Chapter 18
Single Embryo Transfer: Significance of the Embryo Transfer Technique

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Introduction

Elective Single Embryo Transfer

Embryo transfer (ET), an apparently simple technique, constitutes a significant, rate-limiting step that is crucial to the success of any in vitro fertilisation (IVF) cycle. Multiple embryo transfer during IVF increases multiple pregnancy rate, thus also raising maternal and perinatal morbidity [1]. There are several advantages of elective single embryo transfer (eSET); it is the only effective strategy known to minimise the risk of multiple pregnancy that can also be applied to patients aged 36–39 years, thus increasing the safety of ART in this age group [2]. Though a single fresh embryo transfer may be associated with a lower live birth rate than double embryo transfer (DET) [1], no significant differences have been reported in the cumulative pregnancy and delivery rates following eSET compared to DET, accompanied with a significant decrease in the multiple gestation rate with better neonatal and obstetric outcomes [1, 3–5]. Authors have even reported significantly higher cumulative pregnancy rates (54.0 % vs. 35.0 %) and cumulative live birth rates (41.8 % vs. 26.7 %; $p<0.0001$), but lower multiple birth rates (1.7 % vs. 16.6 %; $p<0.0001$) following eSET compared to DET [2]. The comparative efficacy between SET and DET was observed in a natural as well as a hormone-stimulated cycle [1]. In women aged <35 years, a significantly higher rate of 'healthy baby' per transfer cycle has been reported following eSET compared to selective double embryo transfer (sDET), regardless of stage of embryo development [6]. For a woman with a 40 % chance of live birth following a single cycle of DET, the

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chance following repeated SET would be between 30 % and 42 %; for a woman with a 15 % risk of multiple pregnancy following a single cycle of DET, the risk following repeated SET would be between 0 % and 2 % [1].

Factors that Influence the Success of eSET

Failure to achieve a live birth following IVF may be attributed to the embryo transfer stage due to lack of good quality embryo(s), lack of uterine receptivity, or the transfer technique itself [7]. The success of eSET is influenced by the following factors.

Patient Selection

To ensure optimal outcomes with eSET, patient selection plays an important role. Selective application of eSET in a small group of good-prognosis patients may be effective in reducing the overall multiple rate of an entire IVF population without substantially reducing the likelihood of achieving a live birth [8]. Good-prognosis patients may be considered as women aged ≤35 years, in their first or second IVF attempt, and with at least two good quality embryos available for transfer. Women aged 36–37 years may also be considered good-prognosis patients for eSET if good quality embryos, particularly blastocysts, are available for transfer as blastocyst stage embryo transfer generally increases the chance of implantation and live birth compared with cleavage stage embryo transfer. Kresowik et al. [9] reported a live birth rate of 64.6 % and a multiple birth rate of 3.4 % following a mandatory single embryo transfer (mSET) policy for all women aged <38 years, with at least seven zygotes, no prior failed fresh cycle, and at least one good quality blastocyst [9]. In women aged ≥38 years, eSET may result in a significant reduction in live birth rate compared with DET [8].

Embryo Quality

Success with an eSET would be compromised if the embryo quality suffered. Morphological methods used to select the most viable embryos for transfer may be far from predictive of the implantation potential of these embryos. A paradigm shift using morphological factors along with metabolic, protein and genetic markers in culture media aims to enhance embryo selection and IVF success rates [10] and is particularly useful in selecting single embryos for transfer. Several advanced techniques for embryo selection such as rapid polymerase chain reaction (PCR)-based comprehensive chromosome screening (CCS) and trophectoderm biopsy prior to SET have been reported to enhance embryo selection, with a resultant increase in the ongoing pregnancy rate (55.0 % vs. 41.8 %, respectively; $p<0.01$) and a

decreased miscarriage rate compared with traditional blastocyst SET (24.8 % vs. 10.5 %; $p<0.01$). These novel screening techniques may provide a practical way to eliminate multi-zygotic multiple gestation without compromising clinical outcomes per cycle [11]. Image acquisition and time-lapse analysis of the embryos optimise accurate embryo selection of viable embryos by identifying the morphokinetic parameters specific to embryos capable of implanting and thus, make it possible to determine the exact timing of embryo cleavages in a clinical setting [12]. New technology, based on embryo developmental and morphological characteristics, using multilevel images combined with a computer-assisted scoring system (CASS) has the potential to overcome the disadvantages with standard embryo evaluation with a superior ability to predict implantation and live birth [13].

