Akita J Med **46** : 49-55, 2019

A NOVEL THREE-DIMENSIONAL/FOUR-DIMENSIONAL ULTRASOUND ANALYSIS OF THE EFFECT OF THE TRANSFER POSITION IN IN VITRO FERTILIZATION AND EMBRYO TRANSFER ON THE PREGNANCY RATE

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(received 31 May 2019, accepted 20 June 2019)

Abstract

Purpose : To identify embryo transfer position using two proposed new measurements in three-dimensional (3D)/four-dimensional (4D) ultrasonography, and examine its relationship to pregnancy rate.

Methods : We studied 105 cycles of in vitro fertilization and embryo transfer of good quality embryo performed between May and December 2016. Endometrial thickness and volume, distance from the embryo to the fundus (TDF), and as new measures, distance from the embryo to the center of both internal ostia of the fallopian tubes (transfer distance of the lateral position [TDL]), and distance from the embryo to the central endometrium area (transfer distance of the front/back position [TDFB]) were measured. The pregnant and non-pregnant groups were compared.

Results : The pregnancy rate was 23.8%. TDL values in the pregnant and non-pregnant groups were 26.9 ± 16.3 and 18.0 ± 16.0%, respectively, showing a significant difference (p = 0.011). There was a significant positive correlation between endometrial thickness and volume.

Conclusions : The TDL was significantly longer in the pregnant group, implying the possibility that lateral deviation of the embryo transfer position does not affect the pregnancy rate. More accurate evaluation of embryo transfer site using 3D/4D ultrasound analysis may contribute to further improvement in pregnancy rates.

Key words : embryo transfer, in vitro fertilization, pregnancy rate, ultrasonography

Introduction

In in vitro fertilization and embryo transfer (IVF-ET), embryo transfer is a simple but important final step, and the embryo transfer technique has changed since in vitro fertilization began[1,2]. A recent meta-analysis demonstrated that ultrasound-guided embryo transfer leads to higher pregnancy and birth rates, suggesting the efficacy of the combined use of ultrasound during embryo transfer[1]. In addition to two-dimensional (2D) ultrasound imaging, Baba *et al.* studied three-dimensional (3D) ultrasound examination for the first time in 2000 and highly suggested the usefulness and potential of 3D ultrasound examination in embryo transfer[3]. They reported that 3D ultrasound showed the tip of the catheter efficiently

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and accurately, and that it can be effectively used without delay[3]. More accurate and effective sites of embryo transfer have been studied using 2D/3D ultrasound[3-6]. There were cases in which the site of embryo transfer, which was considered optimal in 2D ultrasound, turned out to not be optimal in 3D ultrasound[7]. Currently, 3D ultrasound seems to be more useful than 2D ultrasound for identifying the embryo transfer position[3].

Many studies have shown no significant differences in parameters such as the endometrial thickness, volume, and blood flow between the pregnant and non-pregnant groups[8-11]. However, it has been reported that the lower the endometrial volume, the lower the pregnancy rate[12], and that the flow index for the endometrial blood flow assessment predicts pregnancy most efficiently[13].

In addition, some studies have examined the pregnancy rate using the transfer distance from the fundal endometrial surface to the embryo as a parameter[5,14-17]. Fang *et al.* measured the transfer distance from the fundus (TDF)[5]. The results of these studies vary in the actual transfer distance from the fundal endometrial surface to the embryo. Tiras *et al.* reported a higher pregnancy rate with embryo deposition ≥10 mm (especially 10-20 mm) from the fundus[15]. Valentina *et al.* reported a significantly lower pregnancy rate with embryo deposition ≥15 mm from the fundus[14]. In 2005, Geregely *et al.* identified the maximal implantation potential (MIP) point as the optimal position of 3D ultrasound-assisted embryo transfer[18]. In 2010, through further investigations with an increased number of cases, they found that use of the MIP point increased the pregnancy rate by more than 10%[19]. The MIP point was used as a transfer position, but it was determined based on points along the trajectory from the ampullary segment of the fallopian tube, not on plots created from detailed measurements, which would be more accurate[18,19]. As described above, a consensus on the optimal intrauterine embryo transfer position in the xyz axis has not been reached ; thus, further investigation of the usefulness of the method is required[20,21]. Especially, ultrasound imaging of the site of the embryo in the coronal plane is characteristic to 3D ultrasound. Although some studies have examined the methods of assessment of the transfer position in the coronal plane, a consensus has not been reached[18,19,22,23].

