# The Diagnostic Value of Anti-Mullerian Hormone and Follicular Stimulating Hormone in Polycystic Ovary Syndrome Patients

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***Abstract***

***Objective: To evaluate the anti-mullerian hormone (AMH) and follicular stimulating hormone (FSH) in polycystic ovary syndrome patients (POCS) and their potential as markers of PCOS.***

***Methods: This was a comparative cross-sectional study carried out on 80 women aged 18-50 years: 40 were healthy women with non-polycystic ovary syndrome serving as control group and 40 were patients with PCOS. Fasting venous blood (6 ml) was collected from each individual in the second day of the menstrual cycle and AMH, FSH, luteinizing hormone (LH), prolactin and estradiol (E2) as well as fasting blood glucose (FBG) and lipid profile were measured.***

***Results: Serum AMH was significantly (p = 2x10-4) higher in PCOS patients by 3.5-folds with respect to the control group, whereas FSH was significantly (p = 0.01) lower by 36.5%. In contrast, LH, prolactin, estradiol, FBG and the lipid profile were non-significantly different between the two tested groups. Serum AMH was negatively correlated with age (r = - 0.355, p = 0.001) and FSH (r = - 0.454, p = 2x10-6).***

***Conclusion: This study shows that AMH can be used as a diagnostic and prognostic marker in POCS.***

*Keywords:* **Polycystic ovary syndrome, Anti-Mullerian hormone, Follicular stimulating hormone.**

## I. INTRODUCTION

Polycystic ovary syndrome (PCOS) is a common endocrinopathy that is prevalent in 5-10% of women of childbearing age [1]. It is characterized by oligo-or hyperandrogenemia, and polycystic ovarian ultrasonography [1,2]. It is clinically manifested as menstrual thinning, hemorrhoids, hairiness, obesity and infertility. It is also characterized by abnormal levels of reproductive hormones, which can lead to anovulatory, infertile and menstrual disorders [1,3]. About 50% of women with PCOS fulfill the criteria of metabolic syndrome and that PCOS is frequently associated with insulin resistance accompanied by compensatory hyperinsulinemia, resulting in an increased risk for the development of Type 2 DM and cardiovascular disease [2,4]. In comparison with healthy women, PCOS have higher level of anti–mullerian hormone (AMH) that is a peptide produced by granulose cells of follicles that is widely considered as a highly sensitive marker of ovarian reserve [4]. Anti-Müllerian hormone is a dimeric glycoprotein belonging to the transforming growth factor β (TGF-β) superfamily [5]. It has also been proven that it has a strong influence on the function of ovaries, especially on the growth of follicles [6,7]. Anti-Müllerian hormone is supposed to regulate the number of growing follicles and their selection for ovulation [8] and is a negative regulator of early stages of the follicular development [9]. Moreover, AMH inhibits the recruitment and growth of follicles by restricting growth factors and the effect of gonadotropins, especially the follicle-stimulating hormone (FSH) [10]. The level of AMH can reflect the number of ovarian antrum follicles, ovarian reserve, and ovarian function [11]. Serum AMH is not affected by menstrual cycle and oral contraceptive use, so it has potential as a marker for the diagnosis of PCOS [12]. Studies have shown that the level of circulating AMH is two- to threefold higher in women with PCOS than in healthy women of childbearing age, probably due to increased follicular mass associated with PCOS [8,9]. In view of the fact that the increase in AMH levels has been reported to be associated with PCOS as well as longterm effect pf PCOS, the aim of this study was to explore the diagnostic value of AMH and other hormones in Yemeni patients with PCOS.

