Modified natural cycle for IVF does not offer a realistic chance of parenthood in poor responders with high day 3 FSH levels, as a last resort prior to oocyte donation

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BACKGROUND: The purpose of this study was to evaluate the use of the modified natural cycle (MNC) for IVF in poor responders as a last resort prior to oocyte donation. METHODS: Thirty-two patients with a regular menstrual cycle, FSH levels on day 3 of the cycle >12 IU/l and one or more failed IVF cycles with five or fewer cumulus–oocyte complexes (COCs) retrieved were included in this prospective study. Recombinant FSH 100 IU and GnRH antagonist 0.25 mg/day were started concomitantly when a follicle with a mean diameter of 14 mm was present at ultrasound. HCG 10 000 IU was administered as soon as the mean follicular diameter was ≥16 mm. RESULTS: Twenty-five out of 78 cycles performed (32.1%) did not result in oocyte retrieval. In nine out of 53 cycles (16.9%) in which oocyte retrieval was performed, no COCs were retrieved. Following fertilization, embryo transfer was performed in 19 out of 44 cycles in which COCs were retrieved (43.2%). No ongoing pregnancy was achieved in 78 MNCs (0.0%; 95% confidence interval 0.0–4.7). CONCLUSIONS: MNC does not offer a realistic chance of parenthood in patients with high levels of FSH on day 3 of the cycle and previous poor response to ovarian stimulation, when offered as a last resort prior to oocyte donation.

Key words: GnRH antagonists/modified natural cycle/poor responders

Introduction

The management of poor responders to ovarian stimulation for IVF has always been a challenge (Tarlatzis et al., 2003). Pregnancy rates in this group of patients remain disappointingly low, despite changes in the dose or the type of gonadotropins used or the form of downregulation applied (Surrey and Schoolcraft, 2000). When oocyte donation is not an option and the desire for conception is still present, the lack of effective treatment alternatives is frustrating for both doctors and patients.

The need for lengthy, high-dose stimulation schemes used in poor responders to increase the number of cumulus–oocytes complexes (COCs) retrieved, can be avoided by performing IVF during a natural cycle. It has been suggested that by following this approach, reasonable pregnancy rates can be achieved (Lindheim et al., 1997; Bassil et al., 1999; Feldman et al., 2001).

IVF outcome during a natural cycle, however, can be compromised by the occurrence of premature LH rise (Pelinck et al., 2002). The administration of GnRH antagonists in the mid-follicular phase can eliminate this problem and avoid cycle cancellation (Paulson et al., 1994). However, since GnRH antagonist administration, as well as LH, also suppresses FSH levels (Obery et al., 1999), gonadotropins are initiated at the time the antagonist is started to further sustain follicular development. In this modified natural cycle (MNC) the probability of pregnancy per attempt is theoretically increased (Rongieres-Bertrand et al., 1999).

Assessment of MNC in poor responders in whom oocyte donation appears to be the next step in fertility treatment has so far not been attempted. The purpose of this study was to evaluate the use of MNCs for IVF in patients with FSH levels >12 IU/l on day 3 of the cycle and five or fewer COCs retrieved in a previous unsuccessful IVF attempt.

Materials and methods

Inclusion criteria

Patients included should have a regular menstrual cycle (duration 21–35 days), FSH levels >12 IU/l on day 3 of the cycle and one or more failed IVF cycles in which five or fewer COCs were retrieved using a high gonadotropin dose. Given their bad prognosis, all patients were offered oocyte donation. Patients included in the current study insisted on continuing IVF attempts in order to become parents using their own genetic material or had ethical objections to oocyte donation.

Diagnosis of the patients included is given in Table I. Six patients had performed only one previous IVF trial. Oocyte donation is offered in our unit if the IVF cycle is stopped due to no response.
under the maximum FSH dose in patients with high basal FSH levels. The probability of pregnancy in these patients is disappointingly low (Klinkert et al., 2004).

