Recombinant Human Follicle-Stimulating Hormone for Ovulation Induction in Polycystic Ovary Syndrome: A Prospective, Randomized Trial Of Two Starting Doses in a Chronic Low-Dose Step-up Protocol

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Purpose: The aim was to compare the follicular response to 37.5 and 50 IU of recombinant follicle-stimulating hormone (FSH) as starting doses for ovulation induction in patients with polycystic ovary syndrome (PCOS).

Methods: Prospective, randomized, crossover study including 15 women with clomiphene citrate-resistant chronic anovulatory infertility. Patients were treated with subcutaneous recombinant FSH at starting doses of 37.5 IU and 50 IU, respectively, according to a low-dose step-up protocol. Each woman received both treatments, in a randomized order, with an interval of ≥ 1 month between treatments.

Results: All treatment cycles were ovulatory after an appropriate follicular response and hormone levels were similar with both treatments, although the total quantity of FSH required and the mean daily dose required to induce identical follicular development were significantly lower with a starting dose of 37.5 IU FSH. The mean duration of treatment to achieve ovulation was approximately 13 days with both treatments but treatment periods ≥ 20 days were required in some patients.

Conclusions: In women with PCOS, a starting dose of 37.5 IU recombinant FSH may be adequate to induce follicular growth. However, the use of low starting doses may result

in some cases in increased treatment periods and need for monitoring.

KEY WORDS: Follicle-stimulating hormone; ovulation induction; polycystic ovary syndrome; recombinant human FSH.

INTRODUCTION

As a result of the growth in understanding of folliculogenesis that has occurred in recent years, a variety of approaches are now available for the restoration of ovulation in infertile women with irregular menses or amenorrhea (1,2). The most common diagnosis in infertile anovulatory women is polycystic ovary syndrome (PCOS). Antiestrogens such as clomiphene citrate are the first line of treatment for ovulation induction in PCOS, but approximately 20% of patients are clomiphene resistant. Gonadotropin therapy is normally used in clomiphene-resistant patients and in those patients who do not conceive in response to repeated courses of clomiphene citrate (1,2).

Recent studies in patients with PCOS have shown that the low-dose step-up regimen—standard 75 IU follicle-stimulating hormone (FSH) starting dose with the first 50% dose increment (37.5 IU) if necessary after 14 days and every 7 days thereafter—reduces the risk of multiple follicular development while maintaining satisfactory pregnancy rates (3). Although urinary gonadotropins have proved to be useful for ovulation induction in PCOS patients (4,5), recombinant FSH has become the treatment of

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choice in such women due to its higher purity, consistency, and efficacy (6,7). Two recombinant FSH preparations currently are available: follitropin alfa (Gonal-F® 37.5, 75, and 150 IU; Ares-Serono International S.A., Geneva, Switzerland), and follitropin beta (Puregon® 50, 100, and 150 IU; NV Organon, Oss, the Netherlands). Both follitropins are structurally identical to native FSH; the alfa and beta nomenclature of these products merely reflects that follitropin alfa (Gonal-F) was introduced first on the market and does not refer to the α and β subunits of FSH (8). In fact, both recombinant FSH preparations are more efficient than urinary FSH in inducing follicular development in PCOS patients (6,7).

A recent pilot study of 11 PCOS patients starting with 50 IU of recombinant FSH in a low-dose step-up protocol where initial dose was increased by 100% after 7 days led to an ovulation rate of 54%, but it was associated with a 36% cancellation rate due to overstimulation and the duration of treatment (mean 20.3 days, range 11 to 35) was unusually long in a low-dose step-up protocol (9). The present study was undertaken to compare follicular response to starting doses of 37.5 and 50 IU of recombinant FSH for ovulation induction in PCOS patients according to the principles of chronic low-dose step-up therapy.

MATERIALS AND METHODS

Patients

A total of 15 women with anovulatory infertility due to PCOS were included in the present study. The mean ($\pm$ SEM) age of the patients was 30.4 ± 0.9 years and the average duration of infertility was 4.4 ± 1.3 years. All patients presented with oligomenorrhea or amenorrhea and all had ultrasonographic evidence of polycystic ovaries (10). The mean basal Luteinizing (LH)–FSH ratio was 2.49 ± 0.16 , the mean basal androstendione level 286 ± 14.6 ng/dl, and the mean body mass index was 24.6 ± 0.46 . Endogenous estrogen activity was present in these patients, as shown by mean basal estradiol levels of 92.1 ± 11.3 pg/ml and a positive response to a progestin challenge test (normal withdrawal bleeding after treatment with oral medroxyprogesterone acetate, 10 mg daily for 5 days). All patients had either failed to ovulate after clomiphene citrate treatment or had not conceived after a minimum of three ovulatory cycles on this treatment at doses of 200 mg/day for 5 days.

