

中国肿瘤生物治疗杂志

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419~423.SDF-1y/rhGM-CSF融合蛋白的制备及其趋化作用[J].居小萍,徐新颜,张晓青,刘永明,于春山,曹洋森,卢明智,陈樱.中国肿瘤生物治疗杂志,2011,18(4)

SDF-1γ/rhGM-CSF融合蛋白的制备及其趋化作用 点此下载全文

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基金项目:上海市自然科学基金资助项目(No.06ZR14114),军队医学研究"十一五"计划课题资助项目(No.10MA004)

DOI:

摘要:

目的: 利用基因工程技术制备SDF-1与hGM-CSF的融合蛋白(SDF-1γ/rhGM-CSF),研究该融合蛋白对肿瘤患者造血和免疫功能的增强作用。 方法: 构建表达SDF-1γ/rhGM-CSF融合蛋白的pPIC9k-SDF1-rhGM-CSF1质粒,转染酵母菌,诱导SDF-1γ/rhGM-CSF融合蛋白的表达,Western blotting鉴定SDF-1γ/rhGM-CSF融合蛋白的表达。集落形成实验观察SDF-1γ/rhGM-CSF对骨髓细胞集落形成的影响,趋化实验检测其对未成熟树突状细胞(dendritic cell,DC)的趋化作用。 结果: 成功构建pPIC9k-SDF1-rhGM-CSF1质粒,高表达SDF-1γ/rhGM-CSF融合蛋白,分子量约为25 000,并可被GM-CSF特异性抗体所识别。SDF-1γ/rhGM-CSF融合蛋白能显著刺激骨髓细胞的集落形成,其效果强于GM-CSF(P <0.05)。与SDF-1相比,SDF-1γ/rhGM-CSF融合蛋白可更有效地趋化未成熟DC(P <0.05)。 结论: SDF-1γ/rhGM-CSF融合蛋白可有效促进骨髓细胞的集落形成,趋化未成熟DC,有促进肿瘤化疗患者造血和免疫功能恢复的潜在临床应用前景。

关键词: <u>SDF-1</u> <u>GM-CSF</u> 融合蛋白 <u>肿瘤</u> 造血 <u>免疫</u> 趋化

Preparation of SDF-1\(\bar{\psi}\)/rhGM-CSF fusion protein and its chemotactic effect
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Fund Project: Project supported by the Natural Science Foundation of Shanghai (No.06ZR14114), and the "11th Five-Year Plan" Program of PLA (No.10MA004)

Abstract:

Objective: To prepare the fusion protein of SDF-1 and rhGM-CSF (SDF-1y/rhGM-CSF) by genetic engineering technology, and investigate its hematopoietic and immune promotion functions in tumor patients. Methods: The expression vector for SDF-1y/rhGM-CSF fusion protein, pPIC9k-SDF1-rhGM-CSF1, was constructed and the protein expression was induced by yeast transfection. SDF-1y/rhGM-CSF fusion protein was further identified by Western blotting analysis. Colony-formation assay and chemoattract assay were used to study the roles of the prepared fusion protein in stimulating bone marrow cell colony-formation and in chemoattracting immature dendritic cells. Results: SDF-1y/rhGM-CSF fusion gene vector, pPIC9k-SDF1-rhGM-CSF1, was successfully constructed and expressed high level of SDF-1y/rhGM-CSF fusion gene. The molecular weight of the expressed protein was about 25 000 and was recognized by GM-CSF specific antibody. The fusion protein had a stronger effect in stimulating bone marrow cell colony-formation than GM-CSF (P <0 05) and in chemoattracting immature dendritic cells than SDF-1 (P <0.05). Conclusion: SDF-1y/rhGM-CSF fusion protein can promote bone marrow cell colony-formation and chemoattraction of immature dendritic cells, which might be used for promoting hematopoiesis and immune function of tumor patients after chemotherapy.

Keywords: SDF-1 GM-CSF fusion protein tumor hematopoiesis immunity chemotaxis

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