Determination of reference genes for normalisation of gene expression study of Ganoderma-infected oil palms

ABSTRACT

Basal stem rot (BSR) disease is a major threat to oil palm industry in Malaysia, caused by pathogenic fungus, Ganoderma boninense. Quantitative real-time polymerase chain reaction (qPCR) has become a favourable method in quantification the levels of gene expression involved in disease development. In this study, 14 oil palm reference genes were tested for their suitability as reference genes for qPCR analyses using oil palm root taken from six-month old seedlings (nursery sample) and 15-year old mature palms (field sample). Only six reference genes were subjected to stability test via RefFinder. The GRAS and ACTIN genes were ranked as the best reference genes for nursery sample, whereas, GAPDH and GvHK genes for field samples. These reference genes were used in the qPCR analysis for accurate normalisation. Thus, results obtained in this study emphasise the importance of validating the stability of the reference gene and proving the credibility and reliability of RefFinder in determining the most stable reference genes in each specific experiment or biological setting used.

Keyword: Ganoderma boninense; Normalisation; Oil palm (Elaeis guineensis Jacq.); Quantitative real-time PCR (qPCR); Reference genes; RefFinder