High Level of Anti-Mullerian Hormone (AMH) as Predictor for Polycystic Ovary Syndrome among Women Women of Reproductive Age at Giri Emas Public Hospital

Melissa Edelweishia,[1] Bagus Komang Satriyasa,[2] I Gusti Ayu Widianti,[2] J. Alex Pangkahila,[2] I Nyoman Mangku Karmaya,[2] I Gede Ngurah Harry Wijaya Surya[3]

[1]Biomedical Science Master Study Program, Faculty of Medicine, Udayana University, Denpasar Bali, Indonesia

[2]Faculty of Medicine, Udayana University, Denpasar Bali, Indonesia

[3]Department of Obstetric and Gynecology, Prof.Dr. I.G.N.G Ngoerah Hospital, Denpasar, Bali, Indonesia

[*]Corresponding: Melissa Edelweishia, E-mail: melissa.edelweishia@gmail.com

Abstract

Introduction: Polycystic Ovary Syndrome (PCOS) is the most prevalent endocrine condition in women, affecting 5 – 10% of women who are of reproductive age. Together with other Rotterdam criteria, elevated blood AMH levels are considered a significant diagnostic for PCOS and may be used as a powerful predictor to reflect the certainty of the diagnosis of PCOS in women of reproductive age.

Objective: This study aims to prove high AMH as a predictor for PCOS.

Method: This study is an analytic study with a case-control study design. A total of 30 respondents were divided into PCOS and control groups. All women were subjected to anthropometric assessments such as measurement of height, weight, BMI, and trans-abdominal ultrasonography for ovaries. Data analysis was carried out using independent t-tests and Chi-Square tests.

Result: The data analysis revealed that the PCOS group's mean AMH levels were considerably different ($p<0.05$), with 6.5±1.75 greater than the control group's 3.34±0.64. AMH levels were found to be twice as high in the PCOS group as in the control group. AMH levels and PCOS incidence were compared using the Chi-Square test; the odd ratio is 17.875 (95% CI = 2.73 -116.8; p=0.001).

Conclusion: High levels of AMH at reproductive age can 18 times predict the risk of PCOS.

Keywords: AMH, PCOS, oligomenorrhea, hyperandrogenism, anovulation

Kadar Antimullerian Hormon (AMH) Tinggi sebagai Prediksi Sindrom Polikistik Ovarium pada Wanita Usia Reproduksi di RSUD Giri Emas

Abstrak

Pendahuluan: Sindrom Ovarium Polikistik (SOPK) merupakan kelainan endokrin yang menyerang kira-kira 5 - 10% wanita usia subur dan dianggap sebagai kelainan endokrin yang paling umum pada wanita. Kadar AMH serum yang meningkat saat ini dianggap sebagai penanda penting untuk SOPK dan dapat digunakan sebagai prediktor kuat untuk mencerminkan kepastian diagnosis SOPK pada wanita usia subur bersama dengan kriteria Rotterdam lainnya.

Tujuan: Penelitian ini bertujuan untuk membuktikan kadar AMH yang tinggi sebagai prediktor SOPK.

Metode: Penelitian ini merupakan penelitian analitik dengan desain studi kasus kontrol. Sebanyak 30 responden akan dibagi menjadi kelompok SOPK dan kelompok kontrol. Semua wanita menjalani penilaian antropometri seperti pengukuran tinggi badan, berat badan, BMI, dan ultrasonografi trans-abdominal untuk ovarium. Analisis data dilakukan dengan uji t independen dan uji Chi-Square.

Hasil: Analisis data menunjukkan rerata kadar AMH pada kelompok SOPK lebih tinggi 6,5±1,75 dibandingkan dengan kontrol 3,34±0,64 dan berbeda bermakna ($p<0,05$). Ditemukan kadar AMH pada kelompok SOPK dua kali lebih tinggi dibandingkan dengan kontrol. Untuk mengetahui hubungan kadar AMH dengan kejadian PCOS digunakan uji Chi-Square dan odd ratio menunjukkan 17,875 (95% CI = 2,73 -116,8 ; p=0,001).

