Diseases Caused by Fungi and Fungus-Like Organisms

First Report of Anthracnose on *Camellia japonica* Caused by *Colletotrichum siamense* in Zhejiang Province, China

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Camellia japonica is an attractive flowering woody plant with great ornamental and medicinal value in China. However, typical anthracnose lesions on the leaves are usually observed in summer in Zhejiang province. One hundred trees were investigated with over 70% leaf disease incidence. The symptom initially develops from the tip or edge of the leaf and dark green infected spots appear. The diseased spots expand and become yellow brown. The lesions are covered with abundant, small, black acervuli at the center with yellow edges. The diseased leaves become brittle, cracked, and finally fall off. Sixty leaves with typical anthracnose symptoms were sampled from gardens in Lin'an, Zhejiang province. The diseased tissues were cut into pieces and incubated in moist chambers at 25°C. The spore mass was collected using a sterile needle under a dissection microscope and put on 2% malt extract agar (MEA). The cultures were incubated at 25°C in the dark for one week (Hou et al. 2016). Thirty single spore cultures were obtained and grown on 2% MEA at 25°C for morphological characterization. White aerial mycelia and black conidiomata with orange masses of conidia developed 7 days later. Conidia were cylindrical in shape, 12 to 19 µm, mean lengths ranging from 15.5 ± 1.0 to 16.0 ± 1.2 µm. The morphological characteristics are consistent with those of *Colletotrichum* species (Guarnaccia et al. 2017). DNA was extracted from three selected isolates (HT-71, J-5, J-20) for sequencing. The partial regions of ribosomal internal transcribed spacers (ITS), calmodulin (CAL), glyceraldehyde-3-phosphate dehydrogenase (GAPDH),

actin gene (ACT), beta-tubulin (TUB2), Apn2-Mat1-2 intergenic spacer and partial mating type gene (ApMat), and glutamine synthetase (GS) were amplified as described by Liu et al. (2015). Sequences of the above seven loci for the selected isolates were obtained and deposited in the GenBank database (MZ014901 to MZ014905, MZ514915 to MZ514922, MZ514925 to MZ514930, MZ497332 and MZ497333). BLAST results indicate they represent *Colletotrichum siamense*. Multilocus phylogenetic analysis including ex-type of *C. siamense* (ICMP18578 = CBS130417) and related species was conducted using the Maximum Likelihood method, and *C. acutatum* (CBS 112996) served as the outgroup (Vieira et al. 2019). The three obtained isolates clustered with the ex-type isolate of *C. siamense*. Eight leaves on two *Camellia* plants were inoculated to confirm the pathogenicity in the field. The leaves were surface sprayed with 75% ethanol and dried with sterilized filter paper. The leaves were inoculated using the wound/drop inoculation method: an aliquot of 10 µl of spore suspension (1.0×10^6 conidia per ml) was dropped on the left side of a leaf after wounding once by pin-pricking with a sterilized needle. Sterile water was dropped on the right side of the same leaf in parallel as control (Fu et al. 2019). The initial symptoms were observed 7 days later; all inoculated leaves developed lesions similar to those observed in the field, and no symptoms were observed in the control. The fungus was successfully reisolated only from lesions inoculated with spore suspension exhibiting morphological characteristics resembling those in *C. siamense*, and further confirmed with sequence data. To our knowledge, this represents the first report of anthracnose on *C. japonica* caused by *C. siamense* worldwide. Confirmation of this pathogen in the region will be helpful for the disease management on *C. japonica*, considering the previous report of *C. camelliae-japonicae* on the same host.

References:

- Fu, M., et al. 2019. Persoonia 42:1.
- Guarnaccia, V., et al. 2017. Persoonia 39:32.
- Hou, L. W., et al. 2016. Mycosphere 7:1111.
- Liu, F., et al. 2015. Persoonia 35:63.
- Vieira, A. D. S., et al. 2019. Mol. Phylogenet. Evol. 143:106694.

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