



The Prevalence of Polycystic Ovarian Morphology in Sonographic Evaluations of Iranian Adolescent Girls

Razieh Shahnazari^{1,2}, Farahnaz Farzaneh³ and Yasaman Sharifi^{4*}

¹Department of Radiology, Firoozabadi Hospital, Iran University of Medical Sciences, Tehran, Iran

²Firoozabadi Clinical Research Development Unit (FCRDU), Iran University of Medical Sciences, Tehran, Iran

³Department of Obstetrics and Gynecology, School of Medicine, Iran University of Medical Sciences, Tehran, Iran

⁴Department of Radiology, School of Medicine, Iran University of Medical Sciences, Tehran, Iran

*Corresponding Author: Yasaman Sharifi, Department of Radiology, School of Medicine, Iran University of Medical Sciences, Tehran, Iran.

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Abstract

Objectives: The significance of polycystic ovarian morphology (PCOM) in adolescence remains uncertain. This study aimed to determine the prevalence of PCOM in adolescent girls and to explore the relationship between PCOM and Hyperandrogenism, using clinical diagnostic criteria and testosterone levels as laboratory diagnostic markers in this population.

Methods: One hundred adolescent girls, aged 9 to 19 years, were referred for ultrasonography in this study. Transabdominal ultrasound and blood samples were collected during the follicular phase.

Results: PCOM was observed in 86% of the subjects. Subjects with PCOM showed a higher prevalence of hirsutism with 62.8% versus 14.3% (P-Value = 0.01). Girls with PCOM had a higher prevalence of abnormal total testosterone levels than girls without PCOM (24% versus 4%; P = 0.04). Subjects with PCOM also revealed higher ovarian volume (Right ovary: 14.43 ± 5.8 versus 8.57 ± 1.98 P-Value = 0.01 Left Ovary: 14.66 ± 5.8 versus 8.14 ± 2.26 P-Value = 0.006) and follicle numbers per ovary (Right ovary: 15.6 ± 4.2 versus 9.14 ± 2.85 P-Value = 0.001 Left Ovary: 16.02 ± 4.27 versus 10.86 ± 3.97 P-Value = 0.004) as expected.

Conclusions: The prevalence of polycystic ovary morphology (PCOM) among Iranian adolescent girls is reported to be 86%. Those with PCOM exhibit increased ovarian volumes and higher follicle counts per ovary. Additionally, there is a higher occurrence of hirsutism and elevated total testosterone levels in participants with PCOM.

Keywords: Polycystic Ovarian Morphology; Adolescent Girls; Hyperandrogenism; Ovarian Volume; Follicle Number

Background

An endocrine disorder known as polycystic ovarian syndrome (PCOS) is characterized by specific hormonal and metabolic abnormalities. This condition involves between 5-18% of women of reproductive age and 6-18% of adolescent girls worldwide, making it one of the most widespread medical issues affecting women

and girls globally [1,2]. This condition is a multifaceted disorder of elevated androgen production with recognizable symptoms and a spectrum of difficulties with fertility and reproduction. Additionally, conditions like metabolic syndrome and insulin resistance may coexist with this condition [1-3].

Although PCOS is one of the most common endocrine disorders among women of reproductive age [1], the diagnosis of this syndrome during adolescence, which is known based on the World Health Organization definition of age 10 to 19 years, can be challenging due to the overlapping of clinical symptoms and morphological changes of the ovaries in the sonographic view with the physiological changes of puberty (irregular periodic cycles, acne, and polycystic morphological changes of the ovaries in the ultrasound evaluation) [3,4]. The diagnostic guidelines for polycystic ovary syndrome so far include the National Institute of Health (NIH) guideline in 1990 and the Rotterdam guideline in 2003 [5], as well as the Androgen Excess and Polycystic Ovary Syndrome Society (AE-PCOS) guideline [6] and the latest International evidence-based guideline for the assessment and management of polycystic ovary syndrome [7].

