

OPTIMIZING POLYMERASE CHAIN REACTION (PCR) CONDITIONS FOR DETECTION OF WELIGAMA COCONUT LEAF WILT DISEASE-PHYTOPLASMA IN *PROUTISTA MOESTA*

G.L.B.E. Gunathilaka¹, N. Aratchige² and D.M. De Costa^{1*}

¹*Department of Agricultural Biology, Faculty of Agriculture, University of Peradeniya, Sri Lanka*

²*Crop Protection Division, Coconut Research Institute, Lunuwila, Sri Lanka*
**devikacos@yahoo.com*

Weligama coconut leaf wilt disease (WCLWD) was first reported from Weligama in Matara district of Sri Lanka in 2006. It is a phytoplasma disease and therefore, is expected to be transmitted by phloem feeding insect vectors. *Proutisa moesta* has been identified as a putative vector of WCLWD. Polymerase Chain Reaction (PCR) conditions to detect phytoplasma in *P. moesta* has not been optimized yet. Therefore, experiments were conducted to compare two methods of DNA extraction and to determine the minimum number of *P. moesta* needed for detection of WCLWD phytoplasma in the insect body. *P. moesta* were collected from field cages at Weligama (viruleferous insects) and Bandirippuwa estate, Lunuwila (aviruleferous insects). Two methods were used to extract DNA and out of which CTAB extraction method yielded higher concentration and high quality DNA than that from the modified CTAB extraction method (which has an overnight incubation). DNA extracted from 15 viruleferous *P. moesta* per sample yielded a multiplication with lower cycle threshold values than 15 aviruleferous *P. moesta* per sample in real time PCR using primers and probes specially designed for WCLWD. No amplification was observed when the samples were subjected to normal PCR using the primers PC399 and P1694 which are specific for phytoplasma detection.

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