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HEART RATE VARIABILITY AND LIPID PROFILE IN NON OBESE YOUNG INDIAN WOMEN WITH POLYCYSTIC OVARY SYNDROME

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ABSTRACT: CONTEXT (BACKGROUND): Polycystic ovary syndrome (PCOS) is one of the most common endocrinopathy of premenopausal women and is a most frequent cause of infertility. There are more stringent reports of cardiovascular events and infrequent investigations of lipid levels in lean PCOS. **AIMS:** To assess the cardiac autonomicity using Heart Rate Variability (HRV) parameters and metabolic risk with serum lipid parameters among ideal and lean weight PCOS patients. Settings and Design: case control study. 24 classical PCOS diagnosed by Rotterdam 2003 Diagnostic criteria and were ideal and lean as per WHO criteria and 24 BMI matched, age matched normally menstruating women served as study participants. Methods and Material: 5 min of Electrocardiogram (ECG) was taken which was evaluated for HRV. Power spectral analysis was done to calculate the time and frequency domain measures of HRV. Fasting serum lipid profile was done. Statistical analysis: Independent sample't' test was used. **RESULTS:** The results of the present study showed that cardiac autonomic innervations can be affected in lean and ideal weight PCOS with increased sympathetic and decreased parasympathetic components of HRV. As a result, sympathetic to parasympathetic ratio may increase in PCOS. Fasting blood sugar was also increased. Of the lipid parameters total cholesterol and LDL-C was increased. Other parameters were not altered. **CONCLUSION:** This study gives a solid and strong evidence of altered cardiac autonomic activity and unfavorable metabolic profile, which are the important risk factors for cardiovascular disease even in lean and ideal weight PCOS. Our data suggest that all PCOS patients should undergo periodic screening for normal cardiac activity and lipid profiles irrespective of obesity.
KEYWORDS: Polycystic Ovarian Syndrome, Heart rate variability, Lean and ideal weight patients, metabolic profile, cardiac autonomic activity.

INTRODUCTION: Polycystic ovary syndrome (PCOS) was first described by Stein Leventhal in 1935 and became one of the most common endocrinopathy of premenopausal women, with a prevalence estimated at approximately 25%[1] of the Indian population. PCOS was initially recognized as a clinical combination of anovulation and hyperandrogenism; now it appears to be a new face of metabolic syndrome.[2]

It is not only a reproductive endocrinopathy, but like the metabolic syndrome is associated with long term health risk including insulin resistance, diabetes mellitus, dyslipidemia, hypertension and premature atherosclerosis.[3] Cardiovascular diseases (CVD) remains the leading cause of death in women, and age remains one of the strongest risk factors for the development of atherosclerosis and death from a cardiovascular event. Whether CVD occurs at an earlier age in PCOS is not known, because rates of CVD are very low in premenopausal populations.

Several aspect of cardiovascular involvement has been investigated and many reported some abnormalities in PCOS patients compared to the controls. Although data regarding the cardiovascular

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mortality and morbidity are conflicting,[4] based on the prevalence of risk factors, PCOS patient have an estimated 4-11 fold increased risk for coronary artery disease,[5] high prevalence of diabetes (40%) and hypertension (60%).[6] The abnormalities include decreased cardiac systolic flow velocity,[7] diastolic dysfunction,[8] increased vascular stiffness,[9] endothelial dysfunction,[10] low grade chronic inflammation,[11] increased homocysteine,[12] impaired fibrinolysis[13] and increased tissue plasminogen activator antigen.[14] However, there is only one study reported so far investigating the cardiac autonomic functions in women with PCOS using heart rate variability (HRV) and no study was done in lean and ideal weight PCOS.

A heart rate variability measurement from electrocardiographic recording has been shown to be useful to assess cardiac autonomic function.[15] Although cardiac automaticity is intrinsic to various pacemaker tissues, heart rate and rhythm are largely under the control of the autonomic nervous system. The vagal and sympathetic activity constantly interact, giving rise to RR interval variations. HRV has the potential to provide additional valuable insight into physiological and pathological conditions and to enhance risk stratification. More importantly a reduction of HRV has been reported in several cardiological and non cardiological diseases.[15]

Another independent risk factor for cardiovascular morbidity is alterations in serum lipid levels. Investigations of lipid levels in PCOS are infrequent.[16] There are relative controversies regarding the lipid abnormalities in this group of women. Some studies report only negligible or no effect on lipids in women with PCOS.[17] Hence a substantial portion of women with PCOS may have a completely normal circulating lipid profile or an abnormal lipid profile. May be, the discrepancies could be explained by presence or absence of obesity.

As there is no study reported so far on cardiac autonomic functions evaluated by HRV in young Indian women with PCOS and as there is little published evidence to support that women with PCOS are unduly affected, we determined to study the HRV parameters and metabolic profile in Indian women with PCOS. Therefore our aim was to investigate the cardiac autonomic innervation using HRV parameters and metabolic profiles in lean and ideal weight PCOS patient and to compare them with regularly menstruating apparently healthy controls.

We hypothesized that the PCOS are associated with decreased heart rate variability (HRV) and high risk lipid profile.

MATERIALS AND METHODS: The study was conducted in the department of physiology, PSG IMS&R, after getting clearance from the Institute's Human Ethics Committee and informed consent from both study and control groups. The study group consisted of women who presented to the infertility clinics, gynecologists and family physicians with complaints of dysfunctional uterine bleeding, or infertility and diagnosed to have PCOS by the experts. Diagnosis of PCOS was made with physical findings of hyperandrogenism, oligo / anovluation and ultrasonography, after exclusion of specific ovarian, adrenal and pituitary disorders, according to Rotterdam 2003 diagnostic criteria [18].

SUBJECTS:
  a. **Patient Group:** This group comprised of 24 non-pregnant ideal and lean weight (by BMI- body mass index) women with PCOS. The sample size was calculated according to the prevalence of the disease in India.[1] The required sample size calculated was 18. The number of patients studied were 25 among one was excluded because of presence of ectopic beats. The patients were categorised as lean and normal as per the WHO criteria.[19]

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b. **Control Group:** It consisted of 24 regularly menstruating (every 27-32days) volunteer medical students, doctors, nurses and staff of the hospital and who were matched for age and BMI.

Our inclusion criteria for both groups consisted of young women aged 16 to 35 years, women who are not on any medications affecting lipid or carbohydrate metabolism at least for past 2 months. Our exclusion criteria included, women below 16 and above 35 years, pregnant and lactating women, those who had undergone hysterectomy and had attained menopause, women taking lipid lowering drugs for the past 2 months, women on oral hypoglycemic drugs or insulin sensitizing agents, oral contraceptives and sex steroids for the past 2 months and those who were on current infertility treatment.

After selecting the study and control group according to the selection criteria, written informed consent was obtained. All the participants of the control group were in their follicular phase (6-8 days the start of menstruation) and PCOS were amenorrhaeic during data recording. Antecubital blood sample was taken after at least 12 hours fasting for the lipid profile. A detailed history taking and a complete physical examination was done and a complete record was obtained for future verification.

