Oligo-Amenorrheic Polycystic Ovary Syndrome Patients Have Higher Risk For Cardiovascular Disease Compared to Hirsute Patients and Healthy Control

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OBJECTIVE: To evaluate the clinical, endocrine and cardiovascular disease risk profile differences in polycystic ovary syndrome (PCOS) patients who complain of hirsutism or oligo-amenorrhea.

STUDY DESIGN: A total of 129 consecutive women underwent the screening investigation at Dr. Sami Ulus Women's Health Teaching and Research Hospital between December 2009 and June 2010. There were 48 PCOS patients with hirsutism while 49 PCOS patients with oligo-amenorrhea and 32 healthy women included in the study. Body mass index (BMI), Waist/hip ratio (WHR) serum follicle stimulating hormone (FSH), luteinizing hormone (LH), progesterone, free testosterone, glucose, low density lipoprotein (LDL), total cholesterol, high density lipoprotein (HDL), triglceride (TG), high sensitive C reactive protein (hs-CRP), insulin, insulin sensitivity and carotid intima thickness (CIMT) were compared in PCOS patients who complain of hirsutism or oligo-amenorrhea and control group.

RESULTS: Mean ages were similar among groups. There were significant differences among groups in terms of BMI, WHR, total cholesterol, LDL, TG, LH, fasting glucose, hsCRP, CIMT ($p<0.05$) (table 1). BMI, waist-hip ratio (WHR), total cholesterol, LDL, TG, LH, estrodiol, fasting glucose adjusted mean CIMT values were 0.40±0.01 mm, 0.49±0.012mm, 0.34±0.012mm ($p<0.001$) in hirsute, oligo-amenorrheic and control groups respectively.

CONCLUSION: PCOS patients with oligo-amenorrhea without hirsutism have higher risk for cardiovascular disease (CVD) and abnormal lipid profile than patients with hirsutism without oligo-amenorrhea.

Key Words: Polycystic ovary syndrome, Carotid intima media thickness, Rotterdam criteria

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Introduction

Polycystic ovary syndrome (PCOS) is a multifactorial and polygenic pathology that manifests itself with a wide spectrum of signs and symptoms that are related to the disturbances of reproductive, endocrine and metabolic function at various degrees resulting in a heterogenous presentation of the disease.[1] The diagnostic criteria for polycystic ovary syndrome (PCOS) have undergone several changes in recent years. While the clinical presentation of chronic anovulation and hyperandrogenism has been stressed as the major diagnostic criteria,[2] the presence of normal ovulatory function in women with PCOS has been acknowledged in recent years.[3-5] New diagnostic criteria were established in 2004, placing all these three factors; presence of chronic anovulation, hyperandrogenism and polycystic ovaries together, with a special emphasis placed on existence of polycystic ovaries on ultrasonography.[6,7] PCOS was diagnosed in the presence of two of the three diagnostic criteria.

The triad of hyperandrogenism, insulin resistance and anovulation affect various steps in lipid metabolism resulting in dyslipidemia observed in lean PCOS as well as the obese ones and lower levels of HDL-cholesterol is the most significant change[8] accompained by elevated low-density lipoprotein (LDL), triglyceride levels.[9]

In recent study women with ovulatory PCOS showed milder forms of atherogenic dyslipidemia than anovulatory PCOS.[10] Compared with control women, PCOS patients present with an increased CIMT, independent of obesity and related directly to androgen excess; this suggests that hyperandrogenism is associated with atherosclerosis and cardiovascular risk in these women.[11]

PCOS is associated with a higher risk of diabetes, hypertension, dyslipidemia, metabolic syndrome endothelial dysfunction and cardiovascular disease.[12] There are studies show-

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ing the association between subclinical atherosclerosis, vascular dysfunction and PCOS and they report reduced vascular compliance and vascular endothelial dysfunction in some of the patients with PCOS.[13-16] Measurement of carotid intimal thickness (CIMT), a noninvasive vascular evaluation of atherosclerotic plaque is a valuable tool for evaluation of cardiovascular disease risk in patients with PCOS as increase in CIMT is associated with cardiovascular events, mainly stroke .[17] The aim of this study was to evaluate the clinical, endocrine and cardiovascular disease risk profile differences in polycystic ovary syndrome (PCOS) patients who complain of hirsutism or oligo-amenorrhea.

