

Mitochondria transfer

Chii-Ruey Tzeng

Taipei Medical University Hospital, Taipei, Taiwan

The mitochondria DNA (mtDNA) is not protected by histones and is constantly exposed to a high level of reactive oxygen species (ROS) and free radicals in the matrix of mitochondria. The current study discloses that mtDNA 4977 deletions in compromised human oocytes may range from 32.8% (Brenner et al., 1998) to 49% (Chen et al., 1995). Furthermore, any mtDNA rearrangement is even higher in 50.5% of oocytes (Barrett et al., 2000).

Although a live birth has been achieved following transfer of anucleate donor oocyte cytoplasm into recipient eggs (Cohen et al., 1997), mtDNA inheritance and mtDNA heteroplasmy have been found in the fetal cord blood and placental tissues. However, microinjection of purified mitochondria from granulosa cells could suppress the progression of apoptosis in cultured mouse oocyte by 50% (Perez et al., 2000) indicating the "rescue effect" of cGCs mitochondria for compromised mouse oocyte developed in vitro.

Patients age over 38, recurrent implantation failure, prolonged unexplained infertility, poor fertilization rate, or compromised embryo quality of the previous cycle were subjected to ICSI and mitochondria transfer (MIT) in an IVF laboratory of a university hospital.

Preparation of mitochondria from cumulus granulosa cells (cGCs): mitochondria were prepared by differential centrifugation. The cGCs were trimmed from the oocytes, collected and homogenized with a glass/Teflon at 10^6 cells per ml buffer. The crude homogenate was centrifuged twice for 15 min at 800x g and the pellets were discarded. The supernatant was again centrifuged for 20 min at 9800 xg. The pellet was resuspended very slowly by dropwise addition of 0.5 ul of 150 mM KCl. The mitochondria preparations were maintained at 4°C. Then the mitochondria suspension along with a single sperm was injected into the MII oocyte. The number of mitochondria transferred per oocyte was 3000-5000 during the ICSI procedure.

By the mitochondria transfer (MIT) technique, we were able to achieve clinical pregnancies. Mixing the population of donor and recipient ooplasm has caused extensive debate. By using the transfer of mitochondria from cumulus granulosa cells into oocyte, it can avoid heteroplasmy of donor and recipient mtDNA. This new and evolving treatment may offer enormous potential and the limitless possibilities of rescuing compromised oocyte, normalization of embryo growth and retention of patient's mtDNA fingerprint.

Key words: cumulus granulosa cell / ICSI / mtDNA / mitochondria transfer / pregnancy.