Association of hypovitaminosis D with metabolic disturbances in polycystic ovary syndrome

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Abstract

Objectives: Women with polycystic ovary syndrome (PCOS) frequently suffer from metabolic disturbances, in particular from insulin resistance. Accumulating evidence suggests that vitamin D deficiency may contribute to the development of metabolic syndrome (MS). Hence, the aim of our study was to investigate the association of 25(OH)D levels and the components of the MS in PCOS women.

Methods: 25(OH)D levels were measured by means of ELISA in 206 women affected by PCOS. Metabolic, endocrine, and anthropometric measurements and oral glucose tolerance tests were performed.

Results: The prevalence of insufficient 25(OH)D levels (< 30 ng/ml) was 72.8% in women with PCOS. PCOS women with MS had lower 25(OH)D levels than PCOS women without these features (17.3 vs 25.8 ng/ml respectively; \( P < 0.05 \)). In multivariate regression analysis including 25(OH)D, season, body mass index (BMI), and age, 25(OH)D and BMI were independent predictors of homeostatic model assessment-insulin resistance (HOMA-IR) and quantitative insulin sensitivity check index (QUICKI; \( P < 0.05 \) for all). In binary logistic regression analyses, 25(OH)D (odds ratio, OR 0.86, \( P < 0.019 \)) and BMI (OR 1.28, \( P < 0.001 \)) were independent predictors of MS in PCOS women. We found significantly negative correlations of 25(OH)D levels with BMI, waist circumference, waist-to-hip ratio, systolic and diastolic blood pressure, fasting and stimulated glucose, area under the glucose response curve, fasting insulin, HOMA-IR, HOMA-\( \beta \), triglycerides, and quotient total cholesterol/high-density lipoprotein (HDL) and positive correlations of 25(OH)D levels with QUICKI and HDL (\( P < 0.05 \) for all).

Conclusion: We demonstrate that low 25(OH)D levels are associated with features of MS in PCOS women. Large intervention trials are warranted to evaluate the effect of vitamin D supplementation on metabolic disturbances in PCOS women.

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Introduction

Polycystic ovary syndrome (PCOS) is the most common female endocrine disorder with a prevalence of \( \sim 5\% \) to 10% in women of reproductive age (1–3). PCOS is characterized by increased ovarian and adrenal androgen secretion, hyperandrogenic symptoms such as hirsutism, acne and/or alopecia, menstrual irregularity, and polycystic ovaries. In addition, insulin resistance and hyperinsulinemia are common features in PCOS women (4), who are therefore at an increased risk of type 2 diabetes (5).

Accumulating evidence suggests that vitamin D deficiency might be a causal factor in the pathogenesis of metabolic syndrome (MS) in PCOS (6). This notion is supported by the fact that the vitamin D receptor gene regulates about 3% of the human genome, including genes that are crucial for glucose and lipid metabolism and blood pressure regulation (7–9). Clinical studies have largely but not consistently shown that type 2 diabetes and insulin resistance are associated with poor vitamin D status (10, 11). In addition, some studies suggest that vitamin D insufficiency is also associated with dyslipidemia (6) as well as arterial hypertension, which might be explained by the inhibition of the renin gene expression by vitamin D (9). Obesity is also linked with low 25(OH)D levels in PCOS cohorts (6, 12) and in large study cohorts including various groups of obese and normal weight women and men (13, 14), which might be a consequence of 25(OH)D deposition in the adipose tissue.

So far, the role of 25(OH)D in the development of MS in PCOS is largely unknown. Therefore, the aim of our study was to extend the currently rare knowledge about the association of vitamin D and MS in PCOS women.
Methods

Subjects

We evaluated 206 women with PCOS aged 16–41 years, who were routinely referred to our outpatient clinic. The diagnosis was based on the Rotterdam criteria (15). Two out of three of the following are required to confirm the diagnosis: oligo- and/or anovulation; clinical and/or biochemical signs of hyperandrogenism; and polycystic ovaries (by ultrasound). Disorders with a similar clinical presentation, such as congenital adrenal hyperplasia, Cushing’s syndrome, and androgen-secreting tumors, must be excluded. Oligo- and/or anovulation were defined by the presence of oligomenorrhea or amenorrhea. Hyperandrogenism was defined by the clinical presence of hirsutism (Ferriman–Gallwey score ≥ 6), acne or alopecia, and/or elevated androgen levels. Polycystic ovarian morphology was examined by ultrasound. Polycystic ovaries were defined as the presence of 12 or more follicles in each ovary measuring 2–9 mm in diameter and/or increased ovarian volume (>10 ml) calculated using the formula 0.5×length×width×thickness (15). Hyperprolactinemia, Cushing’s syndrome, congenital adrenal hyperplasia, and androgen-secreting tumors were excluded by specific laboratory analysis (cortisol, ACTH, 17αOH-progesterone, and DHEAS).

