

Normo- and hyperandrogenic women with polycystic ovary syndrome exhibit an adverse metabolic profile through life

Pekka Pinola, Ph.D.,^{a,b} Katri Puukka, Ph.D.,^{b,c} Terhi T. Piltonen, Ph.D.,^{a,b} Johanna Puurunen, Ph.D.,^{a,b} Eszter Vankó, Ph.D.,^{d,e} Inger Sundström-Poromaa, Ph.D.,^f Elisabet Stener-Victorin, Ph.D.,^g Angelica Lindén Hirschberg, Ph.D.,^h Pernille Ravn, Ph.D.,ⁱ Marianne Skovsager Andersen, Ph.D.,^j Dorte Glintborg, Ph.D.,^j Jan Roar Mellembakken, Ph.D.,^k Aimo Ruokonen, Ph.D.,^{b,c} Juha S. Tapanainen, Ph.D.,^{b,l} and Laure C. Morin-Papunen, Ph.D.^{a,b}

^a Department of Obstetrics and Gynecology, Oulu University Hospital, Oulu, Finland; ^b University of Oulu and Medical Research Center Oulu, Oulu, Finland; ^c Nordlab Oulu, Oulu University Hospital and Department of Clinical Chemistry, University of Oulu, Oulu, Finland; ^d Institute of Laboratory Medicine, Children's and Women's Health, Norwegian University of Science and Technology, Trondheim, Norway; ^e Department of Obstetrics and Gynecology, St. Olav's University Hospital Trondheim, Trondheim, Norway; ^f Department of Women's and Children's Health, Uppsala University, Uppsala, Sweden; ^g Department of Physiology and Pharmacology, Karolinska Institutet, Stockholm, Sweden; ^h Department of Obstetrics and Gynecology, Karolinska University Hospital, Stockholm, Sweden; ⁱ Department of Gynecology and Obstetrics, and ^j Department of Endocrinology and Metabolism, Odense University Hospital, Odense, Denmark; ^k Section of Reproductive Medicine, Department of Gynecology, Women's Division, Oslo University Hospital, Oslo, Norway; and ^l Department of Obstetrics and Gynecology, University of Helsinki and Helsinki University Hospital, Helsinki, Finland

Objective: To compare the metabolic profiles of normo- and hyperandrogenic women with polycystic ovary syndrome (PCOS) with those of control women at different ages during reproductive life.

Design: Case-control study.

Setting: Not applicable.

Patient(s): In all, 1,550 women with normoandrogenic (n = 686) or hyperandrogenic (n = 842) PCOS and 447 control women were divided into three age groups: <30, 30–39, and >39 years.

Interventions(s): None.

Main Outcome Measure(s): Body mass index (BMI), waist circumference, blood pressure, glucose, insulin, cholesterol, lipoproteins, triglycerides and high-sensitivity C-reactive protein.

Result(s): Both normo- and hyperandrogenic women with PCOS were more obese, especially abdominally. They had increased serum levels of insulin (fasting and in oral glucose tolerance tests), triglycerides, low-density lipoprotein, and total cholesterol, higher blood pressure, and lower high-density lipoprotein levels independently from BMI compared with the control population as early as from young adulthood until menopause. The prevalence of metabolic syndrome was two- to fivefold higher in women with PCOS compared with control women, depending on age and phenotype, and the highest prevalence was observed in hyperandrogenic women with PCOS at late reproductive age.

Conclusion(s): When evaluating metabolic risks in women with PCOS, androgenic status, especially abdominal obesity and age, should be taken into account, which would allow tailored management of the syndrome from early adulthood on. (Fertil Steril® 2017;107:788–95. ©2016 by American Society for Reproductive Medicine.)

Key Words: PCOS, hyperandrogenism, metabolism, obesity

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Reprint requests: Laure C. Morin-Papunen, M.D., Ph.D., Adjunct Professor, Department of Obstetrics and Gynecology, Oulu University Hospital, PL 23, OYS 90029, Finland (E-mail: Imp@cc.oulu.fi).

Polycystic ovary syndrome (PCOS) is the most common endocrine disorder affecting reproductive-age women (incidence 6%–18% depending on the population and the diagnostic criteria used) (1, 2). It is known to have multifaceted unfavorable effects on women's health. In addition to anovulatory infertility, which is typically associated with the syndrome, women with PCOS exhibit numerous metabolic risk factors, such as abdominal obesity, insulin resistance, impaired glucose tolerance, dyslipidemia, and chronic inflammation (3–6). Hyperandrogenism (HA) plays a central role in the syndrome and has been associated with a more severe metabolic profile in some, but not all, studies (7–11). Obesity is common in PCOS and exacerbates the symptoms and promotes negative health consequences by increasing the risks of type 2 diabetes (T2DM) and cardiovascular disease (CVD). However, regarding the interaction between weight, weight gain, and hyperandrogenism, the results of earlier studies have been inconsistent. Some data suggest that the metabolic risks linked to PCOS are mainly related to obesity and insulin resistance, whereas others support the hypothesis that the syndrome per se, especially HA, independently from BMI, are the most important cardiovascular risks (12–14). According to the results of some studies, the unfavorable metabolic profile linked to PCOS seems to worsen after the menopausal transition, highlighting the lifelong health burden of the syndrome (3, 15). A recent study, however, could not confirm any association between a history of androgen excess and menstrual irregularity with worsening of metabolic health after menopause (16).

In women without PCOS, an impaired metabolic profile is associated with obesity and weight gain. Especially during the menopausal transition, weight gain has been shown to have an adverse effect on glucose metabolism (17). Furthermore, circulating levels of ovarian and adrenal androgens do not seem to have a significant impact on chronic inflammation markers or lipid profile in women in the general population, whereas a low serum level of SHBG is an independent predictor of an increased risk of CVD (18, 19).

Given the lack of data regarding age-related metabolic changes in women with PCOS, the aim of the present study was to explore and compare metabolic profiles in women with PCOS and in control women throughout their reproductive life spans in a cross-sectional dataset. We also aimed to identify specific metabolic risk factors and the impact of hyperandrogenism in PCOS during early adulthood.

