Efficiency of changing the embryo transfer time from day 3 to day 2 among women with poor ovarian response: A prospective randomized trial

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Objective: To compare the outcome of day 2 and day 3 embryo transfers in women demonstrating poor ovarian response.
Design: Prospective randomized clinical trial.
Setting: Private assisted reproductive technology center.
Patient(s): Two hundred eighty-one women demonstrating poor ovarian response to controlled ovarian hyperstimulation.
Intervention(s): Women who were poor responders were randomly allocated to day 2 or day 3 embryo transfer following oocyte retrieval.
Main Outcome Measure(s): Implantation rates and pregnancy rates per oocyte retrieval and embryo transfer.
Result(s): The clinical pregnancy rates per oocyte retrieval (37.2% vs. 21.4%, respectively; $P<.05$) and per embryo transfer (38.9% vs. 24.1%, respectively; $P<.05$) were significantly higher in the day 2 embryo transfer group compared with day 3. On the other hand, implantation rates were not different between groups (23.9% vs. 17.2%, respectively; $P=.08$).
Conclusion(s): Our results demonstrated that transfering embryos on day 2 could provide an alternative to the management of poor responder patients. (Fertil Steril® 2006;86:81–5. ©2006 by American Society for Reproductive Medicine.)

Key Words: Poor responder, embryo transfer, day 2, day 3

In earlier stages of assisted reproductive technology (ART), knowledge of the physiology and energy metabolism of human embryos enabled 4-cell stage embryos to be successfully cultured in vitro (1). Subsequently, better selection of surviving embryos and differentiation of their quality allowed embryo transfer on day 3. Delaying transfer can increase the likelihood of successful implantation owing to the availability of additional morphologic features (2). Thus, various grading systems were developed to identify good-quality day 3 embryos, and, over the last decade, the majority of ART centers have transfered day 3 embryos.

More recently, the use of sequential media has facilitated the in vitro development of early preimplantation human embryos to the blastocyst stage in 40% to 70% of cases (3). Conceivably, extended in vitro culture enabled the selection of embryos with a higher implantation potential, thus decreasing the need for multiple gestations. Several studies comparing the efficiency of day 3 and day 5 transfers have reported better implantation rates for the latter (4–6). It should be noted, however, that exposing human embryos to in vitro conditions may have a negative impact on their developmental potential, and the increased duration of cultivation may lead to a decrease in the number of available embryos for transfer. Therefore prolonged in vitro culture conditions may not be justifiable in all cases, particularly in cycles in which few oocytes and/or embryos are available.

In women undergoing ART, poor response to ovarian stimulation is a therapeutic challenge (7). Although several protocols have addressed this problem, none has proven to be superior (8). Poor responder patients have few embryos available for transfer. Therefore, to save the couple from unnecessary financial and emotional burdens, a decision must frequently be made on whether to proceed or cancel a cycle. Owing to the high rate of cancellation of embryo transfers among poor responders, we proposed shortening the time embryos are exposed to in vitro conditions, thus reducing their potentially deleterious effects. This would result in a reduction in the number of cancelled cycles and may increase pregnancy rates. We therefore designed a prospective study to compare day 2 and day 3 embryo transfers in women demonstrating poor ovarian response.

MATERIALS AND METHODS

Patients

This study was conducted prospectively over a period of 6 months (June–November 2004) in 281 women with five or fewer follicles (>13 mm) at the end of ovarian stimulation (Fig. 1) and in whom only fresh ejaculated sperm was used

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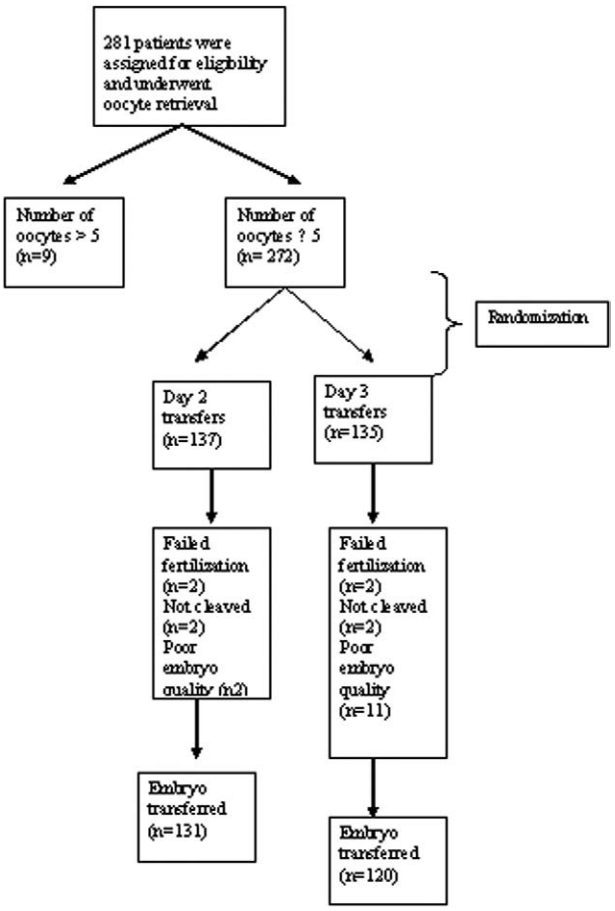
FIGURE 1

