Correlation of Serum Vitamin D and Metabolic Disturbances in Polycystic Ovarian Syndrome

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Abstract
Objective: This study was aimed to determine the correlation between vitamin D and insulin resistance in women with PCOS.
Method: This study was correlational analytic with cross-sectional approach to 34 women diagnosed with PCOS based on ultrasonography. Waist circumference and fasting blood glucose (FBG) represented insulin resistance. Women with hormonal therapy and vitamin D supplementation were not included to this study. This study used consecutive sampling method.
Result: The average of age was 25.6±6.1 years old. Waist circumference and fasting blood glucose (FBG) represented insulin resistance. The average of waist circumference and FBG were 87.6±12.4 cm and 86.2±27.9 mg/dl, respectively. The mean of vitamin D levels was 11,5±3,6 ng/ml. According to Spearman's correlation, vitamin D levels were weak negative correlated with waist circumference (r=-0.2; p>0.05) and FBG (r= -0,1; p>0,05), it statistically was not significant.
Conclusion: There is weak negative correlation between vitamin D and metabolic syndrome in PCOS patients.

Key words: polycystic ovarian syndrome, vitamin D , insulin resistance, metabolic syndrome

Korelasi Kadar Vitamin D Serum dan Gangguan Metabolik pada Sindrom Ovarium Polikistik

Abstrak
Tujuan: Penelitian ini bertujuan untuk menganalisis korelasi vitamin D dengan resistensi insulin pada pasien dengan SOPK.
Metode: Penelitian ini merupakan penelitian analitik korelatif dengan pendekatan potong lintang pada 34 subjek penelitian yang didiagnosis SOPK berdasarkan pemeriksaan ultrasonografi (USG). Lingkar pinggang dan kadar glukosa darah puasa (GDP) diambil sebagai parameter resistensi insulin. Pasien dengan terapi hormon dan suplementasi vitamin D tidak termasuk dalam subjek penelitian. Pengambilan sampel menggunakan teknik *consecutive sampling*.
Hasil: Rerata usia subjek penelitain ini adalah 25,6±6,1 tahun. Lingkar pinggang dan glukosa darah puasa (GDP) diambil sebagai parameter resistensi insulin. Pada penelitian ini subjek memiliki rerata lingkar pinggang 87,6±12,4 dan GDP 86,2±27,9 mg/dl. Rerata kadar vitamin D subjek 11,5±3,6 ng/ml. Berdasarkan uji korelasi Spearman, kadar vitamin D berkorelasi negatif lemah dan tidak signifikan secara statistik baik dengan lingkar pinggang (r= -0,2; p>0,05) maupun dengan GDP (r= -0,1; p>0,05).
Kesimpulan: Vitamin D berkorelasi negatif lemah dengan parameter gangguan metabolik resistensi insulin pada pasien SOPK.

Kata kunci: sindroma ovarium polikistik, vitamin D, resistensi insulin, sindroma metabolik

Introduction

Polycystic ovary syndrome (PCOS) is the most common endocrinopathy in reproductive age women, with prevalence of up to 3—10% in the world.[1] About 50—70% of women with PCOS had insulin resistance. Insulin resistance is defined as the state in which normal concentration of insulin produces subnormal effects on glucose homeostasis and utilization. Insulin resistance triggers the production of excess androgens in adolescent and adult PCOS patients.[2]

Insulin resistance is clinically manifested as metabolic syndrome. Women with PCOS are known to have abnormalities in insulin activity, metabolism, and in control of androgen production.[3] Vitamin D deficiency often occurs in women with PCOS. Approximately 67—85% of women with PCOS have a serum concentration of 25-hydroxy vitamin D 25-(OHD) <20 ng/mL. Vitamin D deficiency can worsen PCOS symptoms. Several studies have shown that vitamin D supplementation can improve insulin resistance and reduce serum androgens level.[4]

Vitamin D supplementation can affect insulin secretion and improve glucose homeostasis in obese people with type-2 diabetes mellitus. The 25-(OHD) concentration is known to be negatively correlated with body mass index (BMI), body fat, and insulin resistance. However, the effect of vitamin D supplementation on the treatment of insulin resistance in women with PCOS remains unclear.[2]

