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Prostaglandin E₂ (EP₁) receptor agonist-induced DNA synthesis and proliferation in primary cultures of adult rat hepatocytes: The involvement of transforming growth factor- β .

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We studied the effects of EP receptor subtype agonists on DNA synthesis and proliferation in primary cultures of adult rat hepatocytes to elucidate their mechanisms of action. Maintained in short-term cultures (i.e., 3.5 h) in a serum-free, defined medium, hepatocyte parenchymal cells underwent DNA synthesis and proliferation in the presence of sulprostone (10^{-6} M), prostaglandin E₂ (PGE₂; 10^{-6} M), and 17-phenyl-trinor-prostaglandin E₂ (17-pt-PGE₂; 10^{-9} M) in a time- and dose-dependent manner. PGE₂ was less potent than 17-pt-PGE₂ in stimulating hepatocyte mitogenesis. Sulprostone (10^{-6} M) and 11-deoxy-PGE₁ (10^{-6} M) showed weak and no significant stimulation, respectively, for hepatocyte mitogenesis. These effects of PGE₂, 17-pt-PGE₂, and sulprostone were abolished by treatment with a specific EP₁ receptor antagonist, SC-51322 or phospholipase C (PLC) inhibitor, U-73122. The effects of these EP₁ receptor agonists were potentiated by ionomycin, and blocked by verapamil. Hepatocyte mitogenesis was almost completely blocked by specific inhibitors of growth-related signal transducers, such as genistein, wortmannin, PD98059, and rapamycin. A monoclonal antibody against transforming growth factor- β (TGF- β) dose-dependently inhibited the PGE₂- and 17-pt-PGE₂-induced hepatocyte mitogenesis. Hepatocyte levels of TGF- β in response to the EP₁ receptor agonists was decreased from 550 pg/mg protein to 100 pg/mg protein within 10 min. The reduction in cellular TGF- β levels was blocked by SC-51322, U-73122, somatostatin, and verapamil, and potentiated by ionomycin. The specific inhibitors of growth-related signal transducers did not affect the hepatocyte TGF- β levels induced by the EP₁ receptor agonists. These results

suggest that the proliferative mechanisms of action of EP₁ receptor agonists is mediated through an increase in the autocrine secretion of TGF- β , which is dependent on the EP₁ receptor/Gq/PLC/Ca²⁺ system. The locally secreted TGF- β , in turn, acts as a complete mitogen rather than incomplete for the primary cultured hepatocytes.