Archives of Gynecology and Obstetrics (2019) 299:565–570
https://doi.org/10.1007/s00404-018-4971-6

GYNECOLOGIC ENDOCRINOLOGY AND REPRODUCTIVE MEDICINE

Accuracy and efficacy of embryo transfer based on the previous measurement of cervical length and total uterine length

Panagiotis Bakas[1] · Mara Simopoulou[1] · Maria Giner[1] · Despina Tzanakaki[1] · Eythimios Deligeoroglou[1]

Received: 14 November 2017 / Accepted: 14 November 2018 / Published online: 21 November 2018

Abstract

Purpose Aim of the study was to examine the accuracy of embryo transfer based on the previous measurement of cervical length and total uterine length.

Methods The study has taken place in an academic-assisted reproduction clinic. 248 patients with infertility requiring IVF/ICSI treatment have been included. All patients underwent IVF/ICSI cycles with antagonist protocol and were treated with controlled ovarian stimulation using recombinant FSH (100–400 IU/day). On the day of oocyte retrieval, all patients were given vaginally natural micronized progesterone in a dose of 200 mg/tds. All patients had transvaginal ultrasound measurement of cervical length and endometrial cavity length prior to embryo transfer and measurement of embryo distance (intrauterine air bubbles) from fundal surface of uterine cavity and internal cervical os immediately after embryo transfer. Embryo transfer was performed on days 2–3. Primary outcome was to estimate the accuracy of embryo transfer based on the measurement of the embryo distance from middle of uterine cavity after embryo transfer and secondary outcome was to assess the effect of embryo distance from uterine fundus and internal cervical os to clinical pregnancy rate.

Results The clinical pregnancy rate was 42.7%. The mean embryo distance from the middle of endometrial cavity was 0.48 ± 0.02 cm, the mean embryo distance from the uterine fundus was 0.88 ± 0.32 cm, and from the internal cervical os was 1.67 ± 0.45 cm. Multiple regression analysis showed that the embryo distance from middle of cavity was related to endometrial cavity length and to the embryo distance from the fundus and it was not related to Cx length, total uterine length, embryo distance from internal Cx os, and embryo transfer length.

Conclusions Embryo transfer with the previous measurement of total uterine length and estimation of embryo transfer length can be performed with very good accuracy by a single operator.

Keywords Embryo transfer · IVF · ICSI · Accuracy · Ultrasound · Blind embryo transfer

Introduction

The development of assisted reproduction techniques (ART) and especially in vitro fertilization (IVF) and intracytoplasmic sperm injection (ICSI) are great scientific advancements in the management of infertile couples. Over the years significant improvements in every aspect of the IVF–ET process have been made that have led to significantly increased efficiency and success [1]. Their outcome of IVF/ICSI techniques is multifactorial including the woman's age, the patient's medical history, ovarian reserve, the quality of gametes and embryos, the endometrial receptivity, and embryo transfer technique [2–5]. The embryo transfer is an important and integral part of the IVF/ICSI treatments, and evidence supports that different methods or physicians performing the embryo transfer can affect the outcome [6, 7]. The introduction of transabdominal ultrasound-guided (TAUG-ET) embryo transfer appears to improve the chance of live birth/ongoing and clinical pregnancies compared with clinical touch, without increasing the chance of multiple pregnancy, ectopic pregnancy, or miscarriage [8, 9]. The use of transvaginal ultrasound guidance (TVUG) for embryo transfer yielded similar success rates compared with the TA ultrasound-guided procedure without requiring the assistance of a sonographer [10]. Furthermore, TVUG-ET for embryo transfer is superior to TAUG transfer in visualization

✉ Panagiotis Bakas
p_bakas@yahoo.com

1 2nd Department of Obstetrics and Gynecology, Aretaieion Hospital, University of Athens, Vas. Sofias Avenue 124A, 11526 Athens, Greece

of ET location and in lessening the pain and discomfort of the patients [11]. Although there is no consensus on the site of embryo transfer [12], several parameters including catheter type, injection speed, catheter withdrawal, embryo transfer depth, location of the catheter tip and location of air bubble, contamination with mucus or blood [13], uterine contractions, and level of difficulty in passing through the cervix [14, 15] may determine the outcome of IVF/ICSI cycle [16, 17].

