Disease Note

FIRST REPORT OF *PHOMA NOVAE-VERBASCICOLA* ON MOTH MULLEIN (*VERBASCUM BLATTARIA*) IN ITALY

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On November 2013, light brown necrotic spots were observed on the leaves of *Verbascum blattaria* seedlings. Necroses reached 10 mm in diameter and dried. Eventually a hole appeared in the internal part of the necrosis. On potato dextrose agar (PDA), a fungus isolated from affected leaves produced globose or subglobose pycnidia measuring 62-164×60-163 (average 111×105 µm), containing ellipsoid, unicellular conidia measuring 2.8-4.4×1.4-2.3 µm (average 3.6×1.8 µm). The internal transcribed spacer (ITS) region of rDNA of the microorganism was amplified using the primers ITS1/ITS4 and sequenced (Altschul *et al*., 1997) (GenBank accession No. KJ192364). BLAST analysis of the 520 bp amplicon showed 100% homology with the sequence KC411473 of *Phoma novae-verbascicola*. Pathogenicity tests were carried out by spraying a conidial suspension of the fungus on three healthy plants of *V. blattaria*. Plants were kept in a moist chamber. First necrotic spots appeared 2 days after the inoculation and *P. novae-verbascicola* was constantly reisolated from affected leaves. *P. novae-verbascicola* has recently been reported on *Verbascum nigrum* (Garibaldi *et al*., 2013), while this is the first report of the pathogen on *V. blattaria* in Italy, as well as in the world.

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Disease Note

FIRST REPORT OF *ALTERNARIA ALTERNATA* CAUSING LEAF SPOT ON HOLLY IN GREECE

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A foliar disease of holly (*Ilex aquifolium*), observed in November 2009 in a natural ecosystem in the region of Thessaloniki (northern Greece), consisted of circular or irregularly shaped necrotic spots with yellow border, 3-7 mm in diameter, present on *ca.* 30% of the leaves. Small pieces of tissue at the margin of the spots were excised, surface-disinfected (1% sodium hypochlorite for 1 min) and plated on potato dextrose agar. Two single-spore cultures in potato carrot agar, gave rise to initially white colonies turning grayish-black due to abundant sporulation. Conidiophores were green to brown, short, septate, branched or unbranched. Conidia, produced in single or more often branched chains, were obpyriform, with a conical or cylindrical beak, ovoid or ellipsoidal, measured 7.7-27.4×5.6-15.0 µm (average 16.3×8.8 µm), and showed 1 to 5 transverse and 0 to 3 longitudinal septa. These morphological traits and measurements tally with those of *Alternaria alternata* (Fr.) Keissl. (Simmons, 2007). The ITS1-5.8S-ITS2 region of the two single-spore isolates, amplified with primers ITS1 and ITS4 and sequenced (GenBank accession Nos. JQ809323, JQ809324), revealed 100% homology with the sequence of various *A. alternata* isolates (e.g. JQ070079). Eight 3-year-old holly plants were sprayed with a 10^6 conidia/ml spore suspension, covered with a polyethylene bag for 48 h and placed in a greenhouse at 23±2°C together with sterile distilled water-sprayed controls. Leaf spots like those observed in the field developed on all inoculated plants but not on controls. *A. alternata* was reisolated from artificially inoculated leaves. Holly infections by *A. alternata* have previously been reported from Poland (Orlikowski and Szkuta, 2004), the USA and Canada (Sinclair *et al*., 1987) but, to our knowledge, never from Greece.

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