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SHORT COMMUNICATIONS

NEW YORK, NEW YORK

Poor Responders to Ovarian Hyperstimulation May Benefit from an Attempt at Natural-Cycle Oocyte Retrieval[1]

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INTRODUCTION

Poor responders to controlled ovarian hyperstimulation (COH) prior to attempts at in vitro fertilization and embryo transfer (IVF-ET) present a clinical challenge. Diminished success rates for pregnancy are associated with decreased oocytes harvested and patients are discouraged from further attempts and often referred for oocyte donation. Undoubtedly, donor oocytes offer patients a better chance for success. However, many patients are not accepting of this option. Alternative attempts to stimulation have included "short flare" with GnRH agonist, human menopausal gonadotropins (hMG) alone, combinations of clomiphene citrate and hMG, and natural or unstimulated cycles (1–4). However, the ideal approach remains elusive.

Typically, poor-response cycles are canceled to allow for the initiation of alternative stimulation protocols in an attempt to optimize the number of recruited follicles. However, on occasion patients with poor stimulation responses elect to proceed with oocyte aspiration with the knowledge of the probable poor outcome. Stimulation cycles that result in few recruited follicles in some respects appear similar to natural IVF cycles in that a single or small cohort of dominant follicle(s) is aspirated and cultured. The early report of a successful pregnancy following natural-cycle IVF was in a poor-responder patient (5).

In select patients with a diminished ovarian reserve (advanced age or intermittently elevated day 3 FSH), an attempt at IVF using a natural cycle may be offered (6) with reasonable pregnancy rates. We present our experience in known poor responders to COH for IVF-ET who subsequently underwent natural-cycle IVF, evaluating clinical outcome measures including fertilization, implantation, and pregnancy rates. We compare these parameters to women with poor-response cycles who elected to have oocyte aspiration.

MATERIALS AND METHODS

All patients who underwent COH for IVF-ET from January 1990 to January 1996 were analyzed. Patients demonstrating a poor response, defined as three or fewer dominant follicles, who still subsequently underwent oocyte aspiration (Group I) and patients previously canceled due to poor responses to COH for IVF-ET who subsequently underwent natural-cycle IVF-ET were evaluated (Group II). Male-factor infertility and women ≥40 years of age were excluded.

Group I received luteal-phase (day 21) gonadotropin releasing hormone agonist down regulation with subcutaneously administered leuprolide acetate (n = 27), 1.0 mg/day, decreased to 0.5 mg/day when serum estradiol was <30 pg/ml, followed by ovarian hyperstimulation with 300 IU/day hMG given as a single dose. Monitoring was performed by serial transvaginal sonography and serum estradiol levels. Dosing was individualized up to 8 ampoules per day to maximize ovarian response. When the dominant follicle(s) was (were) ≥18–20 mm, patients were given human chorionic gonadotropin (hCG), 10,000 IU. Oocyte retrieval was performed by transvaginal ultrasound-guided aspiration 34–36 hr later.

Group II (n = 30) underwent natural-cycle monitoring as described previously (6). Briefly, a baseline

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transvaginal ultrasound was performed during the first 3 days of the menstrual cycle. Patients returned 4 days before the anticipated time of ovulation. Serial transvaginal ultrasonography and serum estradiol measurements were obtained and hCG, 10,000 IU, was administered im when follicle maturity was achieved, as per previously published criteria (6). If a luteinizing hormone (LH) surge was not detected, transvaginal ultrasound-guided aspiration was performed 36 hr after hCG administration.

Oocytes were cultured under standard laboratory conditions using modified Ham's F-10 medium (GIBCO, Grand Island, NY) supplemented with 6% plasmanate human albumin Fraction V (National Hospital Buffer). Sperm samples were processed by a three-Percoll gradient (47, 70, and 90%, respectively). Insemination was done by microdroplet under oil. After fertilization, preembryos were transfered into growth medium (Ham's F10 and 15% maternal serum). Transcervical embryo transfer occurred 48 to 72 hr after retrieval. Serum β-hCG levels were determined 9 and 12 days after transfer. Transvaginal ultrasound confirmed pregnancy viability.

There were no differences with respect to the age of patients enrolled in both groups: Group I, 36.7 ± 1 years; and Group II, 37 ± 3.6 years. Baseline day 3 FSH and estradiol were also similar (Group I, 12 ± 1.2 mIU/ml and 44.6 ± 4.9 pg/ml; Group II, 11.1 ± 1.2 mIU/ml and 41.8 ± 5.6 pg/ml).

Serum samples were assayed for FSH and estradiol using ^{125}IRIA kits. For FSH (Diagnostic Products, Los Angeles, CA), calibrated against the WHO second IRP-hMG, intra- and interassay CV were 3.4 and 4.2%, respectively. For estradiol (Pantex, Santa Monica, CA). Intra- and interassay CV were 5.0 and 8.4%. Statistical analysis was performed using the Student *t* test and chi-square analysis. Significance was expressed as $P < 0.05$.