Objective of the present study was to estimate the accuracy of embryo transfer based on the measurement of the embryo distance from middle of uterine cavity after embryo transfer and embryo distance from uterine fundus and internal cervical os.

Materials and methods

The present study has taken place in the Assisted Reproduction Unit of the 2nd Department of Obstetrics and Gynecology at Aretaieio Hospital, Athens between November 2013 and January 2017. 248 patients have participated in this study. The inclusion criteria were: (1) normal prolactin and TSH levels, (2) FSH levels < 12 IU/L on the days 2–4 of the menstrual cycle, (3) a normal recent hysterosalpingogram or a normal hysteroscopy (< 2 years old), (4) normal menstrual cycle, (5) age ≤ 42 years, and (6) no history of cervical cone biopsy. Exclusion criteria were: (1) FSH > 12 IU/L or estradiol > 80 pg/ml on days 2–4 of a normal menstrual cycle and (2) anti-mullerian hormone levels < 0.3 ngr/ml, and (3) patients with difficult embryo transfer catheter. A transvaginal ultrasound was performed in all patients on days − 7 to + 2 of menstrual cycle (day 0, the day of onset of menses) to exclude the presence of an ovarian cyst. TSH was used to exclude the possible thyroid dysfunction. Approval by the Hospital's ethics committee was obtained and all patients gave their informed consent. All patients enrolled in the study were consecutive patients meeting the aforementioned inclusion and exclusion criteria.

Primary objective was to estimate the accuracy of embryo transfer based on the measurement of the embryo distance from middle of uterine cavity after embryo transfer and secondary outcome was to assess the embryo distance from uterine fundus and internal cervical os.

Controlled ovarian stimulation (COS)

All patients were treated with recombinant FSH (Puregon, MSD, Athens, Greece, or Gonal-F, Merck, Athens, Greece). COS was started on day two of the menstrual cycle with 75–400 IU per day. The starting dose was based on AMH levels, FSH levels on days 2–4 of the menstrual cycle, antral follicle count, patient's age and BMI. All patients were treated using an antagonist protocol. Down-regulation was achieved with the use of Ganirelix (Orgalutran, MSD, Athens, Greece). Ganirelix was started when the leading follicle had reached a size of 14 mm or on the fifth day of the stimulation cycle and it was given as a daily dose of 0.25 mg/0.5 ml. When at least two follicles had reached a size of 18 mm, ovulation triggering was performed with the use of either 250 mcg/0.5 ml of recombinant chorionic gonadotropin alpha that was given subcutaneously (Ovitrelle, Merck Serono Europe Limited, London, UK) or human chorionic gonadotropin (hCG) at a dose of 10.000 IU subcutaneously (MSD, Athens, Greece). Monitoring of the ART/COS cycle was performed with serial transvaginal ultrasound and measurement of E2, progesterone, and LH on day six or day seven of the cycle and it was repeated as required until ovulation triggering. Oocyte retrieval was performed 34–36 h later. Embryo transfer was performed 2–3 days after oocyte retrieval.

All patients were told to use intravaginally natural micronized progesterone at a dose of 200 mg three times daily (Utrogestan capsules, Faran, Athens, Greece) for the support of the luteal phase. This was continued until a serum β-hCG measurement was performed 14 days later and if it was positive it was continued until 9–10 weeks of gestation.

Clinical pregnancy rate was defined as the presence of heartbeat on transvaginal ultrasound that was performed at about 7 weeks of gestation (endovaginal probe, Voluson S8, General Electric).

