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miR-155对人肺癌95D细胞生物学行为的影响 点此下载全文

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摘要：

目的：构建携microRNA-155（miR-155）的真核表达载体并观察其转染高转移性人巨细胞肺癌95D细胞后细胞的生物学行为变化。方法：以95D细胞基因组RNA为模板，经PCR法扩增miR-155的前体序列，由 Bam H Ⅰ和 Hin dⅢ双酶切后将其亚克隆入真核表达载体pcDNA 3.1(-)，并进行双酶切及测序鉴定；将构建成功的pcDNA3.1(-)-pri-miR-155载体（命名为p-miR-155）体外瞬时转染人肺癌95D细胞，利用 Real-time PCR探针法检测miR-155成熟体的表达水平，并利用CCK-8法、克隆形成实验和划痕法检测95D细胞的增殖、克隆形成以及体外迁移能力。 结果： 成功构建携miR-155的真核表达载体；与空白（Mock）和对照组（p-Ctrl）相比，转染后的95D细胞过表达miR-155\[（2.04±0.62) vs (0.76±0.62)、(1.00±0.45)，均 P <0.01\]、p-miR-155载体转染组95D细胞的增殖抑制明显增加\[(46.70±6.89)% vs （3.70±1.40）%、（1.11±0.75）%， P <0.01\]、克隆形成能力（在100和1 000个细胞/孔接种条件下）明显下降\[（12±3) s (34±3)、(35±3)个， P <0.01；(78±4) vs (159±4)、(165±4)个， P <0.01）\]，此外，细胞的迁移细胞数也明显减少\[(110±5) vs (295±5)、(325±5)个， P <0 01\]。 结论： 通过miR-155真核表达载体转染产生的过表达miR-155可显著抑制人肺癌95D细胞的增殖和迁移能力。

关键词：microRNA-155(miR-155) 真核表达 肺癌 95D细胞 增殖 迁移

Biological effects of miR-155 on human lung cancer 95D cell Download Fulltext

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Abstract:

Objective: To evaluate the effect of microRNA-155 (miR-155) on human lung cancer cell behaviors in vitro . Methods: A plasmid vector (p-miR-155) carrying the pri-miR-155 sequence amplified from genomic RNA of human lung cancer 95D cells by PCR was constructed. Lung cancer 95D cells were transiently transfected with p-miR-155. p-miR-155 mRNA abundance in 95D cells was assessed by real-time PCR before and after transfection. Cell proliferation and migration in control, mock-transfected and transfected 95D cells were assessed by CCK-8 assay wound assay respectively. Results: The abundance of miR-155 mRNA was increased significantly in 95D cells transfected with p-miR-155 than in mock-transfected and control cells (2.045±0.62 vs 0.76±0.62, 1±0.45; P <0.01). The proliferation was markedly inhibited in p-miR-155 transfectants as compared in mock-transfected and control cells (\[46 70±6 89\]% vs \[3.70±1.40\]%, \[1.11±0.75\]%; P <0.01). The number of colonies formed was significantly decreased (\[12±3\] vs \[34±3\], \[35±3\]; P <0.01) and so was migration capability (\[110±5\] vs \[295±5\], \[325±5\]; P ＜0.01) in p-miR-155-transfected cells as compared with mock-transfected and control cells. Conclusion: A eukaryotic expression vector carrying human pri-miR-155 sequence is capable of effectively inhibiting lung cancer cell proliferation and migration, thus having a significant clinical implication.

Keywords:microRNA-155 (miR-155) eukaryotic expression lung cancer 95D cell proliferation migration

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