MS was defined by the National Cholesterol Education Program (NCEP) and the Adult Treatment Panel III (ATP III) in subjects presenting at least three of the following criteria: waist circumference > 88 cm; high-density lipoprotein (HDL) cholesterol < 50 mg/dl; triglyceride level > 150 mg/dl; raised blood pressure (systolic > 130 mmHg and diastolic > 85 mmHg); and raised fasting glucose (>110 mg/dl) or impaired glucose tolerance (IGT) during oral glucose tolerance test (OGTT) (16). The study participants did not take any medication known to affect endocrine parameters, carbohydrate metabolism, or serum lipid profile for at least 3 months before entering the study.

The study protocol was approved by the local ethics committee, and written informed consent was obtained from each patient.

Procedures

Standard anthropometric data (height, weight, and waist and hip circumference) were obtained from each subject. Blood pressure was measured after PCOS women had been seated for at least 5 min. The body mass index (BMI) was calculated as the weight in kilograms divided by the square of height in meters. Waist circumference was measured in a standing position midway between the lower costal margin and the iliac crest. Hip circumference was measured in a standing position at the maximum circumference over the buttocks. Hirsutism was quantified with the modified Ferriman–Gallwey score. Moreover, basal blood samples for hormonal (25(OH)D, parathyroid hormone (PTH), total testosterone, free testosterone, sex hormone-binding globulin (SHBG), androstenedione, DHEAS, free triiodothyronine, free thyroxine, TSH, 17αOH-progesterone, and cortisol) and metabolic (glucose, insulin, C-peptide, total cholesterol, HDL cholesterol, low-density lipoprotein cholesterol, and triglycerides) determinations were collected at 0800–0900 h after overnight fast. All participants underwent a fasting 75 g OGTT. Blood samples were drawn after 60 and 120 min for glucose, insulin, and C-peptide determination. Insulin resistance was estimated using the homeostatic model assessment-insulin resistance (HOMA-IR). HOMA-IR was calculated as the product of the fasting plasma insulin value (µU/ml) and the fasting plasma glucose value (mg/dl), divided by 405 (17). Quantitative insulin sensitivity check index (QUICKI) was used to estimate insulin sensitivity. QUICKI was calculated as 1/log fasting insulin (µU/ml)+log fasting glucose (mg/dl). To assess β-cell function, HOMA-β was calculated as (20×fasting insulin (µU/ml))/(fasting glucose (mmol/l)–3.5). Hyperinsulinemia was assessed by calculating the area under the insulin response curve (AUICs). The free androgen index (FAI) was calculated as testosterone (nmol/l)/SHBG (nmol/l)×100.

Biochemical analysis

25(OH)D (normal range 30–60 ng/ml) was measured using a commercially available ELISA (IDS, Boldon, UK) with intra- and inter-assay coefficients of variation (CV) of 5.6 and 6.4% respectively. Insulin (2.0–25.0 µU/ml) and C-peptide (0.5–3.2 ng/ml) were measured by ELISA (DRG, Marburg, Germany) with intra- and inter-assay CV of 4.0 and 2.6, and 5.1 and 8.4% respectively. Fasting glucose (≤115 mg/dl), triglycerides (≤150 mg/dl), total cholesterol (≤200 mg/dl), HDL cholesterol (>40 mg/dl), and C-reactive protein (CRP) (≤8 mg/l) were determined using Modular Analytics SWA (Roche). Free testosterone (0.29–3.18 pg/ml) was determined using a RIA (DSL, Webster, TX, USA). SHBG (19–117 nmol/l), PTH (15–65 pg/ml; Roche), ACTH (10–51 pg/ml), cortisol (43.0–220.0 ng/ml), human growth hormone (HGH) (0.0–7.0 ng/ml; Siemens DPC Bühlmann, Salzburg, Austria), and insulin-like growth factor 1 (100–400 ng/ml), prolactin (2.8–29.2), total testosterone (0.14–0.77 ng/ml), and TSH (0.1–4.0 µU/ml; Bayer) were measured by luminescence immunoassay.