Each patient and control received a detailed clinical examination and underwent a relevant laboratory evaluation. The history focused on age, menstrual pattern, fertility status, duration and treatment of the problem and other relevant drug history. The physical examination, apart from a general review of the systems, focused on the anthropometry (Body mass index and waist circumference).

ANTHROPOMETRY:

a. Body mass Index (BMI): All the study participants were measured for height and weight (wt). Height was measured in centimeters (cms) as the study participants stood in their upright position using the Height measuring scale and the weight was measured using electronic weighing machine. From this BMI was obtained by dividing weight in kilogram (Kg) by square of the height (in meters). According to World Health Organization obesity is graded as underweight (BMI<18.5 kg/m2), normal (18.5–24.9 kg/m2), preobese (25.0–29.9 kg/m2), and class I obese (30.0–34.9 kg/m2).[19]

b. Waist Circumference: Waist circumference established by standard measures [20]. Measurement sites were obtained with the subject assuming a standing position and then the points were marked on the subjects. Waist circumference was measured half way between the lower border of the ribs and the iliac crest in the horizontal plane. 2 measurements to the nearest 0.5 cm were recorded. If the variation between the measurements were >2 cms a third measurements was taken. The mean of the 2 closest measurements was calculated. For females Waist circumference 80–87.9 cms was graded as overweight and ≥102 cms as obese. For the entire study population the BMI and Waist Circumference (WC) was obtained.

c. Resting blood Pressure: At the end of 20 minutes of quiet supine rest, blood pressure was recorded in the supine position, using a manual sphygmomanometer (a Novaphone make). Systolic and diastolic blood pressure was measured with a calibrated manometer from the right arm and recorded to the nearest 2mm Hg.

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d. Metabolic Profile: The fasting blood sample was taken to get the fasting blood glucose and fasting serum lipid profile and analysed using SEAC SLIM Auto Analyser, Italy. Blood glucose was determined using glucose oxidase method. Total cholesterol was determined using CHOD-POD i.e., cholesterol oxidase and peroxidase method (enzymatic colorimetric method). Triglycerides were determined by the method based on the enzymatic hydrolysis of serum or plasma triglycerides to glycerol and free fatty acids by lipoprotein lipase and then using glycerophosphate oxidase and peroxidase enzymes (enzymatic colorimetric method). High density lipoprotein cholesterol (HDL-C) was determined using cholesterol esterase method following selective precipitation of Apolipoproteins B containing lipoproteins with a polyanion solution. Low density lipoprotein cholesterol (LDL-C) was derived from Total cholesterol (TC) and triglycerides (TGL) values. Very low density lipoprotein cholesterol (VLDL-C) was derived from serum triglycerides values. TC/HDL-C was derived from serum total cholesterol and HDL-C values.

HRV Analysis: HRV analysis has now been increasingly recognized as a cardiac autonomic function test (15, 21) it is an assessment of the pliability of the cardiovascular regulatory mechanisms.

The following parameters were recorded initially on both subjects and controls to get the baseline values:
Resting heart rate.
Resting blood pressure.
HRV in the resting supine position.
Low power frequency (LF).
High power frequency (HF).
Low power frequency in normalised units (LF nu).
High power frequency in normalised units (HF nu).
Average of all the NN intervals (Mean RR).

Standard deviation of all NN intervals under consideration. It is a measure of total variability (SDNN).
Square root of the mean of the sum of the squares of differences between adjacent NN intervals (RMSSD).
NN50 is the number of pairs of adjacent NN intervals differing by more than 50.
pNN50 is NN50 divided by the total number of NN intervals (pNN50).

The guidelines provided by the task force on HRV analysis were followed. The tests were done in the morning within 2 - 4 hours after the breakfast. The participant was allowed to lie down quietly in a couch for 20 minutes with the eyes open and not falling asleep. ECG was recorded continuously for 5 minutes. The subject continued to lie quietly in the couch, awake and not making any movements. This ECG was used for the calculation of mean heart rate and HRV during rest in the supine position.

ECG was recorded by placing three disposable adhesion electrodes on the limb in the pattern of lead II configuration, the negative electrode was placed on the right arm, positive electrode on the left arm and reference electrode on the left foot. Baseline electrocardiograms were obtained from all subjects and those with abnormal baseline ECG (including juvenile pattern) were excluded.
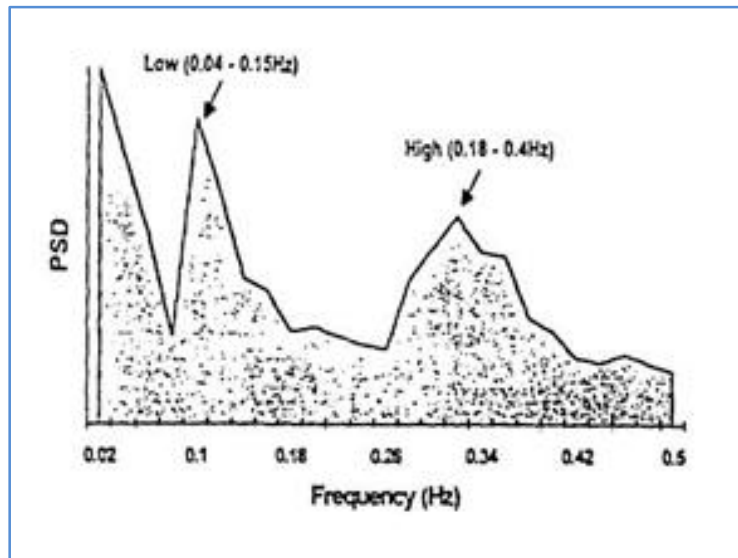
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ECG was obtained by the students Biopac version 1.3. RR intervals were obtained after clearance of noise and baseline fluctuations by digital filters. Those patients who had ectopic beats were excluded from the study. The data was filtered using a digital notch filters with a sampling rate of 1000 samples/sec. The inbuilt software selected the RR peaks and these RR intervals which were obtained as time points were then fed into a Microsoft excel sheet and RR intervals were copied to a notepad file.

Fig. 1: The Tachogram. R-R interval data are represented on a tachogram, in which the y-axis plots the R-R intervals, and the x-axis the total number of beats.
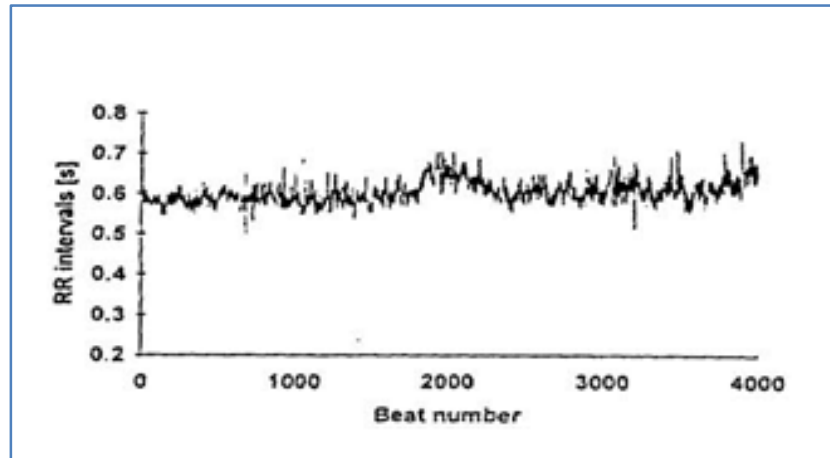
Fig. 1

Fig. 2: Power spectrum of HRV (PSD = power spectral density). Spectral analysis of the tachogram transforms the signal from time to frequency on the x-axis, by representing the signal as a combination of sine and cosine waves, with different amplitudes and frequencies.


