Metformin treatment in patients with polycystic ovary syndrome undergoing in vitro fertilization: a prospective randomized trial

In the present study, we investigated the impact of metformin therapy on in vitro fertilization/intracytoplasmic sperm injection (IVF/ICSI) outcomes in patients with polycystic ovary syndrome (PCOS). Metformin does not lead to any improvement in IVF/ICSI outcomes among patients with PCOS. (Fertil Steril® 2005;84: 798–801. ©2005 by American Society for Reproductive Medicine.)

Polycystic ovary syndrome (PCOS) is the most common cause of anovulatory female infertility (1). A great number of obese and lean women with PCOS have some degree of insulin resistance (2). Insulin resistance in patients with PCOS undergoing infertility treatment might lead to higher doses of gonadotropins administered, higher cancellation rates due to risk of ovarian hyperstimulation, and lower fertilization rates (3, 4).

We hypothesized that the insulin-sensitizing drug metformin, which reduces hyperinsulinemia, insulin resistance, and secretion by increasing insulin sensitivity, might improve the results of IVF or intracytoplasmic sperm injection (ICSI) (5). To our knowledge, there are only three studies in the literature evaluating the effects of metformin on IVF/ICSI cycles, and they report conflicting results (6–8). In this prospective, randomized, double-blind, placebo-controlled study, we investigated the effects of metformin therapy on ICSI outcome in patients with PCOS.

The current study included 110 infertile women with PCOS who were referred to the Centrum IVF Clinic because of failure to conceive by conventional therapies. The study was approved by the local ethics committee, and informed consent was obtained from all participants. All patients had oligomenorrhea or amenorrhea since menarche as a surrogate for oligo-anovulaton and also had at least one of the criteria of hyperandrogenism, including a hirsutism score of >7 [according to Ferriman and Gallway (9)] and/or elevated serum levels of T (≥3.15 nmol/L). All other causes of hyperandrogenism were ruled out before diagnosis of PCOS. Only patients aged <40 years and having PCOS without concomitant causes of infertility and those undergoing first IVF/ICSI attempts were included in the study. The exclusion criteria were previous treatments with hormonal medications and insulin-lowering agents within 3 months.

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Surrogate insulin resistance was defined as a ratio of fasting serum levels of glucose to insulin of <4.5 (10). On the basis of insulin resistance, a heterogeneous group was obtained, hence metformin therapy was started independent from the assessment of insulin resistance. For the randomization process, women were given an envelope containing either metformin (Glucophage Retard 850 mg; Ilsan-Iltaş Pharmaceuticals, Istanbul, Turkey) or placebo (identical to metformin capsules), according to the code provided by computer-generated randomization in blocks. Both patients and investigators were blinded to the content of tablets. Patients were instructed to take a capsule twice or three times daily, according to body mass index (BMI) <28 kg/m^2 or BMI ≥28 kg/m^2 for 8 weeks before their first ICSI cycles, through the luteal phase, and until a positive pregnancy test.

All patients received a standard long protocol of pituitary suppression with GnRH agonist (triptorelin 0.1 mg [Decapeptyl®, Ferring, Kiel, Germany]), followed by administration of recombinant FSH (Gonal F, Serono, Istanbul, Turkey) with a standard initial dose of 2–4 ampules (150–300 IU FSH) per day. Transvaginal follicular aspiration was performed 36 hours after administration of 10,000 IU hCG (Pregnyl, Organon, Istanbul, Turkey). Human chorionic gonadotropin was administered with the detection of a minimum of three leading follicles of ≥18 mm mean diameter and serum levels of E_2 <5,500 pg/mL. All patients were offered ICSI, owing to the poor fertilization rate experienced with IVF in patients with PCOS (11). A maximum number of three embryos were transferred routinely on the third day. Embryo transfer was performed routinely on the third day after a selective assisted hatching procedure with laser. An assisted hatching procedure was applied with laser (Fertilase®, MTM Medical Technologies, Montreux, Switzerland) (n = 16) when the patient was aged >35 years (n = 10), when the zona pellucida was considered to be thick (>17 μm) (n = 3), or when abnormally shaped zona (n = 1), excessive fragmenta-

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TABLE 1

Baseline characteristics, ovarian response, and pregnancy rate in placebo and metformin groups.

