Successful delivery after the transfer of twice-vitrified embryos derived from in vitro matured oocytes: A Case Report

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We report here the first case of successful pregnancy and delivery after the blastocyst transfer of twice-vitrified embryos produced following in vitro maturation (IVM) and ICSI. The patient received 5000 IU hCG on day 12 of the treatment cycle, and oocyte retrieval was carried out 36 h after hCG injection. A total of 22 immature oocytes were obtained. Following incubation for 26 h in IVM medium, 15 oocytes (68.2%) reached metaphase II stage. In total, 13 oocytes (86.7%) were fertilized after ICSI with the husband’s sperm, and 11 embryos at the pronuclear stage and two cleaved embryos on day 2 were vitrified because of thin endometrial thickness. Eight cryopreserved embryos at the pronuclear stage were warmed and cultured until the day 3 stage. Three embryos were transferred, and three embryos were twice vitrified. Unfortunately, these transferred embryos did not implant. Three twice-vitrified embryos were rewarmed and cultured until the day 5 stage, and two embryos were transferred. The second transfer attempt of twice-vitrified embryos resulted in the full-term delivery of a healthy infant. This case report demonstrates that twice-vitrified embryos, developed using an IVM protocol, retain the developmental competence for full-term, healthy infants.

Key words: in vitro maturation/PCOS/repeated cryopreservation/vitrification

Introduction

In vitro maturation (IVM) of immature oocytes has been proposed as a potential alternative for conventional IVF treatment following controlled ovarian hyperstimulation (COH). The protocol is becoming more accepted, and increasing numbers of babies resulting from IVM have been born worldwide (Chian, 2004; Son et al., 2005) since the first report of successful pregnancy following this procedure in 1994 (Trounson et al., 1994).

The capability of human embryo cryopreservation has increased patients’ convenience and safety in fertility treatment, not only reducing the cost and time but also reducing their physical load. It provides opportunities to limit the number of transferred embryos, to store supernumerary embryos or to avoid ovarian hyperstimulation syndrome (OHSS) and so on. Also, regarding the IVM protocol, a report of successful embryo cryopreservation using IVM oocytes (Chian et al., 2001) has expanded the treatment availability. However, to the best of our knowledge, there has been no report of successful pregnancy following repeated cryopreservation of embryos resulting from the IVM protocol.

In this case report, we describe the first successful delivery of a healthy infant from twice-vitrified embryos produced from IVM oocytes.

Case report

A 26-year-old Japanese woman with polycystic ovary syndrome (PCOS) presented at our outpatient clinic with complaints of anovulation and 2 years of infertility. After she had failed to achieve pregnancy following several courses of ovulation induction with gonadotrophins injections, she opted for IVM treatment to reduce the risk of OHSS. Informed consent was provided.

To initiate the treatment cycle, the patient received Norethisterone-Mestranol (Norluten-D®, Shionogi Pharm, Osaka, Japan) for 10 days. On day 12 of withdrawal bleeding, small ovarian follicles were monitored by transvaginal ultrasonography (Toshiba, Tokyo, Japan), confirming that there was no dominant follicle. An endometrial thickness of 5.8 mm was noted. The patient was given 5000 IU of hCG (Profasi, Serono Japan, Tokyo, Japan) 36 h before oocyte retrieval (Chian et al., 1999).
Blastocyst embryo transfer was carried out on the fifth day of Lutrol administration, when the endometrial thickness was 11.5 mm. The serum hCG level was 61.8 IU/l at 9 days after embryo transfer and pregnancy was confirmed. Nineteen days after embryo transfer (5+3/7 weeks of gestation), transvaginal ultrasound revealed a single intrauterine gestational sac and 27 days after embryo transfer an ongoing intrauterine singleton pregnancy with fetal heartbeat was confirmed.

The obstetrical course was uneventful, and a healthy female infant was delivered at 40 weeks of gestation. The weight at birth was 3356 g.

Discussion

Cryopreservation of supernumerary embryos produced during human IVF protocols provides an opportunity for patients to have repeated attempts at conception following a single oocyte retrieval, preventing wastage of valuable genetic material and improving cumulative pregnancy rates. Thus with advances of technology, multiple cryopreservation of human IVF embryos has been contributing to the treatment availability in human assisted reproduction technology (Baker et al., 1996; Farhat et al., 2001; Estes et al., 2003; Smith et al., 2005). We performed primary vitrification at the pronuclear stage and secondary vitrification at the cleavage stage. The viabilities of vitrified and twice-vitrified embryos were both 100% (8/8 and 3/3, respectively). Developmental competences of these embryos to transferable embryos were 75% (6/8) and 67% (2/3, respectively). Our case indicates that vitrification is a robust technique for multiple cryopreservation. It can be a beneficial cryopreservation method for embryos at the pronuclear stage or oocytes (Kuleshova and Lopata, 2002; Liebermann et al., 2003; Walker et al., 2004; Kuwayama et al., 2005). In addition, it has another merit in saving time compared with the slow freezing method (Chen et al., 2005; Huang et al., 2005).

IVM of immature oocytes has been proposed as an alternative for conventional IVF treatment. IVM treatment has several potential advantages including lower external treatment costs (less drugs and monitoring), reduced health risks (reduced incidence of ovarian hyperstimulation syndrome) and increased convenience to the patient (fewer blood tests and ultrasonographic monitoring and no daily injections) compared with conventional IVF treatment. Recently, clinical performance following the IVM protocol has been improved by pretreatment with gonadotrophin injections (FSH or hMG, Wynn et al., 1998), hCG priming before oocyte retrieval (Gomez et al., 1993) and the improvement of culture conditions (Chian, 2004). Application of cryopreservation has also expanded the availability of this protocol (Chian et al., 2001). However, this procedure is performed in only a limited number of institutes worldwide.

We describe here the successful live birth after the blastocyst transfer of twice-vitrified embryos produced following IVM and IVF. Our success could be achieved with the support of a combination of technologies—IVM protocol, vitrification and blastocyst culture. Along with the improvement of IVM protocol, the capability of multiple cryopreservation without impairing embryo quality, even with an embryo derived from...
IVM, could help in promoting the IVM protocol as a more common and more convenient fertility treatment.

This case report demonstrated that embryos produced from IVM oocytes could retain the developmental competence to full-term, healthy infants, even after repeated cryopreservation and blastocyst culture. We envisage that in the future, with a large number of additional studies and the combination of other technologies such as vitrification, the IVM protocol will be proven to be a more functional fertility treatment.

References


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