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Research Article

Assessment and Association between Lipid and Hormonal Profile in Non-Pregnant Females Having Polycystic Ovarian Syndrome

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ABSTRACT

Introduction: PCOS is a gynecological endocrine disorder with ovarian dysfunction, hyperandrogenism, infertility, hirsutism, acne, dyslipidemia, and menstrual irregularities.

Objectives: The aim of our study is to evaluate the fluctuations of hormonal profile regarding PCOS in fasting and random state as well as the possible risk factor of cardiovascular disorders towards PCOS.

Method: Blood was taken by CSLI (Clinical and Laboratory Standards Institution) procedure. Merk and Roach diagnostic systems were used to analyze lipid and hormonal profiles.

Results: Significant differences were observed in cholesterol (p=0.004), HDL, LDL (p<0.001) in fasting PCOS *vs.* control, LDL, HDL (p<0.001), cholesterol (p=0.08), lipid profile ratios in fasting *vs.* random PCOS, FSH (p=0.01), Progesterone (p=0.000), Estradiol (p=0.000), LH/FSH (p=0.04) in fasting PCOS *vs.* control, LH (p=0.04) in fasting *vs.* random PCOS. Significant positive correlation was found among estradiol with cholesterol (r=0.376, p=0.08), LDL (r=0.39, p=0.006) in fasting PCOS, estradiol with cholesterol (r=-0.334, p=0.02), FSH with LDL (r=0.36, p=0.01), progesterone with HDL (r=0.338, p=0.02) in random PCOS.

Conclusion: This study assesses the worth of lipid profile in PCOS towards cardiovascular risk factors because lipid and hormonal profile have a direct correlation with PCOS. Besides CVDs hormonal profile fluctuates frequently in random and fasting state. To diagnose treat PCOS properly, patients should be monitored in the fasting state.

Keywords: Fasting PCOS; Cardiovascular risk factor; Lipid profile; Estradiol; Testosterone

INTRODUCTION

Polycystic Ovarian Syndrome (PCOS) is basically an endocrine disorder in females of reproductive age. Menstrual irregularities and infertility lead to the PCOS. Hormonal and genetic factors participate in the pathogenicity of PCOS. Low progesterone levels result in more formation of estrogen that produces different autoantibodies in PCOS [1]. Deepthi et al. [2] illustrated a strong evidence of developing Cardiovascular Diseases (CVDs) in PCOS by analysing various biochemical parameters such as blood sugar, lipid profiles and hormones such as Follicle Stimulating Hormone (FSH) and Luteinizing Hormone (LH) from the sera and suggested a lifestyle modification (increasing physical exercise) and dietary control to reduce the risk factor of CVDs.

PCOS is a gynaecological, endocrine and metabolic disorder with ovarian dysfunction. About 6%-7% of premenopausal females are affected by this syndrome characterized by ovulatory dysfunction, hyperandrogenism, ovarian cysts and subfertility or infertility hirsutism, acne, and alopecia due to high circulating levels of androgens, menstrual irregularities [3-11].

Low HDL and high TG have been seen in PCOS females [12]. Normal healthy postmenopausal females have increased risk factors for cardiovascular disorders. In PCOS, LDL, BMI, insulin resistance, blood pressure, and waist-to-hip ratio is increased and HDL is decreased. In case of healthy postmenopausal females, ovulatory dysfunction and estradiol is decreased while blood pressure, cholesterol, adipose tissues, and weight is increased that leads to CVDs [13-15].

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Lipid disturbance and insulin resistance are higher in PCOS women as compared to the normal women that lead to atherosclerosis and cardiovascular diseases, this abnormality may affect 60%–80% of women with PCOS and, 95% of obese women with this syndrome [16-18].

PCOS females have higher LDL, cholesterol, triglycerides and HDL level regardless of obesity. Obesity does not affect much on PCOS but high HDL level is a threat towards CVDS [19].

MATERIALS AND METHODS

Subject

One hundred volunteers PCOS patients were selected from Nishtar hospital that came to follow up on the basis of Rotterdam criteria and ultrasound diagnosis. After that their data according to the fasting and random values were sorted out in two groups (fasting and random group) besides the control group was also included in this study. The study samples and the control group were ranged from 18-35 years while the mean ± standard deviations for fasting and random study sample groups were 27.13 ± 5.72 and 25.58 ± 5.46 respectively. Every individual was clinically fit from any viral infection/pathology that was included in the study.