Statistical analysis

For the purpose of this study and according to widely used cut-offs, subjects were divided into groups: vitamin D sufficiency (25(OH)D ≥ 30 ng/ml); hypovitaminosis D (25(OH)D < 30 ng/ml). Furthermore, we defined severe vitamin D deficiency (25(OH)D < 10 ng/ml), moderate
vitamin D deficiency (25(OH)D 10–19.9 ng/ml), and vitamin D insufficiency (25(OH)D 20–29.9 ng/ml) (7, 18). Data are presented as means ± s.d., unless otherwise stated. Kolmogorov–Smirnov test was used to examine for normal distribution, and variables following a skewed distribution were logarithmically transformed before being used in correlation or regression analyses. Pearson correlations and partial correlation analyses were used to determine relationships between variables. Depending on the distribution of data, the Student’s t-test for independent samples and the nonparametric Mann–Whitney U-test for independent samples were applied to test for differences between groups. CRP values within the normal range (<8 mg/l) were included in the analyses. To study seasonal variation, we subdivided the year into 4-month measurement periods: February–May (season 1); June–September (season 2); October–January (season 3) to address the seasonal changes in availability of sunlight (19). Multiple linear regression analyses were calculated with HOMA-IR and QUICKI as dependent variables and 25(OH)D, season, BMI, and age as independent variables. Binary logistic regression analyses were performed to examine the associations of MS (dependent variable) with 25(OH)D, season, BMI, and age (independent variables). Statistical analyses were performed by SPSS version 16.0 (SPSS Inc., Chicago, IL, USA). A P value of <0.05 was considered statistically significant.

## Results

### Findings for entire cohort

Anthropometric and biochemical characteristics of PCOS women stratified by 25(OH)D levels are shown in Table 1. Hypovitaminosis D was present in 150 out of 206 PCOS women (72.8%); 6 out of 206 PCOS women (<30 ng/ml); 74 out of 206 PCOS women (35.9%) had 25(OH)D levels between 10 and 20 ng/ml. Multiple linear regression analyses were performed by SPSS version 16.0 (SPSS Inc., Chicago, IL, USA). A P value of <0.05 was considered statistically significant.

### Table 1 Clinical and biochemical characteristics of polycystic ovary syndrome (PCOS) subjects based on 25(OH)D status.

