ORIGINAL ARTICLE

Transfer of Fresh versus Frozen Embryos in Ovulatory Women

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ABSTRACT

BACKGROUND

Elective frozen-embryo transfer has been shown to result in a higher live-birth rate than fresh-embryo transfer among anovulatory women with the polycystic ovary syndrome. It is uncertain whether frozen-embryo transfer increases live-birth rates among ovulatory women with infertility.

METHODS

In this multicenter, randomized trial, we randomly assigned 2157 women who were undergoing their first in vitro fertilization cycle to undergo either fresh-embryo transfer or embryo cryopreservation followed by frozen-embryo transfer. Up to two cleavage-stage embryos were transferred in each participant. The primary outcome was a live birth after the first embryo transfer.

RESULTS

The live-birth rate did not differ significantly between the frozen-embryo group and the fresh-embryo group (48.7% and 50.2%, respectively; relative risk, 0.97; 95% confidence interval [CI], 0.89 to 1.06; P=0.50). There were also no significant between-group differences in the rates of implantation, clinical pregnancy, overall pregnancy loss, and ongoing pregnancy. Frozen-embryo transfer resulted in a significantly lower risk of the ovarian hyperstimulation syndrome than fresh-embryo transfer (0.6% vs. 2.0%; relative risk, 0.32; 95% CI, 0.14 to 0.74; P=0.005). The risks of obstetrical and neonatal complications and other adverse outcomes did not differ significantly between the two groups.

CONCLUSIONS

The live-birth rate did not differ significantly between fresh-embryo transfer and frozen-embryo transfer among ovulatory women with infertility, but frozen-embryo transfer resulted in a lower risk of the ovarian hyperstimulation syndrome. (Funded by the National Key Research and Development Program of China and the National Natural Science Foundation of China; Chinese Clinical Trial Registry number, ChiCTR-IOR-14005406.)

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In vitro fertilization (IVF) has evolved rapidly since its inception 40 years ago. Advancements include controlled ovarian stimulation with gonadotropin, which multiplied the number of oocytes retrieved, and embryo cryopreservation, which made it possible to conserve surplus embryos for further use, thus increasing cumulative live-birth rates after an IVF cycle. Observational studies[1,2] and small randomized, controlled trials[3-5] have shown higher pregnancy rates and better perinatal outcomes with frozen-embryo transfer than with fresh-embryo transfer. It has been hypothesized that frozen-embryo transfer may provide a more favorable intrauterine environment for embryo implantation and placentation by avoiding the supraphysiologic condition that occurs after ovarian stimulation.

We previously conducted a randomized, controlled trial involving women with the polycystic ovary syndrome, in which we compared fresh-embryo transfer with elective freezing of all embryos followed by frozen-embryo transfer. Women who had been assigned to frozen-embryo transfer had a higher live-birth rate than those who had been assigned to fresh-embryo transfer, which was explained largely by a lower rate of pregnancy loss. Women in the frozen-embryo group also had a lower rate of the ovarian hyperstimulation syndrome, but they had a higher rate of preeclampsia.[6] However, patients with the polycystic ovary syndrome have a different reproductive, metabolic milieu[7] than do other women with infertility; it is characterized by hyperandrogenism and insulin resistance, and these patients typically have a greater ovarian response to gonadotropin stimulation[8] than do ovulatory women undergoing IVF. We designed the present randomized, controlled trial to assess the effects of frozen-embryo transfer, as compared with fresh-embryo transfer, on the rates of live birth and maternal and neonatal complications among ovulatory women.

Material and Methods

Trial Design and Oversight

We conducted a multicenter, randomized, controlled trial involving women who underwent their first cycle of IVF with or without intracytoplasmic sperm injection, comparing fresh-embryo transfer with the freezing of all embryos followed by frozen-embryo transfer. The trial was approved by the ethics committees at the Center for Reproductive Medicine, Shandong Provincial Hospital–Shandong University, and the other 19 participating clinical sites in China. All the couples (i.e., both the female and male partners) provided written informed consent before participation.

All data entry, data management, and analyses were coordinated or performed at Shandong University, which was the data-coordinating center for this trial. The first and the last authors assume responsibility for the accuracy and completeness of the data and analyses, and all the authors vouch for the fidelity of the trial to the protocol (available with the full text of this article at NEJM.org). There was no commercial support for this trial.

