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Polycystic ovary syndrome in adolescents

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Menstrual irregularity and evidence of hyperandrogenism are characteristic features of polycystic ovary syndrome (PCOS) in adolescents. Diagnosis of PCOS is challenging as clinical features cannot be differentiated from the events of normal development. The specific aetiology of PCOS is not known but it is a complex disease resulting from interplay of genetic susceptibility, intra-uterine, extra-uterine and environmental factors. Obesity and insulin resistance are common associations, because of which patients are at high risk for metabolic and cardiovascular diseases. Lifestyle modifications are recommended in all patients with pharmacological agents to control features of hyperandrogenism and menstrual disturbances. This chapter discusses the pathogenesis of PCOS and diagnosis of PCOS in adolescents and the difficulties in diagnosis. In brief the associated co-morbidities and management are discussed.

Introduction

Polycystic Ovary Syndrome (PCOS) affects 3.6–15% of women in the reproductive age group [1,2]. It is the commonest cause of hyperandrogenism and anovulatory infertility in this age group [3,4]. Community based prevalence studies of PCOS in adolescents are scarce. Menstrual irregularities in the form of oligomenorrhoea or amenorrhoea, clinical hyperandrogenism (HA) in the form of hirsutism, acne, androgenic alopecia and anovulatory infertility are the characteristic clinical features of PCOS in women [5]. Rarely, women may present with virilization [6].

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Prevalence rates of PCOS are variable as different diagnostic criteria have been used in different studies. Diagnostic criteria have evolved over time. The National Institute of Health 1990 consensus criteria were the strictest, defining PCOS as clinical or biochemical hyperandrogenism and oligo-anovulation in absence of other endocrinopathies [7]. The Rotterdam consensus in 2003 broadened the criteria to include the presence of any 2 of the following as diagnostic of PCOS 1) clinical or biochemical hyperandrogenism, 2) oligo-anovulation and 3) polycystic ovarian morphology on ultrasound (PCOM) [8]. According to the Androgen-Excess Society criteria, hyperandrogenism is mandatory with either oligo-anovulation or PCOM [9] to diagnose PCOS. The NIH evidence based workshop in 2012 endorsed the Rotterdam 2003 criteria for diagnosis of PCOS but to specify different phenotypes [10]. The recently published international evidence based guidelines have endorsed the Rotterdam criteria 2003 for diagnosis of PCOS in women [11].

PCOS is a heterogeneous disease, in terms of pathophysiology as well as severity of clinical consequences. Not all patients have all the possible manifestations or are exposed to the same degree of long term health risks. Women with phenotype A, who have hyperandrogenism and oligomenorrhoea, have the greatest risk while the non-hyperandrogenic PCOS the least [12,13].

Pathogenesis

The exact etiology of this condition is not clear still but insulin resistance (IR) and compensatory hyperinsulinism is central to its pathogenesis [14]. There is enough evidence to suggest that it is a multifactorial complex disease, resulting from interplay of genetic susceptibility, insulin resistance, pancreatic beta cell dysfunction, abnormal ovarian and adrenal steroidogenesis and steroid metabolism alterations, neuroendocrine influences, environmental factors, epigenetic mechanisms and abnormal adaptation to energy restriction or excess.

Hyperandrogenism

PCOS is primarily an ovarian disease with increased androgen production. Theca cells produce androgen under control of luteinizing hormone (LH) and various intracrine factors. P450c17 enzyme activity is the rate limiting step in androgen synthesis. . Increased expression of CYP17A1 or P450c17 activity is seen in theca cells obtained from PCOS women [15]. Hyperinsulinemia, commonly associated with PCOS exaggerates the response of theca cells to circulating LH. PCOS ovaries also have increased expression of enzymes of the alternate backdoor pathway of dihydrotestosterone production [16] Serum 11-oxygenated androgens 11β-hydroxy androstenedione, 11-ketoandrostenedione, 11β-hydroxy testosterone, and 11-ketotestosterone were significantly higher in PCOS than in control subjects, as was the urinary 11-oxygenated androgen metabolite 11β-hydroxy androsterone and the levels correlated with metabolic risk [17]. In about 20–30% women adrenal androgens dehydroepiandrosterone (DHEA) and its sulfate DHEAS are increased. Zona reticularis cells have an exaggerated response to adrenocorticotrophic hormone [18].