<table>
<thead>
<tr>
<th></th>
<th>All PCOS (n=206)</th>
<th>Hypovitaminosis D (&lt;30 ng/ml, n=150)</th>
<th>Vitamin D sufficiency (≥30 ng/ml, n=56)</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Clinical characteristics</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Age (years)</td>
<td>29 ± 7</td>
<td>29 ± 6</td>
<td>27 ± 7</td>
<td>0.027</td>
</tr>
<tr>
<td>Weight (kg)</td>
<td>72.3 ± 19.7</td>
<td>75.6 ± 21.0</td>
<td>63.7 ± 12.4</td>
<td>0.001</td>
</tr>
<tr>
<td>Height (cm)</td>
<td>166.1 ± 6.4</td>
<td>166.0 ± 6.4</td>
<td>168.2 ± 6.5</td>
<td>0.980</td>
</tr>
<tr>
<td>BMI (kg/m²)</td>
<td>26.2 ± 6.9</td>
<td>27.4 ± 7.4</td>
<td>23.0 ± 4.1</td>
<td>0.001</td>
</tr>
<tr>
<td>Waist circumference (cm)</td>
<td>90 ± 19</td>
<td>94 ± 19</td>
<td>79 ± 10</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Hip circumference (cm)</td>
<td>106 ± 13</td>
<td>108 ± 13</td>
<td>98 ± 10</td>
<td>0.001</td>
</tr>
<tr>
<td>WHR</td>
<td>0.85 ± 0.10</td>
<td>0.86 ± 0.09</td>
<td>0.81 ± 0.10</td>
<td>0.005</td>
</tr>
<tr>
<td>Systolic blood pressure (mmHg)</td>
<td>113 ± 15</td>
<td>114 ± 16</td>
<td>108 ± 12</td>
<td>0.026</td>
</tr>
<tr>
<td>Diastolic blood pressure (mmHg)</td>
<td>75 ± 11</td>
<td>76 ± 11</td>
<td>73 ± 11</td>
<td>0.115</td>
</tr>
<tr>
<td>Metabolic biochemical characteristics</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Fasting glucose (mg/dl)</td>
<td>83 ± 9</td>
<td>83 ± 10</td>
<td>82 ± 9</td>
<td>0.237</td>
</tr>
<tr>
<td>1-h glucose (mg/dl)</td>
<td>115 ± 40</td>
<td>120 ± 43</td>
<td>103 ± 28</td>
<td>0.003</td>
</tr>
<tr>
<td>2-h glucose (mg/dl)</td>
<td>98 ± 30</td>
<td>99 ± 32</td>
<td>93 ± 22</td>
<td>0.259</td>
</tr>
<tr>
<td>AUCgluc</td>
<td>102.5 ± 27.8</td>
<td>105.5 ± 29.9</td>
<td>94.6 ± 18.9</td>
<td>0.046</td>
</tr>
<tr>
<td>HbA1c (%)</td>
<td>5.2 ± 0.3</td>
<td>5.2 ± 0.3</td>
<td>5.1 ± 0.3</td>
<td>0.065</td>
</tr>
<tr>
<td>HOMA-IR</td>
<td>1.72 ± 1.93</td>
<td>1.96 ± 2.16</td>
<td>1.11 ± 0.92</td>
<td>0.002</td>
</tr>
<tr>
<td>HOMA-β</td>
<td>160.2 ± 122.7</td>
<td>173.3 ± 131.1</td>
<td>126.7 ± 90.9</td>
<td>0.014</td>
</tr>
<tr>
<td>QUICKI</td>
<td>0.39 ± 0.09</td>
<td>0.38 ± 0.10</td>
<td>0.40 ± 0.05</td>
<td>0.002</td>
</tr>
<tr>
<td>Fasting insulin (μU/ml)</td>
<td>8.0 ± 7.6</td>
<td>9.0 ± 8.4</td>
<td>5.4 ± 4.0</td>
<td>0.002</td>
</tr>
<tr>
<td>1-h insulin (μU/ml)</td>
<td>58.0 ± 44.1</td>
<td>61.7 ± 45.0</td>
<td>48.9 ± 40.6</td>
<td>0.049</td>
</tr>
<tr>
<td>2-h insulin (μU/ml)</td>
<td>48.1 ± 43.8</td>
<td>52.5 ± 49.1</td>
<td>37.4 ± 24.6</td>
<td>0.267</td>
</tr>
<tr>
<td>AUCins</td>
<td>42.8 ± 31.5</td>
<td>45.9 ± 33.3</td>
<td>35.2 ± 25.3</td>
<td>0.074</td>
</tr>
<tr>
<td>Total cholesterol (mg/dl)</td>
<td>177.5 ± 35.4</td>
<td>174.2 ± 34.9</td>
<td>185.9 ± 35.8</td>
<td>0.088</td>
</tr>
<tr>
<td>Triglycerides (mg/dl)</td>
<td>89.6 ± 44.6</td>
<td>94.8 ± 48.8</td>
<td>76.5 ± 27.9</td>
<td>0.042</td>
</tr>
<tr>
<td>HDL (mg/dl)</td>
<td>64.1 ± 16.2</td>
<td>61.7 ± 15.7</td>
<td>70.0 ± 16.3</td>
<td>0.002</td>
</tr>
<tr>
<td>QChol/HDL</td>
<td>2.9 ± 0.8</td>
<td>3.0 ± 0.8</td>
<td>2.8 ± 0.7</td>
<td>0.106</td>
</tr>
<tr>
<td>LDL (mg/dl)</td>
<td>99.5 ± 29.2</td>
<td>99 ± 29</td>
<td>103.9 ± 31.8</td>
<td>0.221</td>
</tr>
<tr>
<td>CRP (mg/l)</td>
<td>2.3 ± 2.0</td>
<td>2.5 ± 2.1</td>
<td>1.6 ± 1.3</td>
<td>0.030</td>
</tr>
</tbody>
</table>

Comparisons between PCOS women with hypovitaminosis D and sufficient 25(OH)D levels were performed by Student’s t-test or Mann–Whitney U test as appropriate.
and 19.9 ng/ml; 70 women (34.0%) presented with 25(OH)D levels between 20 and 29.9 ng/ml; and 56 women (27.2%) showed sufficient 25(OH)D levels ≥ 30 ng/ml.

