Diseases Caused by Bacteria and Phytoplasmas

First Report of *Pectobacterium polaris* Causing Aerial Stem Rot of Potato in Mexico

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In Mexico, potato (Solanum tuberosum L.) is one of the most important vegetable crops for local consumption and industry. More than 1.8 million metric tons of potatoes are produced annually, of which the state of Sinaloa contributes 21.5% (SIAP 2022). In January 2020, potato plants (cv. FL1867) showing aerial stem rot symptoms were observed in a commercial field from the Santa Rosa Valley, northern Sinaloa, with an incidence of 36%. Putative pectolytic bacteria showing pitting on crystal violet pectate plates were restreaked and purified onto nutritive agar medium at 28°C. Four independent isolates were obtained (L25F-83, L25F-105, L25F-115, and L25F-125) from four symptomatic stems with biochemical and morphological characteristics related to Pectobacterium spp., such as catalase positive, oxidase negative, pectinolytic activity, gram-negative, and nonfluorescent in King's B medium. Bacterial gDNA was used for amplification and sequencing of two housekeeping genes (dnaX and leuS) (Portier et al. 2019). The nucleotide sequence identity between our isolates was 100% with both housekeeping genes (dnaX, OP376536 to OP376539, and leuS, OP376540 to OP376543). BLASTn analysis showed the dnaX gene shared 98.98 and 99.19% identity with two soft rot-causing bacterial strains, NIBIO1006^T (CP017481) and NIBIO1392 (CP017482), respectively, of

Pectobacterium polaris, and the leuS gene shared 99.56% identity with the P. polaris type strain NIBIO1006^T. To further validate the identification, two strains, S5 (isolate L25F-105) and S6 (L25F-125), were selected for wholegenome sequencing. The average nucleotide identity (ANI) values for closely related species were calculated using the Orthologous Average Nucleotide Identity (Ortho-ANI) Software Tool (OAT) (Lee et al. 2016). The Type (Strain) Genome Server was used for accurate genome-based taxonomy (https://tygs.dsmz.de; Meier-Kolthoff and Göker 2019). The genomes of the P. polaris strains S5 (4,811,345 bp, GC = 52%, AULSZ00000000) and S6 (4,809,754 bp, GC = 52%, JAULTA00000000) revealed 96.86 and 96.07% Ortho-ANI and 73.6 and 66.5% dDDH with the P. polaris type strain NIBIO1006^T and the *P. parvum* strain CFBP8630, respectively. Multilocus sequence analysis was performed on concatenated complete sequences of dnaX (OR470476 and OR470477), leuS (OR470484 and OR470485), recA (OR470488 and OR470489), acnA (OR470474 and OR470475), gapA (OR470478 and OR470479), gyrA (OR470480 and OR470481), icdA (OR470482 and OR470483), proA (OR470486 and OR470487), and rpoA genes (OR470490 and OR470491). The consensus tree, constructed using the maximum-likelihood method (MEGA 7.0), clustered the strains S5 and S6 with the *P. polaris* strains NIBIO1006^T and NIBIO1392. The four isolates resulted in pathogenicity in tuber slices and potato seedlings (cv. Fianna) 24 and 72 h, respectively, after being inoculated with 30 µl of bacterial suspension $(1 \times 10^8 \text{ CFU/ml})$ and incubated at 28°C and 85% relative humidity. Bacterial colonies were reisolated from the affected tissue and identified with the same PCR primers as described earlier. Accordingly, the P. polaris isolates S5 and S6 fulfill Koch's postulates for aerial stem rot of potato. To our knowledge, this is the first report of P. polaris causing aerial stem rot of potato in Mexico. This bacterium could be a significant threat to local potato producers; therefore, an accurate and sensitive method of detection and epidemiological studies are needed to support an effective disease diagnosis and management program.

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