Disease Note

MOLECULAR IDENTIFICATION OF ‘CANDIDATUS PHYTOPLASMA ASTERIS’ (16SRI-B) ASSOCIATED WITH THE LITTLE LEAF DISEASE OF POTATO IN INDIA

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Potato (Solanum tuberosum L.) is a widely cultivated and commercially important vegetable crop in India where it is cultivated in an area of about 1.34 million ha with a total production of about 24.7 million tons. Phytoplasma diseases of potato have been reported from several parts of the world as associated with organisms belonging to different 16Sr DNA group/subgroups. In December, 2011, little leaf disease symptoms were observed in potato crops grown in the Kushinagar district of Uttar Pradesh (India). Phytoplasmas were detected by PCR using P1/P7 primers (Deng and Hiruki, 1991; Schneider et al., 1995) which yielded amplicons ca. 1.8 kb in size only from four symptomatic samples. All four amplicons were directly sequenced from both ends and the sequence was deposited in GenBank under the accession No. KC312703. The little leaf potato phytoplasma isolate shared high 16S rDNA sequence identity (99%) with several isolates of Candidatus Phytoplasma asteris (16SrI group) from different parts of the world. Virtual PCR analysis using the web tool iPhyClassifier (Zhao et al., 1999) assigned the isolate to subgroup 16SrI-B. A number of crop and non-crop species have been reported to be infected by Ca. P. asteris in India (Raj et al., 2011). To the best of our knowledge, this is the first report of the occurrence of Ca. P. asteris (16SrI-B) in association with the little leaf disease of potato in India.


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Grapevine leafroll is one of the most important viral diseases of grapevines (Vitis vinifera). To date, 11 grapeleaf-associated viruses (GLRaVs) from the family Closteroviridae have been described. A recent taxonomic revision, however, suggests the existence of only five valid species (GLRaV-1, -2, -3, -4, and -7), whereas GLRaV-4, -5, -6, -9, -Pr, -De and Grapevine leafroll-associated Carnelian virus (GLRaCV) form a uniform phylogenetic clade and represent strains of GLRaV-4 (Martelli et al., 2012). In Spain, IMIDA performs all sanitary tests for certification of candidate clones from all Spanish autonomous communities. The detection of GLRaV-4, -5, -6, -7 and -9 relies on the use of commercial ELISA kits (Bioreba, Switzerland), and the confirmation of positive results by real-time RT-PCR to assess the health status of the clones. In 2011, GLRaV-9 was detected using the GLRaV-9 and TaqMan in a candidate clone of Mantúa (Man086), a traditional cultivar from Extremadura. To confirm the presence of this virus, conventional RT-PCR and sequencing were performed using total RNA from Man086 as template and specific primers designed in the HSP70h domain of GLRaV-9 (Alkowni et al., 2004). Pairwise comparisons of the corresponding sequence (580 bp, GenBank accession No. KC660077) with GLRaV-9 sequences available in GenBank showed the highest nucleotide (97.2%) and amino acid (95.3%) identities with an isolate from Washington state, USA (EU252530). Another set of primers was designed in the RNA-dependent RNA polymerase domain based on accession AY297819 from California, USA. Alignments between the sequence obtained (533 bp, GenBank accession No. KC660076) and AY297819 showed 96.1% and 97.2% nucleotide and amino acid sequence identities, respectively. This is the first report of GLRaV-9 in Spain.
