

## Diseases Caused by Fungi and Fungus-Like Organisms

### First Report of *Colletotrichum gloeosporioides* Causing Leaf Spot on *Cyclobalanopsis glauca* in China

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*Cyclobalanopsis glauca* (Thunb.) Oerst. is one of the most widely distributed species of evergreen broad-leaved tree in subtropical areas of China. It is also grown in Korea, Japan, and India. Because of its beautiful shape, *C. glauca* is commonly used for greening gardens and walkways. In July 2018, leaf spots on *C. glauca* were observed in Zhejiang province (Lishui, N 28°26'6.75"; E 119°54'11.22"), China. About 70% of the trees were found to be diseased, with approximately 50% of leaves showing symptoms. The symptoms on *C. glauca* leaves initially appeared as small brown-yellow spots, which gradually expanded, developing a light brown center and dark brown to black margin. The spots ranged from 4 to 15 mm in diameter. Ten symptomatic fragments measuring approximately 5 × 5 mm from each leaf were surface disinfested with 70% ethanol for 30 s, and then they were rinsed in sterile distilled water and placed on potato dextrose agar medium at 25°C in the dark for 5 days. Segments of colony perimeters were then transferred to new plates. The colonies initially produced white mycelia that later turned gray-white, with pink and occasionally black dots scattered on the surface of the mycelium. Spores were aseptate, cylindrical, 8 to 15 µm in length, and 3

to 5 µm wide, most with rounded ends, a few with one apex round and the other fusiform, as described for *Colletotrichum gloeosporioides* (Penz.) Sacc. (Agostini et al. 1993). The internal transcribed spacer rDNA (ITS, MK758005) and two nuclear protein-coding genes (CHS, MK784770; ACT, MK784769) were amplified with ITS1/ITS4, CHS-79F/CHS-345, and ACT-512F/ACT-783R, respectively (Weir et al. 2012). The sequence had 99.61% identity to GQ485605 for ITS, 99.56% to GQ856782 for ACT, and 100% to GQ856733 for CHS of *C. gloeosporioides* CBS 953.97 in GenBank, respectively. To fulfill Koch's postulates, spores ( $1 \times 10^8$ ) of the isolate were sprayed onto leaves of twelve 2-year-old *C. glauca* plants (at least six leaves per plant). The fungus was inoculated on one side of each leaf, and distilled water was used as a mock-inoculated control on the other side. The plants were cultivated in the greenhouse to maintain high humidity and a temperature near 25°C. After 9 days, 100% of the leaf halves that had been inoculated had symptoms identical to those observed on affected *C. glauca* leaves in the field, whereas no symptoms were observed on the mock-inoculated half of each leaf. The fungus was reisolated from the symptoms and identified as *C. gloeosporioides* using techniques previously described. To our knowledge, this is the first report of *C. gloeosporioides* infecting *C. glauca* in China. This study will establish a foundation for the further study of *C. gloeosporioides* to address the disease effectively.

#### References:

- Agostini, J. P., et al. 1993. *Phytopathology* 82:1177.  
Weir, B. S., et al. 2012. *Stud. Mycol.* 73:115.