Culture Protocols

Improvements in culture protocols facilitate extended culture to the blastocyst stage and by enabling self-selection of viable embryos and improved uterine and embryonic synchronicity, result in higher implantation rates [14, 15]. Excellent pregnancy rates have been reported with SET blastocyst culture with live birth delivery rates comparable to double cleavage stage transfer (27.2 % vs. 24.8 %) and decreased complications related to multiple births [15]. A significant threefold increase in day 5 single embryo transfers over an 8-year period (4.5 % in 2001 to 14.8 % in 2009; $p<0.0001$) has been associated with a significant decrease in the rate of multiple births from 44.8 % to 41.1 % ($p<0.0001$) [3].

Cryopreservation

Elective SET with cryopreservation has been suggested to be more effective in maximising the cumulative live births and significantly less expensive than DET in good-prognosis patients and therefore, from a cost-effectiveness perspective, should be adopted as a treatment of choice [8, 16]. In order to maintain the reduction in the rate of multiples achieved with fresh eSET, eSET should be performed in subsequent frozen-thawed embryo transfer cycles [8]. Patients should be informed of the reductions in both multiple pregnancy rate and overall live birth rate after a single fresh eSET when compared with DET in good-prognosis patients [8].

Significance of the Embryo Transfer Technique

The significance, growing awareness and positive clinical outcomes obtained following SET mandate the performance of a meticulous, atraumatic ET technique that aims to successfully deliver a single good quality embryo in the uterine cavity

without associated difficulty. The significance of the eSET technique stems from the fact that since the possibility of embryo selection in the uterine cavity is eliminated, efforts entailed in the preceding clinical and laboratory protocols and the embryo selection would be rendered useless and the cycle wasted if the ET technique was suboptimal. This is especially true of a fresh first cycle eSET, which can perhaps be salvaged with additional embryos, if available, but will leave fewer embryos for cryopreservation. However, in the case of the unplanned difficult single embryo transfer, the situation may rarely improve and could also compromise the quality of embryo transferred. Though there is no universally acceptable or standard technique for ET, factors documented to have a positive and negative impact on ET must be strictly respected to achieve the desired outcome.

Factors that impact the clinical outcome following ET include (1) routine evaluation of the uterine cavity to detect abnormalities, (2) mock embryo transfer immediately before the actual ET, (3) evaluation of uterine position and dimensions, (4) ultrasound guidance during ET, (5) depositing embryos in the mid-portion of the endometrial cavity, (6) the use of soft catheters, (7) avoidance of uterine contractions, blood, or mucus on the catheter, (8) ensuring an absolutely atraumatic transfer technique and (9) the experience and skill of the clinician performing ET. Evidence detailing the significance of each of these factors is presented below.

Factors that Play an Important Role Prior to ET

Routine Uterine Cavity Evaluation

A routine uterine cavity evaluation enables a thorough exploration of the uterine cavity to check for abnormalities, such as submucosal leiomyomas, adhesions, polyps and congenital abnormalities, that may interfere with a successful outcome. Endometrial cavity abnormalities have been reported with an incidence of 22.9 % following outpatient hysteroscopy in patients with a previous IVF-ET cycle, the correction of which markedly improves the outcome. Sufficient evidence to support the surgical removal of all abnormalities to improve the IVF-ET outcome and the value of performing this procedure before an initial cycle in patients without previous implantation failure is lacking. However, it would seem logical in an effort to minimise the number of cycles a patient must undergo. Three-dimensional saline sonohysterography may be particularly useful in the evaluation [17].

