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DISEASE NOTES

First Report of Colletotrichum theobromicola and C. tropicale Causing Anthracnose on Fruits of Carnauba Palm in Brazil

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The carnauba palm (Copernicia prunifera [Mill.] H.E.Moore) is used for the production of industrial and artisanal commodities (Gomes and Nascimento 2006). Between 1990 and 2006, the northeast region produced 66,067 t of carnauba wax (Alves and Coelho 2008). In a previous report, the etiology of anthracnose on carnauba palm fruits was attributed to C. gloeosporioides (Freire and Barguil 2009). This study aimed to characterize the etiological agents of anthracnose on carnauba fruits collected in Ceará State, Brazil. Carnauba fruits at physiological maturation stage (yellowish-green color), showing severe anthracnose symptoms, were collected in Bela Cruz, Caucaia, and Paraipaba municipalities. A representative isolate to each municipality was selected for the morphological, molecular, and pathogenic characterization (UFCM 0630-Caucaia; UFCM 0631-Bela Cruz; and UFCM 0632-Paraipaba). The isolates were grown on potato dextrose agar medium at 25°C and a 12-h photoperiod and the morphological characteristics were observed after 4 days of incubation. Fungal DNA of the isolates was extracted (Murray and Thompson 1980), and fragments of the ITS rDNA (ITS), actin (ACT), β -tubulin (TUB2), glyceraldehyde-3-phosphate dehydrogenase (GAPDH), and glutamine synthetase (GS) genomic regions (Prihastuti et al. 2009) were amplified and sequenced in both directions (GenBank accession nos. MF289371-85). Multiple alignments of the combined dataset of the isolates and representative sequences obtained from GenBank were phylogenetically analyzed by neighbor-joining and bootstrap with 1,000 replicates. The pathogenicity of the

isolates was assessed on carnauba (n = 10, yellowish-green stage) and mango cv. Tommy

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Atkins (n = 3, yellowish-orange stage) fruits, and on susceptible cashew plantlets (clone BRS 265) (n = 3, 120 days old). Wounded fruits were inoculated with 3-mm agar plugs containing mycelium of the isolates, while control treatments had only the agar. Spore suspensions (2 \times 10⁶ conidia ml⁻¹) were sprayed on young leaves of the cashew plantlets, while only water was sprayed on control plants. The UFCM 0630 and UFCM 0632 isolates formed pale olive-gray aerial mycelium and olive-gray pigment in the back of the colonies. Conidia were cylindrical, 5.4 to 13.8×1.5 to 4.7 (mean = 9.0 × 2.9, n = 50) µm. Appressoria were subglobose to elliptical, 2.8 to 5.8×2.4 to 3.1 (mean = 4.8×2.8 , n = 10) µm. The UFCM 0631 isolate formed dark gray aerial mycelium and dark greenish-gray pigment in the back of the colonies. Conidia were cylindrical, 13.0 to 17.5×2.3 to 5.5 (mean = 12.1×3.7 , n = 50) µm. Appressoria were subglobose to clavate, 6.0 to $9.5 \times$ 4.2 to 7.6 (mean = 7.8×5.9 , n = 10) μ m. In the phylogenetic tree, the isolates of C. prunifera grouped in two clades of the C. gloeosporioides species complex. UFCM 0630 and UFCM 0632 isolates grouped in C. tropicale and UFCM 0631 grouped in C. theobromicola with 100% bootstrap support to both clades. Koch's postulates were completed on inoculated carnauba fruits to all three isolates representing C. tropicale and C. theobromicola. Only UFCM 0630 and UFCM 0632 isolates were pathogenic to mango fruits and cashew plantlets. This study shows evidence that carnauba palm may serve as an alternative host of C. tropicale and inoculum source for mango and cashew anthracnose in orchards established near to natural areas of carnauba. To our knowledge, this is the first report of C. tropicale and C. theobromicola causing anthracnose on carnauba in Brazil.

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