Study of Insulin Resistance and Lipid Profile in Polycystic Ovarian Syndrome

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Abstract- Polycystic ovarian syndrome is most common endocrine disease and metabolic disorder in adolescence and reproductive women. In PCOS women insulin resistance thought to be the uniting pathogenic factor in the development of glucose intolerance, obesity, lipid abnormalities, HTN and coronary artery disease .This study is under taken to measure insulin resistance in PCOS and to see the relationship of insulin resistance with lipid profile. Case control study was done taking 60 women PCOS and 60 age matched healthy women as controls. In all the subjects, concentrations of fasting plasma glucose, serum TG, serum TC and HDL were estimated using enzymatic methods in semiautoanalyser. Fasting serum insulin and measured by CLIA using Lumax-CLIA microplate reader. HOMA IR, serum LDL concentration, were calculated from estimated parameters. The mean concentrations of all the parameters were significantly increased in women with polycystic ovarian syndrome when compared with healthy women except serum HDL concentration, which was significantly decreased. Insulin resistance was significantly positively correlated with serum lipid profile and except with serum HDL, which was significantly negatively correlated. This study suggests that insulin resistance in PCOS is associated with dyslipidemia. Hence in PCOS women if measurement of insulin resistance and serum lipid profile is done early, the disease progression can be prevented and prevent future complications.
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Index Terms- Dyslipidemia, Insulin resistance, Lipid profile & Polycystic ovarian syndrome.

I. INTRODUCTION

Polycystic ovarian syndrome is most common endocrine disease and metabolic disorder in adolescence and reproductive women which is first reason for female infertility [1].According to Rotterdam (2003) polycystic ovarian syndrome is defined as having any two of following:

1) Oligo/anovulation
2) Clinical/biochemical signs of hyperandrogenism
3) Polycystic ovaries by scan with exclusion of other related disorder.[2]

In India polycystic ovarian syndrome affects 5-10% of reproductive age women among them 76.9% manifest insulin resistance[3]. Common age of presentation is 17-36 yrs (mean age 24.8)[1]. Insulin resistance defined as inability of known quantity of exogenous or endogenous insulin to increase glucose uptake and utilization in individual, as it does in normal individual. [1]

In polycystic ovarian syndrome patients there is relative inefficiency of insulin receptors binding to insulin which leads to improper transfer of glucose to intracellular compartment and this result in relative hyperglycemia, in spite of increased insulin producing beta cells. In polycystic ovarian syndrome adipocytes there is significant decrease in number of GLUT4 glucose transporters. [4]

Hyperandrogenism leads to increased hepatic lipase.[1].In polycystic ovarian syndrome there is increased secretion of VLDL particle by liver resulting in elevated TG concentration.[5]Insulin resistance and associated dyslipidemia are independent of obesity markers such as BMI, waist to hip ratio.[3]. Overall prevalence of abnormal OGTT in polycystic ovarian syndrome reproductive age women is 20%. It is probably beneficial to screen IR in this specific group in order to provide optimum management.[6]

This study is undertaken to find out prevalence of insulin resistance in polycystic ovarian syndrome patients and to evaluate relationship of HDL-C, LDL-C, TC, TG with insulin resistance in polycystic ovarian syndrome patients.

II. MATERIALS AND METHOD

A study will be carried out for a period of one year. The patients will be selected from Chigateri General Hospital and Bapuji Hospital, Davangere (both hospitals are attached to the teaching institute JJM Medical College, Davangere).and private hospital in and around JJMMC Davangere.

Study will be carried out in clinically 60 diagnosed cases of polycystic ovarian syndrome and 60 age matched controls will be selected based on inclusion and exclusion criteria.

Inclusion Criteria:

CASES; patients between age 17-36yrs diagnosed as polycystic ovarian syndrome having clinical features

- oligomenorrea (35days)

- amenorrea (6months)

- hyperandrogenisum features like acne, hirsuitism, and

-diagnosed polycystic ovaries by ultrasound

CONTROLS; 17- 36 years females having normal menstrual cycle.

Exclusion criteria.

- Patients having history of diabetes mellitus, impaired glucose tolerance,
- Pregnancy, breast feeding ,non fasting
- Patients with untreated hypothyroidism, those on drug treatment like antihypertensive, antiplatelet, lipid lowering agents, drug affecting glucose &lipid metabolism, congenital adrenal hyperplasia, cushing syndrome, ovarian/adrenal androgen secreting tumors.

After taking informed consent, under all aseptic precautions about 5 ml of venous blood will be collected in a sterile bulb after overnight fasting of 12 hours. 2ml will be collected in EDTA vial (for plasma), 3ml in plain vial for (for serum) , it will be subjected for centrifugation serum and plasma will be separated. Insulin, Serum lipoproteins estimated from serum and fasting glucose from plasma.

