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Reproductive outcomes of retransferring retained embryos in blastocyst transfer cycles

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Objective: To determine the incidence of embryo retention (ER) in the transfer catheter following embryo transfer (ET) in blastocyst transfer and investigate whether retransferring retained embryos has an impact on reproductive outcomes in patients undergoing *in vitro* fertilization-ET.

Methods: We retrospectively analyzed the records of 1,131 blastocyst transfers, which comprised 223 single blastocyst transfer (SBT) and 908 double blastocyst transfer (DBT) cycles. Each SBT and DBT group was classified depending on whether ET was performed without retained embryos in the catheter during the first attempt (without-ER group) or whether any retained embryos were found following ET (ER group) for the purpose of comparing reproductive outcomes in a homogenous population.

Results: The overall incidence of finding retained embryos was 2.8% (32/1,131). There were no retained embryos in SBT cycles. In DBT cycles, implantation rates (30.0% vs. 26.6%), positive β-hCG rates (57.2% vs. 56.2%), clinical pregnancy rates (45.3% vs. 46.9%), and live birth rates (38.9% vs. 43.8%) were not significantly different between the without-ER and ER groups. There were no significant differences in the mean birth weight (g) 2,928.4±631.8 vs. 2,948.7±497.8 and the mean gestational age at birth (269.3±17.2 days vs. 264.2±25.7 days). A total of nine cases of congenital birth defects were found in this study population. Eight were observed in the without-ER group and one in the ER group.

Conclusion: Our results suggest that retransfer of retained embryos does not have any adverse impact on reproductive outcomes in blastocyst transfer cycles. Furthermore, our results support finding that SBT might be advantageous for decreasing the incidence of retained embryos in catheters.

Keywords: Blastocyst stage embro transfer; Cleavage stage embryo transfer; *In vitro* fertilization; Embryo retention

Introduction

Embryo transfer (ET) has been noted as a crucial step that has an influence on pregnancy rates in patients undergoing the *in vitro* fertilization (IVF) process [1,2]. The factors that affect the success of ET include embryo quality, endometrial receptivity, and the ET technique itself [2]. Although ET techniques have improved immensely, the incidence of embryo retention (ER) in the transfer catheter following attempted ET has remained consistent; the reported incidence rates of ER were between 1% and 7% in ET with cleavage-stage embryos [3-8]. There have been several retrospective studies about the effect of retransferring retained embryos on outcomes in IVF-ET with cleavage-stage embryos [3-8]. Among them, several studies [3-6,8] have reported that retransferring retained embryos in the transfer catheter did not affect the IVF outcomes. With great advances in embryo culture and transfer techniques, blastocyst transfer has taken center stage in IVF as an efficient strategy for avoiding multiple pregnancies while improving the pregnancy rate [9,10]. An error during the ET procedure resulting in ER cannot be ignored under the pressure to ensure that only one or two blastocysts are trans-

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ferred and to achieve a high probability of pregnancy. Such an error may lead to impaired IVF outcomes. However, there is a paucity of data about the incidence and reproductive outcomes of retained blastocysts in the transfer catheter. Moreover, some studies have speculated that retained blastocysts had a larger diameter and a more fragile zona pellucida, in which case the ET procedure may cause some trauma to the blastocyst and epigenetic effects on the embryo [11-14]. Keeping these studies in consideration, blastocyst transfer itself could be a factor that may affect the incidence of ER and ER in blastocyst transfer may affect the blastocysts and their subsequent growth in the uterus, which would have a negative effect on IVF and perinatal outcomes.

In this study, we analyzed 10 years of data from Cheil General Hospital to determine the incidence of blastocysts retained in the transfer catheter during Day 5 blastocyst transfer cycles where one or two blastocysts were transferred. We also investigated whether retransferring retained blastocysts had an impact on IVF and perinatal outcomes.

