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A model for pregnancy rates after IVF-ET in patients with infertility and endometriosis

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Abstract. – OBJECTIVE: The objective of the present study was to examine the clinical factors influencing the pregnancy rate of infertile patients with endometriosis and establish a predictive model.

PATIENTS AND METHODS: This study included 158 patients (158 cycles) with infertility and endometriosis who underwent laparoscopic surgery, and *in vitro* fertilization and embryo transfer (IVF-ET) were evaluated retrospectively between January 2019 and December 2020. The clinical factors in the pregnant and non-pregnant group were analyzed by univariate analysis. Statistically significant variables were subsequently used for multivariate logistic regression to establish the prediction model.

RESULTS: Multivariate logistic regression analyses showed that GnRH-a treatment after operation (OR, 6.562; 95% CI: 2.782-15.477; $p<0.01$), ASRM stage (OR, 0.218; 95% CI: 0.093-0.509; $p<0.05$), the number of high-quality transferred embryos (OR, 3.155; 95% CI: 1.647-6.047; $p<0.05$) were independently associated with successful pregnancy. The area under the curve (AUC) of the prediction model was 0.774 (95% CI: 0.700-0.847). According to Hosmer-Lemeshow, the model was well fitted ($p>0.05$). We applied the bootstrapping method to internal validation, and the result showed that the pregnancy rate predicted by the model and the real data were consistent.

CONCLUSIONS: The models for predicting pregnancy rates after IVF-ET in infertility and endometriosis patients showed high accuracy. The effective methods to increase the number of high-quality embryos need further study.

Key Words:

Endometriosis, Infertility, *In vitro* fertilization and embryo transfer, Pregnancy outcome.

Abbreviations

IVF-ET: *in vitro* fertilization and embryo transfer; hCG: human chorionic gonadotropin; EMS: Endometriosis; ICSI: intra-cytoplasmic sperm injection; ASRM: American Society for Reproductive Medicine; GnRH-a: Gonadotrophin releasing hormone agonist; ART: assisted reproductive technologies.

Introduction

Endometriosis (EMS) refers to abnormal growth of endometrium outside the uterine cavity, usually with an associated inflammatory process[1]. Due to the effects of sex hormones, these endometrial lesions periodically become ischemic, detached, and bleeding, resulting in a local chronic inflammatory reaction that may lead to extensive fibrosis and formation of adhesions. Endometriosis affects approximately 10% of reproductive-age women and 5 to 50% of infertile women[2,3]. The relationship between EMS and infertility is not clear. Recent studies[4,5] suggest that infertility in EMS patients may be caused by several factors, including distorted pelvic anatomy, inflammatory changes in the peritoneal fluid affecting the oocytes, sperms, embryos, or fallopian tube function, damaged functional ovarian tissue and reduced uterine receptivity. Endometriosis can also result in lower oocyte production, lower implantation rates and lower pregnancy rates after *in vitro* fertilization (IVF) and intra-cytoplasmic sperm injection (ICSI)[6-8]. Since the birth of the first test-tube baby in human history in 1978, assisted reproductive technology (ART) has been widely used to treat infertility related to endometriosis. We retrospectively reviewed the cases of infertile patients undergoing assisted reproductive treatment after laparoscopic surgery for EMS and analyzed clinical factors that affected pregnancy outcomes.

Patients and Methods

Patients

158 patients (158 cycles) with infertility and endometriosis who underwent laparoscopic surgery and IVF-ET at our hospital were retro-

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spectively evaluated between January 2019 and December 2020. The Hospital Review Board approved our study (Ethical Review No. 2016038), and we obtained informed consent and signatures from all patients. And meanwhile, the privacy of patients was effectively protected through anonymization. The study included clinical data from 143 IVF cycles and 15 ICSI cycles. Patient age ranged between 24 and 43 years and infertility duration ranged between 1 and 16 years. Of 158 patients, 39 were diagnosed with primary infertility and 119 had secondary infertility. The causes of infertility included oligoasthenoteratozoospermia of partner, tubal infertility, adenomyoma, luteinized unruptured follicle syndrome, intrauterine adhesions, unexplained infertility, and others. The basic endocrine levels of all patients were in the normal range.