Ultrasound measurements

Uterine cavity length (UCL) was measured using a transvaginal probe and placing the cursors at the fundal part and at the level of internal cervical os (Fig. 1). Cervical length was measured using the "trace mode'' and the cursors were placed at the internal cervical os and at the external cervical os. The embryo distance from fundal surface of uterine cavity and internal cervical os was measured using transvaginal ultrasound immediately after embryo transfer and as the position of the embryos within the uterine cavity was considered as the hyperechogenic area seen within uterine cavity after embryo transfer "position of air bubbles". In the rare case of two air bubbles, it was estimated from the distance from the middle of the two air bubbles. All ultrasound measurements were performed just before embryo transfer (ET) and immediately after embryo transfer.

The total length of placement and advancement of the inner catheter into the uterine cavity (ETL) was estimated according to the previous measurement of cervical length and uterine cavity length and it was aiming to reach the tip of the inner catheter to the middle of uterine cavity. The ETL was calculated as follows: $A =$ uterine cavity length/2 + cervical canal length (Fig. 1).

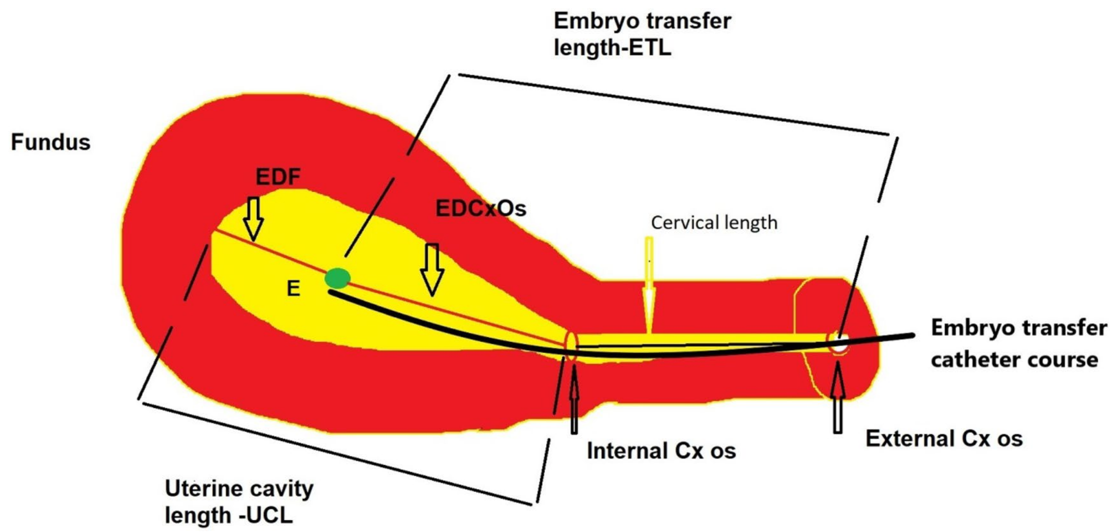

Fig. 1 Explanation of terms and process. Embryo transfer length = UCL/2 + cervical canal length (as measured by ultrasound) *EDF* embryo distance from fundus, *EDCxOs* embryo distance from internal cervical os, *Cx* cervical os, *E* embryos

Table 1 Patients characteristics (mean ± standard deviation)

Age	35.7 ± 2.2
Mean body weight (kg)	66.4 ± 9.4
Mean height (cm)	163 ± 3.6
Number of oocytes retrieved	10.1 ± 2.3
Number of metaphase II oocytes	8.5 ± 0.8
Fertilization rate	79%
Number of embryos G1, G2 on day 3	5.3 ± 1.1
Number of embryos transferred	1.74 ± 0.43
Clinical pregnancy	35.7%
Ongoing pregnancy rate	31.1%
Multiple pregnancy	9
Ectopic pregnancy	2

G1, G2 grade 1 and grade 2 embryo quality based on the morphological characteristics

As total uterine length was considered as the sum of cervical length and endometrial length in cm, as it was estimated by ultrasound (us), while mid-cavity depth was endometrial cavity/2 + cervical (Cx) length (Table 1) (Fig. 1).