Evaluation of Uterine Position and Dimensions

Before proceeding to ET, it is essential to have adequate knowledge about the uterine position, anteverted (AV) or retroverted (RV) by ultrasonography (USG). An RV uterus at mock embryo transfer will often change position at real embryo transfer to

become AV [18], changing the course of the ET catheter. Misdirecting the ET catheter can be avoided by accurate knowledge of the uterine position at the time of embryo transfer. Following a comparative evaluation of 996 consecutive mock and real abdominal ultrasound-guided-(USG)-ET embryo transfer cycles, Henne and Milki [18] demonstrated a highly significant ($p < 0.0001$) change in the position of RV uteri at mock transfer (26 % of 55 % ETs) to AV at the actual transfer compared to the conversion of only 2 % of the 74 % of patients with an AV uterus at mock embryo becoming RV at the actual transfer. The change in uterine position was also noted in frozen-thawed embryo transfers (12 % of AV uteri at mock embryo transfer to RV and 33 % RV uteri to AV at real transfer; $p = 0.01$). Accordingly, patients with an RV uterus at mock embryo transfer should still present with a full bladder for embryo transfer, since a significant number will convert to an AV position [18]. Moreover, ultrasound evaluation of the uterocervical angulation and uterine cavity length prior to the actual transfer can optimise the ET technique and may reduce the rate of ectopic pregnancies [19, 20].

Mock/Trial Embryo Transfer

A mock/trial transfer is essential before actual ET as it enables a thorough knowledge of the uterine position (AV/RV), uterocervical length and angulation that may be of value in accurately guiding the course of the catheter during the actual ET. Additionally, it is of value in revealing intracavitary abnormalities that may interfere with pregnancy and in directing possible surgical management prior to ET. While the value of a mock transfer a few days before the actual procedure has been challenged owing to the change in the uterine position [18], a trial catheterization on the day of ET could prevent most of the unanticipated procedural difficulties during the transfer [17]. Moreover, a USG-trial transfer (UTT) in the office, in preparation for an IVF cycle, has shown to be beneficial as significant differences have been noted between patients when comparing difference in length (DL) to previous pregnancy status and the total cavity depth (sounding depth + DL) ($p < 0.05$) [21].

Hysteroscopic Revision of the Cervical Canal

Cervical stenosis may be associated with a technically difficult ET, reducing the chances of pregnancy after assisted reproductive procedures. Hysteroscopic revision of the cervical canal results in easier ET by facilitating the course of the transfer catheter through the cervical canal and thus, improved pregnancy rates in patients with cervical stenosis and histories of difficult ET [22].

Fluid Volume in the Transfer Catheter

The amount of fluid volume for day 3 transfer has been shown to have a significant impact on pregnancy and implantation rates. A high fluid volume (40–50 μL) for loading the transfer catheter resulted in significantly higher pregnancy (40 % vs. 23 %; $p = 0.012$) and implantation rates (24.4 % vs. 14.7 %; $p = 0.011$) compared to a low fluid volume (15–20 μL; $n = 94$) [23].

Removal of Cervical Mucus

Significantly higher clinical pregnancy rates (39.2 % vs. 22.6 %, respectively, $p < 0.001$), implantation (20.5 % vs. 12.2 %, respectively, $p < 0.001$) and live birth rates have been reported following removal of cervical discharge before ET, compared to patients in whom the cervical canal was not cleansed. This suggests that removal of cervical debris prior to ET may have a significant effect on the rate of implantation, pregnancy and live birth [24].

Bacterial Contamination

Microbial examination of samples from the fundus of the vagina, the cervix, the embryo culture medium prior and post-embryo transfer, the tip of the catheter and the external sheet shows that the presence of vaginal-cervical microbial contamination at the time of ET is associated with significantly decreased pregnancy rates (*Enterobacteriaceae*: 22.2 % vs. 51 %; *Staphylococcus* spp.: 17.6 % vs. 44 %; $p < 0.001$) when compared to negative culture groups [25]. While catheter contamination by upper genital tract microbes has been suggested to affect the success of ET and administration of antibiotics like amoxicillin and clavulanic acid before ET can significantly reduce microbial colonisation and catheter contamination rates [26], this intervention did not translate into better clinical pregnancy rates [26, 27]. Hence, the routine use of antibiotics at embryo transfer prior to ET is not recommended [26, 27].

