First Report of White Rot on *Malus coronaria* caused by *Botryosphaeria dothidea* in Korea

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Sweet crabapple [*Malus coronaria* (L.) Mill.], an ornamental tree with a geographic origin in China, has been widely cultivated in Korea as a street or garden tree, because of its beautiful flowers. White rot has occurred since 1990 on the branches, bark, and fruits of sweet crabapple growing at the campus of Gyeongsang National University, Jinju, Korea. Cankers or warts initially appeared as slightly sunken reddish-brown areas on the bark of sweet crabapple. Cankers expanded to 30 cm in length, and branches became weakened and eventually died (Fig. 1A). The disease mainly occurred on the branches, bark, and fruits, but not on the leaves (Fig. 1B–F). As the fruits enlarge, a brown decayed area appeared. The decay was often characterized by a series of concentric rings, alternating from black to brown on the fruit surface (Fig. 1D and E). Eventually, the rotted fruits dried out and mummified, sometimes hanging on the tree until the next season (Fig. 1F). Pseudothecia, 50–120×50–110 µm in size, were produced on cankered areas of bark and on mummified fruits that had been present on the tree for over a year (Fig. 1G). The asci were cylindrical, club- and banana-shaped, and 80–120×10–12 µm in size (Fig. 1H and I). The ascospores were ovoid and 20–30×7–12 µm in size (Fig. 1I). Pycnidia, 100–300×100–350 µm in size, produced on the PDA plates under natural light (Fig. 2A and B). The pycnidiospores were cylindrical or oval and 5–7×20–30 µm (Fig. 2C). For pathogenicity testing, 5-day-old mycelium grown on PDA was artificially inoculated on fruits of sweet crabapple. The same fruit rot symptoms were reproduced and the fungus was re-isolated from the symptoms to prove Koch's postulates (Fig. 2D).

PCR amplification and sequencing of the complete internal transcribed spacer (ITS) rDNA was performed using the primers ITS1 and ITS4 as described by White et al. (1990). The resulting

Fig. 2. Morphological characteristics of white rot on *Malus coronaria* caused by *Botryosphaeria dothidea*. A: Granular masses resulting in pycnidia on PDA plate. A small box shows pycnidia. B: Pycnidia (bar=100 µm). C: Pycnidiospores (bar=10 µm). D: A fruit of sweet crabapple artificially inoculated with fungal pathogen.

582-bp sequence has been deposited in GenBank as Accession No. HQ231407. Phylogenetic analysis was performed with MEGA4 software using the neighbor-joining method and the Tajima-Nei distance model. The representative isolate showed only one base substitution with sequences from *Botryosphaeria dothidea* (GQ870285), which infects apple trees (Fig. 3). Mycological measurements, taxonomic characters, pathogenicity, and molecular data coincided with those of *Botryosphaeria dothidea* (Moug.) Ces. & De Not. described by Sutton (1990). To our knowledge, this is the first report of the presence of *B. dothidea* on sweet crabapple in Korea (The Korean Society of Plant Pathology, 2009). The representative culture of *B. dothidea* BD001 has been deposited at National Academy of Agricultural Science (KACC 45481). Recent occurrence of the disease suggests that *B. dothidea* is spreading widely and poses a serious threat to the health of sweet crabapple in Korea.

Fig. 3. Phylogenetic relationships among *Botryosphaeria dothidea* isolates, including a canker infecting *Malus coronaria* (bold), based on a similarity analysis of ITS sequences. The numbers above the branches indicate bootstrap values. Bars indicate the number of nucleotide substitutions per site.

Fig. 1. Symptoms and morphological characteristics of white rot on *Malus coronaria* caused by *Botryosphaeria dothidea*. A: The infected sweet crabapple tree showing signs of canker. B and C: Warts on bark and branches of sweet crabapple tree. D and E: Natural symptoms of fruit rot of sweet crabapple. F: A mummified fruit. G: Pseudothecium collected from the diseased fruits (bar=100 µm). H: Asci bearing mature or immature ascospores (bar=20 µm). I: Ascus bearing mature ascospores (bar=10 µm).

References

- Sutton, T. B. 1990. White rot. In: *Compendium of Apple and Pear Diseases*, ed. by A. L. Jones and H. S. Aldwinckle, pp. 76–77, APS Press.
- The Korean Society of Plant Pathology. 2009. *List of Plant Diseases in Korea*, 5th ed. 853 pp. (in Korean).
- White, T. J., Bruns, T., Lee, S. and Taylor, J. W. 1990. Amplification and direct sequencing of fungal ribosomal RNA genes for phylogenetics. In: *PCR Protocols: A Guide to Methods and Applications*, ed. by M.A. Innis, D. H. Gelfand, J. J. Sninsky, and T. J. White, pp. 315–322. Academic Press, Inc., New York.

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