The clinical and laboratory Hyperandrogenism and irregular menstruation at least two years after menarche should be used to diagnose PCOS in adolescents [3,8], and the morphological appearance of PCOM in sonographic evaluation is currently not a diagnostic criterion for PCOS in adolescents [3]. In recent years, upgraded diagnostic criteria for PCOS in adolescent females have been introduced [9]. These new criteria include the specification of irregular menstrual cycles and Hyperandrogenism but exclude polycystic ovarian morphology [9,10].

Studies indicate that one-third of hyperandrogenic or PCOS girls exhibit polycystic ovary morphology (PCOM) [11-13].

According to some studies, PCOM is associated with high anti-Müllerian hormone levels (AMH) in young girls with regular menstruation, as well as greater severity of primary dysmenorrhea in this population [13,14].

Due to the absence of established guidelines for diagnosing polycystic ovarian syndrome in adolescents, we initiated this research. The occurrence of polycystic ovarian morphology among Iranian teenage girls and its correlation with various clinical and laboratory markers has not been previously explored. Recognizing the significance of early identification of polycystic ovarian syndrome and its impact on patients, we investigated the prevalence of PCOM using ultrasound observations in adolescent girls at Firouzabadi Hospital in Tehran.

Materials and Methods

Subjects

In a cross-sectional study, we studied 100 post-menarchal girls between 9 and 19 years old (mean \pm SD 15.66 \pm 2.32 years old). Participants were carefully recruited from Firouzabadi Hospital in the heart of Rey City, Tehran, an area characterized by middle to low-income residents. This strategic selection aimed to ensure diverse representation and address this community's unique health challenges [13]. Inclusion criteria for the study included clinical signs of hyperandrogenism, such as moderate to severe acne or hirsutism, as indicated by a Ferriman-Gallwey (FG) score of 7 or higher. Additionally, an absence of regular menstrual cycles was considered, where regular cycles are defined as those occurring every 21 to 45 days, based on the consensus guidelines from the American College of Obstetrics and Gynecology (ACOG) and the American Association of Pediatrics (AAP) for adolescents [13,15]. The exclusion criteria included the use of oral contraceptives, steroids, or any other medications; the presence of concurrent chronic conditions such as genetic syndromes, coeliac disease, renal disease, liver disease, cardiac disease, or malnutrition; and premature pubertal development, which is defined as the appearance of pubic hair in girls under the age of 8. All participants had normal levels of fasting blood glucose, thyroid hormones, and prolactin. The protocol was approved by the institutional review board (Iran University of Medical Sciences) under the approval ID IR.IUMS.FMD.REC.1401.670. Upon entering the study, parents gave informed consent, and volunteers signed a written consent form. Adolescent girls over the age of 18 signed the consent form.

Study protocol

Girls were evaluated during the follicular phase of their menstrual cycle, specifically from Days 1 to 7 [13]. A thorough clinical and physical examination was performed. The age at menarche was determined retrospectively. Menstrual cycle duration was calculated by averaging the lengths of the previous three cycles. Gynecological age was defined as the number of years since menarche at the time of the participant's entry into the study. Body Mass Index (BMI) and Ferriman-Gallwey (FG) scores were assessed. Overweight was categorized as a BMI in the 85th to 94th percentile, whereas obesity was defined as a BMI exceeding the 95th to 97th percentile, depending on the growth chart utilized. An early-morning blood sample was obtained during the follicular phase (Days 1 to 7 of the cycle) to measure total and free testosterone levels [13].

