Original Article

Obstetric and Perinatal Outcomes After Fresh Embryo Transfer Versus Frozen-Thawed Embryo Transfer With Artificial Hormonal Endometrial Preparation: A Prospective Non-Randomized Study

Ahmed Shoukry, Yasser ElKassar, Mahmoud A. Hammad and Tarek Toppozada

Department of Obstetrics & Gynecology, Faculty of Medicine, Alexandria University, Egypt

ABSTRACT

Background: It is known that obstetric outcomes in singleton ICSI pregnancies are worse than those after spontaneous conceptions. With regard to maternal complications, some studies had showed that FET was associated with lower risks of placenta previa and PTB than fresh ET. On the other hand, FET may contribute to an increased risk of preeclampsia and GDM.

Objective: The aim of this study was to analyze the obstetric and perinatal outcomes in singleton pregnancies achieved after fresh ET versus FET.

Patients & Methods: Prospective cohort, non-randomized study was conducted for 130 eligible singleton pregnant women after a fresh ET (Group A) or a FET (Group B). All the cases in the two study groups were subjected to detailed history taking, antenatal care visits, 2nd trimester fetal anatomical scan, and 3rd trimester fetal Doppler evaluation. All cases were followed up till delivery and development of any obstetric/perinatal complications was observed and managed accordingly.

Results: Regarding the obstetric outcomes that were developed through the pregnancy period include, showed no statistical significance in the group of FET in comparison with the fresh ET group. Also both groups were similar as regards CS rate, mean GA at delivery, and mean neonatal birthweight, and percentage of NICU admission. There was a slight difference in Agpar score at 1 & 5 minutes between the two groups, toward a higher score in the fresh group (p=0.032).

Conclusion: Pregnancies arise after FET show comparable outcomes, so freeze all & FET policy could be considered - when indicated- a safe alternative approach to fresh ET.

Key Words: Frozen; obstetric; outcomes; perinatal; thawed.

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Corresponding Author: Ahmed Shoukry Rageh, Department of Obstetrics & Gynecology, Faculty of Medicine, Alexandria University, Egypt, **Tel.:** +2 010 2458 6399, **E-mail:** ahmedshoukry1989@outlook.com

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BACKGROUND

The percentage of frozen embryo transfer (FET) has increased significantly from 10.1% in 1990s to 42% in 2019[1]. Elective freezing of oocytes or embryos for delayed FET in a later cycle has been used more in ARTs, for a variety of reasons. Large RCTs have shown a number of benefits of eFET vs. fresh ET, including a significant decrease in the risk of moderate and severe OHSS[2,3], as well as a noticeable increase in the LBR for the entire population[4], females with PCOS[3], females with high blood progesterone levels on the day of the ovulation trigger, and cases subjected to PGT for aneuploidy[5].

Though most ART treatments still rely on fresh embryo transfer, as it entails a shorter interval to pregnancy. It has been theoretically hypothesized that the poor uterine environment caused by the supra-physiologic hormone levels seen during COS may have a detrimental effect on embryo implantation and placentation, ultimately leading to unfavorable obstetrical and neonatal outcomes after a fresh embryo transfer[6-8]. On the other hand, FET improves the uterine environment for embryo transfer, which increases endometrial receptivity[9,10]. When compared to fresh transfer cycles, a healthier uterine environment may be associated with better placentation during a FET cycle, improving obstetric outcomes[11,12]. Nonetheless, certain observations have also indicated that FET can potentially have a negative impact on pregnancy outcomes[13,14], such as high birth weights have been linked to singleton ART pregnancies after FET, but preterm births were less common when compared to fresh transfer cycles[14,15].

OBJECTIVE

The aim of this study is to compare the different obstetric outcomes after freshly transferred embryos versus frozen-thawed embryos transferred after artificial hormonal preparation of the endometrium.

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PATIENT AND METHODS

Study design & setting

Prospective cohort, non-randomized study was conducted for eligible women recruited from two IVF centers, Alexandria, Egypt (El-Madina fertility center & Repro fertility center) after approval of the ethics committee of Alexandria faculty of medicine.

