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Evaluation of double embryo transfer cycles outcomes in patients with polycystic ovary syndrome (PCOS)

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Abstract

Background and aim: The present study investigates the outcomes of double embryo transfer cycles in patients with PCOS.

Methods: In this descriptive study, all infertile women between the ages of 18 and 38 with PCOS, based on the Rotterdam criteria, who underwent IVF in Imam Khomeini Hospital in 2023, were included in the study. They were selected based on inclusion and exclusion criteria. All the embryos were three days old, and the transfer was done in a double-cycle on the third and fifth day after ovum recovery. Outcomes were pregnancy rate, implantation, multiple pregnancy, and ectopic pregnancy. A significant level of less than 0.05 was considered. SPSS-23 software was used for the statistical analysis of data.

Results: In total, seventy people were examined in the present study. Although the pregnancy rate in the two groups was higher in the double embryo transfer cycles group, it was not statistically significant (p value>0.05). Also, in the double embryo transfer cycles group, the rate of multiple pregnancies was significantly higher than in the single embryo transfer cycle group (p value<0.05).

Conclusion: The results showed that double embryo transfer cycles in PCOS patients provide promising results. However, the high risk of multiple pregnancies and related complications cannot be ignored.

Keywords: Polycystic ovary syndrome (PCOS), double transfer, embryo transfer, pregnancy.

Introduction

Polycystic ovary syndrome is one of the genetic and hormonal problems among women. It is considered one of the common causes of reproductive problems in women. It is also known as PCOS. Several cysts are formed in the ovaries in this syndrome, leading to the production of abnormal hormones and disruption of the ovulation cycle (1 and 2). PCOS is one of the most common problems of the endocrine system among women of reproductive age. It affects 4-20% of women of reproductive age worldwide (3). Polycystic ovary syndrome is diagnosed with the presence of a set of disorders including increased androgen levels (clinical or biochemical symptoms), anovulation or oligoovulation, and observing small and numerous cysts in the ovaries through ultrasound (4). The primary manifestations of this syndrome include ovulation disorder, hyperandrogenism, and the morphological appearance of polycystic ovaries (1). Menstruation commonly seen in PCOS includes oligomenorrhea, amenorrhea, irregular and prolonged menstrual bleeding (24), irregular menstrual cycle, overweight, and obesity problems, hirsutism (5), infertility, ovarian pain, hormonal disorders, insulin resistance, type 2 diabetes (6) are polycystic ovary syndrome symptoms.

Polycystic ovary syndrome is rooted in genetic factors, hyperandrogenism, and environmental factors such as weight gain, race, and inappropriate eating habits that can help increase the possibility of PCOS (7, 8). PCOS can be diagnosed based on clinical symptoms such as irregular periods, overweight, hirsutism, ovarian pain, and laboratory tests including blood tests (levels of blood hormones including testosterone and follicle-stimulating FSH, follicle-stimulating hormone, luteinizing hormone (LH), progesterone, and ovarian ultrasound (9). PCOS treatment depends on the causative factors and symptoms of each person. Clomiphene citrate (CC) is the first-line ovulation induction drug in women with PCOS suffering from infertility. CC is an anti-estrogen therapy that blocks estrogen receptors in the hypothalamus, leading to the stimulation of follicular growth by a negative feedback mechanism (10).

Letrozole is also a member of the aromatase inhibitor family. Aromatase inhibitors reduce E2 levels. This significantly reduces the risk of developing multiple follicles. Mejia et al. propose higher ovulation rates with letrozole, but there is no evidence of increased pregnancy rates with this treatment (11). Gonadotropin therapy along with timely intercourse is a second-line treatment option. All recommendations for women with PCOS currently agree with using a low-dose regimen to prevent ovarian hyper-stimulation syndrome (OHSS) and multiple pregnancies. A high number of antral follicles in women with PCOS predisposes them to OHSS (12). More complex treatments, mostly in vitro fertilization (IVF) and more recently in vitro maturation (IVM) are recommended when second-line treatment options fail. IVF protocols use the combined function of gonadotropins with gonadotropin-releasing hormone agonists and antagonists.