Factors that Play an Important Role During ET

Ultrasound-Guided ET

The use of ultrasound guidance to perform ET has been one of the most significant advances in the ET technique over the traditional ‘blind’ clinical touch method. Despite the lack of a standard evidence-based protocol, there is substantial evidence

that both transabdominal [6, 7, 28, 29] and transvaginal [30] USG-ETs significantly increase clinical pregnancy, embryo implantation, ongoing pregnancy and live birth rates compared to clinical touch alone [6, 7, 28–30]. Occasional studies have demonstrated no benefit with USG-guided ET over the clinical touch method with reference to clinical pregnancy and implantation rates compared to previous ultrasonographic length measurement [31] and in the hands of an experienced operator [32, 33]. However, success in patients with a prior history of difficult uterine sounding or embryo transfer still relied heavily on USG-ET [32]. Of note, the 25 % chance of pregnancy using the clinical touch method alone increased to 32 % (from 28 % to 46 %) when USG-ET was performed instead [34].

Ultrasound-guided ET brings the following advantages to make this an indispensable technique to achieve an optimal outcome:

- It facilitates an accurate evaluation of the uterine position and cavity length before the actual embryo transfer and, hence, the transfer distance from the fundus (TDF).
- It facilitates the correct placement of the catheter in the endometrial cavity.
- It avoids contact of the catheter tip to the fundus.
- It confirms that the catheter is beyond the internal os in cases of an elongated, cervical canal.
- It allows direction of the catheter along the contour of the endometrial cavity, thereby avoiding disruption of the endometrium, plugging of the catheter tip with endometrium and instigation of bleeding.
- The requirement of a full bladder to perform transabdominal USG-ET is itself helpful in straightening the cervical-uterine access and improving pregnancy rates.
- It may facilitate an uncomplicated access through the cervix to access the uterine cavity, thus overcoming cervical stenosis [35].
- It enables visualisation of the catheter tip during ET and the position of embryo deposition [36].
- It significantly increases the frequency of easy transfers [29, 37] and decreases the incidence of difficult transfers and endometrial injury [38] possibly due to a decrease in cervical and uterine trauma [29] compared to the clinical touch method.
- It may be especially beneficial in patients with previously failed IVF cycles or in patients with previous cycles when embryos were transferred by the clinical touch method [30].

Indeed, tactile assessment of ET catheter placement has been considered unreliable as the outer guiding catheter inadvertently abutted the fundal endometrium in 17.4 % of transfers, indented the endometrium in 24.8 % and the transfer catheter embedded in the endometrium in 33.1 % transfers. Unavoidable sub-endometrial transfers occurred in 22.3 % of transfers, while USG-ET avoided accidental tubal transfer in 7.4 % of transfers [39]. Measurement of cavity depth by USG is clinically useful to determine the depth beyond which catheter insertion should not occur. The transfer distance from fundus (TDF = cavity depth minus depth of catheter insertion), measured

by USG, is highly predictive of pregnancy, unlike that measured by mock transfer as cavity depth by US has been reported to differ from cavity depth by mock by at least 10 mm in >30 % of cases [34]. Moulding the embryo transfer catheter according to the uterocervical angle, measured by ultrasound, increases clinical pregnancy and implantation rates and diminishes the incidence of difficult and bloody transfers compared with the 'clinical feel' method. Patients with large angles (>60°) had significantly lower pregnancy rates compared with those with no angle [40]. Significantly higher pregnancy rates per transfer have been reported when ultrasound visualisation was considered to be excellent/good (when the catheter could be followed from the cervix to the fundus by transabdominal ultrasound with the retention of the embryo-containing fluid droplet), compared to fair/poor transfers (where the sequence of events could not be documented). Performance of embryo transfer with a soft catheter under ultrasound guidance with good visualisation resulted in a significant increase in clinical pregnancy rates [36].

Though transvaginal USG-ET may be associated with increased patient comfort due to the absence of bladder distension, the total duration of transfer is statistically significantly higher compared to transabdominal USG-ET [41].