Ovarian folliculogenesis

In the ovary, follicular development is a well co-ordinated process of recruitment of follicles, growth and finally selection of one dominant follicle for maturation and ovulation. The initial phase of follicular recruitment and development is gonadotropin independent but regulated by autocrine, paracrine and local factors. Anti Mullerian Hormone (AMH) is a glycoprotein of the transforming growth factor beta family [19]. It is secreted primarily by the granulosa cells of the pre-antral and small antral ovarian follicles up to 6 mm in size. It is an important regulator for recruitment of follicles. In the ovaries the imbalance between LH, FSH androgens and AMH leads to follicular arrest [19].

Insulin resistance

Insulin resistance (IR) and hyperinsulinemia have a central role in pathogenesis of PCOS and high prevalence is reported in women and adolescents with PCOS [20] It is seen in both lean and obese

patients, independent of BMI [21] IR in PCOS occurs because of complex interplay of genetic susceptibility, intra uterine factors, adaptation to energy excess, early pubarche and adiposity (Referred to chapter on IUGR). IR is tissue selective and it is present in muscle, liver and adipose tissue mainly to metabolic effects of insulin but ovarian and adrenal tissue are sensitive to the effect of insulin on steroidogenesis [20]. IR is due to a post receptor defect, with serine phosphorylation of tyrosine residues of the insulin receptor rather than threonine [22] Insulin potentiates the effect of LH on theca cells, increasing androgen synthesis and decreasing hepatic synthesis of sex hormone binding globulin (SHBG), thus increasing the level of free androgen.

Obesity is a common feature of PCOS since its original description by Stein-Leventhal in 1930. Obesity, especially abdominal obesity, worsens the clinical and hormonal picture of PCOS. It is reported that nearly 30–40% of adolescent PCOS patients are overweight or obese [23,24]. Despite this high prevalence, obesity or IR are not included in the diagnostic criteria for PCOS. However, they confer a high risk of various metabolic abnormalities like type 2 diabetes, hypertension, dyslipidemia or metabolic syndrome in these patients.

Neuroendocrine axis

Increased LH level, LH pulse frequency and increased LH to FSH ratio have been described well in patients with PCOS, but are not mandatory for diagnosis. There is intrinsic gonadostat alteration with increased GnRH pulse frequency which causes increased LH secretion over FSH from gonadotrophs [25]. In addition, HA blunts the negative feedback of progesterone on GnRH secretion, leading to increased LH secretion. Abnormalities or alterations in other neurotransmitters in the central nervous system are also implicated in pathogenesis of PCOS. Kisspeptin, encoded by gene KISS1 is a major regulator of GnRH neurosecretion and ovulation [26]. The other neurotransmitters involved are neurokinin B (NKB) which stimulates, while dynorphin reduces LH secretion. Some neurons in the arcuate nucleus express all the three neurotransmitters and are called KNDy neurons [27]. KNDy neurons are sensitive to sex steroids and modulate GnRH pulses. Alteration in their function could lead to the neuroendocrine changes seen in PCOS. In rodent models of PCOS, enhanced Gaba-aminobutyric acid drive to GnRH neurons which resulted in increased GnRH secretion has been demonstrated [28]. GnRH neurons express AMH receptor, AMH2. In mice, central injection of AMH causes increased LH pulse, so it is postulated that high levels of AMH in PCOS women may increase LH secretion [29]. Insulin receptors are expressed at various sites involved in the regulation of GnRH secretion, but whether circulating high levels of insulin have a role and the pathways which may be affected are still not clear.