Fig. 2

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HRV analysis was done by feeding this RR interval notepad file to the HRV analysis software, version 1.1 from Biomedical Signal Analysis group, Department of Applied Physics, University of Kuopio, Finland. To analyze data lengths of 5 minutes, 256 seconds of RR interval data was taken from the tachogram and interpolated at 4 Hz to get 1024 points. Power spectral analysis was done by Fast Fourier transformation (FFT) after detrending and removal of the mean from the data points. Hanning window was applied to prevent spectral leakage (default). The power spectral density was obtained by Welch's periodogram, using window width of 512 data points with an overlap of 256 points.

Fig. 3: PC display of the HRV analysis software version 1.1, Power spectral analyses were done by Fast Fourier transformation. The power spectral density was obtained by Welch's periodogram, using window width of 512 data points with an overlap of 256 points.
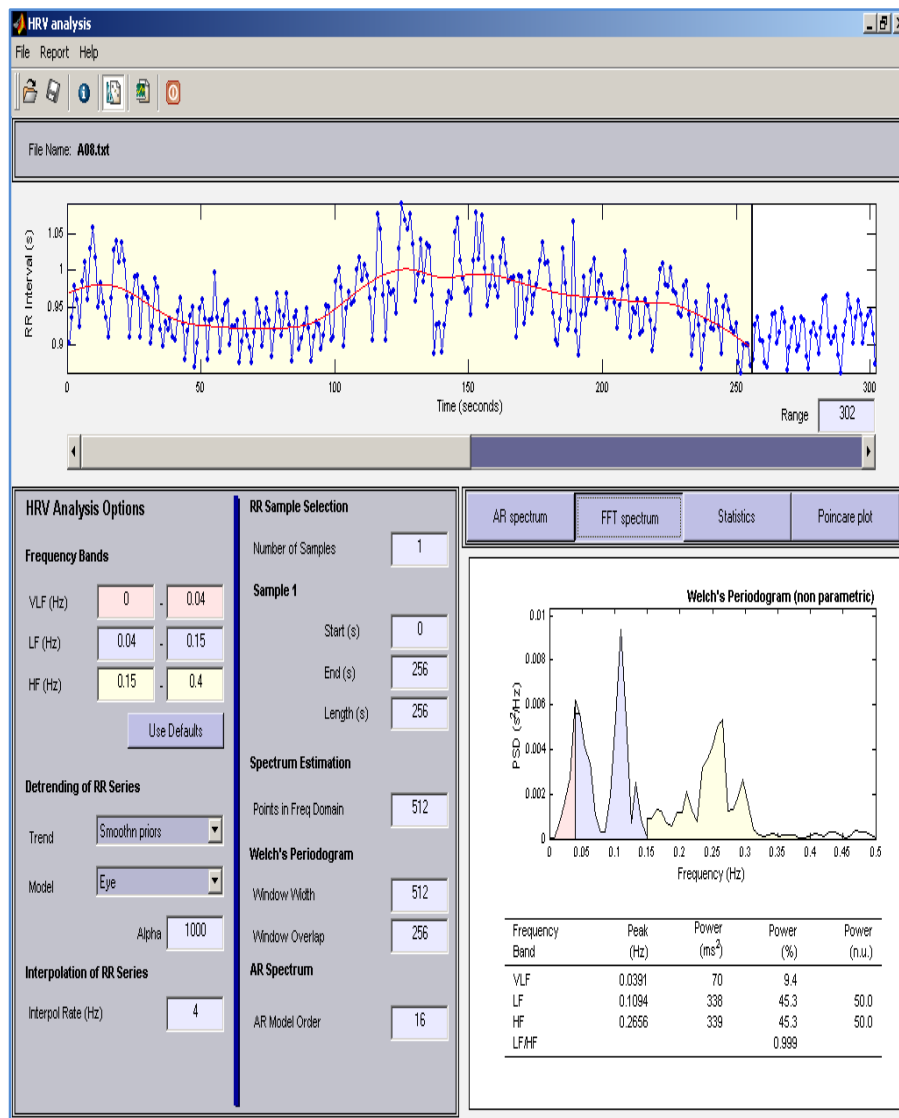
Fig. 3

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Statistical Analysis: The Statistical Package for the Social Science (SPSS 12.0 version for windows) was used for statistical analysis. Independent sample't' test was used to compare the measured parameters of patients with PCOS and control group. ANOVA was used to compare the HRV parameters and the metabolic parameters within the PCOS group. Pearson's correlation analysis was used to investigate the relationship between the metabolic parameters and the HRV parameters. Statistical significance was set at p<0.05.

RESULTS: Analysis between the lean and ideal weight PCOS patient and the control group (study group selected according to BMI) was done. Values were expressed as Mean±SD.

Baseline characteristics of PCOS and control Group: Mean±SD of age of the PCOS was 22.96±3.96 and the control was 24.21±4.69. BMI of the PCOS was 22.12 ± 2.56 and the control was 20.86 ± 2.73. Weight of the PCOS was 53.60 ± 8.86 and the control was 51.09±9.31, with P value 0.324, 0.104 and 0.344 respectively. These shows the mean age, BMI and weight were not significantly different. (Refer table 1).

Resting blood pressure of the lean PCOS and control Group: Mean±SD of SBP of the PCOS was 107.67±10.66 and the control was 106.17±14.30. DBP of the PCOS was 73.58±8.75 and the control was 71.08±9.14. There was no significant difference between systolic and diastolic BP with P value 0.682 and 0.338 respectively (Refer Table 1).

Spectral analysis of Frequency domain HRV parameters of PCOS and control LF of the PCOS was 289.25±256.87 and the control was 348.17±258.48. (Fig. 4) There was no significant difference between the study populations.

