The prevalence of phenotypic subgroups in Greek women with polycystic ovarian syndrome

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Summary

Background: Since 2003, when the American Society for Reproductive Medicine (ASRM) and European Society of Human Reproduction and Embryology (ESHRE) sponsored consensus established criteria for polycystic ovarian syndrome (PCOS) diagnosis, the phenotypic spectrum of the syndrome has been significantly broadened. *Purpose of the study:* This survey makes an effort to distinguish PCOS according to phenotypic expression and to estimate its prevalence in a Greek population. *Materials and Methods:* Greek women from 18 to 35 years of age, who visited the outpatient department, claiming either irregular menstruation (oligo- or anovulation, OA) or clinical manifestations of hyperandrogenemia (HA) were recruited. They gave full disease history and underwent clinical examination, including transvaginal ultrasound (TVUS) scan to identify PCO morphology. Blood samples were collected to perform hormonal and metabolic analyses. Acute or chronic disorders were excluded. Finally, 266 PCOS women constituted the study population. *Conclusions:* The full-blown phenotype (HA+OA+PCO) is the predominant phenotype in this Greek population.

Key words: Polycystic ovary syndrome; Phenotype; Prevalence.

Introduction

Polycystic ovary syndrome or PCOS, is the most common endocrine disorder among women of reproductive age. Criteria for the diagnosis of the particular disorder have been proposed by two different organizations. The initial criteria developed in 1990 by the National Institutes of Health (NIH) conference [1] included 1) oligo- or anovulation (OA) to the exclusion of other disorders and 2) clinical and/or biochemical signs of hyperandrogenemia (HA). Another set of diagnostic criteria were proposed by the American Society for Reproductive Medicine and European Society of Human Reproduction and Embryology (ASRM/ESHRE) sponsored consensus [2] held in 2003 in Rotterdam according to which PCOS is diagnosed when two out of the following three characteristics are present: 1) OA, 2) clinical and/or biochemical signs of HA, and 3) polycystic ovaries as evidenced on ultrasound examination.

When the aforementioned second group of criteria is applied, it yields the following subgroups [3]: A) OA+HA+PCO, that is, the full-blown PCOS phenotype, B) OA+HA, with normal ovarian morphology, C) HA+PCO, with regular ovulation and menstruation, also called "ovulatory PCOS", and D) OA+PCO, without hyperandrogenemia.

The aim of this study was to estimate the prevalence of the aforementioned phenotypes in a large sample of Greek reproductive women.

Materials and Methods

The present study was conducted from September 2005 to September 2009 in the Third Department of Obstetrics and Gynecology of Attikon University Hospital. The study protocol was in accordance with both Greek and European Union Legislations and was approved by the Hospital Ethics Committee. All patients gave informed consent.

Patients

All subjects were recruited from the Gynecological Endocrinology Ambulatory Clinic. The sample consisted of Greek Caucasian women (age range: 18 to 35 years), complaining of irregular menstruation or clinical signs of HA. Patients with positive pregnancy test, personal history of acute or chronic disease, and following treatment with compounds affecting sex hormones (oral contraceptives) within the past six months, were excluded from the study.

Study design

Disease history: A detailed questionnaire addressing subjects' menstrual cycle characteristics (age of first menstrual cycle, frequency of menstruation, qualitative, and quantitative characteristics of menses) was completed by all study participants. Chronic anovulation was defined in the questionnaire as having fewer than eight menstrual cycles per year.

Lifestyle variables among others considered in the present study included: alcohol and tobacco use, extensive physical exercise, and use of hormonal treatment. Also recorded were participants' family history of diabetes mellitus, hypertension or cardiovascular disease, and the presence of any first-degree relatives exhibiting irregular menses.