Material and Method

A total of 129 consecutive women underwent screening investigation at Dr. Sami Ulus Women's Health Teaching and Research Hospital between December 2009 and June 2010. Group of patients with hirsutism consisted of 48 PCOS patients while there were 49 PCOS patients with oligo-amenorrhea and 32 healthy women who were admitted primarily in gynecology outpatient clinic included in compared group. All of the women in the control group had hirsutism score <8. All women in the control group had regular menses, every 21-35 days. None of the women in the control group had polycystic ovary in ultrasound. Women with PCOS presenting with one or two of the following complaints; oligomenorrhoea, hirsutism, infertility were systematically evaluated in our outpatient clinic. Standardized screening was approved by the local Institutional Review Board, and signed written informed consent was obtained from all of the participants. The study protocol was in agreement with the Helsinki Decleration of 1975 (revised version 2000, 52nd WMA General Assembly, Edinburgh).

According to the Rotterdam (ESHRE/ASRM-Sponsored PCOS Consensus Workshop Group, 2004) criteria, PCOS was diagnosed when at least two of the following criteria were present: oligo/amenorrhoea, clinical or biochemical hyperandrogenism and polycystic ovaries on ultrasonography. Paitents with other etiologies mimicking PCOS, like Cushing syndrome, late-onset adrenal hyperplasia or androgen producing neoplasm and thyroid dysfunction or hyperprolactinemia were exclusion criteria. Patients who had taken any medication during the previous 3 months were excluded from the study. Hirsutism was defined as the presence of hirsutism (Ferriman-Galwey score ≥8).[18] Polycystic ovary morphology (PCOM) was established using the criteria of 10 or more peripheral follicular cysts 8 mm in diameter or less in one plane along with increased central ovarian stroma based on the Rott-PCOS criteria (Rotterdam ESHRE/ASRM-Sponsored PCOS Consensus Workshop Group, 2004). Oligo-amenorrhea was defined as having cycles with longer than 35 days or amenorrhea of six months. Medical history; regarding age, race, menstrual cycle pattern, personal and family medical history, any previous or current use of medication, presence of acne and hirsutism were recorded. Menstrual cycle length shorter then 24 days and longer then 34 days were recorded as abnormal.

Body mass index (BMI), waist and hip circumferences were recorded. Fasting early morning endocrine profile (including pituitary hormones, ovarian and adrenal steroids), serum lipids, glucose and insulin levels were measured on day 3-5 of the menstrual cycle or randomly if the patient was amenorrheic. A pelvic ultrasonography was performed and the findings were recorded.

Waist circumference was measured in the standing position, halfway between the lower ribs and the superior anterior iliac spine of the pelvis. The hip circumference was measured at the level of the pubic symphysis. Hirsutism was established by using the Ferriman-Galwey score. Transvaginal ultrasonography was systematically performed by the same investigator using the 7.5 MHz transvaginal probe to a Logic Ultrasound System. Antral follicles were measured in three dimensions and those with a mean diameter of 2-9 mm counted. Plasma glucose was determined with glucose oxidase/peroxidase method (Gordion Diagnostic, Ankara, Turkey).

High-resolution B-mode ultrasound images of the right common carotid artery were obtained with a 7.5-MHz linear array transducer attached to a Logic Ultrasound System. Subjects were placed in a supine position with the head rotated to the left using a 45-degree head block. The jugular vein and carotid artery were located in the transverse view with the jugular vein stacked above the carotid artery. The transducer was then rotated 90 degrees around the central line of the transverse image of the stacked jugular vein-carotid artery to obtain a longitudinal image while maintaining the vessels in the stacked position. The distal common carotid arterial far wall IMT was measured by the radiologist who was blinded to the laboratory findings and PCOS group of the patient with any complaint.

Serum levels of follicle-stimulating hormone, luteinizing hormone (LH), prolactin, dehydroepiandrosterone sulfate (DHEAS), total testosteron (T), insulin, cortisol and thyroid stimulating hormone (TSH) were measured with specific electrochemiluminescence immunoassays (Elecsys 2010 Cobas, Roche Diagnostics, Mannheim, Germany). Serum levels of 17 hydroxyprogesterone and free testosterone were measured by radioimmunoassay. Levels of total-cholesterol, high dansity lipoprotein (HDL) cholesterol, and triglyceride (TG) were determined with enzymatic colorimetric assays by spectrophotometry (BioSystems S.A., Barcelona (Spain)). Low density lipoprotein (LDL) cholesterol was calculated using the Friedewald formula.

