

## African Trypanosomiasis: Detection using Loop-mediated Isothermal Amplification (LAMP) of DNA

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Loop-mediated isothermal amplification (LAMP) of DNA is a novel technique that amplifies target DNA under isothermal conditions. In the present study, LAMP tests to detect sub genus *Trypanozoon*, *Trypanosoma brucei rhodesiense* and *Trypanosoma brucei gambiense* were designed from the repetitive insertion mobile element (RIME LAMP), serum resistance associated gene (SRA LAMP) and the *T.b. gambiense*-specific glycoprotein (TgsGP LAMP) respectively. The assays were performed at 62-63°C for 1 hour in a Rotor Gene 3000 and/or water bath using six primers that recognised eight targets. The template used was 1µl of pure trypanosome DNA [100 pg] and 2µl of supernatant from heat-treated blood samples. The assays were specific and showed lower detection limits than conventional PCR targeting the same genes. The resulting amplicons were detected in real time by fluorescence of SYTO-9 dye, visual observation after addition of SYBR Green I, and after gel electrophoresis. The expected LAMP product was confirmed by specific restriction enzyme digestion, melt curves and sequence analyses. The tests were reproduced in endemic country laboratories, and could be applied on different body fluids. Our studies show that SYTO-9 dye and real time monitoring are amiable to LAMP studies. Further, these RIME, SRA and TgsGP LAMP tests are simple to perform, efficient and specific, and therefore have great potential for the diagnosis of human African trypanosomiasis in endemic regions.