Experimental & Clinical Article

Reproductive Medicine; *Endocrinology and Infertility*

The Effect of Fresh and Frozen Embryo Transfer on Maternal and Perinatal Results in Pregnancy by Assisted Reproductive Therapy

Tugce CIRACI[1,2], Bilge KAPUDERE[1,3], Arif Serhan CEVRIOGLU[3], Ozcan BUDAK[4], Mehmet Suhha BOSTANCI[1], Nermin AKDEMIR[1], Selcuk OZDEN[1]

Sakarya, Türkiye

ABSTRACT

OBJECTIVES: To investigate the effect of different embryo transfer (ET) methods on maternal and perinatal outcomes in vitro fertilization (IVF) pregnancies after fresh ET and frozen embryo transfer (FET).

STUDY DESIGN: In this retrospective cohort study, the files of 1506 patients who underwent ET between April 2017 and March 2022 were scanned. 136 of 147 patients who gave live birth were included in the study, and two groups were formed according to fresh ET and FET. The groups were compared regarding maternal, fetal, and perinatal out-comes.

RESULTS: Fresh ET was performed in 45.3% of the patients, and FET was performed in 54.7%. In the fresh and frozen groups, the biochemical pregnancy positivity rates were 19.21% vs. 17.71%, and the live birth rates were 10.41% vs. 9.22%, respectively. Both were similar, and no significant difference was observed (p=0.457; p=0.721). There was no difference between the fresh ET and FET groups in terms of preterm birth, birth weight, and baby gender. The rate of babies with large gestational age (LGA) was higher in the FET group (p=0.038). Pregnancy-related hypertensive diseases were relatively higher in the FET group (p=0.097). The neonatal intensive care unit (NICU) hospitalization rate was higher in FET cases (p=0.022).

CONCLUSION: It was determined that the risks of pregnancy-related hypertensive diseases, LGA fetuses, and hospitalization in the NICU might be higher in pregnancies following FET after endometrial preparation with the hormonal method (HM) in IVF treatment compared to fresh ET pregnancies. Prospective, randomized, controlled research results are needed to evaluate the subject better.

Keywords: Fresh embryo transfer; Frozen embryo transfer; In-vitro fertilization; Maternal outcomes; Perinatal outcomes

Gynecol Obstet Reprod Med 2025;31(1):40-48

[1] Department of Obstetrics and Gynecology, Sakarya University Faculty of Medicine, Sakarya Tranining and Research Hospital, Sakarya Türkiye
[2] Department of Obstetrics and Gynecology, Kulu Public Hospital, Konya, Türkiye
[3] Department of Perinatology, Bursa City Hospital, Bursa, Türkiye
[4] Department of Histology and Embryology, Sakarya University Faculty of Medicine Sakarya, Türkiye

Address of Correspondence: Bilge Kapudere
Republic of Türkiye Ministry of Health
Bursa City Hospital Doğanköy Mah. Şifa Street No:1 16250 Nilüfer, Bursa, Türkiye
bilgeaksoy@hotmail.com

Submitted for Publication: 11.02.2024 Revised for Publication: 30.03.2024
Accepted for Publication: 05.02.2025 Online Published: 10.02.2025

ORCID IDs of the authors:
TC:0009-0005-8422-4267
BK: 0000-0002-5622-3161
ASC: 0000-0002-3810-6519
OB: 0000-0002-2617-3175
MSB: 0000-0002-4776-6244
NA: 0000-0002-9129-2103
SO: 0000-0002-3346-7227

QR Code	Access this article online
	https://www.gorm.com.tr • gorm@medicalnetwork.com.tr full magazin: https://mndijital.medicalnetwork.com.tr
	DOI:10.21613/GORM.2025.1473

How to cite this article: *Ciraci T. Kapudere B. Cevrioglu AS. Budak O. Bostanci MS. Akdemir N. Ozden S. The Effect of Fresh and Frozen Embryo Transfer on Maternal and Perinatal Results in Pregnancy by Assisted Reproductive Therapy. Gynecol Obstet Reprod Med. 2025;31(1):40-48*

Introduction

Infertility is defined as the inability to get pregnant for more than a year in women under 35 and for more than 6 months in women over 35 years despite regular sexual intercourse 2-3 times a week without protection. Globally 10-15% of couples are affected by infertility. The first pregnancy after in vitro fertilization (IVF) and the first birth from an in vitro fertilized embryo were reported in 1976 and 1978, respectively (1,2). Since then, IVF and other assisted reproductive technologies (ARTs) have developed worldwide, and more than 8 million pregnancies have occurred by ARTs (3). After fertilization in vitro, embryos are transferred, or excess embryos can be cryopreserved for future use. Frozen-thawed embryo transfer (ET) provides for more than one transfer and increases the possibility of pregnancy after a single oocyte pick-up. The first live birth after frozen-thawed ET was reported in 1984. After that, many studies compared perinatal outcomes, implantation successes, and maternal morbidities in fresh and frozen ETs. Previous studies showed that perinatal outcomes are better in frozen than in fresh ET groups (4). Recent trials showed an increased risk of hypertensive disorders and macrosomic fetuses in frozen-thawed ET (5,6).

The differences in perinatal outcomes are thought to be due to various mechanisms. Better embryos may have been selected in frozen ET. It has been proposed that epigenetic changes occurring during the early embryonic period may affect the fetus's growth potential (7).

