Procalcitonin as a Mediator of Chronic Inflammation in Obese Women with PCOS

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Abstract

Background: Polycystic ovary syndrome (PCOS), a common reproductive endocrine condition characterized by hyperandrogenism, chronic anovulation, and obesity. Obesity is associated with a low-grade inflammation of white adipose tissue resulting from chronic activation of the innate immune system and can subsequently lead to insulin resistance, impaired glucose tolerance and even diabetes. In the last few years, adipose tissue emerged as an important source of proinflammatory mediators including TNF-α, IL-6, and procalcitonin.

Objective: to investigate procalcitonin as a marker of chronic inflammation in obese women with PCOS.

Method: a case control study conducted from January 2010 till July 2010. The study involved 20 women with PCOS and 20 control women matched for age and BMI. Waist to hip ratio was measured and blood was drawn from patients and controls and serum levels of FSH, LH, prolactin, testosterone and procalcitonin were estimated by VIDAS using VIDAS kits provided by Biomerieux (France). Student t test was used to evaluate the difference in serum procalcitonin level between the two groups.

Results: compared with control group, patients with PCOS had a significantly higher waist to hip ratio (mean 0.95 ± 0.2 versus 0.8 ± 0.13). Serum procalcitonin level was significantly higher in women with PCOS compared to control group (1.43 ± 0.42 ng/mL Vs 0.19 ± 0.13 ng/mL) P value was < 0.0001. Serum levels of FSH, LH, prolactin and testosterone all were significantly higher in women with PCOS than in the control group (P value < 0.05).

Conclusion: The increase in low-grade chronic inflammation in women with PCOS is primarily associated with increased central fat excess. Procalcitonin represents a novel marker of the inflammatory activity of body fat in PCOS.

Abbreviations

PCOS	Polycystic Ovary Syndrome
TNF-α	Tumour Necrosis Factor-alpha
IL-6	InterLeukine- 6
FSH	Follicle Stimulating Hormone
LH	Leutenising Hormone
mm	millimeter
mmHg	Millimeter mercury
C	Centigrade
Ab	Antibody
Ag	Antigen

الخلاصة:

اشتملت الدراسة على عشرين مريضة تعاني من متلازمة تكيس المبايض و عشرين امرأة طبيعية من نفس الفئة العمرية و لهن أوزان متقاربة. بعد اخذ التاريخ المرضي و اجراء الفحص السريري تم سحب عينات الدم و قياس البروكالسيتونين. و هرمونات ال FSH, LH, prolactin , التستوستيرون

كانت نسبة محيط الخصر: الورك أعلى عند المريضات مقارنة بالنساء الطبيعيات على الرغم من تقارب اوزان اجسامهن مما يدل على تكدس الشحوم في منطقة الجذع . كانت مستويات الهرمونات اعلى عند المريضات و كذلك البروكالسيتونين والذي يعتبرمؤشر لوجود نشاط التهابي في الجسم و الذي بدوره يشكل عامل خطورة للاصابة بأمراض القلب وجهاز الدوران.

Introduction

POLYCYSTIC OVARY SYNDROME (PCOS) is one of the most frequent endocrine disorders, affecting 5–10% of young women [1]. In these patients, an increase in insulin resistance and in central body fat accumulation has been observed independent of obesity [1- 4]. PCOS cases exhibit an adverse coronary heart disease (CHD) profile at an early age, including insulin resistance, dyslipidemia and increased central adiposity [5].

Low-grade chronic inflammation of white adipose tissue WAT, reflected by an increase in highly sensitive serum C-reactive protein (hs-CRP) is closely linked to insulin resistance, to central obesity, and to an increase in cardiovascular risk [6]. WAT is the physiological site of energy storage as lipids. In addition, it has been more recently recognized as an active participant in numerous physiological and pathophysiological processes.

