Antimüllerian hormone levels and cardiometabolic risk in young women with polycystic ovary syndrome

Rebecca A. Feldman, M.D., Kathleen O'Neill, M.D., Samantha F. Butts, M.D., M.S.C.E., and Anuja Dokras, M.D., Ph.D.

Department of Obstetrics and Gynecology, University of Pennsylvania, Philadelphia, Pennsylvania

Objective: To determine the association between antimüllerian hormone (AMH) levels and metabolic syndrome (MetSyn) in young women with polycystic ovary syndrome (PCOS).
Design: Cross-sectional study.
Setting: Academic PCOS center.
Patient(s): A total of 252 women aged 18–46 years with PCOS.
Intervention: None.
Main Outcome Measure(s): Association of AMH with markers of cardiometabolic risk and MetSyn.
Result(s): The median AMH level was 5.1 ng/mL (interquartile range [IQR] 3.0–8.1), and prevalence of MetSyn was 23.8%. AMH levels positively correlated with total T, high-density lipoprotein (HDL) cholesterol, and SHBG and negatively correlated with fasting glucose, homeostasis-model assessment of insulin resistance, body mass index (BMI), and systolic and diastolic blood pressure. A single-unit decrease in AMH was associated with an 11% increase in odds of MetSyn (odds ratio [OR] 1.11, 95% confidence interval [CI] 1.03–1.20); the strength of this association was maintained in the multivariate model (OR 1.09, 95% CI 1.01–1.18) adjusting for age and race. Subjects with AMH values in the lowest tertile were twice as likely as those in the highest tertile to have MetSyn (adjusted OR 2.1, 95% CI 1.01–4.3). Total T was not associated with MetSyn or its individual components.
Conclusion(s): Our findings indicate that in young women with PCOS, low AMH levels predict a greater risk of MetSyn. The role of AMH, an established biomarker of ovarian reserve, in risk stratification of cardiometabolic risk in obese women with PCOS needs to be clarified in longitudinal studies and in the perimenopausal population. (Fertil Steril® 2017;107:276–81. ©2016 by American Society for Reproductive Medicine.)
Key Words: Antimüllerian hormone, polycystic ovary syndrome, metabolic syndrome, cardiovascular disease, testosterone

Discuss: You can discuss this article with its authors and with other ASRM members at https://www.fertstertdialog.com/users/16110-fertility-and-sterility/posts/12554-22343

Women with polycystic ovary syndrome (PCOS) are at an increased risk for insulin resistance (1, 2), metabolic syndrome (MetSyn) (3, 4), type 2 diabetes mellitus (DM), and possibly cardiovascular disease (CVD) (5). Numerous reports have provided evidence that PCOS is associated with subclinical atherosclerosis with increased carotid intima media thickness, coronary artery calcifications (6, 7), and likelihood of hospitalization related to CVD (8). Given the relationship between PCOS and traditional coronary artery disease (CAD) risk factors, identifying biomarkers, especially early in life, that can further stratify the CVD risk is crucial to disease prevention. Antimüllerian hormone (AMH) is a glycoprotein secreted by the granulosa cells in the ovary. Knowledge of its function in the ovary is limited, but it has been demonstrated to be a reliable marker of ovarian reserve and aging (9). AMH levels are significantly elevated in PCOS, secondary to an increased number of pre-antral and antral follicles in the ovary (10, 11), and remain higher in perimenopause (12, 13), reflecting a slower trajectory of reproductive senescence. It is, however, unclear if these higher levels of AMH affect or predict overall cardiovascular (CV) risk in PCOS.

Reproductive aging is an established risk factor for CVD risk in women, as evidenced by the increased incidence of CAD after the menopausal transition. Declining estrogen levels associated with changes in lipid profiles and coronary artery dynamics (i.e.,

Received April 30, 2016; revised October 3, 2016; accepted October 4, 2016; published online November 11, 2016.
R.A.F. has nothing to disclose. K.O. has nothing to disclose. S.F.B. has nothing to disclose. A.D. has nothing to disclose.
Reprint requests: Anuja Dokras, M.D., Ph.D., Department of Obstetrics and Gynecology, University of Pennsylvania, 3701 Market Street, Suite 800, Philadelphia, PA 19104 (E-mail: adokras@obgyn.upenn.edu).

