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Transfer of 2 Embryos Using a Double-Embryo Transfer Protocol Versus 2 Sequential Single-Embryo Transfers: The Impact on Multiple Pregnancy

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Abstract

Introduction: Assisted reproductive technologies (ARTs) are associated with potential risks, mainly related to multiple pregnancies, which are around 20% to 25%. Iatrogenic multiple pregnancies due to ovarian stimulation with multiples embryos transferred can be avoided by the elective single-embryo transfer (eSET), a growing practice worldwide. Adequately applied eSET, which impact on the incidence of complications without compromising treatment success, is still a challenge. The aim of this study was to compare the cumulative success rates of elective transfer of 2 embryos when transferred one by one (eSET), versus the success rates of elective double-embryo transfer (DET) in a single procedure, in a good prognosis population. **Methods:** This study evaluated 610 good prognosis infertile couples undergoing ART, split into 2 groups: eSET group which included those receiving first eSET (n = 237) and for those who did not become pregnant, they could receive a second frozen-thawed SET; and eDET group (n = 373) who received elective transfer of 2 good quality embryos in the first transfer. **Results:** Clinical pregnancy outcomes after a transfer of 2 embryos were similar between the groups (DET: 46.6% vs accumulated SET: 45.9%; $P = .898$). Multiple pregnancy rate was significantly lower in the group receiving transfer of 2 embryos, one by one, compared to DET (DET: 32.2% vs accumulated SET: 6.7%; $P < .001$). **Conclusions:** The eSET policy should be stimulated for good prognosis couples, as it maintains the accumulated clinical pregnancy rates, avoids multiples pregnancies, and consequently the maternal and neonate complication and indirect costs of treatment when considering spending on the obstetrics are reduced.

Keywords

in vitro fertilization, single-embryo transfer, double-embryo transfer, pregnancy rate, multiple pregnancy

Introduction

Since the first baby was conceived by in vitro fertilization (IVF), the number of treatments has been continuously increasing, and more than 200 000 cycles were reported in the United States in 2014. In addition to an increasing number of IVF procedures, maternal and newborn adverse effects have been rising due to an iatrogenic complication of the assisted reproductive technology (ART), namely, the multiple pregnancy. In IVF, 39.4% of newborns come from multiple pregnancies, whereas for natural conceptions, the rate is just 3.5%.[1] On the other hand, when the number of embryos transferred decreases, the multiple pregnancy rate also decreases,[2] achieving approximately the same rate (5%) as that of natural conceptions when a single-embryo transfer (SET) policy is applied.[3] With a decrease in the multiple pregnancy rate, the rates of neonatal complications, such as preterm birth, low birth weight, hemorrhage, and respiratory problems, also decrease.[2-4]

It is clear that a direct association exists between multiple pregnancies and the number of embryos transferred,[5] and the main strategy to combat multiple pregnancies is to limit the maximum number of embryos in 1 embryo transfer (ET). Because the IVF techniques are well established, the objective of IVF is a pregnancy in which the IVF success rate and

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neonatal outcomes are reflected by the delivery of healthy baby.[6] An eSET is established when 1 embryo is transferred, while the spare embryos are cryopreserved. The application of eSET varies among countries depending on factors such as health insurance, legislation, medical society guidelines, and culture. In Europe, an average of 20% of IVF cycles are eSET, but the rate can reach 69% in Sweden. In the United States, the practice of eSET has been increasing, with 28.5% of cycles in women younger than 36 years old reported in 2014.[1] Latin America has reported lower eSET rates, with only 2% of IVF cycles.[7]

The practice of eSET is influenced by regulations on the number of embryos to be transferred in some countries, but in most countries, the decision-making process regarding the number of ET is the responsibility of the doctor and the couple. Awareness that double-embryo transfer (DET) increases the probability of a live birth discourages couples from eSET, and many have strong preferences for twins. Additionally, the media tend to show biases in favor of reporting positive outcomes for twins, and health professionals have different risk perceptions because twin pregnancies are still considered acceptable for IVF.[8]

Double-embryo transfer is the most common practice worldwide, but it seems to promote a small increase in live birth rates and a large impact on twin pregnancies.[9,10] On the other hand, the transfer of 2 embryos in 2 sequential cycles results in similar cumulative live birth rates with reduced multiple pregnancy rates[11,12] and costs.[13] A reduction in the number of ET is a recommendation of the American Society of Reproductive Medicine (ASRM) and Society for Assisted Reproductive Technologies (SART). They consider the prognosis of the couple, embryo quality, and cryopreservation programs while also recognizing difficulties regarding patient information, financial restrictions, and the perceptions of a successful birth outcome for 1 healthy baby.[14]

In Brazil, 80% of IVF treatments are supported by the patients themselves because neither the government nor the private health care provide funding for it,[7] and despite the recommendations of the Federal Medical Counseling to reduce the number of ET, the eSET is not compulsory or even suggested.[15] In light of the financial burden and expectations of couples who seek IVF treatment, the indication of eSET remains a challenge. Faced with this scenario, the aim of this study was to evaluate the cumulative pregnancy rates and impacts on multiple pregnancy after the transfer of 2 embryos by comparing single-cycle DETs to 2 sequential eSETs.

