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论著

免疫刺激序列增强日本血吸虫DNA疫苗的免疫保护作用

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摘要
目的 探讨免疫刺激序列在日本血吸虫*Mr* 23 000膜蛋白 (SjC23)DNA疫苗诱导BALB/c小鼠抗血吸虫感染中的作用。 方法 将SjC23基因片段克隆到增加了免疫刺激序列的真核表达质粒 pcDNA3.1-CpG中,构建pcDNA3.1-SjC23/CpG。 40只雌性BALB/c小鼠随机分为 4组 ,① pcDNA3.1对照组 ;②pcDNA3.1-SjC23组 ;③ pcDNA3.1-CpG组 ;④ pcDNA3.1-SjC23/CpG组。每鼠经两侧股四头肌注射质粒DNA共100 μg ,隔 2周加强免疫 1次 ,共 3次。末次免疫后 4周经腹部皮肤感染日本血吸虫尾蚴 45条 /鼠 ,45d后计数成虫及肝脏虫卵数。首次免疫前和感染前 2d分别经尾静脉采血 ,检测IgG及IgG1、IgG2a。末次免疫后 3周取小鼠脾细胞 ,检测经伴刀豆球蛋白和SjC23重组蛋白刺激后小鼠白细胞介素 2 (IL-2)、白细胞介素 4(IL-4)和γ干扰素 (IFN-γ)。用51Cr释放法检测经SjC23重组蛋白刺激后脾细胞对小鼠淋巴瘤细胞的杀伤作用。 结果 ②组和④组减虫率分别为 2 8.1%和 3 5.1% ,减卵率分别为 2 1.6%和 2 6.5 %。④组减虫率显著高于②组 (*P* <0.0 5)。这两组均检测到特异性IgG ,IgG2a/IgG1比值分别为 10.1和 12.2。脾细胞经伴刀豆球蛋白和SjC23重组蛋白刺激后的IL-2水平 ,②组较①组、④组较③组均有升高。②组脾细胞对靶细胞的杀伤活性为9.7%, ④组为40.0%。 结论 疫苗载体中增加免疫刺激序列，可提高SjC23 DNA 疫苗在BALB/c小鼠中诱导产生的免疫保护作用。

关键词 日本血吸虫 免疫刺激序列 *Mr* 23000膜蛋白 重组抗原 DNA疫苗

分类号

Enhancement of the Protective Effect of SjC23 DNA Vaccine against *Schistosoma japonicum* Infection by Immunostimulatory Sequence

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Abstract
Objective To investigate the effect of immunostimulatory sequence on SjC23 DNA vaccine against *Schistosoma japonicum* infection. Methods SjC23 gene fragment was inserted into pcDNA3.1-CpG to construct pcDNA3.1-SjC23/CpG. BALB/c mice in 4 groups were immunized intramuscularly 3 times at 2 week intervals, with 100 μg plasmid DNA per injection. Four weeks after the 3rd immunization, all mice were challenged with 45±1 cercariae of *S.japonicum* by abdominal skin penetration. After 45 days post-challenge, mice were perfused and the number of recovered worms and of eggs in liver was counted. Blood samples were collected from the tail vein of all mice 2 days before the 1st immunization and before challenge respectively. IgG, IgG1 and IgG2a in sera were detected. Three weeks after the 3rd inoculation, the spleen cells of 2 mice from each group were cultured and stimulated with ConA and recombinant peptide. The supernatant was collected to detect IL-2, IL-4 and IFN-γ. Simultaneously, the cytotoxic activity was detected with 51 Cr release assay in vitro. Results The worm reduction rate in SjC23 group and SjC23/CpG group was 28.1% and 35.1%, the hepatic egg reduction rate was 21.6% and 26.5%, respectively, compared with the control group. The level of protection in SjC23/CpG group was higher than that in SjC23 group (P<0.05). ELISA results indicated that mice immunized with pcDNA3.1-SjC23 and SjC23/CpG produced specific IgG to rSjC23, while mice immunized with pcDNA3.1 and pcDNA3.1-CpG did not. Mice in SjC23 group and SjC23/CpG group also produced IgG1 and IgG2a antibody isotypes, with the ratio of IgG2a/IgG1 10.1 and 12.2, respectively. In comparison with the control, the level of IL-2 and IFN-γ in mice immunized with pcDNA3.1-SjC23 and pcDNA3.1-SjC23/CpG was augmented. The cytotoxic activity of spleen cells from mice in SjC23/CpG group was augmented from 9.7% to 40.0% compared with that in SjC23 group. Conclusion The study indicates that immunostimulatory sequence appears to increase the level of protection induced by immunization with pcDNA3.1-SjC23 vaccine.

Key words *Schistosoma japonicum* Immunostimulatory sequence (CpG) *Mr* 23 000 membrane protein Recombinant antigen DNA vaccine

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