

## First report of alder yellows phytoplasma on common alder (*Alnus glutinosa*) in Serbia

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Alder yellows (AldY) phytoplasma associated with common alder (*Alnus glutinosa*) and grey alder (*A. incana*) belongs to the 16SrV-C group and is closely related to the Flavescence dorée (FD) phytoplasma, a quarantine pathogen of economic importance that affects vineyards of southern Europe including Serbia. AldY phytoplasma has been reported in France, Germany, Switzerland, Austria, Italy and in the Baltic region (Arnaud *et al.*, 2007) where alders are frequently infected, exhibiting symptoms such as yellowing, small leaves, reduced foliage, or sometimes infected trees remain symptomless (Lederer & Seemüller, 1991). During September 2007, leaves with petioles from twelve alder trees showing symptoms of discrete leaf yellowing and multiple shoot growth from the basal part of trunk, were collected from three different sites in the vicinity of Topola (central Serbia) and another twelve samples showing discrete symptoms were collected from one site near Veliko Gradište (east Serbia). Leaves of six symptomless young alder seedlings were used as controls.

DNA was extracted from fresh leaf midribs and petioles from affected and symptomless plants according to previously reported protocols (Angelini *et al.*, 2001). Initial phytoplasma identification was conducted using a nested PCR assay with P1/P7 and 16r758f/M23Sr primers on the 16S rRNA gene, followed by RFLP analysis with *Xba*I restriction enzyme. RFLP profiles showed the presence of phytoplasmas of the 16SrV-C group in all samples with symptoms. Further characterization was performed by amplifying the ribosomal protein gene operon using primers rp(V)F1/rpR1 followed by rp(V)F1A/rp(V)R1A, followed by digestion with *Msp*I (Lee *et al.*, 2004). Two different *Msp*I RFLP profiles were detected among the AldY phytoplasma isolates: one similar to FD-C and one (only in samples from eastern Serbia) similar to the AldY strain previously described by Lee *et al.* (2004). None of the symptomless plants were positive for the presence of phytoplasma.

This is the first report of phytoplasmas associated with common alder in Serbia and of the association of two distinct isolates belonging to rRNA

group 16SrV-C. These phytoplasmas have recently been studied as a possible natural source of phytoplasmas that affect vineyards in France, Italy and Germany (Arnaud *et al.*, 2007). The finding of AldY phytoplasmas infecting alders in Serbia could be of importance in revealing an epidemiological cycle of FD outbreaks in this area.

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## First report of bacterial wilt caused by *Ralstonia solanacearum* biovar 2 on tomato in Turkey

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In the summer of 2006 severe wilt symptoms of tomato plants in some fields in the Canakkale province of the Aegean Region of Turkey were observed. Symptoms started as leaf drooping followed by wilting of whole plants within a few days, leading to total plant collapse. Vascular discoloration and browning of the pith in the lower stem occurred. On the stem of some plants, adventitious roots were formed. White, irregular and fluidal colonies with blood red colouration in the centre were consistently isolated from diseased tissues plated onto mSMSA medium (Englebrecht, 1994; Elphinstone *et al.*, 1996). Isolates were identified as *Ralstonia solanacearum* by biochemical, immunofluorescence (IF) and real-time PCR tests. They utilised maltose, lactose and D(+) cellobiose, but not mannitol, sorbitol and dulcitol in accordance with biovar 2. In IF tests, fluorescing cells with typical morphology were observed at antibody dilutions of 200–12 800. Real-time PCR was performed using biovar 2 specific primers and probe as described by Özakman & Schaad (2003). Ct values of 18.46 and 22.27 were obtained from 2 isolates respectively. Pathogenicity of the strains was confirmed on 2-week old tomato plants (cv. Rio Grande) inoculated with a bacterial suspension containing  $10^6$  cfu mL<sup>-1</sup> using a hypodermic syringe. Control plants were inoculated with

sterile water. Inoculated plants were grown in a growth chamber for 15 days at 25°C and 70–80% humidity. Wilting symptoms were observed 5–6 days after inoculation. No symptoms developed on sterile water inoculated controls. The bacteria were re-isolated and identified as *R. solanacearum* biovar 2. This is the first report of *R. solanacearum* biovar 2 on tomato in Turkey.

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