Designing primers for loop-mediated isothermal amplification (LAMP) for detection of Ganoderma boninense ABSTRACT

Basal Stem Rot (BSR) caused by Ganoderma boninense is one of the most serious diseases of South East Asia's oil palm industry. The losses due to this disease were reported up to RM 1.5 billion a year in Malaysia. Typical methodologies currently used for detection of BSR infection, usually involved visual observation followed by detection of the pathogen using invasive and/or time consuming and expensive instruments. This includes the use of molecular DNA based technique, Ganoderma Selective Media (GSM), molecular techniques. However, most of these methods cannot be performed in situ. Samples need to be sent to the laboratory for testing. In this paper, a diagnostic tool using loop-mediated isothermal reaction (LAMP) is presented for detection of G. boninense. LAMP reaction which consist of a set of four primers, two outer and two inner, was designed specifi cally to recognize the manganese superoxide gene (MnSOD) obtained from NCBI Genbank (Accession no: U56128) of G. boninense, the causal pathogen of BSR. The assay was conducted in the thermal block with temperature 65°C for 50 min and the LAMP products were viewed on agarose gel electrophoresis. This technique removes the need to perform the reaction in thermal cycler as it can be done in a heat block. Results show the ladder-like pattern of bands sizes from 683 bp specifi cally to the gene MnSOD was amplifi ed. Thus, the chosen set of primers can be used for detection of G. boninense in oil palm estates subjected to sensitivity and specifi city.