Diabetes & Metabolic Syndrome: Clinical Research & Reviews 13 (2019) 1065–1069

Contents lists available at ScienceDirect

Diabetes & Metabolic Syndrome: Clinical Research & Reviews

journal homepage: www.elsevier.com/locate/dsx

Original Article

Insulin resistance and hyperandrogenemia independently predict nonalcoholic fatty liver disease in women with polycystic ovary syndrome

Sree Harsha Varma, Sunanda Tirupati*, T.V.S. Pradeep, Vijaya Sarathi, Dileep Kumar

Department of Endocrinology, Narayana Medical College and Hospital, Nellore, 524003, India

ARTICLE INFO

Article history:
Received 4 December 2018
Accepted 26 December 2018

Keywords:
Polycystic ovary syndrome
Nonalcoholic fatty liver disease
Insulin resistance
Hyperandrogenemia

ABSTRACT

Aims: To find the prevalence and predictors of nonalcoholic fatty liver disease (NAFLD) in Asian Indian polycystic ovary syndrome (PCOS) women.

Materials and methods: This is a prospective, cross-sectional study conducted at a tertiary care hospital from South India. Sixty women fulfilling the Rotterdam (2003) criteria for PCOS were recruited for the study. All participants were evaluated with ultrasound abdomen for fatty liver and additional biochemical investigations including fasting plasma glucose, postprandial plasma glucose, serum insulin, lipid profile and liver function tests.

Results: The mean age of the study population was 24.06 ± 5.9 (range: 15–39) years. Oligomenorrhea, hirsutism and acne were present in 58 (96.7%), 37 (61.7%) and 33 (55%) women. Mean BMI of the study population was 29.5 ± 5.28 (range: 19.95 to 45.44) kg/m^2. Fifty (83.3%) women were obese (BMI: ≥ 25 kg/m^2). Twenty-three (38.3%) women with PCOS had NAFLD. Three women each had isolated elevation of alanine transaminase (ALT) and aspartate transaminases (AST) whereas three women had elevation of both. All women with elevated transaminases had NAFLD. By univariate analysis, factors associated with NAFLD were serum total cholesterol, serum insulin, HOMA-IR, hyperandrogenism, ALT and AST. On multiple regression analysis using linear regression, HOMA-IR and hyperandrogenemia were the only significant predictors of NAFLD.

Conclusion: Our study reports NAFLD in more than one third of Asian Indian women with PCOS. In addition to insulin resistance (HOMA-IR), hyperandrogenemia is an independent predictor of NAFLD in women with PCOS.

1. Introduction

Polycystic ovary syndrome (PCOS) is the most common endocrinopathy in reproductive aged women, affecting 6–10% of the population [1]. It is a complex endocrine disorder of uncertain etiology [2]. Clinical features include anovulation, hirsutism, menstrual dysfunction, acne and infertility. It is associated with significant metabolic abnormalities. Metabolic abnormalities associated with PCOS include insulin resistance with compensatory hyperinsulinemia, obesity, impaired glucose tolerance and type 2 diabetes mellitus, dyslipidemia, endometrial carcinoma and possibly cardiovascular disease.

Insulin resistance is the main pathogenic feature of PCOS affecting both obese and lean patients [3,4]. Several studies have shown that about 50% of patients with PCOS fall under the criteria for metabolic syndrome (MS) [5], and MS is the major risk factor for the occurrence of cardiovascular disease in PCOS patients [6]. In recent years epidemiological studies have shown an increased risk of non-alcoholic fatty liver disease (NAFLD) among young women with PCOS [7,8]. Insulin resistance, which is strongly associated with PCOS and MS, also plays a key role in the pathogenesis of NAFLD [9].

