

## DISEASE NOTE

## FIRST REPORT OF *CHILLI VEINAL MOTTLE VIRUS* IN TOMATO IN PAKISTAN

A. Ahmad and M. Ashfaq

Department of Plant Pathology, PMAS-Arid Agriculture University Rawalpindi-46300, Pakistan

*Chilli veinal mottle virus* (ChiVMV, genus *Potyvirus*) infects several solanaceous species worldwide (Zhao *et al.*, 2014). In April 2014, a total of 20 tomato leaf samples with symptoms of mosaic, necrosis and mottling were collected from the research farm of PMAS-Arid Agriculture University Rawalpindi. All disease samples were screened for the presence of potyvirus infection by indirect plate-trapped antigen (PTA)-ELISA using "Poty group test" kit (Bioreba AG, Switzerland). Only 12 samples were positive for potyvirus infection. These samples were further tested for *Potato virus Y* (PVY), *Pepper veinal mottle virus* (PVMV) (Bioreba AG) and ChiVMV (Loewe Biochemicals, Germany) using virus specific DAS-ELISA. Of 12 potyvirus confirmed samples, two were positive for ChiVMV and remaining ten were positive for PVY. No reaction was observed with antibodies against PVMV. The ChiVMV ELISA-positive samples were further confirmed by RT-PCR using ChiVMV specific primers Poty3 and Oligo(dT) (Tsai *et al.*, 2008). Both PCR amplicons were purified using QIAquick® PCR purification kit (Qiagen) and subsequently sequenced in both orientations. A product 759 nucleotides in size (partial CP gene including 3'UTR) was obtained (both clones were 100% identical) and the sequence of ChiVMV isolate tomato (AARTPK) was deposited in GenBank (Accession No. KT876048). BLASTn revealed >90% sequence identity with ChiVMV isolates from India (JN624776, JN692501), China (HQ218936, KF738253, KC711055, JX088636) and Pakistan (KJ472764). This destructive virus has been previously reported from chilli pepper in Pakistan (Shah *et al.*, 2009). To the best of our knowledge, this is the first confirmed report of ChiVMV from tomato in Pakistan.

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- Shah H., Yasmin T., Fahim M., Hameed S., Haque M.I., 2009. Prevalence, occurrence and distribution of *Chilli veinal mottle virus* in Pakistan. *Pakistan Journal of Botany* **41**: 955-965.
- Tsai W.S., Huang Y.C., Zhang D.Y., Reddy K., Hidayat S.H., Srithongchai W., Green S.K., Jan F.-J., 2008. Molecular characterization of the CP gene and 3'UTR of *Chilli veinal mottle virus* from South and Southeast Asia. *Plant Pathology* **57**: 408-416.
- Zhao F.F., Xi D.H., Liu J., Deng X.G., Lin H.H., 2014. First Report of *Chilli veinal mottle virus* infecting Tomato (*Solanum lycopersicum*) in China. *Plant Disease* **98**: 1589.

Corresponding author: M. Ashfaq  
E-mail: mashfaq1642@gmail.com

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## DISEASE NOTE

## OCCURRENCE OF CITRUS YELLOW VEIN CLEARING VIRUS IN CITRUS SPECIES IN IRAN

S.M. Bani Hashmian and S. Aghajanzadeh

Citrus and Subtropical Fruits Research Center, Horticultural Science Research Institute, Agricultural Research, Education and Extension Organization, 46915-335, Ramsar, Iran

Citrus yellow vein clearing virus (CYVCV), a recently described member of the genus *Mandarivirus*, is associated with yellow vein clearing disease (CYVCD), a serious threat of lemon production of the world (Chen *et al.*, 2014). The disease was first reported on its two main hosts, sour orange (*Citrus aurantium*) and lemon (*C. limon*), from Pakistan in 1988. Then it appeared in India, Turkey and China. In 2010 characteristic symptoms of the disease including leaf crinkling, vein clearing with corresponding water-soaked appearance of the lateral veins on the lower surface of the leaves, were first noticed on a sour orange seedling and a lemon tree in Mazandaran province of Iran. A large number of samples was collected from symptomatic trees of sour orange, Eureka lemon and Persian lime (*C. latifolia*) during surveys from citrus orchards of the North of Iran in 2016. RNA extractions by SDS-potassium acetate method (Bernad and Duran-Vila, 2006) were made from one isolate from each of these three hosts. A two-step RT-PCR was performed using Revert Aid Kit and PCR Master Mix (Fermentas) and CYVCV specific primer pair of coat protein gene (Chen *et al.*, 2014). The resulting PCR products were sequenced directly, and multiple alignments revealed 96-98% nucleotide identity among the Iranian isolates (GenBank Accession No. KX902486 to KX902488) and also with the reference sequence of CYVCV (NC\_026592), two sequences reported from Pakistan and five isolates from China. To our knowledge this is the first report of CYVCV in Iran and Persian lime as a susceptible host of the virus.

- Bernad L., Duran-Vila N., 2006. A novel RT-PCR approach for detection and characterization of citrus viroids. *Molecular and Cellular Probes* **20**: 105-113.
- Chen H.M., Li Z.A., Wang X. F., Zhou Y., Tang K.Z., Zhou C.Y., Zhou X.Y., Yue J. Q., 2014. First report of Citrus yellow vein clearing virus on lemon in Yunnan, China. *Plant Disease* **98**: 1747.

Corresponding author: S. M. Bani Hashemian  
E-mail: m.banishemian@areo.ir

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