Materials and Methods

This retrospective observational study evaluated IVF cycles at the Human Reproduction Center, Hospital das Clinicas, Faculdade de Medicina, Universidade de Sao Paulo (HCFMUSP), and a private assisted reproduction center in Sao Paulo, Brazil (Monteleone, Centro de Reproduçao Humana) between January 2010 and March 2016. All procedures in this study are part of the routine care in the assisted reproductive center, and written informed consent was obtained from all patients before treatment. Patients consented to the treatment procedures and to the retrospective data used in the scientific publications (Ethics Committee Proc. Number 1.151.345).

Study Groups

The database included 3451 cycles between January 2010 and March 2016, which were potentially eligible for this study. From those, cycles in which patients received eSET or eDET were first selected (n = 2383) and examined for eligibility. Then, we confirmed eligibility for this study if they met the following criteria: women aged 18 to 38 years who underwent a first or second IVF cycle and oocytes were injected with ejaculated or epididymis sperm, transferred 1 or 2 good-quality embryos, with at least 1 surplus good-quality embryo cryopreserved after transfer. Cycles were excluded if women presented with systemic or infectious diseases.

The final number of cycles included and analyzed in our study was 610 cycles from 572 patients who received an eDET (n = 373) or eSET (n = 237) and attended inclusion and exclusion criteria. Patients could receive a fresh or cryopreserved embryo in their first ET. For patients who received eSET and did not become pregnant, a second frozen-thawed ET was considered (Figure 1).

In Vitro Fertilization Protocol

Briefly, pituitary blockage was obtained either with a GnRH agonist (Lupron kit; Abbot SA Societé Française des Laboratories, France) or a GnRH antagonist (Cetrotide; Serono, Switzerland). Ovarian stimulation was accomplished using recombinant follicle-stimulating hormone (rFSH; Gonal-F; Merck, Germany) with 150 IU/day as the starting dose for women up to 35 years of age and 225 IU/day for women between 36 and 38 years of age. The gonadotrophin doses were adjusted according to the ovarian response. When at least 2 follicles reached a diameter of 18 mm, follicular maturation was triggered with an injection of 250 μg of recombinant human chorionic gonadotrophin (rhCG, Ovidrel; Merck, Germany). Oocyte retrieval was performed after 35 to 36 hours by transvaginal ultrasound-guided aspiration; the luteal phase was supported by 90 mg of daily progesterone (Crinone; Merck, Germany) via the vaginal approach, starting on the day of oocyte retrieval. All oocytes were fertilized by intracytoplasmic sperm injection.[16] Embryos were cultured according to standard methods in a triple gas incubator (90% N_2, 5% O_2, and 6% CO_2) at 37°C. Embryo transfers were performed on day 3 (D3) or day 5 (D5) of development. The embryos on D3 were considered good quality when they presented with 8 to 10 symmetric blastomeres, no multinucleations, and a maximum fragmentation level of 20%.[17] Blastocysts on D5 of development were considered good quality when the presentation expanded (grades 3 or 4), the inner cell mass grades were A or B, and the trophectoderm was A or B.[18] Vitrification of the embryos was performed using the Vitrification Freeze kit (Irvine Scientific, USA) with a Cryotip device