Parameters	Case	Control	P value
Age (years)	22.96±3.96	24.21±4.69	0.324 (NS)
BMI (Kg / m2)	22.12±2.56	20.86±2.73	0.104 (NS)
Weight (Kg)	53.60±8.86	51.09±9.31	0.344 (NS)
Waist Circumference (cms)	60.96±15.97	76.67±8.00	< 0.001***
W / H ratio	0.84±0.005	0.81±0.004	<0.05*
RHR (Beats/min)	82.65±10.87	74.00±6.78	<0.01**
SBP (mm Hg)	107.67±10.66	106.17±14.30	0.682 (NS)
DBP (mm Hg)	73.58±8.75	71.08± 9.14	0.338 (NS)
Table 1: Baseline characteristics of the PCOS and Control group			

* – Significant ** – Moderately Significant *** – Highly Significant NS – Not Significant RHR - Resting heart rate SBP - Systolic blood pressure DBP - Diastolic blood pressure

Parameters	Case	Control	P value
LF	289.25±256.87	348.17±258.48	0.432 (NS)
HF	286.29±299.47	671.13±492.31	<0.01**
LF/HF RATIO	1.66±1.01	0.62±0.41	<0.001***
LF nu	57.02±16.07	35.13±13.39	<0.001***
HF nu	42.98±16.07	64.88±13.39	<0.001***
Table 2: Comparison of Frequency domain measures of the PCOS and Control group			

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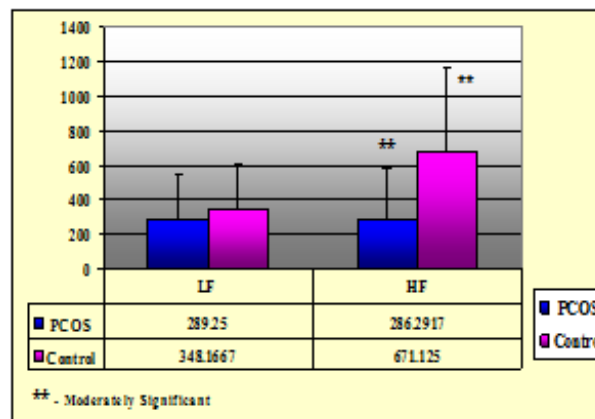** – Moderately Significant *** – Highly Significant NS – Not Significant.

Low power frequency (LF); High power frequency (HF); Low power frequency in normalised units (LF nu); High power frequency in normalised units (HF nu)

Difference between the LF powers of the study population was not significant (P value 0.432). All other parameters were significantly different. HF (286.30±299.47 vs 671.13±492.31 P value 0.002) was moderately significant (Fig: 4), LF nu (57.02 ± 16.07 vs 35.13± 3.39, P value <0.001), HF nu (42.98±16.07 vs 64.88±13.39, P value<0.001) (Fig: 5) and LF/HF ratio were highly significant. (1.66±1.01 vs 0.62±0.41 P value <0.001) (Fig: 6 and 7).

Therefore increased LF/HF ratio, increased LFnu and decreased HFnu shows that there is increased cardiac sympathetic activity even in lean and ideal weight PCOS. (Refer Table 2)

Fig. 4: Comparisons of LF and HF among PCOS and Controls showing moderately significant difference in HF power but LF remained normal. It shows no significant difference between the LF powers of the study population but HF was significantly low among the patients than the controls.

Fig. 4

Fig. 5: Comparison of LFnu and HFnu among PCOS and Controls showing highly significant difference in both HF nu and LFnu. LFnu was significantly higher and HFnu was significantly lower in PCOS indicating altered vagal activity. Therefore increased LFnu and decreased HFnu shows that there is increased cardiac sympathetic activity even in lean and ideal weight PCOS.
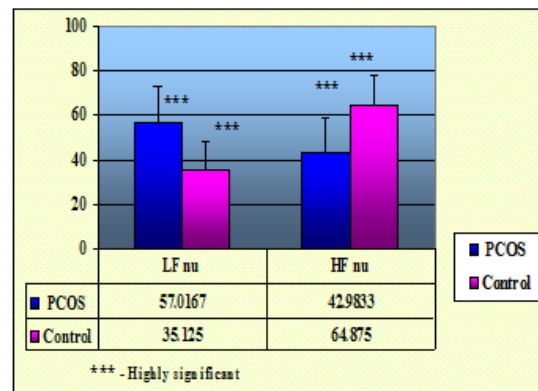

Fig. 5

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Spectral analysis of time domain measures of the PCOS and control group Mean RR (0.74± 0.11 vs 0.82±0.07, P value 0.006), SDNN (0.35±0.02 vs 0.47±0.02, P value 0.011) RMSSD (35.02± 20.23 vs 55.15±23.11, P value 0.002), NN50 (48.54±49.55 vs 94.38±58.84, P value 0.005) and pNN50 (15.38±16.47 vs 31.49±20.57 P value 0.004) are shown in Fig 8 and 9 showing significant decrease in all the parameters (Refer Table 3)

Fig. 6: Comparison of LF/ HF ratio among PCOS and Controls showing highly significant difference in HF/LF ratio infereing increased sympathetic to parasympathetic components of cardiac autonomic activity. LF/HF ratio was highly significant among PCOS. Increased LF/HF ratio, shows that there is increased cardiac sympathetic activity even in lean and ideal weight PCOS.
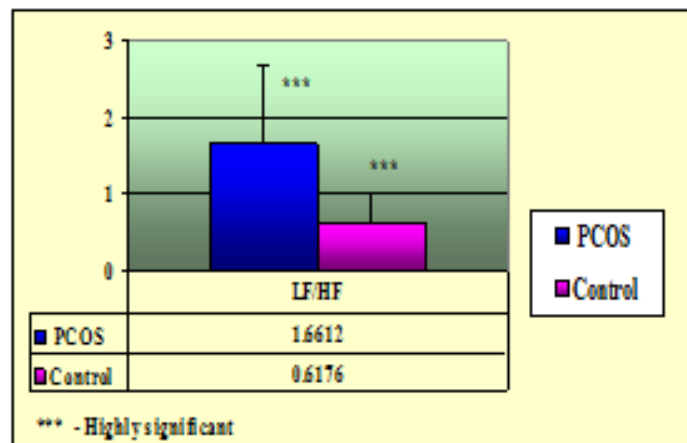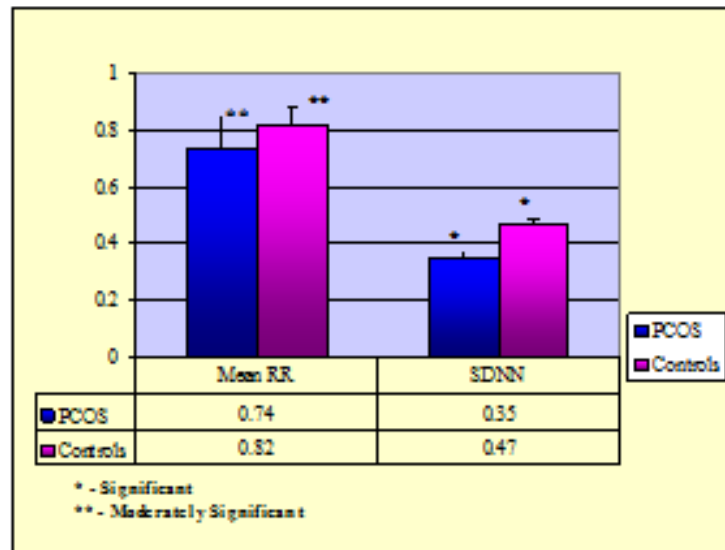

