Diseases Caused by Fungi and Fungus-Like Organisms

First Report of Southern Blight Caused by *Sclerotium delphinii* on *Euonymus fortunei* in China

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Euonymus fortunei is an evergreen shrub-vine in the family Celastraceae, widely used as a groundcover or a vine to climb walls, or traditional herbal medicine in China. In August 2019, typical southern blight symptoms that included basal stem rot and the presence of sclerotia in rotted tissue were observed on *E. fortunei* in Kunshan city, Jiangsu province, China. Disease incidence was estimated at approximately 15 to 20%; meanwhile, approximately 30 to 40% of diseased plants died. The infected plants showed brown to dark stem necrosis near the base, leaf yellowing, and wilting. White mycelia and white to dark reddish-brown sclerotia were observed at the base of the stem and rotten tissue. To isolate the causal organism, infected stem tissue and sclerotia collected from diseased plants in a median strip in Kunshan (31°23'40"N, 120°54'57"E) were disinfected with 70% ethanol for 2 to 3 s, followed by 2 min in 5% NaClO, rinsed three times with sterile water, then plated on potato dextrose agar (PDA) medium, and incubated at 25°C. Isolated colonies were subcultured by needle tip transfer 3 days later. Isolates had white mycelia on PDA, with a radial growth rate of 15.2 to 18.7 mm/day. White and orange sclerotia were developed after 5 to 8 days and eventually turned dark reddish-brown. The sclerotia were globoid or irregular with surface markings (1.4- to 4.3-mm diameter; mean = 2.59 mm; *n* = 50) on PDA, and the average number of sclerotia produced per Petri dish ranged

from 35 to 85 (mean = 52; *n* = 10). Microscopic observations found septal hyphae and clamp connections. These morphological features were identical to the description of *Sclerotium delphinii* (syn. *Sclerotium rolfsii* var. *delphinii*) (Mukherjee et al. 2015; Punja and Damiani 1996; Stevens 1931). A representative isolate, YKY2020.01, was stored in the Key Laboratory of National Forestry and Grassland Administration on Ecological Landscaping of Challenging Urban Sites in Shanghai. For molecular identification, DNA of the isolate YKY2020.01 was extracted using the Fungal DNA Kit (Omega Bio-tek, China). The internal transcribed spacer region (ITS fragment including ITS1, 5.8S rDNA, and ITS2 region) was amplified with primers ITS1/ITS4 (White et al. 1990) and then sequenced by Sangon Biotech (Shanghai, China). BLAST analysis in NCBI found the ITS sequence of YKY2020.01 (MW916955) was 99.84% similar to *S. delphinii* strain CBS272.30 (MH855140). Phylogenetic analysis using the maximum likelihood method placed isolate YKY2020.01 in the same clade as *S. delphinii*. To evaluate pathogenicity, hyphal blocks (0.7-cm diameter) were placed at the base of the stem of healthy *E. fortunei* (*n* = 5 plants). Five healthy plants were inoculated by uncolonized agar blocks as controls. All plants were kept in a greenhouse with a temperature range from 21 to 25.6°C (mean = 24.9°C) and relative humidity of 50%. Inoculated plants were symptomatic after 3 days and wilted after 12 days. Symptoms in inoculated plants were similar to those observed under natural conditions, whereas the control group remained asymptomatic. The fungal pathogen was reisolated from symptomatic tissue and confirmed as *S. delphinii*. To the best of our knowledge, this is the first report of *S. delphinii* causing southern blight on *E. fortunei* in China and worldwide. This finding provides concise and practical information on the newly emerged disease of *E. fortunei*, which is beneficial for future disease management.

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