Day 3 ET, single blastocyst transfer (SBT) or frozen-thawed embryo transfer (FET): which is preferable for high responder patients in IVF/ICSI cycles?

Keliang Wu · Haibin Zhao · Hui Liu · Mei Li ·
Shuiying Ma · Cheng Li · Chang Liu · Zi-Jiang Chen

Received: 19 July 2013 / Accepted: 9 December 2013 / Published online: 5 January 2014
© Springer Science+Business Media New York 2014

Abstract

Purpose To compare the clinical outcomes after day 3 embryo transfer, day 5 single blastocyst transfer (SBT) and frozen-thawed embryo transfer (FET) in high responder patients (>15 retrieved oocytes) undergoing IVF/ICSI treatment.

Methods A retrospective analysis of three embryo transfer strategies for the high responder patients in IVF/ICSI cycles. The 1041 high responder patients diagnosed as primary infertility with more than 15 oocytes retrieved were recruited in Day 3 ET group, 308 patients with more than 15 oocytes retrieved first transferred with one blastocyst in SBT group and 425 patients with more than 15 oocytes retrieved in fresh cycle, first transferred with one frozen-thawed blastocyst were assigned in FET group.

Results In the high responder patients, the clinical pregnancy rate after day 5 SBT was significantly lower than that of day 3 ET (43.18 % VS 57.16 %, $p < 0.05$). In addition, the clinical pregnant rate and implantation rate of FET cycles were significantly higher than SBT cycles (59.06 % vs. 43.18 % and 64.70 % vs. 47.40 %, $p < 0.05$). The multiple pregnancy rate in FET cycles was markedly lower than that of day 3 ET (2.35 % VS 34.97 %, $p < 0.05$).

Capsule FET was the optimal strategy for the high responder patients in IVF/ICSI cycles to lower multiple pregnancy rates and improve clinical pregnancy rate.

Keliang Wu and Haibin Zhao contributed equally to this work.

K. Wu · H. Zhao · H. Liu · M. Li · S. Ma · C. Li · C. Liu ·
Z.-J. Chen (✉)

Center for Reproductive Medicine, Provincial Hospital Affiliated to Shandong University, National Research Center for Assisted Reproductive Technology and Reproductive Genetics, China The Key laboratory for Reproductive Endocrinology of Ministry of Education, Shandong Provincial Key Laboratory of Reproductive Medicine, 324 Jingwu Road, Jinan 250021, China
e-mail: chen zijiang@hotmail.com

Conclusions FET was the preferable strategy for the high responder patients in IVF/ICSI cycles to obtain both desirable clinical outcome and lower multiple pregnancy rates.

Keywords Day 3 embryo transfer · SBT · High responder patients · IVF/ICSI cycles

Introduction

Since the introduction of assisted reproduction, IVF has quickly become one of the most effective and widely used approaches in treating infertility, leading to about 1 % of neonates [16]. However, with the advances in cell culturing and other manipulation technology, higher implantation rates were obtained at the cost of increasing multiple live birth rates. In comparison with single birth, IVF-ET multiple births may lead to increased parental stress, marital discord and financial hardship [5,9,10,13,15,17,21]. Consequently, decreasing the number of embryos transferred is a rational and imperative measure to minimize the incidence of multiple gestations in ART.

Single embryo transfer had been recommended as an approach to decrease the rate of multiple births after IVF [4]. It had been demonstrated that fresh pregnancy rates can be optimized by blastocyst transfer, which results in higher birth rate than single cleavage stage embryo transfer [18,19]. Recent studies had focused on single blastocyst stage transfer in which the embryos were cultured to day 5 enhanced the likelihood of pregnancy compared to day 3 embryo transfer without the complications of multiple pregnancies [12]. It had been shown by another retrospective study that SBT demonstrated a noticeably decrease in twinning in comparison with the double blastocyst group (3 % VS 62 %) and still kept their pregnancy rates at 76 % and 79 %, for donor and non-donor IVF patients, respectively [7].

Although SBT had been reported with so many advantages over day 3 ET, some other aspects should be taken into consideration if ideal clinical outcome want to be obtained, such as receptivity of endometrium which is necessary for the successful implantation of either cleavage stage embryos or blastocysts. It had been demonstrated by Simon that high serum E2 levels ($>3,000$ pg/ml) on the day of hCG administration in high responders were harmful to uterine receptivity [6]. Paulson et al. also reckoned that the disappointing implantation results were due to the quality of endometrial element during COH to increase the number of retrieved oocytes in IVF/ICSI cycles especially in high responder patients [20]. In addition, there were possible adverse effects directly on the embryo that would reduce the chance for blastocyte adhesion and implantation [24]. Therefore, SBT may not necessarily suited for all patients if want to achieve both ideal pregnancy rate and the lower multiple pregnancies for high responder patients.