Implantation effectiveness depends on 3 main parameters; embryo quality, endometrial receptivity, and well-balanced embryo-endometrium interaction (8). Controlled ovarian stimulation leads to supraphysiologic hormone levels (9). Changes in these hormone levels cause accelerated endometrial advancement and asynchrony between the endometrium and transferred embryos, which may cause implantation failure in fresh ET. Uterine receptivity is better in natural than in stimulated cycles (10). It is thought that the absence of supraphysiological hormone application in frozen ET will cause an environment similar to physiological embryo implantation and, as a result, an increase in implantation success. On the other hand, due to the absence of the corpus luteum, some vasoactive products secreted in the early period are not produced, and it has been proposed that the risk of preeclampsia may increase as a result (11).

Maternal and neonatal outcomes in frozen ET continue to be investigated. We aimed to compare the maternal and perinatal outcomes of pregnancies conceived by frozen-thawed or fresh ETs.

Material and Method

This is a retrospective cohort study. We analyzed the data of patients who achieved pregnancy with ART and gave live birth at Sakarya University Education and Research Hospital In Vitro Fertilization Center from April 2017 to March 2022. Ethics committee approval dated 11.01.2023 and numbered 216290-45 was obtained from the Ethics Committee of Sakarya University Faculty of Medicine. Informed consent was obtained from the study participants, and the study was conducted in accordance with the Declaration of Helsinki. We collected the data through the hospital data system and by directly contacting patients.

The files of 1506 patients who underwent either fresh or frozen embryo transfer (ET) were reviewed. Among these, 277 patients had a positive β-hCG test after transfer, and 147 of them progressed beyond the 20th week of pregnancy. Data were available for 136 patients. These 136 patients were categorized based on whether they underwent fresh or frozen embryo transfer. Demographic information including age, height, weight, body mass index (BMI), occupation, duration of infertility, previous pregnancy history (gravida, parity, abortion, and number of living children), existing comorbidity, etiologies of infertility, smoking, and alcohol intake were collected.

Progesterone continued until the 10th week of pregnancy in the fresh ET group. In FET cycles, the hormonal method (HM) was used, and oral estrogen for endometrial preparation was started. In the patients where endometrial thickness reached the desired level, progesterone was added before transfer. In patients who develop pregnancy after HM, estrogen was stopped when fetal heartbeat was detected in the 7th - 8th week of ultrasound control, and progesterone was continued until the 10th week. The data obtained from the patients in both groups during pregnancy, delivery, and 6 months postpartum period was recorded. Obtained data were compared in terms of single/multiple pregnancies, type of delivery, cesarean section indication, gestational age, birth weight, gender, gestational diabetes mellitus (GDM), pregnancy-related hypertensive diseases, premature preterm rupture of membranes (PPROM), cerclage history during pregnancy, gestational cholestasis, and development of placental anomaly. In addition, the presence of neonatal intensive care unit (NICU) hospitalization, duration of hospitalization, and hospitalization indications of live-born babies in these groups were examined.

Normal birth weight was accepted between 2500 grams (g) and 4000 g. Birth weight over 4000 g was classified as large birth weight (macrosomia), 1500 - 2500 g as low birth weight (LBW), and less than 1500 g as very low birth weight (VLBW).

If the birth weight was between 10-90% according to the gestational age, it was specified as appropriate for gestational age (AGA). If the birth weight was more than 90% according to the gestational age, it was specified as large for gestational age (LGA). If the birthweight was less than 10% according to gestational age, it was specified as small for gestational age (SGA). We used Hadlock's calculation of fetal growth (12).

The gestational week was calculated as weeks and days according to the last known menstrual period. Delivery at the 37th - 41st gestational week was considered term delivery, and before the 37th gestational week was considered preterm. Preterm birth weeks were grouped as 32nd - 36th gestational weeks, 28th - 31st gestational weeks, and less than 28th gestational weeks. The presence of water flow before the 37th gestational week was accepted as PPROM. Rates of vaginal and cesarean delivery were evaluated. Cesarean indications were grouped.

Systemic diseases of the patients (diabetes mellitus (DM), chronic hypertension (HT), hypothyroidism, and other systemic diseases) were recorded. Diagnosis of GDM was placed in patients with positive oral glucose tolerance test (OGTT) that was done during 24th - 28th gestational weeks. One or two-step approaches were applied. The one-step approach involves only a diagnostic test, a fasting 75-gram OGTT. Fasting, 1st-hour, and 2nd-hour values are checked. A single elevated value is sufficient for diagnosis. Threshold values are 92 mg/dl for fasting, 180 mg/dL for hour 1, and 153 mg/dLfor hour 2. In the two-step test, screening is first performed with a 50 g OGTT without regard to time of day or previous meals, and the threshold value is taken as 140 in the 1st hour. In the event of a positive result on a screening test, a 100 g oral glucose tolerance test (OGTT) is conducted to confirm the diagnosis. For a definitive diagnosis, an elevation in at least two of the following glucose values is required: fasting glucose level of 95 mg/dL, 1-hour level of 180 mg/dL, 2-hour level of 155 mg/dL, and 3-hour level of 140 mg/dL, which are used as threshold values.

Pregnancy-related hypertensive diseases were classified as chronic HT, gestational hypertension (GHT), preeclampsia, superimposed preeclampsia, and eclampsia. HT in pregnancy is defined as systolic blood pressure of 140 and above and/or diastolic blood pressure of 90 and above. Severe HT is defined as systolic blood pressure of 160 and above and/or diastolic blood pressure of 110 and above. HT should be confirmed by at least two measurements at least four hours apart before beginning treatment. Chronic HT is defined as HT diagnosed or present before pregnancy or first recognized before 20 weeks of gestation. In patients with pregnancy complicated by GHT, HT that persists 12 or more weeks after delivery is also classified as chronic. New-onset HT over 20 weeks is defined as GHT if there is normal protein excretion and no end-organ dysfunction. Preeclampsia is defined as the new onset of HT and proteinuria or the new onset of HT with significant end-organ dysfunction with or without proteinuria in a previously normotensive patient, typically after 20 weeks of gestation or postpartum. Preeclampsia superimposed upon chronic HT is diagnosed when preeclampsia occurs in a patient with preexisting chronic HT. Eclampsia refers to the occurrence of a tonic-clonic seizure in a patient with preeclampsia in the absence of other neurologic etiologies.

