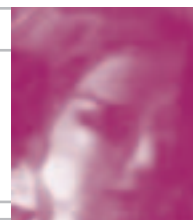


Diagnostic features of polycystic ovary syndrome



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Polycystic ovary syndrome (PCOS) is one of the commonest endocrinopathies in women.¹ It was first described by Stein and Leventhal² in 1935, as the association of infertility, obesity, hirsutism and bilateral enlarged polycystic ovaries. As a syndrome, PCOS has had an interesting history, with much debate and often poor consensus regarding its diagnostic criteria. A variety of histological, biochemical and sonographic features have been described, but until recently there has been no general agreement on definition.

Definition – the diagnostic debate

The National Institutes of Health (NIH) in Bethesda, Maryland, USA held their first international consensus conference on PCOS in April 1990, which ironically made it obvious that there was no true consensus.³ A clinical and working definition emerged from the USA following the NIH conference. This suggested that the diagnosis of PCOS be based on chronic anovulation with biochemical evidence of hyperandrogenism after the exclusion of other causes such as hyperprolactinaemia and non-classic congenital adrenal hyperplasia (NCAH).^{3,4} Ovarian morphology on sonar was not regarded as part of the criteria, making the diagnosis dependent on clinical and biochemical findings.

On the other hand, the predominantly European working definition of PCOS⁵ comprises sonographically diagnosed polycystic ovary morphology, usually using the ultrasound criteria of Adams *et al.*,⁶ associated with oligomenorrhoea or amenorrhoea and/or signs of hyperandrogenaemia.

Rotterdam ESHRE/ASRM-sponsored PCOS consensus workshop

In May 2003 the Rotterdam consensus workshop on PCOS took place, sponsored by the European Society for

Human Reproduction and Embryology (ESHRE) and the American Society for Reproductive Medicine (ASRM). The workshop was attended by well-published authors from both sides of the Atlantic. A consensus statement⁷ was released defining the diagnostic criteria, and proved to be detailed and inclusive. The report was based on clinical trial evidence rather than majority opinion, as had been the NIH classification in 1990.

In essence, there are 3 major criteria, with 2 out of 3 required for diagnosis: (i) oligo- or anovulation; (ii) clinical and/or biochemical signs of hyperandrogenism (with the exclusion of other aetiologies); and (iii) the presence of polycystic ovaries on ultrasound.

They further acknowledged the obvious problems with these criteria as regards trial protocol and data recording. For example, where pregnancy is the trial outcome, the inclusion criteria of anovulation is clearly of significance. However, where clinical improvement of hirsutism is the outcome, less emphasis need be placed on ovulatory function.

The statement contains a detailed discussion of the term 'hyperandrogenism', both clinically and biochemically, with specific reference to the limitations of laboratory measurement of circulating androgens, plus comment that such evidence is not required as proof of clinical hyperandrogenism.

Only with hindsight will it become known whether this carefully researched and constructed document will be used as a general reference in scientific research. Interestingly, Adam Balen from the UK, who participated in the consensus workshop on the revised definitions of ultrasound assessment, also-co-authored an article on the clinical overview on PCOS.⁸ He defined PCOS as a sonographic finding of polycystic ovaries plus either oligo- or amenorrhoea, obesity or hyperandrogenism.

Another prominent figure on the scientific committee of the workshop, Ricardo Azziz from the USA, also published a prevalence study in June 2004,⁹ using the NIH inclusion criteria for his definition. Both these

examples emphasise the problems with adopting a new definition in a scientific field.

Prevalence

Assessment of PCOS prevalence rates is fraught with problems. Data are often difficult to compare from one study to another because of inconsistency in standardisation of diagnostic criteria, making meta-analyses difficult to perform. The inadequacies of the NIH and European systems of classification have become obvious, both in the interpretation of data and in the diagnosis of PCOS.

We know that the finding of polycystic ovaries alone does not necessarily indicate the presence of the syndrome.¹⁰ Prevalence studies for these sonographic ovarian findings place the incidence in the order of 17 - 22%, figures that seem remarkably constant worldwide.¹¹⁻¹⁴ In Polson's 1988 study of 257 women only 7% of the eumenorrhoeic women had polycystic ovaries.¹¹ In contrast, 86% of irregularly cycling women had PCOs. Transvaginal ultrasound places this figure somewhat higher at 21 - 28%, and it appears that younger women have a higher incidence of polycystic ovaries than women over 35 years of age.¹³ Many of the subjects recruited in the Polson study did in fact have clinical problems, although they had not sought medical attention for them, demonstrating the difficulty with performing such studies in a 'normal' population group.

