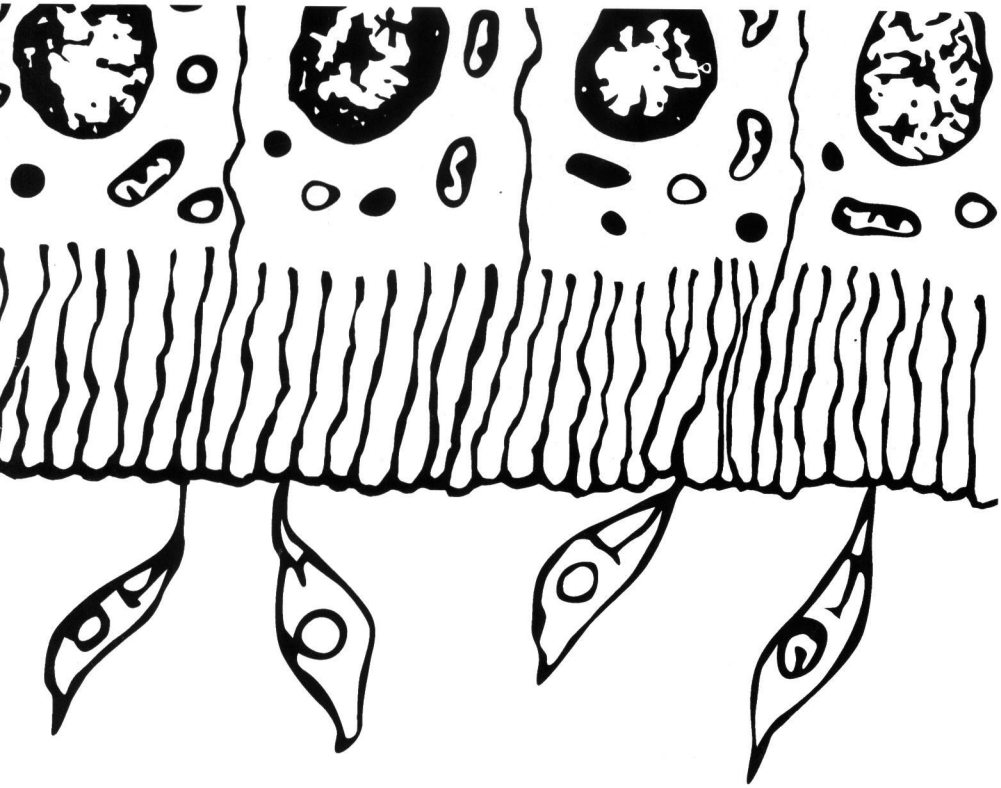

CYTED-RIMLEV

WORKSHOP ON CANINE VISCERAL LEISHMANIASIS

CPqGM-FIOCRUZ (SALVADOR, BA, BRAZIL)

MARCH, 28-30,2012

Performance of recombinant protein-based immunoassays in detecting symptomatic and asymptomatic *Leishmania infantum* visceral infections in dogs

Grimaldi JrG[a], Teva A[b], Ferreira AL[c], Santos CB[c], de-Souza Pinto I[c], de-Azevedo CT[b], Falqueto A[c]

[a]Fundação Oswaldo Cruz, Centro de Pesquisas Gonçalo Moniz, Laboratório de Patologia e Biointervenção, Salvador, Brazil; [b]Instituto Oswaldo Cruz/Fiocruz, Av. Brasil 4365, Rio de Janeiro, RJ 21040-360, Brazil; [c]Centro de Ciências da Saúde, Universidade Federal do Espírito Santo (UFES), Av. Marechal Campos 1468, Vitória, ES 29043-900, Brazil.

Canine visceral leishmaniasis (CVL) is the major source of human VL and is transmitted from dogs to sand flies to humans. To control the spread of this disease, early and accurate detection of infectious dogs is critical but challenging. The diagnosis of CVL remains difficult in rural endemic areas and serological methods still need assessment, as they are not very sensitive to detect early-infected dogs. Previously, we have shown that the recombinant(r) K26 and K39 antigens from *L. infantum* used in ELISAs provide very high sensitivities for the detection of symptomatic dogs (94% and 100%, respectively), followed by the crude soluble antigen, CSA (88%) and the rA2 protein from *L. donovani* (70%). Conversely, rA2 was more sensitive for asymptomatic dogs (88%) than rK39 and rK26 (both 66%) and CSA (30%). Some cross reactivity in sera from dogs with other infections (*L. braziliensis* and *Leptospira interrogans*) was identified, but the rA2 protein provided the greatest specificity (98%). Data further indicate that all three recombinant proteins must be used in parallel to detect essentially all parasite-positive dogs (Clin Vaccine Immunol 2007, 14:544-548). More recently, we reported a study (Trans R Soc Trop Med Hyg 2012, 106:54-59) in which we demonstrated the potential of the Dual-Path Platform (DPP®) CVL rapid test for detecting K26/K39-reactive antibodies in sera from clinically symptomatic ($n = 60$) and asymptomatic ($n = 60$) *L. infantum*-infected dogs. For the specificity evaluation, assays were performed using known negative diagnostic serum samples ($n = 59$) and cross-reaction control sera ($n = 11$) from animals born in a VL-free area of Brazil. The diagnostic kit displayed high specificity (96%) but low sensitivity (47%) in identifying parasite-positive dogs without signs of CVL. However, the test sensitivity was significantly higher (98%) in diseased cases, indicating that this convenient test may be useful to identify the most infectious dogs. Efforts should be pursued to obtain a more sensitive DPP-multiplexed test parameter (i.e. based on simultaneous yet separate antibody detection of carefully selected multiple antigens of diagnostic utility) for effective serodiagnosis of early-infected dogs, as this will likely allow more
efficient canine removal regimens than those used in practice by public health services.

Funding: Fiocruz and the National Council for Scientific and Technological Development of the Ministry of Science and Technology (the PRONEX 3/CNPq-66.1037/1998-3 and the INCT/CNPq-420067/2005-1), Brazil.