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基础医学

STAT3真核表达质粒的构建及对胃癌细胞增殖的影响

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1.山东大学医学院,济南 250012; 2. 山东大学附属济南市中心医院中心实验室,济南250013 摘要:

目的 构建人信号转导和转录激活因子3(STAT3)真核表达载体,研究STAT3基因对人胃癌细胞 ▶参考文献 株MGC803的影响。方法 构建pcDNA3-STAT3真核表达质粒,经脂质体介导转染胃癌细胞株 MGC803。RT-PCR和Western blotting检测转染后STAT3、P-STAT3(Y705)、P-STAT3 (S727)、Survivin 和 PCNA在MGC803细胞中mRNA和蛋白水平的表达,CCK-8法检测 pcDNA3-STAT3对细胞MGC803的影响。结果 测序及酶切鉴定证实, 真核表达质粒pcDNA3-STAT3构建成功。Western blotting和RT-PCR实验结果证实,与转染空载体组相比,转染重组质 》加入引用管理器 粒组STAT3、P-STAT3(Y705)、P-STAT3(S727)、Survivin 和 PCNA在蛋白水平和mRNA水平 的表达均明显升高,差异有统计学意义(P<0.05)。CCK-8实验结果显示,与转染空载体组相 比,转染重组质粒组人胃癌细胞株MGC803增殖能力明显升高,差异有统计学意义(P<0.05)。 结论 真核表达质粒pcDNA3-STAT3可以提高STAT3基因在人胃癌细胞株MGC803中的表达, 并且增强胃癌细胞的增殖能力。

关键词: 胃肿瘤: 信号转导和转录激活因子3; 细胞增殖能力

Construction of STAT3 eukaryotic over-expression vector and its effects on the proliferation of gastric cancer cells

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Abstract:

To investigate the effects of overexpression human signal transducer and activator of transcription 3(STAT3) on human gastric cancer cell line MGC803 by the construction of a STAT3 gene eukaryotic expression vector. Methods The eukaryotic expression vector pcDNA3-STAT3 was constructed and transfected into the gastric cancer cell line (MGC803) by the liposome. The mRNA and protein expression levels of STAT3, P-STAT3(Y705), P-STAT3(S727), Survivin and PCNA in MGC803 were detected by RT-PCR and Western blotting. The effects of STAT3 on MGC803 cell proliferation were detected through the CCK-8 assay. Results
The DNA sequencing and restriction endonuclease identification results confirmed that the eukaryotic expression plasmid pcDNA3-STAT3 was successfully constructed. Western blotting and RT-PCR results confirmed that STAT3, P-STAT3(Y705), P-STAT3(S727), Survivin and PCNA protein and mRNA expression levels were significantly increased in the recombinant plasmid group compared with the empty vector group. CCK-8 results proved that, compared with the empty vector group, the recombinant plasmid group showed higher proliferative capacity, and the difference was statistically significant (P<0.05). Conclusion eukaryotic expression plasmids of pcDNA3-STAT3 can increase the expression of STAT3 gene in human gastric cancer cell line MGC803, and enhance the proliferation of gastric cancer cells.

Keywords: Stomach neoplasm; Signal transducer and activator of transcription 3; Cell proliferating ability

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