Block randomization was performed, with a dynamic block size of two, four, or six, with stratification according to trial site. The randomization sequence was generated and kept by the data-coordinating center and was not accessible to the investigators who enrolled patients. The randomization scheme was entered into an online central randomization database (www.medresman.org). Randomization by means of the online central randomization system occurred on the day of oocyte retrieval. Eligible participants were assigned in a 1:1 ratio to the fresh-embryo group or the frozen-embryo group. After randomization, the patients and their physicians were informed about the trial-group assignment.

Trial Population

This trial included women with regular menses who were undergoing their first cycle of IVF or intracytoplasmic sperm injection because of tubal factors, male factors, or both. Tubal factors included unilateral or bilateral tubal occlusion, peritubal adhesion, unilateral or bilateral salpingectomy, or tubal ligation. Male-factor infertility included oligospermia, asthenospermia, or obstructive azoospermia. Eligible women were 20 to 35 years of age, had a normal menstrual cycle (defined as a spontaneous cycle length of ≥21 days and ≤35 days), and a duration of infertility of more than 1 year. Women with a history of unilateral oophorectomy, recurrent spontaneous abortion, diagnosis of the polycystic ovary syndrome, or uterine abnormality (e.g., müllerian duct anomaly, adenomyosis, submucous myoma, intra-

uterine adhesion, or scarred uterus) were excluded. Women were also excluded if they had a chronic medical condition that has been associated with adverse pregnancy outcomes, such as hypertension, symptomatic heart disease, diabetes mellitus, liver disease or dysfunction (according to the results of serum liver-enzyme testing), renal disease or abnormal renal function, severe anemia, history of deep venous thrombosis, pulmonary embolus, or cerebrovascular accident. All the couples were screened with the use of karyotyping, and those with an abnormal karyotype were excluded.

PROCEDURES

A gonadotropin-releasing–hormone (GnRH) antagonist regimen was used for ovarian stimulation in all the patients. Recombinant follicle-stimulating hormone (Puregon, MSD) at a dose of 75 to 225 IU per day was administered on day 3 of the menstrual cycle, at the discretion of the local investigators. The monitoring of ovarian response, adjustment of the dose of gonadotropin, and timing of the triggering of the final oocyte maturation during ovarian stimulation were done as previously reported.[6] A GnRH antagonist (ganirelix [Orgalutran, MSD]) at a dose of 0.25 mg daily was initiated when at least one follicle was 12 mm or more in the mean diameter (calculated from two diameters in the largest section), and treatment was continued until the day of human chorionic gonadotropin (hCG) administration.

Oocyte retrieval was performed 34 to 36 hours after the administration of hCG. On the day of oocyte retrieval, only patients who had five or more oocytes retrieved underwent randomization. At that time, women who were at high risk for the ovarian hyperstimulation syndrome, as determined by local investigators, were excluded. Embryos were scored according to the morphologic criteria of Puissant et al.[9]

In women who were assigned to the fresh-embryo group, luteal-phase support with vaginal progesterone gel (Crinone, Merck Serono) at a dose of 90 mg daily and oral dydrogesterone (Duphaston, Abbott) at a dose of 10 mg twice daily was started immediately after oocyte retrieval and was continued until the day of serum hCG testing. On day 2 or 3 of the embryo culture, up to two embryos were selected and transferred.[10] Two weeks after transfer, the serum level of hCG was measured. In women with a positive hCG test, the use of vaginal progesterone gel was continued until the clinical pregnancy evaluation was conducted by means of ultrasonography; dydrogesterone was continued until 10 weeks of gestation.

In women who were assigned to the frozen-embryo group, all the embryos were vitrified. Two good-quality embryos were vitrified on day 2 or day 3, and the other embryos could be vitrified at the cleavage or blastocyst stage. At the second spontaneous menstrual cycle after oocyte retrieval, natural ovulation was monitored by means of ultrasonography. Luteal-phase support was started from the day of ovulation with oral dydrogesterone at a dose of 10 mg twice a day and was continued until the day of serum hCG testing. Up to two day 2 or day 3 frozen embryos were thawed and transferred 2 or 3 days, respectively, after ovulation. In women with a positive hCG test, dydrogesterone was continued until 10 weeks of gestation. If the natural ovulation cycle was canceled owing to anovulation or poor endometrial development, an artificial cycle was used for endometrial preparation in the next menstrual cycle. Estradiol valerate (Progynova, Delpharm Lille) at a dose of 4 to 8 mg per day was begun on day 2 or day 3 of the menstrual cycle. When the endometrial thickness reached at least 7 mm, vaginal progesterone gel at a dose of 90 mg per day and oral dydrogesterone at a dose of 10 mg twice daily was added. Up to two day 2 or day 3 frozen embryos were thawed and transferred 2 or 3 days, respectively, after the start of progesterone.

