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DISEASE NOTES

First Report of *Leek yellow stripe virus* in Leek in Serbia

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Leek yellow stripe virus (LYSV), one of the most important and widespread viruses of leek and garlic worldwide, is endemic in various countries of the Mediterranean basin (Katis et al. 2012). During an October 2013 survey for the presence of *Allium* viruses in Serbia, commercially grown leek (*Allium porrum*) plants with virus-like symptoms were observed in Padinska Skela (City of Belgrade District). Initially, the leaf symptoms included irregular chlorotic to light yellow dashes, particularly on the bases of leaves. The lesions later enlarged and coalesced, resulting in large, yellow stripes and the infected leaves turned yellow and flaccid, followed by die-back. Disease incidence in the leek field was estimated at 20%. A total of 15 symptomatic plants were sampled and tested by double-antibody sandwich (DAS)-ELISA test using commercial polyclonal antisera (Bioreba AG, Reinach, Switzerland) for the most important *Allium* viruses: LYSV, *Garlic common latent virus*, *Onion yellow dwarf virus*, and *Iris yellow spot virus* (Pappu et al. 2005, Katis et al. 2012).


Commercial positive and negative controls and extracts from healthy leek leaves were included in each ELISA. All 15 tested leek samples were positive for LYSV and negative for the rest of tested viruses. Five carborundum-dusted plants of each *Chenopodium quinoa* and *A. porrum* 'Varna' were mechanically inoculated with sap prepared from ELISA-positive sample (277-13) using 0.01 M phosphate buffer (pH 7). Chlorotic local lesions on *C. quinoa* and streak mosaic on *A. porrum* 'Varna' were observed 5 and 16 days postinoculation, respectively, on all inoculated plants. Serological results were verified with reverse transcription (RT)-PCR. Total RNAs from all naturally and mechanically infected leek plants were extracted using the RNeasy Plant Mini Kit (Qiagen, Hilden, Germany). RT-PCR was performed using One-Step RT-PCR Kit (Qiagen) and specific primer pair 1LYSV and 2LYSV (Fajardo et al. 2001). An approximately 1000-bp fragment corresponding to the part of nuclear inclusion B (NIB) and coat protein (CP) coding region was obtained from all 20 naturally and mechanically infected leek plants, while no amplicon was recorded in the healthy and water controls. RT-PCR product obtained from one selected isolate (277-13) was purified using QIAquick PCR Purification Kit (Qiagen), sequenced directly in both directions using the same primers as for amplification, and submitted to the GenBank (Accession No. KR075504). Sequence analysis, conducted by MEGA5 software (Tamura et al. 2011), revealed that the leek isolate from Serbia showed the highest nucleotide identity of 94.8% (94.6% amino acid identity) with the sequence of LYSV isolate from leek (X89711). To our knowledge, this is the first report of natural infection of leek with LYSV in Serbia. Leek is an important and traditionally grown vegetable crop in Serbia and the presence of LYSV could cause considerable damage and severe yield losses, resulting in significant economic impact on leek production.

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