First Report of Clover Yellow Vein Virus on Orchid (*Dendrobium* sp.) in South Korea

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Orchid is one of the most popular and commercially important cultivated flowers in the world. Among many orchid species, *Dendrobium* spp. are popular cut flowers and potted plants in South Korea. In March 2019, 10 *Dendrobium* orchid plants in a greenhouse in Daegu, South Korea showed large chlorotic blotches, mosaic, and mottle symptoms. One leaf each from the 10 symptomatic orchid plants was examined by leaf dip preparations and transmission electron microscopy (JEM-1400; JEOL Inc., Tokyo, Japan) after leaf dip preparations (Brenner and Horne 1959; Richert-Pöggeler et al. 2019). Typical potyvirus-like particles of flexuous and filamentous shape and ~760 by 15 nm length/width were observed in all tested samples. The presence of potyvirus was confirmed by serological detection with a commercially available ImmunoStrip for the potyvirus group (Agdia, Elkhart, WI, U.S.A.). In contrast, a negative result was obtained for a virus-free *Dendrobium* plant by the serological test. The two most common viruses in orchids—namely, odontoglossum ringspot virus (ORSV) and cymbidium mosaic virus (CymMV)—in all *Dendrobium* samples were not detected in any samples by an ImmunoStrip for ORSV and CymMV (Agdia). To determine the species of the virus, total RNA was extracted from all 10 ImmunoStrip-positive samples using the RNeasy plant mini kit (Qiagen, Hilden, Germany). Subsequently, reverse-transcription PCR products (~1,625 bp) were amplified using potyvirus-specific primer pairs (Gibbs and Mackenzie 1997) and sequenced by the Sanger method at Macrogen Inc. (Seoul, South Korea).

Sequencing results showed 100% nucleotide identity among 10 samples. Thus, one sequence was chosen for identification of virus species using sequence comparison. BLASTn analysis showed that the nucleotide sequence and its deduced amino acid sequence of the amplicon shared 95.4 to 98.7% and 96.2 to 99.6% identity, respectively, to multiple clover yellow vein virus (CIYVV) sequences (e.g., accession no. AB011819) in GenBank. To further confirm the presence of CIYVV and determine if other viral agents were present in the samples, total RNA from 3 of the 10 symptomatic plants was depleted of ribosomal RNAs and subjected to high-throughput sequencing (HTS) analysis on a HiSeq 4000 platform (Macrogen Inc.). In total, 3,764,432, 4,203,881, and 4,139,775 150-bp paired-end clean reads were obtained for the three samples. After de novo assembly of the reads with Trinity (Haas et al. 2013), five, six, and seven contigs were obtained and searched with BLASTn against the NCBI viral refseq database. Eighteen contigs from all three samples sized at 2,176 to 9,432 nt exhibited 94.0 to 97.9% nucleotide identity with the complete genome sequences of other CIYVV isolates (e.g., accession no. AB011819) deposited in GenBank; no other viruses were identified by HTS. The complete genome sequence (9,585 nucleotides in length) of the CIYVV *Dendrobium* isolate (CIYVV-Den) was determined using CIYVV-specific primers (Uyeda et al. 1997) and the sequence of CIYVV-Den was deposited to GenBank (accession no. LC506604). Together, these results support the theory that symptomatic *Dendrobium* orchids were infected with CIYVV-Den in this study. CIYVV has been previously reported affecting *Calanthe* orchids in Japan (Ikegami et al. 1995; Inouye et al. 1988). Our results suggest that CIYVV may be detrimental to the production of *Dendrobium* orchids or commercial ornamental crops in South Korea. To our knowledge, this is the first report of CIYVV in *Dendrobium* sp. in South Korea.

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