Insulin Resistance (IR) in Women with Polycystic Ovary Syndrome (PCOS)

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Abstract

Background: There are reports which say 65–95% of all the women have PCOS also have insulin resistance which might be due to perturbed receptor tyrosine kinase, or other protein of insulin signaling cascade, modified adipokine signaling, and its secretion when compared to normal women. In addition, the prevalence of type II diabetes is increased in women with PCOS compared with women without PCOS (15% vs. 2.3%). It is now clearly established that insulin resistance is present in obese and non-obese women with PCOS; however, the exact mechanism of insulin resistance in PCOS remain elusive. Recent studies on mechanisms of insulin resistance in PCOS have focused on polymorphisms in genes regulating carbohydrate homeostasis. However, none of these genes have been consistently shown to be related with PCOS. One central paradox regarding insulin resistance in PCOS is the high responsiveness to insulin by the ovary, as opposed to the resistance of the whole body, and this model has been used to explain ovarian hyperandrogenemia as it though to arise from a direct stimulatory effect of insulin on ovarian stromal cells. However, a study from China on women with PCOS appears to contradict this hypothesis suggesting that ovarian cells are also insulin resistant. Insulin resistance is characterized by a post-receptor defect in the action of insulin, the cause of which is still being elucidated. The first step in the action of insulin involves binding to the cell-surface receptor. Following insulin binding, the receptor undergoes auto-phosphorylation on specific tyrosine residues (accomplished by activation of insulin receptor tyrosine kinase IRTK). The activated receptor then activates insulin receptor substrates (such as IRS-1, 2, and 3) that in turn bind to signaling molecules (such as phosphatidylinositol-3 kinase) and activate downstream signaling leading to insulin-mediated glucose transport

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Introduction

The first association between hyperinsulinemia and PCOS was noted by **Burghen et al. (1),** who found a significant positive correlation between insulin, androstene-dione, and testosterone levels among women with PCOS.

Subsequent studies confirmed insulin resistance that is the cause of hyperinsulinemia. It is estimated that 20-40 % of women with PCOS have impaired glucose intolerance, which is approximately seven-fold higher than the rates in age- and weight-matched women **(2).**

There are reports which say 65–95% of all the women have PCOS also have insulin resistance which might be due to perturbed receptor tyrosine kinase, or other protein of insulin signaling cascade, modified adipokine signaling, and its secretion when compared to normal women **(3)**

In addition, the prevalence of type II diabetes is increased in women with PCOS compared with women without PCOS (15% vs. 2.3%) **(4).**

Lean women with PCOS have lower rates of carbohydrate intolerance than obese women with PCOS, although lean women with PCOS even have higher rates than age and weight matched controls. Thus, PCOS is associated with insulin resistance independently of total or fat-free body mass. Obese women with PCOS are more insulin resistant than obese non PCOS or non-obese women with PCOS **(2).**

It is now clearly established that insulin resistance is present in obese and non-obese women with PCOS; however, the exact mechanism of insulin resistance in PCOS remain elusive. Recent studies on mechanisms of insulin resistance in PCOS have focused on polymorphisms in genes regulating carbohydrate homeostasis. However, none of these genes have been consistently shown to be related with PCOS. One central paradox regarding insulin resistance in PCOS is the high responsiveness to insulin by the ovary, as opposed to the resistance of the whole body, and this model has been used to explain ovarian hyperandrogenemia as it though to arise from a direct stimulatory effect of insulin on ovarian stromal cells. However, a study from China on women with PCOS appears to contradict this hypothesis suggesting that ovarian cells are also insulin resistant **(5).**

Ehrmann et al., (6) demonstrated pancreatic β-cell secretory dysfunction in a subset of women with PCOS. This subset is likely to have the highest risk of developing carbohydrate intolerance and type II diabetes.

Insulin resistance is characterized by a post-receptor defect in the action of insulin, the cause of which is still being elucidated. The first step in the action of insulin involves binding to the cell-surface receptor. Following insulin binding, the receptor undergoes auto-phosphorylation on specific tyrosine residues (accomplished by activation of insulin receptor tyrosine kinase IRTK). The activated receptor then activates insulin receptor substrates (such as IRS-1, 2, and 3) that in turn bind to signaling molecules (such as phosphatidylinositol-3 kinase) and activate downstream signaling leading to insulin-mediated glucose transport. Abnormalities in both IRTK activity and in mediators distal to the receptor are present in insulin resistant states**.** Serine phosphorylation of the insulin receptor decreases IRTK activity **(7).**

It has been suggested that serine phosphorylation of the beta chain of the insulin receptor and at the same time of the adrenal and ovarian P450C17 enzyme (the origin or cause of serine phosphorylation is

uncertain, but presumably it would have a genetic basis) would explain both hyperinsulinemia and hyperandrogenism (serine phosphorylation increases and dephosphorylation decreases 17, 20 layse activity and androgen production)**. Zhang et al. (8)** stated simply, serine instead of tyrosine phosphorylation is an "off" mechanism for glucose transport, but an "on" mechanism for P450C17 enzyme activity.