The association of vitamin D and metabolic disorders has been investigated in many studies. The association between vitamin D and insulin resistance has also been extensively studied in diabetic patients, yielding compelling evidence that vitamin D deficiency is inversely related to the severity of insulin resistance,[5] with the 67-85% of women with PCOS having serum concentrations of 25-hydroxy vitamin D 25-(OHD). Further studies are needed to investigate the correlation between vitamin D and insulin resistance in patients with PCOS. This study aimed to determine the correlation between insulin resistance and vitamin D in patients with PCOS.

Method

This study was a correlational analytic study with cross-sectional approach to women diagnosed as PCOS at Dr. Hasan Sadikin General Hospital Bandung from January–December 2019. This study used consecutive sampling method. The inclusion criteria were patients diagnosed as PCOS based on ultrasonography and not undergoing hormonal therapy. Patients taking vitamin D supplements or antihyperglycemic drugs were excluded from the study. Informed consent had been carried out to prospective research subjects. All designs and procedures in this study have been approved by the Research Ethics Committee of Dr. Hasan Sadikin Bandung.

The diagnosis of PCOS was based on the presence of 2 out of 3 symptoms, namely (i) clinical or biochemical signs of hyperandrogenism; (ii) chronic ovulation disorders; and (iii) the presence of polycystic ovary morphology on ultrasound examination.[6]duration >6 months waist circumference and fasting blood glucose (FBG) represented insulin resistance.

Prospective research subjects were screened for vitamin D and fasting blood glucose (FBG) simultaneously. Patients fasted for 8–12 hours before blood tests were drawn. Weight, height and waist circumference were also measured, and the body mass index (BMI) were then calculated.

The threshold values for BMI was ≥23 kg/m^2 for overweight and ≥25 kg/m^2 for obesity, and a waist circumference of ≥80 cm was considered a metabolic risk.

Based on the Endocrine Society Clinical Practice Guidelines, vitamin D deficiency is a level of 25-(OHD) <20 ng/mL, vitamin D insufficiency is a level of 25OHD between 21 and 29 ng/mL, whereas a normal level of vitamin D is defined as a level of 25-(OHD) >30 ng/mL. Vitamin D deficiency is divided into mild25-(OHD) >10–20 ng / mL), moderate 25-(OHD) 5–10 ng / mL), and severe 25-(OHD) <5 ng / mL) deficiency.

Venous blood samples were stored at temperature of -80 °C after centrifugation at 3000 rpm for 10 minutes at 20 °C. Serum 25(OH)D and fasting blood glucose were measured using spectrometry. Measurement of serum 25-(OHD) was carried out by the Laboratory of the Department of Clinical Pathology, Dr. Hasan Sadikin General Hospital. The data collected were processed and analyzed descriptively and analytically.

The mean, standard deviation, median, and range of the data were calculated. Normality test was carried out using Saphiro-Wilk test (n<50). Spearman correlation test was conducted to determine the correlation between variables (p<0.05) was considered statistically significant).

Result

There were 34 patients, with the mean age of 25.6±6.1 years old. Majority of the subjects in this research were in the age group of 20—29 years old. Body mass index was classified into underweight, normal weight, overweight, and obese based on Asian population standards. The majority of the subjects (47.1%) were categorized as obese. The mean waist circumference was 87.6±12.4 cm. Waist circumference was classified as normal (<80 cm) and excessive (≥80 cm) based on Asian population criteria.

A total of 27 out of 34 subjects had excessive waist circumference. The mean fasting blood glucose level was 86.2±27.9 mg/dl. Fasting blood glucose levels were classified based on the threshold value of the metabolic syndrome criteria (100 mg/dL). Only 6 out of 34 subjects had fasting blood sugar levels above 100 mg/dL.

The mean vitamin D level was 11.5±3.6 ng/ml. Vitamin D levels were classified into normal, insufficient, and deficient. Only one subject had normal vitamin D levels and 23 out of 34 subjects (67.6%) were categorized as vitamin D deficiency.