Ultra sonographic study

PCOM, as defined by the Rotterdam consensus, is characterized by either the presence of 12 or more follicles measuring 2 to 9 mm in diameter or an ovarian volume (OV) of 10 ml or more in one or both ovaries. These criteria reflect changes in ovarian structure often linked to hormone imbalances [5,7,13]. On the same day as the blood sample was collected, a single observer (C.V.) underwent transabdominal ultrasonography (TAUS). The examination was conducted using a 5-MHz transabdominal probe with a Medison SonoAce 6000C (Medison, Seoul, Korea). Measurements were obtained in real-time using the highest magnification to examine the ovaries. To accurately calculate the OV, we measured the longest medial axis (length) along with its thickness and width. OV is precisely calculated using the following formula designed specifically for a prolate ellipsoid: $OV = \frac{1}{4} \pi \times \text{length} \times \text{width} \times \text{thickness}$ [13,16]. The follicular number (FN) was evaluated through a detailed examination of each ovary, meticulously scanning from the inner edges to the outer borders in a longitudinal cross-section. To accurately determine the size of each follicle, the mean of the largest diameter and the corresponding perpendicular diameter were calculated. This approach allowed for a precise measurement, enhancing the understanding of follicular development and distribution within the ovarian structure. All follicles between 2.0 and 9.0 mm were counted. The ultrasound showed intra-observer variation coefficients of 3.2% for ovarian volume (OV) and 4.1% for follicular number (FN) [13,17]. When a significant cyst or follicle measuring greater than 10 mm is identified, a follow-up ultrasonography examination is conducted during the subsequent menstrual cycle.

Laboratory assays

Serum testosterone levels, both total and free, were measured using a competitive specific binding radioimmunoassay, which had a coefficient of variation (CV) of 8.1% between tests and 5.3% within tests [13].

Free testosterone was calculated based on total testosterone and Sex Hormone Binding Globulin (SHBG) levels.

A total testosterone level exceeding 0.9 ng/dL is considered elevated and abnormal. Levels of Thyroid Stimulating Hormone (TSH) and prolactin are measured to exclude other conditions that mimic PCOS.

Statistical analysis

Normal distributions were assessed through the Kolmogorov-Smirnov test. To analyze continuous variables in girls who met and did not meet the PCOM criteria, the Student's t-test was employed [13]. The effects of chronological age, age at menarche, gynecological age, and BMI on the presence of PCOM were examined using logistic regression analysis. Pearson's correlation analysis was conducted to evaluate the relationship between continuous variables. A significance threshold of 5% was established. Statistical analyses were performed using SPSS for Windows version 18.0 [13].

Results

Participants included 86 patients with PCOM (adolescent girls) with a mean (SD) age of 15.26 (2.13) years and 14 participants without PCOM with a mean (SD) age of 15.9 (2.44) years. Table 1 displays the clinical characteristics of all participants categorized by the presence of PCOM. Polycystic ovarian morphology (PCOM) was detected in 86% of the adolescent girls included in the study (Figure 1). Generally, subjects with PCOM had higher BMI, and subjects without PCOM had higher age and distance from menarche age but these differences were not statistically significant. Moreover, among participants with PCOM, those with hirsutism generally had a higher frequency but irregular menstruation and severe acne didn't show significant differences among those with and without PCOM.

Table 2 shows the association between ultrasonographic characteristics of girls without PCOM and with PCOM. Subjects with PCOM showed higher uterus size (length, depth, and thickness) and endometrial thickness but these differences were not statistically significant. Among participants with PCOM, ovary size, and FNO measures were significantly associated with a higher value; the corresponding measures and p-values were right ovary volume 14.43 ± 5.8 (0.01), left ovary volume 14.66 ± 5.8 (0.006), right-sided FNO 15.6 ± 4.2 (0.001), and left-sided FNO 16.02 ± 4.27 (0.004) respectively. However, among participants without PCOM, all of these measures were lower and they showed lower ovarian size and lower FNO in each ovary.

Table 3 shows the hormonal profiles among those with and without PCOM. There is no significant association among levels of total, free testosterone, TSH, and prolactin compared between subjects with and without PCOM. However, there is a significant

association between the frequency of people with abnormal total testosterone test levels among groups with PCOM and those without PCOM. The frequency of abnormal testosterone is higher in patients with PCOM (24% vs. 4%) and this difference is statistically significant (P = 0.04) (Figure 2).

