International Journal of Clinical Endocrinology

Research Article

Hypertriglyceridemia in Polycystic Ovarian Syndrome on Oral Contraceptives, Precludes Role in Non-Alcoholic Steatohepatitis with Polycystic Ovarian Syndrome: A Randomized Study -

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Submitted: 27 November 2020; **Approved:** 04 December 2020; **Published:** 05 December 2020

Cite this article: Najar RR. Hypertriglyceridemia in Polycystic Ovarian Syndrome on Oral Contraceptives, Precludes Role in Non-Alcoholic Steatohepatitis with Polycystic Ovarian Syndrome: A Randomized Study. Int J Clin Endocrinol. 2020 Dec 05 ;4(1): 011-015.

ISSN: 2640-5709

ABSTRACT

Background: Polycystic Ovary Syndrome (PCOS) is a common condition that is characterized by menstrual irregularity, hyperandrogenism, and polycystic ovary morphology. Dyslipidemia is seen in 70% of PCOS patients, commonly found metabolic disturbance in PCOS. Non-Alcoholic Fatty Liver Disease (NAFLD) and PCOS are insulin resistant states and in various meta analytical studies, PCOS patients are at high risk of NAFLD.

Materials and Methods: About 100 patients were enrolled in a randomized study and about 98 completed the study who underwent proper clinical, biochemical and Hormonal analysis as per study protocol. We found that there was a significant increase in Triglyceride (TG) levels from baseline 117.67 + 38.71 to 160.66 + 37.42 (p = <0.001) at 6 months of Oral Contraceptive (OCPs) therapy. However, menstrual cycle frequency improved significantly (p = .005) with OCPs, from 8.42 ± 4.80 to 11.11 ± 1.80 cycles per year at 6 months. The hirsutism score decreased (p = .001) gradually from 14.17 ± 4.63 at baseline to 13.53 ± 4.02 at 6 months of therapy. There was no significant effect on Fasting blood glucose, Bilirubin, Total Protein, Albumin, Alkaline phosphatase, aspartate transaminase, Alanine transaminase, Total cholesterol, Low and high density lipoproteins with 6 months of OCPs therapy.

Conclusion: Hypertriglyceridemia is seen in this study in patients with PCOS on combined Oral contraceptives. Hypertriglyceridemia per se is a precursor mediator of inflammation and a major risk factor for Non-Alcoholic Steatohepatitis (NASH) and also Non-Alcoholic fatty liver disease is well known entity in PCOS. Hence precludes the role of Oral contraceptive pills in PCOS patients with Non-Alcoholic Fatty Liver Disease (NAFLD).

Keywords: Hypertriglyceridemia; PCOS; Oral contraceptives; NAFLD

INTRODUCTION

The Polycystic Ovary Syndrome (PCOS) is a disorder that is characterized by hyperandrogenism, ovulatory dysfunction, and polycystic ovarian morphologic features. The diagnostic criteria of the National Institutes of Health (i.e., hyperandrogenism plus ovulatory dysfunction), "classic" polycystic ovary syndrome affects 6 to 10% of women of reproductive age, but the prevalence may be twice as high under the broader Rotterdam criteria. The Rotterdam criteria for PCOS have been endorsed by the National Institutes of Health (NIH). However, Androgen Excess Society (AES) guidelines may correspond better to the pathogenesis of this disorder, as the AES emphasizes the importance of clinical and/or biochemical Polycystic Ovary Morphology (PCOM) [1]. PCOS is inherently associated with metabolic aberrations that include insulin resistance and NAFLD [2,3]. Dyslipidemia is well known risk factor for NAFLD and cardiovascular disorders. The dyslipidemia in NAFLD is characterized by increased serum triglycerides, increased small, dense Low-Density Lipoprotein (LDL) particles, and low High-Density Lipoprotein (HDL) cholesterol [4]. Hypertriglyceridemia and oxidative stress are two independent risk factors inducing inflammatory cascade in the genesis of NASH. In our study we have studied to investigate that hypertriglyceridemia trigger the main events driving the evolution of steatosis to NASH, namely, inflammation and cell death [5]. Based on the pathophysiology of PCOS, various drug regimes have been used as first line therapy in PCOS. Metformin or OCPs have been used to ameliorate the symptoms experienced by women with PCOS. In our study one group of patients received metformin (insulin sensitizing drug) and another group OCPs. We have seen that Oral contraceptive particularly combined ethinyl estradiol and cyproterone acetate has resulted in worsening of lipid profile especially hypertriglyceridemia compared to metformin group with age and BMI matched controls. Hypertriglyceridemia being an important mediator of inflammation and is major risk factor involved in pathway to NAFLD. And we report here that OCPs should be used cautiously in patients with PCOS with NAFLD keeping in view of both being insulin resistant states. Metformin would be drug of choice in these patients.