Fertility and Sterility® Vol. 107, No. 1, January 2017 0015-0282/$36.00

http://dx.doi.org/10.1016/j.fertnstert.2016.10.009

blood flow, plaque formation) support a role for ovarian hormones in CAD. It has been proposed that early changes in atherosclerotic risk may also be apparent by studying the association between AMH and established CVD risk factors. A few studies have examined this with mixed results. In a cross-sectional study of 951 healthy reproductive-age women, low AMH levels were associated with decreased high-density lipoprotein cholesterol (HDL-C), higher waist circumference, and hypertensive status but were not independent from body mass index (BMI) (14). In contrast, a longitudinal study over 12 years including 1,015 premenopausal women demonstrated lower AMH levels to be associated with high low-density lipoprotein cholesterol (LDL-C) and triglycerides (TG), even after controlling for BMI (15). An inverse relationship between AMH and obesity has been described in women with and without PCOS (16), but there are limited data examining the relationship between AMH and other cardiometabolic markers, including glucose and lipids, in women with PCOS. The relationship between AMH and MetSyn, a predictor for risk of type 2 DM and CVD, may be further complicated in PCOS owing to the increased prevalence of MetSyn and higher levels of AMH compared with age-matched control subjects (12, 17). Given the complexity of these relationships and the importance of investigating CVD risk in women with PCOS, we hypothesized that low AMH, an indicator of ovarian reserve, is associated with higher risk of MetSyn. To test this hypothesis we performed a cross sectional study in a well defined population with PCOS to evaluate the relationship between serum AMH levels and MetSyn.

MATERIALS AND METHODS

This study was a retrospective analysis of women 18–46 years of age with PCOS evaluated at the Penn PCOS Center from January 1, 2011, through January 1, 2014. Women are referred to this center from a variety of sources, including gynecologists, primary care and other subspecialty physicians, and self-referral. The Institutional Review Board at the University of Pennsylvania approved this study. PCOS was diagnosed with the use of modified Rotterdam criteria (18) such that all women had ovulatory dysfunction with hyperandrogenism (Ferriman-Gallwey hirsutism score ≥ 8 [19]), an elevated T level (20), or a polycystic ovary according to transvaginal ultrasound.

Laboratory tests and measurements from the physical examination, such as age, blood pressure, height, weight, calculated BMI weight (kg)/height $(m)^2$, total cholesterol, TG, C-reactive protein (CRP), HDL-C, LDL-C, glucose, and insulin, were recorded from the subject's initial evaluation. Additional laboratory tests required to establish the diagnosis of PCOS were TSH, PRL, DHEAS, and 17OH-P. AMH was measured with the use of the Gen II ELISA (Beckmann Coulter), lipids were measured by means of standard enzymatic methods, and total T was measured by means of liquid chromatography–mass spectrometry (ARUP Laboratories). Total cholesterol, TG, HDL-C, and glucose were measured with the use of standard enzymatic methods. LDL-C was mathematically derived. Insulin was measured by means of RIA (EMD Millipore), and plasma glucose was measured by means of the oxidase method. Homeostasis-model assessment of insulin resistance (HOMA-IR) was defined as fasting glucose $\times$ fasting insulin/22.5.

Patients were included if they met the diagnosis of PCOS and had a complete metabolic work up as described above. MetSyn was defined by modified National Cholesterol Education Program–Adult Treatment Panel III criteria with the use of three of the following five criteria BMI ≥ 30 kg/m^2, systolic blood pressure (BP) ≥ 135 mm Hg and diastolic BP ≥ 85 mm Hg or taking antihypertensive medications, fasting glucose ≥ 100 mg/mL or taking medications for DM, TG ≥ 150 mg/mL, and HDL-C ≤ 50 mg/mL (21).

