ejpmr, 2016,3(6), 505-512

SJIF Impact Factor 3.628

EUROPEAN JOURNAL OF PHARMACEUTICAL AND MEDICAL RESEARCH

www.ejpmr.com

Review Article

ISSN 2394-3211

EJPMR

PCOS: A MULTIFACTORIAL HEALTH PROBLEM WITH FEMALES

Nautiyal Himani*, Md. Shane Ali, Bisht Akansha, Aswal Shivani, Joshi Girish and Kumar Ashok

Department of Pharmaceutical Science, Shree Dev Bhoomi Institute of Education, Science and Technology, Village Mazhon P.O Poundha via- Premnagar, Dehradun (Uttrakhand). 248001.

***Corresponding Author: Nautiyal Himani**
Department of Pharmaceutical Science, Shree Dev Bhoomi Institute of Education, Science and Technology, Village Mazhon P.O Poundha via-Premnagar, Dehradun (Uttrakhand). 248001.

Article Received on 22/04/2016 **Article Revised on 13/05/2016** **Article Accepted on 01/06/2016**

ABSTRACT

Polycystic ovarian syndrome, also known as Stein-Leventhal syndrome is a common endocrine and metabolic disorder which is affecting women of reproductive age. The syndrome has complex pattern and multifaced etiology producing imbalance of the hypothalamic-pituitary-adrenal (HPA) axis, thyroid involvement and metabolic syndrome (insulin resistance). PCOS is a common disorder that causes changes in hormone levels or that disrupts the hormone, typically giving up a higher than the normal level of certain sex hormones and insulin, which can triggers the many different symptoms including irregular periods, excess body hair, and/or obesity, acne weight gain, infertility as well as other cardiovascular disease (CVD) risk factors. As a whole PCOD is a route of various health problems in female. It is a life style disorder as the people who are not involved in physical activity and resideing in stressfull environment are more prone to disorder as compare with physically active persons.

KEYWORDS: Polycystic ovarian syndrome, Female metabolic syndrome, 'Stein-Leventhal Syndrome'.

INTRODUCTION

Polycystic ovary syndrome is a heterogeneous disorder associated with ovulatory dysfunction and hyperandrogenism. The condition is one of the most common endocrine and metabolic disorders also called "the female metabolic syndrome", affecting up to 15% of women of reproductive age.[1][2] In actual fact in PCOS, the word 'cyst' simply means an empty egg follicle. Polycystic ovarian syndrome is a common disorder that causes changes in hormone levels or that disrupts the hormone, by which there is an increase in higher than the normal level of certain sex hormones and insulin. These changes trigger the different symptoms including irregular periods, excess body hair, and/or obesity, acne and weight gain. In Polycystic ovaries the follicles may stop growing too early, preventing the release of an egg. Instead of bursting to release the egg, they gradually build up on the ovaries to form lots of small cysts which are actually swollen egg chambers waiting for the right hormone to trigger the maturation and release of an egg. There's a characteristic pattern of ovarian enlargement to 1.5 to 3 times normal size, and a number of small cystic structures of less than 10 mm, which are usually located in a circle around the ovarian surface, commonly called a 'string of pearls.'

History

The term Polycystic Ovarian Syndrome (PCOS) was first described by Irving Stein and Michael Leventhal as a Triad of 'Amenorrhea', 'Obesity' and 'Hirsutism' in 1935, both working at the Department of Obstetrics and Gynecology, Michael Reese Hospital, Chicago, USA, when they observed the relation between obesity and reproductive disorders.[3] It is hence also known as the 'Stein-Leventhal Syndrome' or 'Hyperandrogenic Anovulation' (HA) and this is the most common endocrine ovarian disorder affecting approximately 2-8% women of reproductive age worldwide.[4] Nowadays, it is also referred to as the 'Syndrome O' i.e. Over nourishment, Over production of insulin, Ovarian confusion and Ovulatory disruption. PCOS is currently considered as a lifestyle disorder.[5]

Clinical Pattern of PCOD in fertile age:

Pituitary = ↑LH
↓FSH

Metabolic signs: Diabetes
Obesity [abdominal]
↑ Muscle mass
↓Bone mass

Gynecological sign: Irregular period
Amenorrhea
Infertily
Endometrial cancer

ACNE, HIRSUTISM

Signs and symptoms

1. Menstrual disorders: PCOD mostly produces oligomenorrhea (few menstrual periods) or amenorrhea (no menstrual periods)[6,7]
2. Infertility.[7]
3. High levels of masculinizing hormones: The most common signs are acne and hirsutism (male pattern of hair growth), but it produce hypermenorrhea (heavy and prolonged menstrual periods), androgenic alopecia (increase hair thinning or diffuse hair loss)[6,8]
4. Hyperinsulinaemia, insulin resistance and type 2 diabetes. Type 2 diabetes results primarily from insulin resistance and is correlated with both hyperandrogenaemia and obesity[9]
5. Women with PCOS often develop hypertension and have an increased likelihood of developing cardiovascular risk factors [10]. They also appear to have impaired mental health.[11,12]
6. PCOS-related symptoms lead to higher levels of depression, psychological and psychosexual morbidity, and increased exposure to stressful stimuli.[13]