Kesimpulan: Dapat disimpulkan bahwa pada penelitian ini kadar AMH yang tinggi pada usia reproduksi dapat memprediksi risiko PCOS sebesar 18 kali

Kata kunci: AMH, PCOS, oligomenorea, hiperandrogen, anovulasi

 DOI: 10.24198/obgynia.v8i1.812

Introduction

Polycystic Ovary Syndrome (PCOS) is a group of symptoms and signs of hyperandrogenism and anovulation caused by endocrine system disorders. PCOS is considered a multifactorial disorder with various genetic, endocrine, and environmental.[1]

Polycystic Ovary Syndrome affects about 4% - 12% of women, especially women of reproductive age with unclear etiology. Due to significant variations in the clinical presentation of PCOS, the prevalence may differ among populations. It showed 4.8% in whites and 8% in black women in the southeastern United States, 6.8% in white women in Greece, 6.5% in white women in Spain, 6.3% in South Asia in Sri Lanka, and 5% in Thai women. [2] Meanwhile, in Indonesia, a study in Palembang shows that 78.8% of 279 women who went to the private practice of doctors experienced PCOS.[3]

Moreover, PCOS is associated with several clinical manifestations, including obesity, impaired glucose tolerance, metabolic syndrome, type 2 diabetes mellitus, dyslipidemia, and cardiovascular disease. The severity and presentation of these symptoms can vary greatly between populations and individuals. Current management depends on symptomatic treatment and mitigation of risk factors for associated conditions.[4]

The 2023 updated worldwide evidence-based guideline for assessing and treating PCOS still recommends the Rotterdam criteria for PCOS diagnosis. However, they are now more precisely defined and evidence-based. Two of the three criteria listed below must be met for an adult diagnosis: (1) ovulatory dysfunction (OD), (2) clinical or biochemical hyperandrogenism (HA), and (3) polycystic ovary morphology (PCOM). The 2023 worldwide PCOS guideline includes serum anti-Müllerian hormone (AMH) testing as an alternative to ultrasonography, previously used to assess PCOM. Over the last 20 years, research has shown that serum AMH levels correlate with the number of antral follicles on ultrasonography, making it a useful and affordable diagnostic for identifying PCOM. [5]

PCOS diagnosis among women of the reproductive age using AMH levels has been studied in various populations. The interindividual variability of AMH is high, mainly because of the varying number of follicles in the group of subjects of the same age.[6]

A glycoprotein with a monomer molecular size of roughly 72 kDa and a multimer size ranging from 145 to 235 kDa, anti-Müllerian hormone (AMH), also called Müllerian Inhibiting Substance (MIS), is secreted by the granulosa cells of the small antral and preantral follicles and controls follicular development. AMH levels may rise as a result of PCOS's compromised folliculogenesis, which can cause an excessive buildup of tiny preantral and antral follicles. Previous studies indicate that AMH levels may also be linked to ovarian hyperandrogenism in both healthy women and those with PCOS, and they correlate with the number of ovarian follicles.[6]

Based on this background, this study aims to identify elevated levels of AMH as risk factors for PCOS in women of reproductive age. Together with other Rotterdam criteria, this study sheds light on the pattern and use of serum AMH levels as a diagnostic factor in PCOS patients.

Method

This study is an analytic study with a case-control design. It was conducted at the Obstetrics and Gynecology Polyclinic of the Giri Emas Public Hospital Buleleng. It involved 15 PCOS patients and 15 healthy controls who fulfilled the inclusion and exclusion criteria. It began in September 2022 and was continued until the required number

of samples was obtained. The Faculty of Medicine, Universitas Udayana's Research Ethics Commission (KEP) has approved the ethical viability of this study in the form of a Statement of Ethical Eligibility (Ethical Clearance) Number: 2555/UN.14.2.2.VII.14/LT/2022.

The study's inclusion criteria for the PCOS group are as follows: 1) Women of reproductive age between 16 and 35; 2) Women who have been diagnosed with PCOS according to the Rotterdam ESHRE/ASRM criteria, meaning they meet at least two of the three criteria: oligo/anovulation, clinical or biochemical hyperandrogenism, and polycystic ovaries on sonographic examination; 3) Women who have never undergone surgery for ovarian abnormalities; 4) Women who did not engage in intense aerobic exercise or take medications that impact lipid profiles, carbohydrate metabolism, or reproductive function, such as oral contraceptive pills, at least two months before the study. Meanwhile, the exclusion criteria include 1) A history of diabetes mellitus, 2) thyroid disease, 3) hyperprolactinemia; and 4) Cushing's syndrome. Healthy controls with normal menstrual cycles with 21–35 days intervals satisfied the requirements. Written informed permission was obtained from each individual involved.

Every patient had a complete physical examination and medical history taken. Oligomenorrhea was defined as a greater than 35-day interval between periods or less than eight menses per year. A lack of menstruation for six months or more was regarded as amenorrhea. Each female was assessed anthropometrically by ovarian trans-abdominal ultrasonography, height, weight, and BMI.

