Disease Note

Diseases Caused by Fungi and Fungus-Like Organisms

First Report of Botrytis Blight Caused by *Botrytis cinerea* on *Helleborus niger* in Italy.

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Helleborus niger (Christmas Carol) is a perennial herbaceous plant belonging to the Ranunculaceae family, appreciated for the abundant production of winter flowers. In Italy, it is marketed as potted plant. In January 2021, symptoms of an unknown blight were observed on all the 2-year-old potted plants of *H. niger* grown in a private garden located in Santa Margherita Ligure (Genova Province, northern Italy). Symptoms appeared on stems and petioles that turned brown, rotted, and collapsed starting from the base. The tepals also were affected and desiccated. A soft, gray mycelium appeared on affected tissues and plants totally lost their aesthetic value. Several affected stems and tepals were immersed in a solution of sodium hypochlorite (NaOCl, 1%) for 30 s. Successively, they were washed in sterilized water. Small pieces were excised from the margin of affected tissues and cultured on potato dextrose agar (PDA, Merck KGaA, Darmstadt, Germany) medium. Plates were incubated at temperatures ranging from 20 to 25°C. Gray fungal colonies developed consistently and produced dark, spheroidal to elongated sclerotia that measured 1.0 to 1.5 × 0.5 to 1.3 (mean: 1.1 × 0.9) mm. After 10 days, colonies produced branched conidiophores that supported unicellular, ovoid conidia measuring 7.9 to 12.1 × 5.2 to 9.2 (mean: 10.3 × 7.8) µm. These characteristics permitted us to identify the fungus isolated from *H. niger* as *Botrytis cinerea* Pers.: Fr. (Ellis 1971). The DNA of the isolate 21-1-O1 was extracted with the E.Z.N.A. Plant DNA Kit (Omega Bio-Tek, Norcross, GA, U.S.A.) from a pure culture grown on PDA. Successively, the product obtained from a PCR reaction with primers ITS1/ITS4 (White et al. 1990) was purified and sequenced. A 516-bp sequence (GenBank accession no. MZ417544) was achieved. BLASTn analysis (Altschul et al. 1997) of this sequence had 100% nucleotide identity with the ex-type CBS 156.71 of *B. cinerea* (GenBank accession no. MH860044). The pathogenicity of the isolate 21-1-O1 was confirmed by inoculating three plants of *H. niger* with a conidial and mycelial suspension obtained from 20-day-old cultures grown on PDA at 20 to 24°C. Each plant was sprayed with 20 ml of suspension at the concentration of 5×10^4 CFU (conidia and mycelial fragments)/ml. Three control plants were sprayed with sterile water. All plants were closed in a moistened chamber for 8 days and maintained outdoors, under the shade, at temperatures ranging from 17 to 26°C. First symptoms of browning appeared on tepals of inoculated plants about 6 days after inoculation. Later, symptoms developed on stems and petioles, affected tissues rotted and were covered by a gray soft mycelium. The pathogenicity test was repeated once. *B. cinerea* was reisolated from the affected tissues. Controls remained healthy. *B. cinerea* has been reported on *H. niger* in the United States (Anonymous 1960). To authors' knowledge, this is the first report of *B. cinerea* on *H. niger* in Italy as well as in Europe. The economic importance of this disease is nowadays limited in Italy due to the reduced production of *H. niger*. However, the cultivation of this species might increase, following the selection of genotypes more suitable as potted plants and for cut flower production under the Mediterranean climate.

References:

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