Ubiquitinated sperm mitochondria, selective proteolysis, and the regulation of mitochondrial inheritance in ammalian embryos

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Abstract

The strictly maternal inheritance of mitochondria and mitochondrial DNA (mtDNA) in mammals is a developmental paradox promoted by an unknown mechanism responsible for the destruction of the sperm mitochondria shortly after fertilization. We have recently reported that the sperm mitochondria are ubiquitinated inside the oocyte cytoplasm and later subjected to proteolysis during preimplantation development (P. Sutovsky et al., Nature 1999; 402:371-372). Here, we provide further evidence for this process by showing that the proteolytic destruction of bull sperm mitochondria inside cow egg cytoplasm depends upon the activity of the universal proteolytic marker, ubiquitin, and the lysosomal apparatus of the egg. Binding of ubiquitin to sperm mitochondria was visualized by monospecific antibodies throughout pronuclear development and during the first embryonic divisions. The recognition and disposal of the ubiquitinated sperm mitochondria was prevented by the microinjection of anti-ubiquitin antibodies and by the treatment of the fertilized zygotes with lysosomotropic agent ammonium chloride. The postfecundal ubiquitination of sperm mitochondria and their destruction was not seen in the hybrid embryos created using cow eggs and sperm of wild cattle, gaur, thus supporting the hypothesis that sperm mitochondrion destruction is species specific. The initial ligation of ubiquitin molecules to sperm mitochondrial membrane proteins, one of which could be prohibitin, occurs during spermatogenesis. Even though the ubiquitin cross-reactivity was transiently lost from the sperm mitochondria during epididymal passage, likely as a result of disulfide bond cross-linking, it was restored and amplified after fertilization. Ubiquitination therefore may represent a mechanism for the elimination of paternal mitochondria during fertilization. Our data have important implications for anthropology, treatment of mitochondrial disorders, and for the new methods of assisted procreation, such as cloning, oocyte cytoplasm donation, and intracytoplasmic sperm injection.