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561~565.siRNA对人结肠癌细胞黏附力和侵袭性的抑制作用[J].贾如江,侯丽艳,刘 奇,冯运章.中国肿瘤生物治疗杂志,2008,15(6)

siRNA对人结肠癌细胞黏附力和侵袭性的抑制作用 点此下载全文

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基金项目:

DOI: 10.3872/j.issn.1007-385X.2008.6.012

摘更.

目的: 探讨VEGFR 3对人结肠癌细胞黏附力和侵袭性的影响。方法:构建携靶向 VEGFR 3 基因siRNA(small interfering RNA)表达载体,转染人结肠癌LoVo细胞,半定量RT PCR和Western blotting检测转染前后LoVo细胞 VEGFR 3 mRNA和蛋白表达的变化,基质 黏附实验检测细胞转染后的黏附能力,细胞侵袭实验检测转染后肿瘤细胞侵袭性的改变。结果:携靶向 VEGFR 3 基因siRNA的表达载体成功构建,RT PCR检测转染siRNA后LoVo细胞 VEGFR 3 mRNA表达水平降低;Western blotting检测转染siRNA后72 h LoVo细胞VEGFR 3蛋白表达下降,其表达相对值由(1.26±0.19)降至(0.39±0 12)(P <0.05)。转染siRNA 72 h 后LoVo细胞的黏附能力是著下降\[(0.626±0.047) vs (0.407±0.029),P <0.05\];上oVo细胞穿膜细胞数(6.38±3.25)明显低于空白对照组(24.82±3.44),非特异性对照组(23.58±3.73)(P <0.05)。结论:siRNA能够在LoVo细胞中引发RNA干扰效应,下调VEGFR 3 基因的表达,进而抑制LoVo细胞的黏附能力和侵袭性。

关键词: RNA干扰 血管内皮生长因子受体 3(VEGFR 3) 结肠癌细胞 黏附能力 侵袭能力

targeted siRNA against adherence and invasion of human colon cancer cells

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Abstract:

Objective: To construct a small interfering RNA (siRNA) expression vector (psiRNA VEGFR 3) targeting vascular endothelial growth factor receptor 3 (VEGFR 3) and to investigate the effects of VEGFR 3 siRNA on the adherence and invasion of human colon cancer cells. Methods: A siRNA expression vector (psiRNA VEGFR 3) targeting VEGFR 3 were constructed and was used to transfect LoVo cells via lipofectamine 2000. The mRNA and protein expression of VEGFR 3 were examined after transfection by reverse transcriptase polymerase chain reaction (RT PCR) and Western blotting, respectively. The tumor adhesion ability was detected by cell matrix adhesion experiment and the invasion ability of tumor cells was evaluated by millicell chamber model. Results: The VEGFR 3 siRNA expression vector was successfully constructed. The expression of VEGFR 3 mRNA and protein was inhibited after psiRNA VEGFR 3 transfection. Seventy two hours after psiRNA VEGFR 3 transfection, Western blotting assay showed that the expression of VEGFR 3 protein was decreased from (1.26 ± 0.19) to $(0.39\pm0.12)(P=<0.05)$, the adhesion ability of LoVo cells was also significantly decreased compared with the untransfected group and negative control group $(0.407\pm0.029 \text{ vs} 0.626\pm0.047,0.621\pm0.068, P<0.01)$. The invasion assay demonstrated that the number of LoVo cells penetrating the membrane in the transfection group was significantly lower than those in the untransfected and negative control group $(6.38\pm3.25 \text{ vs} 24.82\pm3.44, 23.58\pm3.73, P<0.05)$. Conclusion: The siRNA of VEGFR 3 gene can effectively inhibit the mRNA and protein expression of VEGFR 3 in LoVo cells, therefore restraining the adhesion and invasion ability of LoVo cells.

Keywords: RNA interference vascular endothelial growth factor receptor 3 (VEGFR 3) colon cancer cell adhesion ability invasion ability

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