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Journal
Liver International, 40(2)

ISSN
1478-3223

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Publication Date
2020-02-01

DOI
10.1111/liv.14279

Peer reviewed

HHS Public Access

Author manuscript

Liver Int. Author manuscript; available in PMC 2021 February 01.

Published in final edited form as:

Liver Int. 2020 February ; 40(2): 355–359. doi:10.1111/liv.14279.

Polycystic Ovary Syndrome (PCOS) Is Associated With NASH Severity and Advanced Fibrosis

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Abstract

Background—Polycystic ovary syndrome (PCOS) affects 10% of reproductive-aged women, and is marked by irregular menses and high androgens. PCOS is a known risk factor for imaging-confirmed steatosis, and we now aim to evaluate whether PCOS influences histologic severity of non-alcoholic fatty liver disease (NAFLD).

Methods—Retrospective study of women ages 18–45 years with biopsy-confirmed NAFLD between 2008–2019. Metabolic co-morbidities were captured within 6 months of biopsy. Histologic features of non-alcoholic steatohepatitis (NASH) were independently evaluated by two pathologists blinded to PCOS status.

Results—Among 102 women meeting study criteria, 36% (n=37) had PCOS; median age was 35 years; 27% were white, 6% black, 19% Asian, and 47% reported Hispanic ethnicity. Women with PCOS had higher LDL (123 vs 101 mg/dL, p=0.02) and BMI (38 vs 33 kg/cm^2, p<0.01). NASH was present in 76% of women with PCOS vs 66% without PCOS (p=0.3), and a higher proportion with PCOS had severe ballooning (32 vs 13%, p=0.02), presence of any fibrosis (84 vs 66%, p=0.06), and advanced fibrosis (16 vs 6%, p=0.10). Adjusted for age and BMI, PCOS remained associated with severe hepatocyte ballooning (OR 3.4, 95% CI 1.1–10.6, p=0.03) and advanced fibrosis (OR 7.1, 95% CI 1.3–39, p=0.02). Among women with advanced fibrosis, median age was 5 years younger in those with as compared to those without PCOS (40 vs 45 years, p=0.02).

Conclusion—PCOS is independently associated with more severe NASH, including advanced fibrosis. Hepatologists should routinely inquire about PCOS in reproductive-aged women with NAFLD, and also evaluate for more severe liver disease in this population.

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Conflicts of interest: There are no other relevant conflicts of interest from co-authors.

Lay summary:

Polycystic ovary syndrome (PCOS) is a common condition in young women, and about half of these women have fatty liver on imaging tests. In the current study we studied young women with liver biopsies confirming fatty liver and found that having PCOS also increased their risk for more severe damage to the liver, including more liver scarring.

Keywords

Women; PCOS; NAFLD; Testosterone; Fibrosis

INTRODUCTION:

Polycystic ovary syndrome (PCOS) is a common endocrinopathy affecting ~ 10% of reproductive-aged women.[1] Most women with PCOS have insulin resistance and elevated androgens, such as high testosterone levels.[2] Other metabolic comorbidities are also more common in women with PCOS, including dyslipidemia and obesity.[2] This aberrant metabolic and hormonal milieu appears to increase their risk for nonalcoholic fatty liver disease (NAFLD), which is present in 40–50% of women with PCOS. [3–5] Indeed, PCOS is now recognized as a distinct at-risk group for NAFLD.[6]

Whether PCOS is also associated with more clinically relevant manifestations of NAFLD, including nonalcoholic steatohepatitis (NASH) and NASH-associated fibrosis is not clear. We therefore aimed to evaluate whether PCOS was associated with more severe NAFLD histology as compared to reproductive-aged women without PCOS. If identified, such differences would support the need for more routine inquiry of PCOS in young women presenting with NAFLD, as well as need to evaluate for more severe liver disease within this young population. Such findings would also support efforts to evaluate NASH treatments in this hormonally and metabolically distinct risk group.