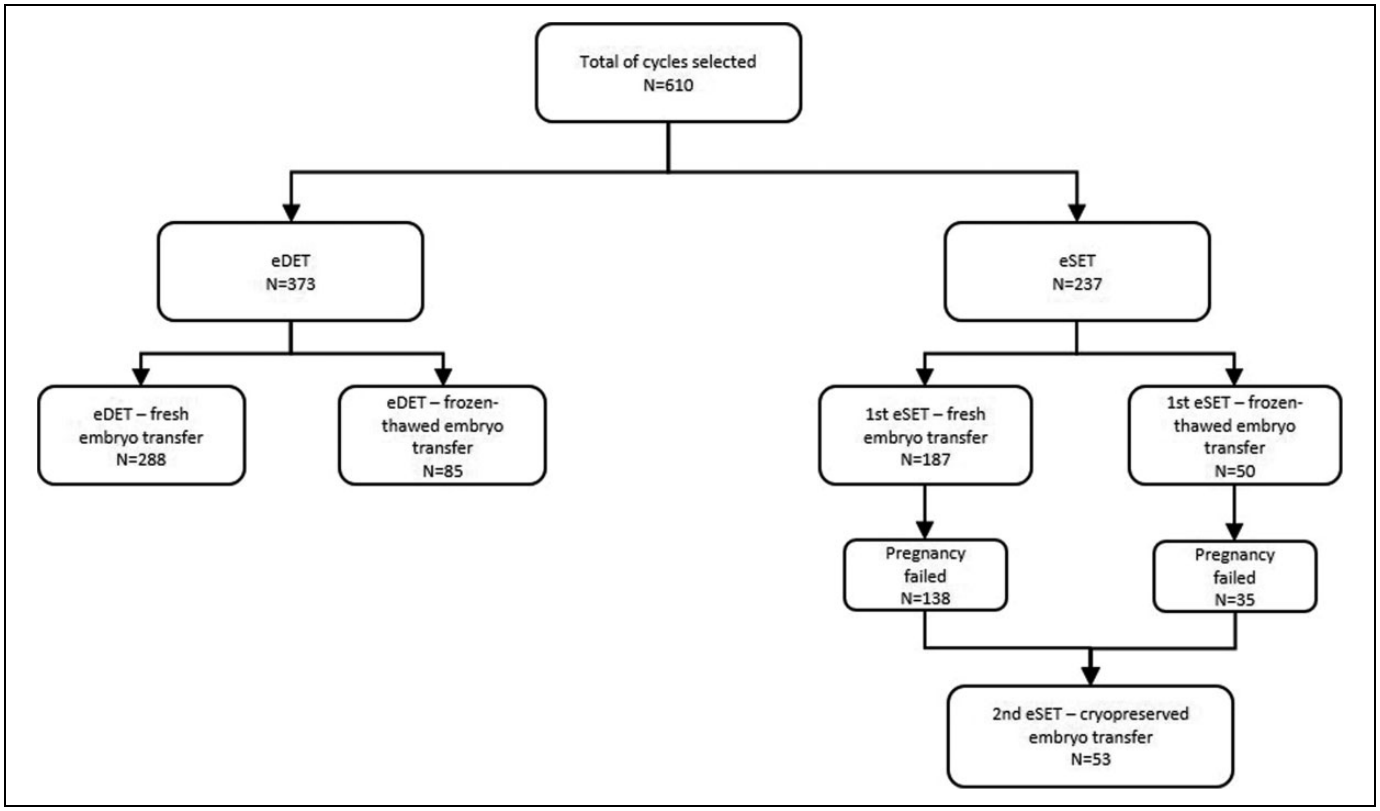

Figure 1. Flow diagram representing the study design.

(Irvine Scientific, USA), following the manufacturer's instructions. For warming, a Vitrification Thaw kit (Irvine Scientific, USA) was used.

For the frozen-thawed ETs, endometrial preparation was conducted with 100 μg of estradiol valerate (Estradot; Novartis, Switzerland) for 14 days plus 600 μg of vaginal micronized progesterone (Utrogestan; Farmoquimica, Brazil) 5 days before the transfer. Blastocysts were warmed, evaluated for survival and morphology, and transferred accordingly 5 days after the use of progesterone was started. Clinical pregnancy was defined by the presence of a gestational sac with a heartbeat at 2 weeks after confirmation of a biochemical pregnancy with a serum β-hCG measurement for fresh or cryopreserved ETs.

Data Collection and Statistical Analysis

Data were obtained from a clinical report form and tabulated for this study. The implantation rate was calculated as the ratio between the number of gestational sacs and the number of embryos transferred, and the pregnancy rate was calculated as the number of patients presenting a clinical pregnancy (defined by presence of gestational sac with a heartbeat) divided by the number of patients with embryos transferred. Additionally, we calculated the cumulative pregnancy rate defined as the pregnancy rate per patient after a fresh SET or after an elective cryopreserved SET. For the calculation of accumulated clinical and multiple pregnancy rates by considering the second ET for patients who did not become pregnant in the first ET, we used a formula previously described by Luke et al.[11] The accumulated clinical pregnancy rate at cycle 2 was equal to the clinical pregnancy rate at the first ET + the clinical pregnancy rate at the second ET × (1 − the clinical pregnancy rate in the first ET). This calculation assumes that there is no contraindication during cycle 1 for continuing into cycle 2. The cumulative multiple pregnancy rate at the second ET was equal to the multiple pregnancy rate at the first ET + the multiple pregnancy rate at the second ET × (1 − the clinical pregnancy rate at the first ET) [11].

Data analyses were performed using SPSS V22 (IBM SPSS Software, USA). Patient demographic data were evaluated using descriptive statistics, which included information on the means and frequencies. Mean comparisons tests (Student *t* test) were used to compare continuous variables, and Pearson chi-square test was used to compare frequencies. Regression analyses were used to evaluate the association between variables, and multivariate models included possible confounders; the results were reported as odds ratios and *P* values. We considered *P* values $\leq .05$ to be statistically significant.