From January 2001 to December 2003, 32 consecutive patients treated at the Centre for Reproductive Medicine of the Dutch-Speaking Brussels Free University met the above criteria and were included in the study. The study was approved by our Institutional Review Board.

Ovarian stimulation and IVF procedure
During a natural cycle, recombinant FSH (rFSH) 100 IU (Puregon; NV Organon, Oss, The Netherlands) and GnRH antagonist garelix 0.25 mg/day (Orgalantrin; NV Organon) were started concomitantly when a follicle with a mean diameter of 14 mm was present at ultrasound. HCG 10 000 IU (Pregnyl; NV Organon) was administered as soon as the mean follicular diameter was ≥16 mm. Hormonal levels were used to exclude a premature LH surge. Estradiol (E2) levels were not considered for HCG administration. Oocyte retrieval was performed by transvaginal ultrasound-guided double lumen needle aspiration 32 h after the HCG injection. The experience in this centre prior to study initiation showed that, despite the absence of LH surge, occasionally no follicle could be seen on the day of oocyte retrieval, 36 h after HCG administration. This has also been reported by Rongieres-Bertrand et al. (1999). ICSI was performed in 25 patients, while in the remaining five patients conventional IVF was performed. ICSI and conventional IVF procedures have been described in detail previously (Van Steirteghem et al., 1993; Devroye and Van Steirteghem, 2004). The decision about the method of insemination in cases of infertility not due to male factor was taken after discussion with the patient about the possibility of fertilization failure, which in the case of MNC means cycle cancellation. Embryos were classified as top quality, medium quality and low quality as described previously (De Vos et al., 1999). Embryo transfer was performed on day 3 after oocyte retrieval.

The luteal phase was supplemented with vaginal administration of 600 mg natural micronized progesterone in three separate doses (Utrogestan; Besins, Brussels, Belgium) starting 1 day after oocyte retrieval and continued until 7 weeks of gestation if pregnancy was achieved.

Hormonal measurements and ultrasound assessment of follicular development
The first hormonal evaluation was performed on day 8 of the cycle. Serum LH, FSH, E2 and progesterone levels were measured by means of the automated Elecsys Immunoanalyser (Roche Diagnostics, Mannheim, Germany). Intra-assay and inter-assay coefficients of variation for FSH were <5% and <10%, respectively. Ultrasound was performed concomitantly with hormonal assessment at each visit. Ultrasound image was frozen when the follicle appeared maximal, and two dimensions perpendicular to each other were measured from which the average follicular diameter was calculated.

Outcome measures
Pregnancies progressing beyond the 12th week of gestation were considered to be ongoing.

Statistical analysis
To the best of our knowledge there are no studies published in patients with poor prognosis using GnRH antagonists/rFSH. Therefore, no data were available by the time the study was initiated to estimate sample size in this population accurately in order to evaluate effectiveness of treatment. An arbitrary decision was taken to perform an initial evaluation after the first 30 patients had started treatment. If the pregnancy rate in this patient population was encouraging this treatment could continue until a calculated larger sample size was reached with which treatment efficiency could be estimated. Alternatively, a comparison with a different treatment could be planned. Values are expressed as mean ± SEM unless stated otherwise.

Results
The mean serum FSH level on day 3 of the cycle prior to initiating the first MNC was 21.9 ± 1.9 IU/l, and the mean age of the patients at initiation of the first MNC was 38.4 ± 0.7 years (Figure 1). The number of COCs retrieved in the last failed IVF cycle was 1.37 ± 0.3, and the mean starting dose of FSH used in the last cycle was 466 ± 17 IU (Figure 1). The patients had performed a mean of 2.5 ± 0.3 failed IVF cycles (range one to eight) before initiating an MNC.

Characteristics of the MNCs that resulted in oocyte retrieval are given in Table II. E2 levels before and after GnRH antagonist administration are shown in Figure 2.

HCG was given in time in cycles that reached oocyte retrieval. No premature LH surge occurred in the present study.