Diagonosis

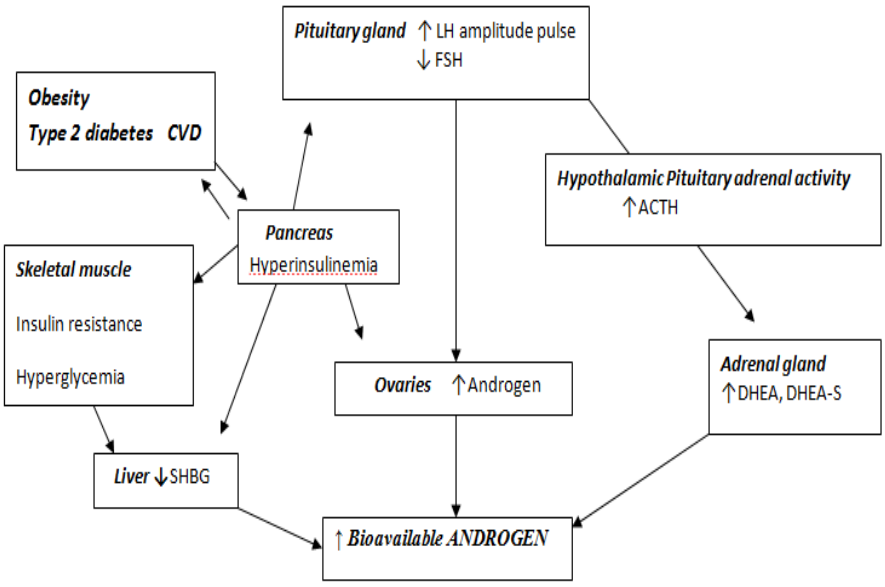
1. Gynecologic ultrasonography, specifically looking for small ovarian follicles. These are believed to be the result of disturbed ovarian function with failed ovulation, reflected by the infrequent or absent menstruation that is typical of the condition.[14]
2. Laparoscopic examination may reveal a thickened, smooth, pearl-white outer surface of the ovary.
3. Serum (blood)levelof androgens (malehormones),including androstenedione and testosterone may be elevated.[6]
4. Anti-Müllerian hormone (AMH) is increased in PCOS, and may become part of its diagnostic criteria.[15,16]

Associated conditions

1. Fasting biochemical screen and lipid profile.[17]
2. 2-Hour oral glucose tolerance test (GTT) in women with risk factors (obesity, family history, history of gestational diabetes)[6] may indicate impaired glucose tolerance (insulin resistance) in 15–33% of women with PCOD.[17]
3. Fasting insulin level or GTT with insulin levels (also called IGTT).

Etiology and pathophysiology of PCOS

The pathogenesis of PCOS is multifactorial and multiple causative mechanisms are involving interactions between certain genes and environmental factors[18,19], dysfunction/regulation by the gonadotropins and intraovarian factors, hyperinsulinemia as well as hyperandrogenism.



Genetics

There is a evidence of genetic component based on the existence of familial clustering[20,21] and twin studies have displayed a twofold increased concordance of PCOS in genetically identical twins compared with non-identical twins.[22] In spite of numerous association studies (mainly focusing on genes associated with the synthesis and metabolism of androgens and insulin), the way in which PCOS is inherited remains unclear.[23] Recent efforts, using modern mapping techniques, have made some progress to identify promising candidate genes. Two promising candidate genes have so far emerged. The first, a locus on chromosome 19p13.2, is associated with high susceptibility to PCOS[24] and the second is the fat-mass and obesity associated gene, whose polymorphism has been found to be associated with PCOS.[25] However, the studies implicating these two locus, needs to be confirmed in larger studies and in other populations.

Gonadotrophins aberrations
The increased frequency of LH pulses from the pituitary gland is secondary to increased frequency of GnRH pulses in the hypothalamus. This leads to increased pituitary production of LH.

Ovarian aberrations
The elevated levels of LH lead to increased androgen production from the theca cells. The relatively lower FSH levels contribute to arrested follicular development in the ovary, which, in turn leads to disturbed negative feedback. This results in continued aberrations in the secretion of LH and FSH.

Aberrations in the adrenal gland:
Impaired adrenal androgen production leads to increased levels of DHEA and DHEAS, which, in turn, also increases the circulating pool of free and bioavailable androgens.

