First Report of Phytophthora pistaciae Causing Root and Collar Rot on Nursery Plants of Pistacia lentiscus in Italy

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In April 2019, a survey carried out in a public forest nursery in Sardinia (Italy) (39°57′39″N; 8°36′02″E) revealed the presence of typical *Phytophthora* symptoms such as leaf reddening, wilted shoots, and dieback associated with root and collar rot on 3-year-old potted seedlings of lentisk (*Pistacia lentiscus* L.). Approximately 60% of 240 potted plants, obtained from seeds collected in natural areas in Sardinia, were symptomatic. To isolate *Phytophthora*, rhizosphere soil and root samples of seven symptomatic seedlings were baited with *Quercus suber* and *Sambucus nigra* leaves as described by Linaldeddu et al. (2014). After 5 days, leaves showing dark spots were plated on potato dextrose agar (PDA) supplemented with 100 ml/liter of carrot juice, 0.013 g/liter of pimaricin, and 0.05 g/liter of hymexazol. *Phytophthora* colonies characterized by an aerial and compact mycelium without a distinct pattern were isolated from all samples. Sporangia produced on PDA plugs immersed in unsterile pond water were nonpapillate, persistent, ellipsoid, rarely obpyriform 41.6 to 100.4 × 25.1 to 59.3 µm, with a length/width ratio of 1.76, proliferating internally and externally. Hyphal swellings were globose to irregular, and no chlamydospores were observed. All isolates produced smooth-walled oogonia (25.1 to 39.5 µm) with chiefly amphigynous antheridia after 2 weeks on carrot agar (CA) at 20°C in the dark. Morphological features were in close agreement with those reported for *Phytophthora pistaciae* Mirabolfathy (Mirabolfathy et al. 2001). The identity of all isolates was confirmed by sequencing the internal transcribed spacer region using the primers ITS1 and ITS4. DNA extraction, PCR amplification reactions, and DNA sequencing were carried out according to Linaldeddu et al. (2016). BLAST searches in GenBank showed 100% identity with reference sequences of *P. pistaciae*, including the ex-type culture P19883 (FJ746648). Two representative isolates of *P. pistaciae* (AM43 and AM53) were stored at 10°C under water at the Culture Collection of the TeSAF Department, University of Padova, and the sequences were deposited in GenBank (MN656156 and MN656157). The pathogenicity of both *P. pistaciae* isolates was evaluated by inoculating 14 lentisk seedlings (3 years old) per isolate. After disinfecting the bark with 90% ethanol and removing a piece of outer bark (5 mm) with a sterile cork borer, the seedlings were inoculated with a same-sized agar-mycelium plug cut from the margin of a 5-day-old colony. Seven control plants were inoculated with a sterile CA plug. All plants were kept in a greenhouse at 20 to 26°C in natural daylight. After 3 weeks, all plants inoculated with *P. pistaciae* showed wilted shoots and inner bark necrotic lesions spreading up and down from the inoculation site. The average lesion size was 5.9 ± 3.1 cm for the isolate AM43 and 4.9 ± 1.4 cm for the isolate AM53. *P. pistaciae* was successfully reisolated from all the inoculated plants, fulfilling Koch's postulates. On control seedlings, only a small brown discoloration restricted to the inoculation point was observed. *P. pistaciae* is reported as one of the main pathogens involved in the etiology of pistachio (*Pistacia vera* L.) gummosis in Iran (Mirsoleimani and Mostowfizadeh-Ghalamfarsa 2013). This is the first report of *P. pistaciae* outside of Iran and as a lentisk pathogen. Considering the high virulence of this exotic pathogen, this discovery could pose a serious threat to lentisk ecosystems and pistachio cultivation in the Mediterranean area.

The author(s) declare no conflict of interest.

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