overweight/obese adult women in Iran.

## Materials and Methods

### *Participants*

This is a randomized clinical trial conducted in Yazd, Iran. Twenty-eight healthy overweight/obese adult women were voluntarily enrolled in this study. The participants were recruited through invitations in local media and on social networks. Then, individuals interested in participating in the current study were first assessed against inclusion and exclusion criteria; those who met the criteria were included. Inclusion criteria in this study were: (1) women aged between 30 and 50, (2) body mass index (BMI) between 25 and 35, (3) being sedentary (<20 minutes of moderate-intensity physical activity on 3 days in a week over the past 3 months). Participants were omitted if they were: (1) suffering from chronic cardio metabolic diseases, cancer, chronic kidney disease (CKD), diabetes mellitus, (2) taking any medications related to chronic diseases, (3) having an acute illness that negatively affects sport performance, (4) taking vitamin D or other nutrient supplements during the past 6 months, (5) adhering to a weight loss diet, (6) and being in pregnancy and lactation periods. During the study periods, the authors excluded participants who did not adhere to the administered interventions, those with bone or muscle injuries during the intervention, those unwilling to continue the trial, and those who took a vitamin D supplement during the trial.

### *Sample size calculation*

Considering a type I error of 5% ($\alpha$=0.05), a type II error of 20% ($\beta$=0.20, power=80%), and



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vitamin D level as the key variable, the authors manually calculated the required sample size using the following formula. The mean and standard deviation of vitamin D levels were extracted from the Malandish *et al.* study (Malandish *et al.*, 2020). Overall, based on the formula and given a 10% dropout rate in each group, the study included 14 individuals per group.

$$n = \frac{2[(a+b)^2 \times \sigma^2]}{(\mu_1 - \mu_2)^2}$$

*n=sample size in each group; µ1=mean for vitamin D level in group 1 (mean=11.42 ng/ml); µ2 = mean for vitamin D level in group 2 (mean=5.34 ng/ml); σ=the highest SD for mean concentrations of vitamin D level (SD=5.28); a=conventional multiplier for alpha = 0.05 that was 1.96; b=conventional multiplier for power = 0.80 that was 0.842.*

### *Study design*

After screening for eligibility, 28 participants were enrolled according to the inclusion/exclusion criteria. All participants underwent medical evaluations by a specialist and completed a sports medicine questionnaire to confirm eligibility. Qualified participants were randomly allocated into two groups: 1) Exercise group (n=14): CrossFit training (3 days/week, 1 hour/session for 8 weeks) and 2) Non-intervention group (n=14): Maintained normal daily activities. Moreover, stratified simple randomization was used for random allocation. Before randomization, women were stratified by age (30 to 40 and 40 to 50 years), and women within the same age group were randomly allocated to the two groups using a lottery. To randomize, an identification code was assigned to each eligible woman, and the code of a woman in the same age group was placed in the lottery container; finally, women in the same age group were randomly assigned to the CrossFit or Non-intervention groups. A person who was unaware of the aim of this study conducted the random allocation. To assess serum 25(OH)$D_3$ levels, a fasting 5 ml blood sample was collected from each participant at the beginning and 48 hours after the last training session. Also, anthropometric measures were assessed at baseline and at the end of the trial (1 day after the last training session). To adjust exposure to sunlight, the training group performed CrossFit training in an indoor gym. Participants were asked not to change their habitual diet or sun exposure throughout the study. In addition, individuals in the CrossFit and Non-intervention groups were asked to continue their usual physical activity throughout the trial, as they had before the trial.

To control dietary intakes during the trial and before the study, all participants completed a validated Food Frequency Questionnaire at the end of the study (Moridpour *et al.*, 2022). Then, the authors converted the dietary reports to grams using household measures available in booklets. To estimate nutrient intakes from these dietary intakes, they used Nutritionist IV software (based on the US National Nutrient Databank) modified for Iranian foods.

### *Intervention*

CrossFit training methodology is fundamentally based on the systematic integration of three primary exercise modalities: weightlifting (utilizing external loads), gymnastics (employing bodyweight resistance), and metabolic conditioning (high-intensity cardiovascular exercises) (Kliszczewicz *et al.*, 2015). These modalities are implemented either independently or in combination to optimize both absolute and relative strength development (Schlegel, 2020). The traditional CrossFit session structure consists of three core components: warm-up, preparatory phase, and the Workout of the Day (WOD), though contemporary programming has expanded to include additional modality-specific skill development segments (Gong *et al.*, 2025).

