Diseases Caused by Bacteria and Phytoplasmas

First Report of 16SrII-V Peanut Witches' Broom Phytoplasma in Snake Gourd (*Trichosanthes cucumerina*) in Taiwan

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Funding: This work was financially supported in part by grants-in-aid from the Ministry of Science and Technology (109-2628-B-005-006) and the Advanced Plant Biotechnology Center from the Featured Areas Research Center Program within the framework of the Ministry of Education (MOE) in Taiwan's Higher Education Sprout Project. Plant Dis. 105:2236, 2021; published online as <https://doi.org/10.1094/PDIS-12-20-2666-PDN>. Accepted for publication 23 March 2021.

Snake gourd (*Trichosanthes cucumerina* L.), an annual climbing plant belonging to the family Cucurbitaceae, is native to Southeast Asian countries (e.g., India, Pakistan, Malaysia, China, and Indonesia). It is commonly consumed as a vegetable and also used as a traditional herbal medicine due to the antidiabetic, anti-inflammatory, antibacterial, hepatoprotective, and cytotoxic activities (Devi 2017). In September 2020, phytoplasma-induced disease symptoms such as little leaf, yellowing, phyllody, virescence, and witches' broom were observed on snake gourd in Yunlin County, Taiwan. The cross-sectional examination of the symptomatic plant by transmission electron microscopy showed typical phytoplasma-like pleomorphic bodies with spherical, oval, and tubular shapes in sieve elements. Further examination by nested

PCR revealed that a 1.2-kb DNA fragment for 16S rRNA gene was only amplified from symptomatic leaf of snake gourd using the phytoplasma universal primer pairs P1/P7 followed by R16F2n/R16R2. BLAST and iPhyClassifier (<https://plantpathology.ba.ars.usda.gov/cgi-bin/resource/iphyclassifier.cgi>) analyses on the amplified DNA fragment (accession no. MW309142) revealed that it shares 100% identity with that of GenBank accession NZ_AMWZ01000008 (complement 31109 to 32640) of peanut witches' broom (PnWB) phytoplasma, a '*Candidatus* Phytoplasma aurantifolia'-related strain (Firrao et al. 2004), and could be classified into the 16SrII-V subgroup. Samples examined by nested polymerase chain reaction (PCR) were further characterized by western blotting using the polyclonal antibody raised against the Imp of PnWB phytoplasma (Chien et al. 2020, 2021). An expected signal of 19 kDa specific for Imp was only detected in the symptomatic snake gourd but not in healthy snake gourd. Because the disease symptoms caused by phytoplasma infection are highly dependent on the secreted effectors (Namba 2019), *phyllogen* gene that is responsible for phyllody and virescence symptoms was amplified from symptomatic snake gourd by PCR. BLAST analysis revealed that *phyllogen* identified in snake gourd is identical with that (accession no. EMR14684) of PnWB phytoplasma. In Taiwan, species of family Cucurbitaceae such as loofah, bitter melon, and pumpkin are commonly infected by 16SrVIII phytoplasma (Davis et al. 2017). In this study, we report for the first time that snake gourd, a species of family Cucurbitaceae, was infected by 16SrII-V PnWB phytoplasma in Taiwan.

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The author(s) declare no conflict of interest.

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Keywords: *Trichosanthes cucumerina*, phytoplasma, snake gourd, peanut witches' broom disease, *phyllogen*

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