

## Metformin and Troglitazone in the Treatment of Female Infertility Associated with Polycystic Ovarian Syndrome

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### ABSTRACT

Polycystic ovarian syndrome is the most common cause of anovulatory infertility and causes menstrual disruption in 5-10% females, and is characterized by insulin resistance, hyperinsulinemia, hyperandrogenism and anovulation. Such factors are responsible for the increased miscarriage and infertility in women with PCOS. Administration of various insulin sensitizing drugs, such as metformin and troglitazone have been shown to decrease serum androgen concentrations and to increase ovulation rates, increase conception and decrease miscarriage in affected women.

**Key Words:** Infertility, Metformin, Ovary, Troglitazone

### INTRODUCTION

Polycystic ovarian syndrome (PCOS) is a common cause of menstrual disruption and it affects 5–10% of women of reproductive age.<sup>1-3</sup> PCOS is characterized by elevated androgen, insulin, and luteinizing hormone level. Patients with PCOS frequently present with hirsutism, amenorrhea and obesity.<sup>1</sup> PCOS, which is characterized by hyperandrogenemia, hirsutism, oligo- or amenorrhea and anovulation is one of the most common endocrinological diseases encountered in premenopausal women.<sup>4,5</sup> PCOS is the most common cause of anovulatory infertility and is conventionally treated with clomifene.<sup>6</sup> In addition to poor conception rates, pregnancy loss rates are high (30–50%) during the first trimester.<sup>7</sup>

### Hyperinsulinemia, Insulin Resistance and Hyperandrogenism in PCOS

Hyperinsulinemic insulin resistance is a key feature of the polycystic ovary syndrome.<sup>8,9</sup> Dehydroepiandrosterone (DHEA) level is increased in PCOS<sup>10</sup> with hyperandrogenism, abnormal maturation of ovarian follicles and anovulation.<sup>11-14</sup> Anovulation in PCOS is associated with hyperinsulinemia and insulin resistance. Insulin has a stimulatory effect on steroidogenesis by granulosa cells of normal and polycystic ovaries and interacts with gonadotropins in an additive or, as in the case of LH, a synergistic manner. Anovulatory women with PCOS showing elevated levels of insulin interacting with LH may contribute to the mechanism of anovulation.<sup>15</sup>

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These actions seem to be mediated specifically by the insulin receptor rather than by cross-reaction with the type I IGF receptor, even in tissue obtained from women with PCOS with biochemical evidence of insulin resistance. Hyperinsulinemia makes a significant contribution to premature arrest of follicle growth, which is characteristic of anovulation in women with PCOS, and the interaction of insulin with LH is a key element in this process. Insulin may also have a role in amplifying LH-induced androgen production by theca cells, which may help explain the prominence of symptoms of hyperandrogenism in obese subjects with PCOS.<sup>16</sup> There is significant decrease in insulin-stimulated glucose incorporation into glycogen in PCOS cells, which is a metabolic action of insulin. There is a selective defect in insulin actions in PCOS granulosa cells, which suggests ovarian insulin resistance, and this metabolic phenotype is associated with an enhanced IGF-1 mitogenic potential.<sup>17</sup> In order to account for the effects of insulin on PCOS, despite peripheral insulin resistance, it has been suggested that insulin may act through the type-I insulin-like growth factor (IGF) receptor and not the insulin receptor.<sup>18</sup>

### Prostaglandin E in PCOS

Prostaglandin (PGE) is not only involved in inflammatory processes and displays immunomodulatory properties, but is also altered in PCOS.<sup>19,20</sup> Studies showed that follicles from anovulatory women with PCOS hypersecrete PGE when compared with size-matched follicles from normal ovaries or polycystic ovaries from ovulatory women.<sup>21-23</sup> Prostaglandins are involved in the paracrine regulation of the rupture of ovarian follicles associated with ovulation<sup>24</sup> and PGE has been reported to have immunomodulatory properties by modulating cytokine production.<sup>25-27</sup> Experimental studies on animal showed that ovarian concentration of PGE was modified by DHEA-induced hyperandrogenization.<sup>28</sup> The fact that the treatment with DHEA reduced ovarian PGE production was an expected result since DHEA-hyperandrogenized animals not only did not start to cycle but also showed increased levels of TNF- when compared with controls, although the authors have previously reported that the treatment with DHEA increased ovarian PGE.<sup>29</sup> This apparently controversial result could be explained by the fact that, in the previous study, the dose of DHEA administered to prepubertal mice was lower than that used in the later study. The dose of DHEA used in the later study correlates better with the concentration of DHEA described in women with PCOS.<sup>10-14</sup> They also found an inverse relationship between the concentration of DHEA and both the ovarian PGE production and the expression of cyclooxygenase (COX) -the enzyme that synthesizes PGE. The last observation is due to the fact that prostaglandins down-regulate their own synthesis.<sup>30</sup> Therefore the higher

dose of DHEA injected in the later study led to an accumulation of ovarian PGE which in turn could inhibit the expression of COX and consequently the production of ovarian PGE.<sup>28</sup>