There was no correlation between different groups of BMI (underweight, normal, overweight, and obese) among subjects with and without PCOM.

	PCOM (-)	PCOM (+)	P-Value
N(%)	14(14)	86(86)	-
Age (years)	15.9± 2.44	15.26±2.13	0.35
Distance from Age of menarche (years)	3.71±1.49	3.26± 1.51	0.46
BMI (kg/m ²)	21.82±4.1	22.78±3.71	0.53
Hirsusism:Yes n(%)	1(14.3)	27(62.8)	0.01
Hirsusism:No n(%)	6(32.7)	16(85.7)	-
Irregular menstruation n(%)	2(28.6)	37(86)	0.33
Regular menstruation n(%)	5(71.4)	6(14)	-
Severe Acne: Yes n(%)	2(28.6)	20(46.5)	0.22
Severe Acne: No n(%)	5(71.4)	23(53.5)	-

Table I: Clinical characteristics defined by ovarian morphology.

Data are shown as means ± Standard deviation. N = Number BMI = Body mass index. PCOM = Polycystic ovarian morphology.

	PCOM (-)	PCOM (+)	P-Value
Right ovary volume (m ³)	8.57± 1.98	14.43±5.8	0.01
Left ovary volume (m ³)	8.14±2.26	14.66±5.8	0.006
FNO right side (n)	9.14 ±2.85	15.6±4.2	0.001
FNO left side (n)	10.86±3.97	16.02±4.27	0.004
Endometrial thickness (mm)	5.92±2.1	7.03±2.21	0.22
Uterus length(mm)	63.29 ±9.63	66.72±9.97	0.40
Uterus depth (mm)	40.14 ±7.19	41.74± 7.44	0.59
Uterus thickness (mm)	28.43± 5.88	29.60±6.54	0.65

Table 2: Ultrasound characteristics of ovaries and uterus based on ovarian morphology.

Data are shown as means ± Standard deviation. FNO = Follicle number per ovary. PCOM = Polycystic ovarian morphology.

	PCOM(-)	PCOM(+)	P-Value
Testosterone total (ng/dL)	0.88±0.67	1.18±0.67	0.27
Testosterone-free (ng/dL)	0.96±0.73	1.11±0.54	0.52
TSH (µU/mL)	1.86±0.69	1.66±1	0.61
Prolactin (ng/mL)	16.22±6.41	16.61±5.47	0.47

Table III: Hormonal profiles defined by ovarian morphology.

Data are shown as means ± Standard deviation. TSH = Thyroid stimulating hormone. PCOM = Polycystic ovarian morphology.

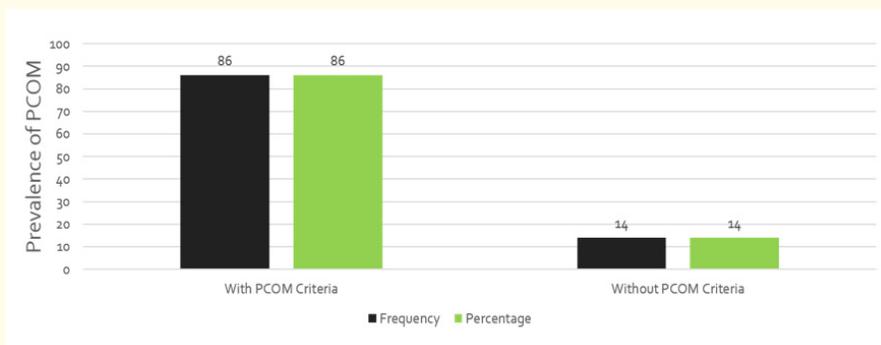


Figure 1: Prevalence of PCOM in adolescent girls. PCOM= polycystic ovarian morphology.

