Comparison of luteal phase profile in gonadotrophin stimulated cycles with or without a gonadotrophin-releasing hormone antagonist

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BACKGROUND: The aim of our study was to explore luteal phase hormone profiles in gonadotrophin-stimulated cycles with or without gonadotrophin-releasing hormone (GnRH) antagonist therapy during intrauterine insemination (IUI). Forty-one infertile couples were recruited in this randomized clinical study. METHODS: The 19 patients included in group A were treated for 21 cycles with recombinant FSH 150 IU/day starting from day 3 of the cycle and with the GnRH antagonist cetrorelix at the dose of 0.25 mg/day starting from the day in which a follicle with a mean diameter of ≥14 mm was seen at ultrasound scan. Cetrorelix was administered until human chorionic gonadotrophin (HCG) administration. The 22 patients included in group B were administered recombinant FSH alone at the same dosage for 27 cycles. RESULTS: The two treatment groups showed a similar increase in progesterone concentration during the luteal phase. In the mid-luteal phase (day 6 after HCG), oestradiol concentrations in group B were significantly higher compared with group A (P < 0.05) but the oestradiol:progesterone ratio was similar in the two groups. Serum LH was completely suppressed during the follicular phase only in group A, concomitantly with GnRH antagonist administration. A total of six pregnancies, all ongoing, were achieved (14.3% per patient and 12.2% per cycle), equally distributed in group A and in group B. CONCLUSION: GnRH antagonists can be safely administered in gonadotrophin-stimulated IUI cycles without luteal phase supplementation because no deleterious effects of GnRH antagonist administration were noted on luteal progesterone concentration or on the duration of the luteal phase.

Keywords: GnRH antagonist/infertility/intrauterine insemination/luteal phase

Introduction

Gonadotrophin-releasing hormone (GnRH) antagonists have been recently introduced in clinical practice for ovarian stimulation in assisted reproduction cycles. The efficacy of these agents in preventing premature LH surge has been demonstrated (Diedrich et al., 1994; Olivennes et al., 1994, 1995) and an advantageous alternative to GnRH agonists in superovulation cycles for assisted reproduction techniques (ART) (Albano et al., 2000; The European Orgalutran® Study Group et al., 2000; Olivennes et al., 2000) but data on the luteal phase during their use are scanty.

Because of the presence on the ovary of GnRH receptors (Clayton and Catt, 1981), and because of evidence showing a negative effect of GnRH agonist on steroidogenesis by the granulosa cells (Tureck et al., 1982) and by the corpus luteum (Casper and Yen, 1979), an inhibitory effect also of GnRH antagonist on steroidogenesis may be postulated. Moreover, an in-vitro study demonstrated an inhibitory effect of GnRH antagonist on gonadal steroid secretion (Spona et al., 1985) and another animal study reported an impaired function of the corpus luteum with reduction of progesterone production in pregnant rats treated with GnRH agonist (Srirdaran, 1987). Evaluating studies in humans, a normal luteal phase was observed in 10 normally ovulating women who underwent GnRH antagonist (Nal-Glu) administration to prevent LH surge and ovulation in natural cycles (Ditkoff et al., 1991). Normal progesterone concentrations in the luteal phase were also recorded after clomiphene citrate and human menopausal gonadotrophin (HMG) plus Nal-Glu administration (Frydman et al., 1991) or after gonadotrophin and cetrorelix stimulation for ART cycles (Diedrich et al., 1994; Felberbaum et al., 1996). Conversely, a reduction of oestradiol concentration in the granulosa cells and a lower in-vivo concentration of oestradiol on the HCG day were recorded in women treated with GnRH antagonist compared with GnRH agonist-treated patients (Minaretzis et al., 1995). Albano et al. found a short luteal phase and low serum oestradiol and progesterone concentration in three out of six patients who did not receive luteal phase hormonal supplementation after stimulation with HMG and cetrorelix for IVF or intracytoplasmic sperm
injection (ICSI) (Albano et al., 1998). In a subsequent paper, the same authors demonstrated that different doses of GnRH antagonist had no impact on the luteal phase of IVF/ICSI cycles when hormonal support was given. (Albano et al., 1999). Luteal phase hormonal supplementation is commonly administered in IVF/ICSI cycles (Edwards et al., 1980), and it is a confounding factor in evaluating any negative influence of GnRH antagonist on corpus luteum function: the aim of this randomized study was therefore to evaluate the luteal phase without any hormonal support, in gonadotrophin and GnRH antagonist stimulated cycles compared with gonadotrophin alone, in an intrauterine insemination (IUI) programme.