Statistical analysis

In this retrospective cohort study, patient data were collected and analyzed from existing medical records. The IBM Statistical Package for the Social Sciences-SPSS v.23 program (IBM, USA) was used for statistical analysis of data. Whether the numerical data fit a normal distribution was decided by looking at the histogram graph, skewness, kurtosis values, and Shapiro-Wilk test (p-value <0.01). It didn't show normal distribution and non-parametric tests were applied. When evaluating research data by using descriptive statistical methods, number, percentage, minimum, maximum, mean, standard deviation, 1st quarter, 3rd quarter, and median values were calculated. The chi-square test (Pearson and Fisher's exact test) was used to compare categorical variables. Fisher's exact test was used when intergroup joins cannot be made in rxc-shaped tables. The Mann-Whitney U test was used to compare continuous variables. Analyses were performed at a 95% confidence interval, and p<0.05 was considered statistically significant.

Results

Between April 1, 2017, and March 31, 2022, a total of 1,506 embryo transfers were performed at the Sakarya Training and Research Hospital Assisted Reproductive Treatment (ART) Center. 54.7% (n=824) of the 1.506 transfers were FET, and β-hCG positivity was detected in 17.7% (n=146) of these transfers, and 9.2% (n=76) completed the pregnancy. Of the 71 births following fresh ET, 88.7% (n=63) were singleton pregnancies and 11.3% (n=8) were multiple pregnancies. All the multiple pregnancies were twin pregnancies, there were no triplets. There was no significant difference in β-hCG positivity, pregnancy outcomes, or the singleton or twin pregnancy with fresh and frozen ET groups.

The data of 7.5% (n=11) could not be reached; therefore, 92.5% (n=136) of 147 births were included in the study. FET was performed in 52.9% (n=72) of 136 patients, and fresh ET was performed in 47.1% (n=64). A total of 155 babies were delivered out of 136 births, 117 of which were singletons and 19 of which were twins.

We didn't observe any significant difference in the characteristics of the groups, such as maternal ages, infertility duration, infertility types, BMI, causes of infertility, FSH and E2 levels, number of babies, and delivery methods. Only AMH values were higher in the FET group (p=0.001) (Table I).

There was no difference in maternal and paternal smoking, alcohol use, maternal occupation, and chronic diseases between fresh and frozen ET groups (p<0.05), and no difference was found in patients' gravida, abortion, ectopic pregnancy, parity, and number of living children in these groups (p<0.05).

Agonist treatment protocols were applied to 6.6% (n=9) of the patients, antagonist treatment protocols to 88.2% (n=120), and dual stimulation treatment protocols to 5.1% (n=7). No significant difference between fresh and frozen ET groups regarding luteal phase support type in singleton and twin pregnancies was observed.

The average number of gestational days of pregnant women was 263.99 ± 15.42 (minimum=180; maximum=286) days. For singleton pregnancies who underwent fresh ET, the number of gestational days was 268.04 ± 10.51 (minimum= 238; maximum=286). For twin pregnancies who underwent fresh ET number of gestational days was 246.38 ± 14.56 (minimum=222; maximum=266). For singleton pregnancies who underwent FET, the number of gestational days was 264.82 ± 1.04 (minimum=180; maximum=284), and for those who gave birth to twins was 251.55 ± 19.88 (minimum=199; maximum=270).

There was no significant difference between the genders, weight groups, weights, birth weeks, and SGA diagnosis of singleton babies. When singleton pregnancies were analyzed in terms of birth weight according to gestational age, no significant difference was observed in fresh and frozen ET groups according to the presence of LGA (p=0.038). 81.8% (n=9) of 11 LGA babies were FET, and 18.2% (n=2) were fresh ET (Table II).

Table I: *Characteristics of the groups*

	Fresh (n=64)	Frozen (n=72)	p
Age, year	31.28 ± 4.10	30.46 ± 4.82	0.253
Infertility duration, year	4.76 ± 2.98	5.79 ± 3.73	0.097
Infertility type		**n (%)**	
Primary	49 (76.6)	50 (69.4)	0.352
Secondary	15 (23.4)	22 (30.6)	
BMI	23.92 ± 3.56	24.32 ± 3.87	0.698
Infertility causes		**n (%)**	
Unexplained	28 (43.8)	34 (47.2)	0.115
Male factor	14 (21.9)	25 (34.7)	
Low ovarian reserve	16 (25.0)	7 (9.7)	
Tubal factor	5 (7.8)	5 (6.9)	
Endometriosis	1 (1.6)	1 (1.4)	
Sex hormones			
AMH	2.51 ± 1.90	4.53 ± 5.01	**0.001**
FSH	6.35 ± 5.60	5.46 ± 2.35	0.094
E2	45.30 ± 25.16	38.32 ± 15.22	0.266
Number of fetuses, n (%)			
Singleton	56 (87.5)	61 (84.7)	0.641
Twin	8 (12.5)	11 (15.3)	
Type of Delivery, n (%)			
NSVD	10 (15.6)	5 (6.9)	0.107
C/S	54 (84.4)	67 (93.1)	

