The Role of Vitamin D in Metabolic Syndrome in Polycystic Ovary Syndrome

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Abstract

Objective: The aim of this study was to assess the women complaining from polycystic ovarysyndrome and the relation of serum vitamin D levels according to the different phenotypes of the disease by a retrospective study.

Methods: Records for 242 infertile women diagnosed with PCOS have been examined. In this retrospective study, 100 of them have been randomly recorded for 4 PCOS phenotypes. 40 normal ovulation women with a male factor history were selectedas the control group.

In four phenotype types including: age, BMIs, the infertility duration, the hormonal profile and the serum vitamin D, the P value of 0.05 was found to bestatistically significant.

Results: In comparison with PCOS patients (P<0.001), the data showed a serum vitamin Dlevel of statistically importance in the control group. Also, four PCOS phenotypeshad no significant variation in the serum vitamin D levels.

Conclusions: There were no significant differences between the serum vitamin D levels of thedifferent PCOS phenotypes. The additional studies of large samples are recommended for determining the role of the serum vitamin D level in PCOS patients.

Keywords: *Infertile women, Polycystic ovary syndrome, Rotterdam Criteria, Vitamin D.*

Introduction

Polycystic ovary syndrome (PCOS) referred to as a condition affecting the endocrine system. Approximately 5-21 percent of women in reproductive age areat risk. The main symptoms of PCOS commonly are ovulatory failure, menstrual cycle defect, and hirsutism. PCOS is a metabolic syndrome of various types withother characteristics including hyperandrogenemia, dyslipidemia and insulin resistance ([1]).

The guidelines for the diagnosis of PCOS are Rotterdam criteria. PCOS is highly accepted to be diagnosed by these criteria. This disease is typically diagnosed in women with two or more of the three symptoms I; polycysts of ultrasound ovaries, II; hyperandrogenemia and III; oligo or anovulation ([2]).

The patient health and infertility risk can be increase by PCOS([3]). The term polycystic ovarian syndrome does not entirely or correctly reflect the complications of the disease. It covers a broad range of diseases and clinicalexpressions.

The indications of reproductive defects is high in women with PCOS, includingtype 2 diabetes mellitus, Insulin resistance, high blood pressure, high LDL,

lowHDL, anxiety and depression([4]).

PCOS in pregnant women have a significant risk of preeclampsia, gestational diabetes, small-for-gestational age infants, fetal macrosomia and perinatal mortality ([5]).

There have been major efforts to classify PCOS types over the last few decades. The initial data comprised the PCOS classification indicator for chronic anovulation and hyperandrogenism.

The data gathered were finally integrated into the Rotterdam criteria([6]). On the basis of the criteria in Rotterdam, four different PCOS phenotypes may be defined including chronic anovulation, polycystic ovary (PCO) and hyperandrogenism. polycystic ovaries and hyperandrogenism but ovulatory cycles; chronic anovulation but normal ovaries, and polycystic ovaries and chronic anovulation butno biochemical or clinical hyperandrogenism ([7]).

Recently, vitamin D supplementation are focused by the clinical researches as an adjuvant therapy for PCOS. Women with PCOS have been confirmed to be high in vitamin D deficiencies. Consequently, a correlation was established with several metabolic symptoms in patients suffering from serum vitamin D.

Recently, vitamin D deficiency has been suggested to be a cause of PCOS([8]).

Numerous studies have demonstrated an adverse correlation of the vitamin D biomarker, e.g. 25-hydroxy vitamin D (25 OH D), with waist circumference, fat level in the body and body mass index (BMI) ([9]-[11]).

A study that examines serum vitamin D in different PCOS phenotypes has been found in the literature review for this study. In this retrospective study, the level ofvitamin D in women with PCOS has been assessed by different phenotypes.

Methods

In this retrospective study, the clinical and laboratory records of 2500 patients have been examined for a 30-month period from April 2017 to August 2019. PCOS was diagnosed in 250 infertile women aged 18-40. In each phenotype group (200/250 women), fifty cases were then randomly enrolled in the study. Fifty normal, ovulatory women with a male history have been chosen as the control group.

Standard ovary reserve (AMH > 1.2 ng/mL) and non PCOS in control group women with regulatory objectives based on selected Rotterdam criteria.

Criteria for inclusion: the diagnosis for polycystic ovary syndrome was made by women aged 18-40 years based on Rotterdam criteria.

