

si RNA阻断Stat3信号促进食管鳞癌细胞株凋亡

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Blocking Stat3 Signal by RNAi Promotes Cell Apoptosis in Esophageal Squamous Cell Carcinoma

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摘要 目的: 研究RNA干扰技术沉默Stat3基因对食管鳞癌细胞 (EC9706) 中持续性激活Stat3信号的阻断作用及对细胞凋亡的影响。方法: 将化学合成的100nM的Stat3 siRNA转染EC9706细胞, RT-PCR检测转染前后Stat3 mRNA的表达, Western blot检测转染前后细胞中Bcl-2、Stat3及磷酸化Stat3 (p-Stat3) 蛋白的表达, 凝胶电泳迁移率 (EMSA) 检测转染前后活化Stat3蛋白的DNA结合活性, 流式细胞术检测转染前后细胞凋亡的改变情况。结果: Stat3 siRNA转染细胞48h后细胞形态发生明显的改变, RT-PCR及Western blot结果显示Stat3 siRNA干扰后Stat3 mRNA及蛋白表达均受到明显抑制, Stat3信号的激活被阻断, 活化Stat3蛋白的核结合活性显著下降。转染后细胞中Bcl-2蛋白的表达明显减少。流式细胞术结果证明Stat3 siRNA可明显促进细胞凋亡。结论: Stat3 siRNA能够特异地阻断食管鳞癌细胞中Stat3 信号的持续性激活并促进肿瘤细胞凋亡。

关键词: 食管鳞癌 信号转导子及转录活化子3 RNA干扰 凋亡

Abstract: Objective: To investigate the blockage of constitutively activated signal transducers and activators of transcription(Stat3) signal and the effect on cell apoptosis by applying RNA interfere technique to silence Stat3 gene in human esophageal squamous cell carcinoma cell line(EC9706). Methods: EC9706 was transfected with chemical syntheswased Stat3 siRNA (100nM), RT-PCR and Western blotting were used to detect the expression of Stat3 mRNA, the protein levels of Bcl-2,Stat3 and phosphorylated-Stat3(p-Stat3) before and after transfection. The change of DNA-binding activity and the cells apoptosis were evaluated by electrophoretic mobility gel shift assay (EMSA) and FCM, respectively. Results: After transfected Stat3 siRNA 48h, cells morphology were significantly transformed.RT-PCR and Western blot showed the expression of Stat3 gene was inhibited at mRNA and protein levels by Stat3 SiRNA and the constitutively activated Stat3 signal was blocked. Stat3-DNA-binding activity was supressedd directly at DNA level comfirmed by EMSA. The expression of Bcl-2 protein was reduced after transfected by Stat3 siRNA.FCM indicated cell that apoptosis was promoted significantly in EC9706. Conclusion: Stat3 siRNA blocked the constitutively activated Stat3 signaling specially and efficiently in EC9706, which promoted cell apoptosis obviously.

Key words: Esophageal squamous cell carcinoma Stat3 RNA interference Apoptosi

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