Diseases Caused by Fungi and Fungus-Like Organisms

Root Rot of *Codonopsis tangshen* Caused by *Ilyonectria robusta* in Chongqing, China

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Codonopsis tangshen Oliv. belongs to the Campanulaceae and is one of the most economically important medicinal materials in China. It is used in medical and agricultural applications (Wu et al. 2020). In August 2019, root rot of *C. tangshen* was first observed in Fengjie, Chongqing City, southwest China (30°45'59" N, 109°36'36" E), causing approximately 20% yield loss. At the initial stage of the disease, the aboveground stems and leaves turn yellow, and brown to black spots of different sizes appear at the base or root of the stem. With further development of the disease, the aboveground leaves gradually turn yellow as the diseased spots rot from bottom to top, so that they die, and the diseased spots on the roots expand and begin to rot. Generally, they gradually rot from the bottom up, but the vascular bundles are occasionally normal. If the symptoms on *C. tangshen* start too late, and the root has not completely rotted by late autumn (late October to early November), the rest of the root will not continue to rot, and it is called half *C. tangshen*. The next spring, the half *C. tangshen* can continue to sprout, but it will continue to rot, which will seriously affect the yield and quality. To identify the pathogen, 25 samples of diseased plants were collected, and symptomatic rhizome tissues were surface disinfected with 0.1% HgCl₂ solution for 30 s, rinsed in sterilized water three times, placed on potato dextrose agar (PDA), and incubated at 25 ± 1°C in the dark. After 7 days of culture on PDA, the center appeared light yellow, the edges were white, and the aerial hyphae were felt-like. The surface of the colony was reddish brown, and the margins were white and regular. The conidiophores were simple, usually born on

the lateral or apical sides of aerial mycelium, and unbranched or minimally branched. Conidia were abundant, cylindrical or rod-shaped, straight or slightly curved, and usually had one to three septa. Macroconidia varied in size depending on the number of cells as follows: one-septate, 15.3 to 26.3 × 4.2 to 7.3 μm (*n* = 50); two-septate, 20.5 to 30.5 × 4.9 to 7.8 μm (*n* = 50); and three-septate, 29.3 to 38.5 × 5.5 to 7.4 μm (*n* = 50) (round at both ends). For molecular identification, DNA was extracted from a representative isolate using a fungus genomic DNA extraction kit (Solarbio, Beijing, China). The internal transcribed spacer (ITS) (ITS1/ITS4) (White et al. 1990), beta-tubulin (TUB2) (BT2A/BT2B) (O'Donnell and Cigelnik 1997), translation elongation factor 1-α (TEF) (EF446F/EF1035R) (Inderbitzin et al. 2010), DNA-dependent RNA polymerase subunit II gene (RPB2) (O'Donnell et al. 2010), and histone H3 (HIS3) (CYLH3F/CYLH3R) (Crous et al. 2004b) were amplified. BLAST results indicated that the ITS, TUB2, TEF, HIS3, and RPB2 sequences (GenBank MW392103, MW386994, MW386995, MW392103, and MW915473, respectively) showed 96 to 100% identity with *Ilyonectria robusta* sequences at NCBI (GenBank KU350726, JF335378, MN833103, MN833113, and KM232336, respectively). A phylogenetic tree was inferred from the combined datasets (ITS, TEF1, TUB, and HIS3) from members of the *I. robusta* species complex analyzed in this study (Cabral et al. 2012). To complete Koch's postulates, a conidial suspension (10⁶ spores/ml) collected from isolate CQ13 was irrigated onto 15 annual *C. tangshen* potted plants. Sterile water was used as a negative control, and the pathogenicity assay was repeated three times. Following inoculation, the plants were cultured for 9 days at 75% relative humidity and 25°C. The inoculated plants showed symptoms similar to those observed in the field, but the negative control plants were healthy and unaffected. *I. robusta* was reisolated from the infected tissues and identified by morphological characteristics and DNA sequence analysis. To our knowledge, this is the first report of *I. robusta* causing root rot disease of *C. tangshen* in China. Our results may help to take appropriate steps to control the disease in the commercial area of *C. tangshen*.

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