Study area

The study was done in MINAR, Nishtar Hospital, Multan, and South Punjab, Pakistan, which provides the medical facility in southern districts of Punjab province. Its geographical location is latitude 30°9'26.8488" N, longitude 71°31' 29.694" E and 129 m high from sea level.

Inclusion/exclusion criteria

The adult's female that were apparently healthy PCOS with Acne, obesity, hirsutism, oligomenorrhea, amenorrhea, and passive smokers were included in this study. Active Smokers, alcoholic, drug-addicted and pregnant subjects were excluded.

Sampling

A single expert phlebotomist drew 3 mL blood from the subject's specimen by venipuncture with BD syringes precision glide needle 23 G 1 TW (0.5×25 mm), (lot no. 6130882, Becton, Dickinson Company, made in Singapore) directly into plain Red Top BD vacutainer (4.0 mL Becton Drive, Franklin Lakes, NJ 07417, USA, lot no. 8130999) with protective glove and under appropriate safety measures, following the guideline of International Standard by Clinical Laboratory Standard Institute and WADA [20,21]. Blood samples were kept at room temperature for proper clotting and, centrifuged at automatic refrigerated centrifuge machine (Hitachi). Serum separated from the samples was stored at -20°C.

Laboratory testing

Lipid profile (cholesterol, triglycerides, LDL, HDL) analysis of PCOS was performed on automatic chemistry analyzer Junior Selectra by Merck. Hormonal profile (LH, FSH, estradiol, testosterone, and progesterone) was done on COBAS machine, which works chemiluminescence technique [20-24]. Appropriate calibration and quality control were run and confirmed their quality control results before performing the test batch using the recommendation of manufacturing [24,25].

Statistical analysis

Statistical analysis was done in Microsoft Excel and verified from IBM SPSS-4. Mean ± SD of lipid and the hormonal profile was measured on the excel sheet. A paired t-test was applied to normal distribution parameters and Wilcoxon rank sum for non-normal distribution parameter after the normality/non-normality distribution analysis by D'Agostino-Pearson test. Pearson correlation for normal distribution and Spearman correlation for non-normal distribution between lipid profile and hormonal profile parameters were established. The significance level was set at $p<0.05$.

RESULTS

The results of our study were calculated on the basis of fasting and random levels. In Table 1, comparison of fasting PCOS *vs.* control, random PCOS *vs.* control, fasting *vs.* random PCOS and ratios of Cho/HDL, Tg/HDL and HDL/LDL for fasting and random PCOS were summarized. Significant results were assessed for cholesterol (p=0.04), LDL (p=0.00), HDL (p=0.00) in fasting PCOS *vs.* control while no significant results were recorded in random PCOS *vs.* control group. When we compared the parameters of fasting and random PCOS, only cholesterol (p=0.008) showed a significant result.

Significant results between the ratios of Cho/HDL (p=0.01), Tg/HDL (p=0.00) and LDL/HDL (p=0.00) in fasting PCOS *vs.* Control was recorded. No significant results of ratios were found in random PCOS *vs.* control. When we compared the ratios of fasting *vs.* random PCOS, only Cho/HDL showed significant variation (p=0.005).

When compared the hormonal level of fasting PCOS *vs.* control, FSH (p=0.01), estradiol (p=0.000), progesterone (p=0.000), LH/FSH (p=0.04) were noted significant variations. Only significant results of LH (p=0.04) was found in fasting *vs.* random PCOS.

In Table 2, significant positive correlation in fasting PCOS were found in estradiol with cholesterol and LDL, (p=0.008, r=0.376) and (p=0.006, r=0.39) respectively. Estradiol with cholesterol (p=0.02, r= -0.0334), FSH with LDL (p=0.01, r=0.36) and progesterone with HDL (p=0.02, r=0.338) were noted significant correlation in random PCOS.

