

论著

阿托品对人视网膜色素上皮细胞D407株表达及分泌TGF β 2的调控

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摘要:

目的: 通过观察不同浓度阿托品联合10-5 mol/L卡巴胆碱干预下D407细胞表达及分泌TGF β 2的变化, 探讨阿托品对RPE细胞表达及分泌TGF β 2的调控作用。方法: 常规培养D407细胞, 药物干预前换用无血清培养基培养, 分为4组。(1) 实验组(A组): A1~A5组依次加入10-4~10-8 mol/L阿托品, 孵育30 min后每组加入10-5 mol/L卡巴胆碱;(2) 阴性对照组(B组): B1~B5组依次加入10-4~10-8 mol/L阿托品;(3) 阳性对照组(C组): 加入10-5 mol/L卡巴胆碱;(4) 空白对照组(D组): 不加药物。干预24 h后采用RT-PCR, Western印迹及ELISA法检测细胞胞浆中TGF β 2 mRNA及蛋白质的表达水平及上清液中TGF β 2的含量。统计学方法采用单因素方差分析。结果: 实验组D407细胞胞浆TGF β 2 mRNA和蛋白质表达水平及上清中TGF β 2蛋白质含量均较阳性对照组低, 10-4 mol/L阿托品可完全阻断10-5 mol/L卡巴胆碱上调TGF β 2表达及分泌的作用, 其效应具有浓度依赖性($F=1\ 056.897, 1\ 320.170, 475.657; P<0.001$)。阴性对照组D407细胞胞浆TGF β 2 mRNA和蛋白质表达水平及上清中TGF β 2蛋白质含量与空白对照组比较差异无统计学意义($P>0.05$)。结论: 阿托品可有效抑制卡巴胆碱促进人RPE细胞表达及分泌TGF β 2的功能, 提示M受体参与介导此过程。

关键词: 视网膜色素上皮 转化生长因子 β 2 卡巴胆碱 阿托品

Expression and secretion of TGF β 2 in human retinal pigment epithelium cell line D407 regulated by atropine

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Abstract:

Objective To investigate the regulation of atropine to the expression and secretion of TGF β 2 in retinal pigment epithelium (RPE) cells by observing the changes of those under different treatments of atropine and carbachol. Methods D407 cells were cultured conventionally and divided into 4 groups as follows: (1) An experimental group (Group A), cells were pretreated with 10-4 10-8 mol/L atropine for 30 min, and then treated with 10-5 mol/L carbachol; (2) a negative control group (Group B), cells were treated with 10-4 10-8 mol/L atropine; (3) a positive control group (Group C), cells were treated with 10-5 mol/L carbachol; (4) a blank control group (Group D). The concentration of TGF β 2 in the supernate, and the level of TGF β 2 mRNA and protein were measured by ELISA, RT-PCR, and Western blot after the 24 hour treatment. The data were analyzed by analysis of variance. Results The levels of TGF β 2 mRNA and protein in the cytoplasm and the concentration of TGF β 2 in the supernate in the experimental groups were lower than those of the positive control group. Atropine at 10-4 mol/L could completely inhibit the effect of carbachol at 10-5 mol/L. The effect of atropine was concentration dependent ($F=1\ 056.897, 1\ 320.170, and 475.657; P<0.001$). There was no change of TGF β 2 level in the cytoplasm and supernate with the treatment of atropine alone ($P>0.05$). Conclusion Carbachol can promote the expression and secretion of TGF β 2 in human RPE cells and atropine could reverse it effectively, suggesting that M receptor may be involved.

Keywords: retina pigment epithelium; transforming growth factor β 2; carbachol; atropine

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