Study design.



Bahceci. Day 2 vs. day 3 transfers in poor responders. Fertil Steril 2006.

for insemination. Patients from whom no oocytes were retrieved were excluded from the study (n = 9). The remaining 272 patients were randomized, using a table of random numbers, for day 2 (n = 137) and day 3 embryo transfer (n = 135). Institutional Review Board approval was obtained for this study, and all participating couples gave informed consent.

Ovarian Hyperstimulation Protocol

Two types of ovarian stimulation protocols were employed. The "long protocol" began with pituitary desensitization using a GnRH agonist (Lucrin; Abbot, Aubonne, France) in the midluteal phase of the preceding menstrual period (9). Administration of gonadotropins (Gonal F; Serono, Aubonne, Switzerland) was initiated on day 3 of the commencing cycle. The "microdose flare-up protocol" began with low-dose oral contraceptive (Desolett; Organon, Istanbul, Turkey), starting on day 1 of the previous menstrual cycle, for 21 days (10). Leuprolide acetate (Lucrin; Abbott) (40 μg SC per day) was initiated on the second day of menstruation, followed by gonadotropins, which were administered on cycle day 3. Gonadotrophin dosage was tailored according to individual ovarian responses, with initial doses ranging from 300 to 600 IU per day. When the leading follicle reached a diameter of 18 mm, 10,000 IU hCG (Pregnyl; Organon) was administered to trigger ovulation. Oocytes were retrieved 35 h after hCG injection and were subjected to intracytoplasmic sperm injection (ICSI).

Embryology

The embryology procedures have been described previously (11). Briefly, development of each fertilized zygote was traced individually in separate 30-μL drops of Quinn's Cleavage Medium (Sage BioPharma, Bedmister, NJ) covered with sterile mineral oil. Embryos were kept in air with 5.5% CO_2. Embryo quality was calculated by multiplying the morphologic grade (1 to 4 grading system of Bolton et al., 1989 (12); in which grade 4 is morphologically the best) with the number of blastomeres (13). Embryos from cycles assigned for transfer at day 3 were not inspected on day 2. Embryo transfers were cancelled in patients whose embryos were all poor quality (grade 1 or 2) after the couple's consent has been obtained.

Embryo transfers were performed using an Edwards-Wallace catheter under ultrasound guidance with full bladder. Patients received supplemental progesterone in oil in a dose of 100 mg/day, starting on the day of oocyte collection through the luteal phase. Clinical pregnancy was defined as the demonstration of gestational sac(s) by transvaginal ultrasonography with rising serum β-hCG levels. All surviving pregnancies beyond 12 weeks of gestation were considered as ongoing pregnancy.

Statistics and Sample Size Calculation

Data were analyzed by Fisher exact test or unpaired Student *t* test, where appropriate. A *P* value of $<.05$ was considered significant.

We calculated that to increase pregnancy rate per oocyte retrieval among poor responders from 15% to 30%, with a power of 80% and 0.05 error, $\geq$133 patients for each group had to be recruited.

RESULTS

Table 1 shows the demographic and clinical characteristics of the participating women. The groups did not differ in mean age, baseline hormonal levels, number of previous ART attempts, or percentage of women >40 years old.

In Table 2, controlled ovarian hyperstimulation characteristics of the two embryo transfer groups are presented. The two groups were administered similar amounts of gonadotropins, which resulted in similar peak E_2 levels. The mean numbers of retrieved, mature (MII), and fertilized oocytes were similar between the two groups. Two cycles in each