Nonalcoholic fatty liver disease (NAFLD) is the most common form of liver disease in the Western world with a prevalence ranging from 5 to 33% [10]. It is characterized by accumulation of fat in the liver, with histology identical to alcoholic liver disease, in patients with no or minimal alcohol consumption. There is a wide

* Corresponding author.
E-mail addresses: shvarma87@gmail.com (S. Harsha Varma), sunanda.tirupati@gmail.com (S. Tirupati), doctvspradeep@gmail.com (T.V.S. Pradeep), drvijayasarathi@gmail.com (V. Sarathi), kdileep_1952@yahoo.co.in (D. Kumar).

https://doi.org/10.1016/j.dsx.2018.12.020
1871-4021/

spectrum of disease ranging from the accumulation of lipid within hepatocytes (steatosis) to inflammation of the liver (non-alcoholic steatohepatitis - NASH) to liver fibrosis and finally to cirrhosis [11]. NAFLD, which was previously considered as a benign hepatic manifestation of MS, is now thought to be pathogenic determinant of MS [12]. In fact, some studies have shown that NAFLD precedes the development of the MS [13]. This raises the prospect of NAFLD being used as an early marker of MS.

Many studies have demonstrated high prevalence of PCOS in Indian population, ranging from 2.2% to 26% [14]. These women are at higher risk for developing MS and NAFLD. There is paucity of data regarding the prevalence of these disorders in South Indian population. To bridge this gap, the current study is undertaken to assess the prevalence and predictors of NAFLD in South Indian women with PCOS.

2. Materials and methods

This is a cross sectional study conducted in the department of Endocrinology, Narayana medical college and Hospital (NMCH), Nellore, between September 2016 and September 2017. Sixty newly diagnosed PCOS women attending out patient services at Endocrinology department were included in the study after obtaining a written informed consent. The study was approved by the institutional ethics committee. The diagnosis of PCOS was based on the 2003 Rotterdam criteria:

(i) Oligo- or anovulation
(ii) Biochemical and/or clinical evidence of hyperandrogenism
(iii) Ultrasound evidence of polycystic ovaries.

In addition to the presence of at least two out of the above listed three criteria exclusion of secondary causes of PCOS was essential to make the diagnosis of PCOS.

Patients with known diabetes mellitus, untreated hypothyroidism, hyperprolactinemia, on treatment with drugs like corticosteroids, antiepileptic or antipsychotic drugs, insulin sensitizers, hormonal contraceptives and anti-tuberculosis drugs were excluded from the study. Patients with chronic alcohol intake and those with history of chronic liver disease, chronic viral hepatitis, autoimmune hepatitis, drug/toxin-induced liver injury or hemochromatosis were also excluded from the study.

A detailed history including menstrual cycles, hirsutism, acne, dietary habits, physical activity, family history of PCOS, diabetes and hypertension was taken. Clinical examination for signs of insulin resistance (acanthosis nigricans), hyperandrogenism (hirsutism and acne) and anthropometric variables including height, weight, body mass index (BMI) and waist circumference (WC) was performed. Hirsutism was quantified according to modified Ferriman-Gallwey method. Patient was considered to have mild hirsutism if score is between 8 and 15 and severe hirsutism if score >15. Overweight, obesity and normal weight are graded according World Health Organization cut offs for Asians. (Overweight > 23 kg/m^2 and obese >25 kg/m^2) [15]. Waist circumference (WC) was measured in standing position midway between the lower costal margin and the iliac crest, at the end of expiration. WC $\geq$ 80 cm was considered abnormal.

2.1. Biochemical assays

Venous blood samples for fasting and postprandial plasma glucose, fasting lipid profile, fasting insulin, serum prolactin, thyroid stimulating hormone (TSH), total testosterone, serum total bilirubin, alanine transaminase (ALT) and aspartate transaminase (AST) were collected after an overnight fast between 2nd to 5th day of a spontaneous or progesterone withdrawal-induced menstrual cycle. Biochemical investigations were performed using Humastar 600 whereas hormonal investigations were performed using Beckman Coulter. For all the above parameters, any value more than the upper limit of the laboratory reference range was used as abnormal. For serum insulin assay, analytical sensitivity, reference range and precision were 0.03 μIU/ml, 1.9–23 μIU/ml and <10% across the assay range whereas for serum total testosterone assay, analytical sensitivity, reference range and imprecision were 0.1 ng/ml, <0.1–0.75 ng/ml and ≤20% at 0.5 ng/ml and <10% from 2 to 10 ng/mL. HOMA-IR was calculated by the following formula: serum insulin (μU/ml) × fasting serum glucose (mmol/l)/22.5. A cut-off of ≥2.7 was used to diagnose of insulin resistance.

