บทฟื้นฟูวิชาการ • Review Article

กลุ่มอาการ Polycystic Ovary Syndrome : มุมมองทางพันธุศาสตร์

สุพัชญ์ สีนะวัฒน์
หน่วยชีววิทยาการเจริญพันธุ์ ภาควิชาสูติศาสตร์และนรีเวชวิทยา คณะแพทยศาสตร์ มหาวิทยาลัยขอนแก่น

Polycystic Ovary Syndrome : The Genetic Aspect

Supat Sinawat
Reproductive Biology Unit Department of Obstetrics and Gynecology, Faculty of Medicine, Khon Kaen University

Introduction

Polycystic ovarian syndrome (PCOS) is a spectrum of diseases comprising the polycystic-like ovary (PCO) together with the symptoms of excess androgen production such as hirsutism, oily skin and acne as well as the manifestations of oligo-ovulation, for instance, oligomenorrhea, secondary amenorrhea, or abnormal uterine bleeding. Patients acquiring this syndrome may also experience difficulty in getting pregnant. The infertility problem in PCOS cases is thought to be due to several causes such as ovulatory disorder and the excess in androgen production resulting in poor quality of follicles and subsequently poor fertilization or low implantation rates. It is noticed for years that PCOS tend to occur in the persons with family history of oligomenorrhea or infertility, and thus the genetic inheritance of PCOS is a reasonable hypothesis. Based on the study of first-degree relatives of women diagnosed with PCOS, it was thought that polycystic ovarian syndrome has some degree of genetic predisposition [1-6]. None of these early family studies, however, convincingly established mode of inheritance because too few families were studied, parental phenotypes were not established, and the male phenotype was uncertain. Despite the lack of comprehensive phenotype information, the older literature clearly suggested clustering of PCOS in families, and suggested a mode of inheritance most consistent with an autosomal dominant pattern[7]. During the past decade, more information regarding the pattern of inheritance of polycystic ovarian syndrome was revealed and thus it is becoming clearer for us to understand the complexity of inheritable route of this syndrome. This article reviews the up-to-date information regarding our current understanding of the genetic component of this syndrome.

Genetic aspect of PCOS: Familial studies

During the 1990s, genetic origin of PCOS attracted the interests of several groups of scientists around the world. Family studies of PCOS were thus re-investigated with more careful phenotyping. Although small numbers of families were investigated the findings were suggestive of an autosomal dominant or modified dominant mode of trait inheritance[8-10]. The study by Legro and colleagues[10] was the largest of these, encompassing a prospective analysis of 80 PCOS women and 115 of their sisters. In this study, 22% of the sisters of the affected women fulfilled the diagnostic criteria for PCOS and an additional 24% were hyperandrogenemic but had more than six menses per year. This led to the conclusions that there is familial clustering of hyperandrogenemia with or without oligomenorrhea, and that hyperandrogenemia is inherited as a dominant trait.

The fact that there is concordance of phenotypes in identical twin pairs provides strong evidence for a genetic component of PCOS. Several case reports describe PCOS in monozygotic twins[11-12]. A more recent study of 19 monozygotic and 15 dizygotic twins revealed that five and six of the twin pairs, respectively, had discordant ovarian ultrasound scans[13], leading the authors to conclude that a PCOS ovarian morphology is not shared by monozygotic

twins and that PCOS is not caused by a single autosomal gene. However, it is important to note that the authors did report a significant genetic influence on levels of androstanediol glucuronide in these twin pairs.

In vitro studies of molecular and biochemical phenotype in PCOS cases

It is reasonable to presume that if PCOS is a genetic disease, cells from PCOS women would show a stable biochemical or molecular phenotype that distinguishes them from normal cells. Studies conducted on thecal cells collected from follicles from PCOS and normal ovaries revealed that PCOS thecal cells retained biochemical and molecular features of increased production of progesterone, 17 α-hydroxyprogesterone and testosterone; increased expression of CYP11a, CYP17 (CYP stands for Cytochrome P450) and type II 3β-hydroxysteroid dehydrogenase, but not steroidogenic acute regulatory protein (StAR) mRna[14]; and increased transcription of the CYP17, but not the StARgene[15]. These alterations parallel findings made on freshly isolated thecal cells studied in short-term culture[16], measurements of mRNAs encoding steroidogenic proteins in freshly isolated theca[17] and ovarian responses to human chronic gonadotropin (hCG) stimulation invivo[18]. Dunaif and colleagues[19] also identified differential patterns of phosphorylation of the β-subunit of the insulin receptor in skeletal muscle cells and fibroblasts from women with PCOS. Approximately 50% of the sample population of PCOS women studied showed the increased insulin receptor serine phosphorylation. This observation was recently confirmed in ovarian tissue[20].

