MECHANISMS OF MITOGEN-ACTIVATED PROTEIN KINASE 3/1 ACTIVATION IN PIG CUMULUS-OOCYTE COMPLEXES MATURED IN VITRO

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Abstract Body

In vitro maturation (IVM) of mammalian oocytes can be stimulated by gonadotropin hormones (FSH, LH) or their intrafollicular mediators, epidermal growth factor (EGF)-like peptides (amphiregulin epiregulin, betacellulin). Signaling pathways stimulated in cumulus cells by FSH and amphiregulin converge in activation of mitogen-activated protein kinase 3/1 (MAPK3/1), also known as Erk1/2. This kinases have a central role in the control of meiosis resumption and cumulus cell expansion by regulating protein modifications and expression of various genes involved in gap junction permeability, synthesis of extracellular matrix and steroidogenesis. We found that FSH induces in pig cumulus-oocyte complexes (COCs) expression and synthesis of amphiregulin and epiregulin that serve as ligands for EGF receptor. The ligand binding results in the receptor autophosphorylation and activation of MAPK3/1 signaling pathway. However, we also found that FSH causes a minute activation of MAPK3/1 in pig cumulus cells, which cannot be explained by the *de novo* synthesis of the EGF receptor ligands. The rapid MAPK3/1 activation required EGF receptor activity, was sensitive to SRC-family kinases inhibitor and protein kinase C inhibitor (PP2 and calphostin C, respectively) and was resistant to inhibitors of protein kinase A (H89) and metalloproteinases (galardin, TAPI-2). We conclude that FSH causes in cultured pig COCs rapid activation of MAPK3/1 by a mechanism involving ligand-independent transactivation of the EGF receptor. The MAPK3/1 activity is maintained in cumulus cells for extended period of time through activation of EGF receptors by *de novo* synthesized ligands.

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