Results

The demographic characteristics of the patients included in the study are presented in Table 1. The groups were similar except for the total gonadotrophin dose administered. However, the gonadotrophin dose was adjusted according to the ovarian response of each patient, aiming for a collection of an adequate

Table 1. Demographic Characteristics of the Patients According to the Study Groups.

	eDET	eSET	*P*
Number of cycles	373	237	
Age, years	33.6 ± 2.9	33.9 ± 3.1	.156
BMI, kg/m^2	23.1 ± 3.4	22.5 ± 3.0	.066
Basal FSH measurement, IU/mL	6.1 ± 3.9	6.3 ± 5.2	.786
Total dose of gonadotrophin administered, IU	1855 ± 500	1709 ± 258	<.001
Number of oocytes recovered	14.0 ± 7.7	13.4 ± 8.1	.344
Number of MII oocytes recovered	11.4 ± 6.4	10.8 ± 6.3	.258
Fertilization rate, %	83.0%	83.8%	.524
Number of embryos cryopreserved	6.6 ± 4.5	7.0 ± 4.5	.216
Cycles with frozen Embryo transfer	85 (22.8%)	50 (21.1%)	.624
Cycles with blastocyst transfer	161 (43.2%)	80 (33.8%)	.021

Abbreviations: BMI, body mass index; eDET, elective double-embryo transfer; eSET, elective single-embryo transfer; FSH, follicle-stimulating hormone.

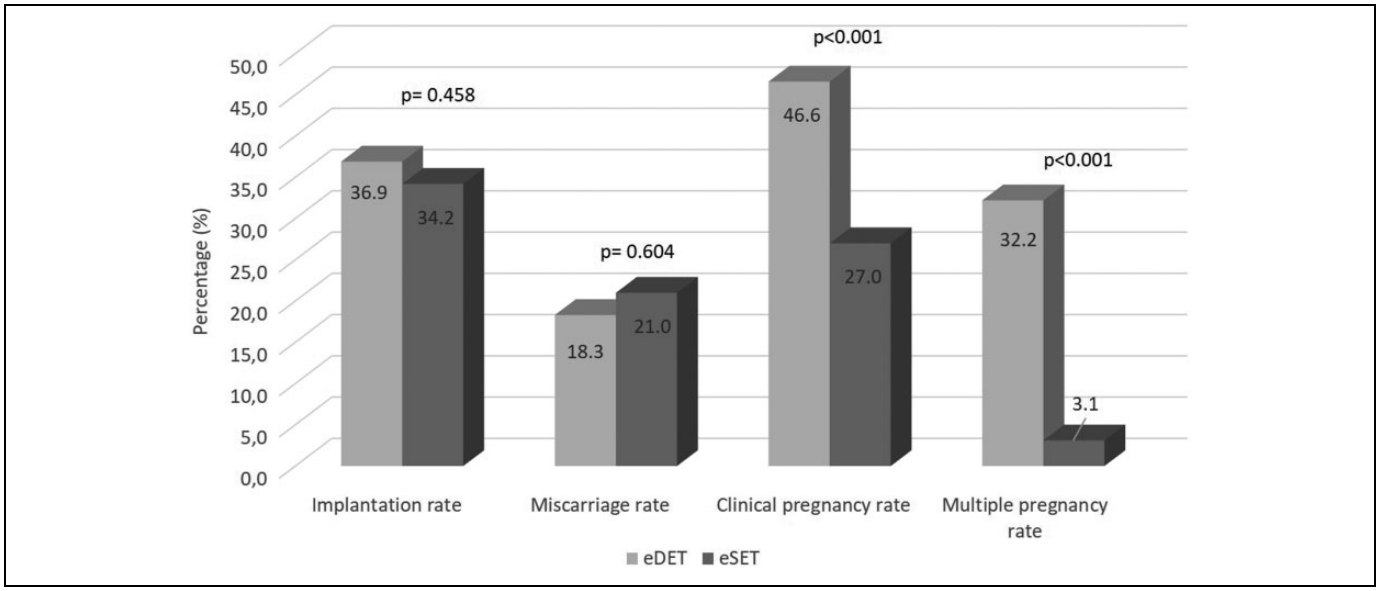

Figure 2. Clinical outcomes after the first embryo transfer (ET) in the elective double-embryo transfer (eDET) and elective single-embryo transfer (eSET) groups.

number of oocytes, which was equivalent between the groups. Thus, this difference was not clinically significant. The infertility factors were classified according to the SART as follows: male factor (35.4%), anatomic female factor (17.9%), endometriosis (17.2%), mixed (9.7%), endocrine female factor (8.5%), unexplained (7.3%), and other (4.0%).

First ET Outcomes

For the first ET, the patients received a fresh or frozen-thawed ET, and the embryos were in the cleavage or blastocyst stage. We compared the eDET versus eSET of the groups independent of whether a fresh or frozen-thawed ET was performed and regardless of the embryo stage. Although the clinical pregnancy rate was significantly lower when 1 embryo was transferred (eSET) compared to 2 embryos (eDET), the multiple pregnancy rate was dramatically higher when 2 embryos were transferred (eDET; Figure 2).