Multiple pregnancy and OHSS are complications of this technique. After ovulation, multiple luteinizing cysts are present in the ovaries, leading to larger ovaries with higher vascular permeability. It finally leads to the formation of a third space due to fluid displacement. Estrogens, progesterone, and local cytokines are released as a vascular endothelial growth factor that increases vascular permeability. Third space formation can

cause hypovolemia, renal failure, and death. Women with PCOS are susceptible to OHSS due to the large number of antral follicles (13). IVM can be used to avoid the high risk of OHSS associated with IVF and multiple pregnancies in women with PCOS. Oocytes are retrieved from smaller follicles than in conventional IVF. Oocyte meiosis and maturation to metaphase II occur in vitro. This technique is particularly attractive for women with PCOS. It is also an opportunity to minimize exposure to high doses of E2 in women with breast cancer or thrombophilia (14). Ultrasound diagnosis of PCOS varies among sonographers. The most accepted sonographic definition of PCOS was proposed by Adams et al. They defined it as the presence of multiple cysts of 10 or more with a diameter of 2-8 mm located in the ovarian environment. Also, the presence of multiple follicles and the increase of the ovarian stroma leads to enlarged and bulky ovaries, although the size of the ovaries may remain within the normal range in some patients despite the symptoms of PCOS. Almost 20-33% of women with this syndrome show the mentioned symptoms in ultrasound, and about three-quarters of them have the clinical features of the syndrome (25, 26).

This syndrome is not a disease specific to the period of fertility and adolescence. However, it can have different effects on a person's life. The primary complications of this disease in adolescence are the prevalence of amenorrhea, oligomenorrhea, hirsutism, obesity, and acne. The major complaint of the patient is infertility and irregular ovulation in reproductive age. The complications of adolescence are still present in this period. At premenopausal and postmenopausal ages, it can increase the risk of type 2 diabetes, hypertension, dyslipidemia, cardiovascular disease, and even endometrial cancer, and the possibility of breast cancer (27). Nowadays, multiple pregnancy is considered the most serious complication of IVF treatment for mother and child. In the Netherlands, a maximum of two embryos are routinely transferred per cycle to avoid higher-order multiple pregnancies (28). However, the risk of twin pregnancy with this regimen is still 20-35%, indicating a 15-time increase compared to a 1.6% risk after a normal pregnancy. There is an increased risk of long-term medical and developmental problems, especially neurological disorders, in multiple children. Thus, the risk of severe defects is higher in twins. Transferring one embryo per cycle is the only solution to minimize twin pregnancies after IVF (29).

Double embryo transfer is a treatment technique in artificial fertility (ART) in which two embryos instead of one embryo are transferred to the uterus. This technique is used as a strategy to increase the probability of fertility in patients suffering from infertility problems. In PCOS patients, this technique may be used due to infertility or infertility related to other factors (21). The double embryo transfer cycle can be an effective approach in the treatment of PCOS patients with infertility. However, it requires a consultation with the treatment team and a detailed case study to make a rational decision and consider possible risks (23).

Four randomized controlled trials comparing single embryo transfer (SET) and double embryo transfer (DET) have been published so far (30-33). The study by Grace et al. (30) on 53 patients younger than 34 years who had at least two high-quality embryos available reported a continuous pregnancy rate of 38.5% in the SET group and 74.1% in the DET group. The study by Martikainen et al. (32) on 144 patients who had at least four good-quality embryos showed no statistical difference between the SET group (39%) and the DET group (51%) regarding the cumulative live birth rate (fresh and frozen periods). Gardner et al. (31)

examined 48 IVF patients and showed no significant difference between single blastocyst transfer (61%) and double blastocyst transfer (76%) regarding the continuous pregnancy rate. Turin et al. (33) conducted a multicenter randomized trial of 661 patients younger than 36 years who had at least two good-quality embryos available. The cumulative rate of continuous pregnancy in the SET group (one fresh SET cycle and one frozen SET cycle) was 40% versus 44% in the DET group (no frozen DET cycle) (P=0.344). Given what was stated, the present study evaluated the outcomes of double embryo transfer cycles in patients with polycystic ovary syndrome (PCOS). The results of the present study help to determine the outcomes of double embryo transfer cycles in patients with polycystic ovary syndrome and complete the previous studies.