Genetics

There is familial aggregation of PCOS phenotype. In a large cohort of twins, it was reported that genetic component contributed to 70% of PCOS pathogenesis [30]. Early studies that focused on candidate genes related to the reproductive axis, insulin action and inflammation yielded inconclusive results. Genome wide association studies (GWAS) conducted in Han Chinese women identified 11 loci with a strong risk for PCOS [31,32]. Results were validated in Caucasian populations as well [33]. The Differentially Expressed in Normal and Neoplastic Development isoform A1 (DDEND1A) gene was identified as a strong risk marker. DDEND1A gene has 2 transcripts and the protein is localized in the cytoplasm and nuclei of theca cell and now shown to be expressed in zona reticularis too [34] DDEND1A.V2, the second transcript is more expressed in theca cells from PCOS ovaries than normal. Overexpression of DDEND1A.V2 in normal theca cells leads to increased expression of CYP17A1 and CYP11A1 and androgen production while knockdown reverses this process [34]. Genes located in the susceptibility locus shown in the GWAS are related to gonadotropins, lipid metabolism and cell cycle regulation. The association of the risk variants with features of PCOS like HA, IR or PCOM needs further study [35].

Diagnosis

Due to a significant overlap in the clinical features of PCOS and changes during normal puberty, diagnosis of PCOS is difficult in adolescents.

All the diagnostic criteria for PCOS unanimously agree that all secondary causes of oligo-anovulation and hyperandrogenism should be excluded by appropriate tests before a diagnosis of PCOS can be made. The conditions which have to be excluded are hypothyroidism (serum thyroid stimulating hormone, TSH), hyperprolactinemia (serum prolactin), Cushing syndrome (overnight dexamethasone suppression test) non classical congenital adrenal hyperplasia (follicular phase early morning 17 hydroxy progesterone) and androgen producing adrenal or ovarian tumours. Non classical CAH is a very common condition resembling PCOS. If the basal 17OHP levelis>6 nmol/L, cosyntropin stimulated 17OHP levels should be estimated to rule out non classical CAH.

Hyperandrogenism

Hyperandrogenism (HA), clinical or biochemical is a key feature of all diagnostic criteria for PCOS. Hirsutism, acne, menstrual irregularity and alopecia are features of HA in adult women. Hirsutism is defined as growth of terminal hair in androgen dependent areas, with a male-like pattern. The modified Ferriman Gallaway (mFG) score, a semi-quantitative scoring of hirsutism, is followed widely. It assesses terminal hair growth (>5 mm thick long black hair) in nine areas considered to be androgen dependent [36]. Each area is visually scored from zero (no terminal hair visible) to four (terminal hair consistent with a well-developed male). But it has limitations as it is subjective, equal score is given to all areas (trunk and face are scored equally) and the rapidity of progression is not taken into account. Prevalence of hirsutism varies in different ethnicities. In adult women an mFG score above the 95th centile for the population would be > 8 in premenopausal White Caucasians, while >9–10 in Mediterranean, Hispanic and Middle Eastern women [37], >2 for Han Chinese women and >5 in Southern Chinese women [38]. There is lack of normative data in adolescence. Sexual hair growth increases through puberty and prevalence of upper lip score of 1–2 increased from 7.3% < 2 years of menarche to 28.2% in girls 2 years after menarche [39]. A score ≥4–6 is considered to be significant [11]. Hirsutism should be differentiated from hypertrichosis which is vellus hair growth over arms and legs, not dependent on androgen. Alopecia is uncommon in adolescents. Comedonal acne is common in adolescents [40]. All girls with moderate to severe inflammatory acne or acne resistant to treatment should be evaluated for HA. Biochemical HA is documented by measuring total or free testosterone or calculating free androgen index (discussed in diagnosis section). About 10–15% patients may have only raised adrenal androgens, DHEAS or androstenedione.

Menstrual irregularity

The median age of menarche in most populations is 12–13 years. Menstrual irregularity is very common at menarche for healthy girls. By 1 year post menarche, 60% girls have 10 cycles per year and 90% achievethis by the end of 3 years [41]but some may take even up to 5 years to become regular in some cases [42]. Most cycles after menarche in the first year vary from 20 to 90 days and girls rarely have amenorrhoea for >90 days [43]. Girls who have amenorrhoea or cycle length >90 days at more than one year post menarche should be evaluated for hyperandrogenism. In some girls dysfunctional uterine bleeding could be a feature of chronic anovulation. Conversely, ovulatory dysfunction may still be present despite having regular cycles in some cases. Documentation of low mid luteal serum progesterone level will confirm anovulation.