MATERIALS AND METHODS

We conducted a study at SMHS hospital, GMC Srinagar where we enrolled 100 patients (young Adolescent females) fulfilling the Rotterdam criteria for PCOS which includes that the diagnosis of PCOS must be based on the presence of at least two of the following three criteria: chronic anovulation, hyperandrogenism (clinical or biological), and polycystic ovaries as suggested by European Society for Human Reproduction and Embryology and American Society for Reproductive Medicine. Menstrual disturbances were classified as Oligo-amenorrhea (≤8 cycles/year or menstrual interval ≥ 35 d) and amenorrhea (absence of menses in last 6 or more months). Also Modified Ferriman-Gallwey score by a single observer was used to assess the degree of hirsutism. A score of at least 8 of a total of 36 was taken as significant. All subjects underwent Anthropometric assessment including measurement of body weight (kg), height (cm), Body mass index (BMI (kg/m^2)), and Waist Hip Ratio (WHR). Waist circumference was measured at the umbilical level and hip circumference at the trochanter region. Blood samples were collected from the patients after an overnight fast for the estimation of Tetraiodothyronine (T4), Thyroid Stimulating Hormone (TSH), Leutinizing Hormone (LH), Follicle Stimulating Hormone (FSH), Prolactin (PRL), blood counts, electrolytes, lipids, liver, and kidney functions. Blood samples for hormonal investigations were collected from d 3-7 (early follicular phase) in subjects with spontaneous menstrual cycles. For insulin estimation, samples were collected in ice and plasma was separated immediately in cold centrifuge and stored at –20°C until the assay. A single observer did transabdominal ultrasonography to demonstrate any suggestion of polycystic ovarian morphology, i.e. presence of 10 or more peripheral follicles each measuring 2–8 mm in size with echogenic ovarian stroma and/or increased ovarian volume. The echogenic theca was considered the most specific finding. The study was approved and conducted according to the guidelines of the Institute's Ethics Committee. Two out of 100 patients were excluded owing to high blood glucose. 98 patients completed the study. They were randomized to two equal groups 49 each received metformin and oral contraceptive respectively.

STATISTICAL ANALYSIS

Data was entered in a Microsoft Excel spreadsheet. Continuous variables were summarized as mean and standard deviation. Categorical variables were summarized as percentages. Independence between two categorical variables were tested using chi-square test. Difference between two independent samples means was tested using

unpaired 't' test. Difference between two paired samples means was tested using paired 't' test. Two sided p values were reported and p values < 0.05 were considered statistically significant. Data was analyzed using Statistical Package for Social Sciences (SPSS Ver. 20.0).

RESULTS AND FOLLOW UP

Metformin group (Clinical profile)

Menstrual cycle frequency improved significantly (p = 0.01) with metformin, from 10.47 ± 1.61 to 11.21 ± 0.32 cycles/y at 6 months (Table 1). There was a significant decrease in BMI (p = 0.005) with metformin, from 24.57 ± 6.65 to 20.32 ± 4.51 at 6 months. There was no significant effect on WHR, systolic and diastolic blood pressure, FG Score with 6 months of metformin therapy.

OCPs group (Clinical profile)

Menstrual cycle frequency improved significantly (p = .005) with OCPs, from 8.42 ± 4.80 to 11.11 ± 1.80 cycles per year at 6 months. The hirsutism score decreased (p = .001) gradually from 14.17 ± 4.63 at baseline to 13.53 ± 4.02 at 6 months of therapy. There was no significant effect on WHR, systolic and diastolic blood pressure and BMI with 6 months of OCPs therapy.

Biochemical profile

Patients followed over a period of 6 months. On follow up, we observed that in:

Metformin group

There is statistically significant decrease in fasting blood glucose with metformin, from baseline 103.10 ± 27.95 to 88.17 ± 8.72 (p = 0.007) at 6 months (Table 2). There was significant increase in bilirubin levels from baseline 0.58 ± 0.13 to 0.93 ± 0.45 (p = <0.001) at 6 months of metformin therapy. There was significant increase in LDL levels from baseline 106.53 ± 0.16 to 108.60 ± 0.12 (p = <0.001) at 6 months of metformin therapy. Also there was significant decrease in total cholesterol levels from baseline 160.80 ± 45.88 to 125.00 ± 45.88 (p = 0.003) at 6 months of metformin therapy. There was no significant effect on TP, ALB, ALP, SGOT, SGPT, TG and HDL with 6 months of metformin therapy.