MATERIALS AND METHODS

Study Population

Women taking part in eight Nordic PCOS studies contributed to the present investigation. Two centers in Finland, three in Sweden, two in Norway, and one in Denmark were involved, and there were totals of 1,550 women with PCOS (age range 14–59 years) and 447 control women without the syndrome (age range 18–62 years) (6,20–27). The women with PCOS were recruited both from hospital gynecology or infertility clinics and from the community with advertisements in local newspapers and the healthy control women from the

community by advertisements in local newspapers. Circulating levels of androgens in a subpopulation of this Nordic PCOS cohort population have been recently published (28).

Diagnoses of PCOS were made according to the Rotterdam criteria (29). Diagnosis of PCOS in peri- and postmenopausal women was based on the presence of oligo-amenorrhea combined with hyperandrogenism (either biochemical or clinical) reported at reproductive age. Thus, retrospectively, all of the women met the Rotterdam criteria. Ovarian morphology was examined by means of transvaginal ultrasonography in all participants. Biochemical hyperandrogenism was defined in relation to the upper limits used in the respective laboratories, depending on the method used, and clinical hyperandrogenism was diagnosed when a subject had a Ferriman-Gallwey score of >7 . The Ferriman-Gallwey scoring was performed by a physician in all studies. The PCOS population was further divided into two groups: hyperandrogenic women with PCOS (HA-PCOS, including biochemical and/or clinical hyperandrogenism) and normoandrogenic women with PCOS (NA-PCOS, including women with both oligo-amenorrhea and polycystic ovaries observed in ultrasonography). The control population consisted of women with normal ovaries (according to ultrasonography) and absence of PCOS-related symptoms, e.g., oligo- or anovulation and/or hirsutism and/or elevated serum T levels. The PCOS and control groups were grouped according to age as follows: <30 years, 30–39 years, and >39 years. Women using hormonal preparations, medication affecting androgen levels and glucose metabolism, as well as statins or antihypertensive drugs were excluded from the study. Alternatively, a 2-month washout period for hormonal preparations or medication affecting androgen levels was required before entering the study. Furthermore, none of the women had T2DM, because having the diagnosis of preexisting diabetes was an exclusion criterion in all studies.

BMI was calculated as kg/m^2 . Waist circumference was assessed according to a generally accepted method. Both systolic and diastolic blood pressures (BPs) were measured in a sitting position after 15 minutes of resting.

A diagnosis of metabolic syndrome (MetS) was made according to the Rotterdam consensus (28), requiring three out of the following five criteria: waist circumference >88 cm and/or $\text{BMI} \geq 30 \text{ kg}/\text{m}^2$, triglycerides $\geq 150 \text{ mg}/\text{dL}$ (1.70 mmol/L), high-density lipoprotein (HDL) $\leq 50 \text{ mg}/\text{dL}$ (1.30 mmol/L), systolic BP $\geq 130 \text{ mm Hg}$ and/or diastolic BP $\geq 85 \text{ mm Hg}$, and fasting glucose $\geq 110\text{--}126 \text{ mg}/\text{dL}$ (6.11–6.99 nmol/L) and/or 2-hour oral glucose tolerance test (OGTT) glucose $\geq 140\text{--}199 \text{ mg}/\text{dL}$ (7.78–11.04 nmol/L).

Laboratory Methods

The metabolic variables (glucose, insulin, cholesterol, lipoproteins, triglycerides, and high-sensitivity C-reactive protein) were assayed by means of the routine methods used in the laboratories of the different study centers (6,19–26). According to the Nordic Reference Interval Project 2000 (30) the reference ranges for the serum lipid levels were similar in all subpopulations of the study.

In five study populations (20–25), assays of T were performed by means of liquid chromatography-mass

TABLE 1**Anthropometric and metabolic parameters in the polycystic ovary syndrome and control women.**

Metabolic parameter	Control		PCOS		<i>P</i> value
	n	Result	n	Result	
Age (y)	447	33.5 (9.9)	1,550	30.0 (7.2)	<.001
BMI (kg/m ²)	447	25.9 (5.4)	1,497	29.2 (6.9)	<.001
Waist (cm)	312	87.6 (14.6)	1,204	92.9 (17.5)	<.001
Testosterone (nmol/L)	433	1.1 (0.5)	1,359	1.8 (1.0)	<.001 ^a
Fasting glucose (mmol/L)	376	5.1 (0.9)	1,104	5.1 (0.6)	NS
Fasting insulin (mU/L)	372	7.4 (6.0)	1,093	12.3 (11.1)	<.001 ^a
Total cholesterol (mmol/L)	364	4.6 (0.9)	982	4.8 (1.0)	.003 ^a
HDL (mmol/L)	346	1.5 (0.3)	960	1.4 (0.4)	<.001 ^a
LDL (mmol/L)	347	2.6 (0.8)	863	2.9 (0.9)	<.001 ^a
Triglycerides (mmol/L)	366	0.9 (0.5)	974	1.3 (0.8)	<.001 ^a
OGTT glucose, 2 h (mmol/L)	140	5.0 (1.3)	681	5.8 (1.8)	<.001
OGTT mean glucose (mmol/L)	140	5.0 (0.8)	681	5.5 (1.1)	<.001
OGTT insulin, 2 h (mU/L)	152	27.4 (20.5)	860	71.7 (73.9)	<.001 ^a
OGTT mean insulin (mU/L)	152	17.2 (12.0)	840	42.6 (41.6)	<.001 ^a
Systolic BP (mm Hg)	318	118.2 (15.6)	1,276	123.43 (16.2)	<.001 ^a
Diastolic BP (mm Hg)	318	74.3 (12.1)	1,276	78.5 (12.1)	<.001 ^a
hs-CRP (mg/L)	159	1.5 (3.0)	761	2.8 (3.8)	<.001

Note: Data presented as n or mean (SD), unless stated otherwise. BMI = body mass index; BP = blood pressure; HDL = high-density lipoprotein; hs-CRP = high-sensitivity C-reactive protein; LDL = low-density lipoprotein; OGTT = oral glucose tolerance test.

^a Statistical significance (*P*<.05) remains after adjustment for age and BMI.

Pinola. Metabolic profile in women with PCOS. *Fertil Steril* 2016.

spectrometry (LC/MS) and SHBG by means of chemiluminometric immunoassay at NordLab Oulu, as reported earlier (28). In the remaining three study sites (not included in our previous publication), T was analyzed with the use of radioimmunoassay (26, 27). To define biochemical hyperandrogenism age, method- and laboratory-specific reference ranges were used in the different subgroups. All of the analyses were also performed in a subpopulation including only the women with T measured with the use of LC/MS.