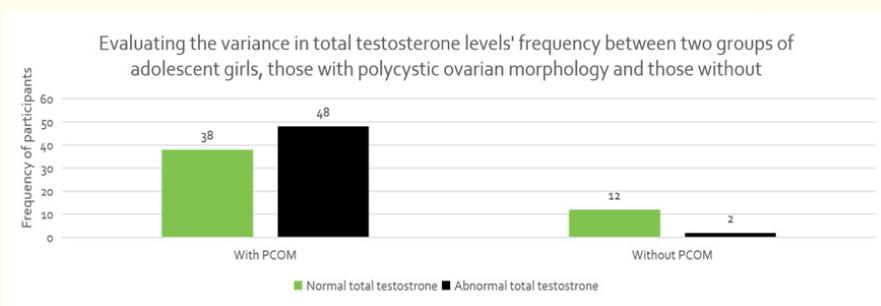


Figure 2: Evaluating the variance in total testosterone levels' frequency between two groups of adolescent girls, those with PCOM and those without. PCOM= polycystic ovarian morphology.

Discussion

We define the prevalence of polycystic ovarian morphology in fifty 9 to 19-year-old adolescent girls presented by gynecologists to the Firouzabadi Hospital between 2022 and 2023. Considering PCOM is a common condition in these girls that is unrelated to Hyperandrogenism or insulin resistance, this ultrasonography pattern may represent a physiological finding in these girls [13].

The prevalence of PCOM among adolescent girls has not been investigated in Iran, and to the best of our knowledge, this study is the first to evaluate this prevalence among Iranian adolescent girls. There is still controversy among various guidelines regarding the diagnostic criteria of PCOM [18]. According to the Rotterdam criteria, polycystic ovaries are characterized by the presence of 12 or more follicles measuring 2 to 9 mm in diameter, or by an ovarian volume exceeding 10 cm³ [19,20]. Polycystic ovarian morphology

is common in adolescent girls, with a prevalence of 30% to 40% [17], often due to hormonal changes associated with puberty [21]. Therefore, ultrasonography should not be used to diagnose PCOS in individuals less than eight years' post-menarche [21]. In our study, we used Rotterdam criteria for diagnosis of the PCOM, and the mean ± SD right and left ovary volume were 13.61 ± 5.81 and 13.75 ± 5.93 respectively. The mean ± SD follicle numbers per ovary of right-sided and left-sided were 14.70 ± 4.63 and 15.30 ± 4.57 respectively. A study by Villarreal et al. showed that girls with PCOM had more follicles with a diameter of 2-5 mm (P = 0.0001) than those without. However, there was no difference in the number of follicles with a diameter of 6-9 mm (P = 0.295) [13].

Elevated testosterone levels, typically two standard deviations above the mean, are commonly used as a hormonal marker

to diagnose Polycystic Ovary Syndrome (PCOS) based on the assay method employed [22]. Most diagnostic criteria for Hyperandrogenism include hirsutism, acne, and androgenic alopecia as indicators [8,20]. Determining biochemical Hyperandrogenism in adolescents is challenging as testosterone levels rise following puberty [23]. The question of whether mild hyperandrogenemia is a typical peri-menstrual condition or if adolescent hyperandrogenemia persists into adulthood remains uncertain [23]. In our current study, we employed a combination of clinical and hormonal profiles for identifying Hyperandrogenism among adolescent girls involving hirsutism, severe acne, and increased total testosterone. According to one study, the raised AMH and reduced FSH levels reported in healthy girls with regular menstrual cycles and PCOM indicate that this ovarian pattern is caused by a greater number of 2-5 mm follicles. Elevated AMH levels can indicate PCOM during adolescence [13]. Adolescents may struggle with ovarian dysfunction, as many experience irregular menstrual cycles at first. True ovarian dysfunction is marked by irregular menstrual cycles that are either shorter than 21 days or longer than 35 days. Menstrual cycles in adolescents are comparable to those in adults, with 75% of cycles ranging from 21 to 45 days in the first year, and 95% stabilizing within that range within 5 years [24,25]. In our study, irregular menstruation was frequent in participants with PCOM, although the differences were not statistically significant. This could be attributed to our research groups' shorter than average 5-year gap from menarche.

