Cumulative live birth rate in freeze-all cycles is comparable to that of a conventional embryo transfer policy at the cleavage stage but superior at the blastocyst stage

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Objective: To determine whether the freeze-all policy ensures a higher efficacy in terms of cumulative live birth rate (CLBR) in comparison with a conventional fresh/frozen embryo transfer (ET) approach in patients with normal ovarian response.
Design: Retrospective, matched, multicenter cohort study.
Setting: Private IVF centers.
Patient(s): This study analyzed 564 completed IVF cycles in which an average of 12–18 oocytes were retrieved. In 435 cycles the conventional strategy was applied, with initial ET followed by frozen embryo replacements, whereas in 129 cycles the freeze-all policy was performed, with elective cryopreservation and deferred use of all viable embryos.
Intervention(s): None.
Main Outcome Measure(s): The primary endpoint was CLBR. The secondary endpoint was cumulative clinical pregnancy rate.
Result(s): Overall, statistically comparable CLBRs were achieved in the fresh/frozen and freeze-all groups (45.5% vs. 53.5%). Stratification of data for age and number of retrieved oocytes confirmed the absence of differences between the two groups. In a subanalysis in which the day of ET and cryopreservation were taken into account, a similar outcome was achieved in cleavage-stage groups (45.6% vs. 46.4%), whereas when ET was performed at the blastocyst stage the CLBR was significantly higher in the freeze-all group (45.3% vs. 66.7%).
Conclusion(s): Our CLBR analysis indicates that clinical performance of the freeze-all policy is equivalent to that of the conventional strategy when ET is carried out at the cleavage stage. However, it seems to be superior if associated with cryopreservation and transfer at the blastocyst stage. (Fertil Steril® 2018;110:703–9. ©2018 by American Society for Reproductive Medicine.)
El resumen está disponible en Español al final del artículo.

Key Words: Blastocyst stage, cumulative live birth, embryo transfer, freeze-all, IVF

In vitro fertilization has resulted in the births of more than 5 million infants worldwide from 1978 (1). Over the years there have been considerable advances in therapies and in laboratory techniques of reproductive medicine that have allowed the individualization of IVF cycles with the aim to maximize results and decrease treatment risks and costs (2).

Treatment with IVF involves a number of consecutive steps. Conventionally the IVF cycle starts with controlled ovarian hyperstimulation (COH) and ends with the transfer at the cleavage or blastocyst stage of the best available embryo(s); all surplus embryos of adequate quality are cryopreserved and stored for later use when no pregnancy has been achieved or when couples wish to achieve a second pregnancy (3). In certain circumstances a fresh transfer cannot be performed, and the entire cohort of viable embryos is cryopreserved. There are different indications for this policy, referred to as "freeze-all": decrease in the risk of ovarian hyperstimulation syndrome (OHSS); postponement of transfer to obtain a more receptive uterine environment in a subsequent

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cycle; fertility preservation; prevention of the effects of premature P elevation; and co-ordination with the treatment options requiring results of genetic tests (4–9).

It is well recognized that cryopreservation is an essential aspect of assisted reproductive technology because it allows increase of the safety and efficacy of IVF treatments (10). The systematic application of cryopreservation is inspired by new indications, such as cycle segmentation (11), oocyte banking (12, 13), and preimplantation genetic testing at the blastocyst stage (14). Cryopreservation techniques also offer the opportunity to improve cumulative results per treatment cycle in terms of pregnancy and live birth rate (LBR), because COH leads to the development and maturation of many follicles and oocytes and consequently the generation of supernumerary embryos.

It is also known, however, that ovarian stimulation is associated with supraphysiologic steroid hormone levels, which are the cause of modifications in the preimplantation endometrium (15, 16), biochemical and morphologic endometrial alterations, and advancement of endometrial receptivity (17, 18). These modifications might affect the success of fresh ET, because embryo implantation depends on embryo quality, endometrial receptivity, and embryo–endometrium interactions (19). Consistent with this view, to date, some authors described comparable or better pregnancy rates in frozen–thawed ET compared with fresh ET (5, 6, 20, 21).

Therefore, the main purpose of this study was to analyze the cumulative success rate of the conventional strategy in comparison with elective cryopreservation of all viable embryos. Data were obtained from a large data set and analyzed in relation to number of oocytes retrieved, because there is a strong correlation between the number of oocytes retrieved and cumulative LBR (CLBR) (22), female age, and day of ET to define the optimal approach for different subclasses of patients. Crucially, data were analyzed also taking into account the stage of ET and cryopreservation.

MATERIALS AND METHODS

We performed a retrospective matched cohort study. Patients underwent IVF cycles between 2012 and 2016, involving five fertility centres in Italy.

We included cycles in which fresh ET were performed (fresh cycles) and cycles in which all embryos were frozen, followed by a later transfer (freeze-all). We included women aged 22–40 years from whom between 12 and 18 oocytes were recovered, because they are patients with better prognosis, in whom embryo cryopreservation is a realistic chance. Only patients who transferred all the viable embryos available and/or achieved a live birth were included. This choice was motivated by the aim to evaluate the CLBR as the primary outcome.

We did not include cancelled cycles, poor-responder patients, and cycles in which preimplantation genetic screening or preimplantation genetic diagnosis was applied. Five hundred sixty-four completed cycles (129 freeze-all and 435 fresh cycles) were suitable for matching and analysis. Characteristics of cycles are showed in Table 1.

Study Protocol

Controlled ovarian hyperstimulation was performed with either recombinant FSH (Gonal-F, Merck Serono) or hMG (Meropur, Ferring), with starting dose ranging from 100 IU to 450 IU per day, according to hormonal and anthropometric parameters. Ovarian stimulation protocol and blastocyst grading procedures were carried out as previously described (23). The gonadotropin dose was adjusted according to the individual follicular response, and GnRH analogues were used to avoid the LH spontaneous surge.

Oocytes were retrieved transvaginally 35 to 36 hours after hCG (Gonasi, IBSA) administration and fertilized using either conventional IVF or intracytoplasmic sperm injection (ICSI). Four to five hours after oocyte pickup, conventional IVF was carried out using a final motile sperm concentration of 200,000–300,000/mL. As for ICSI cases (24), cumulus cells were removed from companion oocytes (25). The embryos obtained were then cultured up to cleavage or blastocyst stage. Grading of embryo quality at cleavage stage was expressed, evaluating and scoring details of their appearance according to cell number, blastomere regularity, and degree of fragmentation. Blastocysts were evaluated according to the degree of expansion and quality of the inner cell mass and trophectoderm cell, as previously described by us (26).

The number of transferred embryos was decided according to patient needs and national guidelines. Surplus oocytes

TABLE 1

Diagnosis of infertility of fresh and freeze-all groups.

Diagnosis	All cycles (%) (n = 564)	Fresh cycles (%) (n = 435)	Freeze-all cycles (%) (n = 129)	*P* value
Uterine	0.7	0.9	0	.27
Idiopathic	23.4	23.2	24.0	.84
Endometriosis	3.9	3.9	3.9	.98
Tubal	13.3	14.3	10.1	.22
Male factor	35.8	37.2	31.0	.87
Diminished ovarian reserve	0.4	0.2	0.8	.36
Genetic	2.0	1.6	3.1	.28
Multiple (female)	2.3	2.3	2.3	.98
Multiple (male and female)	12.9	12.0	16.3	.19
Endocrine/anovulatory	5.3	4.4	8.5	.06

Zacà. Cumulative births in freeze-all cycles. Fertil Steril 2018.