Statistical analysis

A power calculation analysis was performed using the MedCalc software version 17.9.2 to estimate the required sample size. It was estimated that with a null hypothesis of 0 mm distance of the air bubbles from the middle of the cavity, for a mean decline in distance after ET of 3 mm from middle of cavity and a standard deviation of 14 mm, with a type I error 0.05 (significance) were required 231 patients to have a type II error 0.20 (power).

D'Agostino-Pearson test was used to assess if variables were following normal distribution or not. Because not all the examined parameters follow normal distribution, comparison of parameters was performed with the use of either Wilcoxon or Student's *t* test.

Multiple regression analysis was used to analyze the relationship between the examined variables and the ET accuracy. Also, rank correlation which calculates Spearman's rho and/or Kendall's tau rank correlation coefficients was used to assess the degree of association between the variables. A p value < 0.05 was considered statistically significant.

Results

The mean age of patients was 35.7 ± 2.2 (range 28–44), the mean weight was 66.4 ± 9.4 (range 39–110), and the mean height was 163 ± 3.6 cm (range 1.55–1.76).Patient characteristics are presented in Table 1. There was no correlation between patient's weight and height with the cervical length, while there was an association between patient's age and cervical length (correlation coefficient r −0.13 and p 0.037). The endometrial cavity length was not associated with patient's height, weight, and age (multiple regression analysis, p > 0.05). The mean total length of the uterus (UTL) was 6.6 ± 0.45 cm, while the mid-cavity depth was 5.29 ± 0.42 cm. The mean values of uterine parameters examined are presented in Table 2. The mean embryo distance from the middle of endometrial cavity was 0.48 ± 0.02 cm, the mean embryo distance from the uterine fundus was 0.88 ± 0.32 cm, and from the internal cervical os was 1.67 ± 0.45 cm. The mean values of embryo transfer parameters are presented in Table 3.

The clinical pregnancy rate was 42.7%.

Table 2 Means ± SD of measured parameters of uterine characteristics before and after ET

Mid-cavity depth (cm)	Endometrial cavity length (cm)	Cervical length by US (cm)	Uterus total length by US (UTLUS) (cm)	Uterus total length by mock measurement (cm)
5.29 ± 0.42 (3.95–6.60)	2.6 ± 0.49 (1.3–4.0)	3.9 ± 0.38 (3.2–5.4)	6.6 ± 0.45 (4.7–8.3)	7.0 ± 0.54 (5–8.2)

Mid-cavity depth = endometrial cavity length/2 + cervical (Cx) length. Endometrail cavity length: as measured by us from fundus to internal cervical (Cx) os

Uterus total length by US (UTLUS) = endometrial cavity length + Cx length (by US)

Range in brackets

Table 3 The mean values ± SD of measured parameters of embryo position within the uterine cavity after ET

Embryo distance from middle of cavity (cm)	Embryo distance from fundus (cm)	Embryo distance from internal cervical os (cm)	Internal catheter length (ETL) (cm)
0.48 ± 0.02 (– 0.55 to 1.69)	0.88 ± 0.32 (0.1–2.15)	1.67 ± 0.45 (0.6–3.0)	6.04 ± 0.34 (5.0–7.0)

Multiple regression analysis showed that the embryo distance from middle of the cavity was related to endometrial cavity length and to the embryo distance from the fundus and it was not related to Cx length, total uterine length, embryo distance from internal Cx os, and embryo transfer length (Table 4).

On the other hand, embryo distance from the fundus was related to endometrial cavity length, embryo distance from middle of the cavity and embryo distance from internal Cx os and independent of Cx length, total uterine length, and mid-cavity depth.

