

## Functional Analyses of Centrosomal Proteins, *Nek2* and *NuMA* in Development of Mouse Gametes and Early Embryos

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*Nek2* (NIMA-related protein) is a mammalian cell cycle-regulated kinase that involves in chromosome condensation and centrosome regulation and *NuMA* (nuclear mitotic apparatus protein) is involved in spindle assembly during a cell cycle. The cellular distribution and organization of the centrosomal components is completely unknown during fertilization and embryonic development. We examined distribution of two well-known centrosomal proteins, *Nek2* and *NuMA* in mouse gametes and embryos to get an insight in the reorganization of centrosomal proteins during germ cell development and early fertilization. Spermatogenic cells, gametes, and embryos were analyzed with anti-*Nek2* or -*NuMA* antibodies by immunological assay, RT-PCR, and overexpression through gene transfection. Mitotically or meiotically active spermatogenic cells were intensively stained with these antibodies in both centrosomes and cytoplasm, whereas the oocytes showed different staining patterns depending on the meiotic stages. During maturation, GV, GVBD, and MI stage were clearly stained with *NuMA* antibody in the nucleus or cytoplasm at MII. Also, *Nek2* was detectable in cytoplasm as scattered spots or chromosome associated at MII. In early developmental embryo, *NuMA* was detected in nucleus of each blastomere, while *Nek2* was detected in cytoplasm. In contrast to previously reported results, *Nek2* and *NuMA* were detected in both decondensing head, and the centriole of demembrated and decondensed sperm or whole body of trypsin-treated sperm for *Nek2*. During meiotic progress in oocytes, transcripts levels were the highest in MI stage and then downregulated in MII. Also, it shows dramatically change in early developmental embryos, firstly, it was increased until 4 cell stage and reduced in 8 cell stage, and finally, transcript levels were upregulated until blastocyst. This finding suggests that centrosomal component may play an important role in reorganizing of functional centrosome during fertilization process and subsequent development.

Key words) *Mouse, Nek2, NuMA, Spermatogenesis, Fertilization*