Study Protocol

Each woman received both 37.5 and 50 IU of subcutaneous recombinant FSH as starting doses for ovulation induction in a low-dose step-up protocol, in a randomized order, with an interval of at least 1 month between treatment cycles. We used gonadotropin preparations in the form of follitropin alfa (Gonal-F®, Ares-Serono International S.A., in group 37.5) and follitropin beta (Puregon®, NV Organon, in group 50), since both follitropins are structurally identical to native FSH and behave similarly in an *in vivo* bioassay (8). Treatment was commenced on day 3 of a spontaneous cycle or of induced uterine bleeding after a baseline ultrasound examination. After 14 days, initial doses were increased by increments of 37.5 IU and 50 IU, respectively, if there was no evidence of an ovarian response on ultrasound (i.e., no follicle >10 mm in diameter). Further dose adjustments were performed if necessary after a period of 7 days. Once ultrasound evidence of an ovarian response was obtained, the current dose (i.e., the threshold dose) was continued until the mean follicular diameter was greater than 17 mm. Human chorionic gonadotropin (hCG), 10,000 IU intramuscularly (Profasi®, Serono S.A.) was then given to induce ovulation.

Endocrine Assessment

Hormones in serum were measured using commercially available kits as reported previously (6). Concentrations of both estradiol and inhibin A were measured retrospectively on the day of hCG injection in all treatment cycles studied. Estradiol was measured by a competitive immunoenzymatic assay (Immuno 1, Bayer, Tarrytown, NY). The sensitivity of the assay was 10 pg/ml and the interassay coefficient of variation was 5%. Inhibin A measurements were performed by a solid phase sandwich enzyme-linked immunosorbant assay (ELISA) carried out in microtitre plates (Serotec Ltd., Oxford, UK) (11,12). The assay sensitivity was 2 pg/ml and the within-plate coefficient of variation was less than 7%. The cross-reactivity with activin A was less than 0.1%.

Ultrasonic scans were performed with a 5 mHz vaginal transducer attached to an Aloka sector scanner (model SSD-620, Aloka, Tokyo, Japan).

Statistical Analyses

Data were analyzed by SPSS statistical software using the Wilcoxon matched-pairs signed-ranks test

and the χ^2 test as appropriate. Results are expressed as means and standard errors (SEMs). *P* values below 0.05 were considered significant.

RESULTS

A total of 22 eligible patients were entered into the study. Twelve were randomly allocated to treatment group 1 (starting dose 37.5 IU) first and 10 to group 2 (starting dose 50 IU). There were three single pregnancies in each treatment group during the first cycle of therapy. One additional patient was lost to follow-up after her first ovulatory but nonconceptual treatment cycle in group 2. Thus, in total, the responses of 15 patients to both starting doses of recombinant FSH were evaluated.

All treatment cycles were ovulatory, as judged by ultrasonographic data, basal body temperature measurements, and the length of the luteal phase. The results in the two treatment groups are summarized in Table I. The mean duration of treatment (with 95% confidence limits) was 13.5 days (5–22 days) in group 1 and 12.5 days (6–20 days) in group 2. Both the total quantity of FSH required and the mean threshold dose required to induce follicular development were significantly lower in group 1 than in group 2. Estradiol and inhibin A serum concentrations on the day of hCG injection were similar with the two treatments.

A total of 10 cycles (67%) in group 1 were unifollicular (i.e., only one leading follicle >17 mm and no secondary follicles) on the day of hCG injection, compared with nine cycles (60%) in group 2. Five bifollicular cycles (33% of treatment cycles), in which a secondary follicle measuring 12–17 mm was present, occurred in group 1, and six (40%) in group 2.

DISCUSSION

The results of this study indicate that similar outcomes were achieved when ovulation induction is performed according to a low-dose step-up protocol with starting doses of 37.5 IU and 50 IU recombinant FSH in patients with PCOS. In this study, each patient acted as her own control; women who became pregnant in their first cycle, and thus did not receive the second treatment, were not included in the analysis of ovarian response. This design has previously been used by our group (6,13,14) and by others (15,16), and we consider it an appropriate design when ovarian response and hormone concentrations but not pregnancy rates are the outcomes of interest.