Ultrasonography was performed by the same radiologist for all 60 cases. A high-resolution Philips HD 5 with a 3.5 Hz convex-array probe was used. Hepatic steatosis was defined as diffuse increase in fine echoes in liver parenchyma with impaired visualization of intra-hepatic vessels and the diaphragm.

2.2. Statistical analysis

All the data values were entered in to MS-Excel worksheet. Statistical analysis was done by using IBM SPSS Version 21.0. Categorical variables were represented as frequencies and percentages whereas continuous variables were expressed as mean and standard deviation (SD). chi-square test was used to compare the categorical variables whereas unpaired t-test was used to compare continuous variables. Odds ratio for NAFLD was calculated for each variable and multiple regression analysis was performed using linear regression method to identify predictors of NAFLD. p value < 0.05 was considered as statistically significant.

3. Results

The study included a total of 60 PCOS women. The mean age of the study population was 24.06 ± 5.9 (range: 15–39) years. Oligomenorrhea, hirsutism and acne were present in 58 (96.7%), 37 (61.7%) and 33 (55%) women. Family history of diabetes mellitus was present in 24 (40%) women. Fifty-six (93.3%) women were consuming non-vegetarian diet. Acanthosis nigricans was present in 53 (88.3%). Mean BMI of the study population was 29.5 ± 5.28 (range: 19.95 to 45.44) kg/m^2. Fifty (83.3%) women were obese whereas another five (8.3%) women were overweight.

Out of the 60 cases, 23 (38.30%) had hepatic steatosis on ultrasound; three women each had isolated elevation of ALT and AST whereas three women had elevation of both enzymes. All women with elevated transaminases had fatty liver.

Oligomenorrhea, hirsutism, acne, non-vegetarian diet habit, family history of diabetes and acanthosis nigricans were not significantly different between NAFLD and non-NAFLD groups. Clinical parameters such as age, BMI, WC, systolic blood pressure and diastolic blood pressure also did not differ between the two groups (Table 1).

By univariate analysis, factors associated with NAFLD in PCOS patients were serum total cholesterol, serum insulin, HOMA-IR, hyperandrogenism, ALT and AST whereas age, BMI, WC and serum triglycerides were not associated with it (see Table 3).

On multiple regression analysis using linear regression, HOMA IR (standardized coefficient β: 0.642, p: < 0.0001) and hyperandrogenemia (standardized coefficient β: 0.2, p: 0.03) were the only significant predictors of NAFLD.

4. Discussion

The present study reports the prevalence of NAFLD along with

Table 1
Comparison of clinical characteristics between NAFLD group with Non NAFLD group.

Parameters	Total	NAFLD group	Non-NAFLD group	P value
Oligomenorrhea	58 (96.7%)	21 (91.3%)	37 (100%)	0.06
Hirsutism	37 (61.7%)	17 (73.9%)	20 (54.1%)	0.12
Acne	33 (55%)	15 (65.2%)	18 (48.6%)	0.21
Non-vegetarian diet	56 (93.3%)	22 (95.7%)	34 (91.9%)	0.57
Family H/o diabetes	24 (40%)	10 (43.5%)	14 (37.8%)	0.66
Acanthosis nigricans	53 (88.3%)	22 (95.7%)	31 (83.8%)	0.16
Age (Years)	24.06 ± 5.96	23.91 ± 6.54	24.16 ± 5.66	0.87
BMI (Kg/m^2)	29.56 ± 5.28	30.58 ± 5.82	28.93 ± 4.89	0.24
Obese: nonobese	50:10	20:3	30:7	0.55
Waist circumference (cm)	95.58 ± 12.95	96.87 ± 13.12	94.78 ± 12.96	0.54
Systolic blood pressure (mmHg)	109.87 ± 10.49	111.74 ± 11.54	108.65 ± 9.76	0.27
Diastolic blood pressure (mmHg)	73.17 ± 8.33	73.91 ± 7.82	72.70 ± 8.70	0.58

