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论文

# 重组金葡菌肠毒素O的克隆表达及生物学活性分析

潘映秋;丁丁;孙红颖;陈枢青

浙江大学 药学院 药物毒理与生化药学研究所， 浙江 杭州 310058

摘要：

克隆金葡菌肠毒素O(SEO)的全长基因，实现其可溶性表达,并对纯化的表达产物进行生物学活性分析。从金葡菌FRI 100菌株基因组中得到SEO基因，克隆至谷胱甘肽S-转移酶(GST)融合表达载体pGEX-4T-1，转化大肠杆菌，获高效表达。融合蛋白GST-SEO经Glutathione Sepharose 4B亲和纯化和凝血酶消化获重组SEO(rSEO)后，MTT法检测脾淋巴细胞的增殖作用，分析纯化后rSEO的生物学活性。测序结果表明，得到正确的肠毒素SEO基因序列，并获高效表达的融合蛋白；MTT结果表明，rSEO具有与SEC相当的显著的促淋巴细胞增殖以及抑制肿瘤细胞生长的能力。本研究成功克隆、表达、纯化了具有抗肿瘤生物学活性的rSEO蛋白，为进一步研究该蛋白的抗肿瘤机制奠定了基础，并有望成为一种新的超抗原制剂用于肿瘤的临床治疗。

关键词： 肠毒素 超抗原 重组蛋白 细胞增殖

# Expression and bioactivity of the cloned staphylococcal enterotoxin O

PAN Ying-qiu; DING Ding; SUN Hong-ying; CHEN Shu-qing

Abstract:

This study is to clone the gene of staphylococcal enterotoxins O, obtain recombinant protein (rSEO) and investigate its activity on mice lymphocyte. *Staphylococcus aureus* O gene is cloned into GST gene fusion vector pGEX-4T-1. The resultant plasmid pGEX-4T-SEO was used to transform *E.coLi* BL21, where the GST-SEO fusion protein was expressed efficiently. Then SEO was purified by Glutathione Sepharose 4B affinity column and digested with thrombin. The bioactivity of SEO was analyzed by MTT assay on mice lymphocyte and tumor cells. The nucleotide sequence was confirmed to code for the protein correctly, and soluble SEO was expressed efficiently in *E.coli* BL21 with pGEX-4T-SEO. The protein purified by affinity chromatography resulted to be one single band by SDS-PAGE detection. The MTT assay of the purified rSEO demonstrated that its abilities of stimulating T cells and inhibiting the proliferation of K562, K562-ADM and B16 cells were equivalent to that of SEC *in vitro*. The expression plasmid pGEX-4T-SEO was constructed and the recombinant superantigen was expressed successfully, which may provide a foundation for the further research of the anticancer activity of SEO.

Keywords: superantigen recombinant protein cell proliferation enterotoxin

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作者简介:

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