The exclusion criteria were: women with a history of recurrent miscarriage, endometriosis, hyperprolactinemia and a severe male factor (PESA), oligospermia,testicular sperm extraction (TESE).

Based on the Rotterdam criteria, PCOS was diagnosed([2]). If at least two of the following criteria were fulfilled, PCOS would be diagnosed: Clinical and/or biochemical hyperandrogenism (defined by overall concentrations of circulating testosterone over a maximum of 35 days (0,481 ng/mL) are more than 95 percent of the levels detected in the women's group without clinical evidence of menstrual disorders or hyperandrogenism taking no hormonal medication), 'oligomenorrhea/anovulation' (defined as the delay in menses over 35 days or less than 8 spontaneous hemorrhagic episodes per year), polycystic ovary on ultrasonography (≥ 12 small follicles measuring 2-9 mm in at least one ovaryand/or the ovarian volume > 10 cm^3).

The patients were classified in the following defined criteria into four groups: polycystic ovary (PCO), chronic anovulation, and hyperandrogenism; polycysticovaries and hyperandrogenism but ovulatory cycles, chronic anovulation and hyperandrogenism but normal ovaries, and polycystic ovaries and chronic anovulation but no biochemical or clinical hyperandrogenism.

Data from the hospital records were collected. The ELISA kit and range of detection of this test was between 1.56 to 100 ng/ml for biochemical testing in order to determine vitamin D level. Specific ELISA kits were used for other hormones.

The data from patients included duration of infertility, age, hormone profile, body mass index BMI, and serum vitamin level D.

Statistical Analysis

Using the Social Sciences Package 20.0 data were analyzed (SPSS, SPSS Inc, Chicago, Illinois). Continuous data was presented to the independent student t-test as a mean ± standard deviation (SD). A post-hoc LSD with analysis of variance (ANOVA) was used for comparison of the mean of pair parameters. Less than 0.05 p-value was regarded statistically important.

Results

In this study, data were analyzed for 200 PCOS cases compared to 50 cases in control group for male factor infertility. Table 1 summarizes the patient characteristics. Both groups showed a similar average age and duration of infertility. The serum vitamin D was statistically significantly higher in the control group than in patients with PCOS ($P < 0.001$) on statistical analysis. Other factors such as BMI: body mass index; AMH: anti-Mullerian hormone; FBS: fasting blood sugar: FSH: follicle stimulating hormone; LH: luteinizing hormone were significantly different between the two groups (Table 1).

Table 1 Comparison between PCOS and control characteristics of women

p-value	Control (n = 50)	PCOS (n = 200)	Variables
NS	28.46 ± 4.18	28.19 ± 5.37	Women's age (years)
NS	5.44 ± 3.53	5.82 ± 3.78	Infertility duration (years)
0.001	29.08 ± 5.50	20.60 ± 9.22	Vit D3 (ng/mL)
0.001	3.76 ± 1.22	8.70 ± 4.07	AMH (ng/mL)
0.001	23.96 ± 2.63	28.54 ± 3.45	BMI (kg/m2)
0.001	5.36 ± 2.96	9.14 ± 5.51	LH (IU/l)
0.001	6.66 ± 1.69	5.42 ± 2.21	FSH (IU/l)
0.001	0.88 ± 0.44	1.97 ± 2.06	LH/FSH ratio ≥2.5
0.001	97.56±14.12	99.16±17.76	FBS (mg/dl)

Note: Mean ± SD values are shown in a single way P-values obtained from adifference between media were tested on ANOVA for importance.

PCOS: polycystic ovarian syndrome; AMH: anti-Mullerian hormone; BMI: bodymass index; FSH: follicle stimulating hormone; LH: luteinizing Hormone;

FBS: fasting blood sugar.

The parameters have been compared between the PCOS phenotypes, as shown in Table 2. The serum vitamin D level of the four PCOS phenotypes did not differ significantly. The four phenotypes demonstrated a significant three-parameter variation of AMH, LH and testosterone. Major parameter differences and comparison results between the PCOS phenotypes pair showed by letters.

Important differences in the AMH-related phenotypes were noted between A and B (P-value = 0.02). Moreover, LH differs markedly between A and C (P-value + 0,007), as well as between A and D (P-value = 0,08). The difference in the FBS parameter between B and C was also marked (P-Value = 0.008) (Table 2).

