Diseases Caused by Fungi and Fungus-Like Organisms

First Report of *Colletotrichum siamense* Causing Leaf Spot in *Aloe vera* in Bangladesh

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Aloe vera (L.) Burm.f. is a perennial succulent plant commercially cultivated in tropical and subtropical regions for its medicinal, cosmetic, and ornamental uses. During November 2018, leaf spot symptoms were observed on approximately 2- to 3-year-old *A. vera* plants growing in the experimental plots of the National Institute of Biotechnology Campus, Dhaka, Bangladesh. Nearly 100% of the plants were infected. The characteristic symptoms appeared as small (2 to 4 mm) circular dark green lesions found on the upper surface of the leaves. These lesions gradually turned into brown to dark brown spots surrounded by a watery zone. The infected tissue was cut into pieces (~4 mm²), surface sterilized in 3% (w/v) sodium hypochlorite for 2 to 3 min, rinsed with sterilized water three times, plated onto potato dextrose agar medium, and incubated at 28°C for 4 to 7 days in the dark. The fungal colonies were round in shape, cottony in texture, and white to gray in color. The conidia were single-celled, hyaline, smooth-walled, cylindrical with rounded ends, measuring 10.0 to 17.0 µm (mean 14.2 ± 1.3 µm; *n* = 100) in length and 4.5 to 6.2 µm (mean 5.5 ± 0.8 µm; *n* = 100) in width. The morphological characteristics were

consistent with the description of *Colletotrichum siamense* (Weir et al. 2012). For accurate identification, genomic DNA was extracted from a representative isolate, and the internal transcribed spacer (ITS), actin (ACT), glyceraldehyde-3-phosphate dehydrogenase (GAPDH), beta-tubulin (TUB2), microsatellite loci (Apn25L), and calmodulin (CAL) regions were amplified by PCR. The NCBI BLAST comparison of the sequenced PCR products revealed 99.3 to 100% matching with *C. siamense* (NCBI accessions and identity percentage are given in the supplementary material). For the pathogenicity test, a small portion of healthy leaves were gently scratched by emery paper to slightly disrupt the cuticles, and an agar block with the fungal culture was placed on it. The leaf of the inoculated plant was covered with a polythene bag and kept in the greenhouse at 25 to 30°C and 75 to 85% relative humidity. This experiment was performed on three leaves of each of five healthy 2- to 3-year-old plants. After 4 days, characteristic symptoms appeared on the inoculated plants similar to those seen in the field. However, symptoms did not appear on control plants inoculated with agar plugs without fungus. The reisolation of the fungus from the inoculated plants showed identical morphology, which fulfilled Koch's postulates. *C. siamense* exhibits pathogenicity to a number of plants across the world including cacao (Serrato-Diaz et al. 2020), *Euonymus japonicas* (Wu 2020), partridge tea (Liu et al. 2018), and cassava (Oliveira et al. 2018). Because they are genetically very close and significantly differ in infectivity and fungicide sensitivity (Hu et al. 2015; Munir et al. 2016), accurate molecular identification of *Colletotrichum* species is essential in the management of the disease. To our knowledge, this is the first report of *C. siamense* causing leaf spots of *A. vera*.

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