

Monozygotic Twin Birth After the Transfer of a Cleavage Stage Embryo Resulting from a Single Pronucleated Oocyte

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Purpose: To present a case involving the transfer of a single pronucleated oocyte resulting in a monozygotic twin pregnancy.

Method: A descriptive case report of a single patient.

Results: The patient conceived and was found to have a monochorionic diamniotic pregnancy which resulted in the birth of normal identical twin boys at 32 weeks of gestation.

Conclusions: The case report addresses an issue that has not received proper attention in the literature. It illustrates that observing a single PN in an oocyte at fertilization check should not be an absolute deterrent to transferring the resulting embryo even in an older patient with a high FSH level. This report also suggests that single observations, especially at the assessment of fertilization, in the IVF laboratory are limited when evaluating embryo potential and normalcy.

KEY WORDS: Embryo transfer; IVF; monozygotic twin; single pronucleus.

INTRODUCTION

In the course of in vitro fertilization (IVF) treatment normal fertilization is assessed by observing two pronuclei which can be present in the oocyte as early as 13 h following insemination or intracytoplasmic sperm injection (ICSI). Single pronuclei are sometimes observed and cause a concern in the IVF laboratory as these may be the result of parthenogenetic activation, failure of either gamete to progress with pronucleus formation, disynchronous appearance of pronuclei, or male and female pronuclear fusion. Although some of these oocytes may actually be normally fertilized and diploid, due to the uncer-

tainty as to their genetic normalcy, they are often discarded (1).

The ability of embryos derived from single pronucleated oocytes to cause normal pregnancies and births has been infrequently described in the literature. Here we describe our experience with a successful monozygotic twin pregnancy from the transfer of a single embryo developed from a single pronucleated oocyte.

CASE REPORT

The patient is a 41-year-old woman with secondary infertility due to tubal disease and decreased ovarian reserve with a day 3 FSH level of 23.5 mIU/mL confirmed on two occasions. She declined using oocyte donation although she was counseled about the very low odds of success if she pursued IVF treatment using her own oocytes. She underwent controlled hyperstimulation using a microdose flare protocol with 450 IU of rFSH and 150 IU of HMG. Two oocytes

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were recovered by transvaginal ultrasound guided aspiration of two follicles. The oocytes were placed into pre-equilibrated 150 μ L droplets of P1 medium (Irvine Scientific—Santa Ana, CA) supplemented with 10% SSS v/v under mineral oil. The petri dish was incubated in a humidified environment of 37°C, 5% CO₂/5% O₂/90% N₂. The oocytes were inseminated conventionally with approximately 100,000 motile sperm per milliliter. At fertilization check, 17 h post insemination, the oocytes were denuded mechanically and observed using an inverted microscope at X200 magnification with Hoffman modulation optics. A single pronucleated oocyte and a tripronucleated oocyte were observed. On day three the single pronucleated oocyte had developed into an eight-cell embryo with equal sized blastomeres and less than 10% fragmentation. The patient was counseled about the dearth of information in the literature about outcomes after the transfer of embryos derived from single pronucleated oocytes. She elected to proceed with the embryo transfer. Assisted hatching with acidified Tyrode's solution was performed in view of her age and elevated FSH level.

The patient conceived and was found to have a monochorionic diamniotic pregnancy that resulted in the birth of normal identical twin boys at 32 weeks of gestation.

DISCUSSION

The appearance of single pronucleated oocytes have been reported to occur in frequencies varying from 1.6% to 7.7% of inseminated oocytes (2–7). Although these oocytes are usually discarded, due to the belief that they are the result of parthenogenetic activation, it is possible that some may be normally fertilized (1). Staessen and Van Steirteghem found that 48.7% of embryos developing from single pronucleated oocytes were diploid after IVF and 27.9% after ICSI (7). The potential of embryos derived from such oocytes to cause a normal pregnancy has been the subject of only three reports in the literature (1,6,8). The initial report by Staessen *et al.* described two births out of 38 transfers of a single embryo resulting from a zygote with one pronucleus (6). Barak *et al.* reported a birth with the transfer of four embryos all derived from single pronucleated oocytes following intracytoplasmic injection of round spermatids (8). Gras and Trounson also described a birth of a healthy child after transferring a day six blastocyst

following extended culture of a single pronucleated oocyte (1).

Our case report adds to further support a flexible policy in offering to transfer embryos resulting from single pronucleated oocytes, especially in situations when these are the only embryos available. However, patients should be counseled about the risk of genetic abnormalities with such embryos and the scant information available in the literature with regard to the potential for a successful outcome.

Gras and Trounson suggested extended culture beyond the cleavage stage in an effort to assess the potential of these embryos to develop to the blastocyst stage as a better predictor of genetic normalcy (1). In the present report, our patient was 41 years old and had a markedly elevated FSH level. In view of reports in the literature suggesting that patients with borderline embryos fare better with the "uterine rescue" of a day three transfer compared to extended in vitro culture (9), we decided that a day three transfer with assisted hatching may be more beneficial. The occurrence of monozygotic twins has been described to occur more commonly after IVF (10–15). Although we have not found an increased incidence after assisted hatching in our own program (10) a large nationwide study suggested that assisted hatching was a risk factor for the development of monozygotic twins after IVF (16).

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