Statistical Analysis

Continuous parameters were summarized as mean $\pm$ SD and median (interquartile range [IQR]). Relationships between continuous variables were tested with the use of Spearman correlations and simple linear regression as appropriate. Continuous variables were further compared according to grouping variables with the use of the Wilcoxon rank sum test. Categoric variables were compared with the use of χ^2 tests. Logistic regression was used to evaluate the relationship between MetSyn and AMH while adjusting for relevant confounders. Model covariates selected for inclusion were retained in the final model if their P value was $\leq .10$ or if the main effect estimate changed by $>15\%$ when the covariate was added. All statistical tests were performed with the use of Stata 12. A P value of $<.05$ was considered to be the threshold for statistical significance.

RESULTS

Two hundred fifty-two women meeting criteria for PCOS with available AMH levels and complete evaluation for MetSyn were included in the study. The mean age of the group was 28.4 $\pm$ 5.6 years, 49.2% were nonwhite, and 11.5% were smokers (Table 1). Of these women, 73.8% met the National Institutes of Health criteria for PCOS. The median AMH level was 5.1 ng/mL (IQR 3.0–8.1), median T level was 49 ng/dL (IQR 33.5–64.5), and the prevalence of MetSyn in this cohort

TABLE 1

Demographic characteristics of 252 women with PCOS included in study [mean ± SD or % (n)].

Age, y	28.4 ± 5.6
Race	
White	50.8% (124)
Nonwhite	49.2% (120)
Hispanic	2.8% (7)
Nulligravid	66.2% (147)
Ferriman-Gallwey score	11.1 ± 7.8
Total T, ng/dL	52.5 ± 26
Free T, pg/mL	6.8 ± 4.7
AMH, ng/mL	6.6 ± 6.1
Current smokers	11.5% (27)

Note: AMH = antimüllerian hormone; PCOS = polycystic ovary syndrome.

Feldman. AMH and cardiometabolic risk in PCOS. Fertil Steril 2016.

TABLE 2

Metabolic characteristics of women with PCOS [mean ± SD or % (n)].

BMI, kg/m^2	33 ± 9.5
Obese (BMI ≥30 kg/m^2)	58.7% (145)
Systolic BP, mm Hg	124.5 ± 14.3
Diastolic BP, mm Hg	73.7 ± 9.7
Total cholesterol, mg/dL	183.7 ± 38.7
LDL-C, mg/dL	110.5 ± 31.7
HDL-C, mg/dL	52.6 ± 16.6
Triglycerides, mg/dL	108.5 ± 68.5
Fasting glucose, mg/dL	86.4 ± 17.5
Fasting insulin, mg/dL	14.7 ± 13.07
SHBG, nmol/L	61.0 ± 51.1

Note: BMI = body mass index; BP = blood pressure; HDL-C = high-density lipoprotein cholesterol; LDL-C = low-density lipoprotein cholesterol; PCOS = polycystic ovary syndrome.

Feldman. AMH and cardiometabolic risk in PCOS. Fertil Steril 2016.

was 23.8%. Table 2 presents the metabolic parameters in the study group.

The correlation between AMH and selected markers of cardiometabolic risk are shown in Figure 1. AMH levels positively correlated with total T ($P<.001$) and free T ($P<.04$). On examination of individual components of MetSyn, AMH levels positively correlated with HDL-C ($P=.009$) and SHBG ($P<.05$) and negatively correlated with fasting glucose ($P=.002$), total insulin ($P<.0001$), HOMA-IR ($P<.001$), BMI ($P<.0001$), and systolic and diastolic BP ($P=.0001$ and $P=.004$, respectively). AMH did not have a significant relationship with smoking status, LDL-C, TG, CRP, or DHEAS. On examining the individual criteria of MetSyn, mean AMH was significantly higher in subjects with HDL ≥50 mg/dL ($P<.008$) and significantly lower in subjects with BP ≥135 mm Hg ($P<.007$) but this relationship was attenuated by BMI. Total T was not associated with MetSyn or its individual components.