Table 1 Characteristic of the Research Subjects

Characteristic	N=34
Age, mean±SD (years)	25.6±6.1
<20 years old, n (%)	11(14.7)
20-29 years old, n (%)	18(52.9)
≥30 years old, n (%)	5(32.4)
Job	
Housewife, n(%)	30(88.2)
Laborer, n(%)	4(11.8)
Menstrual cycle	
Irregular	20(58.8)
Secondary amenorrhea	14(41.2)
Body Mass Index, mean±SD (kg/m^2)	24.78±5.46
Underweight, n(%)	4(11.8)
Normal weight, n(%)	10(29.4)
Overweight, n(%)	4(11.8)
Obese, n(%)	16(47.1)
Waist circumference, mean ±SD (cm)	87,6±12.4
<80 cm, n (%)	7(20,6)
≥80 cm, n (%)	27(79,4)
Fasting Blood Glucose Levels, mean±SD (mg/dL)	86,2±27,9
<100 mg/dL, n(%)	28(82,4)
≥100 mg/dL, n(%)	6(17,6)
Vitamin D Levels, mean±SD (mg/dL)	11,5±3,6
Vitamin D deficiency	23(67,6)
Vitamin D insufficiency	10(29,4)
Normal	1(2,9)

Correlation coefficient of r = –0.1 (p> 0.05) between vitamin D levels and fasting blood glucose was obtained using Spearman correlation test (Figure 1). This indicates that there was a weak correlation which was not statistically significant between vitamin D levels and fasting blood glucose levels in patients with PCOS.

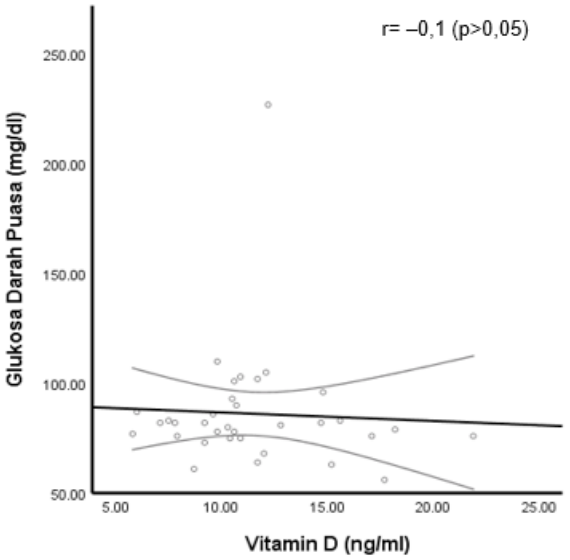

Figure 1 Scatter Plot Showing the Correlation between Vitamin D levels and Fasting Blood Glucose Levels

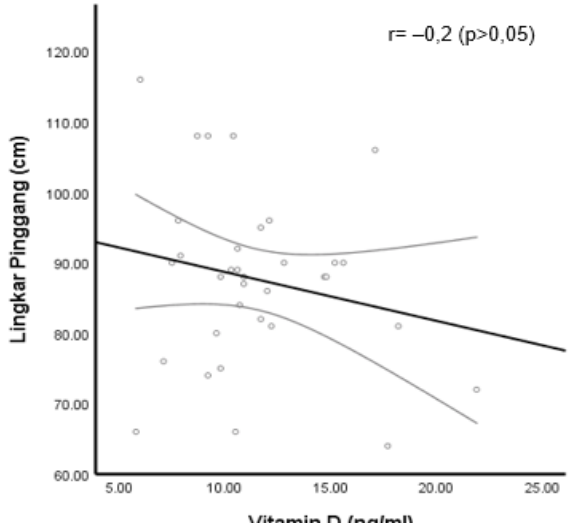

Figure 2 Scatter Plot Showing the Correlation between Vitamin D levels and Waist Circumference

Based on Spearman correlation test the correlation coefficient between vitamin D levels and waist circumference was r = -0.2 with p value > 0.05. This indicates that there was a weak correlation which is not statistically significant between vitamin D levels and waist circumference in patients with PCOS. (Figure 2),