The results of comparisons between vitamin D concentrations associated with the PCOS variables indicate that the serum vitamin D levels did not differ significantlyfrom the study parameters. (Table 3).

Table 2 Study variables comparison in four PCOS phenotypes

p-value	Phenotype D (n = 50)	Phenotype C (n = 50)	Phenotype B (n = 50)	Phenotype A (n = 50)	Variables
NS	**27.20 ± 6.13**	**28.12 ± 4.81**	**29.44 ± 4.57**	**27.98 ± 5.71**	**Women's age (years)**
NS	5.28 ± 4.04	6.00 ± 3.91	6.08 ± 3.40	5.90 ± 3.80	Infertility duration (years)
NS	19.97 ± 9.33	20.54 ± 9.53	20.92 ± 9.43	20.97 ± 8.80	Vit D3 (ng/mL)
0.001	9.71 ± 5.39b	8.53 ± 3.33	7.13 ± 2.35a, b	9.43 ± 4.19a	AMH (ng/ml)
NS	28.27 ± 4.30	28.44 ± 3.57	28.43 ± 3.03	29.00 ± 2.75	BMI (kg/m2)
0.01	8.54 ± 5.46d	7.61 ± 5.07c	9.26 ± 4.87	11.14 ± 6.12c, d	LH (IU/l)
NS	5.39 ± 2.26	5.19 ± 1.88e	5.94 ± 2.62e	5.16 ± 1.97	FSH (IU/l)
NS	2.16 ± 3.48	1.52 ± 1.06	1.73 ± 0.92	2.41 ± 1.66	LH/FSH ratio
0.01	98.04 ± 10.66	102.30 ± 21.85	91.86 ± 12.85	98.52 ± 16.73	FBS (mg/dl)
0.001	0.56 ± 0.28f	0.75 ± 0.41	0.94 ± 0.35	0.91 ± 0.38f	Testosterone (IU/l)

Note: Mean value ± SD is shown. The importance of ANOVA was tested in ranks by one way, P-values derived from the difference of means. A statistical difference for comparison between PCOS phenotypes is indicated in different letters: -

a: Significant between A and B phenotypes. c: Significant between A and C; d: Significant between A and D phenotypes; e: Significant between B and C phenotypes; b: Significant between B and D phenotypes; f: Significant between A and D phenotypes.

FSH: follicle stimulating hormone; LH: luteinizing hormone; BMI: body mass index; AMH: anti-Mullerian hormone; FBS: fasting blood sugar.

Table 3 Comparison of the levels of PCOS-related vitamin D

p-value	Toxicity level (>100 ng/mL) (n = 0)	Sufficiency (30 to 100 ng/mL) (n = 74)	Insufficient (20 to <30 ng/mL) (n = 99)	Deficiency (<20 ng/mL) (n = 27)	Variables
NS	-	**28.04 ± 5.10**	**28.30 ± 5.91**	**28.15 ± 3.93**	**Women's age (years)**
NS	-	5.36 ± 3.58	6.30 ± 5.82	5.26 ± 4.07	Infertility duration (years)
NS	-	8.61 ± 4.08	8.73 ± 3.80	8.84 ± 5.09	AMH (ng/mL)
NS	-	28.22 ± 2.86	28.76 ± 3.21	28.57 ± 5.35	BMI (kg/m2)
NS	-	8.74 ± 5.57	9.06 ± 5.41	10.51 ± 5.73	LH (IU/l)
NS	-	5.29 ± 2.09	5.44 ± 2.35	5.68 ± 2.22	FSH (IU/l)
NS	-	1.80 ± 1.14	2.09 ± 2.70	2.02 ± 1.20	LH/FSH ratio
NS	-	98.68 ± 12.91	97.05 ± 17.58	97.26 ± 20.59	FBS (mg/dL)
NS	-	0.85 ± 0.39	0.77 ± 0.36	0.71 ± 0.44	Testosterone (IU/l)

Note: Mean value ± SD is shown. Analysis of variance has been tested for significance in ranks by one-way P-values obtained from difference betweenmeans.

Discussion

The focus of this study was on serum vitamin D and the prevalence of vitamin D deficiency among PCOS patients. The results showed that the total serum level of vitamin D and vitamin D deficit are significantly different for women with PCOS and controls. However, the serum vitamin D level of different PCOS phenotypes did not differ significantly.