25(OH)D levels were determined between October and January (season 3) in the majority of PCOS women (n = 93, 45.1%). Blood samples of 76 women (36.9%) were collected between February and May (season 1), and 37 women (18%) were examined between June and September (season 2). 25(OH)D levels were significantly higher in season 2 when compared with seasons 1 and 3 (P < 0.05 for all).

25(OH)D levels correlation was significantly negative with BMI (Fig. 1a), weight, waist circumference, hip circumference, waist-to-hip ratio (WHR), systolic and diastolic blood pressure, fasting and stimulated glucose, area under the glucose response curve (AUCgluc: Fig. 1b), fasting and stimulated insulin, AUCins, PTH, triglycerides, quotient total cholesterol (QChol)/HDL, and CRP (Table 2). We found a significantly positive correlation between 25(OH)D levels and HDL cholesterol (Table 2). These latter results did not materially change when controlling for season and age in partial correlation analyses. However, the associations of 25(OH)D with anthropometric parameters, glucose, insulin, and triglycerides were no longer significant after additional adjustment for BMI. The positive correlation of 25(OH)D with HDL remained significant after adjustment for BMI (r = 0.303, P = 0.006).

In addition, we found a significant correlation between 25(OH)D and HOMA-IR (Table 2), QUICKI (Fig. 1c), as well as HOMA-β (Fig. 1d). To explore the association of 25(OH)D and HOMA-IR, we performed a multivariate regression analysis including HOMA-IR as dependent variable and BMI, age, 25(OH)D, and season as explanatory variables. In this analysis, 25(OH)D (P = 0.036) was a significant and independent predictor for HOMA-IR, along with BMI (P < 0.001). Furthermore, we performed a multivariate regression analysis including QUICKI as dependent variable and BMI, age, 25(OH)D, and season as explanatory variables. 25(OH)D (P = 0.047) and BMI (P < 0.001) were significant predictors of QUICKI, along with the other covariates explaining 18% of variation in QUICKI.

Thirty PCOS women (15%) presented with IGT. Women with IGT had significantly lower levels of 25(OH)D than women with normal glucose tolerance (20.2 and 25.6 ng/ml respectively; P = 0.026). In PCOS women with vitamin D deficiency (< 20 ng/ml), 16 women (20.0%) had IGT; in PCOS women with vitamin D insufficiency (20–29.9 ng/ml), 12 women (17.1%) showed IGT; and in PCOS women with sufficient 25(OH)D levels (> 30 ng/ml), 2 women (3.6%) had IGT.

To address the heterogeneity of PCOS, subgroup analyses of PCOS women with and without a family history of type 2 diabetes were performed (20, 21). Data on family history of type 2 diabetes were available in 158 PCOS patients. Family history of type 2 diabetes...
was present in 47 (29.7%) out of 158 PCOS patients, and these PCOS women had significantly lower 25(OH)D levels than PCOS women without the MS (17.3 vs 25.8 ng/ml respectively; P < 0.001).

In logistic regression analyses, MS was associated with BMI (odds ratio (OR) 1.28, 95% confidence interval (CI) (1.15–1.42), P < 0.001) and 25(OH)D (OR 0.86, 95% CI (0.75–0.98), P = 0.019).

Findings stratified by vitamin D sufficiency and hypovitaminosis D

Clinical and biochemical characteristics of PCOS women with vitamin D sufficiency (≥30 ng/ml) and hypovitaminosis D (<30 ng/ml) are shown in Table 1. PCOS women with hypovitaminosis D had significantly higher age, weight, BMI, waist and hip circumference, WHR, and systolic blood pressure than women with vitamin D sufficiency (P < 0.05 for all). Furthermore, the hypovitaminosis D group had significantly higher levels of 1 h glucose, AUCh-gluc, HOMA-IR, HOMA-β, fasting insulin, 1 h insulin, triglycerides, and CRP, and significantly lower levels of QUICKI, HDL, and SHBG than the vitamin D sufficient group (P < 0.05 for all).

