Diseases Caused by Bacteria and Phytoplasmas

First Report of *Xanthomonas hortorum* Causing Bacterial Leaf Spot of Lavender (*Lavandula × intermedia*) in Ohio

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In July 2018, a sample of lavender variety Grosso (*Lavandula × intermedia* ‘Grosso’) from Miami County, OH, was received by The Ohio State University Vegetable Pathology Laboratory in Wooster. Lavender plants were field grown in sandy clay soil with plastic mulch under drip irrigation. Disease incidence ranged from 0 to 32% depending on variety. Leaves and stems showed dark necrotic lesions that varied from roughly circular (~0.3 to 0.5 mm diameter) to large coalesced necrotic areas surrounded by a water-soaked halo. Bacterial streaming from lesions was observed microscopically. Leaf tissue pieces (~0.5 cm²) were surface sterilized in 70% ethanol for 30 s and rinsed in sterile deionized water. The tissue was sliced aseptically into smaller sections in 100 µl of sterile water, and the bacterial suspension was streaked on yeast dextrose calcium carbonate agar medium. Ten yellow *Xanthomonas*-like colonies were selected after 72 h of incubation at 28°C in the dark. Strains were gram negative, oxidase negative, and caused hypersensitive reactions on *Nicotiana benthamiana* (L.). All strains were genotyped after whole-cell DNA extraction by BOX-PCR (Louws et al. 1994) and had the same banding profile. Four 8-week-old lavender plants (*Lavandula dentata* and *Lavandula × gingsii* ‘Goodwin Creek Gray’) were spray-inoculated with a 10⁶ CFU/ml suspension of strain SM175-2018 in sterile water. Control plants were sprayed with sterile water. Plants were kept in plastic bags for the first 48 h at 28°C with a 14-h photoperiod. Water-soaked necrotic

lesions appeared 14 days after inoculation with SM175-2018, whereas mock-inoculated plants did not show symptoms. Bacterial isolation from symptomatic leaf tissue was carried out as described above. The BOX-PCR profile of the reisolated strain was identical to that of SM175-2018. Multilocus sequence analysis of the housekeeping genes *fuyA*, *gyrB*, and *rpoD* was performed (accession nos. MT764834 to MT764836). The resulting concatenated data set was used to perform a phylogenetic analysis using maximum likelihood criteria to evaluate relationships with closely related *Xanthomonas* spp. using published reference sequences (Young et al. 2008). SM175-2018 was assigned to the *Xanthomonas hortorum* clade (Morinière et al. 2020) with strong bootstrap support. The strain was subjected to whole-genome analysis. Genomic DNA was extracted using a Qiagen Genomic DNA buffer set with genomic-tip 100/G following the manufacturer’s protocol and sequenced using the iSeq-100 Illumina platform with the Nextera DNA Flex Library Prep protocol kit and Nextera DNA CD indexes. Average nucleotide identity (ANI) analysis was performed with the ANI-Matrix software Enveomics tool (Rodriguez-R and Konstantinidis 2016) using the sequenced genome (NCBI GenBank Biosample no. SAMN11831455) and those of other *X. hortorum* (Vauterin et al. 1995) bacteria (pvs. *hederae*, *carotae*, *vitians*). SM175-2018 shared a 96% ANI with other *X. hortorum* strains. *X. hortorum* is associated with bacterial leaf spot of carrot (Scott and Dung 2020) and also reported on ornamental plants (Klass et al. 2019; Mirik et al. 2010; Oliver et al. 2012; Roberts and Parkinson 2014); however, additional research is needed to establish the host specificity of lavender strains. To our knowledge, this is the first report of *X. hortorum* causing bacterial leaf spot of lavender in Ohio. The disease may negatively impact the yield and quality of flowers used in production of lavender oils and essences.

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