New Disease Reports

First report of *Pseudomonas mediterranea* causing tomato pith necrosis in Argentina

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During the summers of 2007 and 2008 fruiting tomato plants (*Solanum lycopersicum* cv. Orco) from commercial greenhouses near La Plata, Argentina (35 ºS 57 ºW) showed abundant adventitious root production, apical chlorosis of leaves and a brown discoloration of the stem pith (Fig. 1). These symptoms were similar to those reported by López et al. (1994) and Catara et al. (2002) on tomatoes affected by *Pseudomonas corrugata* or *Pseudomonas mediterranea*. Bacteria consistently isolated from stem lesions formed cream-coloured, glistening, convex colonies on sucrose peptone agar (SPA) and were non-fluorescent on King's medium B (KMB). Four isolates were selected for further study. All were aerobic, Gram-negative rods with PHB inclusions. In LOPAT tests, all induced a hypersensitive response in tobacco plants, were oxidase positive, did not cause soft rot of potato tubers, and were negative for levan and arginine dihydrolase. Colonies developed at 28ºC and 37ºC but not at 41ºC. Additional characterisation was achieved by API 20 NE tests strips (Biomerieux®, Argentina). Reference strains 536.7 (Spain), 592.4 (Spain) and CFBP 10906 (France) of *P. mediterranea* and strain NCPPB 2445 of *P. corrugata* were included in all tests for comparison.

Further identity was confirmed by PCR, using species-specific primers PC5/1-PC5/2 for *P. mediterranea* and primers PC1/1-PC1/2 for *P. corrugata* (Catara et al., 2002). All the strains were identified by the amplification of a 600 bp DNA fragment characteristic of *Pseudomonas mediterranea* (Fig. 2; Catara et al., 2002). The isolates of *P. mediterranea* were also differentiated from those of *P. corrugata* by PCR/RFLP analysis of 16S rDNA gene by using endonuclease AluI (Fig. 3). Pathogenicity was verified on four-week-old tomato plants (cv. Presto) by injecting bacterial suspensions at 10^7 cfu/ml or sterile distilled water for controls, after which the plants were kept for 72 h in a humid chamber before incubation at 25°C. After 45 days inoculated plants showed necrotic pith symptoms similar to those observed on field-grown plants, whereas no lesions were observed on controls. Pith necrosis caused by *P. corrugata* and *P. viridiflava* has been previously reported in Argentina (Alippi et al., 1993 Alippi et al, 2003). This is the first report of a disease caused by *P. mediterranea* on greenhouse-grown tomatoes in Argentina and South America.

Figure 1: Figure 1: Brown discoloration of the stem pith of tomato cv. Orco induced by *Pseudomonas mediterranea*

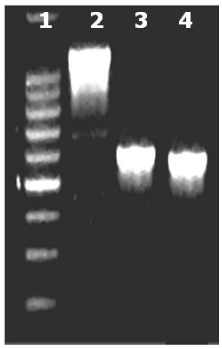

Figure 2: Figure 2: Amplification of 1,100 bp and 600 bp DNA fragments of *Pseudomonas corrugata* and *Pseudomonas mediterranea* by PCR using species-specific type I primers PC5/1-PC5/2 and type II primers PC1/1-PC1/2. Lane 1: Molecular marker 100 bp Promega®, lane 2: *P. corrugata* NCPPB 2445, lane 3 *P. mediterranea* strain 536.7, lane 4. *P. mediterranea* F-431 from Argentina.

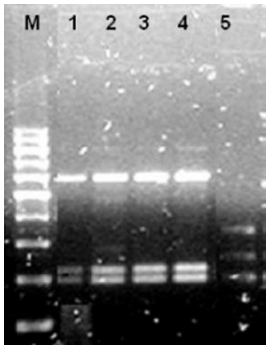

Figure 3: Figure 3: Gel electrophoresis of PCR-amplified 16S rDNA fragment of 1,500 bp with primers rD1 and fD1 and digested with Alu I. Lane 1: *P. mediterranea* strain 536.7; lanes 2, 3 and 4 Argentinian isolates F-431, F-432 and F-433; lane 5: *P. corrugata* NCPPB 2445.

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References

Alippi AM, Dal Bo E, Ronco LB, López MV, López AC, Aguilar OM, 2003. Pseudomonas populations causing pith necrosis of tomato and pepper in Argentina are highly diverse. *Plant Pathology* **52**, 287-302.

Alippi AM, Ronco L, Alippi HE, 1993. Tomato pith necrosis caused by *Pseudomonas corrugata* in Argentina. *Plant Disease* **77**, 428.

Catara V, Sutra L, Morineau A, Achouak W, Christen R, Gardan L, 2002. Phenotypic and genomic evidence for the revision of *Pseudomonas corrugata* and proposal of *Pseudomonas mediterranea* species sp. nov. *International Journal of Systematic and Evolutionary Microbiology* **52**, 1749-1758.

López MM, Siverio F, Albiach MR, García F, Rodríguez R, 1994. Characterization of Spanish isolates of *Pseudomonas corrugata* from tomato and pepper. *Plant Pathology* **43**, 80-90.