IMMUNOGENICITY AND VACCINE EFFICACY OF A MULTIEPITOPE ANTIGEN DESIGNED FROM *Leishmania infantum* EXOANTIGENS

KARINE FERREIRA LOPES[1], MARIANA LOURENÇO FREIRE[1], ROSIANE APARECIDA DA SILVA-PEREIRA[1], DANIELA DE MELO RESENDE[1], DANIEL MOREIRA DE AVELAR[1], MÁRCIO SOBREIRA SILVA ARAÚJO[1], VANESSA PERUHYPE-MAGALHÃES[1], JULIANA MARTINS RIBEIRO[1], DIRCE RIBEIRO DE OLIVEIRA[2], ELIANE DE MORAIS TEIXEIRA[1], SILVANE MARIA FONSECA MURTA[1], EDWARD JOSÉ DE OLIVEIRA[1]

[1]INSTITUTO RENÉ RACHOU, MINAS GERAIS, BRAZIL, [2]UNIVERSIDADE FEDERAL DE JUÍZ DE FORA, GOVERNADOR VALADARES, MINAS GERAIS, BRAZIL

In Brazil, visceral leishmaniasis (VL) is caused by *Leishmania* (*Leishmania*) *infantum* and primarily controlled through the euthanasia of seropositive dogs, a strategy with limited efficacy. In this context, prophylactic vaccines represent a promising alternative for disease control. This study evaluated the immunogenicity and vaccine efficacy of a multiepitope antigen, PQ-4, composed of sequences from main exoantigens (Exo-Ags) secreted by *L. infantum*. Exo-Ags were obtained from promastigotes and analyzed using two-dimensional electrophoresis and Western blotting with sera pools from mice immunized with Exo-Ags, patients with VL, control mice and healthy individuals. Immunoreactive proteins were identified by mass spectrometry, and their amino acid sequences were subjected to epitope prediction. Selected epitopes, combined with specific spacers, were used to design PQ-4, which was produced in *Escherichia coli*. BALB/c mice were immunized three times with PQ-4, with or without saponin, challenged with *L. infantum*, and evaluated 42 days post-challenge. PQ-4 was immunogenic, both alone and with saponin, inducing increased frequencies of naïve and effector $CD4^+$ and $CD8^+$ T lymphocytes, as well as total IgG. Afterward the experimental challenge, an increase in effector CD4+ T lymphocytes, effector memory $CD4^+$ and $CD8^+$ T lymphocytes, naïve $CD8^+$ T lymphocytes, and memory B lymphocytes was observed, along with elevated plasma interleukin-6 levels. Furthermore, the splenic parasite burden was reduced by 54.3% and hepatic burden by 37%, with no significant differences between PQ-4 alone or combined with saponin. These findings emphasize the potential of exoantigens and bioinformatics tools for the rational development of vaccines against VL, highlighting the efficacy of PQ-4 in inducing robust immune responses and reducing parasite burden in an experimental model.

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