

DISEASE NOTE

OCCURRENCE OF APPLE STEM GROOVING VIRUS IN TUNISIAN APPLE ORCHARDS

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Apple is known to host many viruses and mixed infection by these viruses can induce significant yield reductions. Apple chlorotic leaf spot virus (ACLSV), apple mosaic virus (ApMV) and apple stem pitting virus (ASPV) have been reported previously in Tunisia (Mahfoudhi *et al.*, 2013). Apple stem grooving virus (ASGV), the type member of the genus *Capillovirus*, family *Betaflexiviridae*, is widely distributed in apple trees and has been associated with tree decline and graft union necrosis in sensitive combinations of scion and rootstock (Yanase *et al.*, 1990). To investigate the presence of ASGV in Tunisian apple orchards, a survey was conducted in the main apple growing regions and leaf samples were randomly collected in spring 2016 from 90 trees representing four important cultivars (Golden, Lorka, Richared, Anna). Total nucleic acids were extracted from leaf veins and purified according to Foissac *et al.* (2001). All samples were tested by RT-PCR for the presence of ASGV using specific primers (Menzel *et al.*, 2002) for the amplification of a 273 bp fragment from the coat protein coding region. Results showed that 31% of the tested samples were infected by ASGV. The infection rate ranged from 25% (cv. Richared) to 50% (cv. Golden). To confirm the identity of this virus, two isolates of ASGV were sequenced and sequences were compared to those available in GenBank. The nucleotide sequence of Tunisian ASGV isolates R161 (LT882718) and G171 (LT882719) showed identities of 96 and 99% with ASGV iso3_IN/GARD (LT574875), respectively. To our knowledge this is the first report of ASGV in Tunisian apple orchards.

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DISEASE NOTE

FIRST REPORT OF POTATO VIRUS S INFECTING TOMATO IN SLOVAKIA

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In 2016, during a survey aimed to assess the health status of tomatoes (*Solanum lycopersicum* L.) in Slovakia, a sample labelled as T62, collected from cultivated tomato grown in a garden in western Slovakia was analysed by Illumina next-generation sequencing of cDNA libraries prepared from ribosomal-depleted total RNAs isolated from leaves. *De novo* assembling of ca. 2.8 Mb of reads and subsequent mapping enabled the identification of a single potato virus S (PVS, genus *Carlavirus*) variant. Besides PVS, the T62 tomato showing slight veinal chlorosis on the leaves was found coinfecting with cucumber mosaic virus (CMV). Comparative analysis showed that the T62 genome (deposited as MF346599) was 78.7-95.8% identical to the 21 PVS complete genome sequences available in GenBank. Until now, however, the T62 PVS sequence is the only one originating from tomato. In the phylogenetic analysis, T62 forms a subcluster with Hungarian HF571059 and Ukrainian LN851189 and LN854492-93 isolates, all from potato. To further investigate the presence of PVS in tomato, 66 additional samples from seven localities in western Slovakia were tested by RT-PCR using a set of newly designed primers from PVS sequences retrieved from GenBank (accessed in May, 2016). The primers PVS_7833F (5'-AGGCY-ATGGGATTCAATGG-3', sense) and PVS_8386R (5'-TG-GTATCACCTCAGTTACTC-3', antisense) amplified a 533 nt long 3' region of the genome spanning the end of the CP and complete 11K protein genes. RT-PCR confirmed the presence of PVS in 21% of tomato plants tested. Comparison of 13 partial sequences obtained by Sanger sequencing of these amplicons (MF346600-MF346613) showed limited genetic variability (3.1%) among Slovak isolates. The phylogenetic analysis of obtained and additional available 47 sequences encompassing the target region showed that PVS isolates cluster in two main groups, previously designated as PVS^O (ordinary) and PVS^A (Andean) (Cox *et al.*, 2010). All Slovak PVS isolates were assigned to the major PVS^O strain, although divided between two distinct subclusters, without correlation to either geographic origin or biological properties.

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