METHODS:

Study Design and Patient Population-

This was a retrospective, single center study of reproductive-aged women (ages 18–45 years) with biopsy-confirmed NAFLD between 2008–2019 (n=110). Presence of PCOS was identified by chart review of corresponding ICD-9/10 diagnosis codes and confirmation of PCOS status by Rotterdam criteria.[7] Women with suspected PCOS without confirmed diagnosis by Rotterdam criteria were excluded. Metabolic co-morbidities were captured from most recent timepoint within 6 months before or after liver biopsy, including body mass index (BMI), fasting lipid panels (low-density lipoprotein (LDL), high-density lipoprotein (HDL), triglycerides), and diabetes history (types 1 or 2). Women with other causes of fatty liver were excluded, such as more than one alcoholic drink per day, human immunodeficiency virus, use of tamoxifen, valproic acid, or diltiazem within 6 months prior to liver biopsy. PCOS-related medications including hormonal contraception or metformin use were captured as “ever” if any documentation of prior use, or ”recent” if at least one month of use within 6 months prior to liver biopsy.

NAFLD Assessment-

Histologic assessment including, NASH diagnosis, was determined by consensus between two independent pathologists blinded to PCOS status. Findings were scored using the NAFLD Activity Score (NAS), a composite score ranging from 0 to 8 points composed of steatosis (0–3), hepatocyte ballooning (0–2) and lobular inflammation scores (0–3).[8] Ballooning was also classified as “severe” or “not severe” based on detection at low power magnification (i.e. with a 4x objective). Stage of fibrosis was also assessed on each biopsy according to the Nonalcoholic Steatohepatitis Clinical Research Network (NASH CRN) methodology.[8]

Statistical Analysis-

Group comparisons were performed by two-sided t tests, Mann-Whitney rank sum tests, and $\chi 2$ tests as appropriate. Results were reported as median (interquartile range, (IQR)). Logistic regression was used to assess the association of PCOS with presence of NASH and advanced fibrosis, though given similar proportion of patients with NASH by PCOS status, this model was not further pursued. As a post hoc analysis, the association of PCOS with severe hepatocyte ballooning was also evaluated given observed differences in this histologic feature by PCOS status. A subgroup analysis was also performed among women with hyperandrogenic PCOS defined by Rotterdam criteria including either biochemical evidence (ever elevated androgen level above reference range) or clinical evidence of hyperandrogenism (androgenic alopecia or hirsutism)(n=31).[9] P-values <0.05 were considered statistically significant. Analyses were performed using Stata 15.0.

RESULTS:

Of the 102 reproductive-aged women meeting study inclusion criteria, 36% (n=37) had PCOS. Women with PCOS were younger (median age 33 vs 36 years p=0.02), had higher median BMI (38 kg/m^2 versus 33 kg/m^2, p<0.01), LDL levels (122mg/dL vs 102mg/dL, p=0.05), and numerically more diabetes (41% vs 31%, p=0.3) (Table 1). Race/ethnicity and frequency of other metabolic comorbidities were similar between women with and without PCOS. Among women with PCOS, 68% (n=25) had prior use of hormonal contraception, with 67% having at least one month of use within 6 months prior to biopsy. Prior metformin use was present in 65% of women with PCOS (n=24), with 43% (n=16) having at least one month of use during the 6 months prior to biopsy.

Histologic criteria for NASH (i.e. steatosis >5%, ballooned hepatocytes, and lobular inflammation) were met in 76% of women with PCOS vs 66% without PCOS (p=0.3), and median NAFLD Activity Scores (NAS) were similar (Table 2). The proportion with severe steatosis was similar between groups. The proportion of women with any ballooned hepatocytes was also similar, though more women with PCOS had evidence of severe hepatocyte ballooning (32% vs 13%, p=0.02). Presence and severity of lobular and portal inflammation was also similar (p values > 0.5). A numerically higher proportion of women with PCOS had hepatic fibrosis (84% vs 66%, p=0.06) and advanced fibrosis, defined as stage 3 or 4 (16% vs 6%, p=0.10). Importantly, among women with advanced fibrosis, the

median age of those with PCOS was 5 years younger than women without PCOS (40 vs 45 years, p<0.01).