Fasting plasma glucose and Serum triglycerides measured by GOD POD method by Erba mannheim chem5 plus Semi Auto – analyzer".Normal Fasting plasma glucose 70-110mg/dl. Normal Serum TG levels **:-** 40-150 mg/dl .Serum Total cholesterol AND serum HDL measured by CHOD PAP METHOD .Erba Mannheim Chem5 plus Semi Auto – analyzer"Normal Serum TC levels **:-** 150 -200 mg/dl, "Normal Serum HDL levels **:-** 35-75 mg/dl. serum LDL cholesterol calculated by [7]:Friedwalds formula

$$\text{LDLcholesterol(mg/dl)} = \text{Total cholesterol} - \text{HDL cholesterol} - \frac{[\text{triglycerides}]}{5}$$

Normal serum LDL cholesterol concentration range**:** < 130 mg/dl serum fasting insulin estimated by chemiluminescence immunoassay. Normal value of fasting insulin: 5-25µIU/ml.

Calculation of insulin resistance by using HOMA model (HOMA -IR). [8]

$$\text{HOMA -IR} = \frac{\text{fasting plasma glucose in mg/dl} \times \text{fasting serum insulin in µiu/ml}}{405}$$

the subject is considered to have insulin resistance if HOMA-IR value is more than 2.7.

Statistical analysis was done using SPSS software, version 17.0. Descriptive data were presented as mean ± SD and range values. Unpaired student t test used to compare between cases and controls. Relationship between insulin resistance and serum lipoproteins, was assessed by Pearson's correlation coefficient. For all the tests, the probability value (p-value) of less than 0.05 was considered statistically significant.

III. RESULTS

In the present study, a total number of 120 subjects were included. They were divided into 2 Groups:

Controls – It consisted of 60 healthy women

Cases-It consist of 60 PCOS cases

The present study shows that the mean levels of fasting insulin ,fasting plasma glucose and HOMA IR are significantly increased in subjects with PCOS when compared to healthy controls ($p < 0.001$).(Table 1)

Table 1: Comparision of Fasting plasma glucose, Fasting serum Insulin and insulin resistance in study groups. Results expressed as Mean ± SD.

	Fasting serum Insulin (µIU/ml)	Fasting plasma glucose mg/dl	HOMA-IR
Control	9.33 ±3.08	94.38±10.36	2.16±0.67
PCOS Cases	24.50 ±10.03	114.20±30.38	7.29±4.08
p value	<0.001 HS	<0.001 HS	<0.001 HS

Unpaired student's t-test; $p<0.001$: Highly significant; $p<0.05$: significant

Prevalance of insulin resistance in PCOS cases

In insulin sensitive indivials HOMA IR value found to be <2.77.In this study out of 60 PCOS cases, 45 cases HOMA IR value found to be more then 2.77. So prevalance of insulin resistant cases in PCOS cases in this study is 45/60 i,e 75%.

The present study shows that the mean serum levels of triglycerides, total cholesterol and LDL cholesterol are significantly increased in PCOS cases when compared to healthy controls ($p<0.001$,$p<0.05$&$p<0.05$ respectively) except the mean serum HDL cholesterol level which is significantly decreased ($p<0.05$). (Table 2)

Table2: Comparison of serum concentration of triglycerides(TG), total cholesterol (TC), HDL cholesterol and LDL cholesterol among study subjects. Results expressed as Mean ± SD

	TG(mg/dl)	TC (mg/dl)	HDL (mg/dl)	LDL (mg/dl)
Control	124.97±13.87	152.43±25.78	53.84±5.18	73.59±27.45
PCOS cases	202.44±30.15	210.05±36.96	33.71±6.74	135.85±38.60
P value	<0.001 ,HS	<0.05 ,S	<0.05,S	<0.05,S

* Unpaired student's t-test; $p<0.001$: Highly significant; $p<0.05$: significant

There is significant positive correlation between HOMA IR levels and all lipid profile parameters stated above except serum HDL cholesterol, which is negatively correlated. (Table 3)

Table 3: Pearson Correlation of HOMA IR levels with lipid profile.