If conception occurred, transvaginal ultrasonography was performed 3 weeks later to confirm clinical pregnancy, which was defined as the detection of an intrauterine gestational sac. Ultrasonography was repeated at 11 weeks of gestation to confirm ongoing pregnancy, which was defined as a viable pregnancy with a fetal heartbeat. Information regarding the outcome of the pregnancy and regarding obstetrical and perinatal complications was obtained by means of review of obstetrical medical records and neonatal medical records.

OUTCOMES

The primary outcome was live birth after the first transfer (fresh- or frozen-embryo transfer). Live birth was defined as the delivery of any viable neonate who was 28 weeks of gestation or

older. Prespecified secondary efficacy outcomes included biochemical pregnancy, implantation, clinical pregnancy, ongoing pregnancy, pregnancy loss, and birth weight. Safety outcomes included moderate or severe ovarian hyperstimulation syndrome,[11] ectopic pregnancy, congenital anomaly, and obstetrical and perinatal complications (i.e., gestational diabetes, gestational hypertension, preeclampsia, placenta previa, placental abruption, preterm delivery, neonatal hospitalization for >3 days, and perinatal death). Definitions of the secondary efficacy and safety outcomes are provided in Table S1 in the Supplementary Appendix, available at NEJM.org.

STATISTICAL ANALYSIS

According to U.S. registry data in 2012, the live-birth rate after fresh-embryo transfer was 46% among women younger than 35 years of age.[12] This trial was designed to detect an absolute difference of 10 percentage points in the live-birth rate between the two groups, with 90% power at a significance level of 0.01. Assuming a live-birth rate of 55% in the frozen-embryo group and 45% in the fresh-embryo group, we calculated that the minimal sample would be 742 participants in each group. Assuming a 10% rate of withdrawal, we planned to enroll 1650 patients. However, the actual enrollment speed was faster than anticipated. Because there was a substantial time lag between screening (patients signed the informed-consent forms before treatment was initiated) and randomization (which occurred on the day of oocyte retrieval), we enrolled more patients who underwent randomization than we had planned; all the patients who signed the informed-consent form before the date of closing enrollment were allowed to participate.

The primary analysis was performed according to the intention-to-treat principle. Continuous variables were represented as means and standard deviations; differences in these variables were compared by means of Student's t-test. Categorical variables were described as frequencies and percentages, with the between-group difference tested by means of the chi-square test and by means of Fisher's exact test when the number of events was less than 5. Relative risks and 95% confidence intervals, as well as absolute differences and 95% confidence intervals, are presented. We also performed secondary per-protocol analyses that were based on the actual treatment that the patients received and that included patients who adhered completely to the protocol. Two-sided P values of less than 0.05 were considered to indicate statistical significance with respect to the primary outcome. Our protocol did not include a plan to adjust for multiple testing for secondary outcomes. All the analyses were performed with the use of SAS software, version 9.4 (SAS Institute).

RESULTS

PATIENTS

The enrollment of patients began in March 2015 and was completed in November 2015. Follow-up regarding the primary outcome of live birth was completed in March 2017. The baseline characteristics of the 2157 trial participants (Table 1) and the characteristics of ovarian stimulation and the IVF procedures (Table 2) were similar in the fresh-embryo group and the frozen-embryo group. A total of 165 of 1080 patients (15.3%) in the fresh-embryo group and 203 of 1077 (18.8%) in the frozen-embryo group withdrew from the trial or had a deviation from the protocol (P=0.03) (Fig. 1).

LIVE BIRTH AND SECONDARY OUTCOMES

There was no significant difference in the rate of live birth between the frozen-embryo group and the fresh-embryo group (48.7% and 50.2%, respectively; relative risk, 0.97; 95% confidence interval [CI], 0.89 to 1.06; P=0.50) (Table 3). The rates of biochemical pregnancy, clinical pregnancy, implantation, ongoing pregnancy, and overall pregnancy loss also did not differ significantly between the two groups. However, a post hoc analysis showed that the rate of second-trimester pregnancy loss was lower in the frozen-embryo group than in the fresh-embryo group (1.5% vs. 4.7%; relative risk, 0.33; 95% CI, 0.16 to 0.68; P=0.002). The mean birth weight did not differ significantly between the frozen-embryo group and the fresh-embryo group.