Surgery for Endometriosis

All patients with endometriosis were treated with laparoscopic surgery. If pelvic adhesions were identified during the operation, they were separated first to restore the normal anatomical structure of the pelvic cavity. Any visible lesions were destroyed, ablated, or excised at the time of laparoscopy. Excision of the cyst was the first choice for an ovarian endometriosis cyst. Briefly, adhesions with the surrounding areas were first separated, the fluid in the cyst was then aspirated, and the cyst wall was removed after the inner wall of the cyst was washed clean. Low-power electrocoagulation or sutures were used to stop bleeding from the wounds. Attempts were made to preserve and protect the normal ovarian tissue as much as possible.

IVF-ET Method and Embryo Quality Assessment

We performed controlled ovarian hyperstimulation, oocyte retrieval, IVF or ICSI, embryo cultures and embryo transfer, according to the usual methods. Progesterone was used to support the function of the corpus luteum after egg retrieval, and 1-2 third days of embryos were selected to transfer into the uterine cavity. After 14 days of embryo transfer, we underwent blood tests for human chorionic gonadotropin (hCG). HCG-positive patients underwent ultrasound tests to observe the indicators of clinical pregnancy, such as gestational sacs and the fetal heartbeat after 5 weeks of the transfer. According to the study about embryo evaluation criteria set by Van Royen et al[9], the high-quality embryo can produce 4-5 blastomeres on the second day, >/=7 cells on the third day, no more than 20% of anucleated fragments and absence of multinucleated blastomeres.

Grouping

158 patients (158 cycles) in our study were divided into two groups. Clinically pregnant patients were assigned to the pregnancy group, while non-clinically pregnant patients (including biochemical pregnancy or failed implantation) were assigned to the non-pregnancy group. The clinical factors between the two groups were compared: infertility (type and duration), age, GnRH-a treatment after the surgery, stage of endometriosis, endometrial thickness on the day of hCG administration, number of high-quality transferred embryos, etc. Endometriosis stage was determined based on the revised American Society for Reproductive Medicine (ASRM) classification, the revised American Fertility Society (rAFS) stage in 1996[10].

Statistical Analysis

We performed statistical analyses using 24.0 SPSS software (IBM Corp., Armonk, NY, USA). The mean ± standard deviation was expressed for measurement data with normal distribution, the median [min-max] was described for measured data with abnormal distribution, and the count data as numbers and percentages. Normally distributed continuous variables were analyzed by the independent *t*-test or Mann-Whitney U test and categorical variables were analyzed by Pearson's Chi-Square test or Fisher's exact test. Multivariate logistic regression analyses were conducted for the variables with significant differences. We used a Forward Selection (Likelihood Ratio) method for multivariate analysis and a cut-off p-value of 0.2 to omit non-independent risk factor from the model.

We used AUC and Hosmer-Lemeshow goodness-of-fit test to evaluate the performance of the predicted model. The Bootstrap repeated sampling method (sampling 1,000 times) was used to conduct internal verification of the model and make a calibration diagram. According to the prediction model, the rms program package in R3.6.2 software was used to establish the prediction line chart model. $p<0.05$ indicated statistical significance.

Table I. Comparison of clinical indicators in the groups with different pregnancy outcomes.

