Emerging Diseases in Oregon

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Abstract

Oregon is a major producer of nursery plants and seeds