Patients in the frozen-embryo group had a lower risk of moderate or severe ovarian hyperstimulation syndrome than did patients in the fresh-embryo group (0.6% vs. 2.0%; relative risk, 0.32; 95% CI, 0.14 to 0.74; P=0.005) (Table 4). The incidence of obstetrical and perinatal complications, congenital anomaly, and neonatal death did not differ significantly between the two

Table 1. Characteristics of the Participants at Baseline.*

Characteristic	Frozen-Embryo Group (N=1077)		Fresh-Embryo Group (N=1080)	
	No. of Patients†	Value	No. of Patients†	Value
Age — yr		28.5±3.0		28.4±3.1
Body-mass index‡		22.0±3.0	1079	22.2±3.1
Blood pressure — mm Hg				
Systolic		118.6±11.9		118.4±12.4
Diastolic		73.0±8.3		72.7±8.4
Fertility history				
Duration of attempt to conceive — yr		3.4±2.0	1079	3.4±2.1
Previous conception — no. (%)		368 (34.2)		399 (36.9)
Indications for IVF — no. (%)				
Tubal factor		665 (61.7)		660 (61.1)
Male factor		277 (25.7)		280 (25.9)
Combined factors		135 (12.5)		140 (13.0)
Ultrasonographic findings				
Antral follicle count in both ovaries	1053	15.6±5.2	1050	15.4±5.2
Endometrial thickness — mm	1039	6.0±2.4	1032	5.9±2.3
Laboratory tests				
Follicle-stimulating hormone — IU/liter		6.7±1.6	1079	6.6±1.5
Luteinizing hormone — IU/liter	1076	4.9±1.9	1079	4.8±2.1
Estradiol — pg/ml	1070	37.0±17.9	1072	36.3±17.5
Total testosterone — ng/ml	1038	0.28±0.13	1036	0.28±0.14
Prolactin — ng/ml	1054	18.1±7.7	1055	17.8±7.8

* Plus–minus values are means ±SD. There were no significant differences between groups (P>0.05) in any of the baseline characteristics. To convert the values for estradiol to picomoles per liter, multiply by 3.671. To convert the values for total testosterone to nanomoles per liter, multiply by 3.467.
† The number of patients who were included in each analysis is provided if it differs from the total number in the trial group.
‡ The body-mass index is the weight in kilograms divided by the square of the height in meters.

groups (Table 4, and Tables S2 and S3 in the Supplementary Appendix).

In the per-protocol analyses, we compared the two groups with respect to pregnancy outcomes according to the actual treatment that patients received and with respect to pregnancy outcomes in only patients who adhered to the protocol. The results were generally consistent with those of the primary analysis, except that the birth weight of twin infants was higher in the frozen-embryo group than in the fresh-embryo group (mean difference, 80 g) and the between-group difference in the risk of the ovarian hyperstimulation syndrome was no longer significant. Details are provided in Tables S4 through S7 in the Supplementary Appendix.

DISCUSSION

In this multicenter, randomized trial involving ovulatory women, we found no significant difference in the rate of live birth with frozen-embryo transfer as compared with fresh-embryo transfer. The risks of obstetrical and neonatal complications and the mean birth weight also did not differ significantly between the two groups, although frozen-embryo transfer led to a lower risk of moderate or severe ovarian hyperstimulation syndrome.