RESULTS

Clinical results are depicted in Table I. Group I had significantly greater peak estradiol values on the day of administered hCG (678 ± 77 pg/ml) compared to Group II (300 ± 12 pg/ml; $P < 0.01$). Six cases did not result in embryo transfer due to failed fertilization in Group I; five cases in Group II were canceled due to spontaneous LH surge and another two did not result in embryo transfer due to failed fertilization. Significantly more oocytes were obtained at retrieval in Group I (2.3 ± 0.1) compared to Group II (1.1 ± 0.1 $P < 0.01$). However, fertilization rates (FR) were significantly lower in Group I (48.0 ± 8%) compared to Group II (89.1 ± 6%; $P < 0.05$). Although the numbers of embryos transferred were similar in each group (Group I, 1.14 ± 0.2; Group II, 1.1 ± 0.1, the implantation rates (IR) were significantly lower in Group I (7 ± 4%) compared to Group II (33 ± 9%; $P < 0.01$). Ongoing pregnancy rates (PR) per retrieval and per embryo transfer were 7.4% (2/27) and 9.5% (2/21) for Group I and 16.6% (5/30) and 17.9% (5/28) for Group II. Differences in FR, IR, and PR were not seen whether one, two, or three oocytes were retrieved.

Table I. Results of Poor Responders Using COH Compared to the Natural Cycle

	Group I (n = 27 cycles)	Group II (n = 35 cycles)
Peak estradiol (pg/ml)	678 ± 77[a] (282–1407)	300 ± 12.4[a] (180–420)
Cancellation rate (%)	0.0 (0/27)	14.3 (5/35)
No. oocytes	2.3 ± 0.1**	1.1 ± 0.1
FR (%)	48 ± 8*	89.1 ± 6
No. embryo transfers	114 ± 0.2	1.1 ± 0.1
IR (%)	7 ± 4**	33 ± 9
PR (%) per retrieval	7.4 (2/27)	16.6 (5/30)
Per embryo transfer	9.5 (2/21)	17.9 (5/28)

[a] SE.
* $P < 0.05$.
** $P < 0.01$.

DISCUSSION

Optimal stimulation protocols for poor responders remain a clinical challenge. Cycles resulting in poor responses are typically canceled. It is generally believed that collection of a sufficient number of good-quality oocytes is a prerequisite for success (7); otherwise, the chance for achieving a pregnancy is significantly reduced. However, even in "normal" cycles, where ample numbers of oocytes are recruited, embryo implantation rates are decreased following the use of menotropins (8), implying a decrease in endometrial receptivity following the use of fertility medications.

The use of ovulation induction for IVF-ET incurs great expense and added risk including ovarian cancer (9), unlike the natural cycle, which results in cost savings, is time efficient, and avoids many of the issues of risks and complications related to medication. Though the primary indication for natural-cycle IVF remains tubal disease, there is an early report of preg-

nancy following IVF in an unstimulated cycle in a patient failing to respond to ovarian hyperstimulation (6).

Our results indicate that, despite the great expense and effort, ovulation induction cycles, which appear to be similar to natural cycles in the low number of oocytes retrieved, do not result in better clinical outcomes. In fact, the greater number of oocytes retrieved in Group I results in significantly lower fertilization and implantation rates. This suggests that oocyte and embryo quality are lower in stimulated cycles of poor responders than in natural cycles. A decrease in endometrial receptivity cannot be completely discounted because of the significantly reduced implantation rates, despite similar pregnancy rates for each group. The similar pregnancy rates could be explained on the basis of a Type II error (β error) due to our small sample size, especially in light of the reduced implantation rates, suggesting a decrease in endometrial receptivity for all patients using human menopausal gonadotropins. Therefore, because of suboptimal stimulation protocols, patient reluctance to proceed with ovum donation, and the advantages the natural cycle offers including acceptable pregnancy outcome, it appears to be better to proceed to natural cycles than to waste time and financial resources with controlled ovarian hyperstimulation in patients who are under 40 years of age and without male factor.

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Amplification of the RB1.20 Polymorphism in Single Spermatozoa

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INTRODUCTION

The RB1.20 polymorphic locus identified in intron 20 of the retinoblastoma (RB1) gene (1) is a length polymorphism consisting of a variable number (n = 14–26) of the repeat $\{CTTT(T)\}_n$. This class of short tandem repeats (STR) is usually highly informative, thus providing valuable tool for DNA linkage studies. Brandt *et al.* (2) described a nonradioactive protocol for detecting the RB1.20 polymorphism in the genomic DNA extracted from peripheral blood lymphocytes