Z502 Effect of Lysophosphatidic Acid on Proliferation and Differentiation of Rat Skeletal Myoblasts in Culture

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Lysophosphatidic acid (LPA; 1-acyl-glycerol-3-phosphate) has been known as an intercellular phospholipid messenger with a wide range of biological activities. In this study, the effect of LPA in both the proliferation and differentiation of rat E63 myoblasts has been investigated. In the serum-free Insulin-Transferrin-Selenium (ITS) media, the proliferation of E63 cells was largely restricted. Addition of LPA into the ITS media strongly promoted the cell proliferation and resulted in two to four fold increase of cell number. Furthermore, it appeared to increase the percent fusion in a dose-dependent manner up to 15 µg/ml. The synthesis of myosin heavy chain (MHC) was increased by LPA as well. These results indicated that LPA is able to promote both cell proliferation and differentiation in rat E63 myoblasts. Suramin, known to have a uncoupling activity of growth factor-receptor interaction, was tested for antagonistic activity in myoblast proliferation and differentiation. Myoblasts grown in the ITS medium containing LPA were able to proliferate well even in the presence high concentration of suramin, whereas myoblast differentiation was completely blocked by 30 μg/ml of suramin. The inhibitory effect of suramin on the myoblast differentiation was completely reversible by removing the suramin. This result indicates that the intracellular signaling pathway of LPA leading to cell proliferation might be distinct from that leading to cell differentiation on E63 myoblasts. Also the antagonistic effect of suramin suggests that the differentiation activity elicited by LPA might be mediated by a specific G protein-coupled receptor.

Purification and Characterization Allatostatin and Allatostatin-Producing Neurons in the Brain from *Bombxy mori*

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Allatostatin inhibiting juvenile hormone biosynthesis in corpora allata was purified and characterized from brain and retrocerebral complexes of the silk worm *Bombxy mori*. The antiserum in this investigation used analyzed the number and localization pattern of allatostatin-producing neurons in brain and retrocerebral complex. There are two types of allatostatic neuropeptides I and II. Both of them show a highly conserved C-terminal sequence (-Tyr-Xaa-Phe-Gly-Leu-NH₂) with allatostatin from cockroach, blowfly and other moths. Using anti-allatostatin antiserum several pairs of allatostatin-immunoreactive neurons were found in both sides of protocerebrum. Axons of the neurons showing strong immunoreactivity project out to the nervi of corpora cardiaci I ipsilaterally. Allatostatin-immunoreactive axons bypass the corpora cardiaca (CC) and then terminate in the corpora allata (CA), strongly indicating that they may be directly involved in inhibition of juvenile hormone biosynthesis in the CA.