Fig. 6

Fig. 7: Comparison of LF/ HF ratio among entire PCOS and Controls. LF/ HF ratio among entire PCOS and Controls indicating high variations among patients than controls.
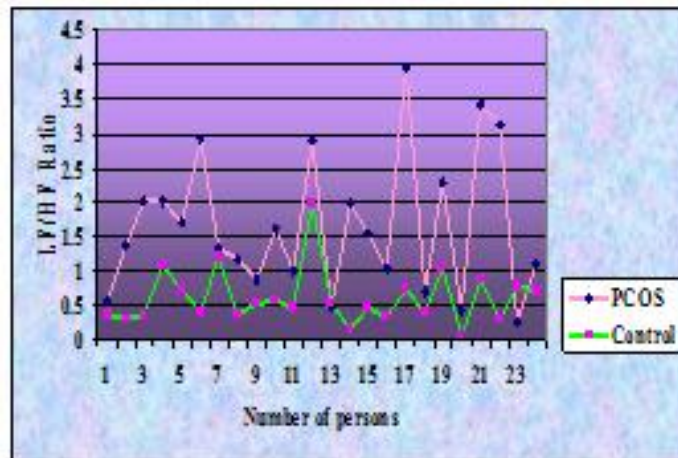
Fig. 7

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Fig. 8: Comparison of Time domain measures of the PCOS and Control groups It shows significant decrease in Mean RR and SDNN among the patients than the controls.

Fig. 8

Comparison of metabolic profile of the lean and ideal weight PCOS and control Group: Fasting blood sugar of the PCOS was 93.59 ± 18.15 and the control was 82.16 ± 10.12. The difference was significant with a p value of 0.0110. On analyzing the fasting lipid profile which included Triglycerides (TGL), total cholesterol (TC), HDL and LDL cholesterol (HDL–C and LDL–C), VLDL–C and the lipid ratio (TC/HDL-C), we found significant difference in TC and LDL – C and with a p value of 0.028 and 0.020 respectively. TC of the PCOS was 166.34 ± 28.12 and the control was 146.42 ± 32.70. LDL–C of the PCOS was 79.92 ± 29.02 and the control was 100.10 ± 28.89. Other parameters which included TGL, HDL–C, VLDL– C and TC/HDL–C ratio did not show any significant difference with a p value of 0.327, 0.968, 0.068 and 0.903 respectively. (Table 4; Fig 10)

Parameters	Case	Control	P value
Mean RR	0.74±0.11	0.82±0.07	<0.01**
SDNN	0.35±0.02	0.47±0.02	<0.05*
RMSSD	35.02±20.23	55.15±23.11	<0.01**
NN50	48.54±49.55	94.38±58.84	<0.01**
pNN50	15.38±16.47	31.49±20.57	<0.01**
Table 3: Comparison of Time domain measures of the PCOS and Control group			

* – Significant ** – Moderately Significant

Mean RR - Average of all the NN intervals; SDNN - Standard deviation of all NN intervals under consideration. RMSSD - Square root of the mean of the sum of the squares of differences between adjacent NN intervals; NN50 is the number of pairs of adjacent NN intervals differing by more than 50. pNN50; NN50 divided by the total number of NN intervals

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Fig. 9: Comparison of Time domain measures of the PCOS and Control groups there was significant decrease in RMSSD and pNN50 among the patients than the controls.
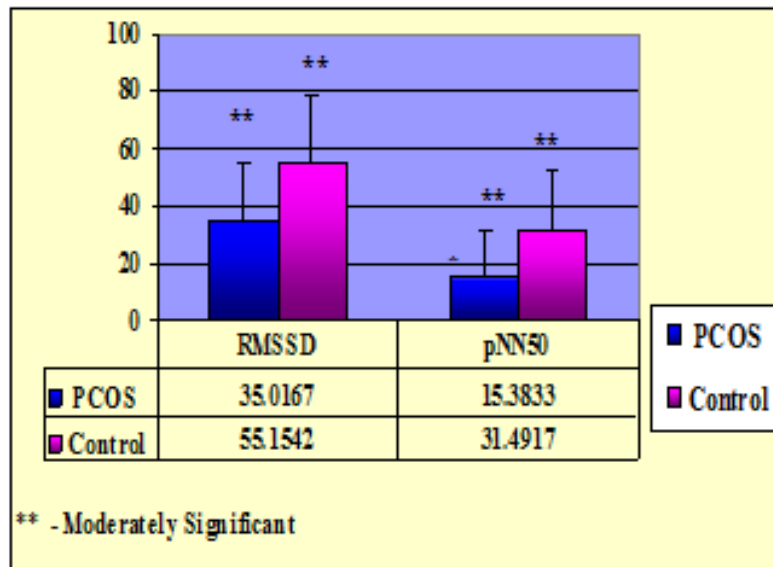
Fig. 9

Correlation analysis of the HRV parameters and metabolic profile between the PCOS and the Control group

In order to find if there is any relationship between the HRV parameters and lipid profile, we did a correlation analysis between the HRV parameters and lipid profile. Correlation analysis between the frequency domain measures and the metabolic parameters of the lean and ideal weight PCOS population (n = 48) are shown in Table 5. There was a correlation between triglycerides with LF nu, HF nu and LF/HF ratio. Other parameters did not show any correlation. (Table: 5)

| Parameter	Case	Control	P value
FBS	93.59±18.15	82.16±10.12	<0.05*
TC	166.34±28.12	146.42±32.70	<0.05*
TGL	97.98±36.44	107.52±29.86	0.327 (NS)
HDL – C	46.66±11.01	46.78±9.19	0.968 (NS)
LDL – C	100.10±28.89	79.92±29.02	<0.05*
VLDL – C	21.44±13.05	21.07±6.56	0.903 (NS)
TC / HDL ratio	3.78±1.47	3.22±0.92	0.068 (NS)
Table 4: Comparison of Metabolic profile of the PCOS and Control group			

* – Significant NS – Not Significant

FBS- Fasting blood sugar; TC - Total cholesterol TGL–Triglycerides; HDL-C - High density lipoprotein cholesterol LDL-C- Low density lipoprotein cholesterol; TGL -Triglycerides; VLDL-C-Very low density lipoprotein cholesterol (VLDL-C)