Pancreatic aberrations
The increased levels of bioavailable androgens lead to increased insulin resistance in peripheral tissues (mostly in the skeletal muscle). This leads to hyperinsulinemia, which, by facilitating the stimulatory role of LH, leads to increased ovarian androgen production. Moreover, increased release of free fatty acids from adipocytes is seen, due to insulin resistance and hyperandrogenism.[26]

Prenatal exposure to androgens
Excess fetal exposure to maternal androgens is thought to contribute to inducing the PCOS phenotype in offspring/children, based on experimental data from animal studies as well as clinical material of pathological conditions in human populations (i.e., congenital adrenal hyperplasia).[27,28] In humans, higher testosterone levels, which were elevated to male levels, have been found in the umbilical vein in female infants born to mothers with PCOS.[29] However, the only prospective study of the relationship between prenatal androgen exposure and the development of PCOS during the human female adolescence did not confirm any association between these variables.[28]

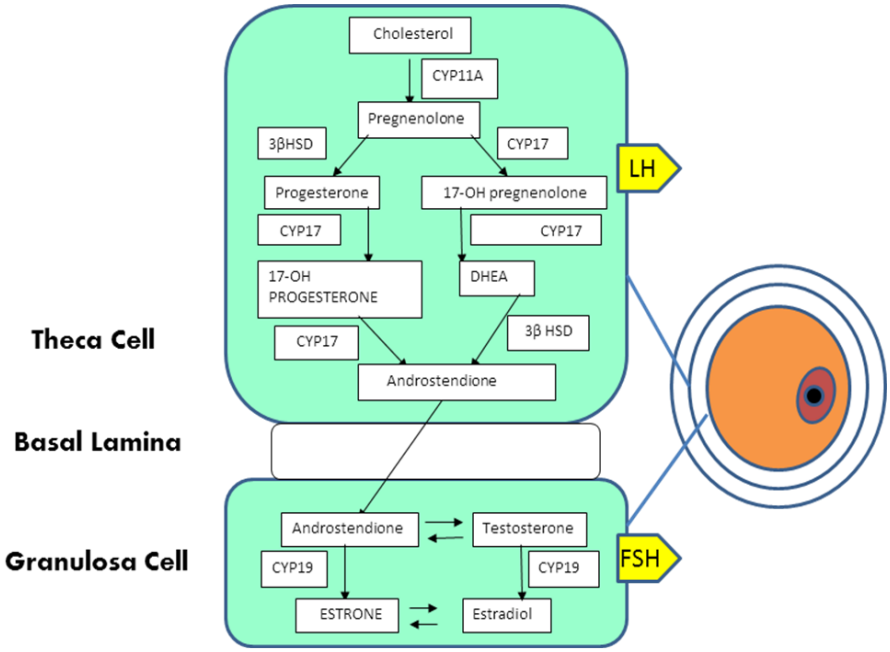
Obesity
Obesity has a considerable effect on the manifestation of PCOS[30] and family studies have implied that weight gain may promote the PCOS phenotype in a predisposed population.[31] Weight gain is usually associated with a worsening of symptoms, while weight loss usually ameliorates the symptoms and the endocrine/reproductive and metabolic disturbances.[32,33]

Hypothalamus/pituitary - ovarian axis dysfunction
A large proportion of women with PCOS have increased levels of LH[34,35] and normal/decreased levels of FSH[36,37], resulting in the classical hormonal hallmark of an increased LH/FSH ratio. The prevalence of an increased LH/FSH ratio is partly related to BMI, and it is more prevalent in PCOS of normal weight and less common with increasing BMI.[38] The increase in LH is explained by an increased pulse frequency of the hypothalamic gonadotropin-releasing hormone (GnRH)[35], which may favour the production of the β-subunit of LH over the β-subunit of FSH [39], and/or by increased pituitary sensitivity to GnRH stimulation.[40] The increase in LH causes the ovaries to favour the production of androgens from the theca cells carrying LH receptors. Partly due to the increased LH stimulation there is increased ovarian production of androgens mainly from the theca cells. The theca cell layer in follicles of Polycystic ovary has been shown to be thicker[41] and androgen hypersecretion and increased expression/efficacy of the key enzymes participating in the synthesis of androgens has been verified.[42,43] The follicular steroid secretion follows the two-cell cooperation, where LH-stimulated theca cells produce mainly androstenedione from the steroid precursor cholesterol and via pregnenolone, progesterone and 17-OH-progesterone. Androstenedione is converted to testosterone after diffusion through the basal lamina to the granulosa cells (GC). This cell compartment is rich in aromatase and consequently androstenedione is aromatized to estrone or testosterone. Testosterone is aromatized to estradiol.[41,44], see Fig. The androgens, and in particular androstenedione, is taken up by diffusion to the capillaries of the theca and may then undergo aromatization in skin, liver and adipose tissue to estradiol after conversion to testosterone[45]. In addition, insulin increases the response of the theca cells to LH, resulting in increased androgen production[46,47] and hyperinsulinemia is common in women with PCOS.[48] In the GCs, FSH stimulates the expression of enzymes that metabolize androstenedione to estradiol.[44] Studies of follicular fluids and in vitro studies of GCs from anovulatory PCOS women demonstrate that GCs, for the most part, remain steroidogenically active with increased aromatase activity, compared with similarly sized follicles from non-PCOS women. Thus, increased estradiol production in PCOS is dependent on the ovulatory status of the patient[49], but also on body weight.[50] Consequently, also normal estradiol levels have been found in PCOS.[50,51] Anti-Mullerian hormone (AMH) is a specific hormone of small growing follicles being produced in GCs of primary follicles and the growing follicles continue to express AMH until the time they are selected for dominance by FSH.[52] After the selection of the dominant follicle, the GCs normally start to produce inhibins and estradiol that cause a progressive decline in FSH by negative feed-back.[53] In PCOS, the primary follicle pool is much higher than in normal women and the number of antral follicles, as assessed by ultrasound, is shown to correlate tightly with the serum AMH levels, which also has been found to be 2-3 times higher than in non-PCOS women.[54,55] The increased AMH levels could be one factor that influences follicular maturation [56]. The high androgen levels and this AMH-related mechanism may be factors behind the follicular arrest, which is the basis of the characteristic appearance of PCO with arrested multiple small follicles <10 mm in

