The relationship of serum 25-dihydroxy vitamin D3 concentrations with metabolic parameters in non-obese women with polycystic ovarian syndrome

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Abstract  Introduction: Polycystic ovarian syndrome (PCOS) is one of the most common endocrine disorders among women of reproductive age, which is associated with an increased risk of developing metabolic syndrome (MBS). Recently, many studies have started to dig up the dramatic effect of vitamin D deficiency on PCOS patients suffering from the MBS. However, little is known about the role of this fat-soluble agent in non-obese PCOS women. Aim: To investigate the relationship between serum 25-dihydroxy vitamin D3 [25(OH) D] levels and metabolic parameters in non-obese women with PCOS. Materials and methods: Eighty-eight non-obese women who underwent PCOS were experimentally evaluated. Patients were divided into two categories according to their body mass index (BMI): patients with normal weight and overweight patients. The serum levels of 25(OH) D, FBS, CRP, TC, TG, LDL, HDL, insulin as well as the insulin resistance indexes were assessed in all the study patients. Results: The mean age of the patients was 27 years, 36 (40.9%) had normal weight and 52 (59.1%) were overweight. Vitamin D deficiency was observed in 84.1% of the patients, but its variations between both groups were not significantly different. A significant correlation was found between 25(OH) D levels and the patients’ age, as well as the HDL serum concentration. Furthermore, there was no significant association between 25(OH) D levels and other metabolic parameters.

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1. Introduction

Polycystic ovarian syndrome (PCOS) is one of the most common endocrine disorders among women of reproductive age. Its worldwide prevalence has been estimated between 2.2% and 26%, which is reported about 7.1% among Iranian population (1). These patients generally are more likely have irregular menstruation, hyperandrogenism, and defects in ovulation and polycystic ovaries (2). Although several diagnostic criteria have been reported for PCOS, Rotterdam is known to be the most important prognostic factor in such patients. Based on this criteria, the diagnosis was made with the presence of at least two of the following features: irregular or failed ovulation, clinical symptoms and laboratory findings of hyperandrogenism or the presence of polycystic ovarian morphology confirmed by ultrasonography (3).

PCOS women have an increased risk of developing metabolic syndrome (MBS), as they show up two times higher prevalence of this disorder compared to normal population (4). Insulin resistance, obesity, impaired glucose metabolism, dyslipidemia and hypertension are the main characteristics of MBS. It has also been shown that the insulin resistance has a pivotal role in the pathogenesis of PCOS and MBS (5), so 50–80% of PCOS patients suffer from insulin resistance syndrome (6). Hypoglycemia is one of the most common complications of insulin resistance, which in turn can lead to hyperinsulinemia, enhanced LH hormone action and elevated androgen levels, in the following. Thus, a sequence of point events should be classified as underlying causes of PCOS (7). More recently, it has been demonstrated that vitamin D deficiency is associated with insulin resistance and particularly affects the amount of insulin, released from pancreatic beta cells (8). However, the prevalence of vitamin D deficiency is a common finding in PCOS women, as approximately 67–85% have a serum 25(OH) D concentration of less than 20 ng/ml. Ovulation problems, menstrual irregularities and infertility are subsequent challenges encountered during the periods of low vitamin D status (9).

Obesity is one of the fastest growing health concerns worldwide. This medical condition, with an impact on hormonal activity and ovulation, represents one of the most important risk factors and also a predictor for progression of PCOS (10). Results of several studies have brought important evidence linking obesity with vitamin D deficiency (11). Therefore, current findings have raised the question of whether obesity and vitamin D deficiency might have different levels of efficacy in the development of PCOS among susceptible subjects. The purpose of this study was to determine the effect of serum 25(OH) D concentration on metabolic parameters and insulin resistance in non-obese patients with PCOS.

2. Materials and methods

This experimental study was performed on 88 non-obese women with PCOS, ranging in age from 19 to 40 years. We used Rotterdam criteria for the diagnosis of PCOS, including menstrual problems such as oligomenorrhea (fewer than 6 periods per year) or amenorrhea (the absence of menstrual periods for more than 6 months), clinical symptoms and laboratory findings of hyperandrogenism in addition to sonographic evaluation of PCOS (the presence of > 12 follicles measuring 2–9 mm in diameter or an ovarian volume more than 10 cm³) (3). Patients with nonclassic congenital adrenal hyperplasia, thyroid disorders, androgen-secreting tumors, Cushing’s syndrome and hyperprolactinemia, as well as cases with a history of drug use within the past three months, affecting calcium and carbohydrate metabolisms and endocrine parameters, are excluded from the study. Our research was approved by the Ethics Committee of Ahvaz Jundishapur University of Medical Sciences and all the patients signed an informed consent before entering the study. Body mass index (BMI) was calculated from adjusted weight and height, with unit kg/m². The population selected was then divided into two groups: normal weight (BMI ≤ 25 kg/m²) and overweight (BMI > 25 kg/m²).

