

论著

猪囊尾蚴特异性抗原的筛选、鉴定和生物信息学分析

方文¹,包怀恩^{2*},肖靛靛²,牟荣²

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摘要

目的 对猪囊尾蚴的特异性抗原进行筛选、鉴定和生物信息学分析。方法 四川省雅江县呷拉乡猪带绦虫病患者,口服槟榔-南瓜子驱虫,收集、制备虫卵悬液(8万个/ml)。将6头20 d龄三元杂交乳猪均分实验组和空白对照组,实验组每猪灌胃虫卵悬液1 ml。40 d后,分别制备实验组心脏血混合血清及对照组心脏血混合血清;收集、制备实验组猪囊尾蚴蛋白,进行双向电泳(2-DE)分析。将凝胶蛋白斑点转移至聚偏氟乙烯膜(PVDF膜),用实验组混合血清及对照组混合血清(均为1:10)为一抗,羊抗猪HRP-IgG(1:200)为二抗,进行蛋白质印迹(Western blot-ting)分析,比较两组杂交蛋白点的差异,确定猪囊尾蚴抗原抗体阳性杂交斑点,据此挖取双向电泳与之对应的蛋白点,用电喷雾离子阱型质谱仪(ESI-Trap MS)进行质谱鉴定。搜索美国国家生物技术信息中心(NCBI)网站信息进行生物信息学分析。结果 双向电泳凝胶共检测到207±9个蛋白质斑点,相对分子质量为Mr 14 400~94 000,等电点(pI)为3.0~10.0。筛选出7个特异性抗原,其中的4个分别为AF239799-1膜联蛋白(AF239799-1 annexin)、细胞支架肌动蛋白-2(cytoskeletal actin-2)、原肌球蛋白(tropomyosin)和肌动蛋白-1(actin-1)。前1个蛋白已被鉴定为猪带绦虫特异性抗原,后3个蛋白抗原与NCBI网上登录的亚洲带绦虫成虫蛋白一致,为猪囊尾蚴特异性抗原。结论 共获得3个猪囊尾蚴特异性抗原,即细胞支架肌动蛋白-2、原肌球蛋白和肌动蛋白-1。

关键词

猪带绦虫 囊尾蚴 抗原 双向电泳 蛋白质印迹 质谱

分类号

Screening, Identification and Bioinformatic Analysis of *Taenia solium* Cysticercus

FANG Wen¹,BAO Huai-en^{2*},XIAO Liang-liang²,MU Rong²

Abstract

Objective To screen and identify specific antigens of *Taenia solium* cysticercus, and predict the function of target proteins using bioinformatics method. Methods Patients infected with *Taenia solium* were dewormed by decoction arecae and pumpkin seeds to collect worms, and eggs were then prepared. Six three-way crossed hybrid pigs were randomly divided into experimental group and control group, and each experimental pig was infected with 80 000 *T. solium* eggs. Serum samples were collected at 40 days after infection. The total protein of *T. solium* cysticercus was separated by two-dimensional electrophoresis, and Western blotting was performed to find out distinct antigens. Proteins from the two groups were identified by ESI-Trap MS. Query in NCBI database was made to confirm function of the proteins. Results 207±9 spots were detected through Coomassie brilliant blue-stained gels with Mr 14 400-94 000 and pI 3.0-10.0. Western blotting showed 7 specific antigen spots with pool sera of infected pigs. Four of the 7 antigens with known functions were respectively ascribed to cytoskeletal actin-2 (adult-specific), tropomyosin (cysticercus-specific), AF239799-1 annexin (cysticercus-specific) and actin-1 (cysticercus-specific). Conclusion Three specific antigens of *Taenia solium* cysticercus have been identified.

Key words

Taenia solium Cysticercus Antigen Two-dimensional electrophoresis Western blotting MS/MS spectrometry

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方文¹;包怀恩^{2*};肖靓靓²;牟荣²