

Development of a Recombinase Polymerase Amplification (RPA) for the detection of *Schistosoma mansoni* infection

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Schistosomiasis is a parasitic disease associated to poverty and low sanitation condition. It is estimated that nearly 240 million people are infected in the world and over 25 million people live in high-risk areas in the Americas. Almost 1.6 million people are infected in Brazil, allopatric for *Schistosoma mansoni*, that causes the intestinal schistosomiasis and is transmitted by freshwater snails from the genus *Biomphalaria*. Among the strategies currently available, sensitive, and specific diagnostic tests followed by the timely treatment of the population, is a strategy for the prevention of potential complications together with reducing transmission. Also, accurate mapping and monitoring of snail breeding sites to detect active transmission areas are needed. The Recombinase Polymerase Amplification (RPA) is an isothermal method that has been piloted for urogenital schistosomiasis, and its simplicity, low resource needs, and speed have highlighted its utility for use in the field at the point-of-care. This research aims to evaluate the performance of RPA to support the diagnosis of infection with *S. mansoni* in humans and snails. For that, primers and probe targeting a repeated region of *S. mansoni* mitochondrial DNA have been designed and are being tested for the standardisation of the assay. Each RPA reaction is being performed using half volume of the rehydrated pellet from the TwistAmp exo kits, with fluorescent signal detection via a portable reader. The specificity of the assay was accessed using gDNA of *Schistosoma* species, and other helminths that are co-endemic with *S. mansoni*. The analytical sensitivity was determined using serial dilutions of *S. mansoni* gDNA. The use of fresh and frozen crude *S. mansoni* eggs was also tested as well as urine and stool samples spiked with gDNA and eggs, respectively, and infected and noninfected *Biomphalaria* snails. The developed assay, named SmMIT-RPA, presented promising results being specific to *S. mansoni*, sensitive enough to detect a single egg and up to 1fg of DNA. Positive results were also obtained from urine samples spiked with 0.01pg of *S. mansoni* DNA, stool samples spiked with *S. mansoni* eggs, and prepatent experimentally infected *Biomphalaria* snails. Further analysis will be conducted in order to optimise and validate the use of the SmMIT-RPA for *S. mansoni* diagnosis using clinical and field samples collected in low-endemic areas of the state of Minas Gerais (MG), Brazil. Results from this work will be later compared to data from reference tests previously applied to the same samples. It is expected that SmMIT-RPA will allow a more accurate and rapid diagnostic in order to improve the decision-making process for a more appropriate destination of public funding aiming the elimination of schistosomiasis as a public health problem.

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