Accepted for publication 14 January 2012. Published 26 February 2013.

Calonectria pseudonaviculata can Cause Leaf Spot and Stem Blight of *Pachysandra procumbens*

J. A. LaMondia and **D. W. Li,** The Connecticut Agricultural Experiment Station, Valley Laboratory, Windsor, CT 06095

Corresponding author: J. A. LaMondia. james.lamondia@ct.gov

LaMondia, J. A., and Li, D. W. 2013. *Calonectria pseudonaviculata* can cause leaf spot and stem blight of *Pachysandra procumbens*. Online. Plant Health Progress doi:10.1094/PHP-2013-0226-01-BR.

Calonectria pseudonaviculata (Crous & al.) L. Lombard & al. (syn= *Cylindrocladium pseudonaviculatum* Crous, J.Z. Groenewald & C.F. Hill, *Cylindrocladium buxicola* Henricot) was recently reported infecting boxwood *Buxus spp*. L. in North Carolina and Connecticut. This was the first report of this disease in North America (5). The pathogen caused significant losses in container nurseries and in the landscape in both states and a number of boxwood species were shown to be infected. Henricot et al. (4) reported that all *Buxus* spp. tested and a *Sarcococca* Lindl. (sweet box) sp. tested were all susceptible to this pathogen. Plants in the *Buxaceae* that are either native or grown as ornamentals in North America include *Buxus*, *Sarcococca*, and *Pachysandra* Michx. *Calonectria pseudonaviculata* was reported to cause disease on Japanese spurge, *Pachysandra terminalis* Siebold & Zucc., in laboratory and greenhouse pathogenicity tests (6) and pathogenicity was subsequently confirmed by the discovery of natural infections in landscapes in association with diseased boxwood plants (2). To date, over 20 cases of natural landscape infections in *P. terminalis* have been confirmed in Connecticut (S. M. Douglas, *personal communication*). The discovery of landscape infections of *P. terminalis* resulted in important modification of best management practices for management of this disease in the landscape (3).

Allegheny spurge, *Pachysandra procumbens* Michx., is a native plant that is also grown as an ornamental ground cover in nurseries and landscapes. *P. procumbens* is primarily reported as a perennial woodland herb or subshrub from the southeastern United States, from Louisiana to Florida and north to Indiana and Pennsylvania. It is relatively rare in nature with locally common populations (1). It is hardy far north of its natural range and is propagated and sold as an ornamental groundcover in the nursery trade.

Our objective in this study was to determine the susceptibility of *P. procumbens* to *C. pseudonaviculata*. To this end, we isolated the anamorphic stage of the pathogen from leaf and stem lesions of *B. sempervirens* and obtained single-spored cultures on half-strength potato dextrose agar (½PDA). The pathogen had been identified by morphological characteristics and by amplification of a portion of the β -tubulin gene from isolates Cps-CT-L1 and Cps-CT-S1 (5). Three healthy *P. procumbens* plants per 10-cm-diameter pot were inoculated with water alone or a conidial suspension of *C. pseudonaviculata* isolate Cps-CT-L1 (ATCC MYA-4891) (1.3×10^6 conidia per plant) using a hand-held sprayer until runoff. Plants in five replicate pots were kept moist in a plastic bag for 48 h at ambient laboratory temperature (about 20°C) and then transferred to the greenhouse. The experiment was conducted in April 2012. Circular lesions (1-4-mm diameter) were evident on leaves within seven days after inoculation. Leaf lesions were necrotic and often had darker margins and a chlorotic area around the lesion (Fig. 1). Elongate necrotic lesions were also observed running vertically along the stems (Fig. 2). All fifteen

inoculated plants developed lesions within seven days, and no lesions were observed on non-inoculated plants. Leaves and stems with lesions were surface sterilized in 0.5% NaOCl for 30 sec, rinsed twice in sterile distilled water and lesion margins plated onto water agar or ½PDA. The pathogen was re-isolated from all plants (and from a total of 19 of 21 leaf and stem lesion isolation attempts). Koch's postulates were tested an additional time in May 2012 as previously described with isolate Cps-CT-S1 (ATCC MYA-4890), inoculated with 1.0×10^6 conidia per plant on both *P. procumbens* and *P. terminalis*. The pathogen was again re-isolated from all plants (and from 15 of 18 leaf and stem lesions in *P. procumbens*). In pathogenicity tests with both isolates, leaf and stem lesions continued to expand. Stem lesions girdled the plant after two weeks and resulted in stem blight and shoot/plant death of *P. procumbens*, but not *P. terminalis*. Under humid conditions, heavy sporulation of the anamorphic stage of *C. pseudonaviculata* was observed on both leaf and stem tissues of *P. procumbens*. Sporulation also occurred to a lesser extent on *P. terminalis*. Microsclerotia were observed in infected leaves and chlamydospores, in infected leaves and stems using both tape lifts and epidermal peels at 400× magnification.

Fig. 1. *Cylindrocladium pseudonaviculatum* infection of *Pachysandra procumbens* leaves.

Fig. 2. *Cylindrocladium pseudonaviculatum* infection of *Pachysandra procumbens* stem tissue.

Calonectria pseudonaviculata has now been shown to cause disease on all common ornamental species in the Buxaceae grown in North America. *Pachysandra procumbens*, while not as common as *P. terminalis*, typically grows in environments conducive for the development of disease and may also serve as a reservoir of inoculum for the boxwood blight pathogen in cultivated landscapes and in nature. In addition, *P. procumbens* is listed by the USDA Natural Resources Conservation Service as endangered in states such as Florida and Indiana and *C. pseudonaviculata* leaf spot and stem blight may further threaten this species in its native habitat.

Acknowledgements

The authors thank Michelle Salvas for technical assistance. This research was supported by grants from USDA-APHIS and the Horticultural Research Institute.

Literature Cited

1. Dirr, M. A., and Alexander, J. H. III. 1979. The Allegheny Pachysandra. Arnoldia 39:16-21.
2. Douglas, S. M. 2012. Natural infection of pachysandra with boxwood blight in Connecticut landscapes. CAES Fact Sheet. Connecticut Agric. Exp. Station, New Haven, CT.
3. Douglas, S. M. 2012. Guidelines for Reporting and Managing Boxwood Blight in Connecticut Landscapes Ver. 2.0. CAES Fact Sheet. Connecticut Agric. Exp. Station, New Haven, CT.

4. Henricot, B., Gorton, C., Denton, G., and Denton, J. 2008. Studies on the control of *Cylindrocladium buxicola* using fungicides and host resistance. Plant Dis. 92:1273-1279.
5. Ivors, K., Lacey, W., Milks, D., Douglas, S. M., Inman, M. K., Marra, R. E., and LaMondia, J. A.2012. First report of boxwood blight caused by *Cylindrocladium pseudonaviculatum* in the United States. Plant Dis. 96:1070.
6. LaMondia, J. A., Li, D. W., Marra, R. E., and Douglas, S. M. 2012. First report of *Cylindrocladium pseudonaviculatum* causing leaf spot of *Pachysandra terminalis*. Plant Dis. 96:1069.