Moreover, the height and weight were assessed upon enrollment to determine the current BMI. The following classification was used: BMI <18.5 kg/m^2= underweight, BMI ≥18.5 to 22.9 kg/m^2 = normal, BMI 23 to 24.9 kg/m^2= overweight, and BMI ≥25 kg/m^2= obese.

The electrochemiluminescence immunoassay "ECLIA," which is designed to be used with Elecsys and Cobas e-immunoassay analyzers (Elecsys AMH), was used to assess anti-Mullerian hormone. AMH values were shown as nanograms per milliliter. Based on several fertility studies, we chose serum AMH >3.5, which is regarded as high, for this investigation.[5]

SPSS version 23.0 was used for all statistical analyses. Mean ± SD was used to express AMH values. For continuous variables, data were displayed as mean ± SD; categorical variables were displayed as numbers and percentages. The independent t-test was used for the statistical analysis, and a significance threshold of $p<0.05$ was set. The odds ratio was computed to determine the association between high AMH.

Results

Most of the PCOS participants were in the 16–25 age group (9 individuals, or 60%), with the lowest percentage being in the 26–35 age group (6 individuals, or 40%). In the control group, the highest percentage was in the 26–35 age group (10 individuals, or 66.7%), and the lowest percentage was in the 16–25 age group (5 individuals, or 33.3%). Furthermore, according to the Body Mass Index (BMI) standards, the PCOS research subject group was distributed as follows: norm weight (4 individuals, 26.7%), overweight (3 individuals, 20%), and obese (8 individuals, 53.3%). Five individuals (33.3%) in the control group were norm weight, four (26.7%) were overweight, and six (40%) were obese. Amenorrhea was reported in 6 individuals (40%), oligomenorrhea in 9 individuals (60%), and polycystic ovarian morphology from ultrasonography was identified in all PCOS patients in 15 individuals (100%), as

Table 1 Distribution of Patients According to the Variables of the Study.

Variables	PCOS (n=15)		Control (n=15)	
	N	%	N	%
Age groups (years)				
16-25	5	33,3	9	60
26-35	10	66,7	6	40
Body Mass Index (BMI)				
Normoweight	4	26,7	5	33,3
Overweight	3	20	4	26,7
Obese	8	53,3	6	40
AMH (ng/ml)				
<3.5	2	13,3	11	73,3
>3.5	13	86,7	4	26,7
Menstrual Period				
Amenorea	6	40	0	0
Oligomenorea	9	60	0	0
Normal	0	0	15	100
Hirsutism				
Yes	7	46,7	2	13,3
No	8	53,3	13	86,7
Morphology PCO				
Yes	15	100	0	0
No	0	0	15	100

Table 2. Distribution Characteristic BMI on PCOS and Control Group

Variables	PCOS (n=15)		Control (n=15)		P
	Mean	SD	Mean	SD	
Age (year)	27,40	3,08	25,20	4,19	0,113
Weight (kg)	64,87	10,67	61,40	11,30	0,395
Height (cm)	158,13	6,48	157,2	6,47	0,698
BMI (kg/m2)	25,97	3,70	24,77	4,50	0,433

shown in the respondents' distribution table (Table 1).

In this case-control study, the variables of age, height, weight, and BMI were examined using the independent t-test. It shows that the PCOS group and the control group had the same subject characteristics based on BMI. Since the results of the independent t-test indicate that there is no appreciable difference between the two study groups' attributes ($p>0.05$), they are considered equal and worthy of comparison. Table 2 shows that the PCOS group had a statistically higher BMI than the control group (25,97 ± 3,70 vs. 24,77 ± 4,50 kg/m2; $p<0.05$).

Using the Enzyme-Linked Immunosorbent Assay (ELISA) technique, AMH levels were measured in both research groups. Table 3 indicates that the mean AMH levels in the PCOS group were greater than those in the controls. Based on statistical analysis utilizing the independent t-test, the

Table 3 Mean AMH Levels in the PCOS Group and Control Group

Variable	PCOS (n=15)		Control (n=15)		P
	Mean	SD	Mean	SD	
AMH	6,50	1,75	3,34	0,64	0,001

Table 4 AMH on PCOS and Control Group

Variable	Group		OR	CI 95%	p
	Case	Control			
AMH > 3,5 ng/ml	13	4	17,875	2.73-116.8	0,001
AMH < 3,5 ng/ml	2	11			

mean AMH levels of the PCOS and control groups differed significantly, with a p-value <0.05 (p=0.001).

