LAMP-BASED *Leishmania* DNA DETECTION TO DIFFERENTIATE BETWEEN *L. braziliensis* AND *L. guyanensis* FOR THE FAST DIAGNOSTIC OF CUTANEOUS LEISHMANIASIS

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Abstract

Differential diagnosis is crucial in cutaneous leishmaniasis (CL) due to its resemblance to diseases such as sporotrichosis, lupus, and skin cancer, especially in overlapping endemic areas. Additionally, treatment outcome varies between *L. guyanensis* and *L. braziliensis*, considering their differences in therapeutic response and the risk of progression to more severe forms. Loop-mediated isothermal amplification (LAMP) is a fast and straightforward method for DNA amplification under constant temperature conditions, eliminating the need for complex equipment. Its simplicity makes it a valuable tool for the molecular diagnosis of leishmaniasis. In this study, we designed specific primers to differentiate *L. braziliensis* and *L. guyanensis*. These primers are a component of LeishID, a LAMP-based molecular diagnostic solution developed by our group, which demonstrated promising results with high sensitivity and specificity at clinically relevant concentrations. We selected species-specific targets from 26 publicly available *Leishmania* genomes, representing at least 16 different species. The accessory genome was filtered to identify unique sequences for each species. LAMP primers were then designed, and the LAMP assay was conducted using the WarmStart Colorimetric LAMP mix (NEB). The primer set demonstrated a detection limit of 0.1 pg and was able to accurately differentiate the DNA of *L. guyanensis* from *L. braziliensis* in 30 min., with no cross-reactivity observed with other trypanosomatid species, such as *T. cruzi*, *Crithidia*, *Endotripanum*, and *Herpetomonas*, nor with mammalian DNA. The primer set showed promising performance with high specificity in differentiating between *L. braziliensis* and *L. guyanensis*. This is a valuable tool for the accurate molecular diagnosis of cutaneous and mucocutaneous leishmaniasis

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