Antigonadotropic action of adenosine triphosphate in human granulosa-luteal cells: involvemnet of protein kinase Calpha

戴承杰

Tai CJ;Kang SK;Choi KC;Tzeng CR;Leung PC

摘要

Abstract

The presence of P2U purinoceptor in human granulosa-luteal cells (hGLCs) indicates a potential role of ATP in regulating ovarian function. In this study an inhibitory effect of ATP on hCG-induced cAMP production was observed. Extracellular ATP has been shown to activate protein kinase C (PKC) after binding to a purinoceptor. To understand the role of PKC in mediating ATP action, hCG-stimulated cAMP level was examined in the presence of the PKC activator, 1 µmol/L phorbol 12-myristate 13-acetate (PMA), or the PKC inhibitor, 1 µmol/L staurosporin or 1 µmol/L bisindolylmaleimide I. PMA, like 10 µmol/L ATP, significantly reduced hCG-evoked cAMP production. In addition, the inhibitory effect of ATP was reversed by staurosporin and bisindolylmaleimide I. To further investigate the involvement of PKC isoforms in mediating the inhibitory effect of ATP, the presence of PKC isoforms in cultured hGLCs was examined by Western blot using monoclonal antibodies against specific isoforms. Translocation of PKC isoforms from cytosolic fraction to membrane fraction was studied to identify the active PKC isozymes subsequent to ATP treatment. The change in PKC isoform in PKC-depleted cells (achieved by exposure to PMA for 18 h) was also examined. Our results demonstrated the presence of PKC, -, -, and - isoforms in hGLCs and the translocation of PKC subsequent to ATP treatment. In PKC-depleted cells the PKC level was reduced, and no significant effect of ATP on hCG-stimulated cAMP production was observed. To our knowledge, this is the first demonstration of PKC isoforms in hGLCs and the involvement of activated PKC in mediating the antigonadotropic effect of extracellular ATP. Taken together, these results further support a role of this neurotransmitter in regulating human ovarian function.

.