The international consensus guidelines on diagnosis of PCOS state that persistence of menstrual intervals shorter than 20 or longer than 45 days, 2 years post menarche provide evidence of oligo-anovulation [44]. Girls with consecutively cycle length >90 days require further evaluation regardless of the years post menarche. Diagnosis of PCOS should be considered in girls who have attained final height and pubertal development but who have primary amenorrhoea and [45]. The same has been endorsed by the recent evidence based guidelines of PCOS [11].

Polycystic ovarian morphology

Rotterdam 2003 criteria included PCOM as one of the criteria for diagnosis of PCOS. PCOM is diagnosed if, on a transvaginal ultrasound, either one of the ovaries has a volume >10 ml or there are

more than 12 follicles 3–9 mm in size arranged peripherally in the absence of a dominant follicle [46]. With advancement of technology and 3 dimensional imaging, the Androgen excess-PCOS task force suggested to increase follicular number to >24 [47] There are issues with using PCOM as a criterion for diagnosis of PCOS in adolescents. Transvaginal ultrasound is not possible in girls who are not sexually active. Moreover, a multi-follicular pattern, defined as > 6 follicles 4–10 mm in size distributed throughout the ovary which is not associated with HA, is seen in healthy adolescents. Various studies have reported PCOM prevalence in normal healthy girls as 35–40% [23,48]. Ovarian volume increases with pubertal onset, reaches a maximum just after menarche and then stabilizes. AE_PCOS task force concluded that in adolescents, when ultrasound is done transabdominally, ovarian volume >10 ml of either ovary should be used to diagnose PCOM [47]. If a girl has complaints of menstrual irregularity with evidence of hyperandrogenism, ovarian ultrasound is not required.

Laboratory evaluation

Hyperandrogenism

Serum levels of total testosterone, free testosterone, DHEAS or androstenedione can be measured to document biochemical hyperandrogenemia. Commercially available assays for testosterone lack sensitivity at the low levels found in patients with PCOS. A study comparing three different methods of testosterone estimation in 500 samples of adult women showed poor precision and variability in the results, highlighting the inadequacies of available assays [49]. Liquid chromatography-tandem mass spectrometry (LC-MS/MS) is the gold standard for measurement of all steroids. Because of this, the Endocrine Society had recommended that all steroid measurements should be done by LC-MS/MS [50], but this is expensive and not widely available. Testosterone is bound to sex hormone binding globulin (SHBG) in circulation. SHBG concentration determines the level or percentage of free testosterone. Free testosterone assays are even less standardized than the total testosterone assays and more unreliable. Equilibrium dialysis is the ideal method for free testosterone estimation. But free testosterone or free androgen index can be calculated by total testosterone and SHBG concentration. Testosterone levels begin to rise with the onset of puberty and reach peak levels a few years after menarche. There is no absolute value of testosterone above which hyperandrogenism can be diagnosed.

Testosterone should be considered elevated when it is greater than the adult female normal range according to the assays performed in the laboratory with a well defined range. For an assay using extraction method total testosterone concentration >1.82 nmol/L [51]or using an LC-MS/Ms assay >1.79 nmol/L is considered elevated [52]. About 20–30% women with PCOS have an elevated adrenal androgen, DHEAS.

There is no role for measuring gonadotropins for diagnosis of PCOS. An elevated LH:FSH ratio may be seen but is not diagnostic.

Anti-mullerian hormone (AMH)

AMH reflects the primordial ovum pool and its level correlates with antral follicular count as well as ovarian volume [53]. AMH levels do not vary with menstrual cycle or previous oral contraceptive use and require one time sampling. AMH levels are increased in patients with PCOS compared to normal ovulatory subjects. Studies have shown that the AMH level correlates with testosterone [54], and with different phenotypes as well as severity of PCOS [55].