Sample size calculation

Sample size was calculated using Power Analysis and Sample Size Software (PASS 2020) "NCSS, LLC. Kaysville, Utah, USA, ncss.com/software/pass". A minimal total hypothesized sample size of 120 eligible singleton pregnant women achieved pregnancy through ICSI cycles either by transfer of fresh embryos or frozen-thawed embryos for various indications (60 per group) is needed to analyze the obstetric outcomes in singleton pregnancies achieved after fresh embryo transfer versus transfer of frozen-thawed embryo; taking into consideration 95% confidence level and 80% power using Chi Square-test.

Inclusion criteria

Singleton pregnant women achieved pregnancy through ICSI cycles either by transfer of fresh embryos or frozen-thawed embryos for various indications. After confirmation of clinical pregnancy, visualization of fetal cardiac activity by TVUS women were divided into two groups:

Group A: pregnancy after fresh embryo(s) transfer.

Group B: pregnancy after frozen-thawed embryo(s) transfer after artificial hormonal preparation of the endometrium.

Exclusion criteria

Women with known uterine malformations, multifetal gestations, women with history of repeated pregnancy loss (RPL), history of preterm labour (PTB), and women with pre-exiting medical disorders such as hypertension, DM, systemic lupus, etc. were excluded from the study.

Methods

All the cases in the two study groups were subjected to detailed history taking, routine antenatal care visits at regular interval, routine 2nd trimester fetal anatomical scan, and 3rd trimester fetal Doppler evaluation. All cases were followed up till delivery and development of any obstetric complications were noted and managed such as: abortion, hypertensive disease, gestational diabetes, placental abnormalities, fetal congenital malformations, fetal growth disorders (IUGR &Macrosomia), amniotic fluid aberrations, preterm birth, and still birth, caesarean section rate, and neonatal outcomes.

Statistical analysis of the data

Data were fed to the computer and analyzed using IBM SPSS software package version 20.0. (Armonk, NY: IBM Corp) Qualitative data were described using number and percent. The Kolmogorov-Smirnov test was used to verify the normality of distribution Quantitative data were described using range (minimum and maximum), mean, standard deviation, median and interquartile range (IQR). Significance of the obtained results was judged at the 5% level.

The used tests were, Chi-square test for categorical variables, to compare between different groups. Fisher's Exact or Monte Carlo correction for Correction for chi-square when more than 20% of the cells have expected count less than 5. Mann Whitney test for abnormally distributed quantitative variables, to compare between two studied groups. Student t-test for normally distributed quantitative variables, to compare between two studied groups.

RESULTS

The Flow chart of the study is shown in (Figure 1). This prospective study was conducted for 130 women undergoing ICSI, after confirmation of clinical pregnancy eligible women were divided into two groups, Group A: pregnancy achieved after fresh embryo transfer (n=67) & Group B: pregnancy achieved after frozen-thawed ET in artificially prepared endometrium (n=63).

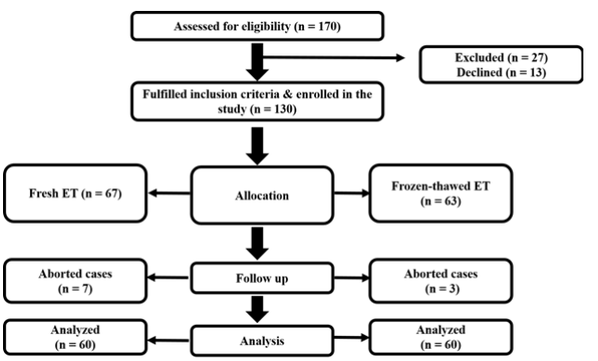

Fig. 1: Flow chart of the study

Regarding demographic data & baseline characteristics, (Table 1) demonstrate the comparison between the two groups. Women in the two study group were matched as regards mean age, mean BMI, categorizes (%) of BMI. Mean AMH was significantly higher (p=0.048) in group of frozen embryo transfer.

Regarding obstetric history of women in both groups, (Table 2) shows that there were no statistically significant differences in gravidity, parity, and previous abortion(s) between the two groups.