It has been hypothesized that frozen-embryo transfer may provide a more physiologic uterine environment for embryo implantation than fresh-embryo transfer.[13] Most randomized trials com-

Table 2. Outcomes of Controlled Ovarian Hyperstimulation.*

Characteristic	Frozen-Embryo Group (N=1077)		Fresh-Embryo Group (N=1080)	
	No. of Patients†	Value	No. of Patients†	Value
No. of days of ovarian stimulation		9.6±1.5		9.6±1.5
Gonadotropin dose — IU		1509±450		1521±452
Estradiol level on hCG trigger day — pg/ml	1037	3188±1558	1053	3110±1525
Progesterone level on hCG trigger day — ng/ml	1073	1.0±0.4	1072	1.0±0.5
Endometrial thickness on hCG trigger day — mm	1066	10.8±1.9	1064	10.8±1.8
Regimen of endometrial preparation for frozen-embryo transfer‡				
Cycle — no./total no. (%)				
Natural		680/917 (74.2)		NA
Artificial		237/917 (25.8)		NA
Endometrial thickness before transfer — mm	924	10.0±1.7		NA
No. of oocytes retrieved		12.5±5.1		12.3±5.2
No. of good-quality embryos on day 3		5.0±3.5		4.9±3.5
Timing of embryo transfer — no./total no. (%)§				
Day 2		28/1028 (2.7)		35/1048 (3.3)
Day 3		944/1028 (91.8)		965/1048 (92.1)
Day 5		56/1028 (5.4)		48/1048 (4.6)
No. of embryos transferred				
Mean	1028	1.9±0.3	1048	1.9±0.2
One embryo — no./total no. (%)		90/1028 (8.8)		69/1048 (6.6)
Two embryos — no./total no. (%)		938/1028 (91.2)		979/1048 (93.4)
Reason for not undergoing embryo transfer — no. (%)		49 (4.5)		32 (3.0)
No embryo to transfer or freeze — no.		32		27
No embryos for transfer after thawing all available embryos — no.¶		0		1
Oocyte cryopreservation — no.		3		2
Personal issue — no.		9		2
Spontaneous conception after oocyte retrieval — no.		5		0

* Plus–minus values are means ±SD. There were no significant differences between groups (P>0.05) in any of the outcomes of controlled ovarian hyperstimulation. To convert the values for progesterone to nanomoles per liter, multiply by 3.180. The term hCG denotes human chorionic gonadotropin, and NA not applicable.

† The number of patients who were included in each analysis is provided if it differs from the total number in the trial group.

‡ Data regarding the regimen of endometrial preparation were missing in 7 of 924 patients who underwent frozen-embryo transfer.

§ Local investigators had the option to transfer the day 2 embryos if the number of embryos was less than three on day 2. Transfer of day 5 embryos were performed in cases of poor embryo quality or at the request of the patient.

¶ Two cleavage-stage embryos were frozen together in one straw. If at least one embryo survived after thawing, the surviving embryo or embryos were transferred. Otherwise, another straw was thawed until a viable embryo was obtained. The viable embryos after thawing were defined as having at least 50% of their cells intact. According to this procedure, there were also 14 patients who had one of their two thawed embryos that lost viability and 7 patients who had two thawed embryos that lost viability; further embryos were thawed and transferred.

paring frozen-embryo transfer with fresh-embryo transfer have involved women with (expected) high responses to ovarian stimulation.[3,5,6] In this trial, we found that the rate of live birth among women with regular ovulation and a normal response to ovarian stimulation was similar in the frozen-embryo group and the fresh-embryo group. We found a lower risk of second-trimester pregnancy loss in the frozen-embryo group than in the fresh-embryo group, but caution is warranted in the interpretation of this finding because the analysis was post hoc and the overall rates