In the 8-week intervention protocol, each training session followed a standardized structure beginning with a 10-minute dynamic warm-up comprising stretching and locomotor exercises. The core training component involved circuit-style implementation of combined modalities performed in multiple rounds with active recovery intervals. Sessions concluded with a 10-minute cool-down period. In addition, exercise intensity was progressively increased from 60-65% of target heart rate and 60% of One-repetition maximum (1RM) in



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weeks 1-2 to 65-70% of target heart rate and 70% of 1RM in weeks 3-8. The program incorporated diverse movement patterns, including weight-bearing exercises (squats, deadlifts, thrusters), bodyweight movements (push-ups, sit-ups, back extensions), and cardiorespiratory activities (running, rope skipping), performed in both loaded and unloaded variations.

***Vitamin D assessment***

After 12 hours of fasting, a 5 ml blood sample was collected from all participants at study baseline and 48 hours after the last training session. To control for the effect of physical activity on $25(OH)D_3$ levels, participants in both groups were asked to avoid strenuous physical activity 24 hours before blood sampling. Serum was isolated from the whole blood and was stored at -20 °C until further analysis. A direct competitive enzyme-linked immunosorbent assay (ELISA) method was then used to measure serum concentrations of $25(OH)D_3$ (Pishtazteb, Iran) (Hollis and Horst, 2007). According to the results, vitamin D levels of <20 ng/ml were considered deficient, 20-30 ng/ml insufficient, 30-100 ng/ml sufficient, and >100 ng/ml toxic (Holick *et al.*, 2011).

***Anthropometric measures***

Weight was measured using a digital scale, with the subject in a state of minimum clothing and without shoes, to the nearest 100 grams. Standing height was measured using a standard stadiometer, without shoes, to the nearest 0.5 cm. BMI was calculated as weight (in kilograms) divided by height (in meters) squared. To measure other anthropometric measures, including waist-to-hip ratio (WHR), lean body mass (LBM), and body fat mass (BFM), the bio-electrical impedance analysis (BIA) method using a Tanita body composition analyzer (Model BC-418MA) was used.

***Ethical considerations***

All participants provided oral and written informed consent after reading and understanding the details of the exercise programs and the experimental procedures. The study was approved by the Ethics Committee of Islamic Azad University, Yazd, Iran (registration No: 392435).

***Data analysis***

All analyses were conducted in SPSS version 21 (SPSS, Inc., Chicago, IL, USA). The authors first examined the normality of continuous variables using skewness and performed a Log transformation for non-normally distributed variables. The authors also used a paired-samples t-test to assess the effect of CrossFit training on outcome variables within a group. To assess differences between the CrossFit and Non-intervention groups on continuous variables and changes in outcome variables, the researchers used independent-samples *t-tests*. In addition, Analysis of covariance (ANCOVA) was used to assess the effect of CrossFit training on outcome variables by controlling for baseline measurements and energy intake during the trial. In addition, since the baseline weight was significantly different between the two groups, the authors controlled for the variable in the analysis of vitamin D. For anthropometric measures, baseline weight was not adjusted because it was collinear with baseline values of the anthropometric measures. Since dietary intakes of some nutrients differed between the CrossFit and control groups, the researchers controlled for energy intake to account for these differences. Also, a p-value less than 0.05 was considered significant.

## Results

From the 28 participants at baseline, two in the CrossFit training group (due to Non-adherence to the study protocol) and two in the Non-intervention group (due to personal reasons and unwillingness) were excluded from the study. Therefore, 24 participants [CrossFit training group (n=12) and Non-intervention group (n=12) completed the trial (**Figure 1**). In this trial, all participants in the intervention group attended all training sessions, resulting in a 100% adherence to the intervention protocol.

The baseline data of participants in the intervention and Non-intervention groups are shown in **Table 1**. There was no statistically significant difference in age, height, and BMI



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between the two groups. However, women in the CrossFit group had significantly greater weight than the control group (*P*=0.02). The dietary intakes of women in the CrossFit training and Non-intervention groups are presented in **Table 2**. Accordingly, women in the CrossFit training group had higher intakes of energy, carbohydrates, fat, calcium, vitamin C, saturated fatty acids (SFAs), monounsaturated fatty acids (MUFAs), and polyunsaturated fatty acids (PUFAs) than those in the Non-intervention group. No significant differences were found for other dietary factors.