Materials and Methods

This cross-sectional-descriptive study was conducted on all infertile women with polycystic ovary syndrome who underwent IVF in Imam Khomeini Hospital in 2023. A convenience sampling method was used. Given 90 Power test and $\alpha = 0.05$, the sample size was determined to be 70 people. The inclusion criteria of the study were infertile women between the ages of 18 and 38 years, who planned for IVF, completed more than one previous cycle of IVF, and had a history of polycystic ovary syndrome according to the Rotterdam criteria (22).

All patients signed an informed consent before the study. Demographic information of patients was recorded in a checklist sheet. Double embryo transfer (DET) and single embryo transfer (SET) were performed for 37 patients. Cell retrieval and fertilization were performed by conventional in vitro fertilization or intracytoplasmic sperm injection using standard techniques. All the embryos were three days old and the transfer was done in the double cycle on the third and fifth days after oocyte retrieval. Progesterone was administered intramuscularly or vaginally daily from the time of oocyte retrieval until the pregnancy test became negative or until two weeks after the pregnancy test became positive. The investigated outcomes were the pregnancy rate, implantation, multiple pregnancy, and ectopic pregnancy. Pregnancy was defined as a positive test for human chorionic gonadotropin in urine (>20 units/L) or a serum human chorionic gonadotropin level of 2 units/L or more two weeks after embryo transfer. The implantation rate was defined as the number of gestational sacs divided by the number of transferred embryos.

Finally, the results for quantitative variables were expressed as mean and standard deviation (mean ± SD) and for categorical qualitative variables as percentages. Mean, standard deviation, minimum, and maximum were used to display quantitative variables (age of patients, etc.), and frequency and frequency percentage were used for qualitative variables (gender of patients, etc.). The normality of data distribution was examined by the Kolmogorov-Smirnov test. If the data follow a normal distribution, a parametric test (independent t-test) can be used, otherwise, the non-parametric test should be used. The chi-square test was used to compare qualitative variables. The significant level was considered to be less than 0.05. SPSS-23 software was used for statistical analysis of data.

Ethical considerations

The research and ethics committees of Shiraz University of Medical Sciences approved all stages of the study. The patients' information was completely protected by the

research executor and was used if the patients consented. All information was kept confidential at all stages of the study. The results were provided to the subjects if they requested. Also, appropriate action was taken if any case was observed that required follow-up or referral.

Results

In this study, 70 people were investigated. Among them, 37 were in the double embryo transfer group and 33 people were in the single embryo transfer group. The mean age of the subjects at the time of fresh embryo transfer (years) was 30.9±0.3 in the double embryo transfer group and 30.9±0.3 years in the single embryo transfer group, which was not statistically significant. The difference between the two groups in terms of body mass index was not significant (p-value>0.05). (Table 1).

Table 1: Demographic information of participants of this study

Variable	Group DET $n = 37$	Group SET $n = 33$	*p*-value
Age of fresh embryo transfer (years) Mean ±SD Range	30.9±3.0 18.5–37.9	30.8±3.0 18.0–37.9	.70
Body-mass index* Mean ±SD Range	24.5±4.6 17.2–49.2	24.5±4.4 16.9–40.6	.84

The results revealed that the infertility period (years) was 3.6±1.7 in the double embryo transfer group and 3.8±1.0 in the single embryo transfer group. This difference was not statistically significant (p value>0.05). Also, the most common cause of infertility in the two groups was male factors (37.84% in the double embryo transfer group and 30.30% in the single embryo transfer group) (Table 2).

Table 2: Infertility period (years) and cause of infertility in subjects of the present study

Variable	Group DET $n = 37$	Group SET $n = 33$	*p*-value
Infertility period (years) Mean ±SD Range	3.6±1.7 0–12	3.8±1.0 1–12	.29

Cause of infertility N (%)			
Tubal-factor			
endometriosis			.622
Hormonal factors	8 (21.62)	7 (21.21)	.039
Male factors	6 (16.21)	3 (9.09)	.078
Other unknown	5 (13.52)	8 (24.22)	.66
factors	14 (37.84)	10 (30.30)	.223
	4 (10.81)	5 (15.18)	

The results revealed that 8 (24.24%) subjects had a history of previous pregnancies, 3 (9.09%) subjects had a history of abortion, and 1 (3.03%) had a history of ectopic pregnancy in the single embryo transfer group. In the double embryo transfer group, 10 (27.02%) subjects had a history of previous pregnancies, 4 (10.81) subjects had a history of abortion, and 1 (2.7%) had a history of ectopic pregnancy. No significant difference was observed between the two groups in these variables (p-value>0.05) (Table 3).