This led to the surmise that AMH is a biomarker for PCOM and could be used in the diagnosis of PCOS. However, because of challenges in the AMH assay like differential response to preanalytical proteolysis, conformational change in AMH dimer, sample to sample variability and conversion factor anomalies in-between assays [56], there is no consensus on use of AMH for diagnosis of PCOS. With improvisation and standardization of assays and validation of normative data in different populations and age groups AMH could be used for diagnosis of PCOM. Recent evidence based guidelines also state that AMH should not be used as an alternative to PCOM or as a single test for diagnosis of PCOS [11].

Associated comorbidities

Health consequences of PCOS are due to the associated obesity, insulin resistance and hyperandrogenism. There is increased prevalence of impaired glucose tolerance and diabetes [57], metabolic syndrome [58] and cardiovascular disease (CVD) risk factors [59]. All adolescents with PCOS should be screened and evaluated for these conditions with measurement of blood pressure, oral glucose tolerance test and a lipid profile [11]. Thereafter follow up assessment should be done annually in a patient who has a high risk related to overweight/obesity, ethnicity or who has a family history of diabetes and CVD. An important issue at this young age would be emphasis on maintaining a healthy lifestyle together with periodic assessment of comorbidities to prevent long term morbidity. Although adolescents and women with PCOS have higher prevalence of risk factors for CVD, there is no evidence of increased CVD events or mortality. Well designed long term studies are needed to address this.

Treatment

There is no specific treatment for the underlying cause and pathophysiology of PCOS. There are 2 components of therapy, the first being control of symptoms of hyperandrogenism (hirsutism, acne, irregular cycles or infertility) and the second improving and preventing long term morbidity associated with PCOS (metabolic syndrome, type 2 diabetes, emotional wellbeing and self esteem). Consultation with the adolescent herself as to her perceived concerns is central to engagement and maintenance of co-operation with a treatment plan. The various options available are lifestyle intervention, combined oral contraceptive pills (COCP), antiandrogens, metformin, bariatric surgery and local cosmetic therapies.

Lifestyle intervention

This is the key component of therapy in all patients. A couple of small randomized control trials in adolescents [60,61] have shown that healthy lifestyle (dietary restriction with intensive exercise) increases the number of menstrual cycles, decreases hirsutism score, decreases testosterone level with increase in SHBG. Even 5–10% weight loss in young women who have excess weight has shown beneficial effects [62] and is considered successful weight reduction in 6 months, but there is paucity of data on which type of diet or exercise is beneficial in PCOS, as to whether this weight loss may lead to sustained benefits in menstrual irregularity, increase ovulation rates and eventually increased pregnancy rates. It is not known if it is effective in normal weight adolescents. All these programs have a high dropout rate. Behavioral therapy and family support are cornerstones of success of lifestyle intervention program. With healthy lifestyle the benefit of improving cardiovascular fitness is equally important even if it does not lead to amelioration of symptoms of HA.

Combined oral contraceptive pill (COCP)

COCPs which contain estrogen and progesterone should be considered as first line drugs in adolescents who have a clear diagnosis of PCOS, for control of HA symptoms and/or to regularize menstrual cycles [11]. The estrogen component of COCPs increases SHBG, thereby decreasing free androgen. Progestins decrease LH secretion and reduce androgen production. Some progestins like drosperinone have anti-androgenic properties. However, studies have not shown clinical benefit of use of COCPs with these progestins. Evidence is lacking on which is the best combination of COCPs or for how long treatment should be given? Use of COCPs is associated with worsening of metabolic profile, increase in triglycerides and insulin resistance and increase risk of thromboembolism. The lowest effective dose of estrogen (20–30 ug of ethinyl estradiol or its equivalent) or natural estrogen is preferred [11].

Antiandrogens

Androgen receptor blockers like spironolactone, flutamide or third generation progestin (cyproterone acetate) and 5 alpha reductase enzyme blockers (finastride) are the antiandrogens available.

