

## Diseases Caused by Fungi and Fungus-Like Organisms

### First Report of *Diaporthe eres* Causing Stem Blight and Dieback on Highbush Blueberry (*Vaccinium corymbosum*) in Italy

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*Vaccinium corymbosum* L. is a woody deciduous shrub in the Ericaceae family. During the last decade, Northern Italy has become a major area for blueberry cultivation and production in Europe, supplying other countries with high-quality fruit (FAOSTAT 2020). In June 2020, plant stem and branch wilting with brownish necrotic internal lesions were observed on 20% of around 250 plants of the cultivar Draper cultivated in a soilless culture system in Peveragno (Piedmont, Cuneo Province, 44°21'05.6"N, 07°37'23.2"E). Dieback and death of the plants also occurred. Fifteen symptomatic plants were collected. Wood pieces (5 to 10 mm) excised from diseased stems and branches were sterilized in 1% sodium hypochlorite for 1 min, rinsed in sterile distilled water, and dried on sterile absorbent paper. Small fragments (2 to 3 mm), obtained from the edge of the necrotic tissues, were plated on potato dextrose agar (PDA) amended with 25 mg/liter of streptomycin sulfate. Plates were incubated at 25 ± 1°C under a 12 h photoperiod and, after 5 days, colonies with the same characteristics of *Diaporthe* spp. were established from single hyphal tip transfers. Two representative strains (CVG1130 and CVG1131) were grown on PDA from single hyphal-tip transfers. After 7 days, white colonies with fluffy, aerial mycelium reaching 8.3 cm were observed. Colonies turned gray after 3 weeks producing dark brown pycnidia. Aseptate, hyaline, fusiform to ellipsoidal, 6.3 to 8.4 × 2.2 to 3.0 µm alpha conidia were produced. No beta conidia were observed. The DNA of the same strains was extracted and PCR

was performed for the ITS (primer set: ITS1-ITS4) (GenBank accession nos. ON834528; ON834529), *tef* (EF1-728/EF1-986) (accession nos. ON843715; ON843716), and *tub2* (T1/Bt2b) (accession nos. ON843713; ON843714) regions, in accordance with previous studies (Bezerra et al. 2021; Gomes et al. 2013). Sequences analyzed with the BLASTn algorithm (Altschul et al. 1997) exhibited 98% identity with the extype strain CBS 138594 of *Diaporthe eres* for ITS (GenBank no. KJ210529), 100% identity for *tub2* (GenBank Accession no. KJ420799), and 99% identity for *tef* (GenBank no. KJ210550). The maximum likelihood method based on combined sequences of ITS, *tef*, and *tub2* loci was performed, and the isolates CVG1130 and CVG1131 clustered with several reference strains of *D. eres*. To fulfil Koch's postulates, pathogenicity tests were performed on 1-year-old blueberry potted plants cv. Draper and Duke. A small piece of the bark tissue was removed with a sterile-bladed scalpel to expose the cambium. Mycelium plugs (5 mm diameter) from 10-day-old cultures of the strains CVG1130 and CVG1131 were placed with the mycelium in contact with the internal plant tissues. Three plants were used for each isolate and the inoculation points were sealed with Parafilm. Three plants treated with sterile PDA plugs were used as a control. Plants were placed in a growth chamber at 25°C for 3 weeks. After this period, symptoms similar to those observed in the field appeared on the inoculated plants, while control plants remained healthy. A fungus with the same morphological characteristics of *D. eres* was reisolated from inoculated plants and identified by sequencing the *tub2* gene to confirm Koch's postulates. *Diaporthe eres* was previously reported on different *Vaccinium* spp. in Chile, Lithuania, the Netherlands, and the United States (Farr and Rossman 2022). To our knowledge, this is the first report of *D. eres* associated with stem blight and dieback of highbush blueberry in Italy. Duke and Draper are among the most cultivated blueberry cultivars in Piedmont, where the spread of *D. eres* could represent a serious threat.

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#### e-Xtra

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