First report of Tomato blistering mosaic virus infecting tomato in Argentina.

L. Ferrand, Centro de Investigaciones de Fitopatología, Universidad Nacional de La Plata, CONICET, Argentina; C. Nome, IPAVE-CIAP-INTA, Córdoba, Argentina; A. F. Orílio, Dept. de Biología Celular, Universidade de Brasília, Brasil; M.L. García, Instituto de Biotecnología y Biología Molecular, Universidad Nacional de la Plata, CONICET, Argentina; T. Nagata, Dept. de Biología Celular, Universidade de Brasília, Brasília, Brasil; L. B. Ronco, and E. Dal Bó, Centro de Investigaciones de Fitopatología, Universidad Nacional de La Plata, CIC, Argentina.

Tomato (*Solanum lycopersi*cum) ranks as the most important crop among greenhouseplanted crops in terms of cultivated areas (17,800 ha) and production in Argentina.

In September 2014, farmers reported that tomato plants of cv. Elpida (Enza Zaden) showed mosaic and blistering on leaves in La Plata (Buenos Aires green-belt). The diseased plants occurred together in the same line, showing a typical pattern of mechanically transmitted pathogens. Although incidence did not exceed 1.5%, it was distributed in several distant locations within the district, posting a serious threat to the tomato production.

Five samples were collected from symptomatic plants and tested for tospovirus, begomovirus and *Cucumber mosaic virus* by reverse transcription polymerase chain reaction (RT-PCR) (Dewey et al., 1996), PCR (Rojas et al., 1993) and enzyme-linked immunosorbent assay (ELISA), respectively. All results were negative. The samples were then examined under transmission electron microscopy. Isometric virus-like particles of 25-30 nm in diameter were seen in leaf-dip preparations and isometric virus-like particles aggregated in the cytoplasm of mesophyll cells, and alterations of chloroplast membrane in association to virus like particles, were observed in ultrathin sections suggesting it might be a tymovirus. To identify this virus, leaf and stem samples were dot blotted on a nitrocellulose membrane and incubated with *Tomato blistering mosaic virus* (ToBMV) antiserum, kindly provided by Dr. Alice K. Inoue-Nagata (Embrapa-Horticulture, Brazil). The samples reacted to the ToBMV antiserum, indicating the presence of ToBMV or a ToBMV-related virus.

Sap of the diseased samples was used to inoculate tomato cv. Elpida and several indicator plants (*Datura stramonium*, *Nicotiana benthamiana*, *Nicotiana tabacum* 'TNN', *Capsicum annuum* cv. Almuden, *Chenopodium quinoa*). The inoculated tomato plants showed foliar chlorosis, mosaic, necrosis and blistering, similar to those observed in the greenhouses, and the indicator plants reproduced the symptoms reported by de Oliveira et al. (2013).

Furthermore, two samples were subjected to RT-PCR using primers specifically designed to amplify the complete capsid protein (CP) from ToBMV: TymoF3RaceFor 5-TCCAATCCATGCCTTTTGAT-3 and ToBMV3EndNdelRev 5'-TTCATATGGTTTCCGATACCCTCG G -3'. DNA fragments of expected size (1000 bp) were amplified, cloned in pGEM-T easy (Promega) and sequenced. Sequence analysis showed the nucleotide sequences of the two isolates were identical, suggesting that they are the same variant (Accession number: KT235894). This isolate shares 92% identity with tomato isolate SC50 (Nicolini et al., 2015), and tobacco isolate BR001 (Melo et al. 2014), both from Brazil.

These results indicate that the tomato disease in Buenos Aires green-belt is caused by ToBMV.

To our knowledge, this is the first report of ToBMV infecting tomato in Argentina. This finding has an important epidemiological impact since ToBMV represents a new problem to tomato crops in this country.

References

De Oliveira VC, Nagata T, Guimarães FC, Ferreira FA, Kitajima EW, Nicolini C, de Oliveira Resende R, Inoue-Nagata AK. Virus genes 46:190-194. 2013

Dewey R, Semorile LC, Grau OI. J Virol Methods 56: 19-26. 1996

Melo FL, Fernandes JEA., Ribeiro BM, Ribeiro SG. Genome Announc. 2(4) e 00701-14. 2014

Nicolini C, Inoue-Nagata AK, Nagata T. Arch Virol. 160:609-12. 2015

Rojas MR, Gilbertson RL, Russell DR, Maxwell DP. Plant Dis. 77, 340-347. 1993



163x109mm (300 x 300 DPI)



109x163mm (300 x 300 DPI)