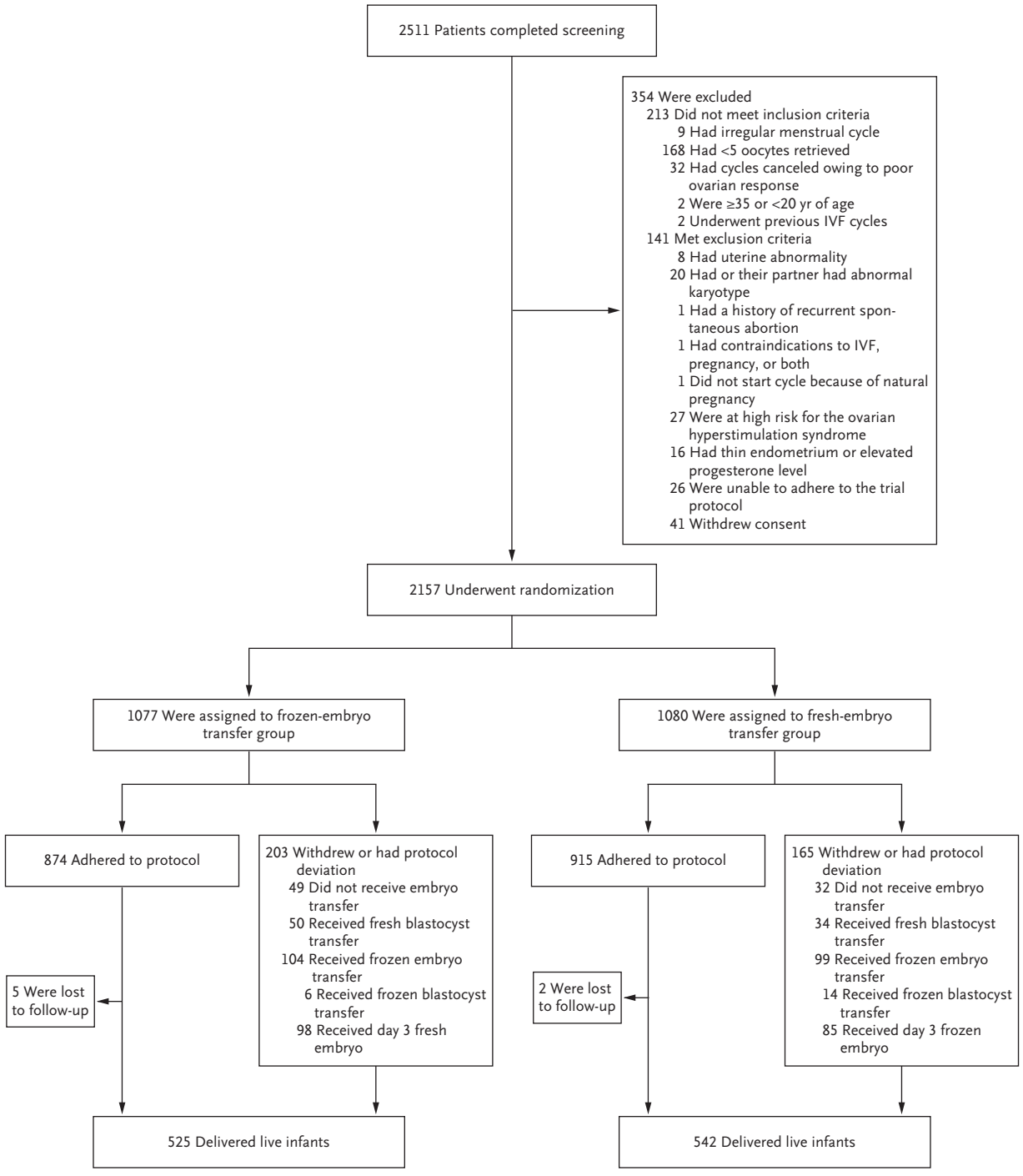

Figure 1. Trial Enrollment and Outcomes.
IVF denotes in vitro fertilization.

Table 3. Live Birth, Birth Weight, Pregnancy, and Pregnancy Loss.*

Outcome	Frozen-Embryo Group (N = 1077)	Fresh-Embryo Group (N = 1080)	Absolute Difference (95% CI)†	Rate Ratio for Frozen- vs. Fresh-Embryo Transfer (95% CI)	P Value
			percentage points		
Primary outcome: live birth — no. (%)	525 (48.7)	542 (50.2)	−1.5 (−5.7 to 2.8)	0.97 (0.89 to 1.06)	0.50
Singleton	337 (31.3)	367 (34.0)	−2.7 (−6.7 to 1.3)	0.92 (0.82 to 1.04)	0.18
Twin	187 (17.4)	173 (16.0)	1.4 (−1.8 to 4.5)	1.08 (0.90 to 1.31)	0.40
Triplet‡	1 (0.1)	2 (0.2)	−0.1 (−0.4 to 0.2)	0.50 (0.05 to 5.52)	1.00
Birth weight — g§					
Singleton					
Mean	3373±515	3380±502	−7 (−83 to 68)	—	0.85
No. of observations	335	366			
Twin					
Mean	2670±397	2616±427	54 (−6 to 115)	—	0.08
No. of observations	374	344			
Secondary outcomes					
Biochemical pregnancy — no. (%)¶	671 (62.3)	696 (64.4)	−2.1 (−6.2 to 1.9)	0.97 (0.91 to 1.03)	0.30
Implantation rate — no./total no. (%)‖	809/1966 (41.1)	857/2027 (42.3)	−1.2 (−4.2 to 1.9)	0.97 (0.90 to 1.05)	0.47
Clinical pregnancy — no. (%)**	586 (54.4)	615 (56.9)	−2.5 (−6.7 to 1.7)	0.96 (0.89 to 1.03)	0.24
Ongoing pregnancy — no. (%)††	540 (50.1)	573 (53.1)	−3.0 (−7.1 to 1.3)	0.95 (0.87 to 1.03)	0.18
Pregnancy loss — no./total no. (%)					
Among biochemical pregnancies	67/671 (10.0)	69/696 (9.9)	0.1 (−3.1 to 3.3)	1.01 (0.73 to 1.39)	0.96
Among clinical pregnancies	55/586 (9.4)	71/615 (11.5)	−2.1 (−5.6 to 1.3)	0.81 (0.58 to 1.13)	0.22
First trimester	46/586 (7.8)	42/615 (6.8)	1.0 (−1.9 to 4.0)	1.15 (0.77 to 1.72)	0.50
Second trimester	9/586 (1.5)	29/615 (4.7)	−3.2 (−5.1 to −1.2)	0.33 (0.16 to 0.68)	0.002

