## Morphological aspects of human aged oocytes: an in vivo and in vitro ultrastructural study

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Maternal age significantly affects the outcome of ART by lowering oocyte quality and competence. However, the morphological ultrastructure features of aged human oocytes have been not extensively studied and understood. In this study we analyzed oocytes donated by women <35 (-35) and ? 35 (+35) years old, enrolled in an IVF program, after informed consent. MII oocytes, fixed at time of pick up (in vivo) or after 24 hours culture (in vitro) were evaluated by light and transmission electron microscopy.

IN VIVO samples of both -35 and +35 showed a spherical oocyte surrounded by regular ZP and expanded cumulus, and an ooplasm rich in organelles provided with a homogenous matrix. Oocyte mitochondria-SER aggregates (M-SER), that were numerous in -35, appeared significantly reduced in number/size and partially replaced by mitochondria-vesicle complexes (MVC) in +35. Cortical granules, normally represented in -35, appeared abnormally distributed in +35. Ooplasm microvilli significantly decreased in density and shortened in +35.

IN VITRO samples showed several changes. All samples showed a compact cumulus with occasional cumulus cell-oocyte contacts. The inner ZP showed an increased density in +35. Small sized M-SER were occasionally detected in -35 and were almost absent in +35. MVC were present in both -35 and +35, increased up in number and often showed abnormal vesicles in +35. All samples showed numerous lysosomal structures and reduction of cortical granules. Ooplasm microvilli significantly decreased in density and shortened in +35.

This study demonstrated several significant ultrastructural changes occurring in fresh oocytes (in vivo) from older patients and in all 24 hours in vitro cultured oocytes. The altered ultrastructure observed in in vitro cultured oocytes resembles in vivo aging-related changes and it is significantly more evident in oocytes from older patients. The changes described in the present study may be considered as ultrastructural markers of oocyte aging.

Keywords: aging, oocyte, ultrastructure

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