Diseases Caused by Fungi and Fungus-Like Organisms

First Report of *Epicoccum poaceicola* Causing Leaf Spot on *Phyllostachys viridis* in China

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Phyllostachys viridis (Poaceae) is a bamboo species broadly distributed in Asia. It is used as raw material in papermaking and food processing and as an evergreen ornamental in parks and landscapes. In February 2019, a leaf spot disease was found on *P. viridis* in Jurong, Zhenjiang, Jiangsu, China. The incidence rate was ~53%. The disease primarily appeared as orange chlorotic spots on leaves. The spots enlarged and coalesced to form irregular brown necrotic lesions. Symptomatic leaves were collected; 12 cuttings were made from the margin of the lesions (~3 × 3 mm), surface sterilized using 75% ethanol for 1 min, rinsed three times in sterile distilled water, and placed on 2% PDA in Petri plates for 5 days at 25°C. Twenty-two fungal isolates were single spored and stored in the Forest Pathology Laboratory at Nanjing Forestry University. Pathogenicity of all isolates was tested on leaves of *P. viridis* using mycelial discs. Mycelial plugs (5-mm-diameter discs) of each isolate were taken from the margins of colonies grown on PDA for 5 days and inoculated onto healthy leaves that were surface sterilized using 75% ethanol and wounded with a sterilized needle. PDA discs without fungi were used to treat the control leaves. The inoculated whole plants were kept in a growth chamber at ~25°C at 85% relative humidity. The experiment was repeated twice, and there were at least five replicates for each treatment. After 5 days postinoculation, the leaves inoculated with isolate PV2-9 showed necrotic lesions. No lesions were found on the plants inoculated by other isolates and the control PDA

plugs. The pathogen was reisolated from the infected leaves, which shared similar morphological characteristics of colonies, aerial mycelia, and conidia with the original inoculated isolate PV2-9. Identification of the fungus was based on morphological characteristics and molecular methods. Colonies of PV2-9 on PDA were white, floccose, round, and pale gray with pale-reddish pigmentation near the center. The colonies attained 65 ± 0.3-mm diameter after 7 days at 25°C. Abundant conidiomata were observed over the medium after 4 weeks. Conidiomata were pycnidial, solitary or gregarious, scattered, semi-immersed to erumpent, or superficial uniloculate to multiloculate, globose to subglobose, ostiolate, 105.2 to 162.1 × 90.3 to 116.2 µm ($\bar{x}$ = 122.6 ± 14.4 × 97.9 ± 6.8 µm, n = 30). Pycnidial wall comprised three to seven layers, dark brown to lightly pigmented, 13.4 to 19.7 µm thick ($\bar{x}$ = 16.4 ± 1.6 µm, n = 30). Conidiogenous cells were phialidic, hyaline, smooth, flask-shaped to doliform, lining the conidiomatal cavity. Conidia were ellipsoidal to cylindrical, smooth, hyaline, aseptate, 4.0 to 6.2 × 2.4 to 3.3 µm ($\bar{x}$ = 4.8 ± 0.5 × 2.5 ± 0.3, n = 30), with zero to three guttules. These morphological characteristics matched the *Epicoccum poaceicola* described by Thambugala et al. (2017). The internal transcribed spacer (ITS) region of rDNA, 28S large subunit ribosomal RNA gene (LSU), RNA polymerase II second largest subunit gene (RPB2), and β -tubulin gene (TUB) were amplified using the primers published in Raza et al. (2019). The ITS (MT110918), LSU (MT110921), TUB (MT119149), and RPB2 (MT119150) showed 99.3, 99.8, 98.1, and 99.8% identity to the corresponding sequences of *E. poaceicola* ex-type MFLUCC 15-0448, respectively. Phylogenetic analysis using concatenated sequences of ITS, LSU, TUB, and RPB2 also showed that PV2-9 clustered monophyletically with *E. poaceicola* MFLUCC 15-0448, supported with a high bootstrap value (98%). Based on morphological and phylogenetic data, PV2-9 was identified as *E. poaceicola*. *E. poaceicola* has been collected from dead leaves of Poaceae sp. and described as a new species by Thambugala et al. (2017). To our knowledge, this is the first report of *E. poaceicola* causing leaf spot on *P. viridis* in China. Further studies are needed to assess its impact on *P. viridis* as well as to monitor and control this disease.

References:

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