Clinical Indicator	Pregnancy group (n = 65)	Non-pregnancy group (n = 93)	$t/\chi^2/Z$	*p*-value
Age (years)	30 [24, 39]	31 [24, 43]	-2.106	0.035
≥ 35 years	12 (18.46%)	26 (27.96%)	1.889	0.169
Infertility duration (years)	3 [1, 12]	3 [1, 16]	-0.842	0.400
Primary Infertility	53 (81.54%)	66 (70.97%)	2.300	0.129
GnRH-a treatment after operation	31 (47.69%)	22 (23.66%)	9.916	0.002
ASRM stage (I/II)	26 (40%)	21 (22.58%)	5.555	0.018
ASRM stage (III/IV)	39 (60%)	72 (77.42%)		
Basal FSH	7.45 [1.41, 14.77]	7.25 [1.57, 13.76]	0.877	0.380
Basal FSH/LH	2.02 [0.77, 9.23]	2.19 [0.80, 22.20]	-1.355	0.175
Basal estradiol (ng/ml)	45.72 [10, 166.20]	50.97 [10.00, 281.00]	-1.548	0.122
Body mass index (kg/m^2)	20.32 [16.65, 25.91]	20.45 [16.81, 36.73]	-1.129	0.259
Endometrial thickness (mm) on hCG trigger day	11.54 ± 1.75	11.20 ± 1.66	-1.264	0.208
Number of transferred embryos (one)	2 [1.2]	2 [1.2]	1.373	0.170
Number of high-quality transferred embryos (one)	2 [0.2]	2 [0.2]	2.923	0.003

GnRH-a, Gonadotrophin releasing hormone agonist; ASRM, American Society for Reproductive Medicine; FSH/LH, follicle-stimulating hormone/luteinizing hormone; hCG, human chorionic gonadotropin.

Results

Pregnancy Outcomes and Univariate Analysis Between the Pregnancy and Non-Pregnancy Group

In a total of 158 patients, 65 patients were clinically pregnant, with a clinical pregnancy rate of 41.1%, and 93 patients showed non-implantation or biochemical pregnancy. We observed no significant differences in infertility type and duration, body mass index, basal FSH, basal estradiol, basal FSH/LH, endometrial thickness on hCG trigger day and the number of transferred embryos between the two groups (all $p>0.05$). Positive treatment outcome (pregnancy) rate significantly correlated with the lower age of the patients (Z=-2.106, $p=0.035$), GnRH-a treatment after the operation ($\chi^2=9.916$, $p=0.002$), and lower ASRM stage (I/II) ($\chi^2=5.555$, $p=0.018$). The number of high-quality transferred embryos in the pregnancy group was different from the non-pregnancy group (Z=2.923; $p=0.003$) (Table I).

Multivariate Logistic Regression Analysis

Age, GnRH-a treatment after surgery, ASRM stage, and the number of high-quality transferred embryos were analyzed using multivariate logistic regression analysis. The results showed that GnRH-a treatment after operation (OR, 6.562; 95% CI: 2.782-15.477; $p<0.01$), ASRM stage (OR, 0.218; 95% CI: 0.093-0.509; $p<0.05$), and the number of high-quality transferred embryos (OR, 3.155; 95% CI: 1.647-6.047; $p<0.05$) significantly affected pregnancy outcomes (Table II). Age of patients in both groups was not statistically different ($p>0.05$) and therefore did not affect pregnancy outcomes.

Table II. Multivariate logistic regression analysis of age, GnRH-a treatment after operation, ASRM stage, and the number of high-quality transferred embryos.

Index	Constant	GnRH-a treatment after operation	ASRM stage	Number of high-quality transferred embryos
B	-0.280	1.881	-1.524	1.149
Wald	0.112	18.463	12.411	11.990
OR value	0.756	6.562	0.218	3.155
95% CI	-	2.782-15.477	0.093-0.509	1.647-6.047
p-value	0.737	< 0.001	< 0.001	0.001

GnRH-a, Gonadotrophin releasing hormone agonist; ASRM, American Society for Reproductive Medicine; OR, Odds Ratio; CI, confidence interval.

