Diseases Caused by Bacteria and Phytoplasmas

First Report of Bacterial Speck Caused by *Pseudomonas syringae* pv. *tomato* Race 1 Affecting Tomato in Different Regions of Chile

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Funding: This study was supported by Fondo Nacional de Desarrollo Científico y Tecnológico (Fondecyt de Iniciación 11200593) (MV) and Programa de Investigación Asociativa, Agencia Nacional de Investigación y Desarrollo (PIA Anillo GAMBIO ACT172128) (MS, MV). Plant Dis. 106:1979, 2022; published online as <https://doi.org/10.1094/PDIS-11-21-2436-PDN>. Accepted for publication 14 December 2021.

In Chile, tomato is one of the most widely cultivated vegetables, with around 5,000 ha for the fresh market and 8,000 ha for the processing industry. During recent years, symptoms of bacterial speck caused by *Pseudomonas syringae* pv. *tomato* have been observed more frequently in tomato plants in different regions of Chile. This pathogen was first identified in Chile in 1987 (Latorre and Lolas 1988), and the presence of an apparent new variant was reported in 2004 (Besoain et al. 2004). To characterize the pathogen that was affecting this crop, samples of diseased tomato plants were taken in three regions of Chile. The samples were collected in 2016 in Northern Chile in Lluta Valley from the Arica y Parinacota Region, in Central Chile in 2014 in Limache from the Valparaíso Region, and in 2015 in Pichidegua from the O'Higgins Region. Affected tomato plants exhibited dark brown to black lesions surrounded by yellow halos in the leaves and dark brown to black lesions in the stems, pedicels, and peduncles. Plants tissues were macerated, and the suspension was spread on King's B medium, resulting in fluorescent colonies visualized under 366-nm UV light. LOPAT test results of three selected isolates from different regions were: levan production (+), oxidase reaction (–), potato soft rot (–), arginine dihydrolase production (–), and tobacco hypersensitivity (+) (Lelliott et al. 1966). Molecular identification was carried out by amplification and sequence analysis of housekeeping

genes *cts*, encoding citrate synthase; *gyrB*, encoding DNA gyrase B; and *rpoD*, encoding sigma factor 70 (Hwang et al. 2005; Sarkar and Guttman 2004) (GenBank accession nos. OK001658 to OK001666). BLAST analysis of the *cts* and *rpoD* genes of the three isolates resulted in a match with a 100% identity (919 bp and 491 bp, respectively) with *P. syringae* pv. *tomato* strain B13-200 (GenBank: CP019871.1). BLAST analysis of the *gyrB* gene of two isolates resulted in a match with a 100% identity (684 bp) and one isolate with 99.85% (683 bp) identity with *P. syringae* pv. *tomato* strain B13-200. To identify race 1, each strain was inoculated in five tomato plants cultivar San Pedro, which is susceptible to both races of *P. syringae* pv. *tomato*, and cultivar Rio Grande, which is resistant to race 0. The tomato plants were slightly wounded with a metal sponge and then sprayed with a bacterial suspension (10^8 CFU ml^{–1}) of each isolate, including the reference strain DC3000 (race 0). Negative controls were sprayed with water. The plants inoculated with Chilean strains in both cultivar San Pedro and cultivar Rio Grande showed symptoms of bacterial speck after 7 days. Plants inoculated with the DC3000 strain showed symptoms only in cultivar San Pedro, whereas control plants remained asymptomatic. Strains were reisolated from symptomatic plants and identified by gene sequence analyses as *P. syringae* pv. *tomato*. This is the first report of *P. syringae* pv. *tomato* race 1 in Chile. Race 1 was previously reported in Canada (Lawton and MacNeill 1986), Italy (Buonaurio et al. 1996), California (Arredondo and Davis 2000), Portugal (Cruz et al. 2010), and other states in the United States and countries in South America, Europe, Africa, and Australia, becoming the most commonly isolated race today (Cai et al. 2011). These results will be the basis for future studies of epidemiology, characterization, and virulence in order to explain the outbreak of this disease and the severity of symptoms observed.

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The author(s) declare no conflict of interest.

Keywords: bacterial speck, Chile, *Pseudomonas syringae* pv. *tomato*, race 1

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