Logistic regression analysis was used to analyze binary data. No relation was found between the endometrial cavity length, embryo distance from the uterine fundus, embryo distance from the middle of the uterine cavity, and patient's weight with the clinical pregnancy rate. On the contrary, endometrial thickness (p 0.007) was found to be related to the clinical pregnancy rate (Table 5).

Comment

Embryo transfer is a crucial part of IVF/ICSI treatment and many factors can affect its effectiveness and technical completeness. Embryo transfer under transabdominal ultrasound guidance (TUSG-ET) has been considered as a significant step towards improvement of the process by increasing clinical pregnancy and live birth rate compared to clinical touch [8, 9]. But, TUSG-ET has requirements such as an adequately filled urinary bladder or an antiverted uterus to provide good picture, which is not the case in all patients. Furthermore, a recent meta-analysis has suggested that both US-guided transfer and clinical touch should be considered acceptable, as the benefit of US is not large and should be balanced against the increased cost and the need to change catheter type [18]. Recently, the introduction of TVUG-ET appears to have similar results with TUSG-ET without the necessity for an ultrasonographer [10] and is superior to TAUG-ET in visualization of ET location and in lessening the pain and discomfort of the patients [11]. In the

Table 4 Correlation coefficients and statistical significance between embryo distance from middle of cavity and independent variables

Independent variables	Coefficient	Std. error	$R_{partial}$	T	P
Constant	0.2379				
Cervical_length	0.05089	0.04714	0.06965	1.079	0.2815
Cavity_length	0.4092	0.05633	0.4252	7.263	< 0.0001
Embryo_distance from_Cx_os	– 0.03687	0.05391	– 0.04419	– 0.684	0.4947
Embryo_distance_from_fundus	– 0.7621	0.06182	– 0.6235	– 12.328	< 0.0001
Mid_cavity_depth	– 0.06121	0.04607	– 0.08561	– 1.328	0.1853

Table 5 Correlation coefficients and statistical significance between clinical pregnancy rate and independent variables

Independent variables	Coefficient	Std. error	$R_{partial}$	T	P
Constant	– 0.05512				
Cavity_length	0.1612	0.1298	0.08057	1.242	0.2155
Cervical_length	– 0.1181	0.08085	– 0.09463	– 1.460	0.1456
Distance_form_middle_of_cavity	– 0.2160	0.1469	– 0.09526	– 1.470	0.1428
Embryo_distance_from_Cx_os	– 0.05746	0.1200	– 0.03116	– 0.479	0.6324
Embryo_distance_from_fundus	– 0.08313	0.1754	– 0.03084	– 0.474	0.6359
Endometrium	0.06874	0.02594	0.1700	2.650	0.0086

past, several studies have tried to perform embryo transfer by measuring total uterine length and the position of the tip of the inner catheter [12, 19, 20] in an attempt to obviate the need for ultrasound guidance during embryo transfer [20]. Objective of the present study was to investigate if it is possible to perform embryo transfer based on pre-transfer measurement with transvaginal ultrasound of the total uterine length, the endometrial cavity length, the cervical length, and the embryo transfer length based on the equation that has been described in "Materials and methods". Aim was deposition of the embryos in the middle of the uterine cavity (mid-cavity).

It has been found that the mean embryo distance from the middle of uterine cavity was 0.48 ± 0.02 cm (range -0.55 to 1.69 cm). In 29.2% of cases (76/260) the embryo distance from mid-cavity was less than 3 mm. The decline from exact mid-cavity transfer implies that it is not possible to accurately estimate the exact total length of the uterus using ultrasound measurement but, embryo transfer can be performed with very good approximation of the middle of uterine cavity estimating uterine dimensions.

