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392~396.siRNA沉默livin的表达对人结肠癌HT-29细胞增殖、凋亡及侵袭的影响[J].何文静,黎军和,赵清梅,熊建萍,彭小东.中国肿瘤生物治疗杂志,2012,19(4)

siRNA沉默livin的表达对人结肠癌HT-29细胞增殖、凋亡及侵袭的影响 点此下载全文

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基金项目: 国家自然科学基金资助项目(No. 30960440)

DOI: 10.3872/j.issn.1007-385X.2012.4.009

摘要:

目的: 探讨小干扰RNA(small interference RNA,siRNA)沉默人结肠癌HT-29细胞livin表达对HT-29细胞增殖、凋亡和侵袭的影响。 方法: 合成靶向livin 的双链siRNA(livin-siRNA),转染HT-29细胞,RT-PCR及Western blotting检测HT-29细胞中livin mRNA及蛋白的表达,MTT实验检测HT-29细胞的增殖,流式细胞术检测HT-29细胞周期分布及凋亡,细胞侵袭实验检测HT-29细胞侵袭性的变化,caspase-3活性检测试剂盒检测caspase-3活性的变化。 结果,Livin-siRNA转染后48 h,与空白组、阴性对照组及脂质体组相比,livin-siRNA转染组HT-29细胞中livin mRNA水平明显下降(0.073±0.007 vs 0.395±0.082、0.423 ±0.025、0.418±0.032, P <0.05),其蛋白表达也明显下调(0.106±0.003 vs 0.456±0.065、0.473±0 078、 0 491±0.045, P < 0.05)。转染96 h后,livin-siRNA组HT-29细胞增殖能力明显低于对照组及脂质体组(0.564±0 102 vs 0 833±0.127、0.860±0.153, P <0.0 5),且细胞凋亡率升高\[(16.5±2.8)% vs (2.4 ±0.5) %、(3.7±1.0) %, P <0.05\]。侵袭实验显示,livin-siRNA转染后,穿过Matrigel膜的HT-29细胞数量明显少于对照组及脂质体组\[(31.3±4.5) vs (101.3±8.6)、(97.4±7.8)个, P <0.05) \]。livin-siRNA组HT-29细胞的caspase-3活性低于对照组(0.160 ±0.023 vs 0.347±0 058, P <0.05) 。结论:siRNA沉默livin的表达可抑制HT-29细胞的增殖,诱导细胞凋亡,抑制细胞的侵袭。

关键词: 结肠癌 livin 小干扰RNA 基因治疗

Effects of siRNA silencing livin expression on proliferation, apoptosis and invasion of human colon cancer cell line HT-29 Download Fulltext

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Fund Project: Project supported by the National Natural Science Foundation of China (No. 30960440)

Abstract:

Objective: To explore the effects of small interference RNA (siRNA) targeting livin on the proliferation, apoptosis and invasion of human colon cancer cell line HT-29. Methods: Chemically synthetic double-strand siRNA targeting livin (livin-siRNA) was transfected into HT-29 cells, and then RT-PCR and Western blotting were used to detect the expression of livin mRNA and protein in HT-29 cells. MTT assay was performed to analyze the proliferation of HT-29 cells. The cell apoptosis and cell cycle distribution were analyzed by flow cytometry. The invasion assay and caspase-3 detective kit were used to detect the change of invasion and caspase-3 activity in HT-29 cells. Results: Forty-eight hours after transfection, there was a significant decrease in the expressions of both livin mRNA $(0.073\pm0~007~vs~0.395\pm0~082, 0.423\pm0.025, 0.418\pm0.032, P~<0.05)$ and livin protein $(0.106\pm0.003~vs~0.456\pm0~065, 0.473\pm0~078, 0.491\pm0.045, P~<0.05)$ in the livin-siRNA group, compared with the blank and negative control and liposome groups. Ninety-six hours after transfection, the growth of HT-29 cells in the livin-siRNA group was significantly lower than that in the control and liposome groups $(0.564\pm0.102~vs~0.833\pm0.127, 0.860\pm0.153, P~<0.05)$, and the rate of apoptosis was obviously increased $(\[16.5\pm2.8 \] \%~vs~ \[2.4\pm0.5 \] \%, \[3.7\pm1.0 \] \%, P~<0.05)$. The invasion assay demonstrated that the number of the migration cells was lower in the livin-siRNA group than in the control and liposome groups $(31.3\pm4.5~vs~101.3\pm8.6, 97.4\pm7.8, P~<0.05)$. The activity of caspase-3 in the livin-siRNA group was decreased compared with that in the control group $(0.160\pm0.023~vs~0.347\pm0.058, P~<0.05)$. Conclusion: The siRNA silencing livin expression in HT-29 cells can suppress the proliferation, induce the apoptosis and inhibit the invasion of HT-29 cells.

Keywords:colon cancer livin small interference RNA gene therapy

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