Diseases Caused by Bacteria and Phytoplasmas

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

Jinhui Wang,¹ Wanxin Han,¹ Zheng Li,¹ Jianing Cheng,² Yang Pan,¹ Dongmei Zhao,¹ Dai Zhang,¹ Qian Li,¹ Zhihui Yang,^{1,3,†} and Jiehua Zhu^{1,3,†}

¹ Hebei Agricultural University, College of Plant Protection, Baoding 071001, China

² Hebei Agricultural University, Agricultural Business Training and Entrepreneurship Center, Baoding 071001, China

³ Technological Innovation Center for Biological Control of Crop Diseases and Insect Pests of Hebei Province, Baoding 071001, China

J. Wang and W. Han contributed equally to this research.

Funding: This research work was supported by China Agriculture Research System of MOF and MARA. Plant Dis. 106:755, 2022; published online as <https://doi.org/10.1094/PDIS-06-21-1269-PDN>. Accepted for publication 17 August 2021.

In July 2020, potato plants (cv. Xisen 6) showing characteristic symptoms of aerial stem rot were observed in a field in Fengning Manchu Autonomous County, Chengde, Hebei Province (northern China). The disease incidence in that field (5 ha in size) was more than 50%. Aerial stem rot of potato has increased in prevalence over recent years in Chengde, and it can cause significant yield loss on susceptible cultivars such as Xisen 6 and Huangxin 226. Affected stem (light brown and water-soaked stem sections) pieces ~0.5 cm in length were surface sterilized by dipping them in 75% ethanol for 1 min followed by three successive rinses with sterile distilled water. Then, the tissues were soaked in 200 µl of 0.9% saline for 20 min. Aliquots (20 µl) of three 10-fold dilutions of the tissue specimen soaking solution were plated onto crystal violet pectate (CVP) medium. The CVP plates were incubated at 28°C for 48 h. Colonies producing pits were restreaked and purified on Luria-Bertani agar plates. The bacterial gDNA was extracted using the Easy-Pure Bacteria Genomic DNA Kit (TransGen Biotech, Beijing, China). The 16S rDNA region was amplified by PCR using the universal primers 27F/1492R (Weisburg et al. 1991) and sequenced. Results of the BLASTn analysis of the 16S rDNA amplicons (MZ348607, MZ348608) suggested that the isolates FN20211 and FN20222 belonged to the genus *Pectobacterium*.

Housekeeping genes including *acnA*, *gapA*, *icdA*, *mdh*, *proA*, and *rpoS* were also amplified using a set of primers (Ma et al. 2007; Waleron et al. 2008) followed by sequencing (MZ356250 to MZ356261). To determine the species of the stem rot *Pectobacterium* isolates, multilocus sequence analysis (MLSA) was performed with six housekeeping genes, and a phylogenetic tree was reconstructed using RAxML (<https://github.com/stamatak/standard-RAxML>). No sequence variation was observed at any MLSA locus between FN20211 and FN20222. The result of phylogenetic analysis showed that the isolates clustered with *Pectobacterium polaris* type strain NIBIO1006^T, which was isolated from potato (Dees et al. 2017). And the concatenated sequence of the six loci of isolate FN20211/FN20222 is 100% identical to those of the strains PZ1 (CP046377.1) and WBC1 (GCF_011378945.1), which were isolated from potato in South Korea and from Chinese cabbage in China, respectively. Potato seedlings (cv. Xisen 6 and Favorita) were inoculated with the isolates FN20211 and FN20222 by injecting 100 µl of bacterial suspensions (10⁸ CFU/ml) into the upper parts of the stems of potato plants, or injecting with 100 µl of 0.9% saline as a control. The seedlings were grown at 25°C and 50% relative humidity. Three days after inoculation, only the bacteria-inoculated seedlings showed disease symptoms resembling those observed in the field. Bacterial colonies were obtained from the infected stems and were identified using the same PCR primers as described above. Therefore, *P. polaris* isolates FN20211 and FN20222 fulfill Koch's postulates for aerial stem rot of potato. *P. polaris* causing blackleg and soft rot on potato plants has been reported in European countries including Netherlands, Norway (Dees et al. 2017), and Poland (Waleron et al. 2019) and also in Pakistan (Sarraz et al. 2019) and Russia (Voronina et al. 2021). To our knowledge, this is the first report of *P. polaris* causing aerial stem rot of potato in China. The stem rot poses a significant threat to the local potato industry, and further research on epidemiology and disease management options is needed.

References:

- Dees, M. W., et al. 2017. Int. J. Syst. Evol. Microbiol. 67:5222.
- Ma, B., et al. 2007. Phytopathology 97:1150.
- Sarraz, S., et al. 2019. Plant Dis. 103:1405.
- Voronina, M. V., et al. 2021. Plant Dis. 105:1851.
- Waleron, M., et al. 2008. Eur. J. Plant Pathol. 121:161.
- Waleron, M., et al. 2019. Plant Dis. 103:1409.
- Weisburg, W. G., et al. 1991. J. Bacteriol. 173:697.

The author(s) declare no conflict of interest.

e-Xtra

Keywords: potato, *Pectobacterium polaris*, stem rot

[†]Indicates the corresponding authors.

Z. Yang; bdyzh@hebau.edu.cn, and J. Zhu; zhujiehua@hebau.edu.cn