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Detection of powdery scab pathogen (*Spongospora subterranea* f. sp. *Subterranea*) in potato using RT-qPCR

K.P.L. Arosha^{1*}, K. Pakeerthan¹, C. Ranasinghe²

¹Faculty of Agriculture, University of Jaffna, 44000, Sri Lanka ²Agriculture Research Station, Department of Agriculture, Sita-Eliya, 22200, Sri Lanka

*lakminiarosha191@gmail.com

A protozoan pathogen named Spongospora subterranea f. sp. subterranea, cause powdery scab in potato (Solanum tuberosum). It is one of the major problems for potato production in Nuawara Eliya, Bandarawela region of Sri Lanka. This parasite infects underground parts of the crop, roots, stems, tubers and stolon and lesions on the tubers by this pathogen diminish the quality and marketability; carry infection to subsequent crops when the potatoes are used as seed and the pathogen acts as the vector of the potato mop-top furovirus disease. Real time q-PCR could be used as a reliable tool for the detection and quantification of this pathogen. The Applied BiosystemsTM Analysis Software was used to calculate presence/absence either by analyzing the change in normalized fluorescence before and/or after the PCR (the Rn/ Δ Rn method) and by comparing threshold cycle values generated from the amplification data (the CT/CRT method). Two primer sets were used: Reverse primer SsTQR 1 5/ - CGG GCG TCA CCC TTC A -3/ Forward primer SsTQF 1 5/ -CCG GCA GAC CCA AAA CC -3/ primer pair from literature and Left primer -5/ - GCG AAT TGC AGA ATT CAG TG -3/, Right primer - 5/- CCG GGT TGG ATA ATC TTT CA -3/ designed primer pair. Perfect sigmoidal curves were obtained from the designed primer pair which indicates a healthy, good RT q-PCR reaction. Therefore, the primers used for the analysis have the possibility to detect powdery scab pathogens in the samples. Though there are no effective management practices for this unculturable, quarantine pathogen, these designed primers can be used in RT-qPCR to detect the pathogen and it will help to avoid the losses in potato production in Sri Lanka.

Keywords: Primer, quantification, real time q- PCR, Solanum tuberosum, threshold cycle