Postprandial plasma glucose, serum total cholesterol, serum insulin, HOMA- IR, serum testosterone, serum total bilirubin, AST, and ALT were significantly more in the NAFLD group than the non-NAFLD group (Table 2).

Table 2
Comparison of biochemical characteristics between NAFLD group with Non NAFLD group.

Parameters	NAFLD group	Non-NAFLD group	Total	P value
Fasting plasma glucose (mg/dl)	89.04 ± 9.07	85.48 ± 8.02	86.85 ± 8.55	0.11
Postprandial plasma glucose (mg/dl)	120.35 ± 17.15	109.95 ± 19.83	113.93 ± 19.3	0.04
Serum total cholesterol (mg/dl)	170.35 ± 30.05	150.97 ± 26.77	158.40 ± 29.40	0.01
Serum triglycerides (mg/dl)	143.00 ± 61.82	122.68 ± 55.41	130.47 ± 58.29	0.19
Serum thyroid stimulating hormone (μIU/ml)	2.56 ± 1.43	2.54 ± 1.57	2.55 ± 1.51	0.96
Serum prolactin (ng/ml)	13.00 ± 4.16	11.29 ± 4.32	11.95 ± 4.30	0.13
Serum insulin (μIU/ml)	21.62 ± 4.18	11.86 ± 4.20	15.60 ± 6.34	<0.01
HOMA-IR	4.73 ± 0.91	2.53 ± 1.02	3.37 ± 1.45	<0.01
Serum total testosterone (ng/ml)	0.74 ± 0.24	0.50 ± 0.15	0.59 ± 0.22	<0.01
Serumtotal bilirubin (mg/dl)	0.66 ± 0.24	0.53 ± 0.18	0.58 ± 0.21	0.03
Aspartate transaminase (IU/L)	25.85 ± 7.65	19.49 ± 4.28	21.93 ± 6.54	<0.01
Alanine transaminase (IU/L)	26.87 ± 9.65	18.70 ± 5.34	21.83 ± 8.25	<0.01

Table 3
Univariate analysis of risk factors for NAFLD.

Variable	Odds Ratio	95% confidence interval	P value
Age (years)	0.99	0.90–1.08	0.87
Weight (kg)	1.03	0.99–1.07	0.09
Body mass index (kg/m2)	1.06	0.95–1.17	0.24
Waist circumference (cm)	1.01	0.97–1.05	0.54
Serum total cholesterol (mg/dl)	1.02	1.00–1.04	0.01
Serum triglycerides (mg/dl)	1.00	0.99–1.01	0.19
Fasting plasma glucose (mg/dl)	1.05	0.98–1.21	0.12
Postprandial plasma glucose (mg/dl)	1.03	0.99–1.06	0.053
Serum insulin (μIU/ml)	1.82	1.32–2.51	<0.01
HOMA-IR	9.59	2.98–30.87	<0.01
Hyperandrogenism	13.46	2.59–68.83	<0.01
Serum total bilirubin (mg/dl)	0.93	0.74–1.18	0.59
Aspartate transaminase (U/L)	1.21	1.07–1.35	<0.01
Alanine transaminase (U/L)	1.16	1.06–1.28	<0.01

clinical and biochemical risk factors for NAFLD in South Indian PCOS women. Prevalence of fatty liver in the present study was 38.3% which is similar to other studies by Cerda et al. [16] (41.46%), Ma et al. [17] (39%), Qu et al. [18](33%) and Vassilatou et al. [19] (36.84%) but less than the prevalence seen in two previous Indian studies by Karoli et al. [20] (66.67%) and Prasad et al. [21](68%). We used ultrasonography, the most widely used imaging method for detecting fatty liver. It is cost effective, easily available, non-invasive and has an acceptable sensitivity of more than 80% (in the presence of >30% of fatty infiltration) for detecting fatty liver. Although liver biopsy remains the gold standard for NAFLD diagnosis there is still no consensus regarding the absolute need of liver biopsy to confirm the diagnosis of NAFLD [22,23].

