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论著

恶性疟原虫乳酸脱氢酶的表达及免疫活性鉴定

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摘要

目的 在大肠杆菌中表达恶性疟原虫乳酸脱氢酶 (LDHp)与谷胱甘肽S 转移酶 (GST)融合蛋白 ,测定重组蛋白的免疫活性。方法 采用PCR方法特异性扩增恶性疟原虫 (海南株)乳酸脱氢酶基因 ,经双酶切后克隆入 pGEX 4T 1表达载体中 ,重组蛋白纯化后免疫小鼠制备特异性血清 ,并用琼脂双向扩散法检测效价 ,ELISA、Western bloting检测重组抗原的免疫活性。结果 得到了重组表达的蛋白抗原 ,表达产物能与兔抗恶性疟原虫血清发生反应 ,并能诱导小鼠产生特异性体液免疫应答 ,免疫琼脂扩散法抗体滴度为 1∶16。结论 LDHp在大肠杆菌中获得高效表达且表达产物具有良好的抗原性。

关键词 恶性疟原虫 乳酸脱氢酶 表达 免疫活性

分类号

Expression and Immunocompetence Characterization of Plasmodium falciparum Lactate Dehydrogenase

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Abstract

Objective To express lactate dehydrogenase (LDH) gene of Plasmodium falciparum FCC1/HN in the E. coli TG1 and analyse its immunocompetence. Methods The LDH gene of the P. falciparum was specifically amplified by polymerase chain reaction, and the recovered gene fragment was cloned into pGEX 4T 1 vector for expression of fusion protein with glutathione S transferase(GST). The recombinant plasmid was transformed into the E. coli TG1. Four mice (Kunming strain) were immunized with purified expressed protein(antigen) and the polyclonal antibodies were collected. The immunocompetence of recombinant protein was analysed by ELISA and Western blot. Results The LDH gene of P. falciparum was successfully expressed in the E. coli TG1. The expressed protein exhibited a specific reaction with immune sera obtained from rabbits immunized with P. falciparum . The specific humoral responses were induced in mice and the titer of the specific antibody was 1∶16 by two dimensional diffusion assay. Conclusion The LDH gene of P. falciparum has been successfully expressed in the E. coli TG1 and the expressed protein has high antigencity.

Key words Plasmodium falciparum lactate dehydrogenase expression immunocompetence

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