In the eSET group, 173 patients did not become pregnant, and of these patients, 11 did not receive a second ET, 109 patients received 2 frozen-thawed embryos (eSET-DET), and 53 patients received another frozen-thawed SET (eSET-SET). A comparison of these 2 subgroups demonstrated a higher implantation rate in the eSET-SET subgroup but similar pregnancy rates. Moreover, the eSET-DET subgroup had a higher multiple pregnancy rate as expected (Figure 3).

Accumulated Outcomes

We then compared the accumulated outcomes of the eSET-SET group to those of the eDET group and noted similar clinical pregnancy rates after the transfers of 2 embryos during 1 ET (eDET) or over 2 ETs (eSET-SET; Figure 4). If we consider only single pregnancies as a successful outcome, the patients who received 2 embryos over 2 ETs (eSET-SET) had a better

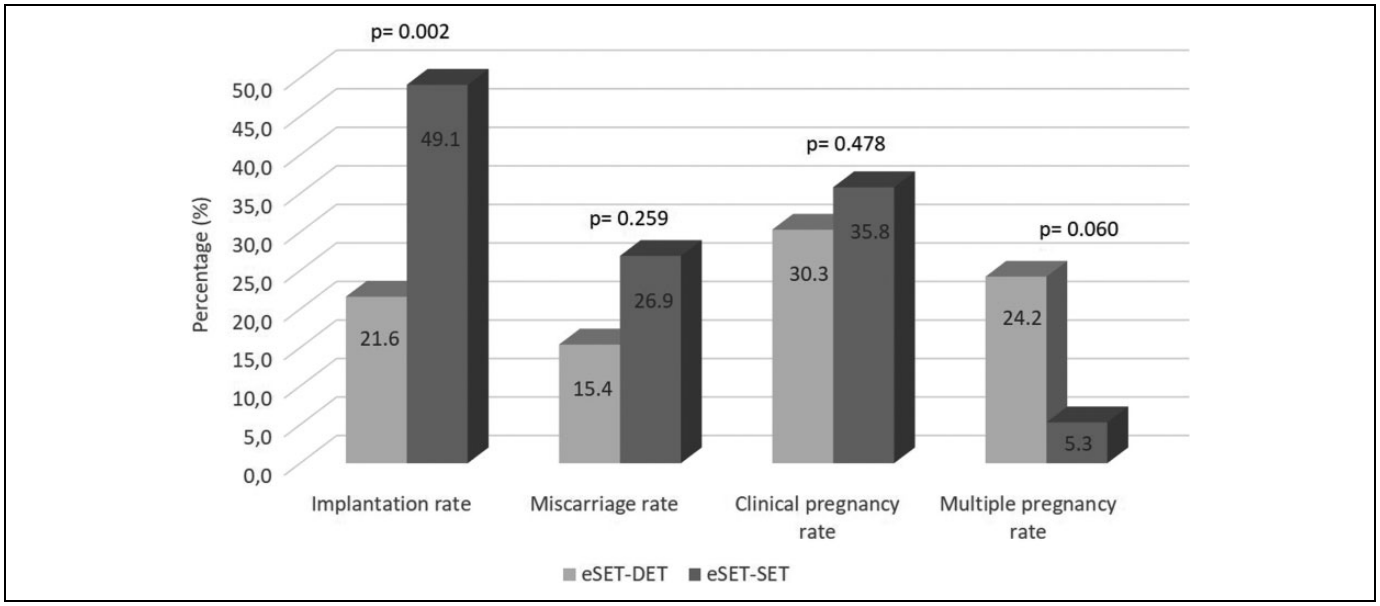

Figure 3. Clinical outcomes of the patients in the elective single-embryo transfer (eSET) group who did not become pregnant after the first embryo transfer (ET) and who received 2 (eSET–double-embryo transfer [DET]) or 1 (eSET-SET) ET as the second ET.

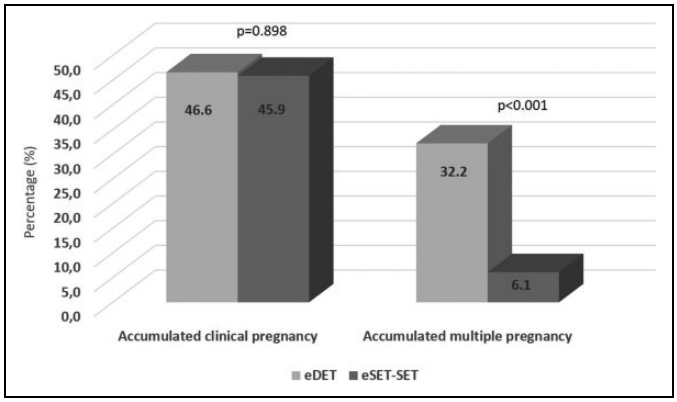

Figure 4. Clinical outcomes after the transfer of 2 embryos for patients in the eDET group compared to the accumulated clinical outcomes in the elective single-embryo transfer (eSET)-SET group.