Serum c-reactive protein (CRP) was determined using high-sensitive CRP (hsCRP) immunonephelometry (BN, Dade-Behring; Marburg, Germany).

Insulin resistance was calculated by using the homeostatic model assessment insulin resistance index (HOMA-IR) according to the formula, HOMA-IR: fasting plasma glucose (mmol/L)x fasting serum insulin (mU/mL)/22.5.[19] Hyperandrogenemia was defined as serum-free testosterone greater than 3.2 pg/ml (normal range 0.8 -3.2 pg/ml). Insulin resistance was defined as HOMA-IR ≥ 3.5.[20] None of the patients with hirsutism has oligo-amenorrhea and patients with abnormal menstrual cycles consist of patients with Ferriman gallwey score lower than eight. Both groups have polycystic ovaries in sonografic evaluation.

Statistical analysis

Means were compared by ANOVA, catagorical variables were compared by chi-square test, regression analysis was performed to determine associations and ANCOVA was used for statistical adjustment for parameters.

Results

Mean ages were similar among groups. There were significant differences among groups in terms of BMI, waist-hip ratio (WHR), total cholesterol, LDL, TG, LH, estrodiol, fasting glucose, hsCRP, CIMT ($p<0.05$) (table 1). BMI, waist-hip ratio (WHR), total cholesterol, LDL, TG, LH, estrodiol, fasting glucose adjusted mean CIMT values were 0.40±0.01 mm, 0.49±0.012mm, 0.34±0.012mm ($p<0.001$) in hirsute, oligoamenorrheic and control groups respectively.

Regression analyses revealed that; BMI ($p=0.001$) and estrodiol ($p=0.002$) levels were positive predictors for CIMT after adjustment for age, WHR, free testosteron, fasting glucose, total cholesterol, LDL, TG, HDL.

PCOS patients were divided into two groups according to HOMA –IR (higher than 3.5 and lower than 3.5) and groups were found to be similar in terms of all the parameters except for fasting insulin levels. There were 11 (24%) patients with high HOMA-IR in hirsutism group while 2 (4%) patients in oligoamenorrheic group and 3 (9%) in healthy women ($P=0.03$). Highest insulin resistance was observed in hirsute group.

Another comparison was performed between patients with hyperandrogenemia (determined by free testosteron level ≥3.2 pg/ml) and without hyperandrogenemia (free testosteron level <3.2 pg/ml). There were 12 (25%) patients in hirsute group with high free testosterone level while 6 (12%) patients in oligoamenorrheic group and 1 (3%) in control group ($p=0.013$). Hyperandrogenemic and normoandrogenemic patients were similar according to all parameters.

Age, CIMT, fasting glucose, WHR, TG were higher in patients with BMI>25 and P values were 0.003, 0.019, 0.009, 0.047 respectively. There were 16 (32%) patients in hirsute group with high free testosterone level while 21 (42%) patients in oligoamenorrheic group and 6 (18%) in control group ($p=0.116$).

Table 1: The demographic and biochemical / hormonal values of the PCOS patients and healthy control

Variable	Oligoamenorrheic group (n = 49)	Hirsutism group (n = 48)	Healthy women (n = 32)	P
Age, years	23.4±3.9	22.7±3.8	24.1±4.7	0.467
BMI, kg/m^2	24.9±4.0	24.3±3.7	24.1±4.2	0.041
Waist/hip ratio	0.75±0.05	0.79±0.06	0.76±0.05	0.002
Fasting insulin, µ IU/ml	11.7±6.6	11.07±6.3	10.2±3.8	0.587
Fasting glucose, mg/dl	83.0±8.8	94.7±11.4	89.2±11.8	<0.001
HOMA-IR	2.4±1.3	2.5±1.4	2.2±0.8	0.609
Total cholesterol, mg/dl	190.8±36.5	172.1±42.3	154.1±31.4	<0.001
Triglyceride, mg/dl	118.5±67.8	106.2±65.1	75.7±31.4	0.013
HDL, mg/dl	58.4±32.4	54.8±10.7	53.8±16.1	0.626
LDL, mg/dl	116.5±30.5	96.6±31.8	85.9±27.4	<0.001
hsCRP, mg/L	0.70±1.5	2.5±4.6	1.6±1.8	0.039
Estrodiol, pg/ml	77.4±69.7	67.8± 89.7	124.4±80.9	0,013
FSH, m IU/ml	4.9±1.8	5.6±2.5	6.02±3.4	0.170
LH, m IU/ml	12.0±7.0	6.6±5.2	8.9±6.1	<0.001
Free Test,pg/ml	1.8±1.7	2.5±1.16	2.2±0.96	0.071
Carotid-IMT	0.48±0.54	0.41±0.05	0.33±0.04	<0,001

