Diseases Caused by Fungi and Fungus-Like Organisms

First Report of *Colletotrichum brevisporum* Causing Anthracnose of *Dalbergia odorifera* in China

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Dalbergia odorifera T. Chen (family Fabaceae) is one of four prized species of mahogany plant in China. In June 2017, an investigation of the condition of anthracnose was carried out on approximately 333 ha of *D. odorifera* plantations in Haikou City, Hainan Province (110.19°E, 20.03°N). Approximately 40% of *D. odorifera* plants had disease symptoms. Lesions on leaves were brown to grayish-white, containing black dots and dark-brown borders, occasionally surrounded by a yellowish-green halo. Leaf spots generally occurred along the edge of the leaf. Severely infected leaves became withered and died. Hyphal growth was recovered from symptomatic leaf tissue, surface sterilized with a 75% ethanol solution for 30 s, rinsed with sterile distilled water, plated on potato dextrose agar (PDA), and incubated at 26°C in the dark. The representative isolate JXHTC19 was recovered by transferring a hyphal tip to a fresh PDA plate to obtain a pure culture. Fungal colonies had white aerial mycelium initially, turning pale gray after 3 days. At 7 days, colonies had a cottony appearance ranging from white to dark gray with orange masses of conidia. The colony surface was slimy, and aerial mycelium was sparse. Isolates displayed single-celled, cylindrical, hyaline conidia that were rounded at both ends, and were 9.7 to 16.4 µm long (average 13.5 µm) × 3.6 to 6.2 µm wide (average 4.8 µm) ($n = 100$). To further identify the fungus, genomic DNA was extracted from single conidial cultures of JXHTC19. The *rDNA internal transcribed spacer (ITS)* region, *glutamine synthetase (GS)* gene, partial sequence of *glyceraldehyde-3-phosphate dehydrogenase-like*

(*GAPDH*) gene, *actin (ACT)* gene, and *beta-tubulin (TUB2)* gene were amplified using the primer pairs ITS4/ITS5, GS-F/GS-R, GDF1/GDR1, ACT-512F/ACT-783R, and TUB2-T1/Bt-2b (Weir et al. 2012), respectively. The results showed that the *ITS*, *GS*, *GAPDH*, *ACT*, and *TUB2* genes of the target strain (JXHTC19) have 100, 95, 100, 97, and 98% sequence homology with *C. brevisporum*, respectively. The sequences obtained were deposited in GenBank (MF993572, MN737615, MN737614, MG515612, and MG515615). All five sequences were analyzed together with representative sequences from type or ex-type specimens of the *Colletotrichum* genus (Weir et al. 2012; Yang et al. 2011), and a phylogenetic tree was generated via the neighbor-joining method using MEGA6. The tree placed the isolate in the same group as *C. brevisporum*. Thus, both morphological and molecular characteristics identified the pathogen as *C. brevisporum*. To verify Koch's postulates, 2-year-old leaves of healthy potted *D. odorifera* plants ($n = 6$) were inoculated with a spore suspensions of JXHTC19 that contained 10^5 conidia/ml. Plants were sprayed with water to serve as mock-inoculated controls (Garibaldi et al. 2020). Six plants per treatment were used in each test. The test was repeated once. Plants were incubated in moist chambers at 26°C and monitored daily for symptom development. After 5 days, 11 of 12 isolates caused lesions on all inoculated plants, whereas no symptoms developed on the mock-inoculated controls. Koch's postulates were fulfilled by reisolating the same fungus and verifying its colony and morphological characters as *C. brevisporum*. To our knowledge, this is the first report of this species causing anthracnose of *D. odorifera* in China. Corresponding measures must be adopted to manage this disease, such as reducing the planting density of *D. odorifera* and increasing the species diversity of undergrowth vegetation. These results could help develop better monitoring and management practices for this disease.

References:

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