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RNA干扰CXCR4表达抑制乳腺癌MDA-MB-231细胞的增殖、黏附和迁移 点此下载全文

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摘要:

目的:构建CXC趋化因子受体4(CXC chemokine receptor 4, CXCR4)RNA干扰真核表达载体,研究其对人乳腺癌细胞MDA-MB-231增殖、黏附及迁移能力的抑制作用。方法:构建针对CXCR4的带发夹结构的小RNA干扰序列,并连接到pGCsi-U6-Neo-GFP载体中,转染293T细胞,筛选出干扰效率最高的表达载体。脂质体法转染MDA-MB-231细胞。利用CCK8法、细胞-基质黏附实验和划痕修复实验检测shRNA干扰CXCR4表达对MDA-MB-231细胞增殖、黏附和迁移能力的影响。结果:成功构建CXCR4-shRNA重组质粒,并转染293T细胞,利用RT-PCR及Western blotting检测发现CXCR4沉默效率最高可达81.3%。CXCR4-shRNA转染能显著抑制MDA-MB-231细胞的增殖(P<0.05)以及细胞与细胞外基质的黏附(P<0.05)。CXCR4-shRNA转染组MDA-MB-231细胞的迁移距离明显低于对照质粒组和空白对照组(P<0.01)。结论:CXCR4-shRNA干扰载体能特异性抑制CXCR4的表达,从而抑制乳腺癌MDA-MB-231细胞的增殖、黏附及迁移。

关键词: CXC趋化因子受体4 RNA干扰 真核表达载体 乳腺癌

Interfering CXCR4 expression inhibits proliferation, adhesion and migration of breast cancer MDA-MB-231 cells Download Fulltext

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Abstract:

Objective: To construct short-hairpin RNA (shRNA) eukaryotic expression vector targeting CXC chemokine receptor 4 (CXCR4), and to observe its impact on the proliferation, adhesion and migration of human breast cancer MDA-MB-231 cells. Methods: The fragments of CXCR4 shRNA were synthesized and cloned into pGCsi-U6-Neo-GFP vector. The recombinant plasmids were transfected into 293T cells and the most effective interfering vector was selected. MDA-MB-231 cells were transfected by liposome assay. The effects of silencing CXCR4 expression by shRNA on the growth, adhesion and migration of MDA-MB-231 cells were determined by CCK8, cell-matrix adhesion and wound healing assays, respectively. Results: The shRNA eukaryotic expression vectors targeting CXCR4 (CXCR4-shRNA) were successfully constructed and transfected into 293T cells. RT-PCR and Western blotting results showed that the maximum inhibitory rate of CXCR4 expression was 81.3%. CXCR4-shRNA transfection significantly inhibited the proliferation of MDA-MB-231 cells (P<0.05) and the adhesion between MDA-MB-231 cells and extracellular matrix (P<0.05). Wound healing experiment showed that the migration distance of MDA-MB-231 cells in CXCR4-shRNA transfection group was significantly lower than those in the control plasmid and the blank control group (P<0.01). Conclusion: CXCR4-shRNA interfering vector can specifically inhibit CXCR4 expression, proliferation, adhesion and migration of MDA-MB-231 cells.

Keywords: CXC chemokine receptor 4 RNA interference eukaryotic express vector breast cancer

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