Materials and methods
Between March and July 2000, 41 infertile patients were recruited for the IUI programme at the Infertility Unit, Department of Obstetrics and Gynaecology of the University of Milan. Patients included suffered from unexplained or mild male factor infertility. Inclusion criteria were women ≤38 years of age, primary or secondary infertility lasting for at least 24 months, body mass index between 19 and 25 m²/kg, normal prolactin (PRL) levels, normal thyroid function, normal uterine cavity and bilateral tubal patency assessed by hysterosalpingography and/or laparoscopy with chromosalpingography and hysteroscopy. Patients with polycystic appearance of the ovaries at ultrasound according to the defined criteria (Adams et al., 1986) were excluded from the study.

After inclusion in the study, patients underwent an ultrasound scan on day 3 of the cycle and were randomized by means of a computer generated list into two groups. Patients in group A were administered recombinant (r)FSH (Gonal-F, Serono, Geneva, Switzerland) 150 IU/day starting from day 3 of the cycle until HCG administration, individualizing the dose according to ovarian response at ultrasound, and GnRH antagonist cetrorelix (Cetrodite, Serono) at the dose of 0.25 mg per day (multiple dose protocol) starting from the day in which a follicle ≥14 mm in mean diameter was visualized at ultrasound scan. Cetrorelix was administered until HCG administration (leading follicle with a mean diameter of 18–20 mm). Patients included in group B were administered recombinant FSH alone at the same dosage and HCG was administered when the leading follicle reached a mean diameter of 18 mm. Ovarian stimulation in both groups was monitored by daily ultrasound scans starting from the day 8 of the cycle (day 5 of ovarian stimulation). Blood sampling for oestradiol, progesterone and LH assessment was performed on the days in which at least one follicle with a mean diameter of 16 mm was visualized. Hormonal assessment was repeated daily until HCG administration and on alternate days during the luteal phase. Luteal phase duration was calculated from the day of HCG administration to the first day of the subsequent menstrual cycle or from the day before the evidence of positive urinary LH test. All hormonal assessments were obtained retrospectively and did not influence clinical decisions regarding ongoing cycles which were made on the basis of ultrasound only. In group B evidence of positive urinary LH test lead to immediate HCG administration; in these cycles IUI was then performed the subsequent morning. IUI was performed as previously described (Ragni et al., 1999). No luteal phase supplementation was administered in any of the patients. IUI insemination was not performed if there were more than six or two or less follicles with a mean diameter ≥15 mm, in order to maximize pregnancy chances and to reduce the risk of multiple pregnancy.

Data were analysed using χ² test with Yates’ correction, Student’s t-test and Mann–Whitney test (SPSS/Windows, Chicago, IL, USA)

Results
Nineteen patients were randomized in group A and 22 in group B and a total of 48 IUI cycles were initially considered. Clinical characteristics of patients and data regarding treatment cycles are reported in Table I. In group A the mean starting day of GnRH antagonist therapy was day 9 (range 6–12) and a mean of 3.6 ampoules were administered (range 3–6). In group B patients started with LH urinary stick on day 8 (range 6–10) and the mean number of sticks used was 4.2 (range 1–7).

