First report of “*Candidatus Phytoplasma oryzae*” associated with lemongrass in India

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Cymbopogon citratus (DC. ex Nees) Stapf commonly called lemongrass is a species of Poaceae family. It is cultivated as culinary and medicinal herbs because of its lemon like scent and antiseptic chemical constituents. During survey of botanical garden at DDU Gorakhpur University Campus, Gorakhpur, Uttar Pradesh, India in August 2017, *C. citratus* plants showed little leaves and chlorosis symptoms with an incidence of 40%. Total DNA was extracted from both symptomatic (five) and asymptomatic (two) samples with CTAB enrichment method. Ribosomal DNA (16S rDNA) of phytoplasma was amplified using primer pairs P1/P7 in first round and R16F2n/R16R2 in nested PCR assay (Deng and Hiruki 1991; Gundersen and Lee 1996). For negative control we used DNA extracted from asymptomatic lemongrass. Amplification reactions were carried out in a Eppendorf Thermal cycler, Germany and PCR amplified products were purified using the Wizard® SV Gel and PCR Clean-up System (Promega). The amplified fragments (~1.2 kb) of phytoplasmal rRNA gene obtained from the five *C. citratus* little leaf (Cll) phytoplasmas Gorakhpur (GKP) isolates were directly sequenced in both directions. They shared 100% sequence identity among them and two of them were submitted to the GenBank (Accession Nos. MT127617, MT127618). Pairwise sequence analysis of the 16S rRNA gene sequence of the Cll 1 and 2 phytoplasma isolates revealed 100% sequence identity with ‘*Candidatus*

Phytoplasma oryzae’ Sugarcane grassy shoot phytoplasma strain (MN719898). Phylogenetic analysis also confirmed that the 16S rRNA gene sequence of Cll isolates clustered together with phytoplasma strains belonging to 16SrXI group (‘*Ca. P. oryzae*’). Further virtual RFLP analysis of 16SrRNA gene sequence with online *iPhyClassifier* tool were performed using seventeen restriction endonucleases, which allowed affiliation of the Cll isolate with 16SrXI-B subgroup phytoplasma strain with similarity coefficient of 1.0. ‘*Ca. P. oryzae*’ (16SrXI-B subgroup) is one of the important phytoplasma group that infects rice, sugarcane, arecanut, vegetables and ornamentals in India. Until now, 16SrII phytoplasma group has been reported on lemongrass from Haryana, state of India (Madhupriya and Rao 2014). Hence, this is the first report of the association of ‘*Ca. P. oryzae*’ (16SrXI-B subgroup) with *C. citratus* (lemon grass) little leaf in India.

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References

- Deng S, Hiruki C (1991) Amplification of 16S rRNA genes from culturable and non-culturable mollicutes. J Microbiol Methods 14: 53–61
- Gundersen DE, Lee IM (1996) Ultrasensitive detection of phytoplasmas by nested PCR assays using two universal primer pairs. Phytopath med 35(3):144–151
- Madhupriya MR, Rao GP (2014) ‘*Candidatus Phytoplasma aurantifolia*’ related strains affecting two important medicinal plants (*Cymbopogon citratus* and *Tylophora asthmatica*) in India. Indian Phytopath 67(3):303–307

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