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从小鼠胎儿基因文库中筛选分离免疫球蛋白重链Cε基因

杨建清, 杨志兴, 杨学成, 曲宝兰, 董甲有, 王润芝, 刘伟民

黑龙江省应用微生物研究所，哈尔滨

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摘要　本文阐述了以缺口翻译制备的pIgB DNA为探针(4.4×10 4 cpm/μg DNA),用吸印法从小鼠儿基因文库中筛选分离出免疫球蛋白重链Cε基因克隆。同时，从扩大培养的噬菌体中提取出DNA，并用EcoRI酶消化，0.7%琼脂糖凝胶平板电泳分离，分离的DNA片段转移至硝酸纤维素滤膜上，以pIgB DNA探针杂交，进而确证有两条阳性带，为免疫球蛋白重链Cε基因。电泳法测定其大小分别约为3.5kb 和3.0kb。也显示出在小鼠胎儿中的未分化型免疫球蛋白重链Cε基因内有一个限制性内切酶Eco RI位点。

关键词

分类号

Screening and Isolating of Mouse Immunoglobulin Heavy Chain C8 Gene from New Bor n Mouse DNA Library

Yang Jianqing, Yang Zhixing, Yang Xuecheng, Qu Vaolan, Dong Jiayou, Wang Runzhi, Liu Weimin

Heilonjiang Applied Microbiological Institute,Harbin

Abstract

Although immunoglobulin IgE has an important biological function as the mediator of allergic reaction,we know little about the genetics of the C8.For its further study we have screened and isolated the C8 gene from the new born mice DNA library using the following methods:Denton Davis screen,Nick translation Southern blotting,hybridization,digested cloning C8 DNA with EcoRI and electrophoresis on the agarose plate.We have got two positive C8 gene clones from 10¡Á10 6 phages by pIg ¦Å DNA probe.During the process of rescreening we got more than 95% positive plaques on a plate as shown in Fig.2. After that we extracted cloning C8 DNA and had them digested with EcoRI.Then we electrophoresised them on 0.7% agarose plate and Southern blotting again,hybridized with pIg ¦Å DNA and autoradiographied as shown in Fig. 3(2).Finally we have got immunoglobulin C¦Ågene fragments that were cut into two fragments by EcoRI so as to find another EcoRI cut site on them.

Key words

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