## II. MATERIALS AND METHODS

The study was a comparative cross–sectional study that was carried out on 80 women aged 18-50 years: 40 were healthy women with non-polycystic ovary syndrome serving as control group and 40 were patients with PCOS attending the University of Science and Technology Hospital and C-Pluse Hospital in Sanaa city for infertility evaluation during the period from September 2015 to February 2016. PCOS was diagnosed based clinical and biochemical signs of hyperandrogenism (HA); and presence of polycystic ovaries (PCOs), defined as the presence of ≥ 12 follicles measuring 2-9 mm in diameter in each ovary and/or increased ovarian volume (> 10 ml). The exclusion criteria were as follows: Cushing's syndrome, dysfunctional uterine bleeding, primary amenorrhea, adrenal cortical hyperplasia or tumor, thyroid

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dysfunction. The study protocol was approved by the institutional review board (IRB) of the Faculty of Medicine and Health Sciences, Sana'a University. Informed consent form was obtained from all individuals after explaining the purpose and nature of the study. The patients' height and weight were measured and body mass index (BMI), defined as weight (kg)/ height squared (m2), was calculated. Standardized questionnaire(s) was administered to collect participants' demographic and clinical data. Venous blood (6 ml) was collected from each individual after an overnight fast of more than 10 hours in the second day of the menstrual cycle and divided into two vacuumed tubes; 4 ml into plain tubes for immediate measurement of AMH, FSH, Luteinizing hormone (LH), prolactin and estradiol (E2); and the remaining 2 ml for biochemical analysis. The serum from each sample was separated within 30 minutes and immediately stored at 20ºC.

## III. BIOCHEMICAL ANALYSIS

Fasting blood glucose (FBG), triglyceride (TG), total cholesterol, HDL-cholesterol (HDL-c), and LDL-cholesterol (LDL-c) were measured on an automated analyzer, the Cobas Integra 400/400 (Roche Diagnostic, Germany), using the respective Roche Diagnostic kits. The AMH, FSH, LH, prolactin and E2 were measured by Electrochemiluminescence reagent kit on the Cobas e411 Immunoassay (Roche Diagnostics, Germany).

## IV. STATISTICAL ANALYSIS

The statistical analyses were performed on Social Package of Social Sciences (SPSS) version 2000 (SPSS Inc, Chicago, IL, USA). Results were expressed as means ±SD and analyzed by independent sample t-test. Pearson correlation used to measure the correlation as well as the sensitivity, specificity and accuracy of AMH, FSH, and LH. The significant difference was indicated if p value was < 0.05.

## V. RESULTS

The results presented in Table 1 show the biochemical and hormonal parameters in polycystic ovary syndrome patients. Anti-mullerian hormone was significantly (p = 2x10-4) higher in PCOS patients by 3.5-folds as compared with the control group, whereas FSH was significantly (p = 0.01) lower by 36.5%. In contrast, LH, prolactin and estradiol were non-significantly different between the two tested groups. Similarly, FBG, TG, cholesterol, HDL-c and LDL-c levels were non-significantly different between the two groups.

**Table 1:** Biochemical and hormonal parameters of PCOS patients and control subjects

| | Control (n = 40) | PCOS (n = 40) | p-value |
|---|---|---|---|
| **Age (years)*** | 32.20±5.68 | 30.43±4.58 | 0.12 |
| **BMI (kg/m$^2$)*** | 24.67±3.98 | 25.67±5.03 | 0.32 |
| **Triglyceride (mmol/L)** | 1.14 (1.0 – 1.3) | 1.17 (1.0 – 1.3) | 0.87 |
| **Cholesterol (mmol/L)*** | 4.24±0.96 | 4.22±0.85 | 0.92 |
| **HDL-c (mmol/L)** | 1.0 (0.88 – 1.23) | 1.0 (0.8 – 1.54) | 0.21 |
| **LDL-c (mmol/L)*** | 2.70±0.9 | 2.60±0.8 | 0.69 |
| **FBG (mg/dl)** | 93.0 (79.4 - 100) | 100.0 (100 - 109) | 0.19 |
| **AMH (ng/ml)** | 1.27 (1.25 – 2.0) | 4.5 (3.8 – 6.0) | **2x10$^{-4}$** |
| **FSH (MIU/ml)** | 7.40 (5.6 -9.5) | 4.7 (3.8 – 6.0) | **0.01** |
| **LH (IU/L)** | 6.1 (5.0 – 7.7) | 7.5 (6.3 – 10.0) | 0.19 |
| **Prolactin (ng/ml)** | 14.7 (12.5 – 15.8) | 16.9 (12.5 – 19.9) | 0.26 |
| **Estradiol (pg/ml)** | 47.86 31.6 – 63.0) | 57.54 (39.8 – 63.0) | 0.23 |