On univariate analysis PCOS was associated with severe hepatocyte ballooning (OR 3.4, 95% CI 1.2–9.2, p=0.02, which persisted after adjusting for age and BMI (OR 3.4, 95% CI 1.1–10.6, p=0.03) (Table 3). On unadjusted analysis the association of PCOS with advanced fibrosis did not reach statistical significance (OR 3.0, 95% CI 0.78–11.2), p=0.11), but was significant on adjusted analysis (AOR 7.1, 95% CI 1.3–39, p=0.02) (Table 4). A sensitivity analysis comparing 31 women with confirmed hyperandrogenic PCOS to women without PCOS was also performed. Women with hyperandrogenic PCOS had numerically more NASH (81% vs 67%, p=0.12) and were more likely to have severe hepatocyte ballooning (29% vs 12%, p=0.05). A higher proportion also hepatic fibrosis (87% vs 66%, p=0.03), with numerically higher number with advanced fibrosis (13% vs 6%, p=0.26) (Supplemental Table 1). Estimates for severe hepatocyte ballooning were attenuated on adjusted analysis (OR 3.1, 95% CI 0.93–10.3, p=0.07), although hyperandrogenic PCOS maintained a statistically significant association with advanced fibrosis on adjusted analysis (OR 9.5 95% CI 1.4–63, p=0.02) (Supplemental Table 2).

Among women with PCOS, the proportion with severe hepatocyte ballooning was similar by ever (35% vs 25%), or recent hormonal contraception use (32% vs 25%), as well as ever (30% vs 38%), or recent metformin use (35% vs 29%), all p values >0.60. A numerically smaller proportion of PCOS patients had advanced fibrosis among those with ever (12% vs 25%), or recent hormonal contraception use (13% vs 25%), p values > 0.32, while a higher proportion had advanced fibrosis with ever (26% vs 0%, p=0.12), or recent metformin use (25% vs 10%, p=0.21).

DISCUSSION:

PCOS is a recognized risk group for imaging-confirmed NAFLD[6], although less is known about its association with the more clinically relevant manifestation of NASH. In the current study of reproductive-aged women with biopsy-confirmed NAFLD, PCOS was independently associated with more severe NASH and advanced fibrosis. Among women with advanced fibrosis, those with PCOS were also five years younger than those without PCOS, suggesting a potential “head start” in NASH progression.

We found that over one third of women with biopsy-confirmed NAFLD carried a diagnosis of PCOS in our study, which is higher than the ~ 10% estimate of PCOS prevalence in the general population.[1] Our findings likely relate to shared metabolic risk factors for PCOS and NAFLD, as insulin resistance is a hallmark feature of PCOS, with a 2-fold higher rate of progression from insulin resistance to frank diabetes than age-matched women without PCOS.[10] Other NAFLD risk factors are also more common in women with PCOS, including higher prevalence of obesity and dyslipidemia than age-matched non PCOS controls.[11] This adverse metabolic profile likely sets the stage for early onset NAFLD, which is seen in 40–55% of these young women.[4,5] The current study expands upon our existing knowledge of NAFLD in women with PCOS by further demonstrating their increased risk for more severe NASH histology.