Discussion

IThe aim of this study was to evaluate the clinical, endocrine and cardiovascular disease risk profile differences in PCOS patients who complain of hirsutism versus oligo-amenorrhea. We also assesed metabolic factors in PCOS patients divided according to their BMI (>25 kg/m^2) and serum androgen levels (≥3.2 pg/ml). In this study, the menstrual abnormality of oligo-amenorrhea was found to be major determinant for high cardiovascular diseaese (CVD) risk in young age PCOS patients. Oligo-amenorrheic and hirsute groups were similar in terms of high BMI (p=0.737) and HOMA-IR (p=0.820). This CVD risk difference was thought to be due to abnormal cycles itself free from BMI and HOMA-IR. Free androgen levels were higher in hirsute group and adjustment only for free androgen levels did not change the mean CIMT values between groups. High serum androgen levels with regular cycles have found to cause less detoriation in metabolic factors than menstrual irregularity without hirsutism.

Polycystic ovary syndrome, the commonest endocrine disorder of women, is currently emerging as a potential facet of the metabolic syndrome (MBS) in women. Available data suggest that the MBS or, alternatively, individual metabolic risk factors may be overly present and most importantly that MBS may arise at a significantly younger age among PCOS women. The concept that a conventionally considered reproductive disorder may entail a significant metabolic impact on affected women has warranted medical interest on the mechanisms underlying the multiplicative sequelae of PCOS. Although obesity indisputably compounds the clinical course of women with PCOS, this appears to be just the tip of the iceberg. Insulin resistance and hyperinsulinemia have been intuitively involved as a critical link due to their contribution to the pathophysiology and clinical presentation of both PCOS and MBS. Hyperandrogenemia, the predominant endocrine hallmark of PCOS, has also been implicated as a contributing factor to the suggested interrelationship.[21]

Hypoandrogenemia in men and hyperandrogenemia in women are associated with increased risk of coronary artery disease but also with visceral obesity, insulin resistance, low HDL, elevated TG, LDL and plasminogen activator inhibitor (PAI-1). Gender differences and confounders render the precise role of endogenous androgens in atherosclerosis unclear.[22] Young women with chronic hypoestrogenism or hyperandrogenism have plasma concentrations of homocysteine similar to those of healthy women with normal menstrual cycles.[23] Correction of hyperandrogenemia by laparoscopic ovarian cautery in women with polycystic ovarian syndrome is not accompanied by improved insulin sensitivity or lipid-lipoprotein levels.[24]

Consistent to previous studies in this study no association was found between hyperandrogenemia and CVD risk.This hyperandrogenemia included both clinical hyperandrogenemia and hyperandrogenemia confirmed by free testosteron level.

Obesity and insulin resistance are both powerful predictors of CVD risk, and insulin resistance at any given degree of obesity accentuates the risk of CVD and type 2 diabetes.[25]

Of the metabolic syndrome components, hyperglycemia in younger subjects and hypertension in elderly subjects were major factors for ischemic ECG changes, whereas hypertriglyceridemia was not an independent risk factor in any age group.[26] The data suggest that improved glycaemic control has the potential to significantly reduce the risk of micro-and macrovascular disease when instigated early in the disease course. However, in more advanced diabetes, the benefits of improved control appear to be less evident.[27]

In our study population fasting glucose levels were all within normal limits and body mass indices were low, thus we have not found any association between increased HOMA-IR and CIMT. These results led us to hypothesize that normoglycemia with increased insulin levels in early ages of PCOS with normal BMI does not have major impact on increased CVD risk.

