Diseases Caused by Fungi and Fungus-Like Organisms

First Report of Stem Rot of Huangjing (*Polygonatum sibiricum*) Caused by *Athelia rolfsii*

Qingqun Tan,¹ Dailin Zhao,¹ Xuehui Yang,¹ Na Wang,^{2,†} and Haiyong He^{1,†}

¹ Guizhou Institute of Plant Protection, Guizhou Academy of Agricultural Sciences, Guiyang, 550006, China

² Research Institute of Pomology, Chinese Academy of Agricultural Sciences, Xingcheng 125100, China

Funding: This work was supported by Science and Technology Support Program, Guizhou, China ([2020] 4Y105). Plant Dis. 105:1197, 2021; published online as <https://doi.org/10.1094/PDIS-09-20-1891-PDN>. Accepted for publication 29 October 2020.

Huangjing (*Polygonatum sibiricum*) is a medicinal plant widely distributed in China, Japan, and Korea. The dried rhizome of huangjing has been reported to have many pharmacological applications and biological activities, such as antioxidant, immunity enhancement, antifatigue, antiosteoporosis, and antiaging activity (Cui et al. 2018). In June 2018, we observed some wilted huangjing plants in commercial plantings in Shuicheng, Guizhou, China (26.22 N, 104.76 E). Symptoms began as moderate to severe wilting of stems and necrosis of leaves, followed by the death of plants. Collar rot appeared on the stem near to the soil. When incubated at 28°C and 100% relative humidity for 8 to 10 days, the infected stem produced brown sclerotia. We picked the sclerotia and cultured them onto potato dextrose agar (PDA) supplemented with 50 µg/ml of streptomycin. The hyphal tips generated by the sclerotia were isolated under microscopic field and transferred to fresh PDA. Three isolates (HJ-1, HJ-6, and HJ-10) came from the hyphal tips and formed the typical clamp connection structure at 6 to 7 days postincubation, and the sclerotia produced were white and the late ones turned dark brown. The matured sclerotia were globular, 1.5 to 3.3 mm (average 2.2) in diameter. The morphologic observation revealed that the three isolates were consistent

with *Athelia rolfsii* (Curzi) C.C. Tu & Kimbr. (Paul et al. 2017). To further confirm the fungal species, the ribosomal internal transcribed spacer (ITS) sequences were amplified and sequenced. Primers and PCR amplification were referenced as previously described (Paul et al. 2017). The sequences were compared with type sequences in GenBank. The ITS sequences (GenBank accessions MT478452, MT949696, and MT949697) of the isolates (HJ-10, HJ-1, and HJ-6) were 99% identical with strain 13M-0091 (GenBank accession KT222898) of *A. rolfsii*, respectively (Paul et al. 2017). A maximum likelihood tree was constructed using MEGA-X version 10.1.6 (Kumar et al. 2018) based on the ITS sequences of the three strains (HJ-10, HJ-1, and HJ-6) and that of *Athelia* spp. previously deposited in GenBank (Paul et al. 2017). Phylogenetic analysis showed that the isolates (HJ-10, HJ-1, and HJ-6) belong to the *A. rolfsii* clade. Based on morphology and DNA sequencing, the isolates (HJ-10, HJ-1, and HJ-6) were identified as *A. rolfsii*. To verify pathogenicity, huangjing seedlings were inoculated with colonized agar discs of the isolates. Additional huangjing plants inoculated with uncolonized agar discs were used as the control. After inoculation, huangjing seedlings were moved to the inoculation chamber under high humidity and 28°C for 3 days and then transferred to a greenhouse. Typical wilting symptoms appeared 8 days after inoculation and were similar to those observed in the field, whereas control plants remained symptomless. The causal agent was isolated from the lesions, and the ITS sequences of them were sequenced again. The alignment analysis of the ITS sequences showed the causal agents are consistent with the original isolates. These studies fulfilled Koch's postulates. To our knowledge, this is the first report of *A. rolfsii* causing stem rot on huangjing.

References:

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The author(s) declare no conflict of interest.

Keywords: *Polygonatum sibiricum*, ITS, stem rot, *Sclerotium rolfsii*

[†]indicates the corresponding authors.

E-mail: N. Wang; Na.Wang18@outlook.com and H. Y. He; Haiyong.He@hotmail.com