Table 2. Multiple Logistic Regression Model to Evaluate the Association Between the Transfer of 2 Embryos Over 3 ETs and the Chance of Becoming Pregnant, Adjusted for Confounders.

	Coefficient	SE of the Coefficient	*P*	OR
Transfer of 2 embryos over 2 ETs (eSET-SET)	−0.451	0.314	.151	0.637
Age of women, years	−0.026	0.037	.482	0.974
Dose of gonadotrophin administered, IU	0.000	0.000	.581	1.000
Number of embryos cryopreserved	0.012	0.027	.671	1.012
Transfer of frozen-thawed embryos in the first ET	0.230	0.337	.494	1.259
Transfer of embryos in the blastocyst stage	−0.018	0.234	.938	0.982
Constant	0.458	1.300	.725	1.580

Abbreviations: ET, embryo transfer; eSET, elective single-embryo transfer; SE, standard error; OR, odds ratio.

result (93.9%) than those who received 2 embryos in 1 ET (DET; 67.8%; $P < .001$).

To adjust the chance of becoming pregnant to possible confounders, we built a multiple logistic regression model evaluating the association between the transfer of 2 embryos over 2 ETs and the chance of becoming pregnant by adjusting for the ages of the women, dose of gonadotrophin administered, number of embryos cryopreserved, transfer of frozen-thawed embryos in the first ET, and transfer of embryos in the blastocyst stage in the first ET (Table 2). The model confirmed that the chance of becoming pregnant was not associated with the transfer of 2 embryos over 2 ETs (eSET-SET) compared to one ET (eDET). On the other hand, when we used the same logistic regression model with multiple clinical pregnancy as the dependent variable (Table 3), the transfer of 2 embryos over 2 ETs (eSET-SET) demonstrated a chance of multiple pregnancy that was 8.3 times lower (odds ratio = 0.120; $P = .043$). Table 4 describes the general outcomes of groups, containing frequencies and proportions of clinical pregnancy, miscarriages, ongoing pregnancies, and multiple pregnancies.

Discussion

It is clear that ART is associated with an increasing prevalence of multiple pregnancies that are related to the number of ETs[5] and consequently, with higher maternal and newborn consequences[19-21] and significantly enlarged costs of hospitalization during the first 5 years.[22] Given this scenario, this study confirmed the benefits of transferring 2 embryos over 2 cycles

Table 3. Multiple Logistic Regression Model to Evaluate the Association Between the Transfer of 2 Embryos Over 2 ETs and the Chance of a Multiple Clinical Pregnancy, Adjusted for Confounders.

	Coefficient	SE of the coefficient	*P*	OR
Transfer of 2 embryos over 2 ETs (eSET-SET)	−2.122	1.048	.043	0.120
Age of women, years	−0.046	0.063	.459	0.955
Dose of gonadotrophin administered, IU	0.000	0.000	.724	1.000
Number of embryos cryopreserved	0.018	0.044	.686	1.018
Transfer of frozen-thawed embryos in the first ET	−0.130	0.536	.808	0.878
Transfer of embryos in the blastocyst stage	0.508	0.375	.176	1.661
Constant	0.713	2.154	.741	2.040

Abbreviations: ET, embryo transfer; eSET, elective single-embryo transfer; SE, standard error; OR, odds ratio.

Table 4. General Clinical Outcomes in the Study Groups.

	eDET	eSET (First Transfer)	eSET-SET (Second Transfer)
Number of cycles	373	237	53
Biochemical pregnancies	237 (63.5%)	92 (38.8%)	29 (54.7%)
Clinical pregnancies	213 (57.1%)	81 (34.2%)	26 (49.1%)
Miscarriage	39 (18.3%)	17 (21.0%)	7 (26.9%)
Ongoing pregnancies	174 (46.6%)	64 (27.0%)	19 (35.8%)
Multiple pregnancies	56 (32.2%)	2 (3.1%)	1 (5.3%)

Abbreviations: eDET, elective double-embryo transfer; eSET, elective single-embryo transfer.

[a]The table shows frequencies and percentages. Biochemical, clinical, and ongoing rates of pregnancies were calculated per total number of cycles. Number of miscarriages per number of clinical pregnancies calculated the miscarriage rate; number of multiple pregnancies per number of ongoing pregnancies calculated the multiple pregnancy rate.

toward a good prognosis for infertile couples, focusing on decreasing the incidence of multiple pregnancies.

For this study, we considered couples with a good prognosis, which we defined as couples undergoing their first or second IVF cycle, couples who had 1 (for the eSET group) or 2 (for the eDET group) good-quality embryos with at least 1 surplus good-quality embryo cryopreserved after transfer, and women up to 38 years of age, independent of other characteristics. Despite the strict inclusion criteria, the retrospective design of this study introduced the possibility of confounders, so the outcomes were also evaluated by performing multiple logistic regression analyses that were adjusted for the age, dose of gonadotropin administered, transfer of frozen-thawed embryos, stage of the ET, and number of embryos cryopreserved, aiming to eliminate possible biases. However, none of these variables was associated with the outcomes.