The mean OGTT plasma glucose level and mean OGTT serum insulin levels were calculated as the means of concentrations at different time points ([basal + 2-hour]/2).

Conversion factors ([SI unit]/[conversion factor] = metric unit) to metric units for the laboratory parameters were as follows: glucose 0.056 (mg/dL), triglycerides 0.011 (mg/dL), and total cholesterol, HDL, and low-density lipoprotein (LDL) 0.026 (mg/dL).

Statistical Methods

Comparisons of continuous variables were carried out with the use of Student *t* test or the (nonparametric) Mann-Whitney *U* test, depending on the distribution of the variable. One-way analysis of variance was used in multiple group analyses with Scheffé and Tukey post hoc analyses. Adjustments for BMI and age were applied with the use of linear regression analysis. For categoric variables, the χ^2 -test was used. Statistical analyses were performed with the use of SPSS 21.0 and 22.0 software. Of note, the number of participants varied between analyses owing to the lack of measurements in some cases.

Informed consents were obtained from all women at the original study sites, and the study was approved by the Ethics Committees of the respective study sites.

RESULTS

The women with PCOS (n = 1,550) had higher BMIs and waist circumferences than the control population (n = 447). They also presented with significantly lower levels of HDL and higher levels of insulin (fasting and during OGTTs), total cholesterol, LDL, triglycerides, and BP (both systolic and diastolic) compared with the control women after adjusting for BMI (Table 1).

The anthropometric and metabolic parameters in the HA-PCOS group (n = 842), the NA-PCOS group (n = 684), and the control women (n = 447) are presented in Table 2 and Figures 1 and 2. Only the results that remained significant after adjustment for BMI and age are presented in the text.

Comparisons Between the HA-PCOS, NA-PCOS, and Control Groups

BMI and waist circumference. BMIs and waist circumferences were significantly greater in both the HA- and the NA-PCOS subpopulations compared with the control group. In addition, BMI was greater in the HA-PCOS women compared with the NA-PCOS women (Table 2).

In age-group analyses, women in the HA-PCOS and NA-PCOS groups had greater BMIs and waist circumferences in all age groups compared with the control group. In the age groups of 30–39 years and >39 years, the women in the HA-PCOS group had higher BMIs and waist circumferences than the women in the NA-PCOS group (Fig. 1).

Glucose tolerance and insulin resistance. Women in the HA-PCOS and NA-PCOS groups were more hyperinsulinemic (higher fasting and OGTT insulin levels) than the control women, and women in the NA-PCOS group were less glucose

TABLE 2

Anthropometric and metabolic parameters in the study populations.

Metabolic parameter	Control		NA-PCOS		HA-PCOS		<i>P</i> value ^a	<i>P</i> value ^b	<i>P</i> value ^c
	n	Result	n	Result	n	Result			
Age (y)	447	33.5 (9.9)	684	29.9 (7.0)	842	30.0 (7.4)	<.001	<.001	NS
BMI (kg/m ²)	447	25.9 (5.4)	666	28.8 (7.0)	811	29.4 (6.7)	<.001	<.001	.027
Waist (cm)	312	87.6 (14.6)	590	92.1 (17.8)	604	93.6 (17.0)	<.001	<.001	NS ^d
Testosterone (nmol/L)	433	1.1 (0.5)	617	1.4 (0.5)	742	2.1 (1.1)	<.001 ^d	<.001 ^d	<.001 ^d
Fasting glucose (mmol/L)	376	5.1 (0.9)	542	5.1 (0.6)	552	5.1 (0.6)	NS	NS	NS
Fasting insulin (mU/L)	372	7.4 (6.0)	544	12.0 (11.4)	537	12.4 (10.7)	<.001 ^d	<.001 ^d	NS
Total cholesterol (mmol/L)	364	4.6 (0.9)	368	4.7 (0.9)	603	4.8 (1.0)	.041	.004 ^d	NS
HDL (mmol/L)	346	1.5 (0.3)	364	1.3 (0.5)	586	1.4 (0.4)	<.001 ^d	<.001 ^d	.013
LDL (mmol/L)	347	2.6 (0.8)	349	2.8 (0.8)	504	2.9 (0.9)	<.001 ^d	<.001 ^d	NS
Triglycerides (mmol/L)	366	0.9 (0.5)	367	1.2 (0.7)	596	1.2 (0.8)	<.001 ^d	<.001 ^d	NS
OGTT glucose, 2 h (mmol/L)	140	5.0 (1.3)	442	5.9 (1.7)	238	6.0 (1.9)	<.001 ^d	<.001	NS
OGTT mean glucose (mmol/L)	140	5.0 (0.8)	442	5.4 (1.0)	238	5.5 (1.1)	<.001	<.001	NS
OGTT insulin, 2 h (mU/L)	152	27.4 (20.5)	476	67.8 (69.4)	376	76.0 (79.0)	<.001 ^d	<.001 ^d	NS
OGTT mean insulin (mU/L)	152	17.2 (12.0)	465	40.9 (40.1)	367	44.5 (43.3)	<.001 ^d	<.001 ^d	NS
Systolic BP (mm Hg)	318	118 (16)	605	123 (16)	657	124 (17)	<.001 ^d	<.001 ^d	NS
Diastolic BP (mm Hg)	318	74 (12)	605	78 (12)	657	79 (12)	<.001 ^d	<.001 ^d	NS
hs-CRP (mg/L)	159	1.5 (3.0)	468	2.8 (3.6)	291	2.9 (4.0)	<.001	<.001	NS

Note: Data presented as n or mean (SD), unless stated otherwise. HA = hyperandrogenic; NA = normoandrogenic; NS = not significant; other abbreviations as in Table 1.

^a *P* value between reference group and NA-PCOS.^b *P* value between reference group and HA-PCOS.^c *P* value between NA- and HA-PCOS groups.^d Statistically significant (*P*<.05) after adjustment for age and BMI.Pinola. Metabolic profile in women with PCOS. *Fertil Steril* 2016.

tolerant (higher 2-hour OGTT glucose levels) than the control women (Table 2).

