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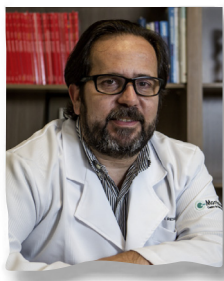
# Outcomes of elective cryopreserved single or double embryo transfers following failure to conceive after fresh single embryo transfer



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**Abstract** The main adverse effect of IVF is the high multiple pregnancy rate resulting from the transfer of two or more embryos. The objective was to evaluate pregnancy rates in infertile women with a good prognosis who failed to conceive in a fresh elective single embryo transfer (eSET) and had a second cycle with elective double vitrified-warmed embryo transfer (eDFET) compared with elective single vitrified-warmed embryo transfer (eSFET). A total of 142 intracytoplasmic sperm injection cycles using a conventional protocol were evaluated. Good-prognosis patients underwent eSET in a fresh cycle, and those who failed to conceive underwent a second vitrified-warmed embryo transfer: eDFET ( $n = 102$ ) or eSFET ( $n = 40$ ). Embryos were transferred and vitrified on day 5 of development. Patients who received eDFET had fewer implantations (30.9%) than eSFET (52.5%;  $P = 0.004$ ); pregnancy rates were similar (eDFET: 35.3%, eSFET: 42.5%). Patients with the eSFET had one monozygotic twin (5.9%), and 22.2% of eDFET patients had multiple pregnancies. Patients with a good prognosis who failed to conceive in the first fresh eSET did not have an advantage when receiving eDFET in the second cycle, as pregnancy rates were similar; 22.2% of patients in the eDFET group had multiple pregnancies.



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**KEYWORDS:** Infertility, IVF, Single embryo transfer, Double embryo transfer, Multiple pregnancies

## Introduction

The demand for assisted reproductive techniques has increased in the past 3 decades owing to a number of factors. These include more older women wishing to become pregnant, more sexually transmitted diseases, higher prevalence of obesity and medical issues such as endometriosis and polycystic ovary syndrome. Despite its success, IVF causes high rates of multiple pregnancies resulting from the transfer of two or more embryos after ovarian stimulation with the aim of increasing the chance of a pregnancy (Naasan et al., 2012; Kupka et al., 2014; Ishihara et al., 2015). Multiple pregnancies are the main adverse effect of IVF and are associated with a high risk of complications to both the mothers and fetuses, as well as potential long-term health issues for both. Multiple pregnancies raise the rate of premature births and intrauterine growth retardation, which contribute to the significantly higher rate of morbidity and mortality (Zollner and Dietl, 2013). Prematurity is six times more frequent; therefore, birth weight is significantly lower, which exposes twins to prematurity-related disorders (respiratory, cardiovascular, infectious) and long-term complications (especially neurological disabilities) (Giuffre et al., 2012).

It is therefore in the interests of public health to reduce multiple pregnancy rates in IVF cycles. The fewer number of embryos transferred is encouraged, as is the subsequent reduction in multiple pregnancies. The average number of embryos transferred, however, varies widely among countries. Reasons for this are multifactorial but consumer affordability will affect access to assisted reproduction techniques (Chambers et al., 2014). In general, the proportion of elective single embryo transfers (eSET) has increased (mean of 23.4% of cycles), and higher rates are seen in Sweden and Finland, which reported eSET rates in 2010 of 73.3% and 67.5%, respectively, with no reduction in pregnancy rates (around 30%) (Kupka et al., 2014). Other countries, such as the USA and Brazil presented lower eSET rates of 10% (Ishihara et al., 2015).

In a meta-analysis, eSET was shown to reduce the risk of multiple pregnancies and decrease live birth rates compared with elective double embryo transfer (eDET) (Baruffi et al., 2009; Pandian et al., 2009). Other studies have shown that when eSET is carried out, and the subsequent cryopreserved embryo transfers are taken into account, the cumulative pregnancy and live birth rates are similar to eDET (McLernon et al., 2010; Pandian et al., 2013). Therefore, in 2009, the Human Fertilization and Embryology Authority introduced a policy to encourage routine use of eSET, with a resulting reduction of multiple births from 24% in 2009 to 10% in 2012 (Harbottle et al., 2015). More recently, in IVF patients with good prognosis, specifically women aged younger than 37 years in their first or second IVF cycle and along with good-quality embryos, eSET is recommended by the Practice Committee of American Society for Reproductive Medicine (Practice Committee of American Society for Reproductive Medicine, Practice Committee of Society for Assisted Reproductive Technology, 2013).

In clinical practice, when an eSET results in failure, the decision to use eDET of cryopreserved embryos is common. On this basis, we hypothesized that an eDET is not beneficial in improving pregnancy rates in good-prognosis patients,

even after an eSET failure. Hence, the aim of this study was to evaluate the pregnancy outcomes of IVF cycles of patients who failed to conceive in the fresh eSET and underwent a following elective double-vitrified-warmed embryo transfer (eDFET) or an elective single-vitrified-warmed embryo transfer (eSFET) and to compare the rates of pregnancies and multiple pregnancies.

## Material and methods

In this retrospective observational study, IVF cycles were evaluated at the Human Reproduction Centre, Hospital das Clínicas, Faculdade de Medicina, Universidade de Sao Paulo, and a private assisted reproduction centre in Sao Paulo, Brazil (Monteleone, Centro de Reprodução Humana) between 2007 and 2015. All of the procedures in this study are part of routine care in the assisted reproductive centre, and written informed consent was obtained from all patients before treatment. Patients consented to the treatment procedures and retrospective data use in scientific publications (Ethics Committee Proc. Number 1.151.345).

## Study groups

Patients were designated for eSET according to the criteria of the study centre and were considered to have a good prognosis if they met the following criteria: patients aged between 18 and 38 years undergoing first or second fresh IVF cycle; at least four oocytes collected characterizing no poor responders at ovarian stimulation; and good-quality blastocysts available for transfer with at least two surplus good-quality blastocysts cryopreserved after transfer.

In Brazil, the law states that patients younger than 38 years can transfer a maximum of three embryos. The risks and benefits of transfer of one or more embryos were explained and the couples then decided on the number of embryos to transfer. Two hundred and thirty-four patients received a fresh eSET, and 58 became pregnant (24.8%). Of the 176 patients who failed to conceive, 142 underwent a second cycle by frozen embryo transfer (FET). It was defined as eSFET ( $n = 40$ ) and eDFET ( $n = 102$ ) patients who had at least two spare good-quality blastocysts that were cryopreserved and who had one or two cryopreserved good-quality blastocysts transferred, respectively (Figure 1).

## IVF protocol

Briefly, pituitary blockage was obtained either with a GnRH agonist (Lupron kit™, Abbot SA Société Française des Laboratoires, France) or a GnRH antagonist (Cetrotide®, Serono, Switzerland). Ovarian stimulation was accomplished using recombinant FSH (rFSH, Gonal-F®, Serono, Switzerland). When at least two follicles reached a diameter of 18 mm, follicular maturation was triggered with an injection of 250 µg recombinant HCG (rhCG, Ovidrel®, Serono, Switzerland). Oocyte retrieval was carried out after 35–36 h by transvaginal ultrasound-guided aspiration; the luteal phase was supported by 90 mg of daily progesterone (Crinone®, Serono,

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