Feline Visceral Leishmaniasis: Evaluation of a heterologous Protein-Based ELISA for Diagnosis

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Visceral leishmaniasis, caused by the protozoan *L. infantum*, is a zoonotic disease in which canines serve as the primary reservoirs. Consequently, most preventive and therapeutic measures target canine populations to reduce human infection risks. However, emerging evidence suggests that felines may also act as reservoirs, contributing to the persistence of the parasite in endemic regions. Despite this concern, feline leishmaniasis (FeL) remains underdiagnosed, underscoring the need for enhanced surveillance, prevention, and diagnostic approaches. Developing sensitive and specific diagnostic tests is crucial for improving FeL detection and refining control strategies. This study evaluated the performance of an enzyme-linked immunosorbent assay (ELISA) based on a heterologous protein (Protein D) for diagnosing FeL. The protein used in this study was obtained through bioinformatics analyses, with the sequence optimized by *in silico* tools. The epitopes composing the protein are derived from *L. infantum*. The sequence was inserted into a plasmid, which was produced by a commercial company. This plasmid containing the sequence was inserted into the *E. coli* Shuffle strain, responsible for protein expression. Serum samples from 16 cats in a community shelter in Aracaju-SE, Brazil, were analyzed. Results were compared with quantitative polymerase chain reaction (qPCR) and ELISA using crude *L. infantum* extract. The ELISA-Protein D assay successfully identified 100% (13/13) of positive cases detected by qPCR and crude extract ELISA. In the negative samples, its performance was similar to the crude extract ELISA, detecting 66.7% (2/3) of the negative cases. These findings suggest that the ELISA-Protein D assay has high sensitivity and potential as a valuable tool for FeL screening. However, additional studies involving a larger and more geographically diverse sample are needed to refine the assay's specificity, minimize false positives, and improve overall diagnostic accuracy.

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