Methods

A total of 1,131 patients, comprising 776 fresh ET cycles and 355 frozen-thawed ET cycles, were recruited retrospectively at Cheil General Hospital from January 2004 to December 2014. Cycles for preimplantation genetic diagnosis and donor oocyte cycles were excluded. Among the patients included, 223 single blastocyst transfers (SBTs) and 908 double blastocyst transfers (DBTs) were performed. In fresh cycles, every patient had undergone controlled ovarian stimulation. Ovulation triggering was performed with the administration of 250 μg of human chorionic gonadotropin (hCG) (Ovidrel, Merck Serono, Darmstadt, Germany) as soon as two to three follicles 17 mm in diameter were observed by transvaginal ultrasound. Transvaginal oocyte retrieval was performed approximately 36 to 38 hours after the administration of hCG. In frozen-thawed cycles, we used 6 to 8 mg/day of oral estradiol valerate (Progynova, Bayer Schering Pharma, Berlin, Germany) for endometrial preparation and we used intramuscular progesterone (50 mg in oil, daily) for luteal support. One or two blastocysts that survived vitrification were transferred into the patient's uterus 5 days after the initiation of progesterone treatment. The embryos obtained were scored according to the modified Gardner and Schoolcraft grading system [15].

ET was performed with a full bladder under transabdominal ultrasound guidance. A Cook Sydney IVF catheter (Cook Medical, Bloomington, IN, USA) was used. The patient was placed in the lithotomy position without any sedation or anesthesia. After the removal of cervical mucus, an empty outer transfer catheter was passed through the external cervical os to the level of the internal cervical os. Then, the embryos were loaded into an inner catheter with approximately 20 μL of media. The inner catheter contained a 5-μg air bubble at the tip to aid visualization during ultrasound guidance. The embryologist checked the catheter microscopically for the presence of retained embryos. If any embryo was found to have been retained in the catheter, the retained embryos were immediately reloaded by the embryologist and a second transfer was performed. Then, the catheter was checked again.

A pregnancy test was performed by determining the serum β-hCG level 12 days after the oocyte retrieval day in fresh ET cycles or 12 days after the initiation of progesterone treatment in frozen-thawed cycles. If the pregnancy test was positive, patients underwent an ultrasound scan 3 weeks later to establish the number of gestational sacs and embryo viability, and to exclude ectopic pregnancy.

The implantation rate was calculated as the number of gestational sacs per the number of transferred embryos. Clinical pregnancy was defined as the presence of an intrauterine gestational sac with a yolk sac, a fetal pole, and fetal heart pulsations. Live birth was defined as the birth of a live infant at $\geq$ 24 weeks of gestation.

We used a chi-square test and Fisher's exact test, as appropriate, to compare pregnancy and live birth and multiple gestation rates. A student's *t*-test was used to evaluate continuous parameters. A *p*-value < 0.05 was considered statistically significant. The statistical analysis was performed using SPSS ver. 22.0 (IBM Co., Armonk, NY, USA). In comparing pregnancy rates, we did not distinguish between patients with one or more than one retained embryo.

Results

A total of 1,131 ET procedures with single or double blastocysts, with Day 5 blastocysts, were performed by eight experienced physicians from January 2004 to December 2014. The cycle characteristics of single and double groups (SBT and DBT, respectively) are presented in Table 1. SBT and DBT groups were similar with regard to body mass index, basal estradiaol and FSH, endometrial thickness at day of hCG injection, and number of retrieved oocytes. Patients in the DBT group were significantly younger and had experienced fewer previous ET cycles ($p < 0.001$ and $p < 0.001$, respectively).

The overall incidence of finding retained embryos during the study period was 2.8% (32/1,131). Only two ET cycles required a third transfer attempt. There was no case of ER during transfer in the SBT group. Therefore, to compare IVF outcomes, only the DBT group was classified by whether the embryos were successfully transferred at the first attempt or not. The cycle characteristics of the without-ER group and the ER group are presented in Table 2. The without-ER group and ER group were similar with regard to age, body mass index, the number of previous ET cycles, basal E2 and FSH, endometrial thickness at day

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Table 1. Baseline characteristics and clinical features in the patients of SBT and DBT groups