Clinical examination

Each woman in the present study underwent a physical examination conducted by two gynecologists with experience in reproductive disorders. Data were collected concerning women's waist and hip circumference, body weight, height, and blood pressure, while body mass index (BMI-weight in kgs divided by the square of height in m^2) and waist-to-hip ratio were also calculated. The amount of excess terminal hair growth was assessed by using the Ferriman-Gallwey scale (FG), based on whole body overview, with patients scoring 8 or higher considered as hirsute [4]. Finally, the presence of acne vulgaris, androgenetic alopecia or acanthosis nigricans, and a cutaneous sign of hyperinsulinemia, were also recorded, however with no particular scoring technique applied.

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Table 1. — *Prevalence of PCOS phenotypes in different study populations*.

	Nationality	A	B	C	D
Hsu *et al*. (2007) [6]	Taiwan	51.8	8.8	21.2	18.2
Barber *et al*. (2007) [7]	U.K.	61.8	0	24.6	13.6
Dewailly *et al*. (2006) [8]	France	60.6	6.7	16.5	16.3
Pehlivanov *et al*. (2007) [9]	Bulgary	58.6	11.4	20	10
Chae *et al*. (2008) [10]	Korea	52.4	13.9	2.4	31.3
Diamanti-Kandarakis *et al*. (2007) [11]	Greece	45.5	40.2	7.4	6.9
Shroff *et al*. (2007) [12]	U.S.A.	58.1	14.3	13.2	14.3
Belosi *et al*. (2006) [13]	Italy	73.6	7.5	5.5	13.3
Welt *et al*. (2006) [14]	U.S.A-Iceland	71.3	1.7	18.4	8.6
The present study	Greece	44.4	18	26.3	11.3

A: HA+OA+PCO, B: HA+OA, C: HA+PCO, D: OA+PCO; HA: Hyperandrogenemia; OA: Oligo-anovulation, PCO:polycystic ovarian morphology (by U/S).

Transvaginal ultrasound (TVUS) scans

Three-dimensional ovarian morphology and size was recorded on the sixth to eighth days of patients' menstrual cycle by using the same operator [5]. The presence of ≥ 12 follicles with a diameter of two to nine mm or increased ovarian volume (> 10 cm^3) established a sonographic diagnosis of PCO.

Biochemical measurements

Venous blood samples were drawn from subjects early in the morning following an overnight fast between the third and sixth day after the onset of a spontaneous or progesterone-induced menstruation.

Blood samples were centrifuged, and serum was drawn off and frozen at -70°C until analyzed. In order to rule out abnormal thyroid function, hyperprolactinemia and Cushing syndrome, thyroid tests (FT3/FT4/TSH) were performed and levels of prolactin (PRL) and cortisone were estimated, respectively, as part of the differential diagnosis work-up. The 17-α-OH-progesterone (17-OHP) was also measured. For women who exhibited plasma levels higher than 1.5 ng/ml, a Synacthen test using tetracosactide was conducted (Novartis Pharma S.A.) enabling to exclude patients with congenital adrenal hyperplasia from the study.

Ultimately, participants enrolled in the study were 266 women.

The parameters below were also assessed in the sample, providing thus a basis for conducting comparisons between different phenotypic groups:

– follicle-stimulating hormone (FSH), luteinizing hormone (LH), and estradiol (E2);

– total testosterone, free testosterone, Δ4-androstenedione (Δ4-A), and dehydroepiandrosterone-sulfate (DHEA-S);

– sex-hormone binding globulin (SHBG).

Table 2. — *Age, BMI, and BMI categories of PCOS subgroups*.