Beyond concurrent metabolic risk factors, the high testosterone state of PCOS may further promote NAFLD/NASH in these young women. We have previously shown higher levels of testosterone in women to confer increased risk of imaging-confirmed NAFLD, independent of insulin resistance, obesity, and dyslipidemia.[3] Interestingly, visceral adiposity appears to be an important mediator of this relationship between testosterone and NAFLD in women.[3] Likewise, a recent study from the United Kingdom included over 63,000 women with PCOS as well as non-PCOS controls, and found the high testosterone phenotype of PCOS to confer a more than 2-fold higher odds of prevalent NAFLD.[12] Given the potential role of testosterone in NAFLD, we also conducted a sensitivity analysis among women with hyperandrogenic PCOS. We did find even more pronounced association of hyperandrogenic PCOS with advanced fibrosis, though given smaller sample size, confidence intervals were wide. Our findings do lend credence to prior publications supporting a role of testosterone in the pathogenesis of NAFLD in PCOS, and the need for larger studies evaluating the potential mechanism linking testosterone to NASH histology in women.

There have been limited prior studies of histologically-confirmed NASH, or biomarkers of NASH in women with PCOS. In a study by Hossain et al, 25 women with PCOS and biopsy-confirmed NAFLD were matched to 25 non-PCOS controls, and NASH was identified in twice as many women with PCOS (44% vs 21%, p=0.08).[13] Additional histologic features were not reported in that study. In the current study we noted a higher proportion of women with NASH, present in 76% of women with PCOS and 66% of non PCOS controls, which may relate to differences in study settings, as our sample derived from a tertiary care center. Interestingly, our study sample was younger, though baseline alanine aminotransferase levels were ~ 70IU/ml compared to median of 40 IU/ml in the study by Hossain et al, suggesting more severe disease in our population. The proportion of women with diabetes was similar, though our cohort did have higher triglyceride levels. Two prior studies have reported caspase cleaved fragment cytokeratin 18 (CK18) levels as a biomarker of NASH in women with PCOS. In a smaller, uncontrolled study of women with PCOS and without known liver disease, ALT and/or CK18 levels were elevated in ~ 20% of women.[14] In a controlled study including 192 women with PCOS and 73 age-matched controls, CK18 levels were significantly higher in women with PCOS, independent of BMI, suggesting their higher risk for NASH.[15] The current study supports these findings by confirming the increased risk for histologically significant liver injury and fibrosis in women with PCOS.

There are some notable limitations as well as strengths of our study. Given the modest sample size, we were unable to adjust for comprehensive metabolic covariates beyond age and BMI, although only these two variables were significantly different between groups. Insulin resistance is a hallmark feature of PCOS, and we lacked fasting insulin and glucose levels to assess for insulin resistance, which may be an important driver of our findings. There was a numerically higher proportion of PCOS patients with advanced fibrosis among those with metformin use, which likely reflects a surrogate of insulin resistance. Our study was also conducted at a tertiary care center with a dedicated PCOS clinic, therefore the high proportion of women with PCOS (36%) could reflect referral bias. Nonetheless, having a study population enriched with PCOS increased our power detect important histologic differences between groups. Additional strengths of our study include independent slide review by two pathologists blinded to PCOS status, as well as with expertise in NAFLD

pathology to facilitate calculation of NAFLD Activity Scores and comprehensive histologic features of NAFLD. Inclusion of a non-PCOS control population also lends confidence to our findings. Though our sample was modest, this is the largest study to date characterizing histologic features of NAFLD in women with PCOS.

In summary, women with PCOS and fatty liver have more severe NASH histology than women without PCOS, as well as more advanced disease at a younger age. PCOS appears to represent a distinct at-risk group, not only for imaging-confirmed steatosis, but for more severe histologic manifestations of NAFLD. Consequently, NASH treatment studies should consider this metabolically and hormonally distinct group of young women, who appear at early risk for disease progression. Hepatologists should also inquire about irregular menses and hirsutism in their evaluation of reproductive-aged women with suspected NAFLD, given the potential implications of PCOS on long-term liver health.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgements:

We also thank the UCSF Liver Center for research support (P30 DK026743).

Funding support: This study was funded with support from a K23 award from National Institutes of Health to M.S (DK111944).

