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论著

恶性疟原虫TRAP/CSP融合抗原的构建及表达

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摘要

目的 构建恶性疟原虫红前期融合蛋白 4(PfCP 4)。 方法 通过甘氨酸 脯氨酸 甘氨酸 (GPG)接点将恶性疟原虫 3D7株血凝素相关匿名蛋白 (TRAP)膜外区序列 (氨基酸 2 6～ 3 3 0)和环子孢子蛋白 (CSP) 19个 4肽重复区及其羧基末端序列 (氨基酸 199～ 3 83)连接 ,采用不对称PCR法人工合成 15 77bpPfCP 4基因。将PfCP 4基因克隆在pQE表达质粒上 ,转化大肠杆菌SG13 0 0 9后进行诱导表达 ,用抗CSP的免疫血清进行免疫印迹检测。 结果 免疫印迹检测显示在 5 7kDa处出现特异的表达条带 ,其大小与推算的PfCP 4分子量一致 ,表明PfCP 4合成基因能在大肠杆菌中表达分子量为 5 7kDa的PfCP 4重组蛋白。 结论 成功构建了PfCP 4。

关键词 恶性疟原虫 融合蛋白 环子孢子蛋白 血凝素相关匿名蛋白 基因表达

分类号

Construction and Expression of a TRAP/CSP Chimeric Protein of Plasmodium falciparum

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Abstract

Objective To construct a chimeric protein of Plasmodium falciparum pre erythrocyte stage (named as PfCP 4). Methods Thrombospondin related anonymous protein (TRAP) and circumsporozoite protein (CSP) of Plasmodium falciparum have been considered important candidates for pre erythrocytic malaria vaccine. The sequences of ectodomain of TRAP (aa: 26-330) and (NANP) 19 repeat region and entire carboxy terminus of CSP were fused to generate the PfCP 4 via a hinge consisting of Gly Pro Gly. The 1577 bp sequence of PfCP 4 was synthesized by asymmetric PCR based method and the synthetic gene was inserted into pQE. The resulting plasmid was transformed into E. coli SG13009 for inducible expression with IPTG. The expression product was detected by Western blotting. Results The result of Western blotting showed that the entire PfCP 4 recombinant protein was produced under IPTG induction whereas no product was detected in the cell without induction. The molecule weight of the protein was 57 kDa which was identical to the expected size, and the product was recognized by polyclonal antibodies against CSP protein. Conclusion A chimeric protein of Plasmodium falciparum pre erythrocyte stage (named as PfCP 4) was constructed successfully.

Key words

Plasmodium falciparum chimeric protein CSP TRAP gene expression

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