Case report

Ongoing pregnancy after ICSI of frozen–thawed PESA-retrieved spermatozoa and IVF in a controlled natural cycle

Dr Kadoch completed his studies in medicine at the Saint-Antoine Faculty of Medicine (Université Paris VI, France) in 1995. After 5 years of residency in Obstetrics and Gynaecology in Paris, he was offered a fellowship position from 2000 to 2002 with Professor René Frydman (Clamart–France), where he acquired training in reproductive medicine and surgery. He spent 2 years as a Clinical Fellow in Reproductive Endocrinology and Infertility in the Centre Hospitalier de l'Université de Montréal (CHUM), and currently he is an Assistant Professor there, in the Obstetrics and Gynaecology department. Dr Kadoch has special interest in the development of new techniques such as natural cycle IVF and IVM to improve the quality of patient care.

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Abstract

The recovery of a mature oocyte from a natural cycle followed by IVF (nIVF) is an attractive alternative to conventional IVF, involving ovarian stimulation, in the treatment of female infertility. Similarly, surgical recovery of spermatozoa from the epididymis by percutaneous sperm aspiration (PESA) has simplified the treatment of men with obstructive azoospermia. A couple sought treatment for diminished ovarian reserve and male factor infertility using IVF. A mature oocyte was retrieved and was inseminated by intracytoplasmic sperm injection (ICSI), following recovery of spermatozoa by PESA. A good quality embryo was transferred. A viable pregnancy was confirmed by ultrasound scan. A healthy baby boy was delivered naturally at 37 weeks gestation. This study reports the first ongoing clinical pregnancy and subsequent birth resulting from ICSI of spermatozoa retrieved by PESA into an oocyte recovered during a natural cycle. The use of a combination of less invasive assisted reproductive techniques (PESA and nIVF) can overcome barriers to fertility.

Keywords: *ICSI, IVF, natural cycle, PESA*

Introduction

The first successful IVF was performed in the natural cycle of an infertile woman with tubal factor infertility (Steptoe and Edwards, 1978).

Ovarian stimulation can increase oocyte and embryo numbers as well as pregnancy rates (Fischel *et al*., 1985). Hence, ovarian stimulation became a widely used method in the treatment of infertility, and natural IVF (nIVF) was soon abandoned in favour of stimulated IVF (sIVF). sIVF increases the probability of obtaining more than one fertilizable oocyte as well as that of conception. sIVF treatment requires ovarian follicular stimulation to increase the number of oocytes and the successful treatment outcome (Healy *et al*., 1994).

However, improvements in laboratory techniques and methods of follicular aspiration have created renewed interest in nIVF. Gonadotrophin-releasing hormone (GnRH) antagonists induce a reversible medical hypophysectomy, which prevents the occurrence of premature LH surges and thus increases the likely success of a cycle of nIVF (Rongieres-Bertrand *et al*., 1999).

IVF with intracytoplasmic sperm injection (ICSI) has been introduced as a potential treatment for severe male infertility, and high rates of fertilization and pregnancy have been reported (Palermo *et al*., 1992; Van Steirteghem *et al*., 1993). In the absence of spermatozoa in the ejaculate (azoospermia), the introduction of micro-epididymal and percutaneous epididymal sperm aspiration (PESA) followed by intracytoplasmic injection (ICSI) also appeared to be potential

breakthroughs for the treatment of severe male infertility (Shrivastav *et al*., 1994). The few drops of fluid obtained during a PESA procedure may contain sufficient spermatozoa to be frozen for several ICSI procedures.

Case report

A 35-year-old woman and her 43-year-old spouse were referred for assisted reproduction due to decreased ovarian reserve diagnosed by low antral follicle count on ultrasound examination and male infertility. Semen analysis revealed an ejaculate volume of 5.5 ml (normal >2 ml), a sperm concentration of 23.1 mol/ml (normal >20 mol/ml) and necrozoospermia.

The couple had had two children together previously and the male partner underwent an unsuccessful vasectomy reversal.