Collectively, the family studies and the experiments conducted on cells in culture are consistent with the concept that genetic factors contribute to PCOS. However, the number of genes involved is not known, although the preponderance of data suggests a dominant or modified dominant pattern of inheritance indicative of an oligogenic disease.

Molecular alterations in other cell types besides reproductive organs

It is clear that, although its reproductive features characterize PCOS, multiple organs are affected[21]. Excessive thecal cell androgen secretion is characteristic of PCOS[14,16,18]. Alterations in the profile of luteinizing hormone (LH) secretion (pulse frequency and amplitude) and excessive adrenal androgen production[22] are also common in women with PCOS. Insulin resistance can be demonstrated in 50% or more of PCOS women[22-23], the primary tissues affected being skeletal muscle and adipose tissue. Pancreatic β-cell dysfunction, independent of obesity, is also associated with PCOS[23-24]. The insulin resistance and β-cell dysfunction are undoubtedly linked to the increased risk of type II diabetes mellitus in PCOS[24]. Higher rates of early pregnancy loss have been documented in PCOS[25-26]. Moreover, there is increased risk of endometrial adenocarcinoma. The early pregnancy wastage has been attributed to an abnormal steroidal milieu resulting from hypersecretion of LH[27]. The increased incidence of endometrial adenocarcinoma is usually ascribed to the unopposed action of estrogen on the endometrium [28-29]. However, an alternative explanation, yet to be explored, is that an intrinsic abnormality in endometrial cell function causes early pregnancy loss and neoplasia.

One commonly asked question is that whether there is any evidence for common metabolic/molecular abnormalities among different cell types in PCOS. The preliminary studies by Wood and colleagues demonstrated altered expression of the secreted frizzled-related protein 4 in cultured thecal cells, cultured dermal fibroblasts, freshly isolated adipocytes and skeletal muscle biopsies, as well as cultured skeletal muscle myocytes from women with PCOS[30]. This suggests that expression of a single gene can be dysregulated in multiple cell types. Whether the multiple endocrine abnormalities are directly linked, or are independent features reflecting the contribution of diverse genetic and environmental factors, has considerable significance for studies on the genetics of PCOS. Hypotheses built upon tight association of these disturbances are most compatible with the notion that PCOS is caused by a limited number of genetic/ environmental factors. The alternative raises the possibility of a polygenic/multifactorial disorder that would require comprehensive phenotyping of subjects to facilitate identification of causative molecular and environmental factors in specific disease subsets, as in the case of the genetics of type II diabetes[31].

PCOS and metabolic imprinting

Besides the genetic hypothesis, another alternative to the origin of PCOS is that PCOS is caused by environmental factors that result in a stable metabolic

imprint. The idea of permanent metabolic imprinting is attaining acceptance in the biomedical community, based largely on the examination of the consequences of intrauterine environment on subsequent adult disease. It is noteworthy that prenatal exposure of female rhesus monkey fetuses to testosterone has been reported to promote the development of PCOS-like biochemical features, including increased LH levels and impaired insulin secretion and sensitivity in adult life.[32-33] The notion of a stable metabolic (non-genetic) imprint arising during development, which might influence organogenesis or cellular composition (e.g. β-cell hypoplasia or hyperplasia), is not difficult to envision. However, a stable metabolic (epigenetic) imprint in dividing cells is more difficult to explain, unless there are alterations in expression of imprinted genes or post-translational modifications of long-lived proteins that are passed down to daughter cells in sufficient quantities to alter cellular function. Alternatively, an extra-genomic replicating molecule would have to be contemplated. These considerations lead us to favor a primary role for genetic factors in the pathophysiology of PCOS, although environmental factors (e.g. obesity) could clearly exacerbate metabolic consequences of a variant genotype predisposing to PCOS.