Furthermore, we performed subgroup analyses of lean (BMI ≤ 25, n = 116, 56.3%) and obese (BMI > 25, n = 90, 43.7%) PCOS women. When lean PCOS women were analyzed separately, we observed no significant differences for all parameters included in Table 1 between PCOS women with hypovitaminosis D and vitamin D sufficiency (data not shown). We found a significant positive correlation of 25(OH)D with HDL in lean PCOS women (P < 0.05, data not shown), whereas all other correlations were not statistically significant. In the subgroup of obese PCOS women, we found significantly increased HbA1c levels and waist circumference in the hypovitaminosis D group when compared with the vitamin D sufficiency group (P < 0.05 for all, data not shown), whereas all other parameters were not significantly different between groups. In obese PCOS women, there were significantly negative correlations when controlling for BMI (r = −0.367; P = 0.002). Furthermore, hirsute PCOS women had significantly lower 25(OH)D levels (21.4 ng/ml) than PCOS women without hirsutism (26.8 ng/ml; P = 0.001).

Levels of PTH were significantly positive correlated with AUCh-gluc (r = 0.235; P = 0.036) and diastolic blood pressure (r = 0.278; P = 0.006).

Metabolic syndrome

Presence of MS was analyzed in a subgroup of 174 out of 206 PCOS women (85.4%). MS was evident in 25 women (12.2%). Out of 25 PCOS women, 18 with MS (72%) presented with deficient 25(OH)D levels, 7 women with MS (28%) had insufficient 25(OH)D levels, and none had a sufficient 25(OH)D level (>30 ng/ml). PCOS women with the MS had significantly lower 25(OH)D levels than PCOS women without the MS (17.3 vs 25.8 ng/ml respectively; P < 0.001).

In logistic regression analyses, MS was associated with BMI (odds ratio (OR) 1.28, 95% confidence interval (CI) (1.15–1.42), P < 0.001) and 25(OH)D (OR 0.86, 95% CI (0.75–0.98), P = 0.019).
of 25(OH)D with 1-h glucose, 2-h glucose, AUCgluc, 
HbA1c, HOMA-β, fasting insulin, 1-h insulin, and 
AUCins, and significantly positive correlations of 
25(OH)D with QUICKI and HDL (P < 0.05 for all, data 
not shown).

Discussion

Our results indicate that low 25(OH)D levels are 
significantly associated with components of the MS 
and insulin resistance in women with PCOS. 25(OH)D 
was an independent predictor of insulin resistance and 
insulin sensitivity in a multivariate regression analysis.

Low 25(OH)D levels have been linked to an increased 
risk for cancer (22), autoimmune diseases, diabetes, and 
cardiovascular diseases (7, 22, 23) indicating the 
importance of sufficient 25(OH)D levels. Although 
there is no consensus on optimal levels of 25(OH)D, 
a level of 30 ng/ml can be considered to indicate 
sufficient vitamin D status (7). In our study, 72.8% of 
PCOS women showed 25(OH)D values below this 
recommended level.

Our data demonstrate a significant association of low 
25(OH)D levels with increased levels of fasting and 
stimulated glucose, AUCgluc, HOMA-IR, and fasting 
and stimulated insulin. Accordingly, Hahn et al. 
reported an association of low 25(OH)D levels with 
insulin resistance in 120 PCOS women (6). Apart from 
these cross-sectional findings, there is one small 
prospective intervention study with vitamin D supple-
mentation, which demonstrates beneficial effects of 
vitamin D on insulin secretion and serum lipids in PCOS 
women (24). In nonPCOS cohorts including subjects 
with various BMI, vitamin D concentration was 
inversely related to the prevalence of diabetes (25), 
plasma concentrations of glucose (26), insulin resistance 
(10, 26), and the MS (8, 10). Besides, the risk of 
future hyperglycemia and insulin resistance was 
associated with hypovitaminosis D (27).

The mechanisms underlying the association of low 
25(OH)D levels and insulin resistance are not fully 
understood. First, vitamin D may have a beneficial effect 
on insulin action by stimulating the expression of 
insulin receptor and thereby enhancing insulin respon-
siveness for glucose transport (8). Secondly, vitamin D 
regulates extracellular and intracellular calcium, which 
is essential for insulin-mediated intracellular processes 
in insulin-responsive tissues such as skeletal muscle 
and adipose tissue (8). Finally, as vitamin D has a 
modulating effect on the immune system (28), hypo-

vitaminosis D might induce a higher inflammatory 
response, which is associated with insulin resistance 
(29). This hypothesis is supported by the results of our 
study indicating an association of low 25(OH)D with 
increased CRP levels.

In turn, an additional mechanism might be seen in 
impaired β-cell function in PCOS women. This is 
underlined by our finding of a negative association of 
25(OH)D levels and HOMA-β and the inverse associ-

ation of stimulated glucose levels and AUCgluc with 
25(OH)D.