The Chi-Square test was performed to ascertain the association between AMH levels and the incidence of PCOS. The results showed that high levels of AMH might predict the risk of PCOS by 18 times, with an Odd Ratio of 17,875 (95% CI = 2.73 -116.8; p=0.001). As seen in table 4.

Discussion

The average AMH level in the PCOS group was 6.5±1.75, whereas the control group's mean AMH level was 3.34±0.64, and the results of the study utilizing the independent t-test showed that these two groups were substantially different (p<0.05). Consequently, it was discovered that the PCOS group's mean AMH levels were greater than those of the control group. In addition, the Chi-Square test was used to analyze the difference in AMH levels between the two groups using a 2 x 2 cross table. The results showed that 13 individuals in the PCOS group had AMH levels > 3.5 ng/ml, and two individuals had AMH levels <3.5 ng/ml, while four individuals in the control group had AMH levels > 3.5 ng/ml, and 11 individuals had AMH levels <3.5 ng/ml. According to the findings of the Chi-Square test analysis, the p-value was 0.001, and the Odd Ratio was 17.875 (95% CI = 2.73-116.8). Accordingly, a rise in AMH levels can 18 times predict the likelihood of PCOS.

AMH was considerably greater in PCOS patients (9.21 ± 0.50 versus 4.40 ± 0.41 ng/ml, p < 0.001) than in healthy controls, according to the same study by Sadiqa-Tuqn et al. (2017). The results showed that the sensitivity and specificity of AMH were 67% and 78.33%, respectively, when the cut-off value was maintained at 3.5 ng/ml. [5]

The granular cells of antral and preantral regions of tiny ovarian follicles produce the glycoprotein known as the anti-Müllerian hormone. Higher levels of serum AMH were linked to ovarian reserve than either FSH or estradiol, and they also showed a substantial correlation with antral follicle count. According to a study by Khmil, women with PCOS had higher blood AMH levels, which supported the findings of prior research. The increased quantity of tiny antral follicles in PCOS may cause this. Obesity and other conditions that influence granular cell activity can also impact on AMH production. The primary cause of changes in serum AMH in PCOS-afflicted women is decreased gonadotropin and steroid hormone production.[7]

According to several studies, women with PCOS have serum AMH levels that are around two to three times higher than those of healthy people. The number of follicles in the

ovaries, as observed on ultrasound, as well as the levels of testosterone, LH, and other variables linked to PCOS, are correlated with AMH levels. Therefore, in situations when a trustworthy ultrasound is unavailable, AMH values may be suggestive of PCOS in women with hyperandrogenism and oligomenorrhea or amenorrhea.[8] At the moment, there isn't a single AMH cut-off point for evaluating PCOM. The revised PCOS guideline has advised using demographic and assay-specific cutoffs due to variations in laboratory techniques across the globe. However, when it comes to establishing diagnostic cut-offs for a clinical illness like PCOS, the conventional definition of the "normal" range—a cut-off of within two standard deviations—is inappropriate. Clustering with other clinical characteristics, including clinical hyperandrogen, oligo-anovulation, or forecasting long-term health effects like fertility, are more crucial factors in this case. Sadly, there isn't much comprehensive research on PCOS that uses cluster analysis.[5]

Although the blood AMH threshold for PCOS diagnosis has not yet been established, an enzyme immunoassay (AMH-EIA) value of 35 pmol/L, or 4.9 ng/ml, has been suggested. It has been proposed that when diagnosing PCOS, oligo or anovulation and hyperandrogenism should be established first, and other differential diagnoses should be ruled out. The AMH value can be utilized to help with the diagnosis if any of the mentioned criteria are absent. It is advised that each healthcare facility employ its assay-specific reference interval for diagnosis until the AMH threshold values are widely accepted. [9]

Ozay et al. (2020) examined the levels of AMH in healthy women and PCOS patients with different phenotypic and polycystic shapes. According to the study's findings, women with PCOS have higher serum AMH levels than both healthy women and individuals with polycystic ovarian morphology alone. Even though AMH is a useful indication for ovarian reserve testing, there is not enough information available to establish a threshold value for PCOS diagnosis or to distinguish between PCOS phenotypes. Higher amounts of AMH were found in the hyperandrogenic PCOS phenotypes (A, B, and C) in the current investigation than in the normoandrogenic phenotype D. All three elements of the syndrome were present in phenotypic A, which had the highest amounts of AMH. Statistically speaking, PCOS phenotypic A had much higher AMH levels than PCOS phenotype D. In their study, Piouka et al. demonstrated that the degree of PCOS was connected adversely with BMI and that the greatest amounts of AMH were seen in phenotypic A. [10]