**Fisher's exact test was used since the number per eye was insufficient.*

Table II. *Birth data of singleton pregnancies*

	Singleton		
	Fresh ET (n=56)**	**FET (n=61)****	**p**
Gender	**n (%)**		
Female	28 (50.0)	28 (45.9)	0.658
Male	28 (50.0)	33 (54.1)	
Birthweight	**Mean**		
	3140.18 ± 479.95	3239.43 ± 666.11	0.266
Weight groups	**n (%)***		
≥4000	1 (1.8)	4 (6.6)	0.413
2500-3999	48 (85.7)	51 (83.6)	
1500-2499	7 (12.5)	5 (8.2)	
≤1499	- (-)	1 (1.6)	
Birthweight by week	**n (%)**		
AGA	47 (83.9)	46 (75.4)	0.360
SGA	7 (12.5)	6 (9.8)	0.647
LGA	2 (3.6)	9 (14.8)	**0.038**
Birth week	**n (%)***		
<28 weeks	- (-)	1 (1.6)	0.728
28-31 weeks	- (-)	- (-)	
32-36 weeks	10 (17.9)	13 (21.3)	
37-41 weeks	46 (82.1)	47 (77.1)	
Birthweek	**n (%)**		
<37 weeks	10 (17.9)	14 (22.9)	0.495
37-41 weeks	46 (82.1)	47 (77.1)	
Type of birth	**n (%)**		
Vaginal birth	10 (17.9)	5 (8.2)	0.11
Birth by cesarean section	46 (82.1)	56 (91.8)	

** Fisher's exact test was used since the number per eye was insufficient. **Column percentage was shown.*

There was no significant difference between the genders, weight groups, weights, birth weeks, and weight classes of twin babies. No significant difference was found between the weights of twin babies according to fresh and frozen ET (p=0.824).

There was a significant difference in birth weights between singletons (median=3200, n=117) and twins (median = 2440, n=38) (p<0.01).

Pregnancy complications which were pregnancy-related hypertensive diseases, DM, PPROM, and cholestasis of pregnancy, developed in 25% of pregnant women (n=34) (27 cases of singletons, 7 cases of twins). Among the singleton pregnancies, these complications were observed at a rate of 16.1% in the fresh ET group and 29.5% in the FET group (p=0.085). Pregnancy-related hypertensive diseases were found to be 3.6% in the fresh ET group and 11.4% in the FET group. Although it was observed more in the frozen group, this was not statistically significant (p=0.097). In the fresh ET group, one patient had both GDM and placental anomaly; in the FET group, DM and GHT were observed together in one patient, and GDM and preeclampsia were observed together in one patient. In twin pregnancies, one patient in the fresh ET group had GDM and gestational cholestasis; in the FET group, one patient developed PPROM after cerclage (Table III).

Twenty-seven of the 155 babies (117 + 19 + 19) were hospitalized in the NICU. Of the 27 babies, 12 are singletons, and 15 are twins. Of the 27 babies remaining in the NICU, 10 were fresh ET, and 17 were FET. One singleton and two twin pregnancies (total of 3) babies died during NICU admission. The cause of death of all babies was prematurity. The average length of NICU stay for 12 singleton babies was 7.08 ± 3.94 (minimum=1; maximum=15) days. The average length of stay in NICU for 15 twin babies was 9.20 ± 6.14 (minimum=3; maximum=27) days. Of the 27 babies needing NICU, 55.5% (n=15) were hospitalized for 7 days or less, and 44.5% (n=12) were hospitalized for 8 days or more.

There was a significant difference between the NICU needs of singleton pregnant women according to their ET types (p=0.02). While the NICU need for babies born with fresh ET was 3.6%, it was 16.4% for babies born with the frozen ET method. There was no significant difference between the number of days of stay in the NICU of singleton pregnant women according to their ET type (p=0.152) (Table IV).

There was no significant difference between the NICU needs of twin pregnant women according to ET types (p=0.65). However, there was a significant difference between the number of NICU days for twin pregnancies according to ET types (p=0.041). The rate of hospitalization for eight or more days in the NICU for twins born with fresh ET was 75.0%, and 14.3% were born with FET (Table IV).

There were no postpartum perinatal deaths in singleton and twin pregnancies in the fresh ET group, whereas, in the FET group, the perinatal mortality rate was 1.63% in singleton pregnancies and 9% in twin pregnancies.

There was a significant difference between the fresh/frozen ET status of patients with and without a pre-pregnancy diagnosis of polycystic ovary syndrome (PCOS), and 72.9% (n=35) underwent frozen ET (p=0.001). There was no statistical difference in the development of pregnancy complications in patients with PCOS compared to patients without PCOS (p=0.214). 25% of the babies of patients with PCOS and 11.4% of the babies of patients without PCOS required NICU (p=0.039).