Obstetric outcomes

After confirmation of clinical pregnancy, 7 cases (10.4%) of group (A) have experienced spontaneous miscarriage versus 3 cases (4.8%) of group (B). There was no significant difference in miscarriage rate between two groups (FEp= 0.327). All cases of abortion have occurred in the first trimester. The rest of obstetric outcomes were studied for 120 women (60 per group) who continued their pregnancy.

Regarding placentation, low lying placenta was diagnosed in 10 women (16.7%) of group (A) versus 5 cases (8.3%) of group (B). Placenta previa was detected in 5 cases of the cohort; 4 cases in group (A) and one case in group (B). There was no statistical difference in placentation between the two groups (p=3.907).

Gestational diabetes was diagnosed in 10 cases (16.7%) of group (A) and 7 cases (11.7%) of group (B). No observed statistical significant difference between both groups (P=0.617).

Concerning hypertensive disorders with pregnancy, 12 cases developed in group (A), 9 diagnosed with preeclampsia (15%), 3 were diagnosed with eclampsia (5 %). While 8 cases developed in group (B), 6 of them diagnosed with preeclampsia (10%), 2 of them were diagnosed with eclampsia (3.33%). There was no statistical significant difference between the both groups concerning hypertension in pregnancy (P=0.327).

Polyhydramnios was detected in 7 women (11.7%) of group (A) and 3 cases (5%) of group (B), the difference did not reach a statistical significance (p=0.186). On contrary, Oligohydramnios was observed in 5 cases (7.5%) in group (A) and in 8 women (12.7%) in group (B), also the difference was not statistically significant (p=0.320).

There were 4 cases (6.6%) of group (A) their fetuses were diagnosed with congenital malformations (one case with VSD, 1 case with multi-cystic dysplastic kidneys, and 2 cases with bilateral pylectasia). In group (B), there were also 4 cases (6.6%) of fetal malformations (one case of esophageal atresia, 1 case of combined VSD&ASD, 1 case ventriculomegaly, and 1 case of congenital ovarian cyst). The difference between the two groups regarding the incidence of fetal malformations was not statistically significant (p=1.00) (Table 3).

Regarding preterm labour (PTL), there were 8 cases (13.3%) of preterm delivery in group (A) versus 6 cases (10%) of group (B). The difference was not statistically significant (p=0.570) (Table 4).

In our cohort, (27/120) cases were diagnosed with fetal growth restriction – defined as fetal abdominal circumference or EFW ≤ 10th centile – of them, 15 (25%) cases were in group (A) versus 12 (20%) cases of group (B), the difference was statistically insignificant (p=0.430). On the other hand, fetal macrosomia (EFW ≥ 90th centile for age) was observed in 5 cases (8.3%) of group (A) and in 4 cases (6.7%) in group (B), there was no statistical significant difference between both groups (p=1.00) (Table 5).

Intrauterine fetal death (IUFD) or stillbirth occurred in 2 cases (3.3%) of group (A) and also in 2 cases (3.3%) of group (B), there was no observed difference between both groups (p=1.00).

When comparing the groups according to caesarean section rate, it was comparable in both groups; 45 women (75%) of group (A) and 44 women (73.3%) of group (B) have underwent caesarean delivery (p=0.835). Mean gestational age of delivery of women in group (A) was 37.30 ± 2.38 weeks versus 37.58 ± 2.20 weeks in women of group (B), there was no statistical significance difference (p=0.499). Mean birthweight of newborns of women in group (A) was 2916 ± 658 gm versus 3023 ± 683 gm of newborns of women in group (B), this difference was not statistically significant (p=0.382).

Regarding neonatal outcomes of newborns in the two groups (n=116, as 4 infants were stillborn), Apgar score was significantly higher in newborns of group (A) with mean of 7.10 ± 1.36 and median (IQR) of 7.0 (6.0 – 8.0) versus mean Apgar of 6.62 ± 0.93 and median (IQR) of 7.0 (6.0 – 7.0) of newborns of group (B), (p=0.032). In group (A), 22 newborns (37.9%) were admitted to NICU versus 14 newborns (24.1%) in group (B), again the difference was not statistically significant (p=0.108).