A new finding in our study was the association of 
increased triglycerides and QChol/HDL with low 
25(OH)D levels. Moreover, we found low 25(OH)D levels 
associated with low levels of HDL, confirming previous 
results in PCOS women (6), but contrasting the lack of 
association in a recent study in a cohort of healthy 
young women (30). In a pilot study, Kotsa et al. showed 
an improvement of HDL and triglycerides after treat-
ment with vitamin D in a small cohort of PCOS women 
(24). Since dyslipidemia should be considered as an 
additional therapeutic target in PCOS (31), vitamin D 
might be useful in the complex treatment of PCOS 
women.

These parameters, i.e. low HDL and elevated 
triglycerides, are central features of the MS. For the 
first time, we observed an association of low 25(OH)D 
levels with the MS independently from BMI, age, and 
seasonal variation of 25(OH)D in PCOS women.

Nevertheless, our data suggest a strong relationship 
of 25(OH)D and BMI in PCOS women, which is in 
agreement with previous studies (6, 12). So far, it is 
not clear whether vitamin D insufficiency results from 
obesity and/or whether obesity is a consequence of 
vitamin D insufficiency. On the one hand, obesity may 
contribute to low circulating vitamin D levels by 
trapping vitamin D in fat tissues. Wortsman et al. 
demonstrated that the increase in 25(OH)D levels 24 h 
after whole-body u.v. light exposure was 57% lower in 
obese than in nonobese subjects (32). On the other 
hand, obese patients may avoid sunlight, which is 
necessary for the synthesis of vitamin D in the skin (33).

This might be especially the case in hirsute PCOS 
women, who tend to hide from the public due to their 
appearance. There is evidence that low vitamin D levels 
are associated with obesity (6) and vice versa low 
vitamin D intake might be an independent predictor of 
obesity (34).

Recent data from an overweight/obese women’s study 
have suggested that women with high 25(OH)D levels 
respond more positively to hypocaloric diets and lose 
more body fat than women with low 25(OH)D levels 
(35). There is evidence that weight loss is probably the 
most effective treatment of PCOS women at the moment 
(36). Thus, vitamin D supplementation might be an 
element in the complex treatment of PCOS women. This 
hyphothesis is supported by the findings of our study 
indicating the association of low 25(OH)D levels with 
obesity, insulin resistance, and MS in PCOS women.

In our study, there was no correlation of 25(OH)D 
levels with FAl, total testosterone, or free testosterone. 
Our observations are in line with a previous study that 
did not find any differences among PCOS and healthy 
control women with respect to 25(OH)D levels (37). One 
previous report on a correlation between vitamin D
levels and FAI (6) could be mediated through the obesity-induced reduction in SHBG. On this note, the significant correlation between SHBG and 25(OH)D in our patients was abolished when controlling for BMI. However, we found an inverse correlation of 25(OH)D with hirsutism score that was independent of BMI, which is in line with previous studies (6, 38). In addition, 25(OH)D levels were significantly lower in hirsute women, which is consistent with the results from a small study in hirsute women (38). This association might be caused by various mechanisms. First, the cosmetic distress may cause hypovitaminosis D due to the decreased sun exposure of hirsute women, as mentioned above. Secondly, the vitamin D receptor is found in keratinocytes of the outer root sheath as well as in cells of the bulge, indicating an important role of vitamin D in hair follicle cycling (28). However, the mechanism by which the vitamin D receptor regulates hair follicle cycling and its potential role in hirsutism remains unclear.

Our study has several limitations that should be noted. First, this study does not include a control group. Thus, we are not able to correlate the effects of low 25(OH)D levels with the metabolic profile of PCOS women specifically or with that of obese women in general. However, we performed subgroup analyses of lean and obese PCOS women with different 25(OH)D levels to address this limitation. Second, there was no information available with respect to calcium and carbohydrate intakes and exercise prior to biochemical measurements, which might influence our association of 25(OH)D levels with metabolic parameters (39, 40).

To the best of our knowledge, we are the first to describe an inverse association of low 25(OH)D levels with impaired β-cell function, IGT, and MS in PCOS women. Further, we confirm previous findings reporting the relationship of low 25(OH)D levels with obesity and insulin resistance women with PCOS. To prove these findings and to find new therapeutic approaches, large intervention trials with vitamin D supplementation are warranted in PCOS women.

Declaration of interest

The authors declare no conflict of interest.

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