

P-7 A MAPK Pathway is Involved in the Control of Cortical Granule Reaction and Mitosis During Bovine Fertilization

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Objectives: In order to understand the mechanism by which MAPK regulates fertilization, we examined the effect of the MAPK pathway inhibitor U0126 on polyspermy, cortical granule reaction and mitosis in bovine oocytes during and after fertilization.

Methods: Exp.I) In order to determine the role of MAPK prior to fertilization, oocytes that had matured in vitro for 22~24 hrs were incubated with 30 μ M U0126 for 30 min, and then inseminated with spermatozoa for 18 hrs. Exp.II) Fifteen hours after insemination, the presumptive fertilized oocytes were cultured for an additional 12 hrs in fertilization media containing 30 μ M U0126.

Results: Western blotting with antibodies that detect active, phosphorylated MAPK revealed that MAPK activity was decreased in U0126 treated oocytes. Oocytes that were treated with U0126 before insemination displayed a significantly higher incidence of polyspermic penetration and incomplete cortical granule reaction than that observed in untreated oocytes ($p < 0.05$). Exposure of oocytes to 30 μ M U0126 15 to 27 hrs after insemination induced aberrant microtubule assembly and cell division, often resulting in the formation of two or three daughter cells with altered shapes and sizes.

Conclusion: These results suggest that an ERK-like cascade is part of a mechanism that controls cortical granule reaction and the formation of the mitotic spindle following sperm penetration in the bovine.