Most of the studies have shown association of NAFLD with obesity in PCOS women [16,17,20,24]. Anthropometric markers of obesity as well as clinical features of PCOS did not differ between PCOS women with and without NAFLD [25]. Lack of correlation between NAFLD and anthropometric markers of obesity may be due to predominance of obese PCOS women in our study. In few studies anthropometric obesity markers were not independent predictors of NAFLD among PCOS women which is in agreement with our observation [26]. Nonobese women with PCOS are also at approximately 2-fold increased risk for NAFLD [27].

Association of NAFLD with lipid parameters in PCOS women is variable across the studies. In a study by Faisal et al., [28] serum total cholesterol but not serum triglycerides, was significantly associated with NAFLD in PCOS women. However, in our study neither serum total cholesterol nor serum triglycerides were associated with NAFLD. Similar finding has been reported previously [29].

The prevalence of elevated liver enzymes in women with PCOS varies among different studies. This may be due to different cut-off levels used and population selected. In the current study, AST and ALT were elevated in 10% each, whereas 5% had elevation of both enzymes. There was a significant difference in ALT and AST levels between those with NAFLD and those without. Similar findings have been reported by most of the previous studies [16,20,21,29,30]. All cases with elevated transaminases had hepatic steatosis but in many patients with NAFLD liver enzymes were normal. This is because enzyme elevation is seen only in patients with steatohepatitis which is advanced stage of NAFLD. It suggests poor sensitivity of liver enzymes in diagnosing NAFLD.

Insulin resistance and the consequent hyperinsulinemia are the major pathogenic factors in the steroidogenic and metabolic dysregulation seen in PCOS. In the current study, both were significantly higher in PCOS women with NAFLD than those without. Like our study several studies have demonstrated lower insulin sensitivity among PCOS women than controls [31,32]. In our study insulin resistance was present in 65% PCOS women. Similar high rates of insulin resistance have been demonstrated in many other studies [31,33]. However, among lean PCOS women prevalence of insulin resistance is less [29]. HOMA-IR was significantly higher among PCOS women with NAFLD than those without and was the strongest predictor of NAFLD in our study. Most of the previously published studies have demonstrated a similar finding [20,21,29,30].

Serum testosterone and frequency of hyperandrogenemia were significantly higher in PCOS women with NAFLD than those without. Hyperandrogenemia was an independent predictor of NAFLD among PCOS women. Many recent studies have demonstrated the association of hyperandrogenemia with NAFLD in PCOS women [20,21,28,34]. A recent meta-analysis has also confirmed hyperandrogenemia as an independent predictor of NAFLD in PCOS women [35]. Association of hyperandrogenemia with other cardiometabolic risk factors such as elevated triglycerides, serum insulin, HOMA-IR, mean arterial pressure, increased glucose and decreased high-density lipoprotein has also been reported. Association of hyperandrogenemia with NAFLD may be a reflection of obesity and insulin resistance in PCOS [36]. However, in our study association of hyperandrogenemia was independent of BMI and HOMA-IR. It has been shown in experimental models that treatment with dihydrotestosterone (DHT) in ovariectomised mice leads to greater visceral fat mass and liver fat associated with triglycerides accumulation. Adenosine monophosphate-activated protein kinase (AMPK) is an enzyme known to inhibit lipogenesis in adipose tissue and its inhibition by excess androgens may be responsible for increased fat accumulation in the liver and visceral fat [37,38]. A recent study has also demonstrated that hCG induced hyperandrogenism strongly aggravates hepatic inflammation which might be mediated by dysregulation of IRS-PI3K-Akt signaling axis [39].