Evaluation of the Prediction Model

With the calculated value of the prediction model as the detection variable and the grouping as the state variable, as indicated by the receiver operating characteristic (ROC) curve, the AUC of the prediction model was 0.774 (95% CI: 0.700-0.847) (Figure 1). The prediction model was better than GnRH-a treatment after operation model (AUC=0.620, 95% CI: 0.530-0.710), ASRM stage model (AUC=0.587, 95% CI: 0.496-0.679) and the number of high-quality transferred embryos model (AUC=0.616, 95% CI: 0.530-0.703). The prediction model fitted well, according to the Hosmer-Lemeshow goodness-of-fit test (p=0.742). We applied the bootstrapping method to internal validation, and the result showed that the pregnancy rate predicted by the model and the real data were consistent (Figure 2). A nomogram was established using R programming language based on the predictive model (Figure 3).

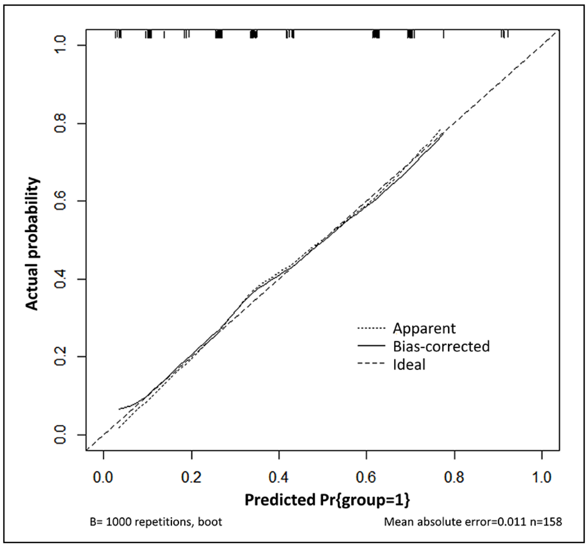

Figure 2. Model calibration.

Discussion

Currently, surgery continues to be one of the main treatment plans for EMS accompanied by infertility[11]. The purpose of the surgery is to remove visible pelvic lesions, restore the pelvic anatomical structure and improve the fertility. Operative laparoscopy could improve the rate of pregnancy in ASRM stage I/II endometriosis for endometriosis-associated infertility[12,13]. However, ovarian endometriosis cyst excision surgery inevitably results in the loss of ovarian tissue, damage to the ovarian function caused by endometriosis itself, and the inflammatory response of the ovary's wound surface after the surgery. All these factors reduce the ovarian reserve function after the surgery, especially in patients with stage III-IV endometriosis[14]. Therefore, effects of laparoscopic surgery on ovarian reserve function in patients with infertility should be comprehensively evaluated before the operation. For patients with poor ovarian function (AFC<5, AMH<0.5-1 ng / ml, or FSH>10 U/L on the 2nd-4th day of menstruation)[15], IVF should be considered to accumulate embryos and preserve fertility before the surgery. Patients with normal ovarian function should be informed about the associated risk of loss of ovarian function, before surgery. In this study, all 158 patients had normal ovarian function before the surgery and signed informed consent.

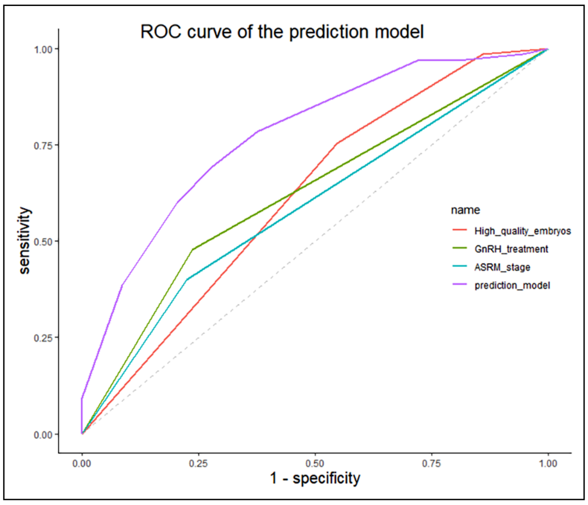

Figure 1. ROC curve of the prediction model.