The aim of this prospective study was to analysis the clinical outcome of the high responder patients when different transfer strategies were applied. We concluded that FET was preferable strategy for the high responder patients in IVF/ICSI cycles.

Materials and methods

Patients

Recruited patients who presented at Anonym. This study was approved by the Reproductive Medical Review Board of Anonym. Informed consent was obtained from all patients who participated in this study. Requirements of patients in this study were: primary infertility, female age ≤ 35 years, the E2 level on the day of hCG administration over 3,000 pg/ml, the number of “ ≥ 14 mm follicles” over 15, more than 15 retrieved oocytes suitable for IVF/ICSI treatment (FSH levels on female cycle day 2–3 ≤ 12 mIU/ml and available spermatozoa in semen).

This study was initiated in January 2012 and ended in November 2012. One thousand three hundred and forty-nine patients in fresh IVF-ICSI cycles and 425 patients in frozen–thawed embryo transfer (FET) cycles were recruited in the study.

Ovarian stimulation

All of the patients underwent controlled ovarian hyperstimulation (COH) using GnRH agonist (triptorelin acetate; Ferring AG, Switzerland) starting on day 21 of the menstrual cycle before the oocyte retrieval cycle. Recombinant FSH (Gonal-F; Serono, Switzerland) was given continuously to the patients from menstrual cycle day 3. Human chorionic gonadotropin (hCG, 6,000–10,000 IU; Lizhu, China) was administered when there were >3 follicles >17 mm in diameter and serum E₂ levels reached 300 pg/ml per follicle in which the diameter was >14 mm. The patients were

scheduled to have oocyte retrieval procedures 36 h after hCG administration.

Insemination and embryo transfer

The COCs were inseminated in 4-well plates with approximately 100,000 motile spermatozoa for each oocyte (IVF) or undergoing intracytoplasmic sperm injection (ICSI). Sequential culture media from Vitrolife (G-IVF, G1 and G2; Scandinavien IVF Science, Goteborg, Sweden) were applied in all steps, and embryo transfer were performed using a Wallace catheter under ultrasound guidance. The embryos transferred in SBT on day 5 were assessed over 4BC grade according to Garden and Lane criteria [11]. In day 3, two high quality embryos were selected for transfer. Good quality embryos on day 3 after oocyte retrieval were defined as ≥ 7 cells with a grade better than 3 according to Puissant's criteria [22].

FET

Patients in the cryopreservation group were down-regulated with leuprolide acetate in a subsequent cycle and received oral E2 (6.0 mg daily) and E2 patches as needed starting 10–14 days before thaw to achieve a target endometrial thickness of at least 8 mm. Vitrified blastocysts were thawed in the morning of transfer day, and all thawed embryos were applied assist hatching half an hour after thawing. The embryos will be incubated for 4–5 h before transfer, only completely expanded blastocysts evaluated over 4BC grade according to Garden and Lane criteria were transferred, otherwise, transfer would be given up.

Diagnosis of pregnancy

A serum hCG level >10 IU/L 14 days after embryo transfer was diagnosed as a biochemical pregnancy and cardiac activity 7 weeks after embryo transfer was defined as a clinical pregnancy.

Statistical analysis

Statistical analysis was performed using a χ^2 test. A P value <0.05 was considered statistically significant.

Results

Day 3 ET and SBT outcome in high responder patients

Clinical pregnancy rate was significantly lower in SBT than day 3 ET groups (43.18 % VS 57.16 %, $p < 0.05$, Table 1), while these two group demonstrated comparable implantation rates (47.40 % VS. 37.18 %, $p > 0.05$). And the multiple pregnancy rate was also very significantly lower in SBT group than day 3 ET group (2.60 % VS. 34.97 %, $P < 0.01$, Table 1)

Table 1 Day 3 ET and SBT outcome in high responder patients

	SBT	ET
No. cycles	308	1041
Age (year)	29.02±2.87	29.75±2.35
No. oocytes retrieved per cycle	19.34±5.73	19.31±5.45
No. embryos transferred per cycle	1	2
Clinical pregnancy (%)	133 (43.18 %)	595 (57.16 %)*
Implantation (%)	146/308 (47.40 %)	774/2082 (37.18 %)
Twin pregnancy (%)	8 (2.60 %)	364 (34.97 %)*

Values are presented as number (%) or mean ± SD

* $P<0.05$ compared to SBT groups

FET outcome of high responder patients

To investigate the clinical outcome of FET in high responder patients, 425 patients in frozen–thawed embryo transfer (FET) cycles were employed in the study. There was no difference in the blastocyst quality (Table 2). The clinical pregnancy rate and implantation rate in FET cycles were significantly higher than that in SBT cycles (59.06 % vs. 43.18 % and 64.70 % vs. 47.40 %, respectively, $P<0.05$) (Table 2). And no significant difference was found in the twin pregnancy rate between the two groups.