PCOS: Association studies and linkage analysis

Association studies and linkage analysis for candidate genes, together with genome-wide scanning using non-parametric methods, are used to identify the genes causing diseases where several different genetic loci are likely to contribute to the phenotype. The latter approach is ideal because it makes no assumptions regarding the gene(s) involved, the mode of inheritance, disease penetrance or prevalence. Unfortunately, no investigative team studying PCOS has been able to assemble a sufficient number of families with affected sib-pairs to perform a genome-wide scanning at the present. Consequently, the existing literature is based exclusively on association and linkage studies for selected candidate genes.[34-35]

This reflects difficulty in accruing families with multiple affected sisters and available parents, despite the fact that PCOS is a common disorder. However, testing available family DNA sets for candidate genes is not an unreasonable exercise in the absence of sufficient populations to conduct a genome-wide scan. Yet, the universe of possible candidates from which to choose in the case of PCOS is potentially large and the number of wrong guesses for PCOS candidate genes is expected to be high. Wrong guesses are not necessarily without value, since the exclusion helps to narrow the field of possibilities and adjusts the level to which the gene products are emphasized in models of PCOS pathophysiology. To date, the candidate genes that have been investigated include those involved in steroid hormone synthesis and action, genes involved in carbohydrate metabolism and fuel homeostasis, genes involved in gonadotropin action and regulation, and genes in the major histocompatability region. Among the most promising candidate genes for which there is evidence for linkage or association are the CYP11a[36], the insulin gene[34,37] and a locus near the insulin receptor on chromosome 19p13.3[35,38,39]. Although mutations in the insulin receptor gene have been found in rare cases in PCOS[40-41], the gene at 19p13.3 is not likely to be the insulin receptor, as it lies 2 meagbases centromeric to the insulin receptor gene. Follistatin, originally thought to be a viable candidate gene, appears less likely to be involved, based on recent studies which failed to identify variants in the follistatin gene strongly associated with PCOS.[35,42] Notably, two different studies have fingered the same locus at 19p13.3, raising considerable interest in this region. Chromosome 19 is gene-rich and a number of putative and known genes are in the vicinity D19S884 and deserve special scrutiny.

Other candidate genes for PCOS

There is a need to increase the number of candidate genes to test for association and linkage with PCOS. One strategy to identify candidate disease genes is to define differential patterns of transcript and protein expression in normal and diseased tissues or cells. By examining those genes whose transcripts or protein products are increased or reduced in the diseased cells, pathways and regulatory networks may emerge which point to the underlying molecular pathology. As noted earlier, PCOS cells in long-term culture have been shown to display stable biochemical and molecular differences from normal cells, allowing for relative comparison of gene and protein expression. The methodology that can be using to make these comparisons include suppression subtractive hybridization (SSH)[43], DNA micro-array analysis and a

novel protein chip technology employing surface-enhanced laser desorption ionization coupled with time-of -flight mass spectrometry[44]. DNA array analysis and SSH may potentially overlook alternative splicing of transcripts. These technologies also cannot detect gene variants that might influence protein function but not level of transcript expression, or differences in post-transnational modification of proteins (e.g. proteolytic processing, phosphorylation). These are also weaknesses that can be overcome by the protein chip methodology.

Conclusion

Polycystic ovarian syndrome comprises a spectrum of manifestations including acne, oily skin, oligomenorrhea, amenorrhea, abnormal uterine bleeding and infertility. The common findings on the investigation include polycystic ovary (PCO), hyperandrogenemia and the increased LH : FSH ratio. This syndrome is among the most common endocrinopathies found in women of the reproductive age and several hypotheses have been proposed regarding the etiology of this syndrome. One of the most likely explanation for the occurrence of this syndrome is genetic alteration that can be passing from one generation to the nexts. The improvement in molecular biology techniques as well as the more information we obtained from the human genome project will, hopefully, lead us to understand more about the possible genetic components of PCOS. The precise understanding of the pathogenesis of this syndrome will, thus, help us to properly tackle with the consequences of this syndrome or even possibly prevent its occurrence in the future generations.