A single-unit increase in AMH was associated with a 10% decline in odds of MetSyn (odds ratio [OR] 0.9, 95% confidence interval [CI] 0.83–0.98; $P=.01$); this association was maintained in the multivariate model (adjusted OR 0.91, 95% CI 0.85–0.99; $P=.02$) that adjusted for age and race. Compared with women with values in the highest AMH tertile, those in the lowest AMH tertile had a greater than twofold increased risk of MetSyn (OR 2.4, 95% CI 1.2–4.6; $P=.02$). Adjusted for age and race, the OR for MetSyn in the lowest AMH tertile was 2.1 (95% CI 1.01–4.3; $P=.046$) compared with the highest AMH tertile. We therefore restricted our analysis to only obese subjects, comparing the lowest and the highest quartiles of AMH, and the OR for MetSyn was 1.1 (95% CI 0.47–2.64); however, the number of subjects in this group was small.

DISCUSSION

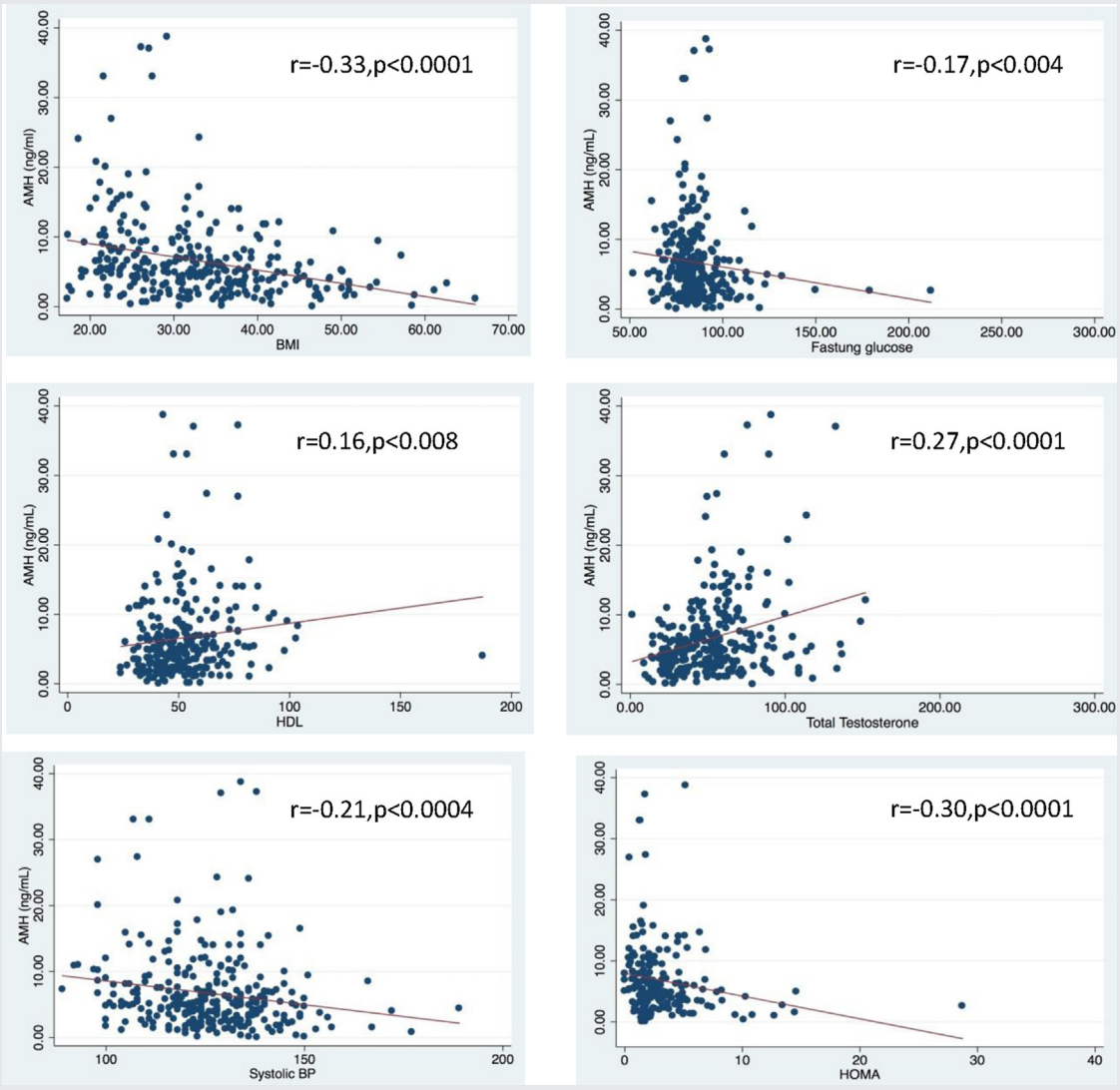
Because young women with PCOS are at an increased risk for MetSyn, it is important to identify factors that might alter this risk. In the present study, AMH levels correlated significantly with traditional CV risk factors such as HDL-C, glucose, HOMA-IR, BMI, and BP. Furthermore, young women with PCOS and low AMH levels had an increased risk of MetSyn, indicating a role for AMH in CV risk stratification in this high-risk population. However, studies with larger numbers of obese subjects are needed to assess the independent association of AMH and MetSyn.