Two-dimensional (2D) USG-ET is the standard for image-guided transfers to monitor catheter passage through the cervix into the endometrial cavity [42], although three-dimensional (3D) USG offers better precision and optimal positioning of uterine catheter tip placement. This is an enhancement in the ET technique and has been shown to improve overall pregnancy rate compared with 2D sonography [42, 43]. Moreover, the disparity of ≥10 mm in transfer distance from the fundus (TDF) between 2D and 3D images may significantly impact the pregnancy outcome [43]. Irrespective of the USG mode used, the important role of USG-ET in optimising pregnancy outcomes warrants perfection in this technical skill.

Catheter Type

The type of catheter used for ET (soft/rigid and echogenic/non-echogenic) may influence the degree of trauma to the endometrial cavity during ET. Significantly higher pregnancy ($p<0.0005$) and implantation rates ($p<0.01$) have been reported with the ultrasoft catheters compared to the more rigid Frydman catheters [35]. A blinded comparison of endocervical and endometrial damage following the use of soft ET catheters [IVF Sydney Set (Cook, Limerick, Ireland), Elliocath (Ellios, Paris, France), Frydman classic 4.5 (CCD, Paris, France)] and rigid ET catheters [Memory Frydman 4.5 (CCD, Paris, France)] demonstrated significantly more frequent endocervical lesions with the soft (63 %) and rigid (85 %) Frydman catheter groups compared to other groups (Elliocath: 29 %, IVF Sydney Set: 26 %; $p<0.0001$). Severe endometrial lesions were significantly less frequently observed when soft catheters were used (85 %, 53 %, 32 % and 11 % for Memory Frydman, Frydman classic, Elliocath and IVF Sydney Set, respectively; $p<0.0001$) [44]. Blood on an ET catheter is a marker for endometrial microtrauma; all ET catheters

can lead to endocervical or endometrial damage, but severe endometrial lesions may less frequently be encountered when soft catheters are used [44]. Though no significant difference in implantation, clinical or ongoing pregnancy rates has been observed following ET with the echogenic catheters (Sure View catheter [44], the echogenic Wallace catheter [45] or the Cook Echo-Tip catheter [46]) and standard catheters without echogenic enhancement, echogenic catheters offer the benefit of superior visualisation due to their ultrasonic contrast properties. This minimises the need for catheter movement to identify the tip [44–46] and significantly shortens the duration of the ET procedure (defined as the interval between when the loaded catheter is handed to the physician and embryo discharge), thus simplifying USG-guided ET [45].

In addition to easy visualisation of the catheter tip, El-Shawarby et al. [47] reported a significantly lower rate of retained embryos in the catheter following ET with the Rocket catheter compared to the Wallace catheter ($p < 0.05$), although there was no difference in clinical pregnancy and implantation rates [47]. The use of a soft pass catheter was the only variable independently and significantly associated with pregnancy success (OR = 2.74) [48].

Depth of Embryo Transfer

Traditionally, ET has been performed blindly with the goal to place the embryos approximately 1 cm inferior to the fundal endometrial surface [49]. The depth of embryo replacement (difference between the cavity depth and depth of catheter insertion) during USG-ET has been shown to have a significant impact on the clinical outcome after controlling for potential confounders [49–53]. Significantly higher ($p < 0.05$) implantation rates (31.3 %, 33.3 % and 20.6 %, respectively) have been reported when embryos were deposited at a distance ≥15 mm (15 ± 1.5 mm or 20 ± 1.5 mm) between the catheter tip and the uterine fundus compared to <15 mm (mean = 10 ± 1.5 mm).