Data are expressed as geometric mean (95% confidence interval of mean); * Expressed as Mean ±SD

Table 2 shows the Pearson correlation between anti-mullerian hormone and other tested parameters in PCOS patients. Of all the parameters tested, AMH was negatively correlated with age (r = - 0.355, p = 0.001) and FSH (r = - 0.454, p = 2x10-6).

**Table 2:** Pearson correlation between anti-mullerin hormone with age and with FSH in PCOS patients and non-PCOS women

| Variables | R | p-value |
|---|---|---|
| **AMH- age** | - 0.355 | **0.001** |
| **AMH – FSH** | **- 0.454** | **2x10-6** |

*P-Value ≤ 0.05 s considered significant.

Table 3 shows the sensitivity, specificity and accuracy of AMH, FSH and LH tests in the diagnosis of PCOS.

**Table 3:** Sensitivity, specificity and accuracy of AMH, FSH and LH tests

| | Sensitivity | Specificity | Accuracy |
|---|---|---|---|
| **AMH** | 60% | 96% | 88.8% |
| **FSH** | 27.5% | 86% | 27.5% |
| **LH** | 27.5% | 93% | 79.9% |

## VI. Discussion

The results presented in our study showed the AMH level to be significantly higher in PCOS patients, which is in agreement with several studies [13-16]. This increase could be attributed to increased production by individual follicles in women with PCOS [17]. Early antral follicles are increased in numbers in women with PCOS that leads to increase production of AMH [18]. Anti-mullerian hormone production was reported to be approximately 75times higher in each polycystic ovarian granulosa cells [13]. Moreover, increased mRNA expression of AMH levels were found in PCOS, also caused by disturbances in folliculalogenesis,, resulting in the accumulation of excessive pre-antral and small antral follicles [19]. Our study also showed significant negative correlation between AMH and age which is in agreement with a previous study [20]. This may be due to decline in the follicular reserve of the ovaries [21]. Moreover, our result showed significant negative correlation between AMH and FSH PCOS, which is in support of the suggestion that AMH inhibits the responsiveness of follicles to FSH mediated by AMH RII that induces adenelyl cycles activation and aromates expiration [18,22,23].

On the other hand, FSH was significantly lower in women with PCOS which is in agreement with an earlier study [18]. This decrease in FSH may be due to suppression effect (decrease production) or increase serum level of AMH in PCOS patients. The increase of AMH inhibits the recruitment of the primary follicle and diminishes the response of selected follicle to FSH stimulation [7]. In contrast, serum levels of LH were non-significantly different in PCOS patients, which are in disagreement with a previous study reporting increased level of LH in PCOS women [24] and suggesting that it was due to arrested folliculogenesis. Moreover, another study demonstrated that AMH directly increased LH pulsality and secretion, machinated by AMHRII receptors on the surface of GnRH neurons [25]. Similarly, estrodial showed no significant difference in the PCOS group, which is disagreement with an earlier study reporting reduction of E2 production in PCOS women [24] due to the increase relationship between serum level of AMH and E2. The AMH affects E2 production by decreasing the aromatase activity [26]. The lack of effect in LH and E2 levels in our study may be attributed to the fact that all our patients were under treatment.

The sensitivity and specificity of AMH was 60% and 96%, respectively, which is in agreement with previous studies [27,28] reporting a sensitivity and specificity of 67% and 92% respectively. On the other hand, the sensitivity of both FSH and LH were less at 27.5%.

## 12. Conclusion

The results presented in this study show that AMH may have potential as a marker of PCOS and therefore can be used as a diagnostic and prognostic marker tool in POCS patients.

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