Serum total testosterone measurement is the most commonly used assay to determine hyperandrogenism. However, it may not be a sensitive marker of hyperandrogenemia due to reduced SHBG in PCOS women. Although, free testosterone is a more sensitive measure of hyperandrogenism, direct free testosterone assays are not used as they are inaccurate and lack standardisation. Hence, free androgen index (FAI) may be more reliable biochemical marker of androgen excess in PCOS women [40]. However, we did not measure free testosterone or FAI in our study. Measurement of FAI might have provided a stronger association of androgen excess with NAFLD.

The study had few limitations. First, sample size of the study was relatively smaller. Second, though the prevalence of NAFLD was high among PCOS women in our study, it is not clear whether it was higher than that in women without PCOS. Third, liver biopsy, the gold standard for diagnosis of NAFLD, was not performed since it is not feasible to perform liver biopsy in all women with PCOS. However, multiple alternative criteria have been proposed including those based on ultrasonogram [41]. In our study ultrasonogram was used as the sole criteria to diagnose NAFLD which might have underestimated the prevalence of NAFLD since the sensitivity of ultrasound for mild NAFLD (<30% hepatocytes involved) is low (60–90%) [41]. Fourth, due to higher prevalence of obese and overweight women in our study, the study findings may not be generalised to lean PCOS.

5. Conclusion

Our study reports NAFLD in more than one third of women with PCOS. In addition to insulin resistance (HOMA-IR), hyperandrogenemia was an independent predictor of NAFLD in women with PCOS.

References

[1] Bozdag G, Mumusoglu S, Zengin D, Karabulut E, Yildiz BO. The prevalence and phenotypic features of polycystic ovary syndrome: a systematic review and meta-analysis. Hum Reprod 2016;31:2841–55.
[2] Azziz R, Woods KS, Reyna R, Key TJ, Knochenhauer ES, Yildiz BO. The prevalence and features of the polycystic ovary syndrome in an unselected population. J Clin Endocrinol Metab 2004;89:2745–9.
[3] Dunaif A, Segal KR, Futterweit W, Dobrjansky A. Profound peripheral insulin resistance, independent of obesity, in polycystic ovary syndrome. Diabetes 1989;38:1165–74.
[4] Diamanti-Kandarakis Christako. Prevalence, definition and clinical manifestations of polycystic ovary syndrome. Endocrinol Nutr 2006;53(Suppl 1): 25–33.
[5] Essah PA, Nestler JE. The metabolic syndrome in polycystic ovary syndrome. J Endocrinol Invest 2006;29:270–80.
[6] Cussons AJ, Stuckey BG, Watts GF. Cardiovascular disease in the polycystic ovary syndrome: new insights and perspectives. Atherosclerosis 2006;185: 227–39.