Discussion

This study aimed to find the correlation between vitamin D levels and insulin resistance, which was represented by fasting blood glucose levels and waist circumference. The mean age of the subjects in this study was 26 years old. This is in accordance with the study conducted by Wahyuni *et al*. which stated that the age group with the highest prevalence of PCOS is between age 21–30 years old.[7]

The mean BMI in this study was 24.78 kg/m2 and almost 80% of the subjects in this study had a waist circumference above the threshold criteria for metabolic syndrome. These results are similar to the study conducted by Musmar *et al*. showed that the mean BMI and waist circumference of patients with PCOS were 23.37±3.85 kg/m2 and 80.08±7.69 cm, respectively.[8] Comparable to our study, the study of Kiranmayee *et al*. reported that about 80% of subjects with PCOS had a waist circumference above the normal threshold and the overweight to obese group accounted for more than half of the subjects with PCOS.[9] These results can be explained by the fact that women with PCOS have abnormalities in insulin activity, metabolism, and in control of androgen production.

The association between PCOS and obesity has a specific role in disrupting insulin sensitivity and insulin resistance in arterial walls whics is known to be associated with decreased synthesis and release of nitric oxide (NO), increased NO inactivation,

increased synthesis of vasoconstricting agents, and impaired insulin vasodilation.

In addition, excess insulin is known to cause a direct hypertrophic effect on vascular endothelium and smooth muscle cells which stimulates endothelin-1 production, thereby causing endothelial dysfunction. Insulin resistance is the most important factor responsible for impaired insulin secretion, which causes the very high incidence of type II diabetes mellitus in women with PCOS. The combination of insulin resistance, obesity, central (visceral) obesity, and type II diabetes mellitus is associated not only with androgen excess and metabolic changes, but also with coagulation and fibrinolysis disorders, anatomical and functional endothelial injury, vascular dysfunction, and subclinical inflammatory states. Taken together, all of these factors may represent an independent risk for cardiovascular disease, particularly after menopause.[10] Visceral adiposity, dyslipidemia, insulin resistance, and hypertension are common in these patients. Unreplaced growth hormone (GH).

Polycystic ovary syndrome arises from the interaction of genetic and environmental factors. Genes involved in steroidogenesis, carbohydrate metabolism and major histocompatibility areas, sex hormone binding globulin (SHBG), insulin receptors, follistatin and CYP17 may be involved in the development of PCOS.[3] Disorders of fat metabolism in PCOS patients may underlie the increasing waist circumference and body mass index results found in this study.

The mean fasting blood glucose level in this study was 86.2±27.9 mg/dl. The threshold for fasting blood glucose levels for patients with suspected metabolic syndrome is ≥100 mg/dl. In this study, only six subjects had fasting blood glucose levels above this threshold. There was a significant difference between the proportion of waist circumference above normal (79.4%) and glucose levels above normal (17.6%). An increase in waist circumference is not always accompanied by an increase in fasting blood glucose levels and vice versa. Previous studies showed that there was only a moderate correlation between fasting blood glucose levels and waist circumference.[11] This result suggested that an increase in blood glucose is not always accompanied by an increase in waist circumference. Thus, the difference in the proportion between waist circumference above normal (79.4%) and glucose level above normal (17.6%) is very likely to happen. The mean level of vitamin D in this study was 11.5 ng/ml which was categorized as mild deficiency. Only one subject had normal vitamin D level. This result is in accordance to the study conducted by Faraji, *et. al*. and Kumar, et.al. Both of these studies agreed that vitamin D deficiency is a common occurrence in patients with PCOS,[6,12] duration >6 months.

The low vitamin D levels found in this study may be explained by the occupation of the study subjects. About 88.2% of the patients were housewives, who may have less sun exposure. These characteristics may explain the low vitamin D levels of PCOS patients in this study.

