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DISEASE NOTES



# First Report of *Colletotrichum musicola* Causing Soybean Anthracnose in Brazil

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Soybean (*Glycine max* L.) is one of the most important crops worldwide as a source of protein-rich foods and animal feeds. Anthracnose, one of the major limiting factors to soybean production (Dias et al. 2016), is caused by species such as *Colletotrichum truncatum*, *C. sojae*, and *C. plurivorum* (Damm et al. 2009, 2019). In December 2016 and 2017, soybean plants of cultivars Monsoy 8768 and Pioneer y-70 with typical symptoms of anthracnose (necrotic and irregular brown lesions on stems, leaves, and pods) were collected in Mato Grosso, Brazil. Commercial fields sampled showed 10 to 15% incidence of anthracnose in 1 ha in each sampled area. In total, 10 different geographic locations were sampled. *Colletotrichum* strains were isolated and cultured on potato dextrose agar at 25°C with a 12-h light photoperiod from surface-disinfected (70% alcohol followed by 0.5% sodium hypochlorite) plant tissues. Among others, three single-spore isolates (LFN0048 from Sinop, LFN0074 and LFN0090 from Lucas do Rio Verde) showed different morphology; isolates LFN0048 and LFN0074 were selected for further characterization. Total DNA was extracted

and partial glyceraldehyde 3-phosphate dehydrogenase (GAPDH), histone H3 (HIS3), and  $\beta$ -tubulin (TUB2) genes were amplified and sequenced. The sequences were deposited in GenBank (accession numbers MN604249 and MK163893 for HIS3, MN604248 and MK142674 for GAPDH, and MN604250 and MK142675 for TUB) and were compared with most similar reference sequences of *Colletotrichum* (Damm et al. 2019). Both isolates clustered with *Colletotrichum musicola* epitype (CBS 132885), showing 100 and 98.5% similarity in GAPDH, 99.5 and 98.9% in HIS3, and 99.2% in TUB2. On PDA, colonies showed dark-gray aerial mycelium with entire margins, reverse violaceous-black. Conidia and ascospore size and shape match those previously described by Damm et al. (2019): 12.12 to 15.86  $\times$  4.93 to 6.95  $\mu$ m and 15.5 to 19.34  $\times$  5 to 7.84  $\mu$ m, respectively ( $n = 100$ ). Appressoria ( $n = 50$ ) were single or in loose groups, violaceous-black with predominant obovoid, truncated, and cylindrical shapes, with smooth, undulate, or lobate margin, and 9.25 to 29.79  $\times$  7.22 to 21.06  $\mu$ m. Perithecia, paraphyses; and unitunicate eight-spored asci were also observed. Asci were cylindrical to clavate, smooth-walled, and 48.12 to 68.78  $\times$  9.59 to 14.47  $\mu$ m ( $n = 50$ ). Soybean anthracnose is seed-borne (Dias et al. 2018; Rogério et al. 2017); therefore, pathogenicity tests were carried out on pregerminated seeds. Five seeds of Brasmax 8579 cultivar were inoculated with 10  $\mu$ l of a conidial suspension ( $10^6$  conidia/ml) that was placed in the emerging radicle, and five mock-inoculated seeds were used as a control. Seedlings were planted in vermiculite and incubated at 25°C with a 12-h photoperiod. After 7 days, inoculated plants showed necrotic lesions on the cotyledons, leaflets, and hypocotyl, whereas control plants remained asymptomatic. The experiment was repeated three times. *C. musicola* was reisolated from the symptomatic tissues, and the identity was confirmed by morphology and multilocus phylogeny. Until now, *C. musicola* has been reported to be associated with *Musa* sp. (Damm et al. 2019) and *Colocasia esculenta* (Vásquez-López et al. 2019) in Mexico, and with *Phaseolus lunatus* in Brazil (Cavalcante et al. 2018). To our knowledge, this is the first report of *C. musicola* joining a group of new and emergent species of *Colletotrichum* causing anthracnose in soybean-producing regions around the world.

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T. R. Boufleur and R. R. L. Castro contributed equally to this manuscript.

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