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Fig. 10: Comparison of Metabolic profile of the PCOS and Control group showing significant increase in FBS, TG and decrease in HDL-C among the patients than the controls.
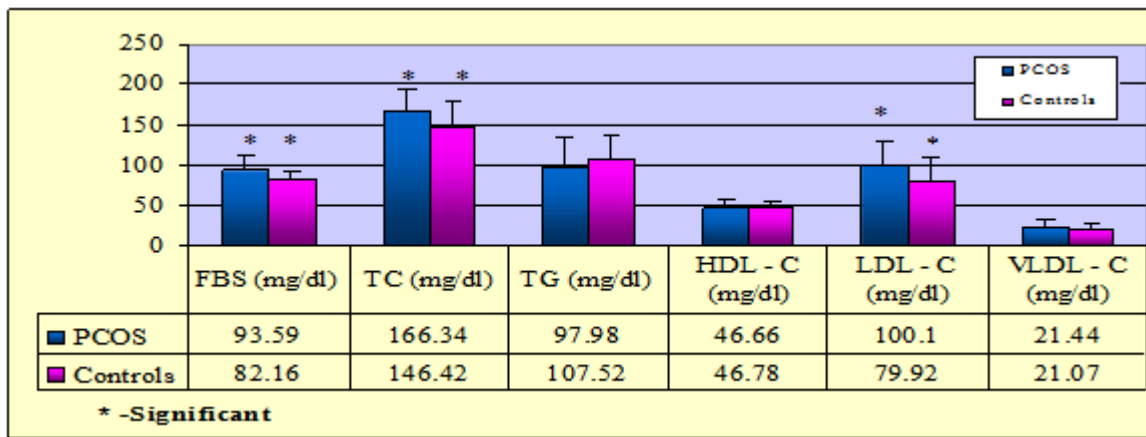
Fig. 10

Correlation analysis between the time domain measures and the metabolic parameters of the lean and ideal weight PCOS study population (n = 48) are shown in Table 6. There was a correlation between RR interval and fasting blood sugar. Also the RR interval was correlated with total cholesterol, LDL cholesterol and the TC/HDL-C ratio. Other parameters did not show any correlation. (Table: 6)

Parameters		FBS	TC	TGL	HDL – C	LDL – C	VLDL - C
LF	R Value	r = -0.173	r = -0.211	r = -0.068	r = 0.133	r = -0.214	r = -0.074
(n=48)	P Value	p = 0.241	p = 0.151	p = 0.644	p = 0.369	p = 0.143	p = 0.617
HF	R Value	r = -0.091	r = -0.187	r = 0.235	r = -0.079	r = -0.163	r = 0.079
(n=48)	P Value	p = 0.539	p = 0.204	p = 0.109	p = 0.592	p = 0.267	p = 0.594
LF/HF	R Value	r = 0.146	r = -0.034	r = 0.393**	r = 0.000	r = 0.032	r = -0.071
(n=48)	P Value	p = 0.321	p = 0.820	p = <0.01	p = 0.998	p = 0.827	p = 0.631
LFnu	R Value	r = 0.047	r = -0.020	r = -.477**	r = 0.069	r = 0.043	r = -0.177
(n=48)	P Value	p = 0.750	p = 0.892	p = <0.01	p = 0.641	p = 0.771	p = 0.229
HFnu	R Value	r = -0.047	r = 0.020	r = 0.477**	r = -0.069	r = -0.043	r = 0.177
(n=48)	P Value	P = 0.750	p = 0.892	p = <0.01	p = 0.641	p = 0.771	p = 0.229
Table 5: Correlation analysis of the HRV parameters and metabolic profile between the PCOS and the Control group							

** – Moderately Significant

DISCUSSION: PCOS is a disorder of chronic anovulation, hyperandrogenism, hirsutism, obesity, sub fertility and insulin resistance. The main feature is the alteration in the endogenous sex hormones and they are known candidates for increased cardiovascular risk, and it has recently been shown that they have systolic dysfunction, an early indicator of hypertension.[22] They have a high incidence of hypertension [23] and are at increased risk of CVD.[24]

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Parameters		FBS	TC	TGL	HDL – C	LDL – C	VLDL - C
RR	R Value	r = -0.297*	r =-0.299*	r = -0.009	r = 0.126	r = -0.326*	r = -0.147
(n= 48)	P Value	p = <0.05	p = <0.05	p = 0.950	p = 0.392	p =<0.05	p = 0.320
SDNN	R Value	r = -0.164	r = 0.189	r = 0.161	r = 0.089	r = -0.219	r = 0.049
(n= 48)	P Value	p = 0.265	p = 0.198	p = 0.276	p = 0.549	p = 0.134	p = 0.740
RMSSD	R Value	r = -0.168	r = -0.168	r = 0.160	r = 0.143	r = -0.225	r = 0.011
(n=48)	P Value	p = 0.253	p = 0.253	p = 0.276	p = 0.333	p = 0.125	p = 0.940
pNN50	R Value	r = -0.132	r = -0.158	r = 0.043	r = 0.122	r = -0.186	r = -0.072
(n=48)	P Value	p = 0.371	p = 0.283	p = 0.769	p = 0.408	p = 0.206	p = 0.627
Table 6: Correlation analysis of the HRV parameters and metabolic profile between the PCOS and the Control group							

* – Significant

Relative studies suggested that though hypertension appears as an integral part of the insulin-resistance syndrome, women with PCOS have only occasionally been noted to have hypertension,[25] and large clinical studies of women with PCOS (based on hyper androgenic chronic anovulation) have reported normal mean baseline blood pressures.[26] Increase in blood pressure, when noted, are usually mild and of questionable clinical significance.[27] In our study population (lean and ideal weight PCOS) resting blood pressure was not elevated (Table 1)

HRV is a statistical measure of the cyclic beat to beat variation of heart rate, which correlates with the individual autonomic tone and is used to quantify risk in a wide variety of both cardiac and non-cardiac disorders. Diminished HRV is associated with increased sympathetic and decreased vagal modulation, and these autonomic changes have been reported to be associated with an increase in the malignant ventricular arrthymias.[5]

As we know obesity is an intriguing factor, we thought to have a closer look at the presence or absence of obesity factor, HRV parameters and metabolic profile in lean and ideal weight PCOS patients. Whether the obesity is a cause of PCOS or obesity is a result of PCOS is unclear, but it seems that the latter is more likely. Our analysis of HRV showed decreased HRV (Table 2 and 3). Of the HRV parameters, frequency domain measures HF power and HFnu was less among patients than the controls, LFnu and LF / HF ratio was high among PCOS than controls.

With regard to LF power it was elevated among controls than patients though not significant (Table 2). This shows that the sympathetic activity was high in the PCOS people and it is consistent with the study done by Aylinrin et al who found autonomic innervations of the heart can be affected in PCOS with increased sympathetic and decreased parasympathetic component of heart rate variability. As a result sympathetic to parasympathetic ratio may increase in PCOS [28]. Of the time domain variables, Mean RR, RMSSD, SDNN, pNN50 and NN50 were less among the patients than controls relating the sympathetic dominance (Table 3).