Characteristics	SBT	DBT	*p*-value
No. of ET cycles	223	908	-
Age (yr)	34.3 ± 3.1	33.2 ± 3.4	0.000
Body mass index (kg/m^2)	20.5 ± 3.7	20.8 ± 3.8	0.393
Prior ET without birth	1.7 ± 2.3	1.1 ± 1.8	0.000
Basal estradiol (pg/mL)	26.0 ± 17.9	26.1 ± 18.4	0.947
FSH (mIU/mL)	8.1 ± 2.7	7.6 ± 2.7	0.135
Endometrial thickness at day of hCG administration (mm)	10.0 ± 2.5	10.3 ± 2.9	0.192
No. of retrieved oocytes	16.6 ± 8.5	17.0 ± 8.1	0.319
Rate of good quality embryo (%)	65.1 ± 43.2	87.1 ± 25.0	0.306

Values are presented as mean ± standard deviation.
SBT, single blastocyst transfer; DBT, double blastocyst transfer; ET, embryo transfer; FSH, follicle-stimulating hormone; hCG, human chorionic gonadotropin.
Student's *t*-test for numeric variables.

Table 2. Baseline characteristics and clinical features in DBT patients of the without-ER group and ER group

Characteristics	Without ER[a)]	ER[b)]	*p*-value
No. of ET cycles	876	32	-
Age (yr)	33.2 ± 3.4	33.0 ± 3.8	0.764
Body mass index (kg/m^2)	20.8 ± 3.8	20.3 ± 3.4	0.492
Prior ET without birth	1.1 ± 1.6	1.2 ± 1.6	0.661
Basal estradiol (pg/mL)	26.2 ± 18.5	22.3 ± 13.2	0.250
FSH (mIU/mL)	7.4 ± 2.5	7.5 ± 2.7	0.834
Endometrial thickness at day of hCG administration (mm)	10.3 ± 2.9	10.9 ± 3.3	0.267
No. of retrieved oocytes	17.0 ± 8.1	18.3 ± 9.5	0.422
Rate of good quality embryo (%)	87.5 ± 25.0	77.2 ± 25.0	0.475

Values are presented as mean ± standard deviation.
DBT, double blastocyst transfer; ER, embryo retention; ET, embryo transfer; FSH, follicle-stimulating hormone; hCG, human chorionic gonadotropin.
[a)]ET was successfully performed without retained embryos at the first attempt; [b)]Any retained embryos in the transfer catheter were found following the first attempt and retransferred immediately.
Student's *t*-test for numeric variables.

Table 3. *In vitro* fertilization-embryo transfer outcomes in DBT patients of the without-ER group and ER group

IVF outcomes	Without ER[a)]	ER[b)]	*p*-value
No. of ET cycles	876	32	-
Implantation rate (%)	30	26.6	0.591
Positive β-hCG rate	57.2 (501)	56.2 (18)	0.916
Clinical pregnancy rate	45.3 (469)	46.9 (15)	0.861
Live birth rate	38.9 (341)	43.8 (14)	0.583
Ectopic pregnancy rate	1.7 (15)	3.1 (1)	0.440

Values are presented as percent (number) unless otherwise indicated.
DBT, double blastocyst transfer; ER, embryo retention; ET, embryo transfer; hCG, human chorionic gonadotropin.
[a)]ET was successfully performed without retained embryos at the first attempt; [b)]Any retained embryos in the transfer catheter were found following the first attempt and retransferred immediately.
Student's *t*-test for numerical variables; chi-square test for categorical variables.

of hCG injection, and number of retrieved oocytes. The pregnancy outcomes of the two groups are described in Table 3. Implantation rates (30.0% vs. 26.6%), positive β-hCG rates (57.2% vs. 56.2%), clinical pregnancy rates (45.3% vs. 46.9%), and live birth rates (38.9% vs. 43.8%) were not significantly different between the two groups (Table 3).

Table 4 shows the perinatal outcomes of children born in this study. Out of 341 deliveries in the without-ER group, 440 babies were born. The mean gestational age was 269.3 ± 17.2 days and birth weight was 2,928.4 ± 631.8 g. There were 92 SBTs resulting in twins. Among them, seven were monozygotic twins. Triplets resulted in two cases.