Table III: *Maternal pregnancy complications seen in singleton and twin pregnancies*

	Singleton			Twin		
	Fresh ET (n=56)	**FET (n=61)**	**p**	**Fresh ET (n=8)**	**FET (n=11)**	**p**
Maternal complication		**n (%)**			**n (%)***	
(-)	47 (83.9)	43 (70.5)	0.085	3 (37.5)	9 (81. 8)	0.074
(+)	9 (16.1)	18 (29.5)		5 (62.5)	2 (18.2)	
Maternal complications		**n (%)**			**n (%)***	
Pregnancy-Related	2 (3.6)	7 (11.4)	0.097	2 (25.0)	1 (9.1)	0.54
Hypertensive Diseases	-	1 (1.6)		-	-	
Chronic Hypertension						
Gestational Hypertension	1 (1.8)	3 (4.9)b		2 (25.0)	-	
Preeclampsia	-	3 (4.9)c		-	1 (9.1)	
Superimposed Preeclampsia	1 (1.8)	1 (1.6)		-	-	
DM	-	1 (1.6)b		-	-	
GDM	5 (8.9)a	6(9.8)c	0.867	2 (25.0)a	-	
PPROM	-	3 (4.9)		-	1 (9.1)b	
Cerclage	1 (1.8)	2 (3, 3)		1 (12.5)	1 (9.1)b	
Placental Anomaly	2 (3.6)a	-		-	-	
Cholestasis of pregnancy	-	-		1 (12.5)a	-	

** Fisher's exact test was used since the number per eye was insufficient. a,b,c More than one complication has been seen in the same patient.*

Table IV: *Postpartum ICU admission status of singleton and twin pregnancies*

	ICU Requirement													
	Singleton							Twin						
	Total		(-)		(+)		p	Total		(-)		(+)		p
Embryo transfer type	n	%	n	%	n	%		n	%	n	%	n	%	
Fresh ET	56	100.0	54	96.4	2	3.6		16	100.0	8	50.0	8	50.0	
Frozen ET	61	100.0	51	83.6	10	16,7	0,22	22	100.0	15	68.2	7	31,8	0,65
	Hospitalization Day													
	Singleton							**Twin**						
	Total		**(-)**		**(+)**		**p**	**Total**		**(-)**		**(+)**		**p**
Embryo transfer type	n	%	n	%	n	%		n	%	n	%	n	%	
Fresh ET	2	100.0	-	-	2	100.0		8	100.0	2	25.0	6	75.0	
Frozen ET	10	100.0	7	70.0	3	30.00	0,152	7	100.0	6	85.7	1	14.3	**0,04**

Discussion

As single ET is preferred due to the complications of multiple pregnancies, the role of cryopreservation has begun to increase. FET is increasingly preferred over fresh ET in most institutions. With single oocyte picking with cryopreservation, the cumulative pregnancy rate increases, and thus, the risk of procedure-related complications and ovarian hyperstimulation decreases. Moreover, FET more closely mimics the physiological conditions of embryo implantation. During fresh ET, high blood estrogen levels due to ovarian hyperstimulation prevent invasion of the spiral arteries, impair placental blood flow, cause placental hypoperfusion, and are thought to affect fetal growth and development adversely (13). Endometrial preparation can be done naturally or artificially in FET, and the endometrium is believed to be much more receptive (14).

In this study, we assessed implantation success and maternal and perinatal outcomes compared to fresh and frozen ET IVF procedures conducted at our institution. In our study, we analyzed the files of 1.506 transferred patients. No statistically significant difference was observed in biochemical pregnancy and live birth rates among patients who underwent fresh and frozen ET. In a single-center study conducted in China, the biochemical pregnancy rate was 33.9% for FET and 38.1% for fresh ET, with no significant difference. Still, the clinical pregnancy rate was significantly lower in the FET group, 26% versus 33% (p=0.035). However, this difference did not affect the number and rates of live births (68 (19.7%) in FET cycles and 91 (24.3%) in fresh ET cycles) (15). In a study published in Iran, clinical pregnancy (35.4% vs. 47.7%) and live birth (15.6% vs. 38.7%) rates were significantly higher in the FET group than in the fresh ET group (p=0.0001; p=0.0001) (16). IVF success is affected by many factors. Although the mean age and types of infertility were similar in both study groups, the higher proportion of patients undergoing IVF due to male factors in the causes of infertility may explain these different results.

In our study, to eliminate the effect of multiple pregnancies on the preterm birth rate and, therefore, on other parameters, multiple pregnancies were excluded, and the results of singleton pregnancies and multiple pregnancies were evaluated separately. Although babies with low birth weight (LBW) (1.500 - 2.500 g) were relatively higher in the fresh ET group with 12.5%, there was no significant difference in terms of birth weight groups (p=0.413). Similarly, although macrosomia (>4.000 g) was more common in the FET group, this difference was not significant (p=0.366). However, when evaluated regarding birth weight according to week, LGA was more common in the FET group and was statistically significant (p=0.038). At the same time, the relatively higher rate of preterm birth in the FET group compared to the fresh ET group coincides with the higher number of babies with a birth weight larger than the gestational age. Yet, in a study, mean birth weight was statistically significantly higher in the FET group (3200 ± 1800 g, 2800 ± 1030 g in the fresh ET group; p=0.001) (16). Another study showed that maternal BMI and frozen ET were independent risk factors for LGA. It has been suggested that placentas are heavier after IVF and that the increase in placental size may lead to the development of LGA by promoting glucose transmission (17). In contrast, according to a retrospective study published in 2022, the incidence of LGA after FET decreased over time; maternal BMI, ethnicity, and parity number were independent risk factors for LGA; the reason for the increased risk of LGA in FET pregnancies is not yet known, so they suggested that other reasons should be emphasized to reduce the risk of LGA (18). In a retrospective cohort study conducted in Madrid, LGA was seen in 6% after fresh ET and 20.6% after FET, and macrosomia (<4.000 g) was seen in 8.2% of FET pregnancies compared to 0.9% in fresh ET (p<0.001). Pregnant women who underwent FET had a 3.4 - fold and 2 - fold increased risk of LGA for ET at the morula stage compared to ET at the blastocyst stage. Although it is predicted that the cause of LGA may be related to cryopreservation methods, some studies have shown that it does not affect (19). In a 10-year data review of 112.432 singleton pregnancies, the risk of LBW in FET pregnancies was lower than in fresh ET pregnancies, and FET was found to be an increased risk factor for macrosomia (20). A meta-analysis of 31 studies found a reduction in LBW rate in pregnancies achieved with FET compared to fresh ET. In addition, a higher risk of LGA was found in the FET group (21). In a study by Aflatoonian et al., LBW was more common in the fresh ET group (57.9% vs. 51.2% in twin pregnancies) (22). In our study, LBW was 56.3% in the fresh ET group and 45.5% in the FET group, like the study of Aflatoonian et al. In a retrospective study by Shavit et al. with 773 twin pregnancies, the mean birth weight was higher in the FET group (p=0.002), and LBW was lower in the FET group with 55% to 64% (p=0.0003) (23).

