Occurrence of Soybean Yellow Common Mosaic Virus in Soybean in China Showing Yellow Common Mosaic Disease

Z. Y. Wei, C. Jiang, C. Y. Mao, H. H. Zhang, R. P. Miao, J. P. Chen,[†] and Z. T. Sun[†]

State Key Laboratory for Managing Biotic and Chemical Threats to the Quality and Safety of Agro-products, Key Laboratory of Biotechnology in Plant Protection of MOA of China and Zhejiang Province, Institute of Plant Virology, Ningbo University, Ningbo, 315211, China

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Soybean yellow common mosaic virus (SYCMV), a positive-sense ssRNA virus classified in the genus *Sobemovirus*, was first reported and characterized in Korea (Nam et al. 2012). Currently, its only known host is soybean (Nam et al. 2012), on which it causes bright yellow mosaic and crinkling of the leaves (Lim et al. 2016). During a field survey in July 2019, bright yellow mosaic and mild crinkling symptoms were observed on soybean leaves (cv. Zhonghuang 13) in the Hubei province of China. To identify the possible pathogen(s) associated to the disease symptoms, leaves from five symptomatic plants were collected and pooled, and total RNA was extracted using TRIzol Reagent (Invitrogen, U.S.A.). A 10-μg sample of the total RNA was purified via magnetic beads (Thermo Fischer Scientific, U.S.A.), and a TruSeq RNA Sample Prep Kit (Illumina, U.S.A.) was then used to construct an RNA sequencing library. Transcriptome sequencing was performed on an Illumina HiSeq 4000 (LC Sciences, U.S.A.). The average insert size for the paired-end library was 300 ± 50 bp. After quality control, a total of 47.5 million clean reads were obtained and assembled using Trinity software (version 2.8.5). The assembled contigs were searched against NCBI virus RefSeqs (<ftp://ftp.ncbi.nlm.nih.gov/refseq/release/viral>) by the BLASTx algorithm with a cutoff E value of ≤10⁻⁵. Twelve contigs sized from 3,421 to 4,093 bp were

found to share a sequence identity of 77.5 to 94.1% with SYCMV isolates from Japan (LC332541) and South Korea (JF495127.1). No other virus matches were identified. The largest contig (4,093 bp, MT816507) covers 99% of the expected complete genome of SYCMV (4,121 bp, KX096577). To verify the accuracy of the sequence assembled, RT-PCR-Sanger sequencing was performed on a single field plant sample using primers designed for SYCMV (forward, 5'-GAACAAAGAGTCTGGATCTT-3'; reverse, 5'-TCCTTCCAAACCTCGCGGG-3'). The sequence of the amplicon (3,854 bp, MT997092) exhibited an identity of 99.9% to the high-throughput sequencing-derived SYCMV contig sequence. Phylogenetic analysis of the amplicon sequence revealed that the SYCMV isolate from China formed a distinct branch in the tree. Sap from symptomatic field plants was used to mechanically inoculate two soybean cultivars (Jiunong 9 and Kefeng 1, 10 plants per cultivar), and leaves inoculated with phosphate buffered saline (0.01 M, pH 7.5) served as a control (three plants per cultivar). All but the control plants developed systemic bright yellow mosaic symptoms 10 days after inoculation. The infection of the soybean plants with SYCMV was confirmed by RT-PCR with the newly designed primers for SYCMV (forward, 5'-CCTACAGG-CATTGGTTTCGT-3'; reverse, 5'-CGTGAGGTTCTTGCTTCACA-3'; anticipated amplicon size, 2,210 bp) and by amplicon sequencing (100% sequence identity with MT997092). In addition, the infection was further confirmed by immuno-blotting using an antibody against SYCMV coat protein (synthesized by GenScript, U.S.A.). Together, the results demonstrate that SYCMV is the causal agent of the bright yellow mosaic symptoms in soybean observed in the field. To the best of our knowledge, this is the first report of SYCMV on soybean in China. These findings shall not only alert local growers to a potential new threat to soybean production in their region but also provide new insights on the transmission, epidemiology, and pathological properties of SYCMV in China.

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[†]Indicates the corresponding authors.

E-mail: jpchen2001@126.com (J. P. C.) and tao198410@163.com (Z. T. S.)