Table 4. Comparison of characteristics of live born babies in DBT patients between the without-ER and ER groups

Outcome	Without ER[a]	ER[b]	*p*-value
Live birth	440	17	-
Gestational age (day)	269.3±17.2	264.2±25.7	0.316
Birth weight (g)	2,928.4±631.8	2,948.7±497.8	0.903
Multiple pregnancy	94	3	-
Twin (monozygotic twin)	92 (7)	3 (1)	-
Triplets	2	0	-

Values are presented as number or mean±standard deviation.
DBT, double blastocyst transfer; ER, embryo retention.
[a]Embryo transfer was successfully performed without retained embryos at the first attempt; [b]Any retained embryos in the transfer catheter were found following the first attempt and retransferred immediately.
Student's *t*-test for numeric variables.

Table 5. Type and prevalence of birth defects in live born babies in the without-ER and ER groups

	Without ER[a]	ER[b]
Type of birth defect	Trisomy 18, VSD, syndactyly, cleft lip, diaphragmatic hernia, omphalocele, inguinal hernia, hydrocephalus	PDA
Total number (%)	8 (1.8)	1 (5.8)

ER, embryo retention; VSD, ventricular septal defect; PDA, persistent ductus arteriosus.
[a]Embryo transfer was successfully performed without retained embryos at the first attempt; [b]Any retained embryos in the transfer catheter were found following the first attempt and retransferred immediately.

In the ER group, out of 14 deliveries, 17 babies were born. The mean gestational age was 264.2±25.7 days and the mean birth weight was 2,948.7±497.8 g. There were three sets of twins. Among them, there was one set of monozygotic twins and there were no triplets. No statistically significant differences in the mean gestational age at birth and the mean birth weight were noted between the without-ER and ER groups ($p=0.316$ and $p=0.903$, respectively).

A classification of all observed birth defects is listed in Table 5. Birth defects were classified according to the criteria of the National Center on Birth Defects and Developmental Disabilities, Centers for Disease Control and Prevention, United States Department of Health and Human Services [16]. A total of nine cases of congenital birth defects were found in this study population. Eight were observed in the without-ER group, and one in the ER group.

Discussion

Finding retained embryos in the transfer catheter may be one of the most stressful and inconvenient situations for both the clinician performing the transfer and the patient. Furthermore, during an ET procedure, it is important that the patient be relaxed, because psychological factors such as stress and anxiety could affect rates of IVF outcomes [17-19].

Over the decades, it has long been debated whether retransferring embryos retained during the transfer could have a negative effect on pregnancy rates in IVF patients. Recent studies [3-6,8] have reported that retransferring retained embryos in the transfer catheter did not have an adverse effect on IVF outcomes. However, all of these studies concerned cleavage-stage embryos. Nowadays, blastocyst transfer with one or two embryos has become common in order to reduce multiple pregnancy rates and to enhance IVF outcomes. However, there has been a lack of consensus regarding the incidence of ER and its influence on blastocyst transfer. Moreover, Lee et al. [3] stated that the difference in the morphological and hydrodynamic features according to the developing embryo stage might have an influence on the incidence of retained embryos. In other words, the larger, less dense, expanded blastocyst compared to the dense, compact character of cleavage-stage embryos might result in an increased risk of blastocyst retention in the transfer catheter. In addition, some studies have speculated that the zona pellucida of the blastocyst is more fragile near the time of hatching, and ET may cause some trauma to the blastocyst [11,12]. Considering these blastocyst features, it seems reasonable to postulate that blastocyst transfer itself might be a risk factor for ER or additional procedures like embryo retransfer might result in compromised IVF and prenatal outcomes. Two prior studies have evaluated ER in blastocyst transfer. Lee et al. [3] compared initial pregnancy rates for Day 3 embryo transfers with ER to those of blastocyst transfer without ER (40% vs. 36%, respectively) and found no statistically significant difference between them. However, they did not present concrete information about blastocyst transfer cycles, such as the number of study cases or the incidence of ER. On the other hand, although Silberstein et al. [6] reported that the embryonic

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stage at ET did not demonstrate a statistically significant difference, they simply compared the incidence of ER in cleavage-stage transfer cycles to that of blastocyst transfer cycles (2.3% vs. 4.4%). Hence, the primary goal of this study was to determine the incidence of retransferring retained embryos and its influence on IVF outcomes in Day 5 blastocyst transfer.