M.S. is a site principal investigator for a clinical trial funded by Zydus Pharmaceuticals. N.T. is on the advisory board at Intercept Pharmaceuticals and has institutionalization grant support from Gilead Sciences.

Abbreviations:

PCOS	Polycystic Ovary Syndrome
NAFLD	Nonalcoholic Fatty Liver Disease
NASH	Nonalcoholic Steatohepatitis
BMI	Body Mass Index
LDL	Low Density Lipoprotein
HDL	High Density Lipoprotein
NAS	NAFLD Activity Score
NASH CRN	Nonalcoholic Steatohepatitis Clinical Research Network
IQR	interquartile range
CK18	caspase cleaved fragment cytokeratin 18

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. Human reproduction. 2016;31(12):2841–2855. [PubMed: 27664216]
2. Goodman NF, Cobin RH, Futterweit W, Glueck JS, Legro RS, Carmina E. American Association of Clinical Endocrinologists, American College of Endocrinology, and Androgen Excess and Pcos Society Disease State Clinical Review: Guide to the Best Practices in the Evaluation and Treatment of Polycystic Ovary Syndrome - Part 1. Endocrine practice : official journal of the American College of Endocrinology and the American Association of Clinical Endocrinologists. 2015;21(11): 1291–1300.
3. Sarkar M, Wellons M, Cedars MI, et al. Testosterone Levels in Pre-Menopausal Women are Associated With Nonalcoholic Fatty Liver Disease in Midlife. The American journal of gastroenterology. 2017;112(5):755–762. [PubMed: 28291240]
4. Cerda C, Perez-Ayuso RM, Riquelme A, et al. Nonalcoholic fatty liver disease in women with polycystic ovary syndrome. Journal of hepatology. 2007;47(3):412–417. [PubMed: 17560682]
5. 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. Clinical gastroenterology and hepatology : the official clinical practice journal of the American Gastroenterological Association. 2007;5(4):496–501. [PubMed: 17287148]
6. Chalasani N, Younossi Z, Lavine JE, et al. The diagnosis and management of nonalcoholic fatty liver disease: Practice guidance from the American Association for the Study of Liver Diseases. Hepatology. 2018;67(1):328–357. [PubMed: 28714183]
7. Rotterdam EA-SPcwg. Revised 2003 consensus on diagnostic criteria and long-term health risks related to polycystic ovary syndrome (PCOS). Human reproduction. 2004;19(1):41–47. [PubMed: 14688154]
8. Kleiner DE, Brunt EM, Van Natta M, et al. Design and validation of a histological scoring system for nonalcoholic fatty liver disease. Hepatology. 2005;41(6):1313–1321. [PubMed: 15915461]
9. Cook H, Brennan K, Azziz R. Reanalyzing the modified Ferriman-Gallwey score: is there a simpler method for assessing the extent of hirsutism? Fertility and sterility. 2011;96(5):1266–1270 e1261. [PubMed: 21924716]
10. Legro RS, Gnatuk CL, Kunselman AR, Dunaif A. Changes in glucose tolerance over time in women with polycystic ovary syndrome: a controlled study. The Journal of clinical endocrinology and metabolism. 2005;90(6):3236–3242. [PubMed: 15797965]
11. Goodman NF, Cobin RH, Futterweit W, Glueck JS, Legro RS, Carmina E. American Association of Clinical Endocrinologists, American College of Endocrinology, and Androgen Excess and Pcos Society Disease State Clinical Review: Guide to the Best Practices in the Evaluation and Treatment of Polycystic Ovary Syndrome - Part 2. Endocrine practice : official journal of the American College of Endocrinology and the American Association of Clinical Endocrinologists. 2015;21(12):1415–1426.
12. Kumarendran B, O'Reilly MW, Manolopoulos KN, 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 medicine. 2018;15(3):e1002542.
13. Hossain N, Stepanova M, Afendy A, et al. Non-alcoholic steatohepatitis (NASH) in patients with polycystic ovarian syndrome (PCOS). Scand J Gastroenterol 2011;46(4):479–484. [PubMed: 21114431]
14. Sarkar M, Terrault N, Duwaerts CC, Tien P, Cedars MI, Huddleston H. The Association of Hispanic Ethnicity with Nonalcoholic Fatty Liver Disease in Polycystic Ovary Syndrome. Curr Opin Gynecol Obstet. 2018;1(1):24–33. [PubMed: 30112518]
15. Tan S, Bechmann LP, Benson S, et al. Apoptotic markers indicate nonalcoholic steatohepatitis in polycystic ovary syndrome. The Journal of clinical endocrinology and metabolism. 2010;95(1): 343–348. [PubMed: 19906783]