The main result of this study was that there was a weak correlation which was not statistically significant between vitamin D levels and fasting blood glucose levels and between waist circumference and vitamin D levels. The results of this study were comparable to several other studies but contradictory to some other studies. The study by Kumar, *et. al*. stated that there was no correlation between vitamin D levels both with glucose metabolism parameters, including fasting blood glucose levels or with anthropometric parameters, including waist circumference.[9] A meta-analysis by He, *et.al*. presented the calculation of six correlational studies between fasting blood glucose and vitamin D. Based on this meta-analysis, the correlation between vitamin D and fasting

blood glucose in polycystic ovary syndrome patients was -0.23 (p=0.009).

This correlation coefficient is similar to the correlation coefficient in this study (-0.2) but the significance of the p value in this study was still above 0.05,[13] and to determine effects of vitamin D supplementation on metabolic and hormonal functions in PCOS patients.

The literature search was undertaken through five databases until 16 January 2015 for both observational and experimental studies concerning relationships between vitamin D and PCOS. A total of 366 citations were identified, of which 30 were selected (n=3182) The study conducted by Hassan, *et.al*. showed similar results to this study, in which vitamin D had a weak negative correlation with fasting blood glucose levels with a statistical test value of p above 0.05 although vitamin D was found to have a strong negative correlation to insulin resistance measured by the parameter homeostatic model assessment for insulin resistance (HOMA-IR).[14] The study conducted byy Kruel-Poel, *et. al*. showed that women with polycystic ovary syndrome had significantly lower serum 25(OH)D levels compared to fertile controls.[15]insufficient (25–50 nmol/l Compromised vitamin D status in women with PCOS is associated with a higher HOMA-IR and an unfavorable lipid profile. Seyyed, *et. al*. demonstrated that 8 weeks of vitamin D supplementation among women with PCOS and vitamin D deficiency led to significant reductions in fasting blood glucose levels and significant increases in levels of HOMA-B (homeostasis model assessment of beta cell function), adiponectin, and vitamin D levels.

Although the vitamin D supplementation group was better than the placebo group in term of stability of insulin resistance, the difference in change from baseline for HOMA-IR and QUICKI (Quantitative Insulin Sensitivity Check Index) was not significant between the two groups.[4] These different results may stem from the different subject characteristics, length of study, form of vitamin D given for supplementation, and VDR (Vitamin D receptor) or VDBG (Vitamin D-binding protein) polymorphism. Vitamin D supplementation in women with PCOS effectively reduces the concentration of fasting blood sugar and HOMA-IR. HOMA-IR was reduced when vitamin D was given daily at low doses, namely <4000 IU/ day.[2]

Vitamin D can affect adiponectin via the renin-angiotensinogen system. Increased activity of the renin-angiotensinogen system is associated with increased angiotensin production, which leads to dysfunctional adipocyte production and ultimately, decreased adiponectin production. Vitamin D may increase serum adiponectin by regulating and decreasing angiotensin production. Insulin resistance and glucose intolerance are inflammatory conditions associated with decreased adiponectin production and increased activity of inflammatory cytokines such as TNF- α and interleukin-1. Tumor necrosis factor-alpha (TNF-α) reduces adiponectin synthesis, while vitamin D may increase serum adiponectin through decreased gene expression.[4]

This research still has several limitations. First, this study only analyzed the data bivariatively, without considering several confounding variables such as age, food intake, sun exposure, and socioeconomic status. Second, this study did not perform other insulin resistance tests such as insulin levels, which if insulin level data was present, HOMA-IR can be calculated. Third, this study did not assess other metabolic syndrome parameters such as total cholesterol and triglyceride measurements. Finally, this study had a small sample size and a cross-sectional research method.

However, the use of fasting blood glucose and waist circumference as parameters were sufficient to describe insulin resistance,

because based on the research of Pourfarzam *et. al*, the correlation of HOMA-IR with fasting blood glucose levels was very strong and waist circumference was also moderately correlated with HOMA-IR.[16]

Conclusion

There was a weak negative correlation which was not statistically significant between vitamin D and fasting blood glucose and between vitamin D and waist circumference in women with polycystic ovary syndrome. Further research with a wider sample size, especially in tertiary non-referral centers are needed.

Conflicts of Interest

We declare that we do not have any conflicts of interest.

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