[7] Daskalopoulos G, Karkanaki A, Piouka A, Prapas N, Panidis D, Gkeleris P, et al. Excess metabolic and cardiovascular risk is not manifested in all phenotypes of polycystic ovary syndrome: implications for diagnosis and treatment. Curr Vasc Pharmacol 2015;13:788–800.
[8] Nascimbeni F, Ballestri S, Di Tommaso L, Piccoli M, Lonardo A. Inflammatory hepatocellular adenomatosis, metabolic syndrome, polycystic ovary syndrome and non-alcoholic steatohepatitis: chance tetrad or association by necessity? Dig. Liver Dis 2014;46:288–9.
[9] Kleiner DE, Brunt EM, Van Natta M, Behling C, Contos MJ, Cummings OW, , et alNonalcoholic Steatohepatitis Clinical Research Network. Design and validation of histological scoring system for non-alcoholic fatty liver disease. Hepatology 2005;41:1313. 12.
[10] Farrell GC, Larter CZ. Nonalcoholic fatty liver disease: from steatosis to cirrhosis. Hepatology 2006;43(2 Suppl 1):99–112.
[11] Levene AP, Goldin RD. The epidemiology, pathogenesis and histopathology of fatty liver disease. Histopathology 2012;61:141–52.
[12] Anstee QM, Targher G, Day CP. Progression of NAFLD to diabetes mellitus, cardiovascular disease or cirrhosis. Nat Rev Gastroenterol Hepatol 2013;10: 330–44.
[13] Lonardo A, Ballestri S, Marchesini G, Angulo P, Loria P. Nonalcoholic fatty liver disease: a precursor of the metabolic syndrome. Dig Liver Dis 2015;47: 181–90.
[14] Nidhi R, Padmalatha V, Nagarathna R, Amritanshu R. Prevalence of polycystic ovarian syndrome in Indian adolescentes. J Pediatr Adolesc Gynaecol 2011;24: 223–7.
[15] Chen MJ, Chiu HM, Chen CL, Yang WS, Yang YS, Ho HN. Hyperandrogenemia is independently associated with elevated alanine aminotransferase activity in young women with polycystic ovary syndrome. J Clin Endocrinol Metab 2010;95:3332–41.
[16] Cerda C, Pérez-Ayuso RM, Riquelme A, Soza A, Villaseca P, Sir-Petermann T, et al. Nonalcoholic fatty liver disease in women with polycystic ovary syndrome. J Hepatol 2007;47:412–7.
[17] Ma RC, Liu KH, Lam PM, Cheung LP, Tam WH, Ko GT, et al. Sonographic measurement of mesenteric fat predicts presence of fatty liver among subjects with polycystic ovary syndrome. J Clin Endocrinol Metab 2011;96:799–807.
[18] Qu Z, Zhu Y, Jiang J, Shi Y, Chen Z. The clinical characteristics and etiological study of nonalcoholic fatty liver disease in Chinese women with PCOS. Iran J Reproductive Med 2013;11:725–32.
[19] Vassilatou E, Lafoyianni S, Vryonidou A, Ioannidis D, Kosma L, Katsoulis K, et al. Increased androgen bioavailability is associated with non-alcoholic fatty liver disease in women with polycystic ovary syndrome. Hum Reprod 2010;25:212–20.