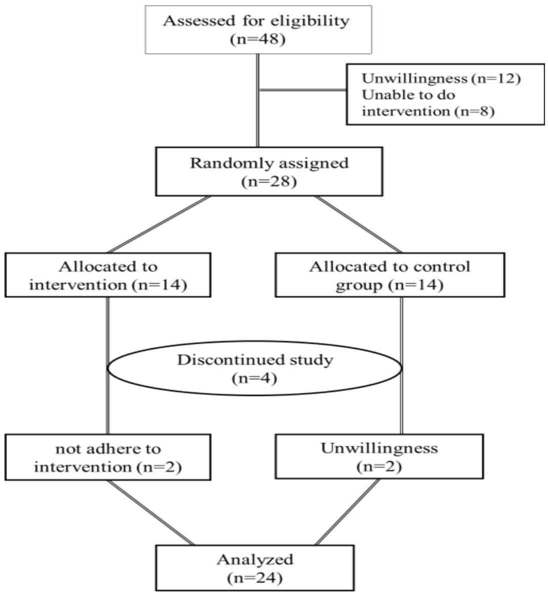


**Figure 1.** Flowchart of selection of study participants

The effects of CrossFit training on serum levels of $25(OH)D_3$ and anthropometric measures in women are shown in **Table 3**. Based on within-group comparison, adherence to the CrossFit training significantly increased $25(OH)D_3$ levels and LBM and reduced WHR, BFM, and BMI throughout the trial. In the between-group comparison, the observed changes in the CrossFit training group were significant compared to the Non-intervention group. When analyses were controlled for baseline measurements and energy intake throughout the trial, we found no changes in any outcomes. However, the reduction in WHR due to CrossFit training was marginally significant (*P*=0.05).

## Discussion

The results of this study showed that CrossFit training significantly increased vitamin D levels compared to the Non-intervention group. The findings also indicated that CrossFit training caused a significant reduction in WHR, BFM, and BMI and a significant increase in LBM in overweight/obese adult women. However, the effect of CrossFit training on WHR was marginally significant.

**Table 1.** Baseline characteristics of women participated in the current study.

| Variables | CrossFit (n=12) | Non-intervention (n=12) | P-value[a] |
|---|---|---|---|
| Age (year) | 38.16±7.98 | 37.75±7.02[b] | 0.89 |
| Weight (kg) | 82.40±13.03 | 71.25±8.43 | 0.02 |
| Height (cm) | 163.00±7.37 | 158.66±5.48 | 0.11 |
| BMI (kg/m$^2$) | 29.72±3.23 | 28.22±2.15 | 0.19 |

***BMI***: *Body mass index;* [a]*: Obtained from the independent sample t test;* [b]*: Mean ± SD.*

**Table 2.** Dietary intakes of women participated in the current study.

| Variables | CrossFit (n=12) | Non-intervention (n=12) | P-value[a] |
|---|---|---|---|
| Energy (kcal) | 2624.07±452.61 | 2246.40±183.51 | 0.01 |
| Protein (g/d) | 82.07±13.80[b] | 74.19±10.79 | 0.13 |
| Carbohydrate (g/d) | 379.86±63.73 | 329.82±51.35 | 0.04 |
| Fat (g/d) | 89.46±19.25 | 72.51±14.43 | 0.02 |
| Fiber (g/d) | 18.60±2.41 | 14.86±1.63 | <0.001 |
| Calcium (mg/d) | 607.15±121.31 | 480.97±79.30 | 0.00 |
| Vit C (mg/d) | 106.73±15.80 | 74.58±13.4 | <0.001 |
| Vit D (µg/d) | 2.24±1.50 | 2.00±0.81 | 0.64 |
| MUFA (g/d) | 32.78±7.96 | 26.71±6.18 | 0.04 |
| PUFA (g/d) | 22.67±4.93 | 17.73±2.90 | 0.00 |
| SFA (g/d) | 24.54±5.18 | 20.41±4.62 | 0.05 |

***MUFA****: monounsaturated fatty acid;* ***PUFA****: Polyunsaturated Fatty Acid;* ***SFA****: saturated fatty acid;* [a]*: Obtained from the independent;* [b]*: Mean ± SD.*