The mean embryo distance from the fundal endometrial surface (EDF) was 0.88 ± 0.32 mm (range 0.1–2.15 cm). There was only one case with 1 mm EDF and two cases with 3 mm EDF. The rest of the cases had a EDF $\geq$ 4 mm. These findings provide a suitable deposition site for the embryos since it is considered that the deposition of the embryos within or very close to fundal endometrial surface significantly reduces the chances of implantation [19]. Also, remains unclear up to now which is the best distance of the embryo deposition from the uterine fundus.

A significant point of distinction of the present study from the previous studies [20–22] is that it does not use clinical touch before embryo deposition or the use of transabdominal ultrasound guidance for embryo transfer [19]. Although, it has been suggested that the uterine length measurement followed by clinical touch provides the same IVF outcomes as transabdominal ultrasound-guided embryo transfer [21] and that the previous uterine length measurement followed by clinical touch showed slightly higher implantation and pregnancy rates, it seems that it would be better if clinical touch could be avoided to prevent any potential damage to the endometrium and blood release to endometrial cavity that could adversely affect the implantation rate.

Potential advantages of this method could be that it can be performed by a single operator obviating the need of an assistant for transabdominal ultrasound-guided embryo transfer and easiness to be taught to trainees since use of transvaginal ultrasound and assessment of the uterus are integral part of everyday clinical practice. Also, it avoids the expertise and dexterity that requires the transvaginal approach of embryo transfer [23]. In terms of better diagnosis and patient treatment, potential advantages could possibly be considered that it does not require full urinary bladder avoiding patient discomfort and it could be applied in patients with retroverted uterus or inadequately filled bladder where there is poor image of transabdominal ultrasound.

Limitations of the study could be the relatively small number of patients for conclusions to be made in relation to the effect of embryo distance from the fundal surface to the outcome of IVF/ICSI.

In conclusion, it appears that embryo transfer with the previous measurement of total uterine length and estimation of embryo transfer length can be performed with very good accuracy by a single operator and without the need for ultrasound guidance or clinical touch maneuver. These findings have to be confirmed by additional future studies.

Compliance with ethical standards

Conflict of interest There was no conflict of interest for any of the participating authors.

References

1. Anderson RE, Nugent NL, Gregg AT, Nunn SL, Behr BR (2002) Transvaginal ultrasound-guided embryo transfer improves outcome in patients with previous failed in vitro fertilization cycles. Fertil Steril 77(4):769–775
2. Spitzer D, Haidbauer R, Corn C, Stadler J, Wirleitner B, Zech NH (2012) Effects of embryo transfer quality on pregnancy and live birth delivery rates. J Assist Reprod Genet 29:131–135
3. Schwartz LB, Chiu AS, Courtney M, Krey L, Schmidt-Sarosi C (1997) The embryo versus endometrium controversy revisited as it relates to predicting pregnancy outcome in in vitro fertilization–embryo transfer cycles. Hum Reprod 12:45–50
4. Raga F, Bonilla-Musoles F, Casan EM, Klein O, Bonilla F (1999) Assessment of endometrial volume by three-dimensional ultrasound prior to embryo transfer: clues to endometrial receptivity. Hum Reprod 14:2851–2854
5. Hoozemans DA, Schats R, Lambalk CB, Homburg R, Hompes PG (2004) Human embryo implantation: current knowledge and clinical implications in assisted reproductive technology. Reprod Biomed Online 9:692–715
6. Levi Setti PE, Albani E, Cavagna M, Bulletti C, Colombo GV, Negri L (2003) The impact of embryo transfer on implantation—a review. Placenta 24(Suppl.B):S20–S26
7. Mains L, Van Voorhis BJ (2010) Optimizing the technique of embryo transfer. Fertil Steril 94:785–790
8. Brown J, Buckingham K, Buckett W, Abou-Setta AM (2016) Ultrasound versus 'clinical touch' for catheter guidance during embryo transfer in women. Cochrane Database Syst Rev 17(3):CD006107. https://doi.org/10.1002/14651858.CD006107.pub4
9. Abou-Setta AM, Mansour RT, Al-Inany HG, Aboulghar MM, Aboulghar MA, Serour GI (2007) Among women undergoing embryo transfer, is the probability of pregnancy and live birth improved with ultrasound guidance over clinical touch alone? A systemic review and meta-analysis of prospective randomized trials. Fertil Steri 88(2):333–341