In this study, we assessed the embryo transfer position by calculating the transfer distance from the fundal endometrial surface to the embryo (TDF) in the sagittal plane. In addition, as new measures, we also calculated the transfer distance from the center of both internal ostia of the fallopian tubes to the embryo in the coronal plane as the transfer distance of the lateral position (TDL) and the transfer distance from the central area of the endometrium to the embryo in the axial plane as the transfer distance of the front/back position (TDFB) using 3D/four-dimensional (4D) transvaginal ultrasound. These new measurements are expressed as percentages.

Materials and Methods

Study population

One hundred five cycles of IVF-ET of good quality embryo (good quality embryo : ≥4 BB) (i.e., 35 cycles of fresh embryo transfer and 70 cycles of frozen-thawed embryo transfer) performed in Akita University Hospital between May 2016 and December 2016 were included in this study. This study was approved by the hospital's ethics committee (register number : 1598), and all patients provided informed consent before participation.

Measurement method

Endometrial thickness in the sagittal plane in embryo transfer was measured using a 3D/4D ultrasound probe. All examinations were performed using a General Electric Voluson Expert series ultrasound machine (Voluson E10) with a 3D/4D RIC6-12 transvaginal probe (GE Healthcare Japan, Tokyo, Japan). After 3D construction of the endometrial part using images of the uterus in the xyz axis (i.e., the sagittal, horizontal, and coronal planes), endometrial volume measurements were performed in the Virtual Organ Computer-aided AnaLysis mode. The site of the embryo was defined as the position of air bubbles that were introduced into the uterine cavity along with the embryo. Then, the following parameters were measured : (i) the transfer distance from the fundal endometrial surface to the air bubble in the sagittal plane (TDF) ; (ii) transfer distance from the center of both internal ostia of the fallopian tubes to the air bubble in the coronal plane (TDL) ; and (iii) transfer distance from the central area of the endometrium to the air bubble in the axial plane (TDFB) (Figs. 1 and 2).

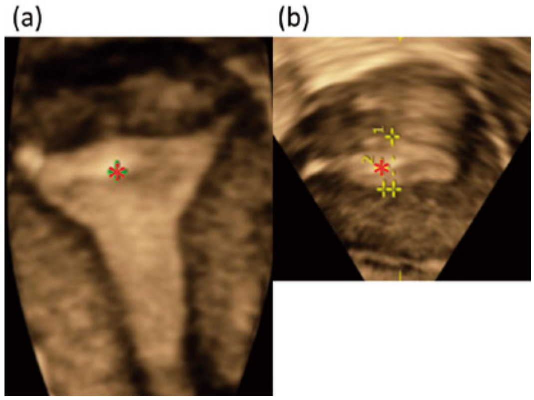

Fig. 1. Example of the embryo transfer position in 3D/4D ultrasound images.
(a) Coronal plane. (b) Axial plane. (*=embryo)

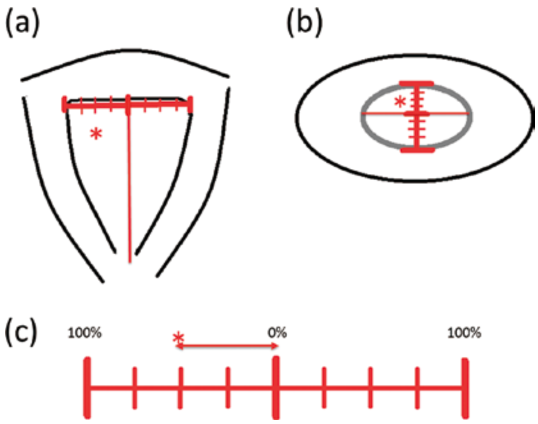

Fig. 2. New measurements assessed the transfer distance of the lateral position (TDL) in the coronal plane and the transfer distance of the front/back position (TDFB) in the axial plane using 3D/4D transvaginal ultrasound. (*=embryo)
(a) Transfer distance from the center of both internal ostia of the fallopian tubes to the air bubble in the coronal plane (TDL).
(b) Transfer distance from the central area of the endometrium to the air bubble in the axial plane (TDFB).
(c) TDL and TDFB are expressed as percentages. The center line is set to 0%, both ends are set to 100%, and the interval is divided into 100%.