OCPs

There was significant increase in TG levels from baseline 117.67 ± 38.71 to 160.66 ± 37.42 (p = <0.001) at 6 months of OCPs therapy. There was no significant effect on BGF, Bilirubin, TP, ALB, ALP, SGOT, SGPT, Total cholesterol, TG, LDL and HDL with 6 months of OCPs therapy.

Table 1: Clinical profile of patients at baseline and at 6 months after treatment

Metformin (0-6 Months)*		Oral contraceptive (0-6Months)*		Variables*
22.53 ± 3.28	22.53 ± 3.28	21.89 ± 5.29	21.89 ± 5.29	Mean age(years)
13.23 ± 1.07	13.23 ± 1.07	13.17 ± 1.48	13.17 ± 1.48	Age at menarche
0.95 ± 0.12	0.95 ± 0.12	1.00 ± 0.16	1.00 ± 0.16	W\H Ratio(cm)
10.47 ± 1.61	11.21 ± 0.32	8.42 ± 4.80	11.11 ± 1.80	Cycles \year
11.46 ± 4.15	11.4 ± 1.61	14.17 ± 4.63	13.53 ± 4.02	FGS
110.06 ± 10.80	110.13 ± 10.74	107.72 ± 10.46	109.17 ± 8.75	SBP(mmHg)
69.60 ± 9.07	70.80 ± 8.04	70.17 ± 8.79	69.72 ± 7.36	DBP(mmHg)
24.57 ± 6.65	20.32 ± 4.51	24.54 ± 3.61	24.14 ± 2.45	BMI Kg\m2

*Results are given in mean ± SD; p = <0.005

Table 2: Biochemical and Hormonal profile of patients at baseline and at 6 months after treatment.

Metformin		Oral contraceptive		
0 months	6 months	0 months	6 months	Variables*
5.07 ± 2.44	4.06 ± 2.46	3.75 ± 1.42	3.45 ± 1.20	LH
6.56 ± 2.16	6.06 ± 2.10	4.70 ± 1.22	5.02 ± 1.09	FSH
0.77 ± 1.12	0.66 ± 1.17	0.79 ± 1.16	0.68 ± 1.10	LH\FSH
0.53 ± 1.52	0.60 ± 0.87	0.34 ± 0.12	0.14 ± 0.35	TESTO
14.60 ± 8.93	14.16 ± 8.52	15.08 ± 7.09	15.74 ± 6.73	PRL
3.17 ± 1.94	3.14 ± 1.72	2.78 ± 2.39	2.99 ± 1.99	TSH
3.57 ± 2.37	3.57 ± 2.11	3.22 ± 2.53	2.96 ± 2.09	INSULIN(f)
1.42 ± 0.91	0.78 ± 0.52	0.71 ± 0.53	0.63 ± 0.45	HOMA-IR
103.10 ± 27.95	88.17 ± 8.72	87.97 ± 8.79	86.64 ± 9.28	BGF
160.80 ± 45.88	125.00 ± 45.88	154.56 ± 21.56	151.67 ± 22.62	CHOL
125.30 ± 44.75	109.00 ± 24.53	117.67 ± 38.71	160.66 ± 37.42	TG
106.53 ± 0.16	108.60 ± 0.12	114.83 ± 19.41	111.14 ± 18.21	LDL
42.93 ± 6.82	43.57 ± 5.21	43.19 ± 10.15	42.92 ± 10.24	HDL

*Variables Units : [LH(IU\L), FSH(IU\L), LH\FSH, TESTOSTERONE(nmol\L), prolactin (ng\dl), TSH(MIU\L), FASTING INSULIN(min\L), HOMA-IR,BGF(mg\dl), chol, TG, LDL and HDL all measured in mg\dl] [Results are given as mean ± SD; p = <.05. Conversion factors: insulin, pmol/L = IU/mL x 7.175; glucose, mmol/L = mg/dL x 0.0555]

