Mouse ovarian follice cryopreservation using vitrification or slow programmed cooling: assessment of in vitro development, maturation, ultra-structure and meiotic spindle organization

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Abstract:

Aim: To compare different outcomes of vitrification and slow freezing of isolated pre-antral follicles and to evaluate different cryo-devices vitrification of isolated follicles. Methods: pre-antral follicles were isolated from mouse ovaries and cryopreserved using vitrification and slow freezing. A preliminary was carried out to select the optimal cryo-devices vitrification of isolated follicles. A total of 414 follicles were randomly distributed among four groups: control (CT) fresh (n=100) , nylon mesh (n=96) , electron microscopy grid (n=120) , and micro-capillary tips (n=116) . Subsequetly, a total of 979 follicles were randomly assigned to three different group: CT fresh (n=256), vitrification (n=399) and Slow freezing (n=324). CT and cryopreserved/thawed follicles were cultured in vitro and examined daily for development . final maturation was triggered with human chorionic gonadotrophin and rates of oocyte maturation were calculated. The Ultra-structure of cryopreserved / thwed follicles was studied using electron microscopy. Meiotic spindle presence and organization in mature oocytes were examined using the oosight imaging system. Result: Micro-capillary tips resulted in poor immediate post-warming survival but no differences were observed in the subsequent in vitro development characteristics between different cryo-devices. Nylon mesh proved to be the easiest carrier, particularly when large numbers of follicles at the end of the culture period (P

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