Diseases Caused by Fungi and Fungus-Like Organisms

First Report of Leaf Spot Caused by *Colletotrichum citricola* on Cavendish Bananas in China

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Cavendish banana (*Musa* spp. AAA group) is an important tropical and subtropical fruit with significant economic value. It is widely planted in Guangxi, Yunnan, Hainan, Fujian, and Guangdong provinces in China. In November 2020, leaf spots were observed on nearly 80% of the plants growing in three Cavendish banana plantations in Chongzuo, Guangxi, China. The symptoms on Cavendish banana leaves initially appeared as small black necrosis spots, which gradually expanded and connected, eventually covering the entire leaf. Three diseased leaves from three plantations were collected, sectioned into small pieces (5 × 5 mm), surface sterilized (10 s in 75% ethanol, followed by 1 min in 1% sodium hypochlorite and rinsing three times in sterile water), and placed on potato dextrose agar (PDA) at 28°C for 5 days for pathogen isolation. The fungal colonies were white, cottony, white on the reverse side, and with concentric circles with yellowish-brown discoloration in 7-day cultures. The conidia were hyaline, aseptate, cylindrical, oval, and measuring 10.3 to 17.71 µm (mean 14.06 ± 1.45 µm; *n* = 200) in length and 4.48 to 9.57 µm (mean 7.46 ± 0.69 µm; *n* = 200) in width. Three

representative isolates (DX1-5, LZ4-5, and FS1-3) were obtained by monospore isolation. The partial internal transcribed spacer (ITS) regions, actin (*ACT*), chitin synthase (*CHS-1*), glyceraldehyde-3-phosphate dehydrogenase (*GAPDH*), calmodulin (*CAL*), and β-tubulin (*TUB2*) were amplified from genomic DNA for the three isolates (Weir et al. 2012). The sequences of the amplified fragments were deposited in GenBank (accessions OL361844 to OL361858 for *GAPDH*, *CAL*, *ACT*, *CHS-1*, and *TUB2* of isolates DX1-5, LZ4-5, and FS1-3; and OL305066 to OL305068 for ITS) and showed over 99% identities with the corresponding sequences of *C. citricola*. A neighbor-joining phylogenetic tree based on the above six genes of type or ex-type specimens of *Colletotrichum* (Fu et al. 2019) was constructed with MEGA 5.2 using the concatenation of multiple sequences (Kumar et al. 2016). All three isolates clustered together with the type culture of *C. citricola* (CBS 134228, CBS 134229, CBS 134230) with 82% bootstrap support in the phylogenetic tree. According to the molecular and morphological characteristics, all three isolates were identified as *C. citricola*. Pathogenicity tests were conducted on 1-month-old, primary hardened tissue culture plantlets. Tender, healthy leaves were gently scratched with a sterile needle, and each wound site was inoculated with sterile cotton impregnated with conidial suspension (10⁶ spores/ml) for each isolate. Wounded leaves were treated with sterile cotton impregnated with conidial suspension of *C. fructicola* as positive controls and sterile water as negative controls. Each isolate was inoculated with three tissue culture plantlets, six inoculated sites on each plantlet, the same as controls. All inoculated tissue culture plantlets were covered with plastic bags to maintain high humidity and placed in a 28°C growth chamber with constant light. Black necrotic lesions were clearly observed on the inoculated leaves and the positive controls after 7 days, whereas no symptoms appeared on the negative control leaves. The fungus was reisolated from inoculated leaves, and these isolates matched the morphological and molecular characteristics of the original isolates, confirming Koch's postulates. To our knowledge, this is the first report of leaf spot caused by *C. citricola* on Cavendish banana worldwide.

References:

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The author(s) declare no conflict of interest.

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