There was no difference between all three transfer groups regarding the main demographic and baseline characteristics of the patients, ovarian response, oocyte retrieval and IVF outcome. Characteristics of embryo transfer and luteal phase support were also similar [49]. While maintaining a uniform method of loading embryos into the embryo transfer catheter and the number and quality of embryos transferred, Pacchiarotti et al. [50] observed significantly higher clinical pregnancy rates (27.7 % vs. 4 %, respectively; $p < 0.05$) when the distance between the tip of the catheter and the uterine fundus at transfer was 10–15 mm compared to ≤10 mm [50]. Tiras et al. [51] buttressed these findings in a large study that included 5,055 USG-ETs in 3,930 infertile couples, observing higher pregnancy and ongoing PRs when the embryos were replaced at a distance >10 mm from the fundal endometrial surface. They suggested that a distance 10–20 mm seems to be ideal for embryo transfer to achieve higher PRs [51]. These findings have been further documented in a very recent study that reported clinical intrauterine pregnancy rates of 65.2 %,

32.2 % and 2.6 % when the distances between the fundal endometrial surface and the tip of inner catheter were <10 mm, 10–20 mm and 20 mm, respectively, suggesting that the optimal distance between the fundal endometrial surface and the tip of inner catheter is 1.5–2 cm [52]. According to Pope et al. [53], for every additional millimetre that embryos are deposited away from the fundus, the odds of clinical pregnancy increased by 11 % [53].

Avoiding Difficult Transfers

It is extremely important to avoid a difficult transfer by preplanning the ET technique, as this may significantly impact the clinical outcome of an eSET. Patients at risk for a difficult ET should be identified so that the ET can be appropriately planned. Embryo transfer is considered difficult if it has been time consuming, the catheter met great resistance, there was a need to change the catheter, sounding or cervical dilatation was needed, there was blood in any part of the catheter [54] or it required at least two attempts [55] and may often be associated with a poor clinical outcome.

In contrast, an 'easy' transfer has been suggested to be an atraumatic insertion of the catheter without touching the uterine fundus. When ET difficulty was evaluated as an independent factor for predicting pregnancy after taking into account the other confounding variables, it was observed that easy or intermediate transfers resulted in a 1.7-fold higher pregnancy rate compared to difficult transfers ($p < 0.0001$; 95 % CI = 1.3–2.2), suggesting that that the degree of difficulty of embryo transfer is an independent factor as regards achieving pregnancy after IVF/intracytoplasmic sperm injection (ICSI) [54]. Hysteroscopic assessment of endocervical and endometrial damage, inflicted by embryo transfer trial, revealed a significant concordance between the perceived difficulty of transfer, presence of blood on the catheter and degree of endometrial damage ($p < 0.05$). There were significantly higher minor and moderate endocervical lesions (35 % and 24 % of cases, respectively) in the difficult transfer group as compared to the easy transfer group (19 % and 3 %, respectively; $p < 0.05$). Within the easy transfer group, 65 % of patients had no endometrial damage, 32 % had minor lesions and 3 % had moderate lesions compared to 42 %, 29 % and 29 % in the difficult transfer group, respectively. Moreover blood on the catheter was noted in 2 %, 56 % and 71 % of the easy, moderate and difficult transfer groups, respectively. The authors concluded that clinical perception of difficulty of transfer and the presence of blood on the catheter are directly associated with endometrial disruption [56].

While the use of external guidance during ET has been shown to significantly reduce live birth delivery rates (LBDR) as compared to an atraumatic ET with a soft catheter (26.0 % vs. 32.5 %, respectively), grasping the portio vaginalis with a tenaculum is reported to result in the lowest clinical pregnancy rates (CPR) and LBDR, compared to ET with a soft catheter, after external guidance or probing of the cervix with a stylet. Though considered to be superior to the use of external guidance in cases of difficult ETs [57], the use of a stylet in the event of a failure of

the soft inner catheter to negotiate the internal os is associated with significantly lower implantation (19.4 % vs. 13.8 %), clinical pregnancy (41.9 % vs. 31.1 %) and live birth rates (37.3 % vs. 27.4 %), compared to ETs without the use of a stylet [58].

Physical contact (such as touching the uterine fundus with the tip of the ET catheter during transfer) results in mechanical stimulation activity of the uterus or junctional zone contractions (JZCs) that may relocate intrauterine embryos. Hence, all efforts should be made to avoid triggering JZCs as this has been implicated in cases of IVF-ET failure or ectopic pregnancy [59]. Embryo transfers that provoke bleeding and those that result in retention of embryos in the cervix and embryo expulsion have all been linked to JZCs [19, 52, 60]. Physicians should use a stepwise approach in difficult embryo transfers [52].