A baseline ultrasound scan was performed on day 3 of menses to exclude ovarian cysts and to ensure that the endometrial lining was <5 mm thick. Serial transvaginal ultrasounds were started on day 6. Follicular diameter was established by calculating the mean value of the two largest measurements perpendicular to each other. Subsequently, the patient was monitored until the leading follicle reached a diameter >14 mm. As soon as the dominant follicle reached a mean diameter of 14 mm, the patient received an antagonist (ganirelix, Orgalutran; Organon Pharmaceuticals, Scarborough, Ontario, Canada) (0.25 mg, s.c.) administered as an s.c. injection to avoid a spontaneous LH surge. Recombinant gonadotrophin (rFSH Puregon; Organon, 150 IU) was administered daily at the time of the first injection of antagonist to prevent a fall in the oestradiol concentration on the following day, and repeated thereafter until human chorionic gonadotrophin (HCG) administration. When the follicle had a diameter of 17 mm, the patient received HCG (Pregnyl; Organon Pharmaceuticals, 5000 IU) administered as an IM injection to achieve final follicular maturation. Transvaginal oocyte retrieval was scheduled 34–36 h after HCG administration, and produced a mature oocyte. Only the dominant follicle was aspirated.

Following retrieval of the oocyte, PESA was performed. A volume of 0.3 ml of fluid was obtained with a sperm count of 13.5 mol/ml and motility of 30%. Spermatozoa were washed in Cook fertilization media (Cook Canada, Toronto, Canada). The oocyte was assessed for maturity and then ICSI was performed according to standard protocols. The remaining sperm specimen was frozen for future use.

Fertilization was confirmed 18 h later with the presence of two pronuclei. Grading of the embryo was performed on day 2 after oocyte retrieval and before transfer. The embryo transferred on day 2 after oocyte retrieval was four cells, of good morphological quality with little fragmentation and equally sized blastomeres. Zona thinning was performed using the Hamilton Thorne ZILOS (Beverly, MA, USA). The transfer of the embryo to the recipient uterus was completed without complication on the first attempt. Unfortunately the patient did not conceive on this cycle. A second cycle was started from the patient's next menses, following the same protocol. A single oocyte was retrieved, fertilized by ICSI using the frozen–thawed PESA sample. On day 2 after oocyte retrieval a 2-cell embryo with even blastomeres and no fragmentation was transferred. The luteal phase was supported by HCG (Pregnyl; Organon Pharmaceuticals) and vaginal natural P micronized progesterone (Prometrium; Schering, Pointe-Claire, Quebec, Canada) (600 mg daily) starting 2 days after oocyte retrieval and continued daily up to menstruation or for at least the first 8 weeks of pregnancy if the patient became pregnant. Serum HCG was collected 15 days after oocyte retrieval. The first ultrasound was performed at a gestational age of 6 weeks. A clinical pregnancy was defined as a pregnancy in which a fetal sac with fetal heart activity was visualized by ultrasound 8 weeks after HCG administration. A healthy baby boy was delivered at 37 weeks' gestation.

Discussion

An ESHRE consensus meeting report recently stated that the essential aim of IVF/ICSI is the birth of one single healthy child, with a twin pregnancy being regarded as a complication (Land and Evers, 2003).

The collection of a mature oocyte from a naturally selected follicle, followed by IVF, has received increasing attention as an alternative to conventional sIVF treatment. The benefits of nIVF compared with sIVF include reduced cost (fewer drugs), reduced health risks (no hyperstimulation syndrome and multiple pregnancies), and increased patient acceptability (no blood tests). The financial benefits of nIVF as compared with sIVF have been reported previously (Daya *et al*., 1995). Although this protocol does require the patient to take some medication, it is greatly reduced as compared with normal stimulation protocols in sIVF, even mild controlled stimulation. The oocyte retrieval procedure with only one follicle is extremely quick and may be performed without anaesthesia or sedation. Since only one oocyte and therefore one embryo is obtained, there is no risk for multiple pregnancy.

So far as is known, this report describes the first ongoing clinical pregnancy and subsequent birth of a healthy baby resulting from ICSI of frozen–thawed PESA-retrieved spermatozoa and nIVF.

In summary, this study indicates that nIVF is a viable option for infertile women who have normal ovulatory menstrual cycles. If necessary, ICSI can be performed using spermatozoa retrieved by PESA. This report illustrates the use of a combination of fewer invasive assisted reproductive techniques in overcoming barriers to infertility. These results also indicate the importance of counselling regarding other possible options, such as sIVF, in cases such as this.

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