Our data showed that the overall incidence of ER in Day 5 blastocyst transfer (2.8%) was similar to the incidence of ER in ET with cleavage-stage embryos. This result was identical with our previous unpublished data regarding the incidence of ER in cleavage-stage ET (2.8%). No ER was observed in SBT, while the incidence of ER in DBT was 3.5%. As expected, the number of transferred blastocysts might be a factor that leads to an increased incidence of ER. From this viewpoint, our results suggest that a lower number of blastocysts transferred has the advantage of decreasing the incidence of retained embryos in catheters.

For the purpose of comparing IVF and prenatal outcomes in a homogenous population between the without-ER and ER groups, we divided patients according to the number of transferred embryos. As there was no case of ER in the SBT group, only the DBT group was split into two groups depending on whether at least one embryo was retained in the transfer catheter after ET. Our data show that IVF outcomes such as the implantation rate, positive β-hCG rate, clinical pregnancy rate, live birth rate, and ectopic pregnancy rate were not significantly affected by retransferring retained embryos during ET.

Some reports have noted a concern about the perinatal outcomes of blastocyst transfer, such as an increased risk for monozygotic twins (1.6%–5.6%) and for congenital malformations (2.5%–6.8%) [13,20-23]. Although the exact mechanism leading to this increased incidence is not yet fully understood, one suggested mechanism has been that micromanipulation of the fragile zona pellucida of the blastocyst may interfere with the natural process of hatching and the normal development of the embryo [13,20]. Considering this mechanism, it seems reasonable to postulate that ER requiring additional procedures might result in compromised IVF and prenatal outcomes. Comparing the perinatal outcomes of blastocyst transfer without ER to those of blastocyst transfer with ER, there were no statistically significant differences in the mean birth weight and the mean gestational age at birth between the without-ER and the ER groups, except for the mean gestational age. There was one case of monozygotic twins in the ER group. Although the incidence of congenital birth defects was higher in the ER group (2.1% vs. 5.8%), the number of cases with ER was too small to perform a proper statistical analysis. Accordingly, our data could not prove that the possibility existed for a lack of increased risk of congenital malformation in the ER group. However, considering the result of this presentation, the apprehensions might prove to be unfounded that blastocyst transfer could influence the incidence of retained embryos or blastocyst transfer and retransfer could be more traumatic to the embryo.

Our results demonstrate that Day 5 blastocyst transfer does not increase the incidence of retained embryos and proper retransfer of retained embryos has no adverse impact on IVF and perinatal outcomes in Day 5 blastocyst transfer cycles. To our knowledge, this study is the first to postulate the prenatal outcomes of blastocyst transfer with ER in addition to evaluating the incidence and IVF outcomes of blastocyst transfer with ER. Thus, we hope that the results of this study could help in resolving some of the debates found in the literature on blastocyst transfer. Furthermore, our results support the conclusion that a lower number of blastocysts transferred, especially with SBT, has the advantage of decreasing the incidence of retained embryos in catheters, in addition to preventing the multiple pregnancies associated with a transfer of multiple embryos.

Conflict of interest

No potential conflict of interest relevant to this article was reported.

References

1. Neithardt AB, Segars JH, Hennessy S, James AN, McKeeby JL. Embryo afterloading: a refinement in embryo transfer technique that may increase clinical pregnancy. Fertil Steril 2005;83:710-4.
2. Mansour RT, Aboulghar MA. Optimizing the embryo transfer technique. Hum Reprod 2002;17:1149-53.
3. Lee HC, Seifer DB, Shelden RM. Impact of retained embryos on the outcome of assisted reproductive technologies. Fertil Steril 2004;82:334-7.
4. Nabi A, Awonuga A, Birch H, Barlow S, Stewart B. Multiple attempts at embryo transfer: does this affect in-vitro fertilization treatment outcome? Hum Reprod 1997;12:1188-90.
5. Tur-Kaspa I, Yuval Y, Bider D, Levron J, Shulman A, Dor J. Difficult or repeated sequential embryo transfers do not adversely affect in-vitro fertilization pregnancy rates or outcome. Hum Reprod 1998;13:2452-5.
6. Silberstein T, Trimarchi JR, Shackelton R, Weitzen S, Frankfurter D, Plosker S. Ultrasound-guided miduterine cavity embryo transfer is associated with a decreased incidence of retained embryos in the transfer catheter. Fertil Steril 2005;84:1510-2.
7. Visser DS, Fourie FL, Kruger HF. Multiple attempts at embryo transfer: effect on pregnancy outcome in an in vitro fertilization and embryo transfer program. J Assist Reprod Genet 1993;10:37-43.
8. Vicdan K, Isik AZ, Akarsu C, Sozen E, Caglar G, Dingiloglu B, et al. The effect of retained embryos on pregnancy outcome in an in