	Placebo (n = 55)	Metformin (n = 53)	*P*
Age (y), mean ± SD	29.76 ± 5.3	29.3 ± 3.9	.625
Duration of infertility (y)	7 (2–22)	7 (2–15)	.63
BMI (kg/m^2)	23.5 (19–34)	25 (19–41)	.97
FSH (IU/L)	5.4 (2.6–11.2)	5.4 (3.1–10.6)	.45
LH (IU/L)	6.1 (1.5–23)	5.2 (2.8–22.3)	.32
E_2 (pg/mL)	59 (18–143)	52 (22–123)	.147
Total T (0.35–3.15 nmol/L)	3.1 (2.4–3.9)	3.1 (2.5–3.9)	.646
PRL (62–1,075 pmol/L)	471 (219–842)	472 (224–866)	.123
DHEAS (0.9–11.2 μmol/L)	5.3 (1.4–8.5)	4.4 (1.6–8.6)	.341
Fasting glucose (mg/dL), mean ± SD	87.2 ± 16.3	81 ± 6.12	.813
Fasting insulin (IU/L)	14 (9.9–23)	14 (9.9–31)	.105
Glucose/insulin ratio	6 (3–10)	6 (2.4–8.8)	.81
Antral follicle(s)	10 (6–16)	11 (7–18)	.23
Duration of ovarian stimulation (d)	9 (7–12)	9 (6–13)	.85
Total amount of FSH (IU)	2,050 (950–5,100)	2,025 (675–5,300)	.31
No. of follicles (10–16 mm)	12 (6–14)	10 (8–13)	.46
No. of follicles (≥17 mm)	7 (3–15)	8 (3–16)	.59
Peak E_2 (pg/mL)	3,946 (1,069–5,592)	3,615 (1,095–5,439)	.97
No. of retrieved oocytes	17 (8–32)	17 (4–35)	.35
No. of metaphase II oocytes	12 (1–30)	13 (2–35)	.49
No. of metaphase I oocytes	5 (0–23)	5 (0–21)	.53
Fertilization rate (%), mean ± SD	71.7 ± 17	73.6 ± 15.5	.56
No. of embryos transferred	2 (1–3)	2 (1–3)	.36
Clinical pregnancy rate, % (n)	40 (22/55)	30.2 (16/53)	.6
Biochemical pregnancy rate, % (n)	5.5 (3/55)	5.6 (3/53)	.75
Abortion rate, % (n)	5.5 (3/55)	5.6 (3/53)	.75
Assisted hatching, % (n)	15 (8/55)	15 (8/53)	.69

Note: Data are presented as mean ± SD or median (range). $P<.05$ considered significant.

Önalan. Metformin and ICSI outcome. Fertil Steril 2005.

tion (n = 1), or slowly developing embryos (n = 1) were noted.

Insulin, LH, FSH, E_2, and PRL levels were assessed with an autoanalyzer (AXSYM; Abbott Laboratories, Abbott Park, IL), with the microparticle enzyme immunoassay method. Total T and DHEAS levels were detected with an autoanalyzer (Immulite One; BIODPC, Los Angeles, CA), with the chemiluminescent method. Statistical analysis was performed with Student *t*, Mann-Whitney *U*, χ^2, and Fisher exact tests, as appropriate. Data are reported as mean ± SD or median (range). A *P* value of <.05 was considered significant.

Only two patients abandoned the treatment cycles. The remaining 108 patients were divided into placebo (n = 55) and metformin (n = 53) groups. There were no statistically significant differences between the two groups regarding baseline characteristics, duration of stimulation, total dose of FSH, number of follicles 10–16 mm and ≥17 mm in diameter, serum peak levels of E_2 on the day of hCG injection, number of retrieved metaphase I and II oocytes, fertilization rate, number of transferred embryos, and total and clinical pregnancy rates (all $P\geq.05$) (Table 1). Patients with a glucose/insulin ratio ≥4.5 had similar measures of outcome in both placebo and metformin groups.