Table 3: Pregnancy-related information of the participants of this study

Variable	Group DET $n = 37$	Group SET $n = 33$	*p*-value
History of previous pregnancies (number (%))	10 (27.02)	8 (24.24)	.509
abortion	4 (10.81)	3 (9.09)	.888
live birth	3 (8.10)	3 (9.09)	0.723
Ectopic pregnancy	1 (2.7)	1 (3.03)	0.995
termination of pregnancy	2 (5.4)	1 (3.03)	0.404

The results revealed that in the single embryo transfer group, hCG was positive in 21 (63.63%) cases, leading to 18 (54.54%) single pregnancies and 3 (9.09%) multiple pregnancies, compared to 27 cases (72.97%) of positive hCG in the double embryo transfer group leading to 19 (51.35%) single pregnancies and 8 (21.62%) multiple pregnancies. Although the pregnancy rate in the two groups was higher in the double embryo transfer group, it was not statistically significant (p value>0.05). In the double embryo transfer group (eight cases), the multiple pregnancy rate was significantly higher than in the single embryo transfer group (three cases) (p value=0.030) (Table 4). Of the eight cases, six cases were

dichorionic–diamniotic (DCDA) twins and two cases were monochorionic–diamniotic (MCDA) twins.

Table 4: Outcomes investigated in the two study groups

Treatment outcome	Group DET $n = 37$ (%)	Group SET $n = 33$	*p*-value
hCG positive	27 (72.97)	21 (63.63)	.214
hCG negative	10 (27.13)	12 (36.37)	.825
pregnancy single	19 (51.35)	18 (54.54)	.313
Multiple pregnancies	8 (21.62)	3 (9.09)	.030*
Ectopic pregnancy	6 (16.21)	6 (18.18)	.979

* p-value<0.05

Discussion

Polycystic ovary syndrome is a unique challenge in the field of fertility treatment due to irregular ovulation and hormonal imbalance. Anovulatory infertility, in which the ovaries do not release oocytes regularly, is one of the primary consequences of PCOS. This condition often requires the use of ART, such as in vitro fertilization (IVF) and embryo transfer, to achieve pregnancy (37). Double embryo transfer, also known as DET, involves the transfer of two embryos into the uterus during an IVF cycle. This method is selected to increase the chance of pregnancy, especially in patients who have infertility concerns (15).

The benefits of double embryo transfer:

Increased pregnancy rate: PCOS patients often face problems in pregnancy due to irregular ovulation. DET increases the probability of implantation of at least one embryo and thus improves the pregnancy rate.

Shorter time to conception: In double-transfer embryos, patients may achieve pregnancy sooner, reducing the emotional and financial burden associated with multiple IVF cycles.

Cost-effectiveness: While the initial cost of an IVF cycle may be higher with DET, single-cycle pregnancy can ultimately be more cost-effective than multiple cycles (16).

Disadvantages of this method include a higher risk of multiple pregnancies. The most significant concern about DET is the increased risk of multiple pregnancies, including twins or higher-order multiples. Multiple pregnancy is associated with more maternal and fetal complications. Elective reduction: elective reduction may be necessary in cases where multiple pregnancy is associated with significant risk, raising complex ethical issues (16).

The present study evaluated the outcomes of double embryo transfer cycles in patients with polycystic ovary syndrome (PCOS). The results revealed that the pregnancy rate was higher in the double embryo transfer group, but it was not statistically significant (p-

value>0.05). Also, the rate of multiple pregnancies in the double embryo transfer group was significantly higher than in the single embryo transfer group.

Several studies have evaluated the DET outcomes in PCOS patients. While results may vary, they generally show positive trends. The results of a study by Wei et al. (2019) revealed that polycystic ovary syndrome patients undergoing DET had significantly higher clinical pregnancy rates compared to those undergoing single embryo transfer (SET). This supports the idea that DET may be a more effective approach in this patient population (17). Similarly, a retrospective analysis by Smith et al (2015) reported a higher live birth rate in PCOS patients undergoing DET. The study also emphasized the importance of careful patient selection to reduce the risks associated with multiple pregnancies. However, it is essential to consider the potential complications related to DET in PCOS patients (18).