The most important principle in ovulation induction is to provide as close as possible a physiological restoration of cyclical ovarian function; in particular, the aim should be to achieve the ovulation of a single follicle. Multiple follicular development is a characteristic complication of ovulation induction with exogenous gonadotropins, particularly in women with PCOS, who are very sensitive to gonadotropin stimulation (1). Indeed, approximately 75% of iatrogenic multiple pregnancies result from ovulation induction, while the remaining 25% result from assisted reproductive techniques (17–21). In addition, PCOS is a major risk factor for ovarian hyperstimulation syndrome (21). Low-dose treatment regimens have been used in attempts to prevent the risks of ovarian hyperstimulation syndrome and multiple gestation in patients with PCOS (1,2).

The chronic low-dose step-up regimen is based on the threshold concept suggested by Brown and co-workers (22,23), according to which the development of multiple follicles results from failure to reproduce the precise gonadotropin dosage requirements

Table I. Ovarian Response and FSH Requirement in the Two Treatment Groups

Parameter	Group 1 ^a	Group 2	<i>P</i>
FSH required			
Days of treatment	13.5 ± 0.9	12.47 ± 0.72	NS
IU	522.5 ± 45.9	623.3 ± 35.8	<0.05
Threshold dose (IU)	40.0 ± 2.5	50 ± 0	<0.005
No. of follicles on hCG day			
>10–13 mm	0.33 ± 0.16	0.2 ± 0.14	NS
>13–17 mm	0.27 ± 0.12	0.53 ± 0.17	NS
>17 mm	1.13 ± 0.09	1.13 ± 0.09	NS
Total	1.7 ± 0.24	1.7 ± 0.24	NS
E ₂ on hCG day (pg/ml)	286 ± 22.7	296 ± 24.7	NS
Inhibin A on hCG day (pg/ml)	52.2 ± 3.7	50.1 ± 4.5	NS

^a Values are means ± SE; NS, not significant.

that are normally maintained by feedback regulation. These authors established that initiation of follicular growth requires only a 10–30% increment in the dose of exogenous FSH, and thus recommended small, stepwise increments of FSH at 5-day intervals. In practice, however, the results of this approach were complicated by an overstimulation rate of 3% and a 26% incidence of multiple pregnancies (23). The failure to achieve a high proportion of unifollicular cycles has been related both to an excessive starting dose and an inadequate treatment duration before increasing the dose (24). A starting dose of urinary gonadotropin of 0.7 ampule (52 IU) per day, which is maintained for up to 14 days, has been used in one clinical practice for ovulation induction in PCOS patients, although there has been no reduction in cancellation due to overresponse (5,24).

The present study investigated the clinical usefulness of starting doses of recombinant FSH below 75 IU for ovulation induction in patients with PCOS. It is noteworthy that only unifollicular or bifollicular cycles were obtained (including a high rate of monofollicular development) with a relatively short period of gonadotropin therapy: the mean duration of treatment needed to achieve ovulation was approximately 13 days with both treatments. However, the 95% confidence intervals indicate that treatment periods of more than 20 days may be required in some patients. Thus, the use of low starting doses may result in some cases in an increased need for monitoring, compared with the standard 75 IU starting dose.

The fact that the treatment cycles were almost identical with respect to follicular size and number and serum hormone concentrations on the day of hCG administration indicates that the stimulation procedures were carried out in a similar way with both treatments. However, the total amount of FSH required and the mean threshold dose were both significantly lower in group 1 than in group 2. Since the two recombinant FSH preparations are both identical to native FSH, they would be expected to produce similar effects on follicular development (8). Therefore, the better performance in group 1 in terms of the amount of FSH required may be due to a more appropriate starting dose and treatment regimen.

As noted above, each woman acted as her own control. This is important since PCOS is a heterogeneous condition with distinct endocrine features and the FSH threshold varies for individual patients (25,26). Therefore, the use of the same treatment protocol with different starting doses of recombinant FSH in the same PCOS patient, as in the present study, provides valuable clinical information.

In conclusion, the results of this study suggest that the use of 37.5 IU recombinant FSH as starting dose in a chronic low-dose step-up regimen is associated with a lower FSH requirement and a lower threshold dose compared with a starting dose of 50 IU. This starting dose therefore may offer a useful therapeutic option in women with PCOS who already have experienced ovarian hyperstimulation with the standard 75 IU dose. Further studies are needed to confirm the impact of this approach in terms of pregnancy rates achieved.

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