As much evidence demonstrates that vitamin D plays an important role in reproductive activities, all patients were infertile in the current study([13]). Studies have shown that vitamin D receptors are present in many tissues, including endometrium, ovaries and placenta in the reproductive system([14]). Vitamin D deficiency has been shown to be associated with calcium dysregulation. This condition increases follicular arrest and leads to menstrual and fertility

disorders inPCOS-patient women.

According to the study, in women with PCOS (n=545) the concentrations of serum vitamin D were lower than in the control group (n= 145) and 25.7 and 32 ng/mL respectively. Many studies suggest that the levels of serum vitamin D in women with or without PCOS are similar([16]-[18]).

In the study for vitamin D-PCOS correlations, only one study found that women with PCOS had a significantly higher serum vitamin D level than control womenof similar age and BMI([19]).

In the literature on serum vitamin D levels, therefore, there are various results forwomen with or without PCOS. The paper about vitamin D assessment were published in 2018 by Newly Davis and cols. where in all PCOS cases and male

infertility as a control group in the intrauterine insemination cycle. They showed a lower level of vitamin D than other PCOS phenotypes in androgen excess([12]).

However, this finding did not show the androgen level to find excessive androgen, but we presented androgen as testosterone levels in 4 phenotypes. These results showed that androgen in patients with deficiency was less than other vitamin D categories, but that difference was not statistically significant (Table 3). In four phenotypes, They also presented fertility hormone levels and this was what made the study strong.

Kim and cols. reported that 2-month therapy of 1,500 mg calcium daily and 50,000 weekly unit vitamin D improved menstrual cycles in 7 of 9 cases in PCOS patients with vitamin D deficiency([18]).) There have been other trials in BMI, body fat, insulin and hyperinsulinemia patients in PCOS that showed negative linkage between the levels of serum vitamin D and metabolic disorder. ([20]-[22]).

Studies showed that PCOS is correlated with other metabolic problems like, dyslipidemia, depression, anxiety, high blood pressure and chronic inflammation. Metabolic disorders in women with PCOS have also been confirmed([23]).

In their study, Hang Wun and cols stated that in Scottish women with PCOS vitamin D was highly prevalent and this rate was higher in the UK than ovular controls. In addition, vitamin-D deficiency has been shown to be correlated with metabolic risk factors, including insulin resistance and low HDL-C levels, regardless of obesity measures. ([17]).

Although no trial to evaluate serum vitamin D levels in a range of PCOS phenotypes was found in the literature review of the current study, some studies evaluated PCOS-Clinical correlations. Reza Ghadimi and cols were found, although PCOS patients had common hypovitaminosis D, it was not correlated to the clinical features to insulin resistance or obesity and complications. In PCOS patients, there has been correlation between the severity of vitamin D deficit and certain PCOS features and complications, including Obesity and Insulin resistance.([24]).

The study of 260 PCOS women (cases) and 221 Normo-ovulatory (checks) whohave been recruited to a clinic for reproductive endocrinology was performed byVakili and cols. They classified the cases according to their clinical and para- clinical characteristics into two groups of serious and mild PCOS phenotypes.

Adenosine was a genotyped PCR–RFLp method for guanine nuclear polymorphism (rs757343) of the VDR gene. They noted that there was no change in the distribution of genotypes and alleles among cases and checks showing a lackof correlation between the single nucleotide polymorphism (SNP) and increasing PCOS risk. However, PCOS phenotype severity was the responsibility of the SNP. The risk that a severe phenotype was present was 74% higher than in other patientsin the case of an allele. They also showed that the genetic version of the VDR was associated with the seriousness of the PCOS' clinical presence, but not PCOS risk([25]).They assessed the correlation between VDR and PCOS in Indian patients ina genetic study

by Dasgupta and cols. They identified a significant relationship between the VDR genotype and some PCOS events([26]).

In conclusion, we found no significant vitamin D differences in various PCOS phenotypes. Other studies recommend additional studies with larger samples to conclude that serum vitamin D is of importance in PCOS patients, including PCOS data phenotypes. The first restriction of insulin resistance and metabolic malformations associated with vit-D deficiency in PCOS was a retrospective study,and the lack of these data was limited by the manuscript.

Conflict of Interest: There is no conflict of interest among the authors.

Funding: Self

Ethical Clearance: This study is ethically approved by the Institutional ethical Committee.

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