Fasting venous blood samples were collected during the follicular phase of the menstrual cycle (between the 3rd and 5th day of menses) and serum was separated, stored at −20 °C until they were transported for analysis of biochemical factors. Serum concentrations of 25(OH) D by radioimmunoassay (DRG, Marburg, Germany), insulin by IRMA assay (BioSource Europe S.A. Nivelles, Belgium), as well as high-sensitivity C-reactive protein (hs-CRP) by Architect c16000 (Abbott) system were measured for all the patients with PCOS. Serum glucose, triglycerides (TG), total cholesterol (TC), high-density lipoprotein (HDL) and low-density lipoprotein (LDL) were also obtained by standard methods (CimnaGen, Tehran, Iran). The insulin resistance index was calculated by homeostasis model assessment (HOMA) as follows (12):

$$\text{HOMA} = \frac{\text{Fasting Insulin (mIU/mL)} \times \text{Fasting Glucose (mg/L)}}{405}$$

Statistical analyses were performed using SPSS 16.0. The difference in mean levels of metabolic factors between normal weight and overweight PCOS subjects was determined by an independent t-test. The Pearson Correlation test was then done to evaluate the relationship between BMI index and metabolic dysfunction. Results were considered significant with a P-value of less than 0.05.

3. Results

Patients’ age ranged from 19 to 40, with a mean age of 27 years. Of these, 36 (40.9%) had a normal BMI of...
Table 1  Comparison of metabolic parameters, serum levels of vitamin D and CRP in PCOS women with normal weight and obesity.

<table>
<thead>
<tr>
<th>Variables</th>
<th>Normal</th>
<th>Over weight</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age</td>
<td>26.67 ± 6.63</td>
<td>27.42 ± 6.027</td>
<td>0.457</td>
</tr>
<tr>
<td>BMI</td>
<td>25 (19–39)</td>
<td>27 (19–39)</td>
<td></td>
</tr>
<tr>
<td>BMI</td>
<td>21.75 ± 2.03</td>
<td>27.59 ± 1.72</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>HOMA-IR</td>
<td>4.46 ± 5.84</td>
<td>5.10 ± 5.39</td>
<td>0.272</td>
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<tr>
<td>Insulin</td>
<td>12.08 (3.3–98)</td>
<td>14.33 (1.41–71.12)</td>
<td>0.62</td>
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<tr>
<td>HOMA-B</td>
<td>2.50 ± 2.13</td>
<td>2.27 ± 1.89</td>
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<tr>
<td>(Total) Chol</td>
<td>1.85 (0.02–10.90)</td>
<td>1.62 (0.01–8.67)</td>
<td>0.507</td>
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<tr>
<td>(HDL) Chol</td>
<td>47.44 ± 9.39</td>
<td>44.79 ± 10.61</td>
<td>0.16</td>
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<tr>
<td>(VLDL) Chol</td>
<td>23.78 ± 10.52</td>
<td>28.71 ± 21.02</td>
<td>0.473</td>
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<tr>
<td>Vitamin D</td>
<td>14.95 ± 10.89</td>
<td>15.44 ± 9.78</td>
<td>0.492</td>
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<tr>
<td>(LDL) Chol</td>
<td>94.39 ± 23.63</td>
<td>88 ± 24.88</td>
<td>0.232</td>
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<tr>
<td>TG</td>
<td>118.47 ± 53.19</td>
<td>149.37 ± 137.99</td>
<td>0.44</td>
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<tr>
<td>FBS</td>
<td>2.53 (0.77-32.67)</td>
<td>3.28 (0.27-23.88)</td>
<td>0.272</td>
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</table>


Table 2  Comparison of the mean serum vitamin D levels in patients with insulin resistance and non-resistance poly cystic ovary syndrome.

<table>
<thead>
<tr>
<th>Vitamin D</th>
<th>HOMA-IR</th>
<th>Mean ± SD</th>
<th>Medline range</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>&lt;2.5 without insulin resistance</td>
<td>15.33 ± 11.07</td>
<td>12.85 (4.85-58.9)</td>
<td>0.806</td>
<td></td>
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<tr>
<td>&gt;2.5 with insulin resistance</td>
<td>15.18 ± 9.64</td>
<td>12.65 (2-51.6)</td>
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Abbreviations: a SD: Standard deviation.

Vitamin D levels were also similar in PCOS patients both with and without insulin resistance (Table 2). Seventy-four patients (84.1%) were vitamin D deficient (serum 25(OH) D level < 20 ng/mL). Mean serum 25 (OH) D levels were 14.9 and 15.4 ng/mL in normal weight and overweight cases, with no significant differences between groups, but were positively correlated with the age of patients and their serum concentration of HDL (Table 3). However, other metabolic factors have no significant association with the levels of this fat-soluble vitamin. Although 25(OH) D levels were positively correlated with insulin resistance and negatively associated with beta cell function (HOMA-B), these values were not statistically different.

Table 3  Relationship between serum vitamin D levels with metabolic parameters in women with PCOS.