Injection Speed

There appears to be an inverse relationship between ejection speed (i.e., the velocity of discharge of embryo/s plus media from the catheter) and the subsequent development rate of the transferred embryo/s. Thus, reducing the ejection speed of the transferred load may help avoid developmental delay and decreases the associated embryo(s) injury. Specifically, the embryo development rate has been found to be the slowest in embryos exposed to a fast ET with a higher mean apoptotic index of embryos compared to the group exposed to a slow ET (17.6 % vs. 5.6 %, respectively) and the control group (2.58 %). Hence, embryos should be transferred with the lowest possible ejection speed [61].

Experience of the Practitioner

Apart from the numerous factors that should be considered while performing an ET, the most influential factor in the outcome is the operator's experience in the use of each system, and not the system itself [62]. The physician factor is an important variable in the overall ET technique and can result in significant differences in clinical pregnancy rates ($p \leq 0.01$), as shown by comparisons between different providers using the same method of loading embryos into the embryo transfer catheter and the same number of embryos transferred [63]. Desparoir et al. [64] demonstrated pregnancy rates of 29.9 % for attending physicians (>20 years of experience), 28.2 % for assistant physicians (2–5 years of experience) and 19.1 % for resident physicians (<6 months of experience) ($p < 0.05$). Resident physicians used tight difficult transfer (TDT) catheter more often than attending physicians: 42 % vs. 21.3 %, respectively ($p < 0.05$), suggesting that resident physicians require monitoring to avoid lower pregnancy rates [64]. Authors have even suggested that in the hands of experienced, skilled operators, neither the choice of transfer catheter and difficulty of transfer nor observations of blood on the transfer catheter will make any significant impact on pregnancy outcomes [65].

Embryo After-Loading

Despite significantly more transfer catheters with mucus contamination compared to direct transfers (25.58 % vs. 5.95 %), there was a trend towards an increase in clinical pregnancy rate following the embryo after-loading technique compared to the direct technique (52.4 % vs. 34.9 %) [21]. However, more evidence is required to substantiate these results.

Blood on the Catheter

The presence of blood on the transfer catheter may be an indication of a difficult transfer or infection. While some studies have demonstrated a significant decrease in the pregnancy and implantation rates in the presence of blood on the catheter [66, 67] or inside the catheter [68] after ET, others have failed to support the association between the presence of any type of contamination, whether macroscopic or microscopic, presence of blood or mucus and pregnancy outcome [69].

Retained Embryos

Immediate retransfer of embryos retained in the catheter following an initial transfer attempt in the absence of blood and mucus in the transfer catheter and other signs of a difficult transfer does not adversely influence the pregnancy outcome in terms of pregnancy, implantation, and delivery rates per embryo transfer [70, 71].

Recent Advances

Despite attempts to standardise the protocol of manually performed conventional embryo transfers, a comparative study that evaluated the injection speeds of simulated conventional embryo transfers by seven laboratory technicians and a pump-regulated embryo transfer (PRET) device demonstrated a large variation in injection speed in manually performed transfers, even after standardisation of the protocol. The recently developed automated PRET device generates a reliable and reproducible injection speed and therefore, brings new possibilities for further standardisation of the embryo transfer procedure. However, additional studies are needed to confirm if the observation mimics real clinical circumstances and if a standardised injection speed results in more exact positioning of the transferred embryos and therefore, higher pregnancy rates [72].

Conclusion

To maximise pregnancy outcomes with a single euploid embryo, we believe it is mandatory to ensure the atraumatic ultrasound-guided delivery of the embryo with a soft echogenic catheter, at a precise position in the endometrial cavity with a receptive endometrium, in a timely manner. The ET technique deserves dedicated attention owing to the number of parameters involved in ensuring a smooth and successful ET as discussed here. Should these factors be neglected, the reproductive outcome may be compromised. Hence, the ET technique must be preplanned to anticipate and avoid difficult transfers and those associated with a negative outcome. The clinician's knowledge of these factors and skill in performing ET is of paramount importance.

Conflict of Interest The authors declare no conflicts of interest.

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