diameter. Concerning androgens and follicular arrest, there is a positive correlation between the number of arrested follicles and androgen levels.[18] These high androgen levels and the excessive stimulation of follicular cells by insulin and LH might produce high levels of cyclic adenosine monophosphate in the GCs, which may result in premature terminal differentiation and, hence, arrest follicular growth.[56]

Taken together, it is likely that the abnormal endocrine environment in PCOS women, with the hypersecretion of LH, androgens and insulin, together with the relative FSH deficiency[57,58] and increased AMH levels, impair the development of the maturing pool of follicles.[54]

Monophosphate, leads to increased expression of cholesterol side chain cleavage cytochrome P450 (CYP11A), 17αhydroxylase/C17, 20 lyase cytochrome P450 (CYP17), and 3β hydroxysteroid dehydrogenase (3β-HSD). The theca cell is then able to synthesize androstenedione from cholesterol. Androstenedione diffuses across the basal lamina into the granulosa cells and in normal ovaries, the major part of androstenedione is converted into estrone by aromatase cytochrome P450 (CYP19) and then to estradiol by 17 β –hydroxysteroid dehydrogenase (17β-HSD). However, in PCOS ovaries, testosterone is produced (by conversion by 17β-HSD) to a larger degree from androstenedione.



Adrenal androgen production

The adrenal cortex synthesizes all the three major androgens; dehydroepiandrosterone sulfate (DHEAS), androstenedione and testosterone, and this is the other major site of female androgen production, besides the ovaries. DHEAS is almost exclusively (97-99%) produced by the adrenal cortex and androstenedione is produced in both the adrenal gland and the ovaries[45], whereas 25% of testosterone is synthesized by the adrenal gland, 25% in the ovary and the remaining part being produced through peripheral conversion from androstenedione in liver, adipose tissue and skin.[45] Around 60-80% of PCOS women have high concentrations of circulating testosterone.[59] In PCOS women, the prevalence of DHEAS excess is 20-30%, depending on ethnicity and DHEAS levels decline up to the age of ~ 45 years.[60] The increased DHEAS levels in PCOS women compared with controls is verified up to the perimenopausal ages.[61] However, the mechanisms of the adrenal androgen excess in PCOS is still unclear, although it has been proposed that it may result from increased metabolism of cortisol, which could lead to decreased negative feedback on ACTH secretion.[62]

SHBG production

SHBG is produced in the liver. Women with PCOS have decreased levels of SHBG, which is caused by inhibitory effects of insulin on the SHBG production.[63,64] In addition, overweight/obesity decreases SHBG production even more.[65] Decreasing SHBG levels result in increased levels of biologically active androgens, as normally about 80% of testosterone[64] and 8% of androstenedione[65] is generally bound by SHBG, with the other main binding protein being the constitutively expressed albumin.[64]

Insulin resistance

Insulin resistance, i.e., impaired stimulation of glycogen formation in all major target tissues (skeletal muscle, adipose tissue, liver, kidney), is a pathogenic characteristic feature of PCOS, particularly among obese subjects.[67] The molecular mechanisms of insulin resistance involve defects in the insulin-receptor signaling pathway in both adipocytes and in skeletal muscle.[68]