After ovarian stimulation, neither IUI nor luteal phase evaluations were performed in 16 cycles due to low response (32.6%) (less than two follicles ≥15 mm in mean diameter: 3 cycles in group A, 2 in group B) or because of hyper-response (more than six follicles ≥15 mm in mean diameter: 4 cycles in group A, 7 in group B).

Four cases of progesterone rise (serum values >1.5 ng/ml) on the day of HCG administration were recorded, all in group B (30.8% of premature luteinization). In three cycles in group B, LH concentrations before HCG administration were >10 IU/l, despite progesterone values <1.5 ng/ml. When a positive urinary LH test was evident in these cycles, HCG was immediately administered and IUI performed the subsequent morning. All these seven cycles were excluded from the final evaluation of the luteal phase. In group A only one patient showed an increase in serum LH (4.7 IU/l after the first cetrorelix administration and 13.6 IU/l on the day of HCG administration) despite regular GnRH antagonist administration: nevertheless serum progesterone values were constantly low (0.5 ng/ml on the day of HCG). In this particular cycle IUI was performed and the luteal phase hormonal profile appeared normal but we prefer to exclude these hormonal data from the final evaluation. Finally, the hormonal profile of 13 cycles in group A and of 11 cycles in group B were evaluated. Progesterone, oestradiol and LH serum values in the luteal phase are shown in Figures 1, 2, and 3. Progesterone rise during the luteal phase was similar in the two groups. Oestradiol showed two different peaks: one on the day of HCG administration and the other six days after HCG administration. In the mid-luteal phase (day 6 after HCG), oestradiol levels in group B were significantly higher compared with group A (P < 0.05) but the oestradiol:progesterone ratio was similar in the two groups (data not shown). All other differences in hormonal concentrations in the two groups were not statistically significant. Serum LH was suppressed during the follicular phase in group A (except in the above mentioned patient), concomitantly with GnRH antagonist administration. In analysed cycles in group B, LH values were >2 IU/l on day –2 and –1. Mean duration of the luteal phase was 13.6 days (range 10–15) in group A and 11.9 (9–14) in group B (considering the luteal phase starting from the day of HCG administration or from the day of LH rise) (P < 0.05). Luteal phase duration was ≥12 days in 90.9% of patients in group A and in 60% of patients in group B. A total of six pregnancies was achieved (14.3% per patient and 12.2% per cycle): 3 pregnancies were obtained in group A (2 singleton and one tubal pregnancy) and 3 pregnancies in group B (2 twins and one triplet). All the intrauterine pregnancies are ongoing, the triplet pregnancy reducing spontaneously to twins.
Table I. Clinical characteristics of patients and data regarding treatment cycles in the two treatment groups

<table>
<thead>
<tr>
<th></th>
<th>Group A</th>
<th>Group B</th>
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</thead>
<tbody>
<tr>
<td>No. of patients</td>
<td>19</td>
<td>22</td>
</tr>
<tr>
<td>Mean age [years ± SD (range)]</td>
<td>33 ± 3.5 (26–38)</td>
<td>32.9 ± 3 (28–38)</td>
</tr>
<tr>
<td>No. of cycles</td>
<td>21</td>
<td>27</td>
</tr>
<tr>
<td>No. of cancelled cycles</td>
<td>7</td>
<td>9</td>
</tr>
<tr>
<td>No. of completed cycles</td>
<td>14</td>
<td>18</td>
</tr>
<tr>
<td>No. of evaluated cycles</td>
<td>13</td>
<td>11</td>
</tr>
<tr>
<td>FSH administered [IU ± SD (range)]</td>
<td>1080 ± 236.8 (750–1650)</td>
<td>1054.2 ± 167.6 (900–1350)</td>
</tr>
<tr>
<td>No. of follicles with mean diameter &gt;15 mm [± SD (range)]</td>
<td>2.7 ± 1.1 (2–6)</td>
<td>3.2 ± 1.4 (2–6)</td>
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</table>

Discussion

GnRH antagonists offer advantages compared with GnRH agonists in ovulation-induction protocols for assisted reproduction techniques: a successful LH suppression with an immediate arrest of gonadotrophin secretion may be obtained with a shorter administration course. Especially in IUI cycles, in which GnRH agonists are not frequently used because of the excessive follicular selection they favour and because of the long pre-treatment period required, GnRH antagonists might be useful in order to lower cancellation rates due to premature LH surge. However, it is unclear whether the luteal phase in GnRH antagonist and gonadotrophin cycles should be supplemented.