* Plus–minus values are means ±SD.
† Absolute differences between percentages are given in percentage points; absolute differences between other values are given in the unit indicated for that value.
‡ Two embryos were transferred, but one developed into monozygotic twins.
§ Data on the singleton birth weight of two infants in the frozen-embryo group and one in the fresh-embryo group were missing. Data on birth weights of a pair of twins in the fresh-embryo group were missing.
¶ Biochemical pregnancy was defined as a serum level of human chorionic gonadotropin of more than 10 mIU per milliliter.
‖ The implantation rate was calculated as the total number of gestational sacs divided by the total number of embryos transferred.
** Clinical pregnancy was defined as the observation of a gestational sac on ultrasonography.
†† Ongoing pregnancy was defined as the presence of a fetal heartbeat on ultrasonographic scan at 12 weeks of gestation.

Table 4. Maternal, Fetal, and Neonatal Adverse Events.*

Event	Frozen-Embryo Group (N=1077)	Fresh-Embryo Group (N=1080)	Absolute Difference (95% CI)	Rate Ratio for Frozen- vs. Fresh-Embryo Transfer (95% CI)	P Value
	no./total no. (%)				
Moderate or severe ovarian hyperstimulation syndrome before biochemical pregnancy	7/1077 (0.6)	22/1080 (2.0)	−1.4 (−2.4 to −0.4)	0.32 (0.14 to 0.74)	0.005
Ectopic pregnancy among biochemical pregnancies	18/671 (2.7)	12/696 (1.7)	1.0 (−0.6 to 2.5)	1.56 (0.76 to 3.21)	0.23
Therapeutic abortion or fetal reduction due to fetal congenital anomalies at 12 to 28 wk of gestation among clinical pregnancies	3/586 (0.5)	4/615 (0.7)	−0.2 (−1.0 to 0.7)	0.79 (0.18 to 3.50)	1.00
Gestational diabetes among clinical pregnancies	18/586 (3.1)	24/615 (3.9)	−0.8 (−2.9 to 1.2)	0.79 (0.43 to 1.44)	0.43
Preeclampsia among clinical pregnancies	26/586 (4.4)	20/615 (3.3)	1.1 (−1.0 to 3.4)	1.36 (0.77 to 2.42)	0.28
Gestational hypertension among clinical pregnancies	5/586 (0.9)	7/615 (1.1)	−0.2 (−1.4 to 0.8)	0.75 (0.24 to 2.35)	0.62
Preterm delivery among clinical pregnancies	91/586 (15.5)	80/615 (13.0)	2.5 (−1.4 to 6.5)	1.19 (0.90 to 1.58)	0.21
Congenital anomalies among live newborns	16/714 (2.2)	26/719 (3.6)	−1.4 (−3.1 to 0.4)	0.62 (0.34 to 1.15)	0.12
Neonatal death among live newborns†	2/714 (0.3)	4/719 (0.6)	−0.3 (−0.9 to 0.4)	0.50 (0.09 to 2.74)	0.69

* A full table of the adverse events is provided in Table S1 in the Supplementary Appendix.
† Neonatal death was defined as the death of a newborn within 28 days after delivery.

