A vaccine composed by the association between a hypothetical protein and eukaryotic initiation factor 5a from *Leishmania braziliensis* protects BALB/c mice against *Leishmania amazonensis* infection

Thaís T. O. Santos[1], Mariana C. Duarte[2], Vívian T. Martins[3], Daniela P. Lage[1], Lourena E. Costa[1], Letícia M. R. Lage[2], Ana Maria R. S. Carvalho[2], Fernanda R. Ludolf[1], Bruno M. Roatt[2], Daniel Menezes-Souza[2], Carlos A. P. Tavares[3], Eduardo A. F. Coelho[1,2,*]

[1] Programa de Pós-Graduação em Ciências da Saúde: Infectologia e Medicina Tropical, Faculdade de Medicina, Universidade Federal de Minas Gerais, Av. Prof. Alfredo Balena, 190, 30130-100, Belo Horizonte, Minas Gerais, Brazil.

[2] Departamento de Patologia Clínica, COLTEC, Universidade Federal de Minas Gerais, Av. Antônio Carlos, 6627, 31270-901, Pampulha, Belo Horizonte, Minas Gerais, Brazil.

[3] Departamento de Bioquímica e Imunologia, Instituto de Ciências Biológicas, Universidade Federal de Minas Gerais, Av. Antônio Carlos, 6627, 31270-901, Pampulha, Belo Horizonte, Minas Gerais, Brazil.

SUMMARY

In the present study, two *Leishmania braziliensis* proteins, one hypothetical (LbHyp) and the eukaryotic initiation factor 5a (EiF5a), were evaluated for the protection of BALB/c mice against *L. amazonensis*. Animals were immunized with the antigens separately or in association, in both cases using saponin as an adjuvant. Spleen cells from vaccinated mice and later challenged produced significantly higher levels of protein- and parasite-specific IFN-γ, IL-12, and GM-CSF, besides of low levels of IL-4 and IL-10. Evaluating the parasite load both by a limiting dilution assay and by RT-PCR, these animals presented significant reductions in their parasite number in the infected tissue and evaluated organs, as compared to the saline and saponin groups. Also, vaccinated animals showed lower footpad swellings when compared to the control groups. The best results in inhibiting the infection were reached when the polyproteins vaccine was administered in the animals. Protection was associated with an IFN-γ production against parasite extracts, which was mediated by both $CD4^+$ and $CD8^+$ T cells and correlated with an antileishmanial nitrite production. In this context, the polyproteins vaccine combining two *L. braziliensis* proteins could be evaluated in the protection against other *Leishmania* species, as well as in other mammalian hosts.

KEYWORDS: Hypothetical proteins; eukaryotic initiation factor 5a; heterologous protection; vaccine; tegumentary leishmaniasis; Th1 immune response.

FINANCIAL SUPPORT

This work was supported by grants from Instituto Nacional de Ciência e Tecnologia em Nano-biofarmacêutica (INCT-NanoBiofar), FAPEMIG (CBB-APQ-00819-12 and CBB-APQ-01778-2014), and CNPq (APQ-482976/2012-8, APQ-488237/2013-0, and APQ-467640/2014-9). EAFC is a grant recipient of CNPq.