DISCUSSION

Polycystic Ovary Syndrome (PCOS) is a common condition that typically develops in reproductive-age women. It is important to emphasize that PCO morphology is itself a common ultrasonic feature in 20% of reproductive-age women, most of whom have minimal or no symptoms of the syndrome [6], therefore most women with PCO morphology do not have PCOS. Furthermore, PCOS is also a condition that associates with obesity, dyslipidemia, insulin resistance and cardiometabolic risk, although these features do not form part of the diagnostic criteria for PCOS [7]. Metabolic syndrome is more common in women with PCOS compared with BMI-comparable control women. Based on National Cholesterol Education Program Adult Treatment Panel III criteria, 34–46% of US-based Caucasian women with PCOS have metabolic syndrome [8]. Not all women with PCOS are insulin resistant. Previously study done by Dunaif has shown that insulin resistance is independent of obesity and compared Oral Glucose Tolerence Test (OGTT) in lean and obese women with PCOS with matched controls and hirsute women wherein he found an abnormal hyperinsulinimic response in lean PCOS. Similar response was seen with other studies done with the euglycemic clamp technique [9]. NAFLD is associated with reduced insulin sensitivity [10,11]. Nonalcoholic Fatty Liver Disease (NAFLD) is an increasing cause of liver disease that is associated with significant morbidity and mortality, with a global prevalence of 25% [12]. NAFLD is defined as the accumulation of excess triglyceride droplets in the liver (>5% of hepatocytes with droplets detected histologically or >5% proton density fat fraction by Magnetic Resonance Imaging [MRI]) in people who consume little or no alcohol [13] Current study emphasizes relation of insulin resistance and NAFLD in PCOS patient [14,15]. Patients with Nonalcoholic Fatty Liver Disease (NAFLD) often have dyslipidemia along with other features of metabolic syndrome such

as obesity, diabetes mellitus, and hypertension. NAFLD prevalence was estimated at 80%-90% in obese adults and up to 90% in patients with hyperlipidemia [16] . In our study, we have seen worsening of triglyceride levels, secondary to PCOS per se and due to Oral contraceptives, as perpetuating factor in the pathogenesis of NAFL to NAFLD. The association of insulin resistance in the pathophysiology of PCOS has given rise to the use of insulin sensitizing drugs in its treatment. Studies of metformin in both obese and lean PCOS women have documented a significant decrease in fasting insulin and androgen levels, as well as a restoration of menstrual cyclicity [17]. Metformin has also been shown to improve hyperandrogenemia, even in non-obese women with PCOS who appear to have normal metabolic insulin sensitivity. Whether it is the correction of abnormal insulin action per se or the reduction of plasma insulin levels that is responsible for these beneficial effects of insulin sensitizers is currently unclear [18]. In our study it was observed that there was a significant decrease in LDL levels from baseline 108.60 ± 0.12 to 106.53 ± 0.16 (p = <0.001) at 6 months of metformin therapy. Also there was also a significant decrease in total cholesterol levels from baseline 160.80 ± 45.88 to 125.00 ± 45.88 (p = 0.003) at 6 months of metformin therapy. These findings are similar to that observed by Mehandiratta R, et al. [19]. However the decrease in TG and increase in HDL was not statistically significant with 6 months of metformin therapy. No significant effect was observed on LH, FSH, LH/FSH, PRL, T, TSH and fasting insulin levels. It has also been seen that the Hyper androgenic patients have significantly higher level of triglycerides, compared to those with normal androgen concentration [20,21]. Oral Contraceptive use is these patients have been found to be associated with an approximate 50% rise in plasma triglyceride and a 5% to 10% rise in cholesterol [22-25]. In our study, we found that there was a significant increase in TG levels from baseline 117.67 ± 38.71 to 160.66 ± 37.42 (p = <0.001) at 6 months of OCPs therapy. Similar findings have been seen by Costello MF, et al. [26]. However, menstrual cycle frequency improved significantly (p = .005) with OCPs, from 8.42 ± 4.80 to 11.11 ± 1.80 cycles per year at 6 months. Similar findings were reported by Al-Zubeidi H, et al. [27]. The hirsutism score decreased (p = .001) gradually from 14.17 ± 4.63 at baseline to 13.53 ± 4.02 at 6 months of therapy. There was no significant effect on BGF, Bilirubin, TP, ALB, ALP, SGOT, SGPT, total cholesterol, TG, LDL and HDL with 6 months of OCPs therapy.

In Summary, considering that insulin resistance is a common feature of both NAFLD and PCOS, it is an important issue which may have relevance for clinical management in terms of when and how to screen for liver disease in patients with PCOS. Also hypertriglyceridemia as is seen in patients of PCOS, have shown worsening on receiving Oral Contraceptive Pills, are at higher risk of NAFLD and are at worse cardiovascular outcome. This article emphasizes against the role of OCPs in Patients with PCOS with NAFLD.

CONSENT

Informed consent was obtained from patients for study.

AUTHOR STATEMENT

This study was approved by the ethics committee of GMC Srinagar 190010, J&K India.

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