Diseases Caused by Fungi and Fungus-Like Organisms

First Report of Leaf Spot Caused by *Fusarium equiseti* on Switchgrass in Shandong, China

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Switchgrass (*Panicum virgatum* L.) is widely cultivated in China as a forage and model bioenergy crop. In August 2019, a new leaf spot disease on 'Alamo' switchgrass was found in Guangrao, Shandong, China, with disease incidence of approximately 20%. Initial symptoms appeared as 2- to 4-mm reddish-brown circular spots on the leaves and subsequently increased in size and coalesced to oblong lesions with 5 to 15 mm length. Affected tissues turned from brown to gray in color as the tissues shriveled. Typical symptoms appeared as reddish-brown, circular or oblong lesions with chlorotic centers on leaves. Symptomatic tissues (5 × 5 mm) were cut from the border between diseased and healthy leaf tissue, sterilized in 75% ethanol for 30 s, soaked in 0.1% HgCl₂ for 2 min, rinsed three times with sterile water, and placed on potato dextrose agar (PDA) at 25°C for 5 days. Colonies of the single-spore isolates were white and fluffy at the beginning

and then changed to dark brown in the middle and beige coloration on the reverse side of PDA. The isolates were transferred to carnation leaf agar (Li et al. 2014) and incubated at 28°C for 10 days. Macroconidia were curved to lunate and had three to five septa, with the size ranging from 21.2 to 59.4 × 2.8 to 5.7 μm (*n* = 100). No microconidia were observed. Chlamydospores with thick, roughened walls were abundant and were always found as clumps or chains, ellipsoidal or subglobose. For molecular identification, rDNA internal transcribed spacer (ITS) region (White et al. 1990) and the transcription elongation factor 1α gene (TEF-1α) (Carbone and Kohn 1999) of isolate DYL6Z were amplified and sequenced. The ITS and TEF-1α sequences of the isolate were submitted to GenBank (MN589985 and MN735219, respectively). BLASTn analysis of the sequences showed 100% identity with *Fusarium equiseti* (MN243680 and MK630325). Phylogenetic analysis using the maximum-likelihood algorithm showed that isolate DYL6Z was clustered with *F. equiseti* isolate (NNGF1). Based on morphological (Leslie and Summerell 2006) and molecular characteristics, the fungus was identified as *F. equiseti*. Pathogenicity tests were performed by spraying a conidial suspension (5 × 10⁵ conidia/ml) on twenty 8-week-old Alamo switchgrass plants, whereas control plants were inoculated with sterile water only. All plants were placed in a growth chamber with a temperature of 22°C and an approximate relative humidity of 95% for 24 h, and they were then transferred to a greenhouse with a temperature of 25 ± 2°C and an approximate relative humidity of 75%. Five days after inoculation, all inoculated leaves showed symptoms similar to those observed in the field, but the control plants remained symptomless. The same fungus was reisolated from inoculated leaves and reidentified, thus confirming Koch's postulates. The pathogenicity test was repeated three times. To our knowledge, this is the first report of leaf spot caused by *F. equiseti* on switchgrass in China.

References:

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