Diseases Caused by Fungi and Fungus-Like Organisms

Ginger Wilt and Rot Disease Caused by *Ceratocystis fimbriata* in China

R. Q. Zhang, K. C. Xu, X. Li, Y. Gao, Y. X. Sun, and Q. Huang[†]

State Key Laboratory for Conservation and Utilization of Bio-Resources in Yunnan, Yunnan Agricultural University, Kunming, 650201, China

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Ginger (*Zingiber officinale* Rosc.) is an herb that has been grown in China for more than 2,500 years. It can be used as both a spice and a therapeutic drug. In July 2013, ginger plants were found to have wilting symptoms and yellowing leaf tips in a farm in Kunming city of Yunnan province (25.02 N, 102.42 E), southwest China, and we also found gray-black lesions on the surface of the harvested ginger in a market in Kunming. Initial symptoms on harvested ginger appeared as gray-black mycelia growth on the surface of the harvested ginger, which enlarged and extended internally. Carrot baiting was used to isolate the pathogen from rotted ginger and from diseased ginger leaves (Moller and Devay 1968). After 2 weeks, spores developing from perithecia on the carrot pieces were transferred to malt extract agar (MEA) and incubated at 25°C constant temperature. Six single-spore isolates (ZOR-1 to ZOR-6) were obtained, and the isolates were stored in 15% glycerol at -80°C in a refrigerator in the State Key Laboratory for Conservation and Utilization of Bio-Resources in Yunnan Agricultural University. Cultures varied in color from white to brownish green to brown. For all measurements, $n = 50$. Blackish brown, globose perithecia (131.9 to 186.0 × 138.5 to 188.3 μm) with a long black neck (400.2 to 644.7 μm) were immersed, partially embedded or superficial on the substrate. Ascospores were globose or had a “hat-like” morphology typical of *Ceratocystis fimbriata* and were 4.0 to 5.3 × 4.8 to 6.2 μm. Endoconidia were cylindrical and clavate (2.9 to 7.4 × 7.5 to 32.8 μm), conidia were barrel-shaped (4.4 to 10.4 × 6.2 to 12.9 μm), and chlamydospores were smooth, blackish brown, ovoid or obpyriform (8.42 to 12.21 × 10.47 to 17.65 μm) (Engelbrecht and Harrington 2005; Webster and Butler 1967). Genomic DNA was extracted from two isolates (ZOR-1, ZOR-

2) using the CTAB method (Lee and Taylor 1990). The internal transcribed spacers (ITS) region of rDNA was amplified using primers ITS1F/ITS4 (Thorpe et al. 2005). The nucleotide sequences of ZOR-1 and ZOR-2 (GenBank accessions KJ511490 and KJ511491) were 100% homologous to those of the isolates of *C. fimbriata* from diseased *Cucumis sativus* L. and *Punica granatum* L. in China (GenBank accessions MH535909 and KT963159). Thus, the pathogen was identified as *C. fimbriata*. Pathogenicity tests were made on fresh ginger rhizomes in the laboratory; the pathogen was cultured for 14 days on MEA (ZOR-1, ZOR-2), which were washed with sterilized water, and the resulting spore suspensions diluted to 1.0×10^6 spores/ml. Wounds (0.5 × 0.5 cm) were made on the surface of healthy mature ginger rhizomes by scraping with a sterile scalpel and were then treated with 100 μl of spore suspension. Control ginger rhizomes were coated sterile water. Ginger rhizomes were stored at room temperature. Each treatment was performed in triplicate. After 5 days, gray-black mycelia developed on the rhizome surface, becoming a visible black mold after 1 week. We reisolated the pathogen from infected tissues but not from the controls. In the greenhouse, 20 ml of 1.0×10^6 spores/ml suspensions from isolates ZOR-1 and ZOR-2, or sterile water, were injected into 2-month-old ginger seedlings in triplicate. The inoculated site on the stem turned black in 5 days. Six weeks after inoculation, the inoculated plants developed yellowing leaves and wilting symptoms. The same fungus was reisolated from inoculated plants but not from the controls. According to Koch's postulates, the inoculated strains of ZOR-1 and ZOR-2 were the pathogens causing ginger wilt and rot disease. To the best of our knowledge, ginger is a new host plant of *C. fimbriata* from China. In recent years, we have found that this disease incidence was approximately 5 to 10% of the farmland and 5 to 15% of the stored condition, respectively, in Yunnan Province. If not prevented, ginger production in China will be affected.

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[†]Indicates the corresponding author.
Q. Huang; huangqiong88hs@163.com