Insulin resistance causes compensatory hyperinsulinemia and might contribute to hyperandrogenism and gonadotropin aberrations through several mechanisms. Insulin may act directly in the hypothalamus, the pituitary or both and thereby contribute to abnormal gonadotropin levels [69]. High insulin can also serve as a co-factor to stimulate ACTH-mediated androgen production in the adrenal glands [70]. As stated above, the stimulation of the ovaries is exerted by a synergistic effect of insulin upon LH stimulation of the theca cells [46, 47] and insulin may also directly stimulate theca cell proliferation.[48] In addition, high insulin concentrations also cause decreased circulating SHBG, thereby increasing the levels of free bioavailable testosterone [63,71]. Administration of insulin in young non-PCOS women resulted in increased LH-puls frequency, thereby implying an association between insulin and hypothalamus-pituitary-ovarian-axis-activity.[72]
Hyperinsulinemia also results in increased levels of free IGF-1 and human theca cells express IGF-1 receptor genes (as well as insulin receptor genes), which is another way in which androgen production is stimulated.[73] In addition, free IGF-1 is a potent growth factor that can induce proliferation of ovarian cells.[74]

In conclusion, excess of androgens in PCOS of ovarian and/or adrenal origin initiates or maintains a vicious circle, where hyperandrogenism leads to hypothalamus/pituitary abnormalities, ovarian dysfunction, insulin resistance and abdominal obesity, which in turn stimulates further androgen production. Polycystic ovary syndrome (PCOS) is a prevalent, complex endocrine disorder characterised by polycystic ovaries, chronic anovulation and hyperandrogenism leading to symptoms of irregular menstrual cycles, hirsutism, acne and infertility.

REFERENCES

1. March WA, Moore VM, Willson KJ, Phillips DI, Norman RJ, Davies MJ. The prevalence of polycystic ovary syndrome in a community sample assessed under contrasting diagnostic criteria. Hum Reprod, 2010; 25: 544-551.
2. Norman RJ, Dewailly D, Legro RS, Hickey TE. Polycystic ovary syndrome. Lancet, 2007; 370: 685-697.
3. Stein IF, Leventhal MN Amenorrhea associated with bilateral Polycystic Ovaries. American Journal of Obstetrics and Gynaecology, 1935; 29: 181.
4. Knochenhauer ES Prevalence of Polycystic Ovarian Syndrome. Journal of Clinical Endocrinology & Metabolism, 1998; 83(9): 3068-3082.
5. NidhiR et al. Prevalence of Polycystic Ovarian Syndrome in Indian adolescents. Journal of Paediatric Adolescent Gynecology, Aug, 2011; 24(4): 223-227.
6. Teede H, Deeks A, Moran L. "Polycystic ovary syndrome: Complexes condition with psychological, reproductive and metabolic manifestations that impacts on health across the lifespan". BMC Med, 2010; **8**(1): 41. doi:10.1186/1741-7015-8-41. PMC 2909929. PMID 20591140.
7. Mayo Clinic Staff (4 April 2011). "Polycystic Ovary Syndrome – All". MayoClinic.com. Mayo Clinic. Retrieved 15 November 2011
8. Christine Cortet-Rudelli, Didier Dewailly (Sep 21, 2006). "Diagnosis of Hyperandrogenism in Female Adolescents".Hyperandrogenism in Adolescent Girls. Armenian Health Network, Health.am. Retrieved, 2006-11-21.
9. Barber TM, McCarthy MI, Wass JA, Franks S. Obesity and polycystic ovary syndrome. Clin Endocrinol (Oxf), 2006; 65: 137–145.
10. Tsilchorozidou T, Overton C, Conway GS. The pathophysiology of polycystic ovary syndrome. Clin Endocrinol (Oxf), 2004; 60: 1–17.
11. Himelein MJ, Thatcher SS. Polycystic ovary syndrome and mental health: a review. Obstet Gynecol Surv, 2006; 61: 723–732.
12. 12.Barnard L, Ferriday D, Guenther N, Strauss B, Balen AH, Dye L. Quality of life and psychological well being in polycystic ovary syndrome. Hum Reprod, 2007; 22: 2279–2286.
13. 13.Carmina E, Legro RS, Stamets K, Lowell J, Lobo RA. Difference in body weight between American and Italian women with polycystic ovary syndrome: influence of the diet. Hum Reprod, 2003; 18: 2289–2293.
14. Richard Scott Lucidi (25 October 2011). "Polycystic Ovarian Syndrome". eMedicine. Retrieved, 19 November, 2011.
15. Dewailly D, Andersen CY, Balen A, Broekmans F, Dilaver N, Fanchin R, Griesinger G, Kelsey TW, La Marca A, Lambalk C, Mason H, Nelson SM, Visser JA, Wallace WH, Anderson RA. "The physiology and clinical utility of anti-Mullerian hormone in women".Hum. Reprod.Update, 2014; **20**(3): 370 doi:10.1093/humupd/dmt062. PMID 24430863.
16. Broer SL, Broekmans FJ, Laven JS, Fauser BC. "Anti-Müllerian hormone: ovarian reserve testing and its potential clinical implications". Hum. Reprod. Update, 2014; **20** (5): 688–701. doi:10.1093/humupd/dmu020. PMID 24821925 .
17. "Polycystic Ovarian Syndrome Workup". eMedicine.25October2011.Retrieved 19 November 2011.
18. Ehrmann DA. Polycystic ovary syndrome. N Engl J Med., Mar 24, 2005; 352(12):1223-36.
19. Legro RS, Spielman R, Urbanek M, Driscoll D, Strauss JF, 3rd, Dunaif A. Phenotype and genotype in polycystic ovary syndrome. Recent Prog Horm Res., 1998; 53: 217-56.
20. Legro RS, Driscoll D, Strauss JF, 3rd, Fox J, Dunaif A. Evidence for a genetic basis For hperandrogenemia in polycystic ovary syndrome. Proc Natl Acad Sci U S A, Dec 8, 1998; 95(25): 14956-60.