of pregnancy loss did not differ significantly between the two groups. In a subgroup analysis of a previous observational study, frozen-embryo transfer was associated with higher rates of implantation and ongoing pregnancy than fresh-embryo transfer among women with 10 to 15 oocytes retrieved but not among those with 4 to 9 oocytes retrieved.[14]

Our previous trial involving women with the polycystic ovary syndrome, which used a protocol that was nearly identical to that used in the present trial, showed a favorable effect of frozen-embryo transfer on the rate of live birth.[6] The reason for the discrepant results in these two trial populations is uncertain. We speculate that the difference is due to the unfavorable uterine environment after fresh-embryo transfer in women with the polycystic ovary syndrome, as shown by a much lower rate of live birth overall in the previous trial than in the present trial. Women with the polycystic ovary syndrome have an intensified ovarian response with an elevated estradiol level and a greater number of oocytes retrieved than do women with regular ovulation.[8] This altered hormonal milieu, as well as the need to initiate an ovarian stimulation cycle with oral contraceptives[15] or progestins,[16] may adversely affect endometrial receptivity after fresh-embryo transfer. A small, randomized trial comparing fresh-embryo transfer with frozen-embryo transfer after preimplantation genetic screening showed a higher rate of live birth after frozen-embryo transfer.[17] However, an oral contraceptive was administered before ovarian stimulation, and the median number of oocytes retrieved was 14 in the fresh-embryo group and 17 in the frozen-embryo group, which indicates higher ovarian responses in that trial[17] than were observed in our trial.

Birth weight did not differ significantly between the frozen-embryo group and the fresh-embryo group. Multiple studies have shown higher birth weights after frozen-embryo transfer than after fresh-embryo transfer.[1,18,19] Two studies showed that birth weight after frozen-embryo transfer differed from that after fresh-embryo transfer only in autologous cycles and not in cycles in which donated oocytes were used,[20,21] which suggests that the difference was due largely to the unfavorable uterine environment in the fresh-embryo transfer cycles. In one study, the risk of small-for-gestational-age status

of the neonate after fresh-embryo transfer was shown to be inversely associated with the estradiol level in early pregnancy.[22] Since ovulatory women usually have a lower stimulated level of estradiol than do women with the polycystic ovary syndrome or those with a high ovarian response, it is possible that the uterine environment of a fresh-embryo transfer cycle may be less affected in ovulatory women with a normal ovarian response.

In contrast to our previous trial involving infertile women with the polycystic ovary syndrome,[6] which showed a higher risk of preeclampsia with frozen-embryo transfer than with fresh-embryo transfer, we did not find a significant between-group difference in the risk of preeclampsia in the present trial. Our previous trial also showed numerically more neonatal deaths in the frozen-embryo group than in the fresh-embryo group, although the between-group difference was not significant[6]; in the present trial, the rates of neonatal death were similar in the two groups. However, the trial was not powered to detect differences in these or other uncommon outcomes, and further studies with a larger sample size or the pooling of multiple trials in a meta-analysis may be necessary to assess these outcomes.

There are limitations in this trial. The rate of withdrawal and protocol deviation was higher than 15%. Switching of groups may have been affected by the lower stimulation rates such that embryo cryopreservation was less medically indicated and fresh-embryo transfer more desired. However, our per-protocol and per-treatment analyses yielded results regarding the primary outcome that were consistent with those of the intention-to-treat analyses. We did not adjust for multiple testing. The only significant between-group difference was in the rate of the ovarian hyperstimulation syndrome ($P=0.005$); this syndrome has been reported previously to be less common after frozen-embryo transfer than after fresh-embryo transfer.[6] For practical reasons, the trial was open label. Bias that was introduced by the lack of blinding cannot be ruled out. Finally, embryos were transferred at cleavage stage in order to minimize the risk of arrested embryo development leading to no embryo transfer. However, blastocyst vitrification has been increasingly used in clinical practice, with higher pregnancy rates after transfer of blastocysts than after transfer of cleavage-stage embryos.[23] Given the differences in the uterine condition and embryo characteristics between cleavage-stage embryo transfer and blastocyst transfer, our results may not be applicable to cycles with blastocyst transfer.

In conclusion, frozen-embryo transfer resulted in a rate of live birth that was similar to the rate with fresh-embryo transfer. The risk of moderate or severe ovarian hyperstimulation syndrome was lower with frozen-embryo transfer.

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APPENDIX

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