In the metformin group, compared with the placebo group, patients with a glucose/insulin ratio <4.5 had lower day-3 serum levels of LH (4.8 IU/mL [range, 2.8–8.1 IU/mL] vs. 6.8 IU/mL [5–13.1 IU/mL], $P=.04$) and E_2 (49 pg/mL [37–68 pg/mL] vs. 68.5 pg/mL [54–88 pg/mL], $P=.002$) and lower numbers of follicles ≥17 mm in diameter (5.5 [3–8] vs. 8 [6–13], $P=.01$). Patients with a BMI <28 had similar measures of outcome in both placebo and metformin groups. Patients with a BMI ≥28 in the metformin group had lower serum levels of LH (4.2 IU/mL [2.8–8.1 IU/mL] vs. 7.5 IU/mL [5.1–13.1 IU/mL], $P=.006$), increased numbers of antral follicles (12 [9–15] vs. 8.5 [7–14], $P=.015$), and increased numbers of follicles ≥17

mm in diameter (9 [6–13] vs. 5.5 [3–10], $P=.046$), compared with those in the placebo group. We did not observe any failure of fertilization. There were four and three cases of mild ovarian hyperstimulation syndrome in the placebo and metformin groups, respectively ($P\geq.05$).

In the present study, metformin had no effect on duration of stimulation, total dose of FSH, number of follicles 10–16 mm and ≥17 mm in diameter, serum peak levels of E_2 on the day of hCG injection, number of retrieved metaphase I and II oocytes, fertilization rate, number of transferred embryos, and total and clinical pregnancy rates. All of these results, except for improved pregnancy rates in patients with normal body weight, are consistent with those from the prospective, randomized, placebo-controlled study by Kjotrod et al. (6). The main differences between the Kjotrod et al. study and ours have to do with indication for IVF/ICSI, dose and duration of metformin treatments, and the presence of concomitant infertility causes of patients. In the Kjotrod et al. (6) study, the metformin group received a dose of 500 mg twice daily for 16 weeks. In the present study, patients had no concomitant infertility causes, and metformin doses of 850 mg twice or three times daily were given, according to patient BMI, for 8 weeks. A limitation of our study is the shorter follow-up period for making comments on ongoing pregnancy rates. Although the dose and duration of metformin might be sufficient to make a difference, changes in insulin concentrations and body weight were not recorded to ascertain an effect at the end of the study.

In the first study on the effects of metformin on IVF/ICSI cycles in PCOS, by Stadtmauer et al. (7), 60 IVF cycles were retrospectively analyzed. Metformin was started independently from insulin resistance with doses of 500 mg twice or three times daily, according to the patients' BMI (<28 kg/m^2 or ≥28 kg/m^2). Intracytoplasmic sperm injection was performed for all patients with PCOS undergoing IVF, owing to the authors' past experience of poor fertilization rates in patients with PCOS. They concluded that metformin might improve IVF outcomes in patients with clomiphene-resistant PCOS.

Another study, a small, open-label cross-over randomized trial demonstrated that administration of metformin as 1,500 mg/day for 3 weeks in only 17 insulin-resistant patients with PCOS increased the number of oocytes retrieved in insulin-resistant, obese patients with PCOS and increased the number of oocytes collected without altering gonadotropin requirements (8).

Poor oocyte quality resulting from extrinsic or intrinsic factors in PCOS (11, 12) might lead to decreased fertilization rates (13) and increased fertilization failure (14) and abortion rates (15). ICSI offers the advantage of bypassing the barriers that might be of oocyte origin could overcome the fertilization rate and high complete fertilization rate in PCOS patient during their IVF cycles (16). Moreover, ICSI might be the cause of the low abortion rate seen in this study (17).

Rather than being the primary cause of anovulation in PCOS, hyperinsulinism and/or insulin resistance might be viewed as a "second hit" that nonspecifically worsens follicular abnormalities (18). This second-hit effect of insulin might be counteracted with long-term GnRH agonist treatment, suppressing endogenous LH secretion, and purified FSH preparation. Furthermore, long-standing hyperinsulinemia might down-regulate insulin receptors in the ovary and as a result reduce insulin's effect on granulosa cells (19, 20). The current study contributes additional data that metformin provides no improvement in IVF/ICSI outcomes in patients with PCOS.

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