DISCUSSION

The polycystic ovarian syndrome is a gynecological and endocrine disorder frequently occurs in the 5%-10% of premenopausal females characterized by obesity, hyperandrogenism, insulin resistance, menstrual irregularities, hirsutism and acne [26]. It is an undefined and multidimensional disorder based on environmental and genetic factors firstly prescribed by Stein and Leventhal in 1935 [3,27]. PCOS females have broader ovaries containing follicles situating in either one or both ovaries. In this condition, the hormones become imbalance which is more to be seen nowadays than earlier females due to less physical activities. Inherited and having diabetic family history, females are susceptible to PCOS that further develop insulin resistance, affects FSH, LH level and menstrual irregularities [28-33].

We have included both fasting and random PCOS and our study showed significant results of Cholesterol, LDL, HDL, Cho/HDL, Tg/HDL and HDL/LDL in fasting PCOS, cholesterol and Cho/HDL in random PCOS similar to the work done by Talbott [34].

Table 1: Comprehensive Results of lipid and hormonal profile of PCOS and control.

Parameters (lipid & hormonal profile)	Fasting PCOS Mean ± Std	Random PCOS mean ± Std	Fasting Control Mean ± Std	Random Control	Fasting PCOS *vs.* Control p Value	Random *vs.* Control p Value	Fasting *vs.* Random PCOS p Value
Cholesterol[a]	173.5 ± 46.57	210.11 ± 64.85	157.3 ± 26.22	147.88 ± 30.15	0.004*	0.108	0.008
Tg[a]	158.7 ± 56.60	181.64 ± 80.15	167.8 ± 15.30	173.67 ± 12.62	0.29	0.04	0.12
HDL[b]	77.56 ± 35.07	76.66 ± 51.55	54.52 ± 5.13	54.92 ± 4.77	0.000*	0.889	0.480
LDL[a]	107.10 ± 43.03	103.94 ± 47.25	170.29 ± 19.15	171.18 ± 19.23	0.000*	0.83	0.75
Cho/HDL ratio	2.64 ± 1.47	3.81 ± 2.34	2.90 ± 0.535	2.71 ± 0.62	0.01*	0.14	0.005*
Tg/HDL ratio	2.39 ± 1.34	3.28 ± 2.31	3.10 ± 0.38	3.18 ± 0.34	0.000*	0.294	0.051*
HDL/LDL ratio	0.869 ± 0.771	0.899 ± 0.651	0.322 ± 0.03	0.322 ± 0.02	0.000*	0.797	0.434
LH[a]	9.03 ± 8.50	11.95 ± 11.81	6.69 ± 3.03	8.28 ± 6.29	0.18	0.21	0.04*
FSH[b]	5.81 ± 6.91	5.36 ± 4.03	6.00 ± 2.08	6.97 ± 2.75	0.01*	0.13	0.29
Testosterone[b]	46.31 ± 50.74	36.25±38.09	23.56 ± 9.56	27.91 ± 12.04	0.06	0.07	0.73
Estradiol[b]	125.70 ± 100.74	138.05 ± 119.09	278.20 ± 130.76	292.12 ± 182.32	0.000*	0.93	0.83
Progesterone[b]	1.61 ± 2.58	1.49 ± 2.35	1.64 ± 0.72	1.46 ± 0.72	0.000*	0.23	1.000
LH/ FSH	1.85 ± 1.06	2.12 ± 0.84	1.27 ± 0.76	1.41 ± 1.37	0.04*	0.93	0.08

Impression: [a]Normal distribution, [b]Non-Normal, *significant value <0.05

Table 2: Correlation of lipid and hormonal profile fasting *vs.* random PCOS.

Parameters	Random group (Hormonal Profile) p value Summary					
Lipid Profile	**LH[a]**	**FSH[b]**	**Estradiol[b]**	**Testosterone[b]**	**Progesterone[b]**	**LH/FSH**
Cholesterol[a]	r=0.049, p=0.75	0.004, 0.97	r=-0.0334, p=0.02**	-0.26, 0.868	0.088, 0.57	-
Triglycerides[a]	0.213, 0.166	0.197, 0.201	0.137, 0.377	0.248, 0.105	0.253, 0.09	-
LDL[a]	0.22, 0.134	r=0.36, p= 0.01**	-0.134, 0.387	-0.043, 0.781	-0.188, 0.222	-
HDL[b]	-0.034, 0.82	-0.152, 0.323	-0.09, 0.56	-0.05, 0.743	r=0.338, p= 0.02**	-
Cho/HDL	-	-	-	-	-	-0.039, 0.801
Tg/HDL	-	-	-	-	-	0.08, 0.591
HDL/LDL	-	-	-	-	-	r=0.004, p=0.97
	Fasting Group					
Cholesterol[a]	-0.02, 0.84	-0.24, 0.08	r=0.376, p=0.008**	0.05, 0.70	-0.135, 0.36	-
Triglycerides[a]	-0.114, 0.44	0.239, 0.102	0.126, 0.392	0.041, 0.78	0.142, 0.337	-
LDL[a]	-0.150, 0.30	0.144, 0.32	r=0.39, p=0.006**	0.034, 0.82	-0.127, 0.38	-
HDL[b]	-0.097, 0.51	r=0.05, p=0.70	0.232, 0.113	-0.095, 0.52	-0.082, 0.51	-
Cho/HDL	-	-	-	-	-	0.086, 0.56
Tg/HDL	-	-	-	-	-	-0.113, 0.444
HDL/LDL	-	-	-	-	-	0.001, 0.994