In obesity, WAT is characterized by an increased production and secretion of a wide range of inflammatory molecules including TNF-alpha and interleukin-6 (IL-6), which may have local effects on WAT physiology but also systemic effects on other organs. Recent data indicate that obese WAT is infiltrated by macrophages, which may be a major source of locally-produced pro-inflammatory cytokines. Interestingly, weight loss is associated with a reduction in the macrophage infiltration of WAT and an improvement of the inflammatory profile of gene expression. Several factors derived not only from adipocytes but also from infiltrated macrophages probably contribute to the pathogenesis of insulin resistance. Most of them are overproduced during obesity, including leptin, TNF-alpha, IL-6 and resistin. Conversely, expression and plasma levels of adiponectin, an insulin-sensitising effector, are down-regulated during obesity. Leptin could modulate TNF-alpha production and macrophage activation. TNF-alpha is overproduced in adipose tissue of several rodent models of obesity and has an important role in the pathogenesis of insulin resistance in these species. However, its actual involvement in glucose metabolism disorders in humans remains controversial. IL-6 production by human adipose tissue increases during obesity. It may induce hepatic CRP synthesis and may promote the onset of cardiovascular complications [7]

In patients with PCOS, circulating levels of TNF-α, IL-6, and hs-CRP as well as white blood cell count (WBC) and neutrophil count have been found to be elevated compared with age and/or body mass index (BMI)-matched controls [8-10].

When comparing patients with PCOS with controls, most previous studies adjusted differences in serum inflammatory markers for BMI or total fat mass. However, the impact of body fat distribution, *i.e.* the central accumulation of body fat, on these markers has never been assessed previously.

Waist to Hip Ratio

Waist-to-hip ratio (WHR) is a way of determining body shape (apple/pear) and thus weight-related health risks.

Apple-Shape or Pear-Shape:

Most people store their body fat in two distinct ways: around the middle (apple shape), or around the hips (pear shape).

For most people, being an apple shape (carrying extra fat around the abdomen) places them in a higher health risk category than being a pear shape (carrying extra weight around the hips or thighs). Other important measurements are Body Mass Index (BMI), Body Fat Percentage and Waist to hip ratio [11].

Aim of study

To assess serum procalcitonin as an inflammatory marker in patients with PCOS.

Patients and Method

We consecutively recruited 20 patients diagnosed with PCOS who presented to our clinic and met the inclusion criteria. Twenty control women with regular menses were simultaneously recruited, with an attempt to match for BMI and age.

The diagnosis of PCOS depended on the definition generated at the 2003 Rotterdam Sponsored PCOS Consensus Workshop, which concluded that PCOS is a syndrome of ovarian dysfunction along with cardinal features of hyperandrogenism and polycystic ovary morphology and was based on a history of oligomenorrhea, oligoovulation and/or hirsutism with or with out ultrasound evidence of polycystic ovaries (eight or more subcapsular follicular cyst ≤ 10 mm in diameter and increased ovarian stroma.) [12]. None of the subjects had diabetes or evidence of cardiovascular disease, or infection and blood pressure was less than 140/90 mm Hg in all participants on screening examination. Subjects were screened by medical history and examination. Body weight and height were measured and BMI calculated. Waist to hip ratio was measured for all subjects. Blood samples were drawn on cycle day two in those with spontaneous menstrual cycles and on cycle day three of withdrawal bleeding induced by 10 mg daily of norethisterone acetate tablets for five days in those with amenorrhea or oligomenorrhea. Blood samples were immediately centrifuged, and serum was aliquoted and stored at –20 C until batch analyzed. FSH, LH, prolactin, testosterone and procalcitonin were estimated by VIDAS using VIDAS kits provided by Biomerieux (France).

Assay: The antigen is first captured by antibody present on solid phase. Enzyme linked antibody (conjugate) is then added making a sandwich like. Substrate that yields a fluorescent product on enzyme action is then added. The amount of light emitted from fluorescent product is directly proportional to the concentration of antigen present in the sample.

Ab + Ag + Enzyme linked Ab (conjugate) + fluorescent substrate

Statistical Analysis: For statistical analysis SPSS (version 10) program was used. The results were represented through mean and standard deviation. Student t-test used to compare between means and study the significance of the difference. P value <0.05 was considered to be statistically significant.

