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## Abstract title:

Impact of embryo replacement position (ERP) on euploid frozen embryo transfer (FET) outcomes

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### Study question:

Does the position of the euploid blastocyst, measured as distance from the fundus (DFF) of the uterine cavity (mm) affect the implantation potential?

### Summary answer:

There is a negative correlation between the DFF and the probability of pregnancy and implantation, while no effect was seen on the miscarriage rate.

### What is known already:

The optimal ERP of a euploid blastocyst is one of the last hurdles of an ART treatment. However, the ideal location within the uterine cavity is still being debated. While it has been claimed that the transfer site does not affect the implantation potential, most of the studies found that the highest pregnancy rates are obtained when the embryo is placed in the upper or middle area of the uterine cavity. Unfortunately, due to heterogeneity of parameters between different studies in which embryos with unknown ploidy state were transferred, objective assessment on the real effect of the ERP is difficult.

### Study design, size, duration:

This single center retrospective cohort study included a total of 455 single/double euploid FET cycles between March 2017 and November 2018. Trophoctoderm biopsy samples were subjected to Next Generation Sequencing to screen the ploidy state. Vitrification and warming were performed using the Cryotop method (Kitazato, Biopharma). The longitudinal distance between the fundal endometrial surface and the air bubble after transfer with a K-soft COOK catheter was measured and pregnancy, implantation and miscarriage rates were recorded.

### Participants/materials, setting, methods:

The following explanatory variables were analyzed: age, Anti Müllerian hormone (AMH), body mass index (BMI), endometrial thickness, quality of the trophoctoderm, difficulty of the transfer (requirement of additional instrumentation), presence of mucus or blood, day 5 or day 6 biopsy, single (SET) or double (DET) embryo transfer, cycle preparation (natural cycle - NC or hormone replacement therapy - HRT). The primary aim was to detect if the implantation potential is affected by the DFF.

### Main results and the role of chance:

The patients were on average 33.5 years old. The FET was performed in a NC (n=137) or HRT cycle (n=318). Of the 455 transfers, 325 (71.4%) resulted in a pregnancy and 262 (57.6%) in a clinical pregnancy leading to an implantation rate of 61.6%. Initially, we performed bivariate basic comparison tests between all explanatory variables and the pregnancy, implantation and miscarriage outcomes. When comparing the cycle preparation, number of embryos transferred and the quality of the embryos, the pregnancy and implantation rates were significantly higher in NC ( $p=0.0117$  and  $p=0.0077$ ), after DET ( $p=0.0049$ ; and  $p=0.0026$ ) and for high quality embryos ( $p=0.0039$  and  $p=0.0072$ ).

After performing a multivariate logistic regression analysis to consider the effect of all explanatory variables on the DFF, a negative effect between DFF and pregnancy ( $p=0.0084$ ) and implantation ( $p=0.0065$ ) was found. When all variables remained constant, the increase of one mm of DFF increases

the odds ratio of pregnancy by 0.902 and of implantation by 0.903, implying that the probability of pregnancy and implantation decreases as the fundus distance increases. No statistical significance of DFF was found for the miscarriage outcome ( $p=0.2166$ ), however, the presence of only 38 miscarriage cases is insufficient to make an adequate evaluation.

**Limitations, reasons for caution:**

Besides the retrospective design of the study, superiorly the full length of the cavity should have been measured to estimate the exact position within different uterine sizes. Also, a higher number of miscarriages is needed to find a possible effect of the distance from the fundus on this parameter.

**Wider implications of the findings:**

The depth of embryo replacement inside the uterine cavity may influence the pregnancy and implantation rates and should be considered as an important factor to improve the success of IVF cycles.

**Trial registration number:**

NA

**Keywords:**

Embryo Replacement Position

pgt-a

Pregnancy

implantation

Miscarriage