J. Pharmacol. and Exp. Ther. **291**, 171-180 (1999)

Stimulation by transforming growth factor-α of DNA synthesis and proliferation of adult rat hepatocytes in primary cultures: Modulation by α- and β-adrenoceptor agonists

Mitsutoshi Kimura (木村 光利), and Masahiko Hagihara (荻原 政彦)

We investigated the effects of transforming growth factor-α (TGF-α) on DNA synthesis and proliferation in primary cultures of adult rat hepatocytes and examined the influence of α- and β-adrenoceptor agonists on the TGF-α-induced responses. TGF-α (1.0 ng/ml) produced a 4.1-fold elevation of DNA synthesis during 3 h of culture and a 1.2-fold increase in the nucleus number (proliferation) during 4 h of culture at a cell density of 3.3×10^4 cells/cm^2. The TGF-α-induced hepatocyte DNA synthesis and proliferation were dose-dependent at EC_{50} values of 0.36 ng/ml and 0.45 ng/ml, respectively. Hepatocyte DNA synthesis and proliferation induced by 1.0 ng/ml TGF-α did not reduce even at higher initial plating densities (5.0×10^4 and 1.0×10^5 cells/cm^2). Increasing concentrations of a β_2-adrenoceptor agonist, metaproterenol (10^{-7} - 10^{-6} M), markedly reduced the proliferative effects of TGF-α, while those of an α_2-adrenoceptor agonist, UK-14304 (10^{-6} -10^{-5} M), and an α_1-adrenoceptor agonist, phenylephrine (10^{-7} -10^{-6} M), significantly potentiated the TGF-α action. The proliferative effects of TGF-α (1.0 ng/ml) were not affected significantly by a monoclonal anti-epidermal growth factor receptor antibody (1-100 ng/ml) and were almost completely blocked by specific inhibitors of signal transducers, such as genistein (10^{-5} M), U-73122 (10^{-5} M), wortmannin (5×10^{-7} M), sphingosine (5×10^{-6} M), PD98059 (5×10^{-5} M), and rapamycin (10 ng/ml). These results suggest that among the elements that link signals of cell surface receptor to the nucleus, the proliferative action of TGF-α is mediated, at least, by tyrosine kinase, phospholipase C, PI (3) kinase, protein kinase C, MAP kinase kinase, and p70 S6K.