It is unlikely that anovulation is the cause of impaired insulin sensitivity. Clinical changes in insulin sensitivity have been reported, but these take the form of reduced sensitivity during the luteal phase of anovulatory menstrual cycle. Furthermore, complete suppression of ovarian steroids does not alter insulin sensitivity. It is more likely that hyperinsulinemia and insulin resistance contribute to the mechanism of anovulation **(9).**

Excess circulating insulin influences the clinical presentation of PCOS in several major ways:

- **(1)** Directly increases ovarian androgen secretion.
- **(2)** Reduces insulin-like growth factor-binding protein (IGF-BP1) production by the liver which augments the thecal androgen response to LH and consequence, increasing bioavailable insulin like growth factor-1 (IGF-I).
- **(3)** Elevation in insulin and bioavailable IGF-I influencing gonadotropin-secretion, adrenal androgen secretion and also contributing to abnormalities in lipids and lipoproteins
- **(4)** Inhibits hepatic synthesis of sex hormone-binding globulin (SHBG). **(9).**

It has been shown that insulin stimulates ovarian androgen secretion; and this effect perpetuates the chronicity of ovarian hyperandrogenism in PCOS. This effect is partially dependent on an increase of LH receptors but may also be linked to a direct effect of insulin on steriogenesis. Because of peripheral insulin resistance, the effect of insulin on ovarian steriogenesis have been ascribed to insulin binding to ovarian IGF-I receptors. Although this is possible, it is improbable given the low affinity of insulin for IGF-1 receptors (100 to 1000-fold less than IGF-I) **(10).** Insulin most likely binds to its own receptor, which in the ovary is functioning normally.

It should be noted that evidence indicates that the endogenous insulin-like growth factor in the human ovarian follicle is IGF-II in both the granulosa and the thecal cell, studies indicating activity of IGF-I with human ovarian tissue can be explained by the type IGF-I receptor, which is structurally similar to insulin receptor **(11).**

Serum IGF-BP1 has been found to be low in PCOS **(12).** The activity of IGF-I inside the ovary is determined by its intraovarian pool which includes the potential modulating influence of six IGF-BPs (insulin like growth factor binding proteins). The reduction of circulating IGF-BP1 is a consistent finding and is probably the consequence of hyperinsulinemia. Regardless of whether IGF-BP1 reduction results in elevated bioavailable IGF-I levels or not, both IGF-I and insulin are able to stimulate ovarian androgen production in vitro. Also there are human in vivo data corroborate the role of insulin and IGF-I in the pathophysiology of PCOS and specially their role in the androgen disturbances **(12).**

Serine phosphorylation, as noted above, has been associated with decreased IRTK auto-phosphorylation. In fact, this is the probable mechanism of tumor necrosis factor α-induced insulin resistance (TNF- α, a cytokine that causes insulin resistance and is secreted by adipose tissue), Since serine phosphorylation of P450C17-α hydroxylase (the key regulatory enzyme of androgen biosynthesis) increases enzyme activity leading to androgen biosynthesis, it is possible that single defect (serine phosphorylation) can produce both insulin resistance and hyperandrogenism in a subgroup of PCOS women **(11).**

There are some data to suggest that insulin enhances the effect of LH on pre-ovulatory ovarian follicles causing premature activation and subsequent follicle arrest **(13).**

It is possible that hyperinsulinemea (due to insulin resistance) drives the LH effect on ovarian theca cells to cause androgen excess that are intrinsically programmed to produce more androgen. Excess androgens are known to interfere with the process of follicular maturation, thus inhibiting ovulation and adding to the production of arrested follicles **(14).**

Insulin resistance has been associated with an increased incidence of cardiovascular disease (CVD) and atherosclerosis and is now considered to be an inflammatory disorder. Insulin resistance has also recently been associated with increased levels of inflammatory mediators in the blood. Therefore, studies have been conducted to review the levels of inflammation in PCOS.There is increase of tumor necrosis factor-α (TNF- α) in women with PCOS compared with controls. Interestingly, lean women with PCOS had higher TNF- α level than lean normal women while the levels were similar in obese women with PCOS and obese controls **(15)**.