References

1. Cooper H, Spellacy W, Prem K, Cohen W. Hereditary factors in the Stein-Leventhal syndrome. Am J Obstet Gynecol 1986; 110:371-87.
2. Ferriman D, Purdie A. The inheritance of polycystic ovarian disease and a possible relationship to premature balding. Clin Endocrinol 1979; 11:219-300.
3. Cohen PN, Givens JR, Wiser W. Polycystic ovarian disease, maturation arrest of spermiogenesis, and Klinefelter's syndrome in siblings of a family with familial hirsutism. Fertil Steril 1975; 26:1228-38.
4. Wilroy RS Jr, Givens GR, Wisler WL. Hyperthecosia: an inheritable form of poly-cystic ovarian disease. Birth Defects 1975; 11:81-5.
5. Govems JR. Familial polycystic ovarian disease. Endocrinol Metab Clin North Am 1988; 17:711-83.
6. Lunde O, Magnus P, Sandvik L. Familial clustering in polycystic ovarian syndorme. Gynecol Obstet Invest 1989; 28:23-30.
7. Legro RS, Spielman R, Urbanek L. Familial clustering in polycystic ovarian syndrome. Gynecol Obstet Invest 1989; 28:23-30.
8. Carey AH, Chan KL, Short F. Evidence of a single gene defect causing polycystic ovaries and male pattern baldness. Clin Endocrinol 1993; 38:653-8.
9. Norman RJ, Masters S, Hague W. Hyperinsulinemia is common in Family members of women with polycystic ovary syndorme. Fertil Steril 1996; 66-942-7.
10. Legro RS,Driscoll D, Strauss JF III. Evidence for a genetic basis for hyperandrogenemai in polycystic ovary syndrome. Proc Natl Acad Sci USA 1998; 95:14956-60.
11. Mcdonough PG, Mahesh VB, Ellegood JO. Steroid, follicle-stimulating hormone, and luteinizing hormone profiles in identical twins with polycystic ovaries. Am J Obstet Gynecol 1972; 113:1072-8.
12. Hutton C, Clark F. Polycystic ovarian syndrome in identical twins. Postgrad Med J 1984; 60:64-5.
13. ahanfar S,Edin JA, Warren P. A twin study of polycystic ovary syndrome. Fertil Steril 1995; 63:478-86.
14. Nelson VL, Legro RS, Strauss JF III. Augmented androgen production is a stable phenotype of propagated theca cells from polycystic ovaries. Mol Endocrinol 1999; 13:946-57.
15. Wickenheisser J, Quinn PG, Nelson VI. Differential activity o th ecytochrome P 450 17α-hydroxylase and steroidogenic acute regulatory protein gene promoters in normal and polycystic ovary syndrome theca cells. J Clin Endocrinol Metab 2000,85:230-11.
16. Gilling-Smith C, Willis DS, Beard RW. Hypersecretion of androstenedione by isolated thecal cells from polycystic ovaries. J Clin Endocrinol Metab 1994; 79:1158-65.
17. Jakimiuk AJ, Weitsman SR, Navab A. Luteinizing hormone receptor, steroidogenesis acute regulatory protein, and steroidogenic enzyme messenger ribonucleic acids are overexpressed in theca and granulosa cells from polycystic ovaries. J Clin Endocrinol Metab 2001; 86:1318-23.
18. Gilling-Smith C, Story H,Rogers Y. Evidence for a primary abnormality of thecal cell steroidogenesis in the polycystic ovary syndrome. Clin Endocrinol 1997; 47:93-9.
19. Dunaif A, Xia J, Book C-B. Excessive insulin receptor serine phosphorylation in cultured fibroblasts and in skeletal muscle. J Clin Invest 1995; 96:801-10.
20. Moran C, Huerta R, Conway-Wyers B-A. Altered autophosphorylation of the insulinreceptor in the ovary of a woman with polycystic ovary syndrome. Fertil Steril 2001; 75:625-8.
21. Strauss JF III, Dunaif A. Molecular mysteries of polycystic ovary syndorme. Mol Endocrinol 1999; 13:800-5.

