

Diseases Caused by Fungi and Fungus-Like Organisms

First Report of *Colletotrichum tropicale* Causing Anthracnose on Pitaya (*Hylocereus costaricensis*) in Brazil

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Pitaya, *Hylocereus costaricensis*, is a species of the Cactaceae family and originated in the Americas (Ortiz and Livera 1995). It has been cultivated in Brazil and has shown great potential for fruit production and is currently present in several markets (Faleiro et al. 2021). In July 2018, infected plants of pitaya with symptoms of anthracnose were obtained from an orchard in Fortaleza, Ceará, Brazil (3°44'24.50"S, 38°34'30.80"W), with 50% disease incidence. The symptoms observed consisted of well-defined and depressed stains that initially appeared as reddish-orange spots and were surrounded by a border of dark-brown color. As the lesion progressed, the center became light brown or whitish in color, with black dots appearing later. Four cladodes were collected with anthracnose symptoms. The pathogen was isolated from symptomatic cladodes, which were surface-disinfected with 1% v/v NaClO and 70% v/v ethanol, rinsed with sterile distilled water, transferred onto potato dextrose agar (PDA) medium, and incubated under a light/dark (12 h/12 h) photoperiod. Two isolates were recovered from the lesions on cladodes. Pure cultures were obtained from single conidia produced on colonies grown on PDA medium using an inoculation needle under a microscope. Colonies on PDA exhibited white aerial mycelia with an orange conidial mass. The colonies were light gray on the front and light orange on the reverse of the plate. Morphological features suggested that the isolates had the same characteristics as previously described for *Colletotrichum* spp. (Weir et al. 2012). In order to identify the species of the isolates, the genomic DNA of the isolates UFCM 0684 and UFCM 0685 was extracted

using the cetyltrimethylammonium bromide method, and the internal transcribed spacer (ITS) region, β -tubulin (*TUB2*), actin (*ACT*), glutamine synthetase (*GS*), and glyceraldehyde-3-phosphate dehydrogenase (*GAPDH*) gene fragments were amplified. PCR products were sequenced, and the resulting sequences were submitted to phylogenetic analyses based on maximum likelihood for the combination of the genes. The isolates grouped within *Colletotrichum tropicale* with 99% bootstrap support. The sequences obtained in this study were deposited in GenBank with accession numbers OL799311 and OL799312 for *ACT*, OL799313 and OL799314 for *TUB2*, OL799315 and OL799316 for *GAPDH*, OL799317 and OL799318 for *GS*, and OL799319 and OL799320 for ITS. After that, the isolate UFCM 0685 was selected to study for further characterization. Conidia ($n = 50$) were 13.7 μ m (length) \times 4.7 μ m (width) on average, hyaline, aseptate, and cylindrical. To fulfill Koch's postulates, pathogenicity tests were performed in a moist chamber for 1 week at 25°C with 80% relative humidity on a 12-h fluorescent light/dark photoperiod. The cladodes were wounded using a sterilized needle and inoculated with 10 μ l of a conidial suspension (1×10^6 conidia/ml) on three cladodes with five wounds each. The same number of uninoculated cladodes served as controls. The experiment was performed twice. Two weeks later, all the inoculated cladodes showed necrotic symptoms, which were similar to the symptoms previously observed in the field. The uninoculated cladode remained symptomless. The fungus was reisolated from the inoculated cladode, and its morphological characteristics were similar to the original isolate. *C. tropicale* has been reported to cause anthracnose on *H. costaricensis* in Mexico (Nuñez-García et al. 2023) and on *H. undatus*, *H. monacanthus*, and *H. megalanthus* (Evallo et al. 2022). To the best of our knowledge, this is the first report of anthracnose caused by *C. tropicale* in *H. costaricensis* in Brazil.

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