As mentioned in the introduction, few studies have examined the relationship between AMH and metabolic parameters in the general population, and results have been mixed (14, 15). In the PCOS population, a positive relationship between AMH and insulin/HOMA-IR has been reported (22, 23). However, the data on comprehensive assessment of the association between AMH and other cardiometabolic risk factors in this population are limited. One study reported a positive correlation between AMH and LDL-C (22), whereas a larger Chinese study did not show an association between AMH and insulin resistance or components of the lipid panel (23). The majority of subjects in the latter study were lean, and differences in AMH assays may explain the differences in the results in these studies. Given the heterogeneity in assessment of individual CVD risk factors, we also examined the association between AMH levels and MetSyn to understand the composite risk in this young population.

Age, T, and BMI are proposed confounders in the relationship between AMH and metabolic risk. Age-related decrease in AMH and increase in MetSyn risk have been clearly described in PCOS (12, 17). Also, a role for androgens is supported by studies demonstrating that women with the hyperandrogenic PCOS phenotype have a higher metabolic risk in both reproductive and perimenopausal periods (24, 25). Interestingly, androgens at high concentrations inhibit AMH mRNA levels in bovine granulosa cells, although the absence of an androgen response element on the AMH promoter suggests that this effect is indirect (26). In our cohort, although T correlated with AMH, it did not correlate with individual components of MetSyn and therefore was not retained in the final multivariable logistic regression model. The correlation between AMH and MetSyn remained significant after controlling for both age and T, suggesting that other mechanisms contribute to the association.

Insulin and obesity also may mediate the relationship between AMH and MetSyn. Obesity, i.e., visceral adiposity, is integral to the definition of MetSyn and is also associated with low AMH levels. As reported previously (16), we also found an inverse correlation between AMH and fasting insulin, HOMA-IR, and BMI. Although insulin regulates androgen production in the ovary, the impact of insulin on AMH production is unclear. Use of metformin in the treatment of obese hyperinsulinemic women with PCOS decreased insulin and AMH levels, suggesting an insulin-mediated effect on AMH production (27). We have recently shown that oral contraceptive pill treatment, resulting in lower serum androgens, and weight loss interventions are both independently associated with a decrease in AMH levels (28). Other investigators have demonstrated a role for leptin in the regulation of AMH production by cumulus granulosa cells obtained from ovarian follicles during in vitro fertilization treatment (29). In the present study, the relationship between individual components of MetSyn and AMH was significantly attenuated by BMI. These findings are similar to those reported in healthy reproductive-

FIGURE 1



Correlations between antimüllerian hormone (AMH) and selected biomarkers of cardiometabolic risk in women with polycystic ovary syndrome (PCOS). BMI = body mass index; BP = blood pressure; HOMA = homeostasis-model assessment of insulin resistance.