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#### e-Xtra

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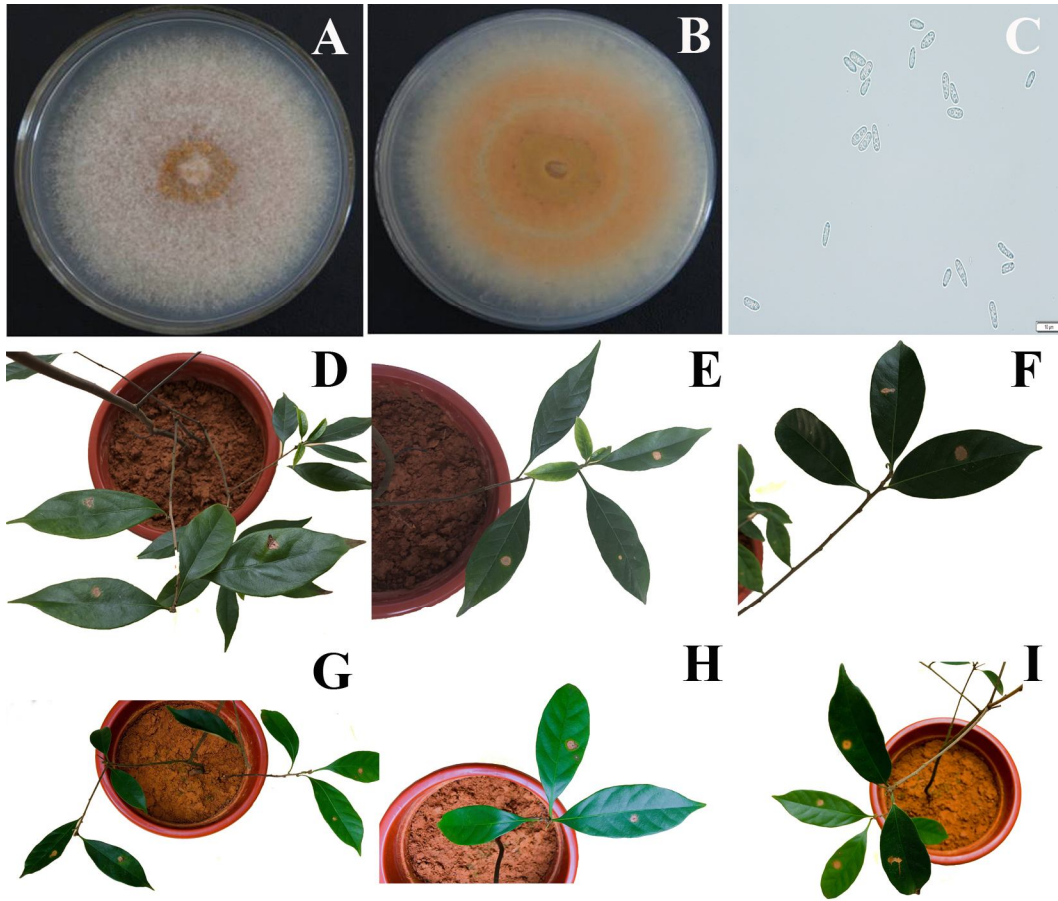


Figure 1. A, B: the colony of *C. gloeosporioides*; C: the conidia of *C. gloeosporioides*; D-I: the plants infected with *C. gloeosporioides* in the greenhouse, one side of each leaf was inoculated with the microorganisms, the other side was sprayed with distilled water as a control.

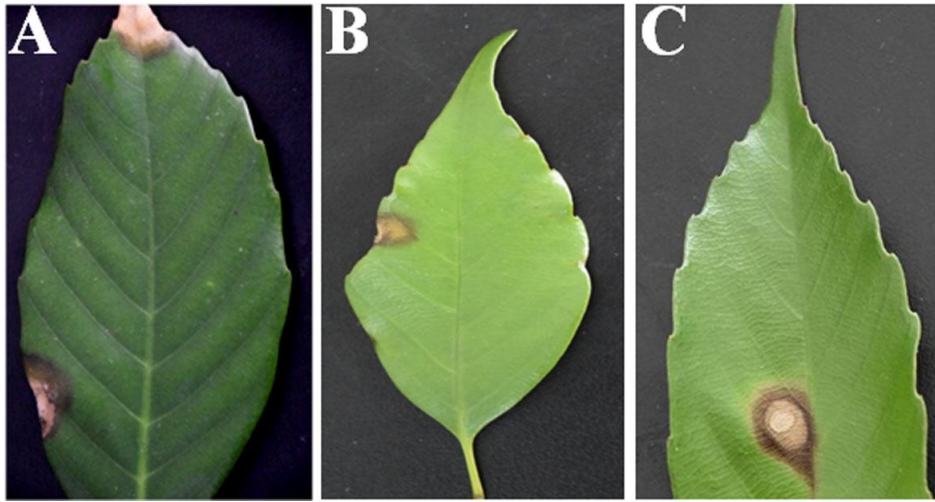


Figure 2. A and B: leaves of *C. glauca* infected with *C. gloeosporioides* in the field; C: the detached leaf inoculated with *C. gloeosporioides*, the left side of the leaf was sprayed with the microorganism, distilled water was used on the right side as a control.