RCTs comparing efficacy of one agent over the other are not available. Spironolactone is the most commonly used one, starting at a dose of 25 mg/day to a maximum of 200 mg/day. Spironolactone is associated with intermenstrual bleeding, breast tenderness, scalp alopecia or fatigue. Flutamide is not widely used because of potential for hepatotoxicity at high doses >250 mg/day but data has shown that 1 mg/kg/day is not hepatotoxic in extended use and is very effective [63]. Hirsutism is significantly reduced with use of antiandrogens as compared to metformin monotherapy. Efficacy is increased when used in combination with COCPs, metformin or 2 antiandrogens. In sexually active adolescents, antiandrogens should be used only after effective contraception to avoid possible feminizing effects on male fetuses.

Metformin

It is an insulin sensitizer that has shown beneficial effect on weight, metabolic parameters especially glucose tolerance and menstrual irregularity in adolescents [61,64]. Most of the studies are limited by small sample size and short study duration (usually 16–24 weeks). Gastrointestinal side effects are associated with metformin use and are dose dependent. No serious adverse events have been reported. The recent guidelines state that metformin, in addition to lifestyle intervention, could be considered in adolescents with a clear diagnosis of PCOS or with symptoms of PCOS before the diagnosis is made [11].

Combination treatment

Lifestyle intervention is the first line treatment in all adolescents with PCOS especially those who are overweight or obese. Addition of COCPs to this would improve symptoms of PCOS. Recent evidence based guidelines recommend that in combination with the COCP, metformin could be considered in adolescents with PCOS and BMI $\geq$25 Kg/m^2, where COCP and lifestyle changes do not achieve desired goals. In addition, combination of antiandrogens with the COCP should only be considered in PCOS to treat hirsutism, after six months or more when COCP and cosmetic therapy have failed to adequately improve symptoms. Also combination therapy could be considered for treatment of androgenic alopecia. A low dose triple drug combination of metformin, flutamide and pioglitazone when compared with COCPs showed no improvement in androgen excess in adolescents but had better effect on lipid abnormalities, carotid intima media thickness and body composition parameters [65].

Contraception

One in 10 women who have oligomenorrhoea ovulate spontaneously. If an adolescent is sexually active, she should follow the general principles of contraception. No specific method/agent is recommended for PCOS girls.

Anti-obesity treatment including bariatric surgery

None of the anti-obesity medications are approved for use in children and adolescents. Bariatric surgery for improvement of PCOS features and infertility is considered as an experimental therapy, even in adult women with PCOS.

Summary

PCOS which is a heterogeneous condition has hyperandrogenism and menstrual irregularity as the characteristic features. The exact aetiology of this condition is not known. Diagnosis of PCOS in adolescents is difficult as the clinical features of PCOS cannot be differentiated from the changes seen during normal puberty. Obesity, insulin resistance and resulting hyperinsulinemia are common associations and play a role in pathogenesis. Because of this patients are at a high risk of developing diabetes, dyslipidemia, hypertension and cardiovascular diseases. Lifestyle modifications are

recommended in all patients with PCOS. Oral contraceptive pills, antiandrogens and metformin alone or in combination are used for treatment of clinical features of hyperandrogenism.

Practice points

- In adolescents diagnosis of PCOS should be made in presence of persistent oligomenorrhoea and hyperandrogenism (clinical or biochemical).
- A fine balance has to be maintained in labeling the clinical features as normal pubertal development and over diagnosis of PCOS which can lead to anxiety and stress.
- Even a single clinical pointer to the diagnosis warrants a follow up because the disease may evolve over time.
- Serum AMH estimation and ovarian ultrasound for polycystic ovarian morphology should not be used for diagnosis of PCOS in adolescents.
- Patients are at a high risk for type 2 diabetes, metabolic syndrome and cardiovascular disease.
- Lifestyle modification is the first line of therapy in all patients.

Research agenda

- Standardization of AMH assays for use in clinical practice in PCOS.
- To classify patients into different phenotypes and long term follow up of patients to understand the implications of these phenotypes.
- Long term follow up to evaluate the cardiovascular disease event rate.
- Effect of lifestyle modifications in normal weight or lean patients.

Conflicts of interest

Nothing to declare.

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