<table>
<thead>
<tr>
<th>Vitamin D</th>
<th>Age</th>
<th>HOMA-IR</th>
<th>HOMA-B</th>
<th>Insulin</th>
<th>FBS</th>
<th>TG</th>
<th>Chol</th>
<th>HDL</th>
<th>VLDL</th>
<th>LDL</th>
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<tbody>
<tr>
<td>Age</td>
<td>0.233</td>
<td>0.036</td>
<td>0.108</td>
<td>0.003</td>
<td>0.123</td>
<td>-0.017</td>
<td>0.113</td>
<td>0.212</td>
<td>-0.015</td>
<td>0.54</td>
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<tr>
<td>HOMA-IR</td>
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<td>Insulin</td>
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4. Discussion

Vitamin D deficiency is a global health issue. Inadequate exposure to sunlight is one of the main causes of this deficiency, since food dietary contains natural sources of vitamin D supplement (13). In addition, older individuals with increased fat deposits are also prone to develop vitamin D deficiency. Hypovitaminosis D may be associated with a number of mental and physical disorders such as MBS, type 2 diabetes, PCOS and cancer (14).

Our results have indicated that 84% of PCOS patients are suffering from hypovitaminosis D, while there is a high prevalence (67–90%) of vitamin D deficiency among Iranian population (15,16). These findings suggest almost equal correlation between the measured parameters in normal weight and overweight women.

20–24.99 kg/m², and another 52 (59.1%) had a BMI of 25–30 kg/m². Seventy-five (85.2%) and seven (8%) patients were classified as having normal and impaired fasting blood glucose concentrations respectively, and 6 (6.8%) patients were suspected to have diabetes. Serum levels of metabolic factors, 25(OH) D and CRP are shown in Table 1. There was no significant correlation between the measured parameters in normal weight and overweight women with PCOS.

Insulin resistance (HOMA index > 2.5) was observed in 52 cases (59.1%) of PCOS, in which 36% had a BMI index less than 25 and the rest were in the range of 25–30. There were no significant differences in insulin resistance indices between normal weight and overweight women (P = 0.28), and serum
different age-group classification is the main reason for this dispute, since skin atrophy due to aging resulted in decreased calciferol syntheses in over one fifth of young people. Further decrease in fat-soluble vitamin intestinal absorption, as well as lower physical activity in older people can lead to inadequate vitamin D intake in such cases (24).

Interestingly, serum levels of 25(OH) D were positively associated with only HDL concentration, among investigated metabolic factors. This result is consistent with similar findings of other studies, so that vitamin D-deficient women are more susceptible to have lower HDL concentration (25–28). Although the exact mechanisms are not known, it has been suggested that vitamin D is essential for maintenance of sufficient Apolipoprotein A-1 (ApoA1) levels, the major component of HDL, throughout the body. Thus, patients with higher levels of 25(OH) D have almost elevated concentration of ApoA1 and HDL in the following survey (29).

Insulin resistance was observed in 59.1% of our patients and only 8% of cases actually had impaired fasting glucose. Evidence from multiple studies has shown that almost 70% of patients diagnosed with PCOS have resistance to insulin (30). On the opposite of our study, Ketel et al. found a significant difference in insulin resistance between normal weight and obese PCOS women, but nearly the same ratio between healthy and normal weight PCOS subjects (31). We also found that serum 25(OH) D levels were not significantly associated with insulin resistance. On the other hand, conflicting results have been reported for such variables, as 25(OH) D deficiency was positively associated with insulin resistance among PCOS women (32–34).

Obesity is a common finding in patients with PCOS and 61–76% of women with this syndrome are reported to have obesity (35). It has also been shown that obesity rates are higher in PCOS patients than in healthy individuals (36). It is known that obesity is the main risk factor for developing insulin resistance, type 2 diabetes, MBS and cardiovascular diseases. One explanation may be that, inflammatory mediators released from adipose tissue immune cells contribute to metabolic disorders in such cases (37). However, this fat-soluble vitamin is stored in the body’s adipose tissue in terms of increased total fat mass, which explains the reason of 35% increase in vitamin D deficiency in obese patients compared to normal weight subjects (11). While several mechanisms have been proposed for the impact of vitamin D supplementation on glucose metabolism, suppression of pro-inflammatory cytokines and increased insulin sensitivity are considered the most important factors for this event (38). It therefore appears that obesity is the main cause of both vitamin D deficiency and insulin resistance in PCOS patients.

In conclusion, our study found an association between the high prevalence of obesity and metabolic dysfunction, as well as insulin resistance in women with PCOS, and vitamin D deficiency is also occurred secondary to obesity in such patients. Thus, vitamin D deficiency cannot be considered as an independent factor through the development of metabolic disorders in PCOS patients.

Evaluation of the effect of vitamin D deficiency on metabolic parameters and insulin resistance in normal weight and overweight women with PCOS was the main purpose of our study; however, future investigations with larger numbers of samples and control groups are also needed to compare the PCOS patients with healthy subjects.

Conflict of interest
We have no conflict of interest to declare.

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Vitamin D and metabolic parameters in PCOS


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