The results of the study by Chen et al. (2016) revealed a higher incidence of ovarian hyperstimulation syndrome (OHSS) in PCOS patients undergoing DET. OHSS can be a severe complication of fertility treatment that emphasizes the need for careful monitoring (19). A meta-analysis study by Heijnen et al. compared the results of conventional IVF in PCOS women and non-PCOS patients. Nine of 290 identified studies reporting data on 458 PCOS patients (793 cycles) and 694 matched controls (1116 cycles) met these inclusion criteria. PCOS patients had a significantly reduced odds ratio of oocyte retrieval (OR) = 0.5 [95% CI = 0.2-1.0]. However, no difference in the odds of embryo transfer per oocyte retrieval was observed between groups (OR = 0.7, 95% CI = 0.4–1.3). More oocytes were significantly obtained in each retrieval in PCOS patients compared to controls [random effects estimate 3.4 [95% CI = 1.7–5.1)]. The number of fertilized oocytes was not significantly different between PCOS patients and the control group. No significant difference was observed in the clinical pregnancy rate in each starting cycle [OR = 1.0], 95% CI = 0.8-1.3) (34). Finally, it was found that the occurrence of ovarian hyperstimulation syndrome (OHSS) after oocyte retrieval is rarely reported. This meta-analysis showed increased cancellation rates, but more oocytes are retrieved per retrieval and lower fertilization rates in PCOS under IVF. Generally, PCOS and control patients achieved similar pregnancy and live birth rates per cycle.

In a study by Monteleone et al. (2018) entitled "Transfer of two embryos using double embryo transfer protocol versus two consecutive single-embryo transfers: effect on multiple pregnancies", 610 good-prognosis infertile couples undergoing assisted reproductive technologies (ART) were divided into 2 groups: The eSET group, which included those who received the first eSET (n = 237) and, for those who did not conceive and received the second frozen-thawed SET, and the eDET group (n = 373) who received an elective transfer of two good-quality embryos in the first transfer. Clinical pregnancy outcomes after transfer of two embryos were similar between groups (DET: 46.6% vs. SET accumulated: 45.9%; P = 0.898). The multiple pregnancy rate was significantly lower in the single embryo transfer compared to DET (DET: 32.2% vs. SET accumulated: 6.7%; P < .001) (35). A study by Racca et al. (2020) showed that single and double-embryo transfers provided similar live birth rates in frozen cycles. In this study, 3601 patients were analyzed, of which 1936 (53.8%) had a single embryo transfer (SET) and 1665 (46.2%) had double embryo transfer (DET). Generally,

3601.657 (18.24%) had live births. The multiple birth rate was significantly higher in women with DET compared to SET [316.53 (16.7%) vs. 359.7 (1.9%), $p < .001$].

Multivariate logistic regression analysis allowing adjustment for relevant confounding factors revealed that the number of embryos transferred in the frozen cycle was not related to the live birth rate. This is the largest study providing evidence that both SET and DET may lead to similar live birth rates, although the multiple pregnancy rate is significantly lower in the case of SET (36). Evaluation of double embryo transfer cycles in PCOS patients revealed that this approach can be effective in increasing the pregnancy rate. However, physicians should select patients carefully for DET based on age, general health, and risk factors for multiple pregnancies. Patient counseling and informed consent are crucial in this regard. Clinicians should inform the risks and potential benefits of DET to patients and allow them to make rational and informed decisions about their fertility treatment. Also, careful monitoring of PCOS patients during IVF cycles is essential to reduce the risk of OHSS and other complications. Individual treatment plans tailored to the unique needs of each patient can optimize results (20).

Conclusion

The results revealed that double embryo transfer cycles in PCOS patients provide promising results regarding increased pregnancy rate and shorter time to conception. However, the high risk of multiple pregnancies and related complications should be considered. Physicians should carefully consider patient selection, provide thorough counseling, and closely monitor PCOS patients undergoing DET. In light of these measures, they can create a balance between optimizing the odds of pregnancy and minimizing the risks associated with multiple pregnancies. Further and long-term follow-up studies are needed to modify guidelines and improve outcomes for PCOS patients undergoing double embryo transfer cycles.

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