Table 1: Demographic data & baseline characteristics of women in both groups

	Group (A) n = 67		Group (B) n = 63		*(p) value*
Mean age (years)	32.52 ± 4.90		31.25 ± 5.77		0.178
Mean BMI (kg/m²)	26.87 ± 4.61		26.67 ± 3.42		0.779
BMI (categorized)	No	%	No	%	
Under weight (<18.5)	1	1.5%	0	0.0%	
Normal (18.5- <25)	20	29.9%	21	33.3%	^{MC}p= 0.616
Overweight (25 - <30)	26	38.8%	28	44.4%	
Obese (≥30)	20	29.9%	14	22.2%	
Mean AMH (ng/dl)	2.81 ± 2.82		3.88 ± 3.24		0 .048*
Indication for ICSI	No	%	No	%	
Primary infertility	41	61.2%	49	77.8%	0.027891
Secondary infertility	15	22.4%	12	19.0%	
PGD	11	16.4%	2	3.2%	

Table 2: Obstetric history of women in both groups

Obstetric history	Group A (n = 67)		Group B (n = 63)		*p*
	No.	%	No.	%	
Gravida					
1	32	47.8	31	49.2	
2	8	11.9	11	17.5	0.572
>2	27	40.3	21	33.3	
Min. – Max.	1.0 – 7.0		1.0 – 7.0		0.827
Median (IQR)	2.0 (4.0 – 7.0)		2.0 (3.0 – 5.50)		
Parity					
Nulli-parous (0)	42	62.7	49	77.8	
Primi-parous (1)	15	22.4	9	14.3	0.167
Multi-parous(≥2)	10	14.9	5	7.9	
Min. – Max.	1.0 – 5.0		1.0 – 4.0		0.851
Median (IQR)	1.0 (1.0 – 3.0)		1.0 (1.0 – 3.0)		
History of Abortions					
No	49	73.1	41	65.1	0.320
Yes	18	26.9	22	34.9	
Min. – Max.	1.0 – 4.0		1.0 – 6.0		0.677
Median (IQR)	1.0 (1.5 – 3.0)		1.0 (1.0 – 1.50)		

Table 3: Comparison of obstetric outcomes between women in both groups

Obstetric outcome	Group A (n = 60)		Group B (n = 60)		*p*
	No.	%	No.	%	
Intrauterine growth retardation	15	25.0	12	20.0	0.512
Placenta					
Low lying	10	16.7	5	8.3	^{MC}p=0.145
Placenta Previa	4	6.7	1	1.7	
Gestational DM	10	16.7	7	11.7	0.432
Preeclampsia	9	15.0	6	10.0	0.327
Eclampsia	3	5.0	2	3.33	
Preterm labour	8	13.3	6	10.0	0.570
Polyhydramnios	7	11.7	3	5.0	0.186
Oligohydramnios	5	7.5	8	12.7	0.320
Congenital anomalies	4	6.6	4	6.6	1.000
Macrosomia	5	8.3	4	6.7	^{FE}p=1.000
Stillbirth or IUFD	2	3.3	2	3.3	^{FE}p=1.000

Table 4: Comparison between two groups according to mode of delivery, GA at delivery, and birthweight.

	Group A (n = 60)	Group B (n = 60)	*p*
Mode of delivery			
CS	45 (75%)	44 (73.3%)	0.835
NVD	15 (25%)	16 (26.7%)	
Gestational age at delivery (weeks)			
Min. – Max.	29.0 – 40.0	29.0 – 40.0	0.499
Mean ± SD.	37.30 ± 2.38	37.58 ± 2.20	
Birth weight (gm)			
Min. – Max.	1490 – 4600	1410 – 4700	0.382
Mean ± SD.	2916 ± 658	3023 ± 683	

Table 5: Comparison between the two studied groups according to APGAR and NICU admission (n = 116)

	Group A (n = 58)	Group B (n = 58)	*p*
NICU admission			
Negative	36 (62.1%)	44 (75.9%)	0.108
Positive	22 (37.9%)	14 (24.1%)	
APGAR score			
Min. – Max.	4.0 – 9.0	4.0 – 9.0	
Mean ± SD.	7.10 ± 1.36	6.62 ± 0.93	0.032*
Median (IQR)	7.0 (6.0 – 8.0)	7.0 (6.0 – 7.0)	