We see that even lean PCOS have high cardiac sympathetic activity, suggesting altered sympathovagal balance excluding obesity factor. They showed enhanced LH / HF ratio and LFnu and decreased HFnu and HF power. Excluding the confounder the obesity factor it is clear that sympathetic hyperactivity occur in the absence of obesity. This finding may have implications for the mechanisms underlying the increased risk of developing hypertension, including the higher incidence

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of CVD later in life, in PCOS women with normal or lean BMI. The greatest potential therapeutic use of HRV analysis is in the risk strategical analysis of patients after myocardial infarction.[29]

Women with androgen excess represent the biological experiment in nature to illustrate the effects of sex steroids on lipids. Disorders of androgen excess are more common than is generally realized. Some elaborate that 15% of women are affected. Women with normal estrogen by androgen profile have less insulin resistance and favorable lipid profile. On the other hand women with insulin resistance as a result of obesity and (or) abnormal androgen / estrogen profile (adipose distribution also may be involved), loose their favorable cardiovascular low risk state.[30]

We know women with polycystic ovary syndrome are hyper androgenic and insulin resistant, which are associated with alterations in circulating lipid and lipoprotein levels. Women with PCOS also have metabolic abnormalities such as insulin resistance,[31] glucose intolerance[32] and dyslipidemia.[33] Increased low-density lipoprotein (LDL) levels are independent of obesity,[34] whereas increased triglyceride and decreased high-density lipoprotein (HDL) levels are found primarily in obese women with PCOS.[35] At the baseline, the obese women had a more atherogenic lipid profile than the non-obese subjects but in our study population (lean and ideal weight population), fasting blood sugar was elevated than the controls. On analyzing the fasting lipid profile which included TGL, TC, HDL–C, LDL–C, VLDL–C and TC/HDL-C, we found TC and LDL-C were higher among the patients TGL, HDL–C, VLDL–C and TC/HDL–C ratio did not show any difference. (Table 4).

Our study was consistent with the study done by Legro et al who found in a large study of non-Hispanic white women, elevations in LDL-C levels were the predominant lipid abnormality in women with polycystic ovary syndrome, independent of obesity,[36] S Dejager et al who suggested androgen excess and mild insulin-resistance may have an early modifying effect on low density lipoprotein levels in polycystic ovary syndrome women,[37] Murat Yilmaz et al who found changes in serum lipid profile were observed in lean PCOS patients[38] and Mattsen et al who found increased triglycerides and very low density cholesterol but no difference in HDL-C in hirsute women with oligomenorrhea[39] even though Ehrmann DA et al showed that there is only negligible or no effect on lipids in women with PCOS.[40]

This study focuses abnormal cardiac autonomic activity and abnormal metabolic profile in younger and lean PCOS patients but long-term studies of well characterized women with PCOS are lacking and the link to primary cardiovascular events such as stroke or myocardial infarction remains more speculative than substantive.

Our study gives a solid and strong evidence of altered cardiac autonomic activity and unfavourable metabolic profile, which are the important risk factors for cardiovascular disease even in lean and ideal weight PCOS. It is a favourable study since we could have significantly younger age group study population because HRV may worsen with age but our study should also be interpreted in light of some limitations. One of the main limitations is the small sample size of study groups. Second is that we were not able to do a direct comparative analysis between obese and lean PCOS and the last is that lean and ideal weight PCOS were categorized based on BMI and not on absence of visceral obesity (According to waist – hip ratio).

Despite the limitations of the study, to our knowledge, this is the first study to investigate the cardiac autonomic innervations in patients with PCOS in Indian subcontinent by means of HRV parameters. Also it is the first study to analyze HRV parameters in lean and ideal weight PCOS. The result of the present study shows that cardiac autonomic activity can be affected in PCOS with

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increased sympathetic and decreased parasympathetic components of HRV. As a result sympathetic to parasympathetic ratio may increase in PCOS. After this study we feel further research with larger sample size will help in better analysis of HRV parameters in this special group of PCOS women. Also further studies in larger group of lean and ideal weight PCOS are warranted. Hence we suggest all PCOS irrespective of obesity should be screened for cardiac dysfunction and abnormal lipid profile and routine investigations should include ECG with HRV.

REFERENCES:
1. Abdul H Zargar, Vipin K Gupta, Arshad I Wani, Shariq R Masoodi, Mir I Bashir, Bashir A Laway, Mohammed A Ganie and Mohammad Salahuddin. Prevalence of ultrasonagraphy proven polycystic ovaries in North Indian women with type 2 diabetes mellitus. Reproductive Biology and Endocrinology 2005; 3: 35
2. Ovalle F, Ricardo A. Insulin resistence, Polycystic ovary syndrome and Type II Diabeyes mellitus. Fertil Steril 2002; 77: 1095-1105.
3. Legro RS. Polycystic ovary syndrome and cardiovascular disease: A premature association? Endo Rev 2003; 24: 302 – 312.
4. Wild S, Pierpoint T, McKeigue P, Jacobs H. Cardiovascular disease in women with polycystic ovarian syndrome at a long term follow up: a retrospective cohort study. Clin Endocrinol (Oxf) 2000; 52: 595-600.
5. E. Dahlgren a, P. O. Janson a, S. Johansson b, L. Lapidus c, A. Od n b S. Polycystic ovary syndrome and risk for myocardial infarction: Evaluated from a risk factor model based on a prospective population study of women. Acta Obstet Gynecol Scand 1992; 71: 599 – 604.
6. Enrico carmino and Rogerio A Lobo. Polycystic Ovary Syndrome (PCOS): Arguably the most common Endocrinopathy is associated with significant morbidity in women. The Journal of clinical Endocrinology and Metabolism. 1999; 84: 1897-1899.
7. GM Prelevic, T Beijic, L Balint-Peric, J Ginsburg - Clinical Endocrinology. Cardiac flow velocity in women with polycystic ovary syndrome. Clin Endocrinol 1995; 43: 677–681.
8. H Yarali, A Yildirir, F Aybar, G Kabakci. Diastolic dysfunction and increased serum homocysteine concentrations may contribute to increased cardiovascular risk in patients with polycystic ovary syndrome. Fertil Steril 2001; 76: 511-516.
9. Christopher J. G. Kelly, Angela Speirs, Gwyn W. Gould, John R. Petrie, Helen Lyall and John M. C. Connell. Altered vascular function in young women with polycystic ovary syndrome. J Clin Endocrinol Metab 2002; 87: 742–746.
10. Giancarlo Paradisi, Helmut O. Steinberg, Annette Hempfling, Jessica Cronin, Ginger Hook, Marguerite K. Shepard, Alain D. Baron, Paradisi G, Steinberg HO, Hempfling A. Polycystic ovary syndrome is associated with endothelial dysfunction. Circulation 2001; 103: 1410–1415.

ORIGINAL ARTICLE

11. Chris C. J. Kelly, Helen Lyall, John R. Petrie, Gwyn W. Gould, John M. C. Connell and Naveed Sattar. Low grade chronic inflammation in women with polycystic ovary syndrome. J Clin Endocrinol Metab 2001; 86: 2453–2455.

12. Giuseppe Loverro, Filomenamila Lorusso, Loredana Mei, Raffaella Depalo, Gennaro Cormio, Luigi Selvaggi. The plasma homocysteine levels are increased in polycystic ovary syndrome. Gynecol Obstet Invest 2002; 53: 157–162.

13. Bulent O. Yildiz, Ibrahim C. Haznedaroglu, Serafettin Kirazli and Miyase Bayraktar. Global fibrinolytic capacity is decreaed in polycystic ovary syndrome, suggesting a prothrombic state. J Clin Endocrinol Metab 2002; 87: 3871–3875.

