

论著

日本血吸虫线粒体相关蛋白的基因克隆及特性鉴定

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摘要

[目的] 探索研制血吸虫病疫苗的新途径, 对日本血吸虫线粒体相关蛋白进行基因克隆及特性鉴定。
[方法] 分析本室筛选日本血吸虫成虫cDNA文库获得的 1个cDNA片段 (Sj338 2 4)的开读框序列, 在其上下游分别设计引物A和B, 并以该cDNA片段为模板进行PCR扩增后, 将该片段重组于pGEM T中并进行DNA测序鉴定及检索。再经酶切后将该基因片段亚克隆入表达载体pGEX 6P 1, 并进行蛋白表达、纯化及抗原性鉴定。[结果] 该目的基因PCR产物全长共 487bp, 其开读框由 45 9bp组成, 编码 15 3个氨基酸经残基组成的多肽。DNA序列同源性分析发现Sj338克隆基因与人及褐鼠的线粒体外膜蛋白的部分编码基因较高度同源。重组质粒pPEX 6P 1 Sj338能高效融合表达, 理论蛋白的分子量为 17kDa。SDS PAGE和Westernblotting检测结果表明, 重组蛋白rSj338具有良好的抗原性。[结论] Sj338可能为日本血吸虫线粒体相关蛋白的基因, 重组蛋白有望成为新的疫苗候选分子。

关键词 [日本血吸虫](#) [线粒体](#) [基因克隆](#) [重组抗原](#) [融合表达](#) [线粒体相关蛋白](#)

分类号

GENE CLONING AND CHARACTERIZATION OF MITOCHONDRIA-RELATED PROTEIN OF SCHISTOSOMA JAPONICUM

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Abstract

[Objective] To subclone and characterize a cDNA clone coding for Schistosoma japonicum (S.j.) mitochondria-related protein. [Methods] The open reading frame of the fragment(Sj338/24) obtained from an adult worm cDNA library of S.j. was analysed, at the upstream and downstream of the open reading frame(ORF) the primers A and B were designed, respectively, and the cDNA fragment was used as PCR template. The Sj338 gene fragment obtained was amplified by PCR method and then subcloned into pGEM-T vector for sequencing. The gene sequence was analyzed and the target fragment was restrictedly digested and subcloned into expression vector pGEX-6P-1. The expressed recombinant protein was purified and characterized. [Results] The cloned Sj338 gene was demonstrated to be 487 bp long containing one 459 bp ORF, encoding a protein with a molecular weight of 17 kDa. The nucleotide sequence of the cloned gene Sj338 had higher homology with those genes coding for mitochondrial outer membrane protein of Homo sapiens and Rattus norvegicus. The recombinant construct of pGEX-6P-1/Sj338 could be expressed efficiently and the antigenicity of its product rSj338 has been demonstrated by Western blotting. [Conclusion] Sj338 may be the gene coding for S.j. mitochondria-related protein and the recombinant protein may be used as a new vaccine candidate.

Key words [Schistosoma japonicum](#) [mitochondria](#) [gene cloning](#) [recombinant antigen](#) [fusion expression](#)

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