DISCUSSION

There is marked advancement in cryopreservation culture procedures for embryos since the success of the 1st pregnancy obtained via FET in the 1980s. FET is adopted by every center and has become a crucial portion of IVF/ICSI therapy. Thus, the high rate of FET has elevated the awareness of the safety of the approach[16]. The current study was a prospective study including one hundred thirty cases, aimed at analyzing the obstetric outcomes in singleton pregnancy achieved following fresh ET versus FET. Cases were classified into 2 groups, the 1st included 67 cases who underwent to fresh ET, whereas the 2nd group that included 63 cases who were subjected to FET.

In this study, regarding the baseline characteristics (age, the type of infertility, BMI, AMH and obstetric history) in both groups were matched. Regarding the obstetric outcomes that were developed through the ongoing pregnancy period including (abortion, intrauterine growth retardation, placentation, gestational DM, preeclampsia, eclampsia, preterm labour, polyhydramnios, oligohydramnios, incidence of congenital malformations macrosomia, and stillbirth) showed no statistical significance in the group of FET in comparison with the fresh ET group. Also both groups were similar as regards cesarean section rate, mean GA at delivery, and mean neonatal birthweight, and percentage of NICU admission. There was a slight difference in Agpar score at 1 & 5 minutes between the two groups, toward a higher score in the fresh group (p=0.032).

In agreement with the findings of the current study, Chen *et al*.,[17] performed a retrospective study that encompassed 1669 women with advanced maternal age performing ICSI. After matching, 345 women and 375 women were eligible in the freeze all group and fresh group respectively. In their research study, they found that in Fresh ET there were 8.5% aborted while in FET group there were 5.8% with non-significant difference in between. Those results coincide with the current study as there were 10.4% in group A aborted while in group B there were 4.8% aborted with non-significant difference in between. In previous studies, FET revealed an increased implantation rate, clinical PR, and LBR in comparison with fresh ET[18,19]. Nevertheless, 2 recent RCTs denoted that FET didn't significantly elevate the LBR[20,21].

Also the current study was in line with the results in the study done by Chen *et al*.[17]. As non-statistically significant differences between the 2 groups regarding to hypertension and preeclampsia during pregnancy. Regarding to macrosomia, there was 8.3% in fresh ET group and 6.7% in FET group had macrosomia with non-significant difference in between and that was in the same way with the results in this study as in Fresh ET there were 5% while in FET group there were 6.2% had macrosomia with non-significant difference in between.

However in the study done by Ishihara *et al*.,[22], Opdahl *et al*.,[23] demonstrated that the risk of developing PIH is elevated in the FET group in comparison with the fresh ET one. Sazonova *et al*.,[13] evaluated such result in about 2,348

singleton pregnancy following FET and 8,944 following fresh cycles were assessed in the study. A higher risk of preeclampsia was demonstrated (OR 1.32, 95 percent CI 1.07, 1.63) in singleton pregnancy following FET in comparison with fresh cycles.

In the current study, there was 16.7% in fresh ET group and 8.3% in FET group had low lying placenta with non-significant difference in between. Ishihara *et al*.,[22]; Kaser *et al*.,[24] concluded that the risk of placenta accreta occurrence was elevated markedly in the FET group in comparison with the fresh ET group. Kaser *et al*.,[24] also denoting FET as a risk factor for placenta accreta and they revealed that FET is a potentially independent risk factor for placenta accreta, even following the control of such conditions that were determined risk factors for such status and other probable complications distinct to ART. They also concluded that the high risk of placenta accreta is accompanied by factors correlated to FET in a direct way and not with patient's criteria. They suggested that the probable mechanisms related the high risk of such pregnancy complication could include decreased blood estrogen level and a thinner endometrium in FET cycles that participate in uncontrolled growth of the extra villous trophoblast into the myometrial layer.