Feldman. AMH and cardiometabolic risk in PCOS. Fertil Steril 2016.

age women when assessing AMH and individual components of MetSyn (8). Because we used BMI as one of the components of MetSyn, we were unable to evaluate the association of AMH and MetSyn independently from obesity. Our data suggest the need for larger studies in obese women with PCOS to precisely define the independent relationship between AMH and cardiometabolic risk.

The present study suggests that low AMH levels may be a marker of higher cardiometabolic risk in reproductive-age women with PCOS. Similarly, other models suggest a link between AMH and CVD risk in women. Two large studies showed that AMH levels were significantly lower in women who developed preeclampsia and hypertensive disorders in pregnancy compared with normotensive women (30, 31). We found an inverse relationship between AMH and systolic and diastolic BP in our population; however this relationship was attenuated by obesity. In women with type 2 DM, one study has reported lower antral follicle counts and higher FSH levels compared with healthy control subjects (32). Finally, AMH levels negatively correlated with subclinical atherosclerosis as measured by iliac artery plaque size in monkeys (33). Regardless, longitudinal studies are needed to understand if AMH, an early marker of ovarian aging, and CV risk are temporally and causally related in PCOS.

Our study has several strengths: To our knowledge it includes the largest cohort of well characterized young women with PCOS and examination of individual components of cardiometabolic risk as well as a composite score, namely, MetSyn. To better understand the interplay between AMH and

MetSyn risk, we controlled for known variables, including age, race, and T levels. We recently reported a significantly higher prevalence of MetSyn in black women compared with white women with PCOS (4), and the present study included a large proportion of black women. A limitation of the present study is the cross-sectional analysis, which prevented us from assessing the longitudinal changes in this relationship. A 10-year longitudinal study in the general population showed that the decrease in AMH levels during perimenopause was significantly associated with increase in BMI (34). Although T levels are normalized in most women with PCOS in perimenopause, the prevalence of obesity remains high in this population (35). In contrast, because women with PCOS have higher AMH levels in perimenopause, it can be postulated that continued secretion of ovarian hormones may have a persistent cardioprotective effect. Therefore, the relationship between AMH and MetSyn needs to be evaluated in women in the menopause transition to better understand the longitudinal impact of our findings. Although we did not have corresponding E_2 levels, the cyclicity of E_2 and lack of menstrual regularity in the PCOS population would limit its utility.

In summary, our findings indicate that in young women with PCOS, low AMH levels, independently from age and race, may predict a higher risk of MetSyn. Although the precise longitudinal impact of the high prevalence of traditional CV risk factors on coronary event rates and mortality in PCOS is unclear (36), appropriate risk stratification in young women with PCOS is recommended (37). AMH is currently an established biomarker of ovarian reserve, but future studies in a perimenopausal cohort of women with PCOS are needed to clarify its precise role in CV risk stratification independently from obesity.