Twenty-five out of 78 cycles (32.1%) did not result in oocyte retrieval due to no follicular development. In nine out of 53 cycles (16.9%) in which oocyte retrieval was performed, no COCs were retrieved (mean number of oocytes retrieved in patients with oocyte retrieval 0.9 ± 0.1 COCs; range 0–2).

Overall, in 34 out of 78 cycles (43.6%), no COCs were available after an MNC. Following fertilization, embryo transfer was performed in 19 out of 44 cycles in which COCs were retrieved (43.2%). From the remaining cycles, oocytes of bad quality were retrieved in five cycles, immature oocytes were retrieved in five cycles, no fertilization occurred in five cycles, the oocyte was degenerated after ICSI in four cycles and poor embryo cleavage was present in six cycles. The 2PN rate per COC retrieved was 60.0% (27/45).

In 17 out of 19 cycles in which embryo transfer was performed, embryos of medium quality were replaced, while one embryo of high and one of low quality were replaced in the remaining two cycles.

Overall, the embryo transfer rate was 24.4% [19/78; 95% confidence interval (CI) 16.2–34.9]. No ongoing pregnancy

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**Table I. Diagnosis of the patients included in the study**

<table>
<thead>
<tr>
<th>Diagnosis</th>
<th>No. patients</th>
<th>%</th>
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<tbody>
<tr>
<td>Andrological</td>
<td>12</td>
<td>37.50</td>
</tr>
<tr>
<td>Female age</td>
<td>9</td>
<td>28.12</td>
</tr>
<tr>
<td>Andrological + age</td>
<td>6</td>
<td>18.75</td>
</tr>
<tr>
<td>Endometriosis</td>
<td>2</td>
<td>6.25</td>
</tr>
<tr>
<td>Tubal</td>
<td>1</td>
<td>3.12</td>
</tr>
<tr>
<td>Andrological + tubal</td>
<td>1</td>
<td>3.12</td>
</tr>
<tr>
<td>Age + endometriosis</td>
<td>1</td>
<td>3.12</td>
</tr>
</tbody>
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was achieved in 78 MNCs (0.0%; 95% CI 0.0–4.7) (Figure 3).

Discussion
The current study has shown that use of MNCs does not offer a realistic chance of parenthood in patients with high levels of FSH on day 3 of the cycle and previous poor response to ovarian stimulation, when offered as a last resort prior to oocyte donation.

To the best of our knowledge, this is the first study to evaluate the use of MNC in poor responders. In the present series, patients evaluated were characterized by high FSH levels on day 3 of the cycle and retrieval of five or fewer COCs using high doses of gonadotropins in the last IVF cycle before initiating MNC treatment.

Unfortunately, no single definition of poor ovarian response exists in the literature, and consequently, poor responders comprise a wide spectrum of patients with probably different prognoses. Generalization of the study findings to poor responders overall is probably not justified. The patients included in the current study are likely to be at the worst end of the poor responder spectrum, as they were characterized by a proven poor ovarian response, a low ovarian reserve, depicted by high basal FSH levels (median FSH 21.9 IU/l), and no acceptance of oocyte donation.

Table II. Characteristics of the modified natural cycles which resulted in oocyte retrieval

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>Median ± SD</th>
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<tbody>
<tr>
<td>Duration of antagonist administration (days)</td>
<td>2.7 ± 1.6</td>
</tr>
<tr>
<td>Total units of rFSH used (IU)</td>
<td>173.5 ± 15.7</td>
</tr>
<tr>
<td>Duration of follicular phase (days)</td>
<td>11.4 ± 0.4</td>
</tr>
<tr>
<td>Hormonal values on the day of HCG</td>
<td></td>
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<tr>
<td>E2 (pg/ml)</td>
<td>270.4 ± 18.3</td>
</tr>
<tr>
<td>Progesterone (ng/ml)</td>
<td>0.5 ± 0.0</td>
</tr>
<tr>
<td>FSH (IU/l)</td>
<td>10.3 ± 0.8</td>
</tr>
<tr>
<td>LH (IU/l)</td>
<td>6.3 ± 0.5</td>
</tr>
<tr>
<td>Mean follicular diameter of the leading follicle on the day of HCG (mm)</td>
<td>17.3 ± 0.3</td>
</tr>
</tbody>
</table>
The fact that the patients included did not accept oocyte donation poses difficulties in generalizing the study findings, as this is a personal decision related to cultural or religious characteristics of the population analysed, which cannot be quantified accurately. However, it can probably be supported that the study findings can be applied to a population of patients with characteristics described in Figure 1.

