Diseases Caused by Fungi and Fungus-Like Organisms

First Report of Cobweb Disease in *Oudemansiella raphanipes* Caused by *Cladobotryum varium* in Beijing, China

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Oudemansiella raphanipes is an edible mushroom with medicinal properties, which has been recently cultivated throughout China (Hao et al. 2016). In October 2019, a disease with symptoms similar to that of cobweb disease (Carrasco et al. 2017) was observed in *O. raphanipes* in the Tongzhou District, Beijing, China, infecting 25% of the fruiting bodies. A white cotton-like net of hyphae was present typically on the casing soil or on the stipe of the fruiting bodies; they gradually spread to the pileus, covering the fruiting body, which eventually wilted and died, resulting in yield reduction and economic loss. Cultures were obtained by aseptically transferring the diseased fruiting bodies onto potato dextrose agar at 25°C; they were deposited in the culture collection (JZBQA1) of the Beijing Academy of Agriculture and Forestry Sciences, China. The colonies were pale white/white, with an occasional formation of yellow diffusing pigments on the reverse side. Conidiophores were *Cladobotryum*-like, phialides were solitary or commonly divergent in whorls of two to three (to four), lageniform to subulate, 20 to 63.5 (to 66) × (3.8 to) 4 to 5.3 (to 9) μm (*n* = 40); conidia were hyaline, oval to ellipsoidal, with one or two septa, (10.4 to) 11.4 to 20 (to 22) × 6.6 to 9.5 (to 10) μm (*n* = 40); chlamydospores were globose or ellipsoidal. The morphological characteristics were consistent with those of *Cladobotryum varium* (Back et al. 2012a, b; Sun et al. 2019). For species-level fungal identification, genomic DNA was

extracted using the DNeasy Plant Mini Kit (Qiagen, U.S.A.). The internal transcribed spacer regions, translation elongation factor 1 alpha exon, RNA polymerase II subunit b, and RNA polymerase I largest subunit genes were amplified using the primer pairs ITS1/ITS4 (White et al. 1990), EF1-983F/2218R (Rehner and Buckley 2005), RPB2-5F/7cR (Liu et al. 1999), and RPB1F1 (5'-GCCGATGAAGTTGGTCTA-3')/RPB1R1 (5'-TATGTTGCGGTGAGCCTT-3'), respectively. A BLAST nucleotide search showed 99.34% (449/452 bp), 99.24% (914/921 bp), 98.08% (1,022/1,042 bp), and 99.66% (588/590 bp) homology, respectively, with those of the ex-type culture of *Hypomyces aurantius* TFC 95-171 (FN859425.1, FN868743.1, FN868679.1, and FN868805.1). The four sequences were deposited in GenBank (accession nos. MW534093, MW560066, MW560064, and MW560065). Phylogenetic trees based on the assessed gene loci revealed that the JZBQA1 strain was closely related to *C. varium*. An in vivo pathogenicity test was performed using the fruiting bodies. Spore suspension (10⁸ spores/ml) of the JZBQA1 strain or sterile distilled water was sprayed on six healthy fruiting bodies, which were maintained in an artificial climate chamber at 24 to 26°C. Cobweb-like features were observed on the fruiting bodies treated with the spore suspension 2 to 3 days postinoculation, whereas those treated with water did not exhibit such features. The same pathogen was reisolated and confirmed from the infected fruiting bodies by integrated analysis of morphological characteristics and gene sequencing data. *Cladobotryum* spp. infects different varieties of cultivated edible mushrooms, resulting in the development of cobweb diseases (Cao et al. 2020; Carrasco et al. 2017). *C. varium* is the causal agent of cobweb disease in *Flammulina velutipes* and *Hypsizygus marmoreus* (Back et al. 2012a, b). To our knowledge, this is the first report of cobweb disease caused by *C. varium* in *O. raphanipes*. This finding is a valuable contribution to the knowledge of cobweb disease development in edible fungi.

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