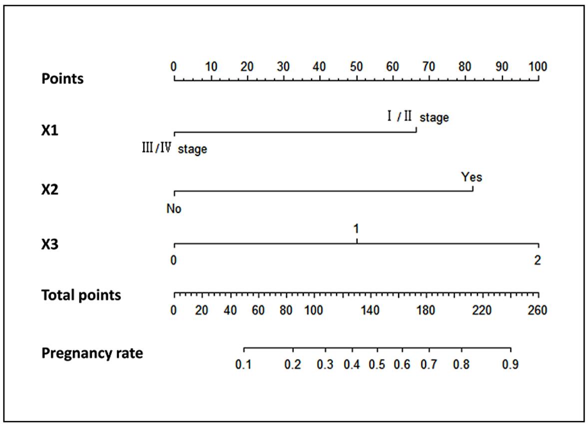

Figure 3. Pregnancy rate assessment tool. X1, ASRM stage; X2, GnRH-a treatment after operation; X3, Number of high-quality transferred embryos (one).

Embryo quality significantly correlates with the outcome of IVF-ET, with high-quality embryo transfer associated with a higher chance of a successful pregnancy. Dobson et al[16] reported a pregnancy rate of 16.3% in a single poor-quality embryo transfer group and a pregnancy rate of 37.6% in a single top-quality embryo group in patients that underwent IVF treatment. Recent studies[17,18] demonstrated that when a poor-quality embryo is transferred together with a good-quality embryo, the poor-quality embryo has no significant effect on the good-quality embryo. In our study, the number of high-quality embryos transferred correlates with the clinical pregnancy rate, which was higher in the pregnancy group (Z=2.923; p=0.003). Furthermore, multivariate logistic regression analysis also showed that higher embryo quality was associated with clinical pregnancy ($p<0.05$).

For patients with severe endometriosis after the operation, the use of GnRH-a before assisted reproductive technology treatment can improve clinical pregnancy rates. A systematic review and meta-analysis included 980 women with endometriosis and 5,934 patients without endometriosis (control) revealed that patients with stage I-II EMS and the control group had similar clinical pregnancy rates. However, the clinical pregnancy rates of patients with stage III-IV EMS were lower, compared to the control group (OR 0.45; 95% CI 0.29-0.70). GnRH-a may improve the symptoms of endometriosis and reverse the negative effects of endometriosis on IVF cycles, including poor folliculogenesis that results in oocytes of reduced quality, anatomical dysfunction of the fallopian tubes and ovaries, affects oocyte pickup and transportation, hostile inflammatory peritoneal environment[19,20]. Kaponis et al[21] reported a prospective, randomized, controlled trial involving 400 infertile women with mild endometriosis diagnosed by laparoscopy, undergoing IVF. Women who received GnRH-a had a statistically significantly reduced concentration of follicular fluid cytokines (interleukin-1β, interleukin-6, interleukin-8, IL-1 receptor antagonist and tumor necrosis factor α), and higher fertilization rate. However, the implantation rate, embryo quality, and clinical pregnancy rates showed no statistically significant difference compared with women who did not receive this treatment. In agreement with these results, our study also indicated that infertile women with endometriosis after surgery, who received GnRH-a, had higher clinical pregnancy rates. Due to very few patients receiving GnRH-a in stage I-II, it was not possible to conduct a stratified study.