In some studies, the incidence of preterm delivery was significantly higher in the fresh ET group (15,16,24). In a 2020 meta-analysis, preterm delivery (RR 1.24, p=0.00001) was more common in pregnancies obtained by fresh ET (25). However, we didn't observe any difference in the preterm birth rate between fresh and frozen ET. Similarly, Shavit et al. observed no significant difference or increased relative risk for preterm labor (8.5% vs. 9.4%) (20).

In our study, preterm delivery in twin pregnancies was 75% higher in fresh ET and 63.6% higher in FET. 50% (8 babies) of babies born after fresh ET and 31.8% (7 babies) of babies born after FET remained in the NICU secondary to preterm birth. The history of NICU stay for more than 7 days was significantly higher in the fresh ET group (p=0.04). In addition, pregnancy complications were observed more frequently in the fresh ET group (62.5%) compared to the FET group (18.2%) in mothers of twin babies (p=0.074). Preterm labor and pregnancy complications may affect postpartum stay in the NICU. However, further studies involving larger populations are necessary due to the limited number of twin pregnancies.

In the literature, no significant difference was found between the two groups regarding stillbirth and perinatal mortality (24,25). In our study, there were no perinatal deaths in the fresh ET group in singleton and twin pregnancies, whereas in the FET group, the rate was 1.63% in singleton pregnancies and 9% in twin pregnancies. Mortality rates were found to be high due to the small size of our study group.

In our study, male babies were observed to be more common in FET pregnancies. There was no significant difference in baby genders (p=0.658). In a study, the male sex ratio was 50.9% after fresh ET, 51.3% after FET, and 51.5% after spontaneous conception (26). In a study by Aflatoonian et al., singleton male babies were higher in the FET group, 58.5% versus 44.6% (p = 0.03). In twin pregnancies, there was no difference in infant gender (42.4% vs. 38.1% for male infants) (p=0.7) (22).

In this study, cesarean delivery rates in singleton pregnancies were 82.1% to 91.8% higher in the FET group compared to the fresh ET group. This high rate can be explained by the fact that patients are afraid of vaginal birth, are worried about the risks of vaginal birth, and want cesarean section as a birth method. In addition, increased maternal and fetal complications associated with ART pregnancies may also be an expanded indication for cesarean delivery.

Our results revealed that placenta previa was observed in 2 nulliparous patients (3.6%) who underwent fresh ET, and placental abruption was not observed in any patient. In a study by Healy et al., the placenta previa rate was 3.0% in fresh ET and 2.2% in FET. This difference is not statistically significant (p=0.05) (27). In the study by Ginström Ernstad et al., FET was associated with a reduced risk of placenta previa and placental abruption (26). In a meta-analysis reported by Yang et al., placenta previa and placental abruption were more common in pregnancies obtained by fresh ET. Postpartum hemorrhage was not significantly different between the two groups (25). Similarly, in another meta-analysis, placenta previa and placental abruption risks were lower after FET (21).

In terms of maternal outcomes, we did not observe any difference between the groups in singleton and twin pregnancies. In a meta-analysis in which maternal pregnancy complications were evaluated; there was no increased risk for GDM and PPROM after fresh and frozen ET (21). In a 2020 meta-analysis, the incidences of GDM and PPROM were not significantly different between the two groups (25).

The etiology of preeclampsia is not fully understood. High E2 levels in IVF pregnancies are thought to cause impaired angiogenesis and abnormal placentation in early pregnancy. Abnormal placentation is believed to result in decreased trophoblastic invasion of the spiral arteries, resulting in persistent vasoconstriction and, consequently, inadequate blood flow to the placenta, leading to spontaneous abortion, stillbirth, SGA, or preeclampsia (28). Fresh ET requires more hormone replacement than FET, and prolonged hormone replacement exposure may affect placentation (29). Studies suggest that the absence of corpus luteum is also a factor in developing preeclampsia (11). In our research, preeclampsia in singleton pregnancies was not seen in those who underwent fresh ET. Preeclampsia was observed in 3 of 61 patients (4.9%) who underwent frozen ET. In addition, a chronic hypertensive patient with frozen ET and a chronic hypertensive patient with fresh ET developed superimposed preeclampsia. In a 2020 meta-analysis, pregnancy-related hypertensive diseases and preeclampsia were lower in pregnancies obtained with fresh ET compared to pregnancies obtained with frozen ET (RR

0.74; 95% CI 0.63-0.87; p=0.0002) (25). In a meta-analysis of 31 studies, maternal pregnancy complications were evaluated; an increased risk of pregnancy-related hypertensive disease (p<0.01) was observed in pregnancies obtained after FET (21).

Along with these, our study has some limitations. It was conducted at a single center, and as a result, the sample size was relatively small. The low number of patients may have contributed to the lack of observed differences in complications. Future studies with a larger sample size could provide more insights. Additionally, the study was not randomized or controlled; however, no significant differences were observed between the characteristics of the groups. The homogeneity between the groups, which was a strength of the study, helped reduce the possibility of errors.