vitro fertilization and embryo transfer program. Eur J Obstet Gynecol Reprod Biol 2007;134:79-82.
9. Cruz JR, Dubey AK, Patel J, Peak D, Hartog B, Gindoff PR. Is blastocyst transfer useful as an alternative treatment for patients with multiple in vitro fertilization failures? Fertil Steril 1999; 72:218-20.
10. 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.
11. Butterworth S. Blastocyst culture: myth or magic? Hum Fertil (Camb) 2001;4:109-16.
12. Sheiner E, Har-Vardi I, Potashnik G. The potential association between blastocyst transfer and monozygotic twinning. Fertil Steril 2001;75:217-8.
13. Milki AA, Jun SH, Hinckley MD, Behr B, Giudice LC, Westphal LM. Incidence of monozygotic twinning with blastocyst transfer compared to cleavage-stage transfer. Fertil Steril 2003;79:503-6.
14. Schoolcraft WB, Surrey ES, Gardner DK. Embryo transfer: techniques and variables affecting success. Fertil Steril 2001;76:863-70.
15. Gardner DK, Schoolcraft WB. In vitro culture of human blastocysts. In: Jansen R, Mortimer D, editors. Towards reproductive certainty: fertility and genetics beyond 1999. Pearl River: Parthenon; 1999. p. 378-88.
16. Dimeglio A, Bensahel H, Souchet P, Mazeau P, Bonnet F. Classification of clubfoot. J Pediatr Orthop B 1995;4:129-36.
17. Facchinetti F, Matteo ML, Artini GP, Volpe A, Genazzani AR. An increased vulnerability to stress is associated with a poor outcome of in vitro fertilization-embryo transfer treatment. Fertil Steril 1997;67:309-14.
18. Gallinelli A, Roncaglia R, Matteo ML, Ciaccio I, Volpe A, Facchinetti F. Immunological changes and stress are associated with different implantation rates in patients undergoing in vitro fertilization-embryo transfer. Fertil Steril 2001;76:85-91.
19. Smeenk JM, Verhaak CM, Eugster A, van Minnen A, Zielhuis GA, Braat DD. The effect of anxiety and depression on the outcome of in-vitro fertilization. Hum Reprod 2001;16:1420-3.
20. Skiadas CC, Missmer SA, Benson CB, Gee RE, Racowsky C. Risk factors associated with pregnancies containing a monochorionic pair following assisted reproductive technologies. Hum Reprod 2008;23:1366-71.
21. Chang HJ, Lee JR, Jee BC, Suh CS, Kim SH. Impact of blastocyst transfer on offspring sex ratio and the monozygotic twinning rate: a systematic review and meta-analysis. Fertil Steril 2009; 91:2381-90.
22. Schwarzler P, Zech H, Auer M, Pfau K, Gobel G, Vanderzwalmen P, et al. Pregnancy outcome after blastocyst transfer as compared to early cleavage stage embryo transfer. Hum Reprod 2004;19: 2097-102.
23. Kallen B, Finnstrom O, Lindam A, Nilsson E, Nygren KG, Olausson PO. Blastocyst versus cleavage stage transfer in in vitro fertilization: differences in neonatal outcome? Fertil Steril 2010;94:1680-3.

http://dx.doi.org/10.5653/cerm.2016.43.2.133