Age is an important factor affecting fertility. Numerous studies[22-24] showed that female fertility begins to decrease significantly after age 32 and declines more rapidly after age of 37. Ovarian reserve function reflects a woman's reproductive potential, which is mainly related to the quality and number of the antral follicles. With the increasing age, the ovarian reserves diminish, and fewer embryos can be obtained. Schwartz et al[23] observed the cumulative pregnancy rate of up to 12 insemination cycles, which was 74% for women under 31 years of age, reduced to 62% for 31-35 years of age, and 54% for women over 35 years of age[23]. In women undergoing ART, the rate of live births per cycle was 40.1% for women under 35 years, and 4.5% for women over 42 years[24]. In our study, the age was older in the non-pregnancy group compared to the pregnancy group (p=0.035), but there was no statistically significant value in the multivariate logistic regression analysis ($p>0.05$), which probably needs a larger sample size to further investigation.

Conclusions

This study confirms that GnRH-a treatment after the operation, ASRM EMS stage, and the number of high-quality transferred embryos are the significant influence factors of pregnancy outcomes for patients with endometriosis who underwent IVF-ET after the operation, and we use multivariate logistic regression to establish the prediction model. The effective methods to increase the number of high-quality embryos need further study.

Conflict of Interest

The Authors declare that they have no conflict of interests.

Acknowledgements

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Availability of Data and Materials

The datasets used and/or analyzed during the current study are available from the corresponding author on reasonable request.

Authors' Contribution

Feipeng Huang was involved in the conception and design of the study and in data collection, and in the writing of the manuscript. Jingsong Yi was involved in the conception and design of the study and in the editing of the manuscript. Xi Xie was involved in the data interpretation and editing of the manuscript. Shengrong Du and Huale Zhang were involved in data collection. All authors have read and approved the final manuscript.

Ethics Approval

The Ethics Committee of the Fujian Maternity and Child Health Hospital approved this retrospective medical record review (approval no. 2016038).

Informed Consent

Informed written consent was obtained from all patients for the surgical procedures. For the present study, patient consent was waived as it was a retrospective analysis, with no direct contact between the authors and the patients, and personal privacy was protected through anonymization.