21. Vink JM, Sadrzadeh S, Lambalk CB, Boomsma DI. Heritability of polycystic ovary syndrome in a Dutch twin-family study. J Clin Endocrinol Metab, Jun, 2006; 91(6): 2100-4.
22. Balen A, ed. Current management of Polycystic Ovary Syndrome. London, UK: RCOG, 2010.
23. 23.Urbanek M, Woodroffe A, Ewens KG, Diamanti-Kandarakis E, Legro RS, Strauss JF,3rd, et al. Candidate gene region for polycystic ovary syndrome on chromosome 19p13.2. J Clin Endocrinol Metab, Dec, 2005; 90(12): 6623-9.
24. Barber TM, Bennett AJ, Groves CJ, Sovio U, Ruokonen A, Martikainen H, et al. Association of variants in the fat mass and obesity associated (FTO) gene with polycystic ovary syndrome. Diabetologia, Jul, 2008; 51(7): 1153-8.
25. Diamanti-Kandarakis E, Papavassiliou AG, Kandarakis SA, Chrousos GP. Pathophysiology and types of dyslipidemia in PCOS. Trends Endocrinol Metab, Sep, 2007; 18(7): 280-5.
26. Vendola KA, Zhou J, Adesanya OO, Weil SJ, Bondy CA. Androgens stimulate early stages of follicular growth in the primate ovary. J Clin Invest, Jun 15, 1998; 101(12): 2622-9.
27. Hickey M, Sloboda DM, Atkinson HC, Doherty DA, Franks S, Norman RJ, et al. The relationship between maternal and umbilical cord androgen levels and polycystic ovary syndrome in adolescence: a prospective cohort study. J Clin Endocrinol Metab, Oct, 2009; 94(10): 3714-20.
28. Barry JA, Kay AR, Navaratnarajah R, Iqbal S, Bamfo JE, David AL, et al. Umbilical vein testosterone in female infants born to mothers with polycystic ovary syndrome is elevated to male levels. J Obstet Gynaecol, 30(5): 444-6.
29. Norman RJ, Masters SC, Hague W, Beng C, Pannall P, Wang JX. Metabolic approaches to the sub classification of polycystic ovary syndrome. FertileSterile, Feb, 1995; 63(2): 329-35.
30. Legro RS, Bentley-Lewis R, Driscoll D, Wang SC, Dunaif A. Insulin resistance in the sisters of women with polycystic ovary syndrome: association with hperandrogenemia rather than menstrual irregularity. J Clin Endocrinol Metab, May, 2002; 87(5): 2128-33.
31. Clark AM, Ledger W, Galletly C, Tomlinson L, Blaney F, Wang X, et al. Weight loss results in significant improvement in pregnancy and ovulation rates in anovulatory obese women. Hum Reprod, Oct, 1995; 10(10): 2705-12.
32. Kiddy DS, Hamilton-Fairley D, Bush A, Short F, Anyaoku V, Reed MJ, et al. Improvement in endocrine and ovarian function during dietary treatment of obese women with polycystic ovary syndrome. Clin Endocrinol (Oxf), Jan, 1992; 36(1): 105-11.
33. Van Santbrink EJ, Hop WC, Fauser BC. Classification of normogonadotropic infertility: polycystic ovaries diagnosed by ultrasound versus endocrine characteristics of polycystic ovary syndrome. FertileSterile, Mar, 1997; 67(3): 452-8.
34. Rebar R, Judd HL, Yen SS, Rakoff J, Vandenberg G, and Naftolin F. Characterization of the inappropriate gonadotropin secretion in polycystic ovary syndrome. J Clin Invest, May, 1976; 57(5): 1320-9.
35. Fauser BC, Pache TD, Lamberts SW, Hop WC, de Jong FH, Dahl KD. Serum bioactive and immunoreactive luteinizing hormone and follicle-stimulating hormone levels in women with cycle abnormalities, with or without polycystic ovarian disease. J Clin Endocrinol Metab, Oct, 1991; 73(4): 811-7.
36. Yen SS. The polycystic ovary syndrome. Clin Endocrinol (Oxf), Feb, 1980; 12(2): 177-207.
37. DeVane GW, Czekala NM, Judd HL, Yen SS. Circulating gonadotropins, estrogens, and androgens in polycystic ovarian disease. Am J Obstet Gynecol, Feb15, 1975; 121(4): 496-500.
38. Haisenleder DJ, Dalkin AC, Ortolano GA, Marshall JC, Shupnik MA. A pulsatile gonadotropin-releasing hormone stimulus is required to increase transcription of the gonadotropin subunit genes: evidence for differential regulation of transcription by pulse frequency in vivo. Endocrinology, Jan, 1991; 128(1): 509-17.
39. Ehrmann DA, Rosenfield RL, Barnes RB, Brigell DF, Sheikh Z. Detection of functional ovarian hyperandrogenism in women with androgen excess. N Engl J Med., Jul 16, 1992; 327(3): 157-62.
40. Magoffin DA. Ovarian theca cell. Int J Biochem Cell Biol., Jul, 2005; 37(7): 1344-9.
41. Nelson VL, Qin KN, Rosenfield RL, Wood JR, Penning TM, Legro RS, et al. The biochemical basis for increased testosterone production in theca cells propagated from patients with polycystic ovary syndrome. J Clin Endocrinol Metab., Dec, 2001; 86(12): 5925-33.
42. Wickenheisser JK, Quinn PG, Nelson VL, Legro RS, Strauss JF, 3rd, McAllister JM.Differential activity of the cytochrome P450 17alpha-hydroxylase and steroidogenic acute regulatory protein gene promoters in normal and polycystic ovary syndrome theca cells. J Clin Endocrinol Metab, Jun, 2000; 85(6): 2304-11.
43. Hillier SG, Whitelaw PF, Smyth CD. Follicular estrogen synthesis: the 'two-cell, two-gonadotrophins' model revisited. Mol Cell Endocrinol, Apr, 1994; 100(1-2): 51-4.
44. Burger HG. Androgen production in women. FertileSterile, Apr, 2002; 77(4): S3-5.
45. Diamanti-Kandarakis E, Argyrakopoulou G, Economou F, Kandaraki E, Koutsilieris M. Defects in insulin signaling pathways in ovarian steroidogenesis and other tissues in polycystic ovary syndrome (PCOS). J Steroid Biochem Mol Biol, Apr, 2008; 109(3-5): 242-6.
46. Nestler JE. Insulin regulation of human ovarian androgens. Hum Reprod, Oct, 1997; 12(1): 53-62.