10. Bodri D, Colodrón M, García D, Obradors A, Vernaeve V, Coll O (2011) Transvaginal versus transabdominal ultrasound guidance for embryo transfer in donor oocyte recipients: a randomized clinical trial. Fertil Steril 95(7):pp. 2263–8, 2268
11. Karavani G, Ben-Meir A, Shufaro Y, Hyman JH, Revel A (2017) Transvaginal ultrasound to guide embryo transfer: a randomized controlled trial. Fertil Steril 107(5):1159–1165
12. Cenksoy PO, Fıcıcıoglu C, Yesiladali M, Akcin OA, Kaspar C (2014) The importance of the length of uterine cavity, the position of the tip of the inner catheter and the distance between the fundal endometrial surface and the air bubbles as determinants of the pregnancy rate in IVF cycles. Eur J Obstet Gynecol RB 172:46–50
13. Alvero R, Hearns-Stokes RM, Catherino WH, Leondires MP, Segars JH (2003) The presence of blood in the transfer catheter negatively influences outcome at embryo transfer. Hum Reprod 18:1848–1852
14. Fanchin R, Righini C, Olivennes F, Taylor S, de Ziegler D, Frydman R (1998) Uterine contractions at time of embryo transfer alter pregnancy rates after in vitrofertilization. Hum Reprod 13:1968–1974
15. Lesny P, Killick SR, Tetlow RL, Robinson J, Maginess SD (1998) Embryo transfer—can we learn anything from the observation of junctional zone contractions? Hum Reprod 13:1540–1546
16. Tiras B, Polat M, Korucuoglu U, Zeyneloglu HB, Yarali H (2010) Impact of embryo replacement depth on in vitro fertilization and embryo transfer outcomes. Fertil Steril 94:1341–1345
17. Schoolcraft WB, Surrey ES, Gardner DK (2001) Embryo transfer: techniques and variables affecting success. Fertil Steril 76:863–870
18. Texeira DM, Dassuncao LA, Vieira VR, Barbasa AP, Coelho Neto MA, Nastri CO, Martins WP (2015) Ultrasound guidance during embryo transfer: a systematic review and metaanalysis of randomized controlled trials. Ultrasound Obstet Gynecol 45:139–148
19. Pope CS, Cook EK, Arny M, Novak A, Grow DR (2004) Influence of embryo transfer depth on in vitro fertilisation and embryo transfer outcomes. Fertil Steril 81(1):51–58
20. Revelli A, Rovei V, Dalmasso P, Genarelli G, Racca C, Evangelista F, Benedetto C (2016) Large randomized trial comparing transabdominal ultrasound-guided embryo transfer with a technique based on uterine length measurement before embryo transfer. Ultrasound Obstet Gynecol 48:289–295
21. Lambers MJ, Dogan E, Kostelijk H, Lens JW, Schats R, Hompes PGA (2006) Ultrasonographic guided embryo transfer does not enhance pregnancy rates compared with embryo transfer based on previous uterine length measurement. Fertil Steril 86:867–872
22. Maldonado LG, Aoki T, Busato W, Pasqualotto FF, Iaconelli A, Borges E (2010) Embryo transfer based on previous uterine length measurement: a prospective randomized study. Hum Reprod 25:i99
23. Anderson RE, Nugert NL, Gregg AT, Nunn SL, Behr BR (2002) Transvaginal ultrasound guided embryo transfer improves outcome in patients with previous failed in vitro fertilization cycles. Fertil Steril 77(4):769–775