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The central role of centrosomes in translocation of mitochondria and centrosome proteins

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Centrosomes are central cell bodies that are predominantly known for their role as microtubule organizing centers. However, by nucleating and organizing microtubules into specific patterns, centrosomes play also a crucial role in directing the translocation of cell organelles, vesicles, and proteins to their functional destinations. Here we review translocation of mitochondria and the nuclear mitotic apparatus protein (NuMA) in somatic cells and during oocyte maturation, parthenogenetic activation, fertilization, nuclear cloning, and development in porcine oocytes.

Our studies have shown that a temporal, spatial, and developmental relationship exists between microtubule organization and the progressive translocation of mitochondria and NuMA allowing dynamic and precisely coordinated cell and molecular interactions. During normal fertilization in most species centrosomes are crucial for the organization of the sperm aster that unites maternal and paternal genomes during fertilization and for the equal distribution of DNA and cell organelles during cell division, cell differentiation, and development [1,2,3,4,5,6]. Mitochondria play a significant role in cellular metabolism by providing energy in form of ATP that is required for specific cell cycle and developmental events. Mitochondria distribution is important for embryo development and abnormal mitochondria distribution has been associated with developmental abnormalities [7,8].

We have revealed the distribution of active mitochondria during pig oocyte maturation, fertilization and early embryo development *in vitro* by using MitoTracker Green staining and confocal laser scanning microscopy. Mitochondria accumulated in the perinuclear area during meiotic progression from germinal vesicle breakdown (GVBD) through anaphase I and we found differences for *in vitro* versus *in vivo* matured oocytes which may correspond to the differences in developmental competence that is lower in *in vitro* matured oocytes [6]. The absence of mitochondria accumulation in the perinuclear region coincided with reduced development. In fertilized eggs, active mitochondria were aggregated in the pronuclear region. Active mitochondria were distributed in both inner cell mass cells and trophectoderm cells of the blastocysts. These studies suggest that in the pig mitochondria accumulate in the perinuclear area from GV stage to anaphase I stage and that mitochondrial translocation in oocytes matured *in vitro* is not as complete as in oocytes matured *in vivo*.

Our studies on NuMA have shown a tight association between NuMA and microtubules during the early stages of mitosis and with centrosomes in anaphase and telophase in somatic cells [9]. During interphase, NuMA plays significant roles in DNA replication and displays

punctate nuclear staining patterns in most cells. To become a functional mitotic protein NuMA moves from the nucleus into the cytoplasm where it associates with microtubules and becomes translocated along microtubules to its functional destination, the centrosomes. During fertilization NuMA is not associated with sperm but it is localized to the meiotic apparatus in matured oocytes. The distribution of NuMA in parthenogenetic pig embryos, in *in vitro* fertilized embryos and in porcine nuclear transfer embryos was determined. Our results revealed that NuMA was localized only in a few of the parthenogenetically activated oocytes while it was detected within the nucleus in most of *in vitro* fertilized and reconstructed oocytes at 25 hours of fertilization and nuclear cloning, respectively.

Taken together, our studies show the importance of centrosomes in directing cell organelles and centrosome proteins such as mitochondria and NuMA, respectively.

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