In the current study, there was 6.7% in fresh ET group and 1.7% in FET group had placenta previa with non-significant difference in between. Healy *et al*.,[25]; Ishihara *et al*.,[22]; Rombauts *et al*.,[26]; Sazonova *et al*.,[13] found that in the 4 studies 36,455 singleton pregnancy were documented following the trasfer of frozen-thawed embryos, and 33,031 emerged after transferring fresh embryos. There was non-significant difference in the risk of Placenta previa development between the fresh ET and FET groups. A decreased rate of Placenta previa was determined in singleton pregnancy after cryopreservation cycles in comparison with fresh cycles[13]. Conversely, many reports have revealed that there's an elevated rate of placenta previa in ART singleton pregnancy in comparison with spontaneously occurred pregnancy.

In our study, there was 25% in fresh ET group and 20% in FET group had intrauterine growth retardation with non-significant difference in between. A lot of studies have reported that birth weights associated with FET are more, in comparison with those after fresh ET[27]. That difference could be due to difference in sample size. Given the little information concerning both groups, large prospective as well as randomized studies should be carried out to confirm our finding. Also in the study done by Chen *et al*.,[17] they reported higher birth weights in the FET group. Nevertheless, there non-significant difference in the risk of macrosomia and LBW was proved. It has been concluded that FET is accompanied by a reduced risk of LBW in 2 meta-analysis studies[27,28].

A meta-analysis carried out on observational studies that compared perinatal outcomes of FET and fresh cycles, ensured that FET decreases the risk of LBW as well as premature babies in singletons[15]. On the contrary, another research reported that LBW and premature babies didn't show any significant differences between fresh and FET groups in singleton as well as multiple pregnancies[29,30].

Shi *et al*.,[29] revealed that babies born following FET had markedly higher weight in comparison with babies delivered following fresh cycles in singleton as well as multiple pregnancies. Another survey denoted that prematurity and LBW are 1.3 times & 1.5 times higher respectively in single pregnancy following fresh ET in comparison with FET. It's proved that prematurity is more in couples with infertility related to women factors than male factors. Nevertheless , in twins, preterm babies and LBW were reduced in ICSI and FET cycles as well as in couples suffering infertility due to male factors[31].

In contrast, another study didn't exhibit any significant differences between 2 compared culture media as regard mean BW weight, yet babies delivered following cryopreservation had a significantly elevated BW in comparison with the fresh group[32,33]. The authors suggested that such changes might be because of the interactions between cryo-protectants with the essential enzyme interfered in epigenetic programming, resulting in normalized imprinting process[34].

A larger and more recent study carried out the Nordic countries Wennerholm *et al*.,[35] has since greatly confirmed the previously mentioned finding. It exhibited that when compared to singleton born following fresh IVF and ICSI, singletons delivered following FET had an increased risk of being LGA and having macrosomia, that's in accordance with finding from other greater population-based studies. Nevertheless , one great population-based study (of 56792 infants) revealed that fresh transfer resulted in LBW in comparison with FET offspring markedly being larger[36].

In the current study, non-statistically significant differences between the 2 groups as regard the preterm labour. 2 randomized trials that involved FET and fresh ET concluded non- significant differences in preterm birth rate. In some studies, FET was accompanied by a reduced risk of low preterm birth in comparison with the ET group[14].

A systematic review and meta-analysis by Maheshwari *et al*.,[15] evaluated findings from eleven observational studies concerning the obstetric and perinatal outcome. They suggested that singleton pregnancy after FET Vs. fresh ET were markedly less susceptible for complication by preterm birth.

Regarding to still birth in our study, non- statistically significant differences between the two groups as regards

still birth were determined which was the same mentioned by Aflatoonian *et al*.,[37] who had the same results.

Our study has the advantage of being one of the few prospective studies that addressed not only the obstetric but also the perinatal outcomes of FET versus fresh ET and was conducted for a fair number of patients. However, limitations of our study include that we compare fresh ET with frozen ET in artificially prepared endometrium, excluding other-more physiological-protocols of endometrial preparation such as true or modified natural cycle and ovarian stimulation.

CONCLUSION

Pregnancies arise from transfer of frozen-thawed embryos show comparable obstetric and perinatal outcomes, so freeze all & FET policy could be considered - when indicated- a safe alternative approach to fresh ET.

CONFLICT OF INTERESTS

There are no conflicts of interest.

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