Conclusion

IVF is an important treatment option for infertile patients. However, it may lead to consequences during and after pregnancy due to the psychological and economic burden it brings with it. This study suggests that pregnancies following FET after endometrial preparation with HM may have a higher risk of PIH in mothers and LGA fetuses in infants compared to fresh ET pregnancies. This indicates that the increasingly prevalent freeze-thaw treatment approach or 'freeze all' protocol is not universally correct, emphasizing the need for treatments to be tailored to each patient. There is a need for prospective, randomized, controlled research to compare data from large series groups undergoing fresh ET, HM FET, and natural cycle FET.

Declarations

Ethics approval and consent to participate: Ethics committee approval dated 11.01.2023 and num-bered 216290-45 was obtained from the Ethics Committee of Sakarya University Faculty of Medi-cine. Informed consent from the study participants was taken, and the study was conducted in ac-cordance with the principles stated in the Declaration of Helsinki.

Availability of data and materials: The data supporting this study is available through the corre-sponding author upon reasonable request.

Competing interests: The authors declare that they have no competing interests.

Acknowledgment: We are grateful for the tireless efforts of the research team members. This manu-script is the result of Tugce Ciraci's thesis.

Funding: No funding was received for this study.

Authors' Contributions: Each author contributed to the manuscript's design, data collection, analysis, and writing.

References

1. Steptoe PC, Edwards RG. Birth after the reimplantation of a human embryo. Lancet. 1978;2(8085):366. Doi: 10.1016/s0140-6736(78)92957-4. PMID: 79723.
2. Steptoe PC, Edwards RG. Reimplantation of a human embryo with subsequent tubal pregnancy. Lancet. 1976; 1(7965):880-2. Doi: 10.1016/s0140-6736(76)92096-1. PMID: 58146.
3. Fauser BC. Towards the global coverage of a unified registry of IVF outcomes. Reprod Biomed Online. 2019;38 (2):133-7. Doi: 10.1016/j.rbmo.2018.12.001. PMID: 3059 3441.
4. Maheshwari A, Pandey S, Shetty A, Hamilton M, Bhattacharya S. Obstetric and perinatal outcomes in singleton pregnancies resulting from the transfer of frozen thawed versus fresh embryos gener-ated through in vitro fertilization treatment: a systematic review and meta-analysis. Fertil Steril. 2012;98(2):368-77.e1-9. Doi:10.1016/j.fertnstert.2012.05.019. PMID: 22698643.
5. Wei D, Liu JY, Sun Y, Shi Y, Zhang B, Liu JQ, et al. Frozen versus fresh single blastocyst transfer in ovulatory women: a multicentre, randomised controlled trial. Lancet. 2019;393(10178):1310-8. Doi: 10.1016/S0140-6736(18)32843-5. PMID: 30827784.
6. Maheshwari A, Pandey S, Amalraj Raja E, Shetty A, Hamilton M, Bhattacharya S. Is frozen embryo transfer better for mothers and babies? Can cumulative meta-analysis provide a definitive answer? Hum Reprod Update. 2018;24(1):35-58. Doi: 10.1093/humupd/dmx031. PMID: 29155965.
7. Wennerholm UB, Bergh C. Perinatal outcome in children born after assisted reproductive technologies. Ups J Med Sci. 2020;125(2):158-66. Doi: 10.1080/03009734.2020.1726534. PMID: 32124667, PMCID: PMC7720966.
8. Achache H, Revel A. Endometrial receptivity markers, the journey to successful embryo implanta-tion. Hum Reprod Update. 2006;12(6):731-46. Doi: 10.1093/humupd/dml004. PMID: 16982667.
9. Roque M, Lattes K, Serra S, Solà I, Geber S, Carreras R, et al. Fresh embryo transfer versus frozen embryo transfer in in vitro fertilization cycles: a systematic review and meta-analysis. Fertil Steril. 2013;99(1):156-62. Doi: 10.1016/j.fertnstert.2012.09.003. PMID: 23040524.
10. Paulson RJ, Sauer MV, Lobo RA. Embryo implantation after human in vitro fertilization: importance of endometrial receptivity. Fertil Steril. 1990 May;53(5):870-4. Doi: 10.1016/s0015-0282(16)53524-9. PMID: 2110078.
11. von Versen-Höynck F, Schaub AM, Chi YY, Chiu KH, Liu J, Lingis M, et al. Increased preeclampsia risk and reduced aortic compliance with in vitro fertilization cycles in the absence of a corpus luteum. Hypertension. 2019;73(3): 640-9. Doi: 10.1161/HYPERTENSIONAHA. 118.12043. PMID: 30636552, PMCID: PMC6434532.
12. Hadlock FP, Harrist RB, Martinez Poyer J. In utero analysis of fetal growth: a sonographic weight standard. Radiology. 1991;181(1):129-33. Doi: 10.1148/radiology.181.1.1887021. PMID:1887021.
13. van Duijn L, Hoek J, Rousian M, Baart EB, Willemsen

SP, Laven JSE, et al. Prenatal growth trajectories and birth outcomes after frozen-thawed extended culture embryo transfer and fresh embryo transfer: the Rotterdam Periconception Cohort. Reprod Biomed Online. 2021;43 (2):279-87. Doi: 10.1016/j.rbmo.2021.04.013. PMID: 340 92521.

