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实验研究

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田鼠巴贝虫可溶性抗原组分分析及其初步应用

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Component analysis and priliminary application of soluble antigens of Babesia microti

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摘要

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摘要 目的 分析田鼠巴贝虫可溶性抗原,寻找可用于免疫诊断的有效抗原组分。方 法 用田鼠巴贝虫感染BALB/c小鼠, 待虫血症达高峰期时, 收集田鼠巴贝虫虫体; 采 用超声法制备可溶性粗抗原(soluble babesia antigens, SBA)并包板, ELISA检测 血清特异性IqG, 评价SBA的免疫反应性、交叉反应性和特异性; 以SDS-PAGE电泳 分析SBA组分, 并进行Western Blot, 分析其与感染鼠血清的反应性。结果 SBA-ELISA法可检测早期感染(7 d)小鼠血清, 且与恶性疟、间日疟弓形虫病阳性血清无 交叉反应,显示出其较好的诊断敏感性和较高的特异性。通过SDS-PAGE分析,获得 5条分子质量为72、66、 60、 53、43 kDa的主蛋白带和介于14.4~116 kDa的 7条次带。经Western Blot分析, SBA有15个抗原组分能被阳性小鼠血清识别, 以 72、53、43、39、30 kDa蛋白组分反应较强。结论 以SBA建立的间接ELISA可 用于巴贝虫感染的有效筛查工具: 田鼠巴贝虫可溶性抗原组分中72、53、43、39、 30KDa为比较理想的抗原组分。

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关键词: 田鼠巴贝虫 可溶性抗原 SDS-PAGE Western bolt ELISA

Abstract: The soluble antigens of Babesia microti were analyzed in this study to screen effective antigen components for immunologic diagnosis. BALB/c mice were infected with Babesia microti (B. microti) and then polypide of B. microti was collected when parasitemia reached the peak. The soluble Babesia antigens (SBA) were prepared by Ultrasonic method. ELISA with SBA was applied to detect specific IgG in sera from B. microti-infected BALB/c mice and to evaluate reactogenicity and specificity of SBA. SDS-PAGE was used to analyze the component of SBA, and Western blot was used to analyze the reactivity with serum of infected mice. Resutls indicated that the established SBA-ELISA method could detect specific antibodies as early as 7 days post-infection in sera from BALB/c mice, which showed no cross reaction with positive serum of Plasmodium falciparum, Plasmodium vivax and Toxoplasma gondii, thus indicating good sensitivity and specialty. In SDS-PAGE, 5 main protein bands with molecular mass at 72, 66, 60, 53, and 43 kDa and 7 minor bands at 14.4-116 kDa were obtained. In Western blot analysis, 15 antigen components in SBA could be recognized by positive serum of mice, during which protein component at 72, 53, 43, 39, and 30 kDa showed strong reaction. The results demonstrated that SBA components of B. microti are complex, during which antigen component at 72, 53, 43, 39, and 30 kDa were ideal, while their characteristics and diagnostic effect need to be further discussed.

Keywords: Babesia microti soluble Babesia antigens SDS-PAGE Western bolt ELISA

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