Prevalence rates of 3 - 11% for PCOS are reported, depending on the criteria used for definition.¹³ A recently published USA study by Azziz *et al.*⁹ on 347 women undergoing pre-employment medical examinations, found the prevalence of PCOS to be 6.6% using modified NIH criteria of oligo-ovulation rather than amenorrhoea. It also emerged that 86% of women presenting with both menstrual dysfunction and hirsutism had PCOS, whereas only 8% with menstrual dysfunction alone had PCOS. In this study, prevalence rates in black and white subjects were not significantly different.

This correlates with a study¹ describing 173 anovulatory women attending a reproductive endocrine clinic, of whom 87% had polycystic ovaries; more than 60% were hirsute, and 93% had biochemical evidence of raised androgens and/or luteinising hormone (LH).

A problem with both the NIH and European definitions arises where the patient clearly has the syndrome but does not comply with the criteria. For example, a woman with PCO and hyperandrogenism who is ovulatory would by NIH criteria not be diagnosed as PCOS, while an anovulatory woman with hyperandrogenism but sonographically normal ovaries would not fit the diagnosis using the European criteria.

Clinical presentation

As the commonest endocrinopathy and reproductive disorder in women, it is essential that we be aware of

PCOS and note the signs to ensure timely diagnosis. Such women present clinically primarily with menstrual irregularity, androgen excess (hirsutism), acne, androgen-dependent alopecia and infertility.⁴

The first of these clinical features is menstrual irregularity, which is secondary to ovulatory dysfunction. This may be defined by a history of 8 or fewer menstrual cycles in a year, or menstrual cycles that are shorter than 26 or longer than 35 days. Alternatively, a biochemical diagnosis can be made where cycle length is 26 - 35 days and a mid-luteal (day 22 - 24) progesterone level less than 4 ng/ml confirms anovulation.⁹

Over the last decade we have become more aware of the increasing prevalence of metabolic problems associated with PCOS,¹⁵ the so-called metabolic syndrome. These women are frequently obese, with increased risk of hyperinsulinaemia, impaired glucose tolerance (IGT), and even frank diabetes. An association with hypertension and dyslipidaemia is also well described.¹⁶ The consequent cardiovascular risk implications make clinical detection of PCOS and further identification of its metabolic sequelae a very relevant health issue. In fact, the ESHRE/ASRM 2003 statement includes a consensus guideline regarding indications for screening for metabolic disorders in PCOS (Table I). Chronic anovulation also implies unopposed oestrogen, and the consequent increased risk of endometrial carcinoma is well recognised.

Azziz¹⁷ discusses an approach to screening of the hirsute woman in clinical practice from a cost-effective perspective. In his guideline he suggests that all hirsute women first be screened for ovulation, even those claiming to be eumenorrhoeic, as in fact 40% of these are oligo-ovulatory. He further recommends that oligo-ovulatory hirsute women be screened using thyroid-stimulating hormone (TSH) levels (for coincident ovulatory dysfunction) and via 17-hydroxyprogesterone (to exclude NCAH). He recommends that routine gonadotrophin testing not be done, since only 50 - 60% of PCOS subjects have an elevated LH/follicle-stimulating hormone (FSH) ratio, and although this may confirm what is suspected, it is often erroneously used to exclude the diagnosis. Screening must be done for diabetes, as 30% of PCOS subjects have IGT and 8%

| Table I. Criteria for the metabolic syndrome in women with PCOS (3 of 5 required for diagnosis of the syndrome) * | |
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| Risk factors | Cut off |
| 1. Abdominal obesity (waist circumference) | > 88 cm |
| 2. Two triglycerides | ≥ 150 mg/dl |
| 3. HDL cholesterol | < 50 mg/dl |
| 4. Blood pressure | ≥ 130/≥ 85 mmHg |
| 5. Fasting and 2-hour glucose from oral GTT | 110 - 126 mg/dl and/or glucose 140 - 199 mg/dl |

HDL = high-density lipoprotein; GTT = glucose tolerance test.
* 2003 Rotterdam PCOS consensus.⁷

have frank type 2 diabetes. Routine sonography of the hirsute patient is not considered mandatory,¹⁷ although of course it stands to reason that where there are other suggestive symptoms of PCOS, ultrasound should form part of the diagnostic workup.¹⁶

Obesity is an important association with PCOS. We know that response to treatment is reduced with increased body mass index (BMI). Weight loss itself may be associated with attenuation of symptoms and reduction of circulating androgens and insulin, and even spontaneous ovulation. However, weight loss has no effect on gonadotrophin secretion.¹⁰

Obese patients may reveal the presence of a cutaneous indicator of hyperinsulinaemia called acanthosis nigricans, an association described in 1980 as the 'HAIR-AN' syndrome (hyperandrogenism, insulin resistance and acanthosis nigricans).⁴

An interesting study¹⁸ assessed the effectiveness of interviewing as a means of predicting PCOS as a less cost-limiting and time-saving approach. The questionnaire centred on androgenic symptoms and was given to patients, their mothers and sisters. The authors found that interviewing via questionnaire was a highly sensitive and specific technique for screening for PCOS, and moreover predicted a high portion of affected family members.