We also evaluated the second ET of 1 or 2 embryos for patients who failed after the first transfer in the eSET group. We confirmed the previous results of our group by demonstrating that for patients with a good prognosis who failed to conceive in the first eSET, there was no advantage of undergoing a DET in the subsequent ET compared to an SET. Additionally, the DET approach for the second ET maintained a high incidence of multiple pregnancies.[23]

The use of eSET in the United States and Latin America (LA) is lower than in European countries, where the government limits the number of embryos transferred and where treatments are financially supported by the public health system. This difference can be attributed to the fact that the decision-making process regarding the number of embryos transferred in the United States and LA is the shared responsibility of the couple and their doctor.[8] While eSET is preferable when possible and agreed upon by the doctor and couple, the transfer of a higher number of embryos is entertained if desired by the patient,[24] and the DET remains the most common practice worldwide.

Considering all of these factors, the eSET policy should be promoted. On the other hand, both provider and patient perceptions of eSET are negative even when their preferences are not favorable to multiple pregnancies.[25] There are a number of strategies and tools that may encourage the more widespread adoption of eSET in clinical practice, including personalized counseling, educational information regarding the risks of multiple pregnancies and births, financial incentives, and tools to help predict the chances of IVF success.[26] These actions seem to be efficient toward reducing the preferences of patients for transferring higher numbers of embryos and, sometimes, for twin pregnancies.[27,28]

The limitations of this study include its retrospective design and the variability in patient profiles. However, the multiple regression analysis assessed possible confounders and showed that they did not influence the outcomes. The choice about the number of embryos to be transferred was performed as a shared decision-making process between patients and doctors, after an explanation of advantages and disadvantages of each situation. This characterizes another limitation of our study, as there were no clear rules to transfer 1 or 2 embryos for each couple. The recommendation about the number of embryos to be transferred in Brazil does not indicate an SET, and a shared decision-making process is routine in reproductive medicine centers. Another flaw is that not all patients in the eSET group who failed after the first ET had a second SET, and some of them received a DET, which resulted in a reduced number of patients evaluated in the accumulated clinical outcome calculations for the eSET-SET subgroup. On the other hand, this situation is also a consequence of a system in which a shared decision-making process regarding the number of ET is current and reflects the perceptions of the couples that the transfer of 2 embryos can increase the chances of a live birth, motivating them to choose a DET after an eSET failure. Studies have shown patient education about the morbidity and mortality of mother and newborns as consequence of multiple pregnancies,

as the costs of those situations, are effective to decrease the preferences for twin pregnancies and stimulate the SETs.[27,28] Finally, the indirect cost of IVF treatment considering the obstetric outcomes could not be evaluated in this study because the patients who became pregnant were not followed in the IVF clinic, and thus, data could not be obtained.

Despite discussions of the advantages and pitfalls of eSET in the literature,[24,29,30] we propose, based on our findings, that the eSET policy should be promoted for couples with a good prognosis. The eSET policy maintains the accumulated clinical pregnancy rates, avoids multiple pregnancies, and consequently avoids the maternal and neonatal complications and indirect costs of treatment such as obstetrics, in which spending is reduced.

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Declaration of Conflicting Interests