In the age-group analyses, 2-hour glucose and mean glucose levels in OGTTs were higher in the HA-PCOS and NA-PCOS groups than in the control women in the age group of 30–39 years (*P*=.001 and *P*=.024, respectively). Two-hour insulin and mean OGTT insulin levels were significantly higher in the women of age \geq 30 years in both PCOS groups compared with the control group (*P*=.002 and *P*=.008, respectively). Fasting insulin levels were higher in women of ages <30 and 30–39 years in the NA-PCOS group (*P*=.041 and *P*=.021, respectively) and in women of age >39 years in the HA-PCOS group (*P*=.003) compared with the level in the control women (Fig. 1).

Lipids. Serum total cholesterol levels were higher in the HA-PCOS group than in the control group. Serum levels of HDL were lower and those of triglycerides and LDL higher in both PCOS populations compared with the control women (Table 2).

In the age group analyses, in women <30 years of age in both the NA- and HA-PCOS groups had worse lipid profiles compared with the control population. Women >39 years of age in the HA-PCOS group presented with higher levels of LDL and triglycerides compared with controls and higher levels of LDL compared with the NA-PCOS population (Fig. 2).

High-Sensitivity C-Reactive Protein

There were no significant differences between study groups in serum levels of high-sensitivity C-reactive protein in any of the age groups after adjustment for BMI.

Blood Pressure

Both systolic and diastolic BPs were significantly increased in both PCOS populations compared with control women (Table 2).

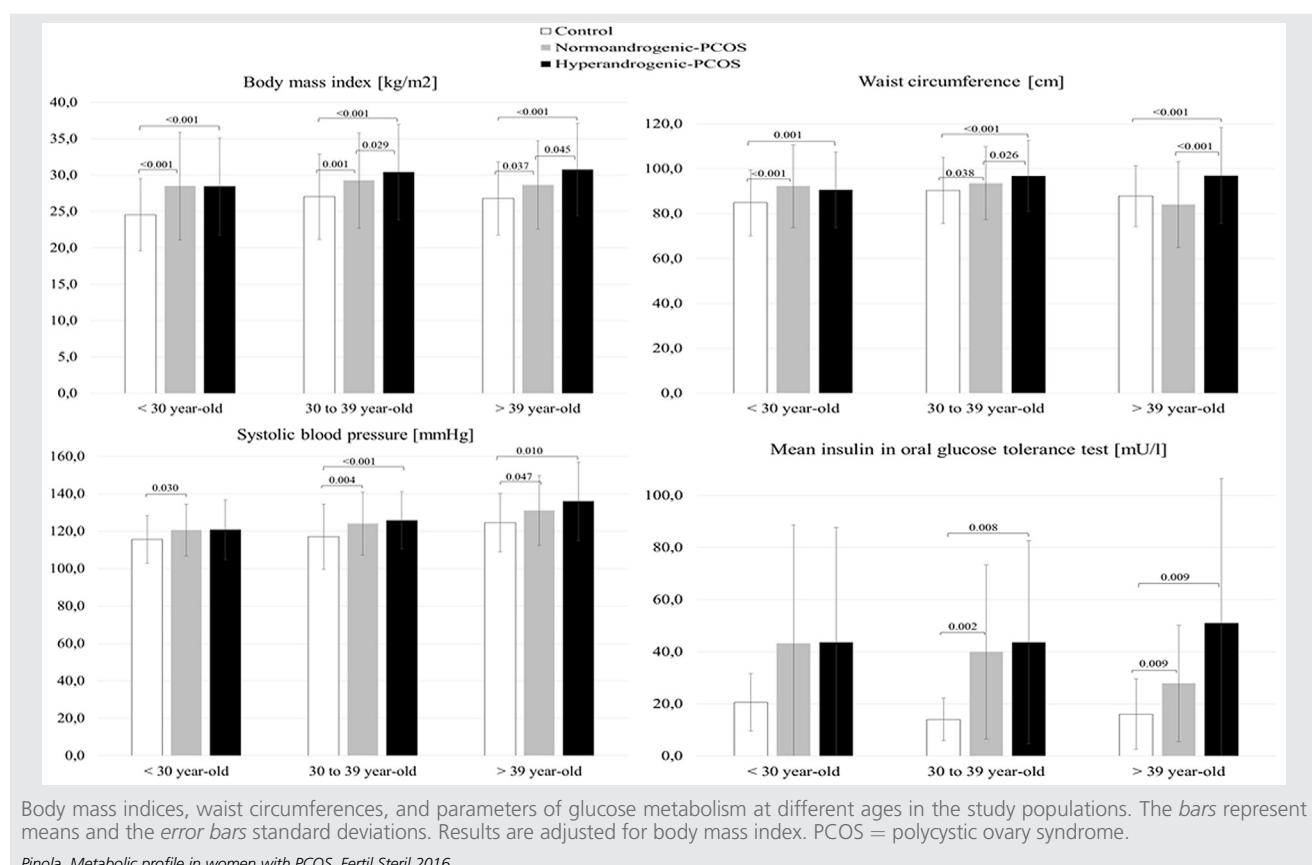
Age group analyses revealed that systolic BP was significantly higher in all age groups of the NA-PCOS group and in women \geq 30 years of age in the HA-PCOS group compared with the control group (Fig. 2). Diastolic BP was higher in all age groups of the HA-PCOS group and in women <40 years of age in the NA-PCOS group compared with control women (data not presented).

Prevalence of Metabolic Syndrome

In women <30 years of age the prevalence of MetS was fivefold higher and in women <40 years of age twofold higher in both PCOS populations compared with the control women. However, in women >39 years of age, the difference between the NA-PCOS and control groups had disappeared, whereas the prevalence of MetS remained twofold higher in the HA-PCOS group compared with the NA-PCOS or control groups (Supplemental Table 1, available online at www.fertstert.org).

In age-group analyses, isolated parameters of the metabolic risk profile did not considerably differ between PCOS subpopulations, except for alterations of glucose metabolism and HDL in the age group of 30–39 years, and higher BMIs and waist circumferences in the age group of >39 years in the HA-PCOS compared with the NA-PCOS group. These results were identical when using International Diabetes Federation criteria for MetS.

FIGURE 1



Analyses in the Subpopulation of Women With Testosterone Measured by Means of LC/MS

All the results were similar except for the differences in serum lipid concentrations between the groups, which became nonsignificant, most probably because of the decreased power for the analysis in this subpopulation (data not presented).