Kelly et al. (16) noted significantly increased levels of C-reactive protein (CRP) and tissue plasminogen activator (tPA) in women with PCOS compared with healthy weight-matched controls. However, when adjusted for insulin sensitivity, CRP was no longer significantly different between groups but the difference in tPA levels remained. Women with PCOS have been shown to have higher plasminogen activator inhibitor type-1 (PAI-1) activity and fibrinogen levels than controls **(17).**

Mechanisms for the Association of Insulin Resistance and PCOS

Insulin as a reproductive hormone

Hyperandrogenemia and ovulatory disturbances are commonly encountered in the syndromes of extreme insulin resistance. There are a number of distinct molecular mechanisms of insulin resistance in these disorders that result in substantial hyperinsulinemia. This observation has led to the hypothesis that hyperinsulinemia causes hyperandrogenemia and anovulation. Similarly, the finding of significant positive correlations between insulin and androgen levels in PCOS has suggested that insulin also contributes to hyperandrogenism in affected women. There is now an extensive body of evidence demonstrating direct ovarian actions of insulin on steroidogenesis as well as the importance of the insulin signaling pathway in the control of ovulation**. (18)**

Insulin receptors are present in normal and polycystic human ovaries. The IGF-I receptor is a tyrosine kinase that shares considerable structural and functional homology with the insulin receptor .The IGF-I receptor is also present in the ovary, and its ligand, IGF-I, is synthesized by the ovary .Insulin can bind to and activate the IGF-I receptor, and IGF-I can bind to and activate the insulin receptor.The affinity of the IGF-I

receptor for insulin is considerably less than it is for IGF-I and vice versa .However, α, β dimers of the insulin and IGF-I receptor can assemble together to form hybrid heterotetramers, which can bind insulin and IGF-I with similar affinity **(19).**

Accordingly, some insulin actions on the ovary may be mediated by the IGF-I or hybrid insulin-IGF-I receptors .Nevertheless, studies using specific anti-insulin receptor antibodies indicate that insulin action on steroidogenesis in granulosa and theca cells isolated from normal and polycystic ovaries is mediated via the insulin receptor .In addition, in granulosa cells from anovulatory PCO, increased insulin levels in synergy with LH may trigger premature LH receptor expression in a subpopulation of small follicles leading to premature granulosa terminal differentiation and the arrest of follicular growth that may contribute to anovulation in this subgroup **(20).**

Ovarian insulin action on steroidogenesis is preserved, despite resistance to insulin's metabolic actions in PCOS. Indeed, in granulosa-lutein cells isolated from ovaries of women with classic PCOS, insulin action on glucose metabolism is significantly decreased, whereas insulin action on steroidogenesis is unchanged compared with granulosa-lutein cells from control women. This observation suggests that in PCOS there is selective insulin resistance in the ovary as there is in skeletal muscle and in skin fibroblasts **(21).**

Genetic susceptibility to PCOS

The possibility that there might be a genetic susceptibility to PCOS, and its associated insulin resistance has been suggested by several observations. First, families with multiple affected women have been reported. Second, the phenotypic similarity between PCOS and the rare syndromes of extreme insulin resistance and hyperandrogenism suggested that insulin receptor mutations might also be present in PCOS. Third,defects in insulin action persist in cultured cells, suggesting that they are genetically determined .Fourth, the fact that insulin resistance could not entirely account for reproductive dysfunction and vice versa suggests that additional factors contributed to the pathogenesis of PCOS **(22).**

Familial clustering of PCOS suggesting a genetic susceptibility to the disorder is now well documented in PCOS. Twin studies have shown heritability of 79% for PCOS with a correlation of 0.71 between monozygotic twins and 0.38 between dizygotic twins, consistent with a major influence of genetic factors in PCOS. Although some studies have suggested that there is an autosomal dominant mode of inheritance these studies have been limited by a lack of prospective design, a failure to examine all first-degree relatives, and the fact that only reproductive-age women can be phenotyped for PCOS. PCOS is more likely a complex genetic disease with at least several susceptibility genes. **(23)**

The intermediate reproductive phenotype of hyperandrogenemia aggregates in PCOS families. About 40% of reproductive-age sisters are affected, but there is phenotypic heterogeneity. Some sisters have classic NICHD PCOS with hyperandrogenemia and oligomenorrhea, whereas others have hyperandrogenemia with regular menses. **(24)**

Hyperandrogenemia is the major underlying reproductive phenotype in PCOS families, and this finding has been replicated in other populations. Affected sisters with either of these hyperandrogenemia phenotypes have insulin resistance metabolic syndrome, and elevated low-density lipoprotein levels. Furthermore, mothers and brothers also have defects in glucose homeostasis and

circulating lipid levels. Therefore, reproductive and metabolic abnormalities are tightly associated in PCOS families, suggesting that they are causally related, have a common pathogenesis, or reflect closely linked genetic traits.