		Phenotype A		Phenotype B		Phenotype C		Phenotype D		P-Pearson's
		N	%	N	%	N	%	N	%	χ^2 test
AGE, mean ± SD, median		25 ± 6	24 (21 - 29)	25 ± 6	24 (21 - 28)	26 ± 6	26 (22 - 29)	24 ± 5	24 (20 - 27)	0.442
BMI, mean ± SD, median		27.1 ± 7.7	24 (21.1 - 32.7)	25.5 ± 6.9	23 (19.6 - 33)	23.7 ± 5	23 (20 - 25.2)	23.3 ± 5.6	21.3 (19 - 24)	0.009
BMI, categories	Normal	62	52.5	27	58.7	50	72.5	23	76.7	0.009
	Overweight	18	15.3	7	15.2	13	18.8	3	10.0	
	Obese	38	32.2	12	26.1	6	8.7	4	13.3	

Table 3. — *Hormonal profile of PCOS phenotypes*.

	Phenotypes								
	A		B		C		D		P-Kruskal-Wallis test
	Mean ± SD	%	Mean ± SD	%	Mean ± SD	%	Mean ± SD	%	
FSH (m/U/ml)	5.7 ± 1.9	5.4 (4.2 - 6.7)	6 ± 2.8	5.9 (4.4 - 6.5)	6.2 ± 2.3	5.7 (4.6 - 7.5)	6.5 ± 2.3	5.6 (5.1 - 7.2)	0.328
LH (m/U/ml)	7 ± 5	5.8 (3.9 - 8.1)	6.5 ± 3	6 (4.4 - 7.8)	5 ± 2.1	4.7 (3.5 - 6.4)	6.1 ± 2.9	5.3 (4 - 8)	0.015
PRL	17.1 ± 9.5	14.9 (10.3 - 21.1)	15.9 ± 12.1	12.4 (9.5 - 18.4)	22.5 ± 29.6	18 (12.3 - 22.7)	17.1 ± 8.6	15.6 (11 - 20.5)	0.063
E2 (pg/ml)	54.3 ± 64.9	39.9 (31 - 51)	45.8 ± 29.5	37.3 (32 - 57)	61.2 ± 89.1	40.8 (32.9 - 54.1)	48.3 ± 35.8	39.6 (29.5 - 49.5)	0.625
Total testosterone (ng/dl)	59.9 ± 25.2	60 (41 - 75)	62.7 ± 23.6	63.7 (44 - 78)	62.1 ± 25.6	63.3 (50 - 73)	37.8 ± 12.2	37 (30 - 47)	< 0.001
Free testosterone (pg/ml)	2.3 ± 1.6	2 (1.2 - 2.8)	2 ± 0.9	2.1 (1.2 - 2.7)	2.1 ± 1.4	1.8 (1.5 - 2.4)	1.6 ± 1	1.6 (0.8 - 2)	0.106
OHP17 (ng/ml)	1.2 ± 0.6	1.1 (0.8 - 1.5)	1.3 ± 0.8	1.2 (0.7 - 1.7)	1.5 ± 1	1.2 (0.7 - 1.9)	0.8 ± 0.5	0.8 (0.5 - 1)	0.023
DHEA-S (μg/dl)	255.2 ± 206.1	214.5 (152.6 - 316.5)	224.3 ± 122.6	206.5 (136 - 303.5)	226.7 ± 114.1	194 (130 - 340.3)	181.7 ± 80.8	192.5 (103.8 - 244.4)	0.234
Δ4 Androstenedione (nmol/l)	2.8 ± 1.4	2.6 (1.9 - 3.4)	2.7 ± 1.3	2.3 (1.9 - 3.6)	2.6 ± 1.3	2.4 (1.7 - 3.3)	2 ± 0.7	2.2 (1.6 - 2.6)	0.086
SHBG (nmol/l)	42.5 ± 23.1	38 (25 - 58.8)	47.2 ± 25.3	38 (29.5 - 58)	52.2 ± 29.6	47 (32.6 - 69)	73.4 ± 72.4	48 (33 - 82)	0.046
Cortizole (mg/dl)	18.3 ± 7.6	17 (13 - 22.1)	18.4 ± 6.2	19.5 (13.6 - 22.5)	16.4 ± 7.7	17 (10 - 22.4)	17.8 ± 8	20.5 (12.6 - 21.8)	0.687
T3 (nmol/l)	1.5 ± 0.5	1.4 (1.1 - 1.9)	1.5 ± 0.4	1.4 (1.2 - 1.9)	1.5 ± 0.5	1.3 (1.2 - 1.9)	1.7 ± 0.5	1.7 (1.1 - 2.1)	0.832
T4 (μg/dl)	7.8 ± 1.6	7.7 (7 - 8.8)	7.6 ± 1.8	7.8 (6.8 - 8.7)	7.6 ± 1.7	7.8 (6.9 - 8.3)	7.8 ± 1.6	7.6 (6.5 - 9)	0.977
TSH (mU/l)	2.2 ± 1.2	2.1 (1.6 - 2.6)	2.4 ± 2.2	1.7 (1.2 - 2.9)	1.9 ± 1.5	1.6 (1.2 - 2.4)	3.2 ± 1.9	2.3 (1.7 - 4.4)	0.006