DISCUSSION

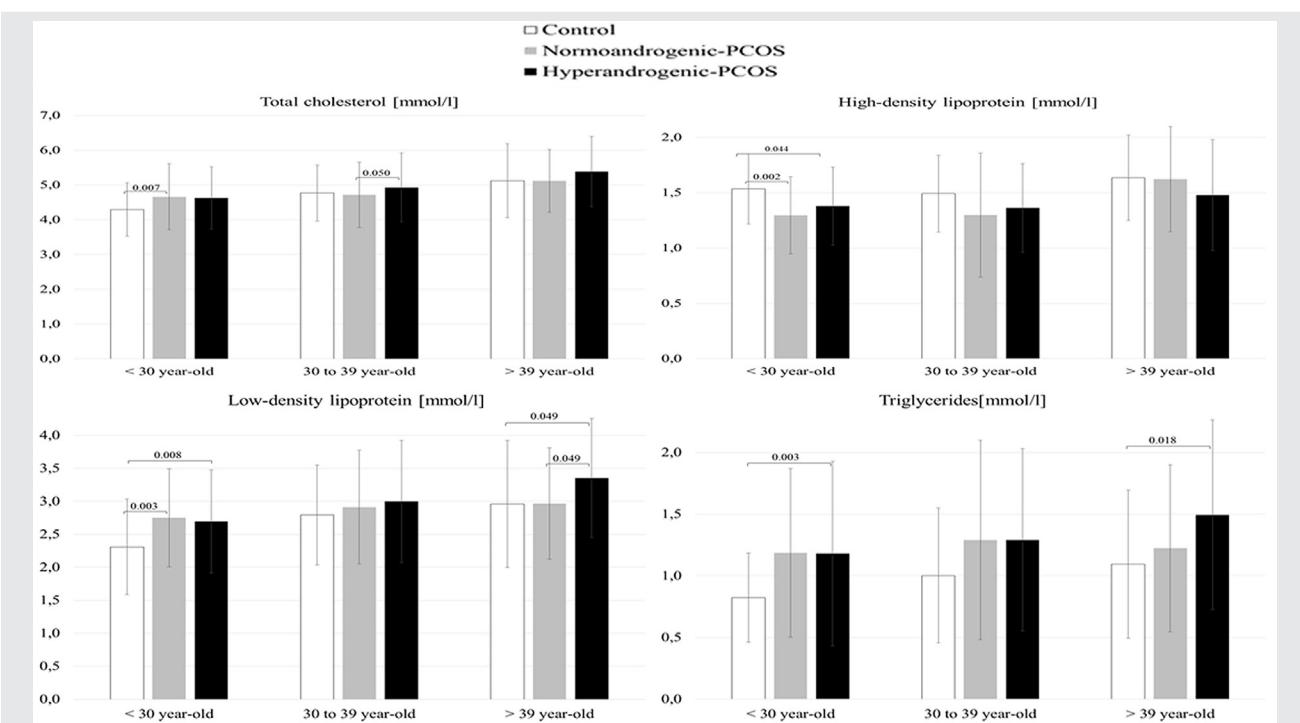
In this large Nordic multicenter collaboration study, we demonstrated that women with PCOS already had an unfavorable metabolic profile in early adulthood, lasting until menopause. Furthermore, we found no significant differences in individual metabolic parameters between the NA-PCOS and HA-PCOS groups after adjustment for BMI, strongly suggesting a predominant role of obesity, specifically abdominal obesity, in the severity of the syndrome. Nevertheless, the prevalence of MetS was twofold higher in the HA-PCOS population compared with the NA-PCOS and control groups during late reproductive years, suggesting that hyperandrogenism also is implicated, at least in the long term, in the adverse metabolic profile seen in PCOS.

The present data indicate that both NA- and HA-PCOS group women were more obese (especially abdominally), hyperinsulinemic, and dyslipidemic and exhibited higher BP

levels throughout their life spans compared with the control population. More specifically, even after adjusting for BMI, the women in the NA-PCOS group were more glucose intolerant, insulin resistant, hyperinsulinemic, and dyslipidemic, especially in early adulthood, compared with the control women, which is in line with earlier data indicating that the differences between normoandrogenic women with PCOS and healthy women, as well as the adverse metabolic outcomes in PCOS, are mainly related to abdominal obesity (31, 32). In some studies, however, oligo-amenorrheic normoandrogenic women with PCOS exhibited a more favorable metabolic phenotype compared with their hyperandrogenic counterparts (7), and other studies have highlighted the role of hyperandrogenism as a cardiovascular risk factor in women with PCOS (5, 33).

Of note, both groups of women with PCOS (HA- and NA-PCOS) exhibited already in early adulthood an adverse lipid profile which, together with BMI, waist circumference, and BP, seemed to worsen with age in both PCOS groups and in control women, supporting the results of earlier studies (5, 26). As for glucose metabolism, only women with PCOS aged 30–39 years exhibited higher glucose and insulin levels in OGTTs compared with control women. The results of follow-up studies up to the postmenopausal period (15, 34) are in line with our data, showing adverse changes in metabolic profile and BP at early ages in women with PCOS

FIGURE 2



Lipids at different ages in the study populations. The bars represent the means and the error bars the standard deviations. Results are adjusted for body mass index. PCOS = polycystic ovary syndrome.

Pinola. Metabolic profile in women with PCOS. *Fertil Steril* 2016.

independently from obesity. In particular, a high level of triglycerides has been shown to be an independent predictor of future risk of myocardial infarction, and low levels of HDL reflect cardiovascular morbidity (35) and are associated with elevated cardiovascular risks in general, especially in women (36). Moreover, the difference in BP (4–5 mm Hg between the PCOS and control groups), though relatively modest, has clinical significance, because it was already present in women <30 years of age and continuing thereafter. It has been estimated that an increase of BP of only 1.5–2 mm Hg could have a large impact on CVD risk at population level, and reduction of mildly and/or moderately elevated BP to normal levels has been associated with a reduced risk of CVD in a large placebo-controlled study (37). The present observations fit with recent epidemiologic data indicating that women with PCOS have increased risk factors of ischemic heart and cerebrovascular diseases, independently from obesity (38). However, in women without PCOS, an impaired metabolic profile has been associated with obesity and weight gain, especially during the period of menopausal transition (17), and this also seems to be the case in PCOS. All in all, the results highlight the importance of screening for PCOS and overweight/obesity in early adulthood to tailor treatment and intervention protocols and reduce the risks of future CVD and T2DM.

