Diseases Caused by Fungi and Fungus-Like Organisms

First Report of Anthracnose of *Gossypium indicum* Caused by *Colletotrichum theobromicola* in Korea

Donghun Kang, Jungyeon Kim, Younmi Lee, Kotnala Balaraju, and Yongho Jeon[†]

Department of Plant Medicals, Andong National University, Andong 36729, Korea

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In September 2020, typical anthracnose symptoms were observed on cotton (*Gossypium indicum* Lam.) leaves growing in Hahoe Village, Andong, Gyeongbuk Province, Korea. The leaves of the infected plants initially showed spots with halo-lesions which became enlarged and spread to the entire leaf surface area. The infected leaves later became yellowish and chlorotic. The disease incidence was at least 90% in the field. For pathogen isolation, fresh samples collected from symptomatic leaves were cut into small pieces (4 to 5 mm²), surface sterilized in 1% sodium hypochlorite for 1 min, rinsed three times, and macerated in sterile distilled water (SDW). They were spread onto potato dextrose agar (PDA) plates and incubated at 25°C for 5 days under a 12-h photoperiod. Five isolates were recovered from the infected leaves. Purified fungal colonies were initially white, then later turned yellow on PDA medium. Conidia were yellow colored, smooth walled, aseptate, straight or slightly distorted, and cylindrical, with one end slightly acute or with broadly rounded ends, and with size ranges from 15.3 to 17.5 µm (length) by 4.5 to 5.2 µm (width). The morphological characteristics of the present isolates were consistent with those of *Colletotrichum gloeosporioides* (Weir et al. 2012). A single isolate, ANUK97, was selected for identification.

The multilocus sequence analysis of the actin (ACT), calmodulin (CAL), glyceraldehyde-3-phosphate dehydrogenase (GAPDH), internal transcribed spacer (ITS) rDNA, and β-tubulin (TUB2) were amplified by PCR with the primer pairs of ACT-521F/ACT-783R, CL1C/CL2C, GDF/GDR, ITS1/ITS4, and T1/T2, respectively (White et al. 1990). The resulting sequences were deposited in GenBank under accession numbers MW580367 (ACT), MW580368 (CAL), MW580369 (GAPDH), MW580370 (ITS), and MW580371 (TUB2). A nucleotide BLAST search revealed that ACT, CAL, GAPDH, ITS, and TUB2 sequences were 99% similar to accession numbers MN307380.1, MH155176.1, MK796226.1, MW580370.1, and JX010377.1, respectively, of *C. theobromicola*. Maximum-likelihood phylogenetic analysis was conducted based on a combined dataset of ACT, CAL, GAPDH, ITS, and TUB2 sequences using MEGA-X 10.1.8. The isolate ANUK97 was clustered with a representative strain *C. theobromicola* CBS124945 100% bootstrap support. For the pathogenicity test, 2-month-old cotton seedlings ($n = 10$) were inoculated with conidial suspensions (10^6 spore/ml) of *C. theobromicola* obtained from 7-day-old PDA cultures at 25°C by spray method. Seedlings treated with SDW served as controls. Inoculated and control cotton plants were incubated in the greenhouse at 25°C under a 12-h photoperiod. After 7 days, necrotic lesions were observed on the artificially inoculated cotton plants, whereas control plants did not develop any disease symptoms. The pathogen was reisolated from infected cotton leaves but not from control plants to fulfill Koch's postulates. To our knowledge, this is the first report of anthracnose of cotton caused by *C. theobromicola* in Korea.

References:

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[†]Indicates the corresponding author.
Y. Jeon; yongbac@andong.ac.kr