Table 1.

Cohort Characteristics in Women With and Without PCOS (n=102)

	PCOS (n=37)	Non-PCOS (n=65)	p-value
Age, years	33 (16)	36 (10)	0.02
Race (%): Non-Hispanic White Hispanic White Non-Hispanic Black Hispanic Black Asian Other-Hispanic Other Non-Hispanic	18.9 8.1 5.4 0.0 29.7 18.9 16.2	13.9 12.3 4.6 1.5 36.9 18.5 7.7	0.83
Body mass index (kg/m^2)	38.2 (12.0)	33.1 (8.8)	<0.01
Triglycerides (mg/dL)	198 (137)	185 (83)	0.32
LDL (mg/dL)	122 (47)	102 (42)	0.05
HDL (mg/dL)	45 (18)	43 (10)	0.64
Diabetes (%)	40.5	30.8	0.32
ALT (IU/ml)	71 (93)	78 (70)	0.91

Median (IQR) unless otherwise specified

Table 2.

NAFLD Histology in Women With and Without PCOS (n=102)

	PCOS (n=37)	Non-PCOS (n=65)	p-value
NASH (%) NAFLD without NASH (%)	75.7 24.3	66.2 33.8	0.32
NAFLD Activity Score	4 (3)	4 (2)	0.58
Severe steatosis (grade 3) (%):	38.9	44.6	0.59
Ballooned hepatocytes (%): Any Severe (grade 2)	48.6 32.4	46.2 12.5	0.75 0.02
Lobular inflammation (%): Any Severe (grade 2–3)	82.9 29.7	77.8 26.2	0.82 0.92
Portal inflammation (%): Any Severe (grade 2)	45.7 10.8	44.4 6.2	0.99 0.70
Fibrosis (%): Any Advanced (stages 3–4)	83.8 16.2	66.2 6.1	0.06 0.10

Median (IQR) unless otherwise specified. “Severe” reflects highest grades for each respective histological category.

Table 3.

Association of PCOS with Severe Hepatocyte Ballooning (n=102)

	Unadjusted Analysis		Adjusted Analysis	
	OR (95% CI)	**p value**	**OR (95% CI)**	**p value**
PCOS	**3.36 (1.22–9.23)**	**0.019**	**3.43 (1.12–10.6)**	**0.032**
Age	1.03 (0.97–1.10)	0.379	1.06 (0.99–1.14)	0.104
BMI	1.07 (1.01–1.14)	0.015	1.06 (1.00–1.13)	0.068

Table 4.

Association of PCOS with Advanced NASH Fibrosis (n=102)

	Unadjusted Analysis		Adjusted Analysis*	
	OR (95% CI)	**p value**	**OR (95% CI)**	**p value**
PCOS	**2.95 (0.78–11.2)**	**0.113**	**7.10 (1.29–39.1)**	**0.023**
Age	1.17 (1.02–1.33)	0.022	1.19 (1.03–1.37)	0.018
BMI	1.04 (0.96–1.12)	0.353	1.00 (0.92–1.10)	0.927

*Adjusted for age and BMI