The prevalence of MetS was significantly higher in women with PCOS (31.5%) throughout their reproductive

life spans compared with control women, who exhibited a MetS prevalence of 12%, as also observed in general Nordic populations (39–41). The prevalence was not increased in women with NA-PCOS compared with control women in the age group >39 years, which may be a result of the relatively low number of women and similar waist circumferences in this age group. Again, this finding supports the postulation that abdominal obesity is the principal determinant of metabolic abnormalities in PCOS (32), but it is also consistent with earlier observations linking hyperandrogenism to metabolic disturbances and a higher prevalence of MetS (11). Even though isolated parameters of the metabolic risk profile did not considerably differ between the NA- and HA-PCOS populations, lifelong exposure to hyperandrogenism may end up being an additional risk factor of MetS later in life. In keeping with this, in our study population, serum levels of LDL were slightly higher in women in the HA-PCOS group compared with the NA-PCOS group in women aged ≥40 years. Hyperandrogenism may directly or indirectly influence metabolic abnormalities and contribute to abdominal obesity. However, controversial data has been published recently indicating that a history of hyperandrogenism is not always associated with metabolic disturbances after menopause (16). More research is needed to provide a better understanding of the interaction between hyperandrogenism, insulin resistance, and abdominal adiposity in PCOS (32).

The strengths of the present study are the inclusion of a large number of cases of HA- and NA-PCOS from a young fertile age to menopause. Moreover, the study population was remarkably homogeneous regarding ethnicity, with all the women being “white.” This is of great importance, because the phenotypic and metabolic profiles of women with PCOS show a high degree of ethnic variation (8, 38). The PCOS cohorts were recruited from eight different study sites, but in all cases the diagnoses of PCOS were made with the use of the Rotterdam criteria. The present results underscore the role of abdominal obesity and, to a lesser degree, hyperandrogenism in the development of lifelong metabolic risks in Nordic women with PCOS, and most probably women in other populations as well. One limitation is the cross-sectional study design, which can cause bias due to heterogeneity in the characteristics of the participants in the different age groups. Furthermore, the control group was smaller than the PCOS group, because not all of the trials contributing to this collaborative study included a control population. The women with T2DM and those using statins or antihypertensive drugs had been excluded from the study populations, which may have decreased the differences between control and PCOS women. The methods for laboratory analyses varied between subpopulations in different study sites, but all were carried out according to accredited methods, and specific reference ranges of the laboratory were used in the definition of biochemical hyperandrogenism. It has to be noted that 52% of the T and 50% of the SHBG measurements were performed at only one study site (28). The upper limit for T in defining biochemical hyperandrogenism did not take into account the physiologic decline in serum levels observed with age. Therefore, some hyperandrogenic women >39 years of age may have been classified as normoandrogenic, which could narrow the differences between the HA- and NA-PCOS groups. Finally, we could not separate the participants into pre- and postmenopausal groups to study more specifically the effect of menopausal transition on metabolic indices, because there were too few women older than 50 years to obtain reliable results in age group comparisons.

CONCLUSION

This Nordic multicenter study showed that women with PCOS presented a worsened metabolic profile compared with a control population from early adulthood to menopause. Even though lifelong exposure to hyperandrogenemia seemed to contribute, at least in part, to a higher prevalence of MetS, metabolic disturbances were present in both the HA- and the NA-PCOS groups, and abdominal obesity appeared to be the principal determinant of metabolic abnormalities in PCOS.

We conclude that when evaluating metabolic risks in women with PCOS, androgenic status, obesity (especially abdominal obesity), and age should all be taken into account to allow tailored management of the syndrome, focusing on prevention of abdominal obesity, starting from early adulthood. However, only a long-term, longitudinal follow-up study will reveal whether the metabolic risk factors linked

to PCOS, and more specifically to HA, translate into later cardiovascular morbidity and mortality.

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REFERENCES

1. Franks S. Polycystic ovary syndrome. *N Engl J Med* 1995;333:853–61.
2. March WA, Moore VM, Willson KJ, Phillips DI, Norman RJ, Davies MJ. The prevalence of polycystic ovary syndrome in a community sample assessed under contrasting diagnostic criteria. *Hum Reprod* 2010;25:544–51.
3. Puurunen J, Piltonen T, Morin-Papunen L, Perheentupa A, Jarvela I, Ruokonen A, et al. Unfavorable hormonal, metabolic, and inflammatory alterations persist after menopause in women with PCOS. *J Clin Endocrinol Metab* 2011;96:1827–34.
4. Daan NM, Louwers YV, Koster MP, Eijkemans MJ, de Rijke YB, Lentjes EW, et al. Cardiovascular and metabolic profiles amongst different polycystic ovary syndrome phenotypes: who is really at risk? *Fertil Steril* 2014;102:1444–51.e3.
5. Johnstone EB, Davis G, Zane LT, Cedars MI, Huddleston HG. Age-related differences in the reproductive and metabolic implications of polycystic ovarian syndrome: findings in an obese, United States population. *Gynecol Endocrinol* 2012;28:819–22.
6. Morin-Papunen LC, Vauhkonen I, Koivunen RM, Ruokonen A, Tapanainen JS. Insulin sensitivity, insulin secretion, and metabolic and hormonal parameters in healthy women and women with polycystic ovarian syndrome. *Hum Reprod* 2000;15:1266–74.
7. Barber TM, Wass JA, McCarthy MI, Franks S. Metabolic characteristics of women with polycystic ovaries and oligo-amenorrhoea but normal androgen levels: implications for the management of polycystic ovary syndrome. *Clin Endocrinol (Oxf)* 2007;66:513–7.
8. Welt CK, Gudmundsson JA, Arason G, Adams J, Palsdottir H, Gudlaugsdottir G, et al. Characterizing discrete subsets of polycystic ovary syndrome as defined by the Rotterdam criteria: the impact of weight on phenotype and metabolic features. *J Clin Endocrinol Metab* 2006;91:4842–8.
9. Barber TM, Vojtechova P, Franks S. The impact of hyperandrogenism in female obesity and cardiometabolic diseases associated with polycystic ovary syndrome. *Horm Mol Biol Clin Investig* 2013;15:91–103.
10. O'Reilly MW, Taylor AE, Crabtree NJ, Hughes BA, Capper F, Crowley RK, et al. Hyperandrogenemia predicts metabolic phenotype in polycystic ovary syndrome: the utility of serum androstenedione. *J Clin Endocrinol Metab* 2014;99:1027–36.
11. Sung YA, Oh JY, Chung H, Lee H. Hyperandrogenemia is implicated in both the metabolic and reproductive morbidities of polycystic ovary syndrome. *Fertil Steril* 2014;101:840–5.
12. Park HT, Cho GJ, Ahn KH, Shin JH, Kim YT, Hur JY, et al. Association of insulin resistance with anti-müllerian hormone levels in women without polycystic ovary syndrome (PCOS). *Clin Endocrinol (Oxf)* 2010;72:26–31.
13. Piouka A, Farmakiotis D, Katsikis I, Macut D, Gerou S, Panidis D. Anti-Müllerian hormone levels reflect severity of PCOS but are negatively influenced by obesity: relationship with increased luteinizing hormone levels. *Am J Physiol Endocrinol Metab* 2009;296:E238–43.
14. Nisenblat V, Norman RJ. Androgens and polycystic ovary syndrome. *Curr Opin Endocrinol Diabetes Obes* 2009;16:224–31.
15. Schmidt J, Brannstrom M, Landin-Wilhelmsen K, Dahlgren E. Reproductive hormone levels and anthropometry in postmenopausal women with polycystic ovary syndrome (PCOS): a 21-year follow-up study of women diagnosed with PCOS around 50 years ago and their age-matched controls. *J Clin Endocrinol Metab* 2011;96:2178–85.
16. Polotsky AJ, Allshouse AA, Crawford SL, Harlow SD, Khalil N, Kazlauskaitė R, et al. Hyperandrogenic oligomenorrhea and metabolic risks across menopausal transition. *J Clin Endocrinol Metab* 2014;99:2120–7.
17. Polotsky HN, Polotsky AJ. Metabolic implications of menopause. *Semin Reprod Med* 2010;28:426–34.