REFERENCES

1. Carmina E, Lobo RA. Use of fasting blood to assess the prevalence of insulin resistance in women with polycystic ovary syndrome. Fertil Steril 2004;82:661–5.
2. 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.
3. Joham AE, Ranasinha S, Zoungas S, Moran L, Teede HJ. Gestational diabetes and type 2 diabetes in reproductive-aged women with polycystic ovary syndrome. J Clin Endocrinol Metab 2014;99:E447–52.
4. Hillman JK, Johnson LN, Limaye M, Feldman RA, Sammel M, Dokras A. Black women with polycystic ovary syndrome (PCOS) have increased risk for metabolic syndrome and cardiovascular disease compared with white women with PCOS [corrected]. Fertil Steril 2014;101:530–5.
5. Dokras A. Cardiovascular disease risk in women with PCOS. Steroids 2013;78:773–6.
6. Shroff R, Kerchner A, Maifeld M, van Beek EJ, Jagasia D, Dokras A. Young obese women with polycystic ovary syndrome have evidence of early coronary atherosclerosis. J Clin Endocrinol Metab 2007;92:4609–14.
7. Talbott EO, Zborowski JV, Rager JR, Boudreaux MY, Edmundowicz DA, Guzick DS. Evidence for an association between metabolic cardiovascular syndrome and coronary and aortic calcification among women with polycystic ovary syndrome. J Clin Endocrinol Metab 2004;89:5454–61.
8. 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.
9. van Rooij IA, Broekmans FJ, Scheffer GJ, Looman CW, Habbema JD, de Jong FH, et al. Serum antimüllerian hormone levels best reflect the reproductive decline with age in normal women with proven fertility: a longitudinal study. Fertil Steril 2005;83:979–87.
10. Iliodromiti S, Kelsey TW, Anderson RA, Nelson SM. Can anti-müllerian hormone predict the diagnosis of polycystic ovary syndrome? A systematic review and meta-analysis of extracted data. J Clin Endocrinol Metab 2013;8:3332–40.
11. Homburg R, Crawford G. The role of AMH in anovulation associated with PCOS: a hypothesis. Hum Reprod 2014;29:1117–21.
12. Piltonen T, Morin-Papunen L, Koivunen R, Perheentupa A, Ruokonen A, Tapanainen JS. Serum antimüllerian hormone levels remain high until late reproductive age and decrease during metformin therapy in women with polycystic ovary syndrome. Hum Reprod 2005;20:1820–6.
13. Tehrani FR, Solaymani-Dodaran M, Tohidi M, Gohari MR, Azizi F. Modeling age at menopause using serum concentration of antimüllerian hormone. J Clin Endocrinol Metab 2013;98:729–35.
14. Bleil ME, Gregorich SE, McConnell D, Rosen MP, Cedars MI. Does accelerated reproductive aging underlie premenopausal risk for cardiovascular disease? Menopause 2013;11:1139–46.
15. Tehrani FR, Erfani H, Cheraghi L, Tohidi M, Azizi F. Lipid profiles and ovarian reserve status: a longitudinal study. Hum Reprod 2014;11:2522–9.
16. de Kat AC, Broekmans FJ, Laven JS, van der Schouw YT. Anti-müllerian hormone as a marker of ovarian reserve in relation to cardio-metabolic health: a narrative review. Maturitas 2015;80:251–7.
17. Dokras A, Bochner M, Hollinrake E, Markham S, Vanvoorhis B, Jagasia DH. Screening women with polycystic ovary syndrome for metabolic syndrome. Obstet Gynecol 2005;106:131–7.
18. Rotterdam ESHRE/ASRM-Sponsored PCOS Consensus Workshop Group. Revised 2003 consensus on diagnostic criteria and long-term health risks related to polycystic ovary syndrome. Fertil Steril 2004;81:19–25.
19. Legro RS, Brzyski RG, Diamond MP, Coutifaris C, Schlaff WD, Alvero R, et al. The Pregnancy in Polycystic Ovary Syndrome II study: baseline characteristics and effects of obesity from a multicenter randomized clinical trial. Fertil Steril 2014;101:258–69.e8.
20. Legro RS, Schlaff WD, Diamond MP, Coutifaris C, Casson PR, Brzyski RG, et al. Total testosterone assays in women with polycystic ovary syndrome: precision and correlation with hirsutism. J Clin Endocrinol Metab 2010;95:5305–13.
21. Grundy SM, Cleeman JI, Daniels SR, Donato KA, Eckel RH, Franklin BA, et al. Diagnosis and management of the metabolic syndrome: an American Heart Association/National Heart, Lung, Blood Institute scientific statement. Circulation 2005;112:2735–52.
22. Skalba P, Cygal A, Madej P, Dąbkowska-Huć A, Sikora J, Martirosian G, et al. Is the plasma antimüllerian hormone (AMH) level associated with body weight and metabolic, and hormonal disturbances in women with and without polycystic ovary syndrome? Eur J Obstet Gynecol Reprod Biol 2011;2:254–9.
23. Cui Y, Shi Y, Cui L, Han T, Gao X, Chen ZJ. Age-specific serum antimüllerian hormone levels in women with and without polycystic ovary syndrome. Fertil Steril 2014;1:230–6.
24. Polotsky AJ, Allshouse A, Crawford SL, Harlow SD, Khalil N, Santoro N, et al. Relative contributions of oligomenorrhea and hyperandrogenemia to the risk of metabolic syndrome in midlife women. J Clin Endocrinol Metab 2012;97:E868–77.
25. Daan N, Louwers Y, Koster M, 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.
26. Crisosto N, Sir-Petermann T, Greiner M, Maliqueo M, Moreno M, Aedo P, et al. Testosterone-induced downregulation of anti-müllerian hormone expression in granulosa cells from small bovine follicles. Endocrine 2009;36:339–45.
27. Romualdi D, de Cicco S, Tagliaferri V, Proto C, Lanzone A, Guido M. The metabolic status modulates the effect of metformin on the antimüllerian hormone–androgens–insulin interplay in obese women with polycystic ovary syndrome. J Clin Endocrinol Metab 2011;96:E821–4.