Measurement items

The measurement items were (1) endometrial thickness, (2) endometrial volume, (3) TDF, (4) TDL, and (5) TDFB. Measurement items (1) to (3) are presented as actual measurement values, whereas (4) and (5) are expressed as percentages in order to make an objective evaluation because there are individual differences in the size and shape of women's uteri. On the other hand, for TDF, it is difficult to evaluate the ratio, so we used the measured value in a previous report[5,14-17]. The distance from the center of both internal ostia of the fallopian tubes to the end of the internal ostia of the fallopian tubes and the distance from the central area of the endometrium to the endometrial surface were divided into 100% (with the center line set to 0% and both ends set to 100%).

Statistical analysis

Each measurement item was compared between the pregnant and non-pregnant groups. The analysis was performed using IBM SPSS Statistics version 24.0 (IBM Corp., Armonk, NY, USA). The Mann-Whitney *U* test and Spearman rank correlation coefficients were also used in the analysis. *P*-values <0.05 were considered statistically significant.

Results

Patient characteristics

The patients were divided into the pregnant group ($n=25$) and the non-pregnant group ($n=80$) and then analyzed. There was no significant difference between the pregnant and non-pregnant groups in the following parameters: age, type of infertility (primary or secondary), infertility duration, cause of infertility (tubal factor, male factor, endometriosis, polycystic ovary syndrome, mixed, or unexplained), type of transplantation (fresh embryo or frozen-thawed embryo) (Table 1).

Statistical results

In 105 cycles, the TDL value was significantly higher in the pregnant group than in the non-pregnant group. For the remaining 4 measurement parameters (endome-

Table 1. Comparison of baseline characteristics between pregnant and non-pregnant group.

	Pregnant (n=25)	Non pregnant (n=80)	P value
Age (years)	36.4±3.0	37.4±3.7	0.082
Type of infertility (n, %)			0.380
Primary	19 (76)	67 (83.75)	
Secondary	6 (24)	13 (16.25)	
Infertility duration (years)	4.3±3.1	4.9±3.4	0.464
Couse of infertility (n, %)			0.544
Tubal	4 (16)	14 (17.5)	
Male	5 (20)	17 (21.25)	
EM	4 (16)	9 (11.25)	
PCOS	0	1 (1.25)	
Mixed	0	9 (11.25)	
Unexplained	12 (48)	30 (37.5)	
Type of ET (n, %)			0.871
Fresh	8 (32)	27 (33.75)	
Frozen	17 (68)	53 (66.25)	

Data are presented as mean±SD or n (%)

Table 2. Comparison of endometrial 3D ultrasound parameters on the day of ET between pregnant and non-pregnant groups.

	Pregnant (n=25)	Non pregnant (n=80)	P value
Endometrial thickness (mm)	11.9 ± 2.8	10.7 ± 2.4	0.061
Endometrial volume (cm^3)	5.1 ± 3.0	4.4 ± 2.2	0.375
TDF (mm)	5.4 ± 1.8	6.1 ± 3.3	0.824
TDL (%)	26.9 ± 16.3	18.0 ± 16.0	0.011
TDFB (%)	11.8 ± 12.3	12.8 ± 9.8	0.365

TDF : transfer distance from fundus, TDL : transfer distance of lateral position, TDFB : transfer distance of front/back position.

trial thickness, endometrial volume, TDF, and TDFB), no significant difference was observed between the pregnant and non-pregnant groups (Table 2).

In subgroups analysis of infertility cause, none of the groups found a significant difference between the pregnant and non-pregnant groups in the 5 measurement parameters.

There was a significant positive correlation between endometrial thickness and volume (Fig. 3).

Discussion

Many studies have examined the pregnancy rate using the transfer distance from the fundal endometrial surface to the embryo as a parameter[5,14-17]. However, a consensus on the optimal site of embryo transfer as a decisive

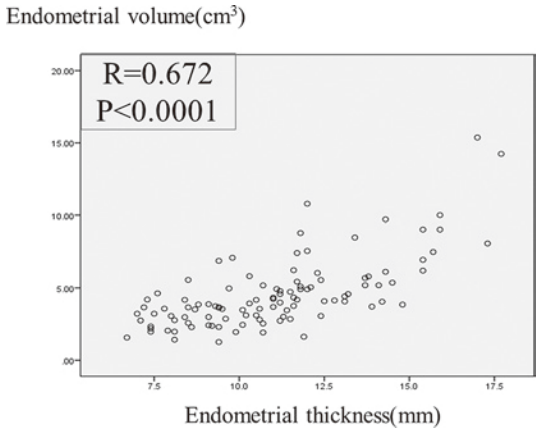

Fig. 3. Positive correlation between endometrial thickness and volume.

parameter using 3D reconstruction of the site of the embryo in the endometrial cavity has not been reached[20,21].