18. Bell RJ, Davison SL, Papalia MA, McKenzie DP, Davis SR. Endogenous androgen levels and cardiovascular risk profile in women across the adult life span. *Menopause* 2007;14:630–8.

19. Wang Q, Kangas AJ, Soininen P, Taininen M, Tynkkynen T, Puukka K, et al. Sex hormone-binding globulin associations with circulating lipids and metabolites and the risk for type 2 diabetes: observational and causal effect estimates. *Int J Epidemiol* 2015;44:623–37.

20. Vanký E, Kjotrod S, Salvesen KA, Romundstad P, Moen MH, Carlsen SM. Clinical, biochemical and ultrasonographic characteristics of Scandinavian women with PCOS. *Acta Obstet Gynecol Scand* 2004;83:482–6.

21. Puurunen J, Piltonen T, Jaakkola P, Ruokonen A, Morin-Papunen L, Tapanainen JS. Adrenal androgen production capacity remains high up to menopause in women with polycystic ovary syndrome. *J Clin Endocrinol Metab* 2009;94:1973–8.

22. Stener-Victorin E, Holm G, Labrie F, Nilsson L, Janson PO, Ohlsson C. Are there any sensitive and specific sex steroid markers for polycystic ovary syndrome? *J Clin Endocrinol Metab* 2010;95:810–9.

23. Hudecova M, Holte J, Olovsson M, Sundstrom Poromaa I. Long-term follow-up of patients with polycystic ovary syndrome: reproductive outcome and ovarian reserve. *Hum Reprod* 2009;24:1176–83.

24. Piltonen T, Koivunen R, Perheentupa A, Morin-Papunen L, Ruokonen A, Tapanainen JS. Ovarian age-related responsiveness to human chorionic gonadotropin in women with polycystic ovary syndrome. *J Clin Endocrinol Metab* 2004;89:3769–75.

25. Piltonen T, Puurunen J, Hedberg P, Ruokonen A, Mutt SJ, Herzig KH, et al. Oral, transdermal and vaginal combined contraceptives induce an increase in markers of chronic inflammation and impair insulin sensitivity in young healthy normal-weight women: a randomized study. *Hum Reprod* 2012;27:3046–56.

26. Glintborg D, Mumm H, Ravn P, Andersen M. Age associated differences in prevalence of individual Rotterdam criteria and metabolic risk factors during reproductive age in 446 caucasian women with polycystic ovary syndrome. *Horm Metab Res* 2012;44:694–8.

27. Nybacka A, Carlstrom K, Stahle A, Nyren S, Hellstrom PM, Hirschberg AL. Randomized comparison of the influence of dietary management and/or physical exercise on ovarian function and metabolic parameters in overweight women with polycystic ovary syndrome. *Fertil Steril* 2011;96:1508–13.

28. Pinola P, Piltonen TT, Puurunen J, Vanký E, Sundstrom-Poromaa I, Stener-Victorin E, et al. Androgen profile through life in women with polycystic ovary syndrome: a Nordic multicenter collaboration study. *J Clin Endocrinol Metab* 2015;100:3400–7.

29. Rotterdam ESHRE/ASRM–Sponsored PCOS Consensus Workshop Group. Revised 2003 consensus on diagnostic criteria and long-term health risks related to polycystic ovary syndrome (PCOS). *Hum Reprod* 2004;19:41–7.

30. Rustad P, Simonsson P, Felding P, Pedersen M. Nordic Reference Interval Project Bio-bank and Database (NOBIDA): a source for future estimation and retrospective evaluation of reference intervals. *Scand J Clin Lab Invest* 2004;64:431–8.

31. Tzeng CR, Chang YC, Chang YC, Wang CW, Chen CH, Hsu MI. Cluster analysis of cardiovascular and metabolic risk factors in women of reproductive age. *Fertil Steril* 2014;101:1404–10.

32. Moran L, Teede H. Metabolic features of the reproductive phenotypes of polycystic ovary syndrome. *Hum Reprod Update* 2009;15:477–88.

33. Macut D, Micic D, Parapid B, Cvijovic G, Sumarac M, Kendereski A, et al. Age and body mass related changes of cardiovascular risk factors in women with polycystic ovary syndrome. *Vojnosanit Pregl* 2002;59:593–9.

34. Pasquali R, Gambineri A, Anconetani B, Vicennati V, Colitta D, Caramelli E, et al. The natural history of the metabolic syndrome in young women with the polycystic ovary syndrome and the effect of long-term oestrogen-progestagen treatment. *Clin Endocrinol (Oxf)* 1999;50:517–27.