[20] Karoli R, Fatima J, Chandra A, Gupta U, Islam FU, Singh G. Prevalence of hepatic steatosis in women with polycystic ovary syndrome. J Hum Reprod Sci 2013;6:9–14.
[21] Srinivas Prasad RH, Balakrishna BV, Kudva N, Sandhya H, Ramakrishna P. Incidence of non-alcoholic hepatic fatty infiltration in women with polycystic ovary syndrome. Journal of Evidence Based Medicine and Healthcare October 15, 2014;1(8):867–75.
[22] Joy D, Thava VR, Scott BB. Diagnosis of fatty liver disease: is biopsy necessary? Eur J Gastroenterol Hepatol 2003;15:539–43.
[23] Lewis JR, Mohanty SR. Nonalcoholic fatty liver disease: a review and update. Dig Dis Sci 2010;55:560–78.
[24] Gambarin-Gelwan M, Kinkhabwala SV, Schiano TD, Bodian C, Yeh HC, Futterweit W. Prevalence of nonalcoholic fatty liver disease in women with polycystic ovary syndrome. Clin Gastroenterol Hepatol 2007;5:496–501.
[25] Daan NM, Jaspers L, Koster MP, Broekmans FJ, de Rijke YB, Franco OH, et al. Androgen levels in women with various forms of ovarian dysfunction: associations with cardiometabolic features. Hum Reprod 2015;30:2376–86.
[26] Macut D, Tziomalos K, Božić-Antić I, Bjekić-Macut J, Katsikis I, Papadakis E, et al. Non-alcoholic fatty liver disease is associated with insulin resistance and lipid accumulation product in women with polycystic ovary syndrome. Hum Reprod 2016;31:1347–53.
[27] Kim JJ, Kim D, Yim JY, Kang JH, Han KH, Kim SM, et al. Polycystic ovary syndrome with hyperandrogenism as a risk factor for non-obese non-alcoholic fatty liver disease. Aliment Pharmacol Ther 2017;45:1403–12.
[28] Faisal A, Gadallah AN, Zyiton A, Eskander A. Liver affection in polycystic ovary syndrome (PCOS). Med J Cairo Univ 2012;80:117–22.
[29] Mehrabian F, Jahanmardi R. Nonalcoholic fatty liver disease in a sample of Iranian women with polycystic ovary syndrome. Int J Prev Med 2017;8:79.
[30] Brown AJ, Tendler DA, McMurray RG, Setji TL. Polycystic ovary syndrome and severe nonalcoholic steatohepatitis: beneficial effect of modest weight loss and exercise on liver biopsy findings. Endocr Pract 2005;11:319–24.
[31] DeUgarte CM, Bartolucci AA, Azziz R. Prevalence of insulin resistance in the polycystic ovary syndrome using the homeostasis model assessment. Fertil Steril 2005;83:1454–60.
[32] Wongwananuruk T, Rattanachaiyanont M, Leerasiri P, Indhavivadhana S, Techatraisak K, Angsuwathana S, et al. The usefulness of homeostatic measurement assessment-insulin resistance (HOMA-IR) for detection of glucose intolerance in Thai women of reproductive age with polycystic ovary syndrome. Internet J Endocrinol 2012, 571035. 6 pages, 2012.
[33] Alebic MS, Bulum T, Stojanovic N, Duvnjak L. Definition of insulin resistance using the homeostasis model assessment (HOMA-IR) in IVF patients diagnosed with polycystic ovary syndrome (PCOS) according to the Rotterdam criteria. Endocrine 2014;47:625–30.
[34] Kumarendran B, O'Reilly MW, Manolopoulos KN, Toulis KA, Gokhale KM, Sitch AJ, et al. Polycystic ovary syndrome, androgen excess, and the risk of nonalcoholic fatty liver disease in women: a longitudinal study based on a United Kingdom primary care database. PLoS Med 2018;15, e1002542.
[35] Rocha ALL, Faria LC, Guimarães TCM, Moreira GV, Cândido AL, Couto CA, et al. Non-alcoholic fatty liver disease in women with polycystic ovary syndrome: systematic review and meta-analysis. J Endocrinol Invest 2017;40:1279–88.
[36] Bohdanowicz-Pawlak A, Lenarcik-Kabza A, Brona A, Kuliczkowska-Płaksej J, Łaczmański Ł, Zaleska-Dorobisz U, et al. Non-alcoholic fatty liver disease in women with polycystic ovary syndrome - clinical and metabolic aspects and lipoprotein lipase gene polymorphism. Endokrynol Pol 2014;65(6):416–21.
[37] McInnes KJ, Corbould A, Simpson ER, Jones ME. Regulation of adenosine 5', monophosphate-activated protein kinase and lipogenesis by androgens contributes to visceral obesity in an estrogen-deficient state. Endocrinology 2006;147:5907–13.
[38] Kanaya N, Vonderfecht S, Chen S. Androgen (dihydrotestosterone)-mediated regulation of food intake and obesity in female mice. J Steroid Biochem Mol Biol 2013;138:100–6.
[39] Zhang Y, Meng F, Sun X, Sun X, Hu M, Cui P, et al. Hyperandrogenism and insulin resistance contribute to hepatic steatosis and inflammation in female rat liver. Oncotarget 2018;9:18180–97.
[40] Cai J, Wu CH, Zhang Y, Wang YY, Xu WD, Lin TC, et al. High-free androgen index is associated with increased risk of non-alcoholic fatty liver disease in women with polycystic ovary syndrome, independent of obesity and insulin resistance. Int J Obes 2017;41:1341–7.
[41] Macut D, Božić-Antić I, Bjekić-Macut J, Tziomalos K. MANAGEMENT OF ENDOCRINE DISEASE: polycystic ovary syndrome and nonalcoholic fatty liver disease. Eur J Endocrinol 2017;177:R145–58.