47. Duleba AJ, Spaczynski RZ, Olive DL. Insulin and insulin-like growth factor I stimulate the proliferation of human ovarian theca-interstitial cells. FertileSterile, Feb, 1998; 69(2): 335-40.
48. Mason HD, Willis DS, Beard RW, Winston RM, Margara R, Franks S. Estradiol production by granulosa cells of normal and polycystic ovaries: relationship to menstrual cycle history and concentrations of gonadotropins and sex steroids in follicular fluid. J Clin Endocrinol Metab, Nov, 1994; 79(5): 1355-60.
49. Franks S. Polycystic ovary syndrome: a changing perspective. Clin Endocrinol (Oxf).1989 Jul;31(1):87-120.
50. Duleba AJ, Spaczynski RZ, Olive DL. Insulin and insulin-like growth factor I stimulate the proliferation of human ovarian theca-interstitial cells. Fertile Sterile, Feb, 1998; 69(2): 335-40.
51. Polson DW, Franks S, Reed MJ, Cheng RW, Adams J, James VH. The distribution of estradiol in plasma in relation to uterine cross-sectional area in women with polycystic or multifollicular ovaries. Clin Endocrinol (Oxf), May, 1987; 26(5): 581-8.
52. Visser JA, Themmen AP. Anti-Mullerian hormone and folliculogenesis. Mol Cell Endocrinol, Apr, 2005: 29234(12): 81-6.
53. Roseff SJ, Bangah ML, Kettle LM, Vale W, Rivier J, Burger HG, et al. Dynamic changes in circulating inhibin levels during the luteal-follicular transition of the human menstrual cycle. J Clin Endocrinol Metab, Nov, 1989; 69(5): 1033-9.
54. Pigny P, Merlen E, Robert Y, Cortet-Rudelli C, Decanter C, Jonard S, et al. Elevate serum level of anti-mullerian hormone in patients with polycystic ovary syndrome: relationship to the ovarian follicle excess and to the follicular arrest. J Clin Endocrinol Metab. 2003 Dec;88(12):5957-62.
55. Laven JS, Mulders AG, Visser JA, Themmen AP, De Jong FH, Fauser BC. Anti-Mullerian hormone serum concentrations in normoovulatory and anovulatory women of reproductive age. J Clin Endocrinol Metab, Jan, 2004; 89(1): 318-23.
56. Franks S, Mason H, White D, Willis D. Etiology of anovulation in polycystic ovary syndrome. Steroids, May-Jun, 1998; 63(5-6): 306-7.
57. Franks S, Stark J, Hardy K. Follicle dynamics and anovulation in polycystic ovary syndrome. Hum Reprod Update, Jul-Aug, 2008; 14(4): 367-78.
58. Nisenblat V, Norman RJ. Androgens and polycystic ovary syndrome. CurrOpin Endocrinol Diabetes Obes, Jun, 2009; 16(3): 224-31.
59. Chang WY, Knochenhauer ES, Bartolucci AA, Azziz R. Phenotypic spectrum of polycystic ovary syndrome: clinical and biochemical characterization of the three major clinical subgroups. FertilSteril, Jun, 2005; 83(6): 1717-23.
60. Kumar A, Woods KS, Bartolucci AA, Azziz R. Prevalence of adrenal androgen excess in patients with the polycystic ovary syndrome (PCOS). Clin Endocrinol (Oxf), Jun, 2005; 62(6): 644-9.
61. Dahlgren E, Johansson S, Lindstedt G, Knutsson F, Oden A, Janson PO, et al. Women with polycystic ovary syndrome wedge resected in 1956 to 1965: a long-term follow up focusing on natural history and circulating hormones. FertilSteril, Mar, 1992; 57(3): 505-13.