HA was defined as serum total testosterone or Δ4-androstenedione level higher than two nmol/l and 10.5 nmol/l, respectively.

Results

The final sample consisted of 266 women with a mean age of 25 years (± 5.6 years). The prevalence of the four different subgroups is shown in Table 1.

The predominant phenotype in this sample was the full-blown phenotype A with a prevalence estimated at 44.4%. Phenotype B represented 18% of the sample, while phenotype C at 26.3%. Phenotype D was the most rare phenotype in comparison with the three others, having a prevalence of 11.3%.

BMI rates differed significantly among the four subgroups. More specifically, the use of Bonferroni adjustment for the level of statistical significance revealed a statistically significant higher BMI for women of phenotype A in comparison with those with phenotype D ($p = 0.009$). In addition, women of normal weight were significantly more in phenotype C compared with those of phenotype A ($p = 0.007$) (Table 2).

Discussion

The prevalence of the different PCOS phenotypes vary according to the different study populations as shown in Table 1 [6-14]. Since these studies have involved women of different ethnicities, ethnic background may also be considered as an important confounding factor.

According to the present results, 44.4% of Greek PCOS women belong to phenotype A, 18% to phenotype B, 26.3% to phenotype C, and 11.3% to phenotype D. With specific reference to the latter phenotype, there is varying evidence in the relevant literature as to whether and to what extent can a woman without HA be diagnosed with PCOS? Other studies have illustrated that women categorized in this group share the same clinical and metabolic characteristics as those of the general population and do not require any treatment in order to modify their hormonal profile [8, 15-18].

This fact has led the Androgen Excess Society (AES) to the proposal of different criteria for the diagnosis of PCOS, according to which the diagnosis of the syndrome is set when a woman fulfills the following two diagnostic criteria: 1) HA, clinical or biochemical and 2) OA as shown by menstrual disorders or polycystic ovarian morphology. Although AES includes PCO as a characteristic of the syndrome, it is not considered as an autonomous criterion [19]. The application of AES criteria leads to the formation of three different subgroups, that is, the same with those of Rotterdam criteria with the exception of phenotype D.

There is a difference in the present results concerning prevalence compared to those of another study of a Greek population conducted by Diamanti-Kandarakis *et al*. [11]. This deviation may be attributed to differences in the study protocol. For the definition of biochemical HA, the authors employed not only the total testosterone levels above the 95th percentile of levels detected in normally-menstruating women, as the aforementioned study did, but also Δ4-androstenedione values greater than 10.5 nmol/l [8]. The latter fact highlights the need for adopting commonly-accepted definitions and standards, primarily for HA, hirsutism, and anovulation.

The sub-classification of PCOS into different subgroups seems clinically significant. According to the literature, not all phenotypes share the same metabolic and hormonal profile (Table 3), indicating that not all PCOS patients need the same therapy or intervention. Clinicians should be aware of the phenotypic expression of the syndrome in order to treat a patient individually and also recognize possible future cardiometabolic risks.

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