22. Rittmaster RS, Deshwal N, Lehman L. The role of adrenal hyperandrogenism, insulin resistance, and obesity in the pathogenesis of polycystic ovarian syndrome. J ClinEndocrinol metab 1993; 76:1295-300.
23. Dunaif A. Insulin resistance and polycystic ovary syndrome: mechanism and implications for pathogenesis. Endrocr Rev 1997; 18:774-800.
24. O'Meara NM, Blackman JD, Ehrmann DA. Defects in β-cell function in functional hyperandrogenism. J Clin Endocrinol Metab 1993; 76:1241-7.
25. Sagle M, Bishop K, Ridley N. Recurrent miscarriage and polycystic ovaries. Br Med J 1988; 1027-8.
26. Balen AH, Bishop K, Ridley N. Miscarriage rates following in-vitro fertilization are increased in women with polycystic ovaries and reduced by pituitary desensitization with buserelin. Hum Reprod 1993; 8:9599-64.
27. Watson H, Kddy DS, Hamilton-Fairley D. Hypersecretion of luteinizing hormone and ovarian sterioids in women with recurrent early miscarriage. Hum Reprod 1993; 8:829-33.
28. Jackson RL, Dockerty MB. The Stein-Leventhal syndrome: analysis of 43 cases with special reference to endometrial cancer. Am J Obstet Gynecol 1957;73:161-73.
29. Smyczek-Gargya B, Gellert M. Endometrial cancer associated with polycystic ovaries in young women. Pathol Res Pract 1992;188: 946-8.
30. Wood JR, Christenson LK, McAllister JM. Suppressive subtractive hybridization reveals up-regulation of the WNT signaling antagonist, secreted frizzled related protein 4, in PCOS theca cells. Presented at the 83rd Annual Meeting of the Endocrine Society (abstract OR39-3), Denver, Colorado, 2001.
31. Almind K, Koria A. Putting the genes for type II diabetes on the map. Nature Med 2001; 7:277-8.
32. Dumesic DA, Abbott DH, Eisner JR. Prenatal exposure of female rhesus monkeys to testosterone propionate increases serum luteinizing hormone in adulthood. Fertil Steil 1997; 67:155-63.
33. Eisner JR, Dumesic DA, Kemnitz JW. Timing of prenatal androgen excess determines differential impairment in insulin secretion and action in adult female rhesus monkeys. J Clin Endocrinol Metab 2000; 85:1206-10.
34. Franks S, Gharani N, Waterworth D. The genetic basis of polycystic ovary syndrome. Hum Reprod 1997; 12: 2641-8.
35. Urbanek M, Legro RS, Driscoll DA. Thirty-seven candidate genes for polycystic ovary syndrome:strongest evidence for linkage is with follistatin. Proc Natl Acad Sci USA 1999; 96:8573-8.
36. Gharani N, Waterorth DM, Batty S. Association of the steroid dynthesis gene CYP11 a with polycysticovary sydrome and hyperandrogenism. Hum Mol Genet 1997; 6:397-402.
37. Waterworth DM,Bennett ST, Gharani N. Linkage and association of insuling gene VNTR regulatory polymorphism with polycystic ovary syndrome. Lancet 1997; 349:986-90.
38. Urbanek M, Legro RS, Driscoll DA. Searching for the polycystic ovary syndrome genes.I J Pediatr Endocrinol Metab 2000; 13:1311-13.
39. Tucci S, Futterweit W, Concepcion ES. Evidence for associationof polycystic ovary syndrome in Causasian women with a marker at the insulin receptor gene locus. J Clin Endocrinol Metab 2001; 86:446-9.
40. Moller DE, Flier J. Detection of an alteration in the insulin receptor gene in a patient with insulin resistance, acanthosis nigricans and polycystic ovary syndrome. N Engl J Med 1988; 319:1526-9.
41. Moller DE, Choen O, Yamaguchi Y. Prevalence of mutations in the insulin receptor gene in subjects with features of the type A syndrome of insulin resistance. Diabetes 1994; 43:247-55.
42. Urbanek M, Wu X, Vicdery KR. Allelic variants of the follistatin gene in polycystic ovary syndrome. J Clin Endocrinol Metab 2000; 85:4455-61.
43. Diatchenko L, Lau Y-FC, Campbell AP. Suppression subtractive hybridization : a method for generating differentially regulated or tissue-specific cDNA probes and libraries. Proc Natl Acad Sci USA 1996; 96:6025-30.
44. Wright GL Jr, Cazares LH, Leung S-M. Protein Chip surface enhanced laser desorption/ionization (SELDI) mass spectrometry: a novel protein biochip technology for detection of prostate cancer biomarkers in complex protein mixtures. Prost Cancer Dis 2000; 2:264-76.