There appears to be a genetic basis for PCOS as evidenced by this familial concordance, with 24% of mothers and 32% of sisters being affected.¹⁸ It appears to have an autosomal-dominant mode of inheritance, with premature balding in men as the putative male phenotype. Genetic linkage with insulin resistance and obesity has been reported via the common allelic variation at the variable number of tandem repeats (VNTR) locus in the promoter region of the insulin gene. Anovulatory hyperinsulinaemic women are more likely to have inherited this class III/III allele, particularly from their fathers. Associations with both low and high birth weights have been reported reflecting the heterogeneity of the condition.¹⁶

Ultrasonography/imaging

The most widely accepted sonographic criteria for polycystic ovary were described by Adams *et al.*⁶ in 1985. The polycystic ovary was defined as the presence, in one plane, of multiple cysts, 2 - 18 mm in diameter, distributed evenly around the ovarian periphery, with an increase in ovarian stroma. The Adams criteria have been adopted by many subsequent studies following this seminal paper.

In 1985 Adams had only transabdominal sonar at his disposal. The advent of transvaginal ultrasound with its greater resolution has today largely superseded the transabdominal approach.¹⁶ The transvaginal approach with modern high-frequency (> 6 MHz) probes provides a more accurate view, and especially in obese patients

avoids the homogeneous appearance of ovaries that may erroneously be found on transabdominal scan.

A paper by Balen *et al.*¹⁶ first presented at the ESHRE/ASRM workshop in 2003, provides a comprehensive view on the current approach to polycystic ovary imaging. It provides a critical discussion on the ultrasonic modalities available today, and defines the criteria for making the diagnosis in women on oral contraceptives and in the menopause.

The revised sonographic criteria of Balen *et al.*¹⁶ define polycystic ovary as the finding of either: (i) 12 or more follicles measuring 2 - 9 mm diameter; or (ii) increased ovarian volume (> 10 cm³).

The presence of a single polycystic ovary is sufficient for diagnosis. The distribution of follicles and the quantification of ovarian stroma are no longer essential to make the diagnosis.

The innovative techniques of 3-D ultrasound and magnetic resonance imaging (MRI) may provide even more sensitive means of detecting the polycystic ovary. The 3-D sonar is limited by the greater cost, training and data analysis it requires. However, excellent correlation between 2-D and 3-D measurements for ovarian volume and morphology were reported at the ESHRE/ASRM workshop.¹⁶

As a diagnostic tool MRI provides superb ovarian imaging, and as such would probably increase the detection rates of abnormal ovarian morphology dramatically, but the cost limits its applicability.¹⁶ Transvaginal colour Doppler has demonstrated that polycystic ovaries have increased blood flow and blood vessels of greater diameter than normal ovaries, in keeping with the well-described feature of ovarian enlargement.¹⁹ A study using dynamic contrast-enhanced (DCE) MRI has shown enhancement behaviour of the ovaries in PCOS women with these findings,²⁰ which may broaden diagnostic and treatment parameters. As a modality DCE-MR imaging has so far been used primarily in the field of breast cancer research, focusing on the assessment of angiogenesis. Increased concentrations of biochemical factors such as vascular endothelial growth factor (VEGF) have been found in polycystic ovaries. Coupled with the finding of increased follicular fluid, VEGF levels in patients with ovarian hyperstimulation syndrome (OHSS is the most serious iatrogenic complication of ovulation induction), DCE-MR imaging may be utilised in predicting OHSS.

Biochemical

The pathogenesis and pathophysiology of PCOS is still incompletely understood. What we do recognise as interrelated characteristics are insulin resistance (IR), hyperandrogenism and altered gonadotrophin dynamics.⁴ This association between PCOS and disordered carbohydrate metabolism was first noted by Achart and Thiers in 1921, as the 'diabetes of bearded

women'. By 1980, Burghen *et al.* demonstrated the association between PCOS and hyperinsulinaemia.^{3,16,21}

IR may be defined as a subnormal biological response to a given level of insulin. In 1989 Dunaif published a now classic study on the association between IR and PCOS, where the extent of IR cannot be explained by obesity alone.³ IR in obese PCOS women was greater than in obese normal subjects. Among non-obese women, those with PCOS had higher IR than the controls (Fig. 1).