Results

Table (1) shows the demographic characteristics of patients with PCOS and the control subjects. Age and BMI were comparable between the two groups waist to hip ratio was significantly higher in PCOS patients (0.95 ± 0.2 Vs 0.8 ± 0.13) and the mean period of infertility was 2.6 years in these women.

Table 1 Demographic characteristics of the patients and controls.

character	PCOS patients (n=20) (mean ± SD)	Control (n=20) (mean ± SD)	P value
Age (yr)	27± 4.7	28.4± 3.7	0.3
BMI (kg/m^2)	28.4± 5.6	27.7 ± 3.8	0.41
Waist-hip ratio	0.95 ± 0.2	0.8 ± 0.13	0.001
Period of infertility	2.6 ± 1.6	1.6 ± 0.4	0.0001

Table (2) shows a comparison of the serum levels of FSH, LH, prolactin and testosterone between PCOS patients and controls. All of them were higher in patients than controls and by using student T test, the difference was statistically significant (P value< 0.05) .

Table 2 Hormonal assay of PCOS patients and control groups

Hormone	PCOS (n=20) (mean ± SD)	Control (n=20) (mean ± SD)	P value
FSH (mIU/ml)	5.25±2.5	6.8±2.8	0.037*
LH (mIU/ml)	9.1±4.3	2.52±0.98	0.0001*
Prolactin (ng/ml)	25.4±15.00	12.3±4.6	0.001*
Testosterone (ng/ml)	0.72±0.21	0.41±0.21	0.0001*

*significant P value < 0.05

Table (3) shows the difference in serum procalcitonin level in patients with PCOS and control subject. The level was significantly higher in PCOS patients (1.43± 0.42 Vs 0.19 ± 0.13). P value = 0.0001.

Table 3 Comparison of serum procalcitonin level in PCOS patients and control subjects.

Subjects	No.	Mean procalcitonin level (pg/ml)	SD(±)	Standard error of the mean	df	t	P value
PCOS patients	20	1.43	0.42	0.0094	19	14.95	0.0001*
control	20	0.19	0.13	0.0030	19		

*significant P value < 0.05

Discussion

Subclinical inflammation and insulin resistance are important predictors of cardiovascular disease [13]. In agreement with previous studies done by Kirchengast et.al and Ibanez et.al, [2, 3], we found that patients with PCOS had an excess of

central fat independent of total fat mass reflected by higher waist to hip ratio when compared to normal subjects matched for BMI. Central fat excess is usually associated with an increase in serum inflammatory markers and in insulin resistance [14].

In our study we found that FSH, LH, prolactin and testosterone are all higher in patients with PCOS than in the control subjects This was in accordance with a study done by Li et.al, who found that FSH, LH, prolactin and testosterone were higher in obese women with PCOS when compared with non-obese PCOS patients and normal control subjects [15].

Also we found that serum procalcitonin was significantly higher in obese PCOS patients with central fat excess when compared to control subjects matched for BMI but with normal waist to hip ratio.

This result was similar to that found in a study done by Pudder et.al who measured procalcitonin and other inflammatory markers like TNF-α, C-reactive protein and white blood cells and found them to be significantly higher in obese women with PCOS compared to control group [16].

Another study done by Samy et.al. who investigated hormonal assay, lipid profile and inflammatory markers in obese and non-obese PCOS patients and compared them to control normal subjects, found that increased levels of testosterone, luteinizing hormone (LH), androstendione and insulin compared to healthy BMI matched controls. High-density lipoprotein (HDL) concentrations were significantly reduced in both patient groups compared to their controls, while triglyceride levels were significantly increased in obese group compared to controls. There is a significant increase in these markers when comparing obese PCOS patients and their matched controls [17].

Conclusion and Recommendations

PCOS and obesity induce an increase in serum inflammatory cardiovascular risk markers. The precise mechanisms underlying these associations require additional studies to clarify the state of the cardiovascular system in women with PCOS compared with controls in large numbers of patients to determine the relative contribution of different factors including insulin resistance, androgen status and BMI.

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