In addition to endometrial thickness and volume and TDF, this study examined the usefulness of TDL and TDFB as additional parameters for identifying the optimal embryo transfer position. In the analysis of 105 cycles of good quality embryo transfer, TDL was significantly longer in the pregnant group than in the non-pregnant group, there is a possibility that lateral deviation of the embryo transfer position does not affect the pregnancy rate. However, there was no significant difference between the pregnant group and the non-pregnant group in any parameters such as endometrial thickness and volume, TDF, and TDFB.

According to reports showing a higher pregnancy rate with embryo deposition at the MIP point[18,19] or the central area of the endometrium and a lower pregnancy rate with embryo deposition on the cervical side, as reported by Saravelos *et al.*[22,23], the closer the embryo deposition is to the central area of the endometrium, the higher the pregnancy rate. However, in this study, there was no significant difference in the transfer distance from the fundal endometrial surface to the embryo between the pregnant and non-pregnant groups. In addition, patients with higher TDL had a higher pregnancy rate, showing inconsistency with the results of other studies.

It is difficult to measure lateral deviation of the embryo transfer position in 2D ultrasonography, this study turned out that there is also variation in actual measurement in 3D ultrasonography. In addition, our findings has a possibility that lateral deviation of the embryo transfer position does not affect the pregnancy rate. Since successful embryo transfer is possible without considering TDL in the current IVF-ET, 2D ultrasound-assisted embryo transfer can be performed based on TDF without affecting the pregnancy rate as long as the site of embryo is near the midcoronal plane. In addition, it is currently difficult to adjust the catheter movement in the lateral position because the catheter could only be adjusted in an "in-out" motion and not in a "left-right" motion[4]. Because there was no ectopic pregnancy in this study, we could not identify embryo transfer positions which should be avoided to reduce this risk.

In addition, in subgroup analysis by cause of infertility, no significant difference was found between the pregnant and non-pregnant groups, suggesting that the cause of infertility and indication for in vitro fertilization do not substantially affect the optimal embryo transfer position.

Further, there was a significant positive correlation between endometrial thickness and volume. In the previous report, the relationship between endometrial thickness and endometrial volume is not clear[6,8,9]. However, from this result, current measures of endometrial thickness, which reflect endometrial volume to some extent, may be useful in the current 2D ultrasound during embryo transfer.

Because the number of cases was small in this study (N=105), future studies should include further examination with a larger sample size. In addition, future studies should investigate whether the results are stable in patients with uterine malformation. Although the assessment of embryo transfer position can be sufficiently performed using the current 2D ultrasound, 3D ultrasound may be useful in the identification of the optimal site of embryo transfer with more detailed settings such as the endometrial blood flow assessment. The endometrial blood flow may be affecting as a reason for the high pregnancy rate of lateral embryo transplantation. In addition, in this study, since the migration of the embryo is not considered as a premise, the assessment of the position of the embryo over time may be a more accurate implantation position.

This study found that there is also variation of lateral embryo transfer position in actual measurement in 3D ultrasonography and the results of this study has a possibility that TDL does not affect pregnancy rates. Although current transplantation techniques under 2D ultrasound guidance may be supported to some extent, more accurate evaluation of embryo transfer site using 3D/4D ultrasound analysis may contribute to further improvement in pregnancy rate.

Acknowledgments

We thank Mibuki Anzai, Kazumasa Takahashi and Mayumi Goto of the Department of Obstetrics and Gynecology, Akita University Graduate School of Medicine, who collaborated in the process of IVF-ET and data analysis, and Fumiaki Takahashi, Iwate Medical University Graduate School of Medicine, who collaborated in the process of medical statistics analysis.

We would like to thank Editage (www.editage.jp) for English language editing.

Disclosures

Conflict of interest : The authors have no conflict of interest to declare.

Human rights and informed consent : All procedures were followed in accordance with the ethical standards of the responsible committee on human experimentation (institutional and national) and with the Helsinki Declaration of 1964 and later versions. This study was approved by the Ethical Review Board of Akita University (No. 1598). Informed consent was obtained from all patients for being included in the study.

Animal studies : This study does not contain any studies with animal subjects performed by any of authors.

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