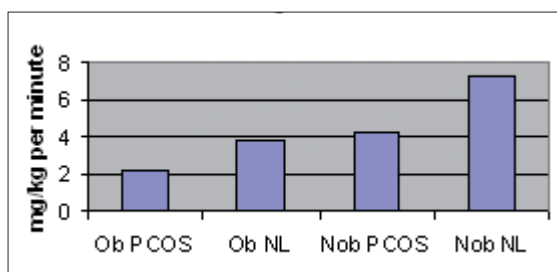


Fig. 1. Insulin sensitivity (inverse of insulin resistance) in obese (Ob) and non-obese (Nob) women with polycystic ovary syndrome (PCOS) and in cycling controls (NL). (Copyright 1989 American Diabetes Association (from Guzick⁴)).

Dunaif³ subsequently sought to demonstrate a causal relationship between insulin resistance and hyperandrogenaemia. Ovarian tissue sensitivity to hyperinsulinaemia appears to drive ovarian and adrenal androgen production, stimulating proliferation of the pilosebaceous unit and suppression of sex hormone binding globulin (SHBG), thereby further increasing the bioavailability of free testosterone. The directionality of this relationship is now accepted as probable,²² but not certain.⁸

We are aware that early detection and treatment of IR and its metabolic sequelae is likely to have far-reaching health benefits, but testing does not necessarily identify women who will respond to insulin sensitizers, nor does treatment usually normalise their endocrine function.²² The assessment of insulin resistance and a clear diagnostic strategy to define its parameters is still an area of debate.

The gold standard for testing IR is the euglycaemic insulinaemic clamp test, in which insulin is administered intravenously at a fixed dose while glucose is simultaneously infused at the rate required to maintain the glucose at a predetermined level. It is expensive, time-consuming and labour-intensive, and therefore inappropriate for an office setting.

Homeostatic measurements of fasting glucose/insulin ratios, such as the homeostatic model assessment (HOMA), and the quantitative insulin sensitivity check (QUICKI), are the most frequently used techniques.

These tests are simple and easy to carry out. The HOMA index is probably the most commonly used formula in our clinical setting, simply calculated by the product of fasting insulin (I_o) and fasting glucose (G_o), divided by a constant of 22.5. A level above 2.5 is generally accepted as consistent with IR.

$$HOMA = [I_o \text{ (uIU/ml)} \times G_o \text{ (mmol/l)}] / 22.5$$

Both these tests are considered to have a good correlation with the clamp technique, and may be used in normo- and hyperglycaemic patients,⁸ although a recent study²³ in Greece on PCOS women failed to demonstrate this correlation. The authors concluded that metabolic or hormonal factors particular to PCOS might have influenced this lack of correlation between their findings and those of other IR groups.

Putative markers of IR²² currently being researched are homocysteine,^{24,25} plasminogen activator inhibitor-1, adiponectin, endothelin-1, SHBG and insulin-like growth factor binding protein-1 (IGF-1). The value of obtaining relatively non-invasive, sensitive and specific serological markers for IR has much appeal. This area of research is of much current interest.

Endocrine

The endocrine hallmarks of PCOS are hyperandrogenaemia and to a lesser extent elevated secretion of the gonadotrophin, LH.¹⁴ Both obese and lean women have an increased 24-hour mean concentration of LH, with an increased pulse frequency and amplitude.⁴ This may suggest the presence of a hypothalamic defect in PCOS,³ but it is more widely accepted that the abnormalities of gonadotrophin release are in fact secondary to ovarian pathology and chronic anovulation, with the polycystic ovary itself central to the pathogenesis of the syndrome.¹⁰

Androgen production by the ovarian theca cells is LH-dependent. It would appear that the excess androgen production is subsequent to elevated LH levels, supported by the finding that suppression of LH by gonadotrophin releasing hormone analogues or oral contraceptives suppresses androgen levels.⁴

FSH concentrations are usually in the midfollicular range of eumenorrhoeic women, but lower than those in the early follicular phase.^{4,14} Whether this relative insufficiency plays a more direct causative role in anovulation is contentious as it has been postulated that threshold levels for initiation of ovulation may be inadequate. The finding that most women with PCOS respond to clomiphene citrate, which itself works by stimulating pituitary release of FSH, provides supporting evidence for this hypothesis.⁴