Impression: [a]Normal distribution and Pearson correlation, [b]Non-Normal and spearman correlation, significant value <0.05*
(p-value represents significance from Pearson correlation statistical analysis).
*Correlation is significant at the <0.05 level (2 tailed).
**Correlation is significant at the <0.01 level (2 tailed).

According to Framingham, HDL, LDL levels and Cho/HDL ratios may lead to coronary artery disease [35-41]. In order to evaluate cardiac risk in PCOS, we have calculated the Cho/HDL, Tg/HDL, HDL/LDL ratios and compare them at fasting and random levels. Ideal values of Cho/HDL and TG/HDL ratios are <3.5 and <2 respectively while present study reported that the mean values of Cho/HDL and TG/HDL ratios were <3.5 and>2 which shows that they are at higher risk of atherosclerotic vascular disease. The lipid profile concentration of females PCOS has higher androgen hormone with an increased risk of atherosclerosis [41]. Moreover, our study also proved by Framingham experience that predicts the link between LDL, HDL, triglycerides, cholesterol levels, Cho/HDL ratios, and cardiovascular disease [37-42].

Most women with PCOS have hyperandrogenemia, elevated Luteinizing Hormone (LH), and normal or decreased Follicle-Stimulating Hormone (FSH) [43]. Increased LH level slows down fertilization as well as gestational chances. There is a direct correlation of elevated LH level and infertility both in healthy women and also in Polycystic Ovary Syndrome (PCOS) [44-48].

Robert A. Wild in 1992 study the mean and std. of LH/FSH ratio of PCOS and control was 2.0 ± 1.0 and 0.6 ± 0.1 while our study shows that the mean ± SD values of LH/FSH ratio of PCOS in fasting and random levels were 1.85 ± 1.06 and 2.12 ± 0.84 respectively which is nearby to the above said PCOS levels.

Results were similar to our investigation showed a significant

variation FSH, estradiol, progesterone and LH/FSH on fasting PCOS and control comparison and LH showed significant difference on fasting *vs.* random PCOS comparison [48,49].

Lipid profile concentrations are sensitive towards estradiol. Estradiol is responsible for the fluctuation of lipid profile concentration. It decreases LDL concentration and increases cholesterol, HDL and triglycerides [50]. A positive correlation of estradiol with cholesterol and LDL that efficiently leads to cardiovascular diseases while positive correlation has been seen in serum leptin level with estradiol level in PCOS patients [51]. Because our study shows a more significant correlation between lipid and hormonal profile so PCOS parameters should be monitored under fasting conditions for cardiac and hormonal diagnosis. In most of the laboratories, there is a general practice of taking blood sample related to hormonal profile to assess randomly (fasting or random) but we have analyzed in MINAR labs that sample should be taken at fasting level because lipid profile has a direct relationship with a hormonal profile in PCOS that leads to CVDs.

CONCLUSION

Lipid profile level usually elevates in PCOS that would be the cause of cardiovascular disorders. In hormonal profile, LH level becomes double as compare to FSH level in PCOS. This fluctuation of LH in response to FSH more clearly is seen at fasting state. Our study also confirms that PCOS parameters (lipid and hormonal profile) should be monitored under fasting state for cardiac and hormonal diagnosis. Further study should be carried out for evaluation of above-said results.

CONFLICTS OF INTEREST

No conflict of interest has been declared.

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