New Disease Reports

First report of Pepper virus A naturally infecting tomato in India

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Tomato (Solanum lycopersicum) is a commercially important vegetable crop grown worldwide. In October 2018, diseased tomato plants were observed in a field from Nashik, Maharashtra state in India. The plants showed symptoms including leaf yellowing, mosaic and mottling (Fig. 1). Leaf samples from four symptomatic plants (NSK-Tom-01-04) were tested with antisera raised against Cucumber mosaic virus, Groundnut bud necrosis virus and Tobacco mosaic virus (TMV), and a broad-spectrum antiserum against Cowpea mild mottle virus (CPMMV; antiserum gifted by George Thottapalli) in direct antigen coating (DAC)-ELISA. Three samples reacted positively with the CPMMV antiserum (NSK-Tom-01, NSK-Tom-02 and NSK-Tom-03) and the fourth sample (NSK-Tom-03) reacted positively only with the TMV antiserum. Mechanical sap transmission of the CPMMV-positive leaf samples produced similar symptoms on tomato plants 8-10 days post-inoculation (Fig. 2). To confirm infection, the inoculated plants were tested using the CPMMV antiserum in DAC-ELISA and they reacted positively.

Total RNA was extracted from the four symptomatic tomato leaf samples using an RNeasy Mini Plant Kit (Qiagen Inc., Chatsworth, USA) and cDNA was synthesized using a High-Capacity cDNA Reverse Transcription Kit (Applied Biosystems) according to the manufacturer's instructions. The RNA samples were tested by RT-PCR using carlavirus (CP)-specific (CP coat protein primers For 5'-AAYKAGTGCDGACACCAAAC-3'; CP Rev 5' -TCATTDTACCCTGTDAYTTGCGT-3') designed using published sequences (GenBank Accession Nos. HQ184471, DQ444266, KC884247, AM493895, AM765838, AM039440 AJ863510, AJ863509, D14449, JN591720, KU726694). An amplicon of the expected size (c. 852 bp) was generated from isolates NSK-Tom-01, NSK-Tom-02 and NSK-Tom-03. The NSK-Tom-04 isolate was negative. The amplicon from the NSK-

Tom-01 isolate was cloned into a pGEM-T Easy vector (Promega, Madison, WI) and sequenced. BLAST analysis of the sequence obtained (MK783696) showed the highest identity for both nucleotide (83%) and amino acid sequences (99%) with a CP sequence of Pepper virus A (PepVA; KU726694 and KU923763) isolated from chilli pepper. A phylogenetic tree based on the amino acid sequences of complete CP gene of Pepper virus A and other carlaviruses constructed using MEGA X (Kumar *et al.*, 2018), revealed that the NSK-Tom-01 isolate of PepVA (MK783696) closely clustered with other isolates of PepVA (KU726694 and KU923763) (Fig. 3)

PepVA belongs to the genus *Carlavirus*, family *Betaflexiviridae*, and infects solanaceous crops (Jo *et al.*, 2018). The impact of PepVA in India and other countries on cultivated crop plants is unknown. To our knowledge, this is the first report of PepVA in tomato in India.

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References

Jo Y, Choi H, Kim SM, Kim SL, Lee BC, Cho WK, 2017. The pepper virome: natural co-infection of diverse viruses and their quasispecies. *BMC Genomics* **18**, 453. <u>http://dx.doi.org/10.1186/s12864-017-3838-8</u>

Kumar S, Stecher G, Li M, Knyaz C, Tamura K, 2018. MEGA X: molecular evolutionary genetics analysis across computing platforms. *Molecular Biology and Evolution* **35**, 1547-1549. http://dx.doi.org/10.1093/molbev/msy096



Figure 1

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