A characteristic finding is the increase of LH relative to FSH. Fifty to sixty per cent of subjects have an elevated LH/FSH ratio,¹⁷ with a ratio greater than 2:1 being commonly accepted as consistent with PCOS.¹⁴ Because

of the pulsatile nature of gonadotrophin release, a single blood assay may fail to detect this.³ Assessment of serum concentrations of gonadotrophins, and LH in particular, is limited by data reflecting divergent results with different assay kits on the same serum sample. Assay-related reference ranges may largely attenuate this problem.²⁴

Serum levels of testosterone, in particular the free testosterone index, are increased in PCOS, averaging 50 - 150% higher than normal.¹⁴ The clinical expression of this hyperandrogenism shows a wide spectrum, with well-documented racial differences. Anovulatory but non-hirsute women with PCOS have similar levels to hirsute women.¹⁴ Testosterone is bound to SHBG, the expression of which appears to be linked to BMI via the insulin mechanism. In women with PCOS, low SHBG levels have been found to correlate with insulin resistance,^{3,10,22} thereby increasing the unbound testosterone fraction with its resultant effects.

Androstenedione (A4) has also been reported as elevated in the PCOS,^{4,10} but the ESHRE/ASRM guidelines exclude it from routine testing in the assessment of hyperandrogenaemia owing to the paucity of data, while there may be evidence indicating elevated levels in NCAH. A small percentage of PCOS patients may exhibit elevated levels of dehydroepiandrosteronesulphate (DHEAS), although again here evidence for routine testing is lacking, according to the consensus statement. Nevertheless, DHEAS and A4 have so far been widely accepted as additional androgens that, like testosterone, may typically be elevated in PCOS, as reported by many investigators.⁹

Oestrogen levels in PCOS follow an acyclical pattern as a consequence of anovulatory cycles.¹⁰ Early and midfollicular levels are normal, but there is no pre-ovulatory or midluteal increase in oestrogen levels.¹⁴ With progesterone deficiency and increased peripheral conversion of androgens to oestrogen by adipose tissue, unopposed oestrogen results in menstrual dysfunction and irregular bleeding, with a long-term increased risk of endometrial carcinoma.⁴

Discussion

It is unclear whether PCOS represents a single disorder or a conglomeration of different disorders with similar clinical presentation. A clinical presentation or phenotype of PCOS may also reflect different aetiology or pathophysiological differences. According to the 1990 National Institute of Child Health and Human Development (NICHD) definition²⁶ PCOS may present as three phenotypes.

In a recent article, Chang *et al.*²⁷ hypothesised that the three clinical phenotypes of PCOS represent different forms of the same metabolic disorder. Three hundred and sixteen women diagnosed as having PCOS were evaluated.

The oligo-ovulation + hyperandrogenism + hirsutism phenotype was present in 48% of subjects, the oligo-ovulation + hyperandrogenism phenotype in 29%, and the oligo-ovulation + hirsutism phenotype in 23%. These three phenotypes did not differ in mean BMI, waist-to-hip ratio, racial composition, degree of oligo-ovulation, prevalence of acne, or family history of hyperandrogenic symptomatology. However, subjects demonstrating the oligo-ovulation + hyperandrogenism + hirsutism phenotype were the youngest and had the greatest degrees of hyperandrogenaemia, hyperinsulinaemia and beta-cell function. Patients with the oligo-ovulation + hirsutism phenotype were the oldest and had the mildest degrees of hyperandrogenaemia, hyperinsulinaemia and beta-cell function. Subjects with the oligo-ovulation + hyperandrogenism phenotype demonstrated intermediate degrees of hyperandrogenaemia and metabolic dysfunction. These data suggest that it is the degree to which the beta cell is able to compensate for the degree of insulin resistance, and not the degree of insulin resistance *per se*, that determines the severity of the phenotype.

The authors also concluded that the lower levels of hyperinsulinaemia are related to lower androgen levels and slightly less severe hirsutism, whereas the greater degrees of hyperinsulinaemia favour the development of hirsutism and frank hyperandrogenism.

Finally, it remains unclear whether the three clinical phenotypes of PCOS described represent a continuum within a single population or are the result of differences in underlying pathophysiological mechanisms, and whether the clinical phenotype predicts differences in the long-term risk of these patients for developing type 2 diabetes mellitus or cardiovascular disease.

The debate regarding diagnosis and what the PCOS encompasses is hopefully becoming clearer. With the revised 2003 guidelines, more accurate prevalence statistics ought to become available, thereby increasing awareness of a common problem that deserves a high index of suspicion in any clinical practice.

The health impact of PCOS is enormous, and with the burgeoning obesity epidemic worldwide, it is likely to increase.

It is of the utmost importance to adhere to current diagnostic guidelines. This will help us to gain information and conduct non-biased research that will hopefully provide some answers to this poorly understood condition.

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