

Sub-group Level Identification of Phytoplasma Strain Associated With Sugarcane White Leaf Disease in Sri Lanka

Y.A.P.K Dayasena^{1*}, P Panda², A.N.W.S Thushari¹ and G.P Rao³

¹Division of Crop Protection, Sugarcane Research Institute, UdaWalawa- Sri Lanka. ²Discipline of Lifesciences, Indira Gandhi National Open University, New Delhi- 110068, India.

³Division of Plant Pathology, Indian Agricultural Research Institute, New Delhi -110012, India.

Corresponding author: prasannaup@hotmail.com

ABSTRACT

Sugarcane white leaf disease (SCWLD) is one of the major sugarcane diseases in the Asian continent caused by phytoplasmas. SCWLD occurred in all sugarcane growing areas in Sri Lanka and causing severe economic losses. But, so far no finer taxonomic classification of SCWLD phytoplasma strain in Sri Lanka has been attempted. Therefore, the objective of the present study was to identify the group and sub-group of the sugarcane white leaf phytoplasma (SCWLP) strain in Sri Lanka and also to identify whether weed grasses as alternative host of SCWLP responsible for natural transmission of the disease. DNA was extracted from forty SCWLD infected sugarcane plants representing all major sugarcane growing varieties and areas of Sri Lanka and symptomatic weeds showing white leaf (*Urochloa distachya* and *Cynodon dactylon*) in sugarcane fields. PCR assays were performed using universal primer pair P1/P7 followed by nested primer R16F2n/R2. Amplicons of ~1.2 kb were consistently amplified from all the symptomatic samples of sugarcane plants and weeds in nested PCR assay. However, no amplification was observed with any of the asymptomatic sugarcane and weed samples. Pair wise sequence comparison and phylogenetic relationships of 16S rDNA sequences of sugarcane white leaf phytoplasma (SCWLP) strain revealed 99% sequence identity of SCWLP isolates with '*Candidatus Phytoplasma oryzae*'. However, the 16S rDNA sequences of the weeds (*U. distachya* and *C. dactylon*) phytoplasma isolates showed 99% sequence identity with '*Candidatus Phytoplasma cynodontis*' strains. Further virtual RFLP analysis with *iPhyClassifier* tool using 17 restriction enzymes allowed affiliation of the SCWLP strains with 16SrXI-B subgroup and in weeds phytoplasma strain with 16SrXIV-A subgroup with a similarity coefficient of 1.0. Based on the sequence comparisons results of the *16S rRNA* gene, SCWLP strains were identified as *Ca. P. oryzae* (16SrXI-B sub-group). However, the identified phytoplasma strain from weeds in the present

study could not match with SCWLP phytoplasma strains, indicating that they were not real natural reservoir of SCWLP in Sri Lanka. Further studies on epidemiology of SCWLD along with identification of natural reservoirs is in progress.

Key Words: Phytoplasma Strain, 16S rRNA Sene, Sri Lanka , Sugarcane White Leaf Disease