Hyperandrogenemia and hyperinsulinemia are heritable traits in the sisters of women with PCOS. In addition, several reproductive phenotypes can occur in reproductive-age sisters, suggesting that some of the phenotypic heterogeneity of PCOS reflects variable expression of the same gene because sisters would be expected to share the same genetic basis for the disorder. Factors that may contribute to phenotypic variation within families include obesity and insulin resistance. There may also be additional environmental factors or genes that modify the phenotypic expression of PCOS**.(25)**

The association of insulin resistance with this reproductive pathology has been well documented. Due to major implication of insulin resistance in PCOS pathobgenesis, insulin reduction strategies were studied as a possible treatment for infertility in PCOS patients. Weight loss was proved to be a simple and efficient method to improve reproductive parameters in PCOS patients and should be recommended to all overweight and obese patients with infertility. **(25)**

Metformin was showed to induce ovulation, at least in a subset of patients with PCOS, but there are not unequivocal proves concerning its efficacy for pregnancies and live–birth rate, mainly because few trials studied this aspect. Therefore, there are not enough evidence to recommend metformin for infertility treatment in PCOS. Few small studies with newer thiazolidindiones suggest their efficacy for ovulation induction, but further extensive studies are needed to confirm these results. In conclusion, reduction of insulin resistance was proved to ameliorate ovulation rate in PCOS patients, but strong evidence to sustain the utility of insulin–sensitizing drugs as a therapeutic option for infertility are lacking. Future studies are needed to elucidate these aspects and to characterize the particular subtype of patients with higher probability to respond to this treatment. **(26)**

The association between insulin resistance and hyperandrogenism has been reported for the first time in 1921, when Achard and Thiers have described ‘diabetes des femmes a barbe’. Nowadays, the relationship between the two conditions is well documented, especialy based on studies showing that PCOS patients have insulin resistance and compensatory hyperinsulinemia. Although insulin resistance is considered an intrinsec feature of the syndrome, independent of body weight, obesity, a frequent feature among PCOS patients (60–70%), has a major contribution to the aggravation of the insulin resistance. Insulin resistance was also reported in lean PCOS patients, but its prevalence was variable, and some studies even failed to find it. **(27)**

Nowadays, PCOS is considered a disease with a complex and heterogeneous pathogenesis, different mechanisms being involved in different ratios in the appearance of the syndrome. Therefore, in certain patients, insulin resistance may be less involved in the pathogenesis.

The etiology of insulin resistance in PCOS, although intensely studied, is not completely cleared up. The mechanisms most probably involved are defects at the post receptor level. Thus, in 50% of the patients autophosphorylation of the insulin receptor (IR), which inhibits the intrinsic tyrosine–kinase activity of the IR, has been described. Another post receptor anomaly has been described in the muscle cells of the PCOS

patients. This consists in the decrease of the insulin–mediated phosphatidylinositol–3–kinase activity associated to IRS1 (insulin receptor substrate 1), involved in the glucose tapping and carbohydrate metabolism **(28)**

The involvement of insulin resistance and secondary hyperinsulinemia in the PCOS pathogenesis is complex, implying the existence of many mechanisms. In vivo and in vitro studies have proved that insulin directly stimulates ovarian streroidogenesis at the level of theca cells, along with LH. This co–gonadotropic effect contributes to the PCOS hyperandrogenism. Furthermore, insulin decreases the sex hormone binding globulin (SHBG) levels by decreasing the liver synthesis, raising the level of free testosterone. **(29)**

Also, it decreases the IGFBP1 liver synthesis and so it increases IGF1 biodisponibility in the ovaries, causing increased production of ovarian androgens through its co–gonadotropic effect. In 50% of the PCOS patients, the adrenal androgens have an important contribution in the appearance of hyperandrogenism, the involvement of hyperinsulinemia in the alteration of the adrenal steroid genesis being demonstrated. Although experimental studies suggested the contribution of hyperinsulinemia to the characteristic changes of gonadotrop secretion in PCOS, the clinical studies didn't succeed to offer solid evidence **(30)**

Recently, it has been demonstrated that insulin action in the ovaries is mediated by inositolglycan at the post receptor level, and not by tyrosine–kinase cascade. This is how we explain why the insulin resistance, through IR autophosphorylation, in other tissues doesn't impede the insulin effects in the ovaries **(31).**

These mechanisms through which insulin resistance contributes to the appearance of clinical and paraclinical changes in PCOS are mostly demonstrated by in vitro studies. In vivo studies have tried to reproduce these mechanisms by acute administration of insulin, but they obtained contradictory results. Of the clinical studies, the most convincing have been those that proved the amelioration of the clinical and paraclinical parameters by decreasing the insulin resistance, thus supporting its pathogenic role **(32)**.

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