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Mitochondrial oxygen consumption rates skyrocket at the blastulation in spite of constant mitochondrial DNA number during preimplantation development.

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Study question:

How do the mitochondrial DNA (mtDNA) copy number and mitochondrial function change in human embryos during preimplantation development?

Summary answer:

Mitochondrial oxygen consumption rates (mtOCRs) rise sharply at the blastulation in spite of constant mtDNA number during preimplantation development. In addition, mitochondria became mature and the CCO activity was detected at blastocyst stage.

What is known already:

OCRs relate to the quantity of ATP production and mitochondrial respiration may be an effective index of mammalian embryo quality. The number of mtDNA has been shown to be a marker of mitochondria number. The number of mtDNA remains constant within preimplantation murine embryos, but not in cattle and pigs decreasing sharply from 2-cell to 4/8-cell stages. Moreover, the changes of mtOCRs of human embryos remain unknown.

Study design, size, duration:

This experimental study was performed after obtaining informed consent of patients and an approval of ethical committee of JSOBGY using 21 MII oocytes, 12 day 3 embryos, 20 day 4 embryos and 20 blastocysts. We assessed the relationship among embryo developmental stage, their mtDNA number, and their mtOCRs.

Participants/materials, setting, methods:

The mtOCRs and the mtDNA copy number of each sample were measured simultaneously using scanning electrochemical microscopy combined with a mito-toxin (cyanide) and by real time PCR, respectively. Morphological changes and cytochrome c oxidase (CCO) activities of mitochondria were assessed using 8 eggs. Data were compared using Tukey–Kramer method.

Main results and the role of chance:

The copy number of mtDNA kept constantly from mature oocytes to blastocyst stage (155,702-222,511 copies). The mtOCRs didn't change from mature oocytes to day 4 embryos (0.5-1.5 fmol/sec). However, the mtOCR in blastocyst (4.3) was higher ($P < 0.05$) than others. Round- or oval-shaped mitochondria transformed into elongated tubular forms, developing well-defined transverse cristae structure toward blastocyst stage at transmission electronic microscopy (TEM) observation. The mitochondrial CCO activity was detected at blastocysts stage based on TEM analysis, not but mature oocytes and cleavage embryos.

Limitations, reasons for caution:

Our data showed the number of mtDNA is constant during preimplantation development. This result is similar to those obtained in murine studies, but not bovine and porcine studies. Further studies need to clarify the species barrier and the relationship between the mtOCRs and mtDNA copy number in mammalian embryos.

Wider implications of the findings:

Data of the present study showed mtOCRs rise sharply at the blastulation, that mitochondria became mature and that the CCO activity was detected at blastocyst stage. Blastulation requires more ATP depending on Na/K-ATPase. Thus, it is reasonable that mitochondria become mature and increase their OCRs at the timing of blastulation. This study provided new insights on the implications of a behavior of mtDNA copy number and mitochondrial function during human embryo development.

Key words: embryo development, mitochondria, mitochondrial DNA, mitochondrial function