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Disease Notes

First Report of *Erwinia amylovora* Causing Fire Blight on Plum (*Prunus domestica*) in Hungary

A. Végh, Zs. Némethy, L. Hajagos, and L. Palkovics Department of Plant Pathology, Corvinus University of Budapest, Ménesi Road 44, H-1118 Budapest, Hungary. The project was funded by TÁMOP-4.2.1./B-09/1-KMR-2010-0005 and TÁMOP- 4.2.2./B-10/1-2010-0023 grants

During July 2011, a severe, unusual disease symptom was observed on young shoots on a 10-year old plum tree (*Prunus domestica* L. 'd'Agen') in the city of Budaörs, near Budapest. The naturally infected shoots showed typical symptoms of fire blight including terminal shoots with brown-to-black necrotic lesions and later, shepherd's crook deformation. Symptoms were the same as fire blight, symptoms reported from other hosts and locations. The first occurrence of fire blight on European plum was recorded in Germany in 2002 (4). Shoots containing regions of dead and healthy tissue were surface sterilized with ethanol (50-mg sample homogenized with 500 µl of sterile water and 50 µl of the homogenate streaked to King's B agar medium). After 48 h of incubation at 26°C, the medium contained pure cultures of a bacterium with white mucoid colonies, which is morphologically consistent with *E. amylovora* (1). Isolates were gram negative and induced a hypersensitive reaction in tobacco (*Nicotiana tabacum* L. 'White Burley') leaves (2). Biochemical tests were also used for identification, and the results of API 20E and API 50 CH kits (Biomérieux, Marcy l'Etoile, France), demonstrated that the bacterium belongs to *Enterobacteriaceae*. Pathogenicity was tested by injecting five healthy young plum shoots from the same tree with a 10-µl bacterial suspension of 10⁷ CFU/ml. Controls were injected with sterile distilled water. Shoots were kept at 26°C and 80 to 100% relative humidity. Five days after inoculation, dark brown-to-black lesions and shepherd's crook symptoms were observed only on inoculated shoots. The bacterium was reisolated from lesions on inoculated shoots, fulfilling Koch's postulates. No lesions were observed on controls. For molecular identification of the pathogen, the 16S rDNA region was amplified from isolate EA-PlumBo1 with a general bacterial primer pair (63f forward and 1389r reverse) (3). The PCR products were cloned into a pGEM T-Easy plasmid vector (Promega, Madison, WI) and were transformed into *Escherichia coli* DH5α cells. A recombinant plasmid (2A2.5) was sequenced by M13 forward and reverse primers. The sequence was deposited in GenBank (Accession No. HE610678) and showed 99 to 100% sequence homology with a number of *E. amylovora* isolates, including type strain AJ233410 with 99% similarity and 100% homology with sequences FN434113 and FN666575, where the complete genomes are known. On the basis of the symptoms, colony morphology, biochemical tests, and 16S rDNA sequence homology, the pathogen was identified as *E. amylovora*. To our knowledge, this is the first report of a natural outbreak of fire blight on plum in Hungary and the presence of the pathogen may seriously influence local stone fruit production in the future.

References: (1) E. O. King et al. J. Lab. Clin. Med. 44:301, 1954. (2) Z. Klement. Nature 199:299, 1963. (3) A. M. Osborn et al. Environ. Microbiol. 2:39, 2000. (4) J. L. Vanneste et al. Acta Hort. 590:89, 2002.

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