### Increased TNF- $\alpha$ in PCOS

TNF has been found to be increased in patients with PCOS.<sup>31</sup> In addition, a mutation of the TNF receptor has been associated with hyperandrogenism.<sup>32</sup> Higher serum TNF- $\alpha$  levels in PCOS correlates both with increased levels of PGE and with anovulatory cycles.<sup>28</sup> It has been reported that TNF- modulates steroidogenesis of both granulosa and theca-interstitial cells by a mechanism independent of those induced by insulin and insulin-like growth factor-I (IGF-I).<sup>33</sup> For this reason, the increase of serum TNF- $\alpha$  observed in animals from the DHEA group would be an additional mechanism to those involved in hyperinsulinemia, which impair ovarian steroidogenesis. The administration of metformin together with DHEA led to serum TNF- $\alpha$  levels similar to control values.<sup>28</sup> In the literature, conflicting results have been reported with regards to the relationship between metformin and the regulation of TNF- $\alpha$ .<sup>34,35</sup>

### Infertility and Miscarriage in Women with PCOS

Both hyperinsulinemia and hyperandrogenism play a pathogenic role in PCOS since they contribute to anovulation.<sup>21</sup> When women with PCOS finally achieve pregnancy (often after a long, arduous, and expensive course of fertility treatments), they are faced with the distressing prospect of a substantially increased risk for miscarriage during the first trimester.<sup>36</sup> Previous studies have suggested that women who hypersecrete LH, a frequent feature of the polycystic ovary syndrome, are at increased risk for miscarriage after either spontaneous or assisted conception.<sup>36,37</sup>

Although the mechanisms linking insulin resistance with anovulatory infertility in PCOS are debatable, proposed mechanisms include a direct stimulation of androgen production from the ovarian stromal cells (thought to directly impair follicle development), impairment of local steroidogenesis mediated via an imbalance in the production of insulin-like growth factors and a direct stimulatory effect on a local (intraovarian) protease inhibitor, plasminogen activator inhibitor-1 (PAI-1), limiting follicle growth.<sup>38</sup> Increased systemic luteinising hormone (LH) and PAI-1 levels have been associated with an increased risk of miscarriage.<sup>6</sup>

Anovulatory infertility due to PCOS should be diagnosed if two of the following three criteria are present, after exclusion of other causes of excess androgen according to the new joint European Society of Human Reproduction and Embryology/ American Society for Reproductive Medicine-sponsored PCOS consensus definition.<sup>39</sup> These

three criteria are: oligo and/ or anovulation, clinical and/or biochemical signs of hyperandrogenism and polycystic ovaries morphology on ultrasound scan, defined as the presence of 12 or more follicles in each ovary (with one ovary sufficient for diagnosis) measuring 2–9 mm in diameter and/or increased ovarian volume (greater than 10 ml).<sup>6</sup>

### Therapeutic Role of Metformin and Troglitazone

Hyperinsulinemia in PCOS impair folliculogenesis and affect follicular development.<sup>40</sup> For these reasons, insulin-sensitizing drugs such as metformin (biguanide) and troglitazone (thiazolidinedione) can improve the rate of spontaneous ovulation.<sup>41</sup> Metformin and the troglitazone in PCOS have provided encouragement that improvement of insulin sensitivity and consequent lowering of circulating insulin levels by these agents may be of therapeutic value in the management of both anovulation and hirsutism.<sup>16</sup>

Administration of various insulin sensitizing drugs, such as metformin, troglitazone and *D-chiro*-inositol have been shown to decrease serum androgen concentrations and to increase ovulation rates in affected women.<sup>7</sup> It has been shown that troglitazone increases the insulin-induced glycogen synthesis but reduces the IGF-1-augmented responses of DNA synthesis in PCOS cells, and it also reverses the expression imbalance between insulin receptor substrate-1 (IRS-1) and IRS-2 in PCOS cells. Troglitazone could divergently alter expression of various IRS molecules and insulin actions and could be used as an ovarian insulin sensitizer and mitogen/steroidogenic inhibitor in PCOS.<sup>17</sup>