14. Roque M, Lattes K, Serra S, Solà I, Geber S, Carreras R, et al. Fresh embryo transfer versus frozen embryo transfer in in vitro fertilization cycles: a systematic review and meta-analysis. Fertil Steril. 2013;99(1):156-62. Doi: 10. 1016/j.fertnstert.2012.09.003. PMID: 23040524.
15. Chen Y, Zhou J, Chen Y, Yang J, Hao Y, Feng T, et al. Pregnancy outcomes after frozen embryo transfer and fresh embryo transfer in women of advanced maternal age: single-center experience. J Clin Med. 2022;11(21): 6395. Doi: 10.3390/jcm11216395. PMID: 36362623, PMCID: PMC9654836.
16. Zargar M, Dehdashti S, Najafian M, Choghakabodi PM. Pregnancy outcomes following in vitro fertilization using fresh or frozen embryo transfer. JBRA Assist Reprod. 2021;25(4):570-4. Doi: 10.5935/1518-0557.20210024. PMID: 34224240, PMCID: PMC8489809.
17. Korosec S, Frangez HB, Steblovnik L, Verdenik I, Bokal EV. Independent factors influencing large-for-gestation birth weight in singletons born after in vitro fertilization. J Assist Reprod Genet. 2016;33(1):9-17. Doi: 10.1007/ s10815-015-0601-4. PMID:26547202, PMCID: PMC471 7143.
18. Roshong AJ, DeSantis CE, Yartel AK, Heitmann RJ, Kissin DM, Pier BD. Factors associated with large-for-gestational-age infants born after frozen embryo transfer cycles. F S Rep. 2022;3(4):332-41. Doi: 10.1016/j.xfre. 2022.09.002. PMID: 36568928, PMCID: PMC9783147.
19. Trobo D, García C, Martínez M, Rodríguez I, Rojo M, Moreno S, et al. Impact of embryo cryo-preservation on large for gestational age babies born by embryo transfer: cohort retrospective study. Reprod Sci. 2023;30(4):1257-65. Doi: 10.1007/s43032-022-01075-x. PMID: 36068375.
20. Maheshwari A, Raja EA, Bhattacharya S. Obstetric and perinatal outcomes after either fresh or thawed frozen embryo transfer: an analysis of 112,432 singleton pregnancies recorded in the Hu-man Fertilisation and Embryology Authority anonymized dataset. Fertil Steril. 2016;106 (7):1703-8. Doi: 10.1016/j.fertnstert.2016.08.047. PMID: 27678031.
21. Sha T, Yin X, Cheng W, Massey IY. Pregnancy-related complications and perinatal outcomes resulting from transfer of cryopreserved versus fresh embryos in vitro fertilization: a meta-analysis. Fertil Steril. 2018;109(2): 330-342.e9. Doi: 10.1016/j.fertnstert.2017.10.019. PMID: 29331236.
22. Aflatoonian A, Mansoori Moghaddam F, Mashayekhy M, Mohamadian F. Comparison of early pregnancy and neonatal outcomes after frozen and fresh embryo transfer in ART cycles. J Assist Reprod Genet. 2010;27(12):695-700. Doi: 10.1007/s10815-010-9470-z. PMID: 20886367, PMCID: PMC2997951.
23. Shavit M, Miller N, Schreiber H, Asali A, Ravid D, Harlev A, et al. Twin pregnancies and perinatal outcomes: a comparison between fresh and frozen embryo transfer: a two-centre study. Reprod Biomed Online. 2019;38(2): 241-48. Doi: 10.1016/j.rbmo.2018.11.004. PMID: 3057 9823.
24. Wennerholm UB, Henningsen AK, Romundstad LB, Bergh C, Pinborg A, Skjaerven R, et al. Perinatal outcomes of children born after frozen-thawed embryo transfer: a Nordic cohort study from the CoNARTaS group. Hum Reprod. 2013;28(9):2545-53. Doi: 10.1093/humrep/ det272. PMID: 23832793.
25. Yang M, Lin L, Sha C, Li T, Gao W, Chen L, et al. Which is better for mothers and babies: fresh or frozen-thawed blastocyst transfer? BMC Pregnancy Childbirth. 2020;20 (1):559. Doi:10.1186/s12884-020-03248-5. PMID:3296 7652, PMCID: PMC7513314.
26. Ginström Ernstad E, Wennerholm UB, Khatibi A, Petzold M, Bergh C. Neonatal and maternal out-come after frozen embryo transfer: Increased risks in programmed cycles. Am J Obstet Gynecol. 2019;221(2):126.e1-126.e18. Doi: 10.1016/j.ajog.2019.03.010. PMID: 30910545.
27. Healy DL, Breheny S, Halliday J, Jaques A, Rushford D, Garrett C, et al. Prevalence and risk fac-tors for obstetric haemorrhage in 6730 singleton births after assisted reproductive technology in Victoria Australia. Hum Reprod. 2010;25(1):265-74. Doi: 10.1093/humrep/dep376. PMID: 19897853.
28. Imudia AN, Awonuga AO, Doyle JO, Kaimal AJ, Wright DL, Toth TL, et al. Peak serum estradiol level during controlled ovarian hyperstimulation is associated with increased risk of small for ges-tational age and preeclampsia in singleton pregnancies after in vitro fertilization. Fertil Steril. 2012;97(6):1374-9. Doi:10.1016/j.fertnstert. 2012.03.028 PMID: 22494926.
29. Blazquez A, García D, Vassena R, Figueras F, Rodriguez A. Risk of preeclampsia after fresh or frozen embryo transfer in patients undergoing oocyte donation. Eur J Obstet Gynecol Reprod Biol. 2018;227:27-31. Doi: 10.1016/j.ejogrb.2018.05.030. PMID: 29879546.