Vitamin D deficiency is common among women, particularly among those with overweight and obese (Rodríguez-Rodríguez *et al.*, 2009). Vitamin D deficiency is associated with an increased risk of osteoporosis, infections, autoimmune and cardiovascular diseases, depression, and mental disorders (Johnson and Thacher, 2023). Vitamin supplementation is the best way to control vitamin D deficiency (Pludowski *et al.*, 2024). However, the influence of other methods, such as physical activity, has long been a topic of debate among researchers (Al-Othman *et al.*, 2012). Previous studies have shown that some types of physical activity, such as HIIT, may favorably affect serum 25(OH)$D_3$ (Haghshenas *et al.*, 2019). Based on the authors' knowledge, no study has investigated the effect of CrossFit training on serum 25(OH)$D_3$ concentrations in obese women. This is the first study on this topic, in which the researchers observed that CrossFit training significantly increased serum 25(OH)$D_3$ levels in obese women. In line with the findings, Haghshenas *et al.* reported that HIIT for 3 days per week over 8 weeks could significantly increase serum 25(OH)$D_3$ levels in adolescents (Haghshenas *et al.*, 2019). Also, in another clinical trial, Sun *et al.* showed that 12 weeks of resistance training could enhance plasma vitamin D levels in young men (Sun *et al.*, 2020).

**Table 3.** The effect of CrossFit training on serum levels of 25(OH)D3 and anthropometric measures in women with overweight or obesity.

| Variables | CrossFit | | | | Non-intervention | | | | P-value[b] |
|---|---|---|---|---|---|---|---|---|---|
| | Before | After | Change | P-value[a] | Before | After | Change | P-value[a] | |
| 25(OH)$D_3$ (ng/ml) | 26.12±12.04 | 27.24±11.99 | 1.11±0.45 | <0.001 | 25.95±9.37 | 26.14±9.62 | 0.19±0.44 | 0.16 | <0.001 |
| Weight (kg) | 82.40±13.03 | 80.15±13.14 | -2.25±1.01 | <0.001 | 71.25±8.43 | 71.40±8.32 | 0.16±0.56 | 0.35 | <0.001 |
| BMI (kg/m$^2$) | 29.72±3.23 | 28.90±3.27 | -0.82±0.37 | <0.001 | 28.22±2.15 | 28.29±2.13 | 0.06±0.20 | 0.27 | <0.001 |
| LBM (%) | 29.55±6.11 | 30.00±6.06 | 0.45±0.23 | <0.001 | 25.43±2.71 | 25.41±2.75 | -0.16±0.32 | 0.86 | <0.001 |
| BFM (%) | 34.55±6.13 | 33.44±6.17 | -1.10±0.17 | <0.001 | 34.64±3.25 | 34.81±3.26 | 0.17±0.64 | 0.37 | <0.001 |
| WHR | 0.89±0.49[c] | 0.88±0.05 | -0.01±0.00 | <0.001 | 0.88 ± 0.06 | 0.89 ± 0.06 | 0.00±0.00 | 0.58 | 0.03 |

*[a]: Paired sample t test; [b]: Independent sample t test; [c]: Date are presented as mean ± SD;* ***BMI****: Body mass index;* ***LBM****: Lean body mass;* ***BFM****: Body fat mass;* ***WHR****: Waist-to-hip ratio.*

**Table 4.** Adjusted mean for the effect of CrossFit training on serum levels of 25(OH)D3 and anthropometric measures in women with overweight or obesity.

| Variables | CrossFit (n=12) | Non-intervention (n=12) | P-value[a] |
|---|---|---|---|
| 25(OH)$D_3$ (ng/ml) | 0.96±0.15[b] | 0.35±0.15 | 0.02 |
| Weight (kg) | -1.97±0.25 | -0.12±0.25 | <0.001 |
| BMI kg/m$^2$) | -0.73 ± 0.08 | -0.02±0.08 | <0.001 |
| LBM (%) | 0.41±0.10 | 0.01±0.10 | 0.02 |
| BFM (%) | -0.89±0.21 | -0.04±0.21 | 0.01 |
| WHR | -0.01±0.003 | -0.001±0.003 | 0.05 |

***BMI****: Body mass index;* ***LBM****: Lean body mass;* ***BFM****: Body fat mass: WHR: waist-to-hip ratio; [a]: covariance (ANCOVA) adjusted for baseline measurements and energy intake during the study. For 25(OH)D3, the authors additionally adjusted for baseline weight; [b]: Mean ± SE.*