Because metformin has beneficial effects on several risk factors for miscarriage in the polycystic ovary syndrome (namely: hyperinsulinemic insulin resistance, hyperandrogenemia, and obesity), it has been hypothesized that decreasing hyperinsulinemic insulin resistance with metformin during pregnancy in women with the disorder would reduce the rate of early pregnancy loss.<sup>7</sup>

Results from the DHEA plus metformin group on animal studies showed a pattern of ovarian PGE synthesis similar to that of the control groups.<sup>28</sup> Although the action of this biguanide on improving lipid metabolism has been widely reported,<sup>42</sup> the data represent the first evidence that metformin modulates the production of ovarian prostaglandins.<sup>28</sup> Thus, the use of metformin is becoming increasingly accepted and widespread though a complete understanding of the mechanisms involved is not clear. However, metformin has been shown to be useful in the reduction of insulin resistance by restoring insulin sensitivity.<sup>41,43</sup> In addition, it has been demonstrated that metformin is able, either directly or indirectly, to regulate

ovarian steroidogenesis.<sup>44</sup> Moreover metformin enhances the tyrosine kinase activity of the insulin receptor by modulating the plasma cell differentiation antigen (PC-1).<sup>45</sup> It also improves insulin sensitivity, lower serum LH, total and free testosterone concentrations and causes an elevation in serum follicle-stimulation hormone and sex hormone binding globulin.<sup>6</sup>

Hyperinsulinemic insulin resistance contributes to early pregnancy loss in the syndrome, and that decreasing hyperinsulinemic insulin resistance with metformin during pregnancy would reduce the rate of early pregnancy loss. Metformin administration during pregnancy reduces first trimester pregnancy loss in women with the polycystic ovary.<sup>7</sup> Study has shown that decreasing hyperinsulinemic insulin resistance with metformin in women with the polycystic ovary syndrome, decreases the rate of early pregnancy loss.<sup>7</sup> It has also been shown to reduce systemic luteinising hormone (LH) and PAI-1 levels, both of which have been associated with an increased risk of miscarriage.<sup>6</sup>

Administration of metformin to pregnant women with PCOS throughout pregnancy was associated with a marked and significant reduction in the rate of early pregnancy loss.<sup>7</sup> Metformin, which is being increasingly used as first-line therapy to treat infertility with PCOS, potentially avoids the risks of multiple pregnancy and ovarian hyperstimulation associated with clomifene.<sup>6</sup> Except for a single baby born with achondrodysplasia, metformin was not associated with any adverse fetal outcomes.<sup>7</sup>

In a metaanalysis of 13 randomised controlled trials, metformin was shown to be effective in achieving ovulation in women with PCOS, with an odds ratio of 3.88 (95% CI 2.25–6.69) for metformin compared with placebo and 4.41 (95% CI 2.37–8.22) for metformin and clomifene compared with clomifene alone.<sup>6</sup> The addition of metformin to clomifene improved ovulation and pregnancy rates in obese women with PCOS.<sup>46,47</sup> This effect was also seen when metformin was added to clomifene in women who were clomifene resistant.<sup>48</sup> Metformin can theoretically be offered as a firstline drug to all women (obese and non-obese) with anovulatory infertility due to PCOS who have been trying to conceive for a year or more.<sup>6</sup> First-line use of metformin could also potentially avoid the risks and costs associated with second- and third-line therapies for women with PCOS who are infertile, such as gonadotrophin ovulation induction, laparoscopic ovarian drilling and *in vitro* fertilisation. However, although there is evidence from a systematic review supporting the first-line use of metformin in inducing ovulation, there is an absence of

robust data indicating the true live birth rates in women with PCOS when metformin is used as a first-line treatment.<sup>51</sup>

## CONCLUSIONS

PCOS is characterized by insulin resistance, hyperandrogenemia, hirsutism, oligo- or amenorrhea and anovulation. It is one of the most common endocrinological diseases in premenopausal women. Hyperinsulinemic insulin resistance is the main feature of PCOS. Both hyperinsulinemia and hyperandrogenism play a pathogenic role in PCOS and are responsible for anovulation. If

women with PCOS achieve conception, they are in the increased risk for miscarriage in the first trimester. Moreover, hyperinsulinemic insulin resistance in PCOS impairs folliculogenesis and affects follicular development which has negative impact on fertility. For these reasons, insulin-sensitizing drugs such as metformin (biaguinide) and troglitazone (thiazolidinedione) can improve the rate of spontaneous ovulation. Metformin and the troglitazone in PCOS have provided encouragement that improvement of insulin sensitivity and consequent lowering of circulating insulin levels by these agents may be of therapeutic value in the management of both anovulation and hirsutism.

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