Discussion

As the development of assisted reproductive technology, the pregnancy rate has been increasing dramatically. However, multiple gestations, the most common adverse outcome associated with ART had been considered the most frustrating aspect we must face. There were many disadvantages with multiple births including low birth weight and increasing prenatal mortality [1–3,14,25]. To obtain desired clinical pregnancy without increasing multiple pregnancies was always ultimate goal for reproductive teams. Many studies demonstrate that ideal clinical outcome can be obtained either on day

3 or day 5 ET [8]. Our previous data also revealed that similar clinical outcomes could be achieved either transferring double embryos on day 3 or SBT on day 5 for normal responder patients. In this study we found that the clinical pregnancy rate after day 5 SBT in high responders was significantly lower than that of day 3 ET although without any difference in implantation rate between them. And interestingly, compared to SBT cycles in our data, the clinical pregnancy rate of day 3 transfer cycles were not declined dramatically, although presenting a downward trend. Although the implantation rate was similar between day 3 transfer cycles and SBT cycles, this not means the embryo transferred had the equal chance to successful implantation between to groups, because not all of the cleavage stage embryos transferred could developed to blastocyst, which may more representative of the totipotency of the embryo. And in our center, about 60 % of good quality day 3 embryos could developed to good blastocyst. Therefore, in the consideration of efficiency of embryo transfer, SBT was not recommended for the high responder patients. In addition, although day 3 ET can obtain ideal clinical outcome, the twin pregnancy rate was really high. Recently, the IVF/ICSI patients are at their own expense both at home and abroad. To gain the widest possible clinical outcome, most patients choose double embryos transferred. Because of this, we advise that reproductive centers could reduce or even render the operations gratis to encourage single embryo transfer when day 3 ET was selected in high responders.

With advances in assisted reproductive technology, not only do we pursue high clinical pregnancies, but also the lower multiple pregnancies. For high responders, although the pregnancy of day 3 ET was acceptable, the multiple gestations were inevitably high. SBT could achieve nearly natural multiple pregnancy rate, however the clinical pregnancy rate was declined dramatically for high responder patients. We must face the paradox. Fortunately, with the application of blastocysts vitrification, FET cycles were used more frequently to patients who failed or without the chance for embryo transfer in fresh cycles. In this study we found FET could obtain both satisfactory multiple pregnancy rate and preferable clinical outcome. This could be supported by Shapiro BS's

Table 2 SBT and FET outcome in high responder patients

	SBT	FET
No. cycles	308	425
Age (year)	29.02±2.87	29.05±2.48
No. oocytes retrieved per cycle	19.34±5.73	19.37±6.25
No. embryos transferred per cycle	1	1
Composition of transferred blastocyst	196 (≥4BB)+112 (4BC)	309 (≥4BB)+116 (4BC)
Clinical pregnancy (%)	133 (43.18 %)	251 (59.06 %)*
Implantation (%)	146/308 (47.40 %)	275/425 (64.70 %)*
No. twin pregnancy cycles (%)	8 (2.60 %)	10 (2.35 %)

* $P<0.05$ compared to SBT groups

study, he found that frozen-thawed embryo transfers significantly improved the clinical pregnancy rate even in normal responder patients, this outcome strongly suggested the impaired endometrial receptivity in fresh ET cycles after ovarian stimulation [23]. So FET may be the preferable strategy for high responders to decline multiple pregnancies and increase the chance of pregnancy.

In conclusion, FET was the optimal strategy for the high responder patients in IVF/ICSI cycles to lower multiple pregnancy rates and improve clinical pregnancy rate.

Acknowledgments This article is funded by Science research foundation item of no-earnings health vocation (201002013) and National Basic Research Program of China (973 Program) (2010CB945002).