Although normal luteal phase duration and normal progesterone values in normally ovulatory women undergoing GnRH antagonist treatment to prevent LH surge in a natural cycle have been shown (Ditkoff et al., 1991), there is evidence of vaginal bleeding and low serum oestradiol and progesterone concentrations in the mid-luteal phase in assisted reproduction cycles using antagonists without luteal supplementation (Albano et al., 1996, 1997). The same authors also reported a reduction in the length of luteal phase in three out of six patients who underwent ovarian stimulation with HMG and the GnRH antagonist cetrorelix without luteal phase supplementation (Albano et al., 1998). In another study on five women stimulated using clomiphene citrate and HMG plus the GnRH antagonist Nal-Glu, normal progesterone concentrations in the luteal phase were recorded (Frydman et al., 1991) and high progesterone serum levels in the luteal phase were obtained in assisted reproduction cycles using cetrorelix at a single 3 mg dose and HCG supplementation (Diedrich et al., 1994; Felberbaum et al., 1996). Moreover, GnRH antagonists do not seem to exert a direct luteolytic effect, as demonstrated in vitro on human granulosa–lutein cells (Orthmann et al., 1998), although an in-vivo study in pregnant rats showed a direct inhibitory effect on corpus luteum of the
GnRH antagonist use in IVF/ICSI cycles is commonly administered in IVF/ICSI cycles (Edwards et al., 1980), especially when a GnRH agonist is used. The rationale for luteal phase normalization derives from concerns that the act of follicular aspiration may disrupt a sufficient volume of granulosa cells to impair luteal steroidogenesis (Kreitmam et al., 1981) and from evidence that the incidence of luteal inadequacy is increased in patients who undergone spontaneous ovulation (Kubik et al., 1986). Although no evidence of a real benefit in luteal phase supplementation has been suggested in the past years by meta-analysis (Daya, 1988) or by randomized studies (Belaisch-Allart et al., 1990; Kupferminc et al., 1990), a more recent reviews of literature suggests that luteal phase support improves the pregnancy rates (Soliman et al., 1994). It should be noted that in gonadotrophin-stimulated cycles serum oestradiol levels are higher than normal and a consequent inadequate oestradiol:progesterone ratio during the luteal phase is possible. HCG triggering might also contribute to lower LH concentration in the luteal phase by increasing steroid levels or by a direct pituitary effect. (Demoulin et al., 1991).

In this study we evaluated the luteal phase both in gonadotrophin plus GnRH antagonist stimulated cycles and in cycles stimulated with gonadotrophin alone and without any luteal phase supplementation to avoid any confounding factor. Although a reduction in mid-luteal oestradiol values was present in GnRH antagonist-treated patients, no detrimental effects of the GnRH antagonist were noted on luteal progesterone concentrations or on pregnancy rate. The lower oestradiol concentration could be due to an ovarian effect of GnRH antagonist (Ortmann et al., 2001). Nevertheless, in this study the duration of the luteal phase appears to be even longer in GnRH antagonist cycles.

It appears that the use of a GnRH antagonist in a multiple dose protocol shows the same progesterone profile in the luteal phase compared with gonadotrophin therapy alone in IUI induction cycles. GnRH antagonists may therefore be safely administered to these patients without luteal phase supplementation. Oestradiol values in the follicular phase of these cycles are probably not high enough to induce a relative progesterone deficiency in the luteal phase: this hypothesis explains why luteal supplementation in these cycles is not crucial.

References
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