Although no data are available on the use of MNCs in poor responders, evaluation of natural IVF cycles has previously been attempted. In a small prospective study, Bassil et al. (1999) analysed 11 patients who underwent 16 natural cycles for IVF. Poor ovarian response was defined as failure to define E₂ concentrations > 200 pg/ml, and either no follicles or a maximum of one follicle during two previous attempts. An ongoing pregnancy rate of 18.6% was achieved; however, both the age (36.6 years) and the basal FSH (12.9 IU/l) of the patients analysed were lower than those in the current study.

Similarly, in a small prospective study, Feldman et al. (2001) evaluated the use of natural cycle for IVF in 22 patients who underwent 44 cycles. A ongoing pregnancy and implantation rate of 20% was observed. However, poor ovarian response was defined solely on the basis of COCs retrieved, and day 3 FSH levels were lower (13.3 IU/l) than those in the present study.

Moreover, in a small retrospective study, 30 patients who had previously been cancelled because of low ovarian response during an IVF cycle, underwent 35 natural cycles achieving an ongoing pregnancy rate of 16.6% per oocyte retrieval and an implantation rate of 33% (Lindheim et al., 1997). However, all patients were < 40 years and had a mean day 3 FSH of 11.1 IU/l.
It can probably be concluded from the above studies that the disappointing result from the application of MNC in poor responders in the present study is related to the stricter inclusion criteria used to define patient population.

On the other hand, MNC has been successfully performed in patients undergoing ICSI aged between 26 and 36 years with normal menstrual cycles, day 3 FSH levels <8 IU/l and fewer than three previous IVF procedures performed (Rongieres-Bertrand et al., 1999). Therefore, treatment failure in the current study is likely to be associated with the characteristics of the patients analysed rather than the method of the modified natural cycle itself.

However, the possibility cannot be excluded that modifications of the protocol applied in the current study could be associated with an improved outcome. HCG administration can be given at a later stage of follicular development, perhaps resulting in a higher proportion of mature oocytes. Criteria used for HCG could incorporate E2 per follicle, although the E2 produced per follicle depends on the degree of LH levels, which vary between patients (Kolibianakis et al., 2003), especially after antagonist administration. Oocyte retrieval can be performed at 36 h after HCG administration instead of 32 h. This might result in a higher proportion of MNCs without a follicle present on the day oocyte retrieval (Rongieres-Bertrand et al., 1999), but might improve oocyte quality. The importance of the criteria used for cycle monitoring has been shown by Vlaisavljevic et al. (2001), although no comparative information currently exists for patients with poor prognosis.

On the other hand, it is also likely that poor ovarian response is not an alteration of ovarian physiology that is amenable to treatment. Poor ovarian response to exogenous stimulation might be more a sign of a failing ovary and the stricter the criteria that are used for its definition, the more unlikely the achievement of conception.

The quality of the embryos transferred in the current study was not optimal. This is probably due the inability to select a morphologically optimal embryo, which is inherent in the MNC, and/or to the advanced reproductive age of the patients analysed, which was reflected in their mean basal FSH and their chronological age.

In conclusion, the current study suggests that the MNC, under the protocol applied, is not an effective treatment for patients with high FSH levels and low numbers of COCs retrieved in a previous IVF cycle. Patients should be counselled appropriately and either proceed to oocyte donation or abandon fertility treatment.

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References

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