However, Butt et al. (2022) discovered that women with PCOS had a longer menstrual cycle, higher blood AMH levels, and a higher chance of menstrual problems. Antral follicle count, a biomarker for ovarian response, is strongly connected with serum AMH. In PCOS women, elevated perantral secretion may be a prediction of oligo-/amenorrhea based on serum AMH levels. [11]

According to the Butt et al. research from 2022, a high mean blood AMH level of 7.23 ± 4.67 ng/ml was also seen in 70.6% of PCOS women. An independent t-test revealed a significant mean difference ($p < 0.05$) in the AMH level between menstrual patterns with oligo-/amenorrhea and those without. There was a significant difference ($p < 0.05$) in the mean group comparison between PCOS women with normal and high AMH levels. [11]

According to the Iranian study, women with PCOS had substantially higher AMH levels (6.21, IQR: 3.90 -9.03 versus 1.70, IQR: 0.85-2.81; $P<0.001$) and were younger (27.9±4.6 compared 33.1±4.6) than the control group. [12]

Conclusions

With mean AMH levels of 6.50 ± 1.75 and the control group's mean AMH levels of 3.34 ± 0.64, respectively, and a significant difference ($p < 0.05$), it may be inferred that the PCOS group had two times higher AMH levels than the healthy women throughout the reproductive phase. High levels of AMH can predict the risk of PCOS by 18 times, according to the findings of the chi-square test of high AMH levels against PCOS, which yielded RO = 17.875.

Reference

1. Oyebanji OG, Asaolu MF, Amonimo EO. Hormonal Imbalance in Polycystic Ovarian Syndrome (PCOS) in Teaching Hospitals in Ekiti State, Nigeria. Open J Obstet Gynecol. 2018;08(13):1456–64.
2. Stracquadanio M, Ciotta L. Metabolic Aspects of PCOS [Internet]. London: Springer International Publishing; 2015. 1–122 p. Available from: http://link.springer.com/10.1007/978-3-319-16760-2
3. Mareta R, Amran R, Larasati V. Hubungan Polycystic Ovary Syndrome (PCOS) dengan Infertilitas di Praktik Swasta Dokter Obstetri Ginekologi Palembang. Majalah Kedokteran Sriwijaya. 2018;50(2):85–91.
4. Lentscher JA, Slocum B, Torrealday S. Polycystic Ovarian Syndrome and Fertility. Clin Obstet Gynecol. 2021 Mar;64(1):65–75.
5. Piltonen TT, Viita-Aho J, Saarela U, Melin J, Forslund M. Utility of Serum Anti-Müllerian Hormone Measurement as Part of Polycystic Ovary Syndrome Diagnosis. Vol. 42, Seminars in Reproductive Medicine. 2024. p. 49–59.
6. Sharma P, Chawla R, Ahuja R, Gupta U. Anti-müllerian hormone as a surrogate marker for hormonal dysfunction and sonographic pattern in polycystic ovarian syndrome. Journal of SAFOG. 2019;11(3):175–80.
7. Khmil M, Khmil S, Marushchak M. Hormone imbalance in women with infertility caused by polycystic ovary syndrome: Is there a connection with body mass index? Open Access Maced J Med Sci. 2020;8(June):731–7.
8. Kohzadi M, Choobsaz F, Khazaei M. New Findings on Anti-Mullerian Hormone in Polycystic Ovarian Syndrome Patients. Gynecol Obstet Open Acc. 2018;02(01):1–7.
9. Toosy S, Sodi R, Pappachan JM. Lean polycystic ovary syndrome (PCOS): an evidence-based practical approach. J Diabetes Metab Disord. 2018;17(2):277–85.
10. Ozay AC, Ozay OE, Gulekli B. Comparison of Anti-müllerian Hormone (AMH) and Hormonal Assays for Phenotypic Classification of Polycystic Ovary Syndrome. Ginekol Pol. 2020;91(11):661–7.
11. Butt MS, Saleem J, Aiman S, Zakar R, Sadique I, Fischer F. Serum anti-Müllerian hormone as a predictor of polycystic ovarian syndrome among women of reproductive age. BMC Womens Health. 2022;22(1).
12. Tehrani FR, Mansournia MA, Solaymani-Dodaran M, Azizi F. Age-specific serum anti-Müllerian hormone levels: estimates from a large population-based sample. Climacteric. 2014 Oct 9;17(5):591–7.