Phage display technology using human peripheral blood mononuclear cells (PBMC) and murine splenocytes applied to select new mimotopes-based vaccine candidates against visceral leishmaniasis

Fernanda F. Ramos[1]; Lourena E. Costa[1]; Beatriz C.S. Salles[1]; Patrícia T. Alves[2]; Thaís T.O. Santos[1]; Mariana P. Lima[1]; Vívian T. Martins[1]; Daniela P. Lage[1]; Miguel A. Chávez-Fumagalli[1]; Daniel S. Dias[1]; Patrícia A.F. Ribeiro[1]; Daniele L. Vale[3]; Grasiele S.V. Tavares[1]; Débora V.C. Mendonça[1]; Antônio L.T. Júnior[1]; Luiz R. Goulart[2]; Eduardo A.F. Coelho[1,3]

[1]Programa de Pós-Graduação em Ciências da Saúde: Infectologia e Medicina Tropical, Faculdade de Medicina. [2]Instituto de Genética e Bioquímica, UFU, Brazil. [3]Departamento de Patologia Clínica, COLTEC, UFMG, Brazil.

The development of prophylactic strategies to prevent visceral leishmaniasis (VL) has become a priority. The present study used a phage display technology based on the selection of epitope-based immunogens, represented by phage-fused peptides that mimic *Leishmania infantum* antigens, to identify new vaccine candidates to be applied against VL. The study identified six bacteriophages clones expressing target peptides, which were immunogenic in two mammalian models, and able to induce the development of a Th1 immune response. These phages induced high production of IFN-gamma both in splenocytes derived from *L. infantum*-infected BALB/c mice and in peripheral blood mononuclear cells obtained from healthy subjects, which were associated with low production of IL-4 and IL-10 cytokines. Then, two clones were selected based on higher IFN-gamma/IL-10 ratio, and they were used to immunize naive BALB/c mice. For this, animals (n=8, per group) were subcutaneously vaccinated with the individual phages (B1 and D11 clones) and, thirty days after the last immunization, they were challenged with stationary promastigotes of *L. infantum*. Regarding immunogenicity, both immunogens were able to induce a high and specific production of IFN-, IL-12, and GM-CSF after *in vitro* stimulation with individual clones or *L. infantum* extract. Additionally, these animals, when compared to control groups (saline, saponin and wild-type phage), showed higher levels of anti-phage IgG2a isotype antibody, when compared with the IgG1 levels. Animals are being followed at this time, and at the end of 45 days after infection, they will be euthanized and the parasite load in different organs by a limiting dilution assay and RT-PCR, as well as the immune response will be evaluated, in order to determine the possible protective efficacy of the phage clones against VL.

Keywords: Phage display; mimotopes; vaccine; spleen cells; visceral leishmaniasis; immune response.

Financial Support: Programa de Pós-Graduação em Ciências da Saúde: Infectologia e Medicina Tropical, Faculdade de Medicina, UFMG. FAPEMIG. CAPES. CNPq.