References

- Alexander GR, Kogan M, Martin J, Papiernik E. What are the fetal growth patterns of singletons, twins, and triplets in the United States? *Clin Obstet Gynecol*. 1998;41:114–25.
- Alexander GR, Slay WM, Salihu H, Kirby RS. Fetal and neonatal mortality risks of multiple births. *Obstet Gynecol Clin North Am*. 2005;32:1–16.
- Blondel B, Kogan MD, Alexander GR, Dattani N, Kramer MS, Macfarlane A, et al. The impact of the increasing number of multiple births on the rates of preterm birth and low birthweight: an international study. *Am J Public Health*. 2002;92:1323–30.
- Bergh C. Single embryo transfer: a mini-review. *Hum Reprod*. 2005;20:323–7.
- Bryan E. The impact of multiple preterm births on the family. *BJOG*. 2003;110 Suppl 20:24–8.
- Simon C, Juna JG, Valbuena D, Peinado J, Pellicer A. Increasing uterine receptivity by decreasing estradiol levels during the preimplantation period in high responderers with the use of a follicle-stimulating hormone step-down regimen. *Fertil Steril*. 1998;70:234–9.
- Criniti A, Thyer A, Chow G, Lin P, Klein N, Soules M. Elective single blastocyst transfer reduces twin rates without compromising pregnancy rates. *Fertil Steril*. 2005;84:1613–9.
- Stoop D, Van Landuyt L, Camus M, Pevroey P. Should a single blastocyst transfer policy be a clinical decision or should it depend on the embryological evaluation on day 3? *Reprod Bio and Endo*. 2011;9:60–8.
- Ellison MA, Hall JE. Social stigma and compounded losses: quality-of-life issues for multiple-birth families. *Fertil Steril*. 2003;80:405–14.
- Ellison MA, Hotamisliligil S, Lee H, Rich-Edwards JW, Pang SC, Hall JE. Psychosocial risks associated with multiple births resulting from assisted reproduction. *Fertil Steril*. 2005;83:1422–8.
- Gardner DK, Lane M. Culture and selection of viable blastocysts: a feasible proposition for human IVF? *Hum Reprod Update*. 1997;3:367–82.
- Gardner DK, Lane M, Stevens J, Schlenker T, Schoolcraft WB. Blastocyst score affects implantation and pregnancy outcome: towards a single blastocyst transfer. *Fertil Steril*. 2000;73:1155–8.
- Glazebrook C, Sheard C, Cox S, Oates M, Ndukwe G. Parenting stress in first-time mothers of twins and triplets conceived after in vitro fertilization. *Fertil Steril*. 2004;81:505–11.
- Gunby J, Daya S. Assisted reproductive technologies (ART) in Canada:2001 results from the Canadian ART Register. *Fertil Steril*. 2005;84:590–9.
- McWhinnie A. Euphoria or despair? Coping with multiple births from ART: what patients don't tell the clinics. *Hum Fertil (Camb)*. 2000;3:20–5.
- National Center for Chronic Disease Prevention and Health Promotion, American Society for Reproductive Medicine, Society for Assisted Reproductive Technology. 2001 Assisted reproductive technology success rates: national summary and fertility clinic reports. Atlanta: Centers for Disease Control and Prevention, December 2003.
- Olivennes F, Golombok S, Ramogida C, Rust J. Behavioral and cognitive development as well as family functioning of twins conceived by assisted reproduction: findings from a large population study. *Fertil Steril*. 2005;84:725–33.
- Papanikolaou E, Kolibianakis E, Tournaye H, Venetis C, Fatemi H, Tarlatzis B, et al. Live birth rates after transfer of equal number of blastocysts or cleavage-stage embryos in IVF. A systematic review and metaanalysis. *Hum Reprod*. 2008;23:91–9.
- Papanikolaou E, Camus M, Kolibianakis E, Van Landuyt L, Van Steirteghem A, Devroey P. In vitro fertilization with single blastocyst-stage versus single cleavage-stage embryos. *N Engl J Med*. 2006;354:1139–46.
- Pinborg A, Loft A, Schmidt L, Andersen AN. Morbidity in a Danish national cohort of 472 IVF/ICSI twins, 1132 non-IVF/ICSI twins and 634 IVF/ICSI singletons: health-related and social implications for the children and their families. *Hum Reprod*. 2003;18:1234–43.
- Paulson RJ, Sauer MV, Lobo RA. Embryo implantation after human in vitro fertilization: importance of endometrial receptivity. *Fertil Steril*. 1990;53:870–4.
- Puissant F, Van Rysselberghe M, Barlow P. Embryo scoring as a prognostic tool in IVF treatment. *Hum Reprod*. 1987;2:705–8.
- Shapiro BS, Daneshmand ST, Garner FC, Aguirre M, Hudson C, Thomas S. Evidence of impaired endometrial receptivity after ovarian stimulation for in vitro fertilization: a prospective randomized trial comparing fresh and frozen-thawed embryo transfer in normal responderers. *Fertil Steril*. 2011;96(2):344–8.
- Valbuena D, Martin J, de Pablo JL, Remohi J, Pellicer A, Simon C. Increasing levels of estradiol are deleterious to embryonic implantation because they directly affect the embryo. *Fertil Steril*. 2001;76:962–8.
- Wright VC, Schieve LA, Reynolds MA, Jeng G. Assisted reproductive technology surveillance—United States, 2002. *MMWR Surveill Summ*. 2005;54:1–24.