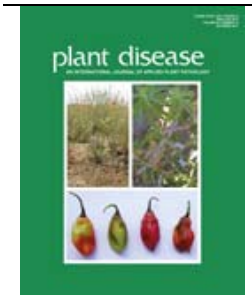

plant disease

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Disease Notes

First Report of the *Plum Pox Virus* Recombinant Strain on Peach in Bulgaria

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Plum pox virus (PPV) causes sharka, the most damaging viral disease of stone fruit species. Seven distinct PPV strains are known; PPV-M, PPV-D, and PPV-Rec are the most common (3). PPV-Rec is a unique recombinant (3) between PPV-M and PPV-D and has been reported from plum, apricot, Japanese plum, myrobalan, and blackthorn in eastern and central Europe, but has never been found in peach as a single natural infection (2). A survey was conducted during spring 2009 in eight peach orchards located in the southwest, southeast, and south central regions of Bulgaria to assess the incidence of PPV infection. A total of 98 leaf samples from individual trees showing PPV-like symptoms were collected and analyzed by triple-antibody sandwich (TAS)-ELISA with the universal monoclonal antibody (MAb) 5B (Agritest, Valenzano, Italy). Sixty one samples reacted positive for PPV (optical density 0.161 to 1.267) and these samples were further analyzed with PPV-M (AL) and PPV-D (4DG5) specific MAbs (1). All 61 samples reacted positively with PPV-M specific MAbs. To distinguish PPV-M and PPV-Rec strains, which are serologically identical, immunocapture (IC)-reverse transcription (RT)-PCR was carried out with PPV-M (CIP-M: 5′-GTC GCA GCA TTT GTA GCC CTT GTT-3′, CIP-MR: 5′-CCA ACA CGT TAA CGC CAT GCT TCA-3′) and PPV-D (CIP-D: 5′-ATG ATG CTG TTT GAC TCG GAG CGA-3′, CIP-DR: 5′-TCG CAA CTG CTT GCA CAC ATT CTC-3′) specific primers targeting the 6K1-CI genomic region. A PCR fragment of ~880 bp amplified with PPV-M specific primers obtained from 59 samples confirmed that these were PPV-M isolates. However, the remaining two samples (both coming from infected tress located in two different orchards in the southwest region) yielded a 468-bp PCR fragment with PPV-D specific primers, suggesting that these two samples belonged to PPV-Rec strain. These samples together with controls of PPV-M, PPV-D, and PPV-Rec strains were further analyzed by RT-PCR using mD5/mM3 primers spanning the recombination breakpoint (4). Both peach samples and the PPV-Rec strain control produced a single 605-bp PCR product. The two peach amplicons were purified and sequenced directly with the same primers. The nucleotide (nt) sequences obtained were 100% identical to each other. BLAST analysis of the two samples with PPV-Rec (No. AF421118.1) showed maximum nt identity of 98%. Percent maximum nt identity with PPV-M (No. AY324837.1) and PPV-D (No. AB576062.1) were 93 and 87%, respectively. The deduced amino acid sequences of the two isolates were 98% identical to PPV-Rec (No. No.

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AF421118.1), 93% identical to PPV-M (No. M92280.1), and 84% identical to PPV-D (No. AB576062.1). Analyzed samples were further transmitted from the diseased trees to peach seedlings (GF 305) by chip-budding in a greenhouse during the fall of 2009. Six months later, faint vein clearing on the leaves of inoculated seedlings was observed. The presence of PPV was confirmed by TAS-ELISA and PPV-Rec presence was shown by IC-RT-PCR (mD5/mM3 primers). One of the generated 605-bp products was sequenced and showed 100% nt identity with the isolate used for inoculation. To our knowledge, this is the first identification of PPV-Rec strain in naturally infected peach trees, a finding that calls for further large-scale investigations of PPV-Rec incidence in peach in Bulgaria.

References: (1) M. Cambra et al. OEPP/EPPO Bull. 24:569, 1994. (2) S. Dallot et al. Acta Hortic. 781:227, 2008. (3). M. Glasa et al. J. Gen. Virol. 85:2671, 2004. (4) Z. Šubr et al. Acta Virol. 48:173, 2004.