Serodiagnosis of human tegumentary leishmaniasis using mimotopes-based antigens selected by phage display by a non-described subtractive technology

Lourena E. Costa[1]; Beatriz C.S. Salles[1]; Patrícia T. Alves[2]; Ana C.S. Dias[2]; Emília R. Vaz[2]; Thaís T.O. Santos[1]; Fernanda F. Ramos[1]; Mariana P. Lima[1]; Daniela P. Lage[1]; Miguel A. Chávez-Fumagalli[1]; Vívian T. Martins[1]; Danniele L. Vale[3]; Daniel S. Dias[1]; Patrícia A.F. Ribeiro[1]; Grasiele S.V. Tavares[1]; Débora V.C. Mendonça[1]; Denise U. Gonçalves[1], Manoel O.C. Rocha[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.

Human tegumentary leishmaniasis (HTL), characterized by skin ulcers that may spread and cause dreadful and massive tissue destruction of the nose and mouth, is considered a neglected tropical disease, and it is a serious threat to global health due to its continuous expansion, favored by the lifecycle of its causative organism that is maintained in domestic animal reservoirs and anthropophilic sand fly species. Serodiagnosis of HTL is a great challenge due to many biological factors, including hampered specificity and/or sensitivity. This investigation addresses the unmet need for new diagnostic markers of HTL, and describes a simple platform to improve the serodiagnosis. A constrained conformational phage display random peptide library combined with a magnetic microsphere-based subtraction strategy was used to identify ligands with potential diagnostic applications. Six clones were selected against IgG antibodies from HTL patients, characterized by sequencing and confirmed by a phage-ELISA using sera from patients developing visceral leishmaniasis (n=20), Chagas disease (n=10), mucosal (n=30) and cutaneous (n=20) leishmaniasis; as well as from healthy subjects living in endemic (n=20) and non-endemic (n=30) areas of leishmaniasis. A wild-type M13-phage clone and a soluble *Leishmania* antigenic extract were used as negative and positive controls, respectively. Three clones reached 100% sensitivity and specificity, without any cross-reactivity with sera from patients with leishmaniasis-related diseases. Briefly, we describe for the first time a set of serological markers based on three immunodominant mimotopes that showed 100% accuracy, and that could be used in a phage-ELISA assay for the HTL serodiagnosis.

Keywords: Phage display; mimotopes; serodiagnosis; tegumentary leishmaniasis; ELISA.

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