In contrast to the mentioned studies, a systematic review and meta-analysis of clinical trials concluded that physical activity interventions have no significant effect on serum 25(OH)$D_3$ concentrations, independent of the time spent outdoors (Khan *et al.*, 2024). The most important reason for the observed disparity is the different types of physical activities administered in the previous studies. For example, most studies included in the systematic review administered low-intensity exercise interventions, whereas all studies that administered high-intensity exercise showed a significant increase in serum 25(OH)D3 concentrations. Also, in most studies included in the systematic review, serum 25(OH)D3 concentrations were assessed as a secondary outcome. In addition, differences in population features, sample sizes, and the lack of adjustment for confounding variables are other reasons for the observed discrepancy.

It has been hypothesized that physical activity



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may influence serum 25(OH)$D_3$ levels by increasing fat metabolism, thereby releasing vitamin D and/or its metabolites from adipose tissue (Vázquez-Lorente *et al.*, 2020). Numerous observational studies have supported this hypothesis, showing positive associations between physical activity and 25(OH)$D_3$ concentrations or vitamin D status (Koda *et al.*, 2023). Moreover, physical activity may indirectly influence vitamin D levels by altering hormonal activity. There is evidence regarding the increasing effect of physical activity on parathyroid hormone (PTH) release. This effect can increase vitamin D absorption in the gut (Peeling *et al.*, 2013). It has also been shown that people with physical training have greater calcium absorption than those with a sedentary lifestyle. This might demonstrate the mediating role of vitamin D levels in the relationship between physical activity and intestinal calcium absorption (Zittermann *et al.*, 2000).

The results showed that an 8-week CrossFit training program can significantly reduce weight, BMI, BFM, and WHR, and increase LBM in obese women. These effects of CrossFit training are consistent with several studies indicating that physical activity improves anthropometric measures in adults. In a systematic review and meta-analysis of randomized clinical trials, Donoghue *et al.* illustrated that physical activity programs can decrease weight and improve other anthropometric measures in adults. Moreover, this systematic review revealed that combining high-intensity aerobic exercise with resistance training has greater effects on anthropometric measures than low-intensity exercises alone (O'Donoghue *et al.*, 2021). Seo *et al*. reported that twelve weeks of resistance training elicited substantial improvements in body composition, muscle hypertrophy, and blood lipid profiles in middle-aged obese women (Seo *et al.*, 2023). Similarly, Feito *et al*. found that an eight-week high-intensity functional training protocol significantly enhanced glucose regulation and reduced adiposity in overweight individuals (Feito *et al.*, 2019). In total, CrossFit training is a good choice for improving anthropometric measurements.

This study had some strengths; it was the first randomized clinical trial to examine the impact of CrossFit training on serum 25(OH)$D_3$ levels in overweight and obese women. In this study, the authors controlled for baseline measurements and energy intake to reduce the effects of confounding variables. In addition, adherence to the intervention was 100% in this study. Despite the strengths, the study had some limitations. The sample size was small, which prevented the researchers from conducting subgroup analyses by obesity status and other important confounders. The usual physical activity during the study could not be assessed; however, participants were asked not to change their physical activity compared to the time before the study. Furthermore, sunlight exposure was not examined in this study, although Iranian women usually have similar exposure to sunlight because of the special clothing they wear. However, individual differences in sunlight exposure may still occur. Variations in outdoor activities, lifestyle, and seasonal sunlight may have influenced serum vitamin D levels, and consequently, the findings of this study.

## Conclusion

It was found that CrossFit training significantly increased 25(OH)$D_3$ in overweight and obese women. In addition, this type of exercise could improve anthropometric measures in this population. Further studies with larger sample size are needed to confirm the findings in this regard. Future studies can examine the influence of indoor and outdoor physical activity on vitamin D levels in adults.

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## Authors' contributions

Emami A and Dehghanpour Farashah F contributed to study conception and study design. Dehghanpour Farashah F contributed to data



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collection and administration of interventions. Emami A and Dehghanpour Farashah F were involved in data interpretation and critically reviewed and revised the manuscript. Emami A and Dehghanpour Farashah F were involved in study conception, data analysis and interpretation, and manuscript drafting, and critically reviewed and revised the manuscript. All authors reviewed and approved the final manuscript. The corresponding authors confirmed that all authors met the criteria for authorship and that no eligible authors were omitted.

## Conflicts of interest

Authors declared no conflicts of interest.

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