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References

1. Sunderam S, Kissin DM, Crawford SB, et al. Assisted reproductive technology surveillance—United States, 2014. *MMWR Surveill Summ*. 2017;66(6):1-24.
2. Kallen B, Finnstrom O, Nygren KG, Olausson PO. Temporal trends in multiple births after in vitro fertilisation in Sweden, 1982-2001: a register study. *BMJ*. 2005;331(7513):382-383.
3. Källén B, Finnström O, Lindam A, Nilsson E, Nygren KG, Otterblad Olausson P. Trends in delivery and neonatal outcome after in vitro fertilization in Sweden: data for 25 years. *Hum Reprod*. 2010;25(4):1026-1034.
4. Klemetti R, Gissler M, Hemminki E. Comparison of perinatal health of children born from IVF in Finland in the early and late 1990s. *Hum Reprod*. 2002;17(8):2192-2198.
5. Kallen B, Finnstrom O, Nygren KG, Olausson PO. In vitro fertilization (IVF) in Sweden: infant outcome after different IVF fertilization methods. *Fertil Steril*. 2005;84(3):611-617.
6. Min JK, Breheny SA, MacLachlan V, Healy DL. What is the most relevant standard of success in assisted reproduction? The singleton, term gestation, live birth rate per cycle initiated: the BESST endpoint for assisted reproduction. *Hum Reprod*. 2004;19(1):3-7.
7. Zegers-Hochschild F, Schwarze JE, Crosby JA, Musri C, Urbina MT; Latin American Network of Assisted Reproduction (REDLARA). Assisted reproductive techniques in Latin America: The Latin American Registry, 2013. *JBRA Assist Reprod*. 2016; 20(2):49-58.
8. Maheshwari A, Griffiths S, Bhattacharya S. Global variations in the uptake of single embryo transfer. *Hum Reprod Update*. 2011; 17(1):107-120.
9. Kissin DM, Kulkarni AD, Kushnir VA, Jamieson DJ; ARTSSG National. Number of embryos transferred after in vitro fertilization and good perinatal outcome. *Obstet Gynecol*. 2014;123(2 Pt 1):239-247.
10. Steinberg ML, Boulet S, Kissin D, Warner L, Jamieson DJ. Elective single embryo transfer trends and predictors of a good perinatal outcome—United States, 1999 to 2010. *Fertil Steril*. 2013; 99(7):1937-1943.
11. Luke B, Brown MB, Wantman E, et al. Application of a validated prediction model for in vitro fertilization: comparison of live birth rates and multiple birth rates with 1 embryo transferred over 2 cycles vs 2 embryos in 1 cycle. *Am J Obstet Gynecol*. 2015; 212(5):676 e1-e7.
12. McLernon DJ, Harrild K, Bergh C, et al. Clinical effectiveness of elective single versus double embryo transfer: meta-analysis of individual patient data from randomised trials. *BMJ*. 2010;341: c6945.
13. Crawford S, Boulet SL, Mneimneh AS, et al. Costs of achieving live birth from assisted reproductive technology: a comparison of sequential single and double embryo transfer approaches. *Fertil Steril*. 2016;105(2):444-450.
14. Practice Committee of Society for Assisted Reproductive, T and M Practice Committee of American Society for Reproductive. Elective single-embryo transfer. *Fertil Steril*. 2012;97(4):835-842.
15. CFM, CFdM, RESOLUÇÃO CFM N° 2.013/13. NORMAS ÉTICAS PARA A UTILIZAÇÃO DAS TÉCNICAS DE REPRODUÇÃO ASSISTIDA. Brasilia; 2013.
16. Palermo G, Joris H, Devroey P, Van Steirteghem AC. Pregnancies after intracytoplasmic injection of single spermatozoon into an oocyte. *Lancet*. 1992;340(8810):17-18.
17. Veeck LLAn atlas of human gametes and conceptuses: an illustrated reference for assisted reproductive technology. *The Encyclopedia of Visual Medicine Series*. New York: Parthenon Pub. Group; 1999:215.
18. 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(6): 1155-1158.
19. Zollner U, Dietl J. Perinatal risks after IVF and ICSI. *J Perinat Med*. 2013;41(1):17-22.
20. Chambers GM, Hoang VP, Lee E, et al. Hospital costs of multiple-birth and singleton-birth children during the first 5 years of life and the role of assisted reproductive technology. *JAMA Pediatr*. 2014;168(11):1045-1053.
21. Murray SR, Norman JE. Multiple pregnancies following assisted reproductive technologies—a happy consequence or double trouble? *Semin Fetal Neonatal Med*. 2014;19(4):222-227.
22. van Heesch MM, Evers JL, van der Hoeven MA, et al. Hospital costs during the first 5 years of life for multiples compared with singletons born after IVF or ICSI. *Hum Reprod*. 2015;30(6): 1481-1490.
23. Monteleone PA, Mirisola RJ, Goncalves SP, Baracat EC, Serafini PC. Outcomes of elective cryopreserved single or double embryo

transfers following failure to conceive after fresh single embryo transfer. *Reprod Biomed Online*. 2016;33(2):161-167.
24. Adashi EY, Gleicher N. Is a blanket elective single embryo transfer policy defensible? *Rambam Maimonides Med J*. 2017;8(2).
25. Rai V, Betsworth A, Beer C, Ndukwe G, Glazebrook C. Comparing patients' and clinicians' perceptions of elective single embryo transfer using the attitudes to a twin IVF pregnancy scale (ATIPS). *J Assist Reprod Genet*. 2011;28(1):65-72.
26. Tobias T, Sharara FI, Franasiak JM, Heiser PW, Pinckney-Clark E. Promoting the use of elective single embryo transfer in clinical practice. *Fertil Res Pract*. 2016;2:1.
27. Murray S, Shetty A, Rattray A, Taylor V, Bhattacharya S. A randomized comparison of alternative methods of information provision on the acceptability of elective single embryo transfer. *Hum Reprod*. 2004;19(4):911-916.
28. Hope N, Rombauts L. Can an educational DVD improve the acceptability of elective single embryo transfer? A randomized controlled study. *Fertil Steril*. 2010;94(2):489-495.
29. Gleicher N, Kushnir VA, Barad DH. Elective single-embryo transfer (eSET) reduces pregnancy rates and should only be used in exceptional circumstances: FOR: the statistically flawed model of eSET. *BJOG*. 2017;124(5):755.
30. Miller LM. 'Elective single embryo transfer (eSET) reduces pregnancy rates and should only be used in exceptional circumstances': AGAINST: SET maintains live birth rates and provides unique advantages. *BJOG*. 2017;124(5):756.