14. Christopher J. G. Kelly, Helen Lyall, John R. Petrie, Gwyn W. Gould, John M. C. Connell, Ann Rumley, Gordon D. O. Lowe and Naveed Sattar. A specific elevation in tissue plasminogen activator antigen in women with polycystic ovary syndrome. J Clin Endocrinol Metab 2002; 87:3287–3290.

15. Task Force of the European society of Cardiology and the North American society of Pacing and Electrophysiology. Heart rate variability Standards of measurement, physiological interpretation and clinical use. Circulation.1996; 93: 1043-1065.

16. RA Wild, PC Painter, PB Coulson, KB Carruth and GB Ranney. Lipoprotein lipid concentrations and cardiovascular risk in women with polycystic ovary syndrome Journal of Clinical Endocrinology & Metabolism, 1985; Vol 61, 946-951.

17. Ehrmann DA, Cavagan MK, Imperial J, Sturis J, Rosenfield RL& Polensky KS. Effect of metformin on insulin secretion, insulin action and ovarian steriodogenesis in women with polycystic ovary syndrome. Journal of Clinical Endocrinology and Metabolism.1997; 82: 524-530.

18. The Rotterdam ESHRE / ASRM – sponsored PCOS consensus workshop group Revised 2003 consensus on diagnostic criteria and long term health risks related to Polycystic Ovary Syndrome. January 2004; Fertility and sterility 81:19–25

19. Tsuguhito Ota, Toshinari Takamura, Nobuyuki Hirai, Ken-ichi Kobayashi. BMI Classification Physical status: Preobesity in World Health Organization Classification Involves the Metabolic Syndrome in Japanese. Doi: 10.2337/diacare.25.7.1252 Diabetes Care July 2002 vol. 25 no. 7 1252-1253.

20. World Health Organization. Obesity – Preventing and Managing the Global Epidemic: Report of a WHO Consultation on Obesity. Geneva: World Health Organization, 1998.

21. Akselrod S, Gordon D, Ubel FA, Shannon DC, Barger AC, Cohen RJ. Power spectrum analysis of heart rate fluctuation: A quantitative probe of beat-to-beat cardiovascular control. Science. 213: 220-222.

22. Tíras MB, Yalcin R, Noyan V, Maral I, Yìldìrìm M, Dörtlemez O, Daya S: Alterations in cardiac outflow parameters in patients with polycystic ovarian syndrome. Human Reproduction 1999; 14: 1949-1952.

23. Dahlgren E, Janson PO, Johansson S, Lapidus L, Lindstedt G, Tengborn L. Hemostatic and metabolic variables in women with polycystic ovary syndrome. Fertil Steril. 1994; 61: 455–460.

24. A. Holmang, J. Svedberg, E. Jennische and P. Bjorntorp. Effects of testosterone on muscle insulin sensitivity and morphology in female rats. Am J Physiol Endocrinol Metab.1990; 259: E555-E560

ORIGINAL ARTICLE

25. Sampson M, Kong C, Patel A, Unwin R, Jacobs HS. Ambulatory blood pressure profiles and plasminogen activator inhibitor (PAI-1) activity in lean women with and without the polycystic ovary syndrome. Clin Endocrinol (Oxf) 1996; 45: 623–629.
26. Legro RS, Kunselman AR, Dunaif A. Prevalence and predictors of dyslipidemia in women with polycystic ovary syndrome. Am J Med 2001; 111: 607–613.
27. Holte J, Gennarelli G, Berne C, Bergh T, Lithell H. Elevated ambulatory day-time blood pressure in women with polycystic ovary syndrome: a sign of a pre-hypertensive state? Hum Reprod 1996; 11: 23–28.
28. Aylin Yildirir, Funda Aybar, Giray Kabakci, Hakan Yarali, and Ali Oto. HeartRate Variability in Young Women with Polycystic Ovary Syndrome. Annals of Noninvasive Electrocardiology October 2006; 11(4): 306–312
29. Fei L, Copie X, Malik M, et al. Short - and long – term assessment of heart rate variability for risk stratification after acute myocardial infarction. Am J Cardiol 1996; 77: 681- 684.
30. Pyorala K. Relationship of glucose tolerance and plasma insulin to the incidence of coronary artery disease: result from two people studies in Fiblanda. Diabetes care 1976; 2: 131 – 41.
31. Chang RJ, Nakamura RM, Judd HL, Kaplan SA.Insulin resistance in nonobese patients with polycystic ovarian disease. J Clin Endocrinol Metab 1983; 57: 356–359.
32. Ehrmann DA, Barnes RB, Rosenfield RL, Cavaghan MK, Imperial J. Prevalence of impaired glucose tolerance and diabetes in women with polycystic ovary syndrome. Diabetes Care1999; 22: 141–146.
33. Wild RA, Painter PC, Coulson PB, Carruth KB, Ranney GB. Lipoprotein lipid concentrations and cardiovascular risk in women with polycystic ovary syndrome. J Clin Endocrinol Metab 1985; 61: 946–951.
34. Graf MJ, Richards CJ, Brown V, Meissner L, Dunaif A. The independent effects of hyperandrogenaemia, hyperinsulinaemia, and obesity on lipid and lipoprotein profiles in women. Clin Endocrinol (Oxf) 1990; 33: 119–131.
35. Conway GS, Agrawal R, Betteridge DJ, Jacobs HS Risk factors for coronary artery disease in lean and obese women with the polycystic ovary syndrome. Clin Endocrinol (Oxf) 1992; 37: 119–125.
36. Richard S Legro, Allen R Kunselman, Andrea Dunaif, Prevalence and predictors of dyslipidemia in women with polycystic ovary syndrome. The American Hournal Of Medicine 1 December 2001; Volume 111, Issue 8, Pages 607-613.
37. S. Dejager, C. Pichard, P. Giral, E. Bruckert, M.C. Federspield, I. Beucler, G. Turpin. Smaller LDL particle size in women with polycystic ovary syndrome compared to controls Clinical Endocrinology 2001; 54 (4): 455–462.
38. Murat Yilmaz, Neslihan Bukan, Goöksun Ayvaz, Ayhan Karakoç, Füsun Töruner, Nuri Çakir and Metin Arslan. The effects of rosiglitazone and metformin on oxidative stress and homocysteine levels in lean patients with polycystic ovary syndrome. Human Reproduction 2005; 20 (12): 3333–3340.
39. Mattson LA, Cullberg G, Hamberg L, Samsioe G, Silferstolve G. Lipid metabolism in women with polycystic ovary syndrome:possible implications for an increased risk of coronory heart disease. Fertil Sertil 1984; 42: 579-84.

ORIGINAL ARTICLE

40. David A. Ehrmann, Melissa K. Cavaghan, Jacqueline Imperial, Jeppe Sturis, Robert L. Rosenfield and Kenneth S. Polonsky. Effects of Metformin on Insulin Secretion, Insulin Action, and Ovarian Steroidogenesis in Women with Polycystic Ovary Syndrome1. The Journal of Clinical Endocrinology & Metabolism 1997; 82 (2): 524-530.

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