In a study by Rickenlund et al the amenorrheic athletes were found to have the most unfavorable lipid profile with significantly higher total cholesterol and LDL, compared with the other athlete groups. The oligomenorrheic athletes had the lowest levels of total cholesterol, low-density lipoprotein, and apolipoprotein B of all groups and significantly lower in comparison with the amenorrheic group. However, with respect to FMD, the oligomenorrheic group represented an intermediate between amenorrheic and regularly cycling subjects. There was a gradual impairment of FMD and the lipid profile to the degree of menstrual disturbance supporting an association with estrogen status. It was concluded that amenorrhea in young endurance athletes is associated with endothelial dysfunction and unfavorable lipid profile.[28]

It is worth noting that data from the Finnish Study involving 1257 healthy men >42 years of age showed that each increase in IMT of 0.1 mm resulted in an 11% increase in the risk of myocardial infarction.[29] When young age of study population is considered 0.1 mm difference of CIMT between groups after adjustment for other risk factors may lead those oligoamenorrheic PCOS patients to higher risk of atherosclerosis.

Recently published study concluded that, for determining the degree of cycle irregularity as a simple clinical parameter might be a valuable instrument to estimate the degree of metabolic and endocrine disorders. Emphasis should be given to those parameters as a first step to characterize PCOS patients with a risk of endocrine and metabolic disorders leading to

consequent detailed examination.[30] Relatively large number of study population and comparison of peer selected PCOS patients to determine the real impact of individual complaint or labarotory findings were major adventages of this study. CIMT values were under cut offs that previously reported, this was disadvantage of this study and it was thought to be due to young age study population. Previous studies concerning metabolic disorders among different PCOS phenotypes compared labile factors like lipid profiles or homocysteine levels, this was major limitation of the studies.[31,18] We have used a valuable stable tool to determine CVD risk in PCOS supported by other labile metabolic factors.[17]

The result of this study shown that menstrual abnormality of oligo-amenorrhea has major role determining the CVD risk in early age PCOS patients.

Oligo-Amenoreik Polikistik Over Hastaları Hirşut ve Sağlıklı Kontrol Grubuna Kıyasla Daha Fazla Kardiyovasküler Hastalık Riski Taşır

AMAÇ: Hirşutizm veya oligo-amenore şikayeti olan polikistik over hastaların klinik, kardiyovasküler ve endokrin profil farklarının değerlendirilmesi.

GEREÇ VE YÖNTEM: Kasım 2009 ve Haziran 2010 tarihleri arasında Dr. Sami Ulus Kadın Hastalıkları Eğitim ve Araştırma Hastanesinde incelenen 129 hasta çalışmaya alındı. Hirşütizm şikayeti olan 48 hasta vardı, oligo-amenore şikayeti olan 49 hasta ve 32 sağlıklı kontrol kıyaslanan gruba dahil edildi. Vücut kitle indeksleri (VKİ), bel-kalça oranı (bel/kal), folikül stimulan hormon (FSH), luteinizan hormon (LH), progesteron, serbest testosteron, estrodiol, düşük dansiteli lipoprotein (LDL), açlık glukoz, yüksek dansiteli lipoprotein (HDL), trigliserid (TG), total kolesterol, insülin, insülin sensitivitesi, sensitif C- reaktif protein (hs-CRP), karotid intima media kalınlığı (CIMT) değerleri hirşut, oligoamenoreik ve kontrol gruplarında kıyaslandı.

BULGULAR: Yaş ortalamaları benzerdi. Gruplar arasında VKİ, bel/kal, total kolesterol, LDL, TG, LH, açlık glukozu, hsCRP, CIMT değerleri bakımından fark saptandı. ($p<0,05$, tablo1). VKİ, bel/kal, total kolesterol, LDL, TG, LH, estrodiol, açlık glukoza göre düzeltilmiş ortalama CIMT değerleri hirsut, oligoamenoreik ve kontrol gruplarında sırası ile 0,40±0,01 mm, 0,49±0,012mm, 0,34±0,012mm idi($p<0,001$).

SONUÇ: Oligo-amenoresi olup hirsurizmi olmayan polikistik over hastaları, hirşütizmi olup oligo-amenoresi olmayanlara ve sağlıklıklı kontrol grubuna kıyasla daha fazla anormal lipid profiline ve kardiyovasküler riske sahiptir.

Anahtar Kelimeler: Polikistik over sendromu, Karotid intima media kalınlığı, Rotterdam kriterleri

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