35. Stampfer MJ, Krauss RM, Ma J, Blanche PJ, Holl LG, Sacks FM, et al. A prospective study of triglyceride level, low-density lipoprotein particle diameter, and risk of myocardial infarction. *JAMA* 1996;276:882–8.

36. Ulmer H, Kollerits B, Kelleher C, Diem G, Concin H. Predictive accuracy of the SCORE risk function for cardiovascular disease in clinical practice: a prospective evaluation of 44,649 Austrian men and women. *Eur J Cardiovasc Prev Rehabil* 2005;12:433–41.

37. Hart R, Doherty DA. The potential implications of a PCOS diagnosis on a woman's long-term health using data linkage. *J Clin Endocrinol Metab* 2015;100:911–9.

38. Glintborg D, Mumm H, Hougaard D, Ravn P, Andersen M. Ethnic differences in Rotterdam criteria and metabolic risk factors in a multiethnic group of women with PCOS studied in Denmark. *Clin Endocrinol (Oxf)* 2010;73:732–8.

39. Ilanne-Parikka P, Eriksson JG, Lindstrom J, Hamalainen H, Keinanen-Kiukaanniemi S, Laakso M, et al. Prevalence of the metabolic syndrome and its components: findings from a Finnish general population sample and the Diabetes Prevention Study cohort. *Diabetes Care* 2004;27:2135–40.

40. Nystrom PK, Carlsson AC, Leander K, de Faire U, Hellenius ML, Gigante B. Obesity, metabolic syndrome and risk of atrial fibrillation: a Swedish, prospective cohort study. *PLoS One* 2015;10:e0127111.

41. Hildrum B, Mykletun A, Hole T, Midthjell K, Dahl AA. Age-specific prevalence of the metabolic syndrome defined by the International Diabetes Federation and the National Cholesterol Education Program: the Norwegian HUNT 2 study. *BMC Public Health* 2007;7:220.

SUPPLEMENTAL TABLE 1

Prevalence of the metabolic syndrome (MetS) and its components in different study populations and different age groups according to the Rotterdam criteria for metabolic syndrome^a.

MetS component	< 30 y			30–39 y			> 39 y		
	Control	NA-PCOS	HA-PCOS	Control	NA-PCOS	HA-PCOS	Control	NA-PCOS	HA-PCOS
Met-S	7 (5.1) ^{b,c}	62 (26.3) ^b	64 (26.7) ^c	16 (18.0) ^{b,c}	57 (37.5) ^b	68 (40.0) ^c	12 (18.2) ^c	8 (19.5) ^d	14 (42.4) ^{c,d}
Waist or BMI ^a	41 (33.6) ^{b,c}	172 (53.6) ^b	159 (50.3) ^c	57 (50.0) ^{b,c}	147 (65.6) ^b	171 (69.8) ^c	35 (46.1) ^c	18 (41.9) ^d	31 (73.8) ^{c,d}
TG ^a	5 (2.9) ^{b,c}	33 (19.0) ^b	54 (17.6) ^c	12 (10.1) ^{b,c}	33 (22.4) ^b	62 (25.2) ^c	10 (13.2) ^c	8 (17.4)	14 (32.6) ^c
HDL ^a	35 (21.9) ^{b,c}	89 (51.1) ^b	138 (45.2) ^c	34 (30.9) ^{b,c}	88 (61.1) ^{b,d}	101 (42.4) ^{c,d}	16 (21.3)	10 (21.7)	13 (30.2)
BP ^a	21 (15.9) ^{b,c}	111 (33.5) ^b	131 (38.0) ^c	28 (25.2) ^{b,c}	108 (47.8) ^b	132 (50.2) ^c	30 (40.5) ^c	23 (50.0)	30 (62.5) ^c
Glucose ^a	4 (8.0)	33 (13.3)	22 (17.2)	5 (15.2)	27 (16.6) ^d	27 (27.6) ^d	15 (21.1)	7 (18.9)	11 (34.4)

Note: Data presented as n (%). BMI = body mass index; BP = blood pressure; HA = hyperandrogenic; HDL = high-density lipoprotein; LDL = low-density lipoprotein; NA = normoandrogenic; PCOS = polycystic ovary syndrome; TG = triglycerides.

^a Rotterdam criteria for metabolic syndrome (MBS), three out of five: waist circumference >88 cm and/or BMI $\geq 30 \text{ kg/m}^2$, triglycerides $\geq 1.70 \text{ mmol/L}$, HDL $\leq 1.30 \text{ mmol/L}$, BP ≥ 130 and/or $\geq 85 \text{ mm Hg}$, fasting glucose $\geq 6.11 \text{ nmol/L}$ and/or 2-hour OGTT glucose $\geq 7.78 \text{ nmol/L}$.

^b Statistically significant difference between reference and NA-PCOS groups.

^c Statistically significant difference between reference and HA-PCOS groups.

^d Statistically significant difference between NA-PCOS and HA-PCOS groups.

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SUPPLEMENTAL TABLE 2

Sources of the subpopulations in the study.

Population	PCOS population			Control population		
	n	Source	Age (y), range (mean)	n	Source	Age (y), range (mean)
Population 1 ^a	319	Clinic	19–39 (28.2)	–	–	–
Population 2 ^{b,c,d}	104	Clinic	18–59 (37.9)	124	Community	19–62 (33.2)
Population 3 ^e	118	Community/clinic	18–38 (29.8)	52	Community/clinic	18–39 (28.6)
Population 4 ^f	91	Community/clinic	18–41 (28.6)	–	–	–
Population 5 ^g	70	Community/clinic	18–58 (42.0)	75	Community/clinic	33–59 (43.2)
Population 6 ^h	488	Community/clinic	15–50 (29.4)	116	Community/clinic	20–54 (30.9)
Population 7 ⁱ	43	Community/clinic	21–39 (30.8)	–	–	–
Population 8 ^j	317	Community/clinic	14–45 (27.8)	80	Community/clinic	19–51 (32.0)

^a Morin-Papunen et al. 2000.^b Puurunen et al. 2009.^c Piltonen et al. 2004.^d Piltonen et al. 2012.^e Stener-Victorin et al. 2010.^f Vanký et al. 2003.^g Hudécová et al. 2009.^h Glintborg et al. 2012.ⁱ Nybacka et al. 2011.^j Unpublished.Pinola. Metabolic profile in women with PCOS. *Fertil Steril* 2016.