28. Legro RS, Dodson WC, Kris-Etherton PM, Kunselman AR, Stetter CM, Williams NI, et al. Randomized controlled trial of preconception interventions in infertile women with polycystic ovary syndrome. J Clin Endocrinol Metab 2015;100:4048–58.
29. Merhi Z, Buyuk E, Berger DS, Zapantis A, Israel DD, Chua S Jr, et al. Leptin suppresses antimüllerian hormone gene expression through the JAK2/STAT3 pathway in luteinized granulosa cells of women undergoing IVF. Hum Reprod 2013;28:1661–9.
30. Yarde F, Maas AH, Franx A, Eijkemans MJ, Drost JT, van Rijn BB, et al. Serum AMH levels in women with a history of preeclampsia suggest a role for vascular factors in ovarian aging. J Clin Endocrinol Metab 2014;99:579–86.
31. Shand AW, Whitton K, Pasfield A, Nassar N, McShane M, Han X, et al. Evaluation of antimüllerian hormone in the first trimester as a predictor for hypertensive disorders of pregnancy and other adverse pregnancy outcomes. Aust N Z J Obstet Gynaecol 2014;54:244–9.
32. Isik S, Ozcan HN, Ozuguz U, Tutuncu YA, Berker D, Alimli AG, et al. Evaluation of ovarian reserve based on hormonal parameters, ovarian volume, and antral follicle count in women with type 2 diabetes mellitus. J Clin Endocrinol Metab 2012;97:261–9.
33. Appt SE, Chen H, Clarkson TB, Kaplan JR. Premenopausal antimüllerian hormone concentration is associated with subsequent atherosclerosis. Menopause 2012;19:1353–9.
34. Freeman EW, Gracia CR, Sammel MD, Lin H, Lim LC, Strauss JF 3rd. Association of antimüllerian hormone levels with obesity in late reproductive-age women. Fertil Steril 2007;87:101–6.
35. Carmina E, Campagna AM, Lobo RA. Emergence of ovulatory cycles with aging in women with polycystic ovary syndrome (PCOS) alters the trajectory of cardiovascular and metabolic risk factors. Hum Reprod 2013;28:2245–52.
36. de Groot PC, Dekkers OM, Romijn JA, Dieben SW, Helmerhorst FM. PCOS, coronary heart disease, stroke and the influence of obesity: a systematic review and meta-analysis. Hum Reprod Update 2011;17:495–500.
37. Wild RA, Carmina E, Diamanti-Kandarakis E, Dokras A, Escobar-Morreale HF, Futterweit W, et al. Assessment of cardiovascular risk and prevention of cardiovascular disease in women with the polycystic ovary syndrome: a consensus statement by the Androgen Excess and Polycystic Ovary Syndrome (AE-PCOS) Society. J Clin Endocrinol Metab 2010;95:2038–49.