References

1) International working group of AAGL, ESGE, ESHRE and WES, Tomassetti C, Johnson NP, Petrozza J, Abrao MS, Einarsson JI, Horne AW, Lee TTM, Missmer S, Vermeulen N, Zondervan KT, Grimbizis G, De Wilde RL. An International Terminology for Endometriosis, 2021. J Minim Invasive Gynecol 2021; 28: 1849-1859.
2) Zondervan KT, Becker CM, Missmer SA. Endometriosis. N Engl J Med 2020; 382: 1244-1256.
3) Shafrir AL, Farland LV, Shah DK, Harris HR, Kvaskoff M, Zondervan K, Missmer SA. Risk for and consequences of endometriosis: A critical epidemiologic review. Best Pract Res Clin Obstet Gynaecol 2018; 51: 1-15.
4) de Ziegler D, Borghese B, Chapron C. Endometriosis and infertility: pathophysiology and management. Lancet Lond Engl 2010; 376: 730-738.
5) Practice Committee of the American Society for Reproductive Medicine. Endometriosis and infertility: a committee opinion. Fertil Steril 2012; 98: 591-598.
6) González-Comadran M, Schwarze JE, Zegers-Hochschild F, Souza M do CB, Carreras R, Checa MÁ. The impact of endometriosis on the outcome of Assisted Reproductive Technology. Reprod Biol Endocrinol RBE 2017; 15: 8.
7) Hamdan M, Omar SZ, Dunselman G, Cheong Y. Influence of endometriosis on assisted reproductive technology outcomes: a systematic review and meta-analysis. Obstet Gynecol 2015; 125: 79-88.
8) Senapati S, Sammel MD, Morse C, Barnhart KT. Impact of endometriosis on in vitro fertilization outcomes: an evaluation of the Society for Assisted Reproductive Technologies Database. Fertil Steril 2016; 106: 164-171.e1.
9) Van Royen E, Mangelschots K, De Neubourg D, Valkenburg M, Van de Meerssche M, Ryckaert G, Eestermans W, Gerris J. Characterization of a top quality embryo, a step towards single-embryo transfer. Hum Reprod 1999; 14: 2345-2349.
10) Revised American Society for Reproductive Medicine classification of endometriosis: 1996. Fertil Steril 1997; 67: 817-821.
11) members of the Endometriosis Guideline Core Group, Becker CM, Bokor A, Heikinheimo O, Horne A, Jansen F, et al. ESHRE guideline: endometriosis. Hum Reprod Open 2022; 2022: hoac009.
12) Bafort C, Beebeejaun Y, Tomassetti C, Bosteels J, Duffy JM. Laparoscopic surgery for endometriosis. Cochrane Database Syst Rev 2020; 10: CD011031.
13) Hodgson RM, Lee HL, Wang R, Mol BW, Johnson N. Interventions for endometriosis-related infertility: a systematic review and network meta-analysis. Fertil Steril 2020; 113: 374-382.e2.
14) Rossi AC, Prefumo F. The effects of surgery for endometriosis on pregnancy outcomes following in vitro fertilization and embryo transfer: a systematic review and meta-analysis. Arch Gynecol Obstet 2016; 294: 647-655.
15) Ferraretti AP, La Marca A, Fauser BC, Tarlatzis B, Nargund G, Gianaroli L; ESHRE working group on Poor Ovarian Response Definition. ESHRE consensus on the definition of 'poor response' to ovarian stimulation for in vitro fertilization: the Bologna criteria. Hum Reprod 2011; 26: 1616-1624.
16) Dobson SJA, Lao MT, Michael E, Varghese AC, Jayaprakasan K. Effect of transfer of a poor quality embryo along with a top quality embryo on the outcome during fresh and frozen in vitro fertilization cycles. Fertil Steril 2018; 110: 655-660.
17) Li J, Du M, Zhang Z, Guan Y, Wang X, Zhang X, Liu J, Pan Z, Wang B, Liu W. Does a poor-quality embryo have an adverse impact on a good-quality embryo when transferred together? J Ovarian Res 2018; 11: 78.
18) Wintner EM, Hershko-Klement A, Tzadikevitch K, Ghetler Y, Gonen O, Wintner O, Shulman A, Wiser A. Does the transfer of a poor quality embryo together with a good quality embryo affect the In Vitro Fertilization (IVF) outcome? J Ovarian Res 2017; 10: 2.
19) Georgiou EX, Melo P, Baker PE, Sallam HN, Arici A, Garcia-Velasco JA, Abou-Setta AM, Becker C, Granne IE. Long-term GnRH agonist ther-

apy before in vitro fertilisation (IVF) for improving fertility outcomes in women with endometriosis. Cochrane Database Syst Rev 2019; 2019: CD013240.

20) Harb HM, Gallos ID, Chu J, Harb M, Coomarasamy A. The effect of endometriosis on in vitro fertilisation outcome: a systematic review and meta-analysis. BJOG Int J Obstet Gynaecol 2013; 120: 1308-1320.

21) Kaponis A, Chatzopoulos G, Paschopoulos M, Georgiou I, Paraskevaidis V, Zikopoulos K, Tsiveriotis K, Taniguchi F, Adonakis G, Harada T. Ultralong administration of gonadotropin-releasing hormone agonists before in vitro fertilization improves fertilization rate but not clinical pregnancy rate in women with mild endometriosis: a prospective, randomized, controlled trial. Fertil Steril 2020; 113: 828-835.

22) American College of Obstetricians and Gynecologists Committee on Gynecologic Practice and Practice Committee. Female age-related fertility decline. Committee Opinion No. 589. Fertil Steril 2014; 101: 633-634.

23) Schwartz D, Mayaux MJ. Female fecundity as a function of age: results of artificial insemination in 2193 nulliparous women with azoospermic husbands. Federation CECOS. N Engl J Med 1982; 306: 404-406.

24) Hornstein MD. State of the ART: Assisted Reproductive Technologies in the United States. Reprod Sci Thousand Oaks Calif 2016; 23: 1630-1633.