62. Tsilchorozidou T, Honour JW, Conway GS. Altered cortisol metabolism in polycystic ovary syndrome: insulin enhances 5alpha-reduction but not the elevated adrenal steroid production rates. J Clin Endocrinol Metab, Dec, 2003; 88(12): 5907-13.
63. Nestler JE, Powers LP, Matt DW, Steingold KA, Plymate SR, Rittmaster RS, et al. A direct effect of hyperinsulinemia on serum sex hormone-binding globulin levels in obese women with the polycystic ovary syndrome. J Clin Endocrinol Metab, Jan; 1991; 72(1): 83-9.
64. Malley B, Strott C. Steroid hormones: Metabolism and mechanism of action. In: Yen, Jaffe, Barbieri, eds. *Reproductive Endocrinology- Physiology, patophysiology and clinical management*. Philadelphia, Pennsylvania, USA: WB Saunders company, 1999.
65. Tchernof A, Despres JP. Sex steroid hormones, sex hormone-binding globulin, and obesity in men and women. Horm Metab Res., Nov-Dec, 2000; 32(11-12): 526-36.
66. Mendel CM. The free hormone hypothesis: a physiologically based mathematical model. Endocr Rev., Aug, 1989; 10(3): 232-74.
67. Dunaif A, Segal KR, Futterweit W, Dobrjansky A. Profound peripheral insulin resistance, independent of obesity, in polycystic ovary syndrome. Diabetes, Sep, 1989; 38(9): 1165-74.
68. Dunaif A. Insulin resistance and the polycystic ovary syndrome: mechanism and implications for pathogenesis. Endocr Rev., 1997 Dec; 18(6): 774-800.
69. 69.Adashi EY, Hsueh AJ, Yen SS. Insulin enhancement of luteinizing hormone and follicle-stimulating hormone release by cultured pituitary cells. Endocrinology, Apr, 1981; 108(4): 1441-9.
70. 70.Moghetti P, Castello R, Negri C, Tosi F, Spiazzi GG, Brun E, et al. Insulin infusion amplifies 17 alpha-hydroxycorticosteroid intermediates response to adrenocorticotropin in hyperandrogenic women: apparent relative impairment of 17,20-lyase activity. J Clin Endocrinol Metab, Mar, 1996; 81(3): 881-6.
71. 71.Yki-Jarvinen H, Makimattila S, Utriainen T, Rutanen EM. Portal insulin concentrations rather than insulin sensitivity regulate serum sex hormone-binding globulin and insulin-like growth factor binding protein 1 in vivo. J Clin Endocrinol Metab, Nov, 1995; 80(11): 3227-32.
72. Moret M, Stettler R, Rodieux F, Gaillard RC, Waeber G, Wirthner D, et al. Insulin modulation of luteinizing hormone secretion in normal female volunteers and lean polycystic ovary syndrome patients. Neuroendocrinology, 2009; 89(2): 131-9.

73. Bergh C, Carlsson B, Olsson JH, Selleskog U, Hillensjo T. Regulation of androgen production in cultured human thecal cells by insulin-like growth factor I and insulin. Fertil Steril, Feb, 1993; 59(2): 323-31.
74. 74.Gambineri A, Pelusi C, Vicennati V, Pagotto U, Pasquali R. Obesity and the polycystic ovary syndrome. Int J ObesRelat Metab Disord, Jul, 2002; 26(7): 883-96.
75. Escobar-Morreale HF, San Millan JL. Abdominal adiposity and the polycystic ovary syndrome. Trends Endocrinol Metab, Sep, 2007; 18(7): 266-72.