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CLINIQUE

DOES EGG VITRIFICATION DISPLACE THE FIRST POLAR BODY-MEIOTIC SPINDLE AXIS?

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OBJECTIVE

To assess whether the polar body – meiotic spindle axis is affected by vitrification.

DESIGN

A prospective observational study carried out in one centre of a donor egg bank programme where donor recruitment, oocyte vitrification and oocyte warming take place in multiple centres.

METHODS

Clinic members of the donor egg bank programme can either be donor processors and receivers of vitrified eggs or simply receivers only. Our clinic is a receiving centre so patients can select their donor and vitrified eggs are shipped from the processing centre. Vitrification, transport, storage and warming are all carried out according to strict egg bank protocols. To maintain quality control, regular checks are carried out on both vitrifying and warming laboratories and staff.

We received 9 donor egg lots from a total of 8 different clinics around the US. The nine cycles of egg warming were carried out between November 2014 and January 2015. At the time of ICSI, all oocytes were assessed for the presence and location of the meiotic spindle in relation to the first polar body using the polscope (Oosight, HTR). The degree of displacement from the first polar body was noted. Fertilisation check was performed 18-20 hours post ICSI and embryo cleavage was assessed a further 24 hours later. Embryo transfer was carried out on day 3 (3 cases) or on day 5 (4 cases) according to the number and quality of the embryos available. One embryo on day 3 or on day 5 was transferred in all cases except one where two day 3 embryos were transferred. Pregnancy was assessed with serum BhCG and clinical pregnancy confirmed by the presence of an intrauterine fetal heart on ultrasound at 7 weeks.

RESULTS

A total of 65 Mill eggs were warmed with a survival rate of 80%. These eggs were assessed at ICSI using a polscope and the meiotic spindle was seen in 84% of the eggs. The spindle was within 45° of the first polar body in 34% of cases and was more than 45° displaced in 50% of eggs. The fertilisation rate was 64% with 7 cases reaching embryo transfer and 5 of these obtaining a clinical pregnancy (71%).

CONCLUSIONS

The successful vitrification of oocytes has opened the possibility for both fertility preservation and donor egg banks without the need to coordinate the cycles of donor and recipient and to permit a greater selection of donor eggs for patients in need of them, in much the same way that donor sperm banks have provided such a service for many years.

The use of birefringence and the polscope was presented by Wang et al (2001) as a useful tool to assess the oocyte at the moment of microinjection. Multiple studies looking at various aspects of birefringence in the oocyte: both of the meiotic spindle and the zona pellucid have since been published. The importance of the length, birefringence value, position and shape of the spindle have all been promoted as oocyte quality selection tools.

In the literature, the spindle is visualised in approximately 80% of fresh oocytes (78%, Rama Ruju 2007) and this compares with the 80% visualisation rate that we saw in cryopreserved-warmed oocytes. Furthermore, the proportion of spindles from fresh oocytes which are within 45° of the first polar body have been reported as approximately 70% (68.8% Moon 2003, 74% Rienzi 2003).

In our study it was noted that the proportion of spindles within 45° of the first polar body was only 34% so half of what would be expected from previous studies on fresh oocytes. Displaced spindles were found to be up to 180° from where they should be located.

This raises the question as to why these eggs demonstrate displaced spindles or perhaps displaced polar bodies. It could be suggested that the method of cumulus cell removal prior to ICSI can have an effect on the displacement of the polar body particularly if a more aggressive

denudation method is employed, however in our study oocytes came from eight different centres all employing their own techniques for egg denudation and the displacement was seen across all supplying centres. In addition, the previous fresh oocyte studies all required cumulus cell removal prior to polscope assessment.

The process of oocyte vitrification causes the ooplasm to shrink and re-expand within the zona pellucida which could cause a mechanical displacement of the polar body as the egg is exposed to cryoprotectants and likewise during warming as cryoprotectants are withdrawn.

It has previously been shown that high doses of FSH can affect the spindle (Madaschi 2009) and it is therefore a possibility that strong ovarian stimulation of egg donors attempting to obtain a maximum of oocytes could have an impact. Unfortunately in these cases, spindle assessment prior to vitrification was not possible. Further studies to eliminate these variables are necessary to confirm that the process of vitrification itself can impact on the placement of the meiotic spindle post warming and therefore the use of a polscope when performing ICSI on these eggs would be highly recommended.

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