Chen et al. conducted a study on the role of ovarian volume in diagnosing polycystic ovary syndrome among Chinese adolescents. The study assessed 69 adolescents with PCOS and 26 control group adolescents using transvaginal ultrasound. It was observed that both the average and maximum ovarian volumes were higher in the PCOS group compared to the control group [26]. Our findings are consistent with the previous study, since the volume of both right and left ovaries in the PCOM group was greater than in the non-affected group.

Tehrani et al. did a review study on the treatment and diagnostic problems of PCOS in adolescent girls, addressing symptoms such as Hyperandrogenism, moderate to severe acne, and hir-

sutism. The study found that the increase in ovarian volume between the commencement of menarche and the age of 16 had not been addressed. It proposed a 12cm^2 criteria for measuring ovarian volume in adolescents [27]. Other studies have demonstrated that adolescents with PCOS exhibit a positive correlation between androgen levels and ovarian characteristics such as area, volume, and follicle count per cross-section [28]. Our findings revealed that PCOM participants had a higher prevalence of hirsutism and abnormal testosterone levels than non-PCOM subjects.

Strengths and limitations

The current study has notable strengths that merit recognition. The foremost strength is our examination of 50 adolescent girls to determine the prevalence of PCOM, as no prior studies have reported on the prevalence of PCOM among Iranian adolescent girls. Secondly, we assess the clinical and hormonal profiles for Hyperandrogenism within our study populations and evaluate the relationship between PCOM and Hyperandrogenism in adolescent girls.

On the other hand, several limitations should be mentioned. We could not perform vaginal ultrasonography on our population study groups since this is against the rule in these adolescent girls before their marriage so the sensitivity and specificity of diagnostic criteria of PCOM had been significantly reduced. The AE-PCOS Society revised the PCOM criteria to include a minimum of 25 follicles in adults [29]. However, the implications of this change for adolescents, who frequently undergo transabdominal ultrasounds and typically exhibit a larger median ovarian size and higher follicle count per section, remain unclear [25]. The results of our study, which observed increased ovarian volume and a higher number of follicles in participants with PCOM, may be attributed to normal physiological changes during puberty. Therefore, it is essential to conduct future research to assess the diagnostic criteria for PCOM in adults compared to adolescents.

Conclusion

In summary, our findings indicate that an increased ovarian volume ($\geq 10\text{ cm}^2$) and a higher number of follicles (≥ 12) are observed in Iranian adolescents with PCOM; however, these expected impacts were not observed in non-PCOM Iranian individuals. Re-

garding Hyperandrogenism criteria, our data showed that participants with PCOM had higher hirsutism scores and abnormal total testosterone levels compared to non-PCOM subjects.

Ethics Approval

The ethics committee of the Iran University of Medical Sciences, Tehran, Iran approved the research protocol. (IR.IUMS.FMD.REC.1401.670).

Availability of Data and Materials

The data sets used and/or analyzed during the current study are available from the corresponding author upon reasonable request.

Competing Interests

The authors have no financial involvement with any organization or any financial interest in or financial conflict with the subject or materials discussed in the manuscript. This manuscript has no relevant financial or other relationships to disclose. Peer reviewers of this manuscript have no relevant financial or other relationships to disclose.

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Declaration

All methods were carried out following relevant guidelines and regulations. This study uses data from patients referred to the Firouzabadi Hospital in Tehran, and informed consent was obtained from all subjects and/or their legal guardian(s).

Authors' Contributions

YSH and RSH raised the presented idea and designed the study. YSH conducted the analyses. YSH and RSH interpreted the results. YSH, RSH, and FF developed the first draft of the manuscript. RSH and FF critically reviewed the manuscript. All authors have read and approved the final manuscript.

Consent of Publication

N/A.

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