Variables	r value	p value
SerumTotal Cholesterol	0.42	0.001*
Serum Triglycerides	0.60	0.00 **
Serum HDL Cholesterol	-0.51	0.00 **
Serum LDL Cholesterol	0.40	0.002 *

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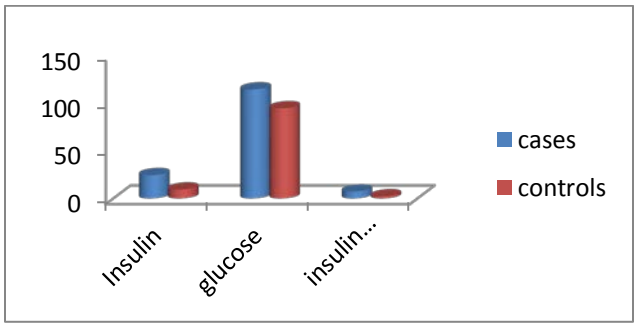
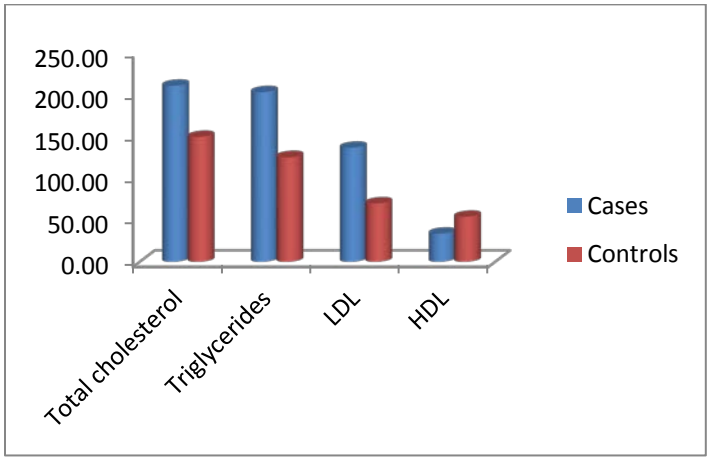
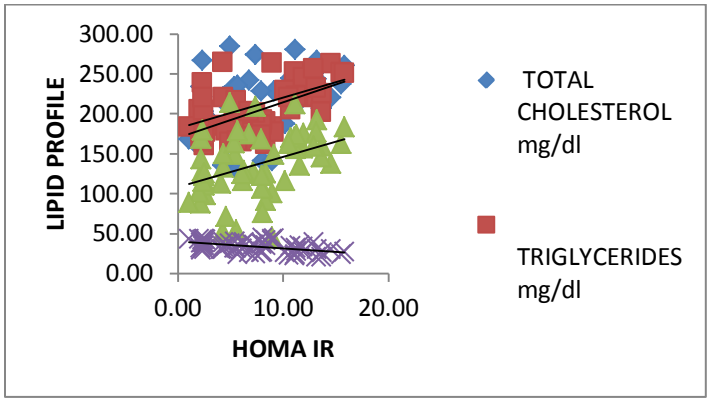
Graph1:Fasting glucose , fasting insulin and insulin resistance HOMA IR in cases and controls



Graph 2: Lipid profile in cases and controls.



Graph 3: Correlation of insulin resistance with lipid profile



DISCUSSION

Polycystic ovary syndrome (PCOS) is the most frequent endocrine disorder seen in pre-menopausal women, affecting 5–10% of this population. It is characterized by menstrual irregularities and biochemical and/or clinical hyperandrogenism such as hirsutism, seborrhoea and acne. Regardless of the presence of obesity, many are also insulin resistant .[9]

PCOS is a heterogenous disorder for which several pathogenic mechanism have been proposed. The main mechanism is abnormal gonadotropin secretion, with excess circulating LH and low FSH. Hypersecretion of androgen by ovarian and adrenal gland which provide substrate for pheripheral aromatisation to estrogen also play role in development of PCOS[10] .PCOS women have insulin resistance, which results in compensatory hyperinsulinemia. The defect in insulin action in PCOS appears to be at the post binding level which involves glucose transport. Insulin resistance may be accompanied by a defect in insulin induced inhibition of lipolysis.[10]

Prevalance in this study is 45/60 i,e 75%. This is similar to anuradhaclara et al[3] where prevalance of IR in PCOS found to be 76.9 % and is in consistant with previous studies that showed Indian PCOS women to be more insuin resistant then there white counterparts[3]. Prevalance of insulin resistance differs by method used to estimate IR. In a study done by pikee saxenaet al[11] prevalance found to be 88%.They used 2 hour post glucose insulin level to identify IR.

In a study done by Jolantaet al[12]insulin resistance was found in 16% of patients according to G0/I0, in 28% of patients according to G120/I120, and in 18% of patients according to HOMA-IR. Here G0/I0 (fasting glucose to insulin ratio; cut-off value 4.5), G120/I120 (glucose to insulin ratio 120 minutes after oral glucose administration; cut- off value 1). The results obtained may be different from the results of other authors, because of the too small number of patients in the study group. QUICKI, HOMA-IR and fasting insulin to fasting glucose ratio are the most useful index in the evaluation of resistance to insulin. However, DeUgarte et al believe that the greatest predictive value is characterized by HOMA-IR.[12]

Presence of insulin resistance is quoted as the main reason for elevation of fasting glucose levels in these studies. S pikee et al[82] found that in patients with normal glucose tolerance, values of glucose remain in the normal range due to adequate insulin response. A woman with IR, normal glycemic levels may be noticed after a glucose challenge because the pancreas will have to secrete excess insulin in order to keep the blood sugar in the normal range. However, with time the β-cells become increasingly dysfunctional and fails to secrete enough insulin to correct the prevailing hyperglycemia , resulting in increased glucose level.[82]

In 2012 Mandrelle K and co-workers conducted study. which aimed to evaluate the prevalence of metabolic syndrome in women with polycystic ovary syndrome. They found that hyperinsulinemia and insulin resistance are the common underlying metabolic abnormalities seen in PCOS and metabolic syndrome 13.

Insulin resistance in PCOS is characterized by decreased sensitivity to insulin in peripheral tissues, notably muscle and adipose tissue, but not in liver that is in contrast to the insulin resistance of type II diabetes where hepatic resistance present. Hyperinsulinemia in women with the polycystic ovary syndrome appears to reflect the hypersecretion of insulin itself, rather than of proinsulin and its split products.[14]

The cellular mechanism of insulin resistance in the polycystic ovary syndrome remains controversial. It may be because of reduced binding of insulin to its receptor, whereas

two recent studies using peripheral adipocytes (recognized target cells for insulin action) have shown normal binding but reduced insulin-mediated glucose transport, suggesting a postr eceptor defect. The mechanism underlying this phenomenon has not been fully characterized, but decreased expression of the insulin-dependent glucose-transporter protein GLUT-4 has been described. [14]

Quantitative insulin sensitivity check index (QUICKI) was developed to improve the sensitivity of fasting measurements. QUICKI has been well correlated to clamp measurements in obese and non-obese patient. QUICKI also demonstrates correlation with HOMAIR[15]. Because of infeasibility of euglycemic insulin clamp method, across globe HOMA method is regarded as gold standard test for measuring insulin resistance in clinical practice and population based research studies .There is a strong positive correlation between HOMA IR and euglycemic insulin clamp IR.[16]

In a study conducted by Cheung L et al showed that high TG and low HDL-C are the characteristic types of dyslipidaemia seen in insulin-resistant subjects although the presence of obesity exacerbates the insulin resistance state associated with PCOS, even lean PCOS women have features of insulin resistance compared to age matched lean control subjects[17].Both insulin resistance and hyperandrogenemia contribute to dyslipidemia in PCOS. Testosterone decreases lipoprotein lipase activity in abdominal fat cells, and insulin resistance impairs the ability of insulin to exerts its antilipolytic effects by altering expression of lipoprotein lipase and hepatic lipase.[18]

In a study conducted by Shabir I et al showed that the maximum subjects PCOS women as well as family members had dyslipidemia in the form of low HDL and high triglycerides.[19]

Insulin plays a key role in TG metabolism as it normally reduces availability of large TGRLs particles, synthesized by a distinct pathway compared with smaller VLDL particles. In insulin resistant subjects, insulin fails to suppress synthesis of large VLDL particles. In addition, insulin resistance is associated with increased flow of free fatty acid to liver, increased lipid synthesis in the liver, and decreased clearance of VLDL particles, all of which increase VLDL concentrations in plasma. Increased secretion of very low density lipoprotein (VLDL) particles by the liver results in elevated plasma TG concentration. Subsequently TGs are exchanged for cholesterol esters by the activity of CE transfer protein. This process results in TG enriched high density lipoprotein (HDL) particle that are catabolised more rapidly, and CE enriched VLDL particle that are converted in to small dense LDL particle. [5,20]

As a consequence insulin resistance contribute to low HDL, high TG and high LDL. Due to reduced levels of HDL cholesterol, reverse transport of cholesterol to liver is impaired leading to its reduced excretion. Therefore these patients have increased serum levels of total cholesterol. Hyperandrogenism has been associated with increased hepatic lipase activity[5,20]. Thus, dyslipidemia may precede the association with insulin resistance and increased risk for CVD.[5,20]

Conclusion

This study shows close relationship between insulin resistance, altered atherogenic lipid profile in PCOS patients. The significantly increased concentrations of fasting plasma glucose and fasting serum insulin along with increased values of HOMA IR are seen in these patients when compared to healthy women. This signifies presence of insulin resistance in women with PCOS.

. Insulin resistance play a role in development of dyslipidemia in PCOS independent of obesity .The variation in lipoprotein causes a pathological status which may lead to damage of tissues, tissue proliferation and cardiovascular inflammation. Since dyslipidemia usally associated with insulin resistance in PCOS, It is of great clinical importance to determine whether the PCOS patients are combined with insulin resistance and to take treatments for insulin resistance for symptom improvement and long-term prognosis in PCOS patients. PCOS symptoms can be improved if insulin resistance is controlled after lifestyle intervention and proper treatment.

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