论著

日本血吸虫表膜蛋白Tetraspanin 2-A基因的克隆、 表达与鉴定

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收稿日期 修回日期 网络版发布日期 接受日期

摘要

目的 扩增并表达日本血吸虫表膜蛋白(SjTsp2-A)基因 SjTsp2。 方法 根据与曼氏血吸虫表膜蛋白 SmT-sp2 基因同源的SjTsp2 序列(编号为AY810722)设计引物,体外扩增目的蛋白基因,并将其亚克隆 入原核表达载体pET28a,转化至大肠埃希菌 (*E. coli*) BL21株,异丙基-β-D-硫代半乳糖苷(IPTG)进行诱导表达,用纯化的重组蛋白免疫小鼠,ELISA检测其血清抗体效价,蛋白质印迹 (Western blotting)分析鉴定其免疫反应性。 结果 PCR扩增获得SjTsp2基因大环核苷酸序列为 228 bp,与曼氏血吸虫表膜蛋白(SmTsp2)的氨基酸序列同源性为 52%。 SjTsp2基因亚克隆入pET28a并可溶性表达,免疫小鼠可产生高滴度(最高1:32 000)特异性抗体。Western blotting分析结果表明,纯化的SjTsp2-A蛋白能被疫区居民血吸虫抗体阳性者血清、急性及慢性血吸虫病患者血清识别,与健康人血清无反应。 结论 日本血吸虫表膜蛋白基因SjTsp2在体外获得表达,其表达蛋白具有一定的抗原性。

Cloning, Expression and Identification of Membrane Protein Tetraspanin 2-A of *Schistosoma japonicum*

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Abstract

【Abstract】 Objective To clone and express a membrane protein(Tetraspanin 2) gene of *Schistosoma japonicum* (SjTsp2). Methods A pair of primers was designed to amplify the SjTsp2 gene which was subcloned into prokaryotic plasmid pET28a(+). The recombinant plasmid was transformed into *E. coli* BL21(D3) and followed by expression of the protein induced by IPTG. The protein was purified by affinity chromatography and used to immunize BALB/c mice. Dilution of antibody against SjTsp2 was determined by ELISA. The protein was also identified by Western blotting. Results Big loop of SjTsp2-A, 228 bp, was amplified in vitro by PCR. Its deduced amino sequence shared 52% similarity with SmTsp2. The soluble recombinant SjTsp2-A was expressed in the experiment and high dilution antibody against the recombinant (1 : 32 000 in maximum) was produced in immunized mice. SjTsp2-A reacted positively with sera of acute and chronic schistosomiasis patients but not with sera from healthy persons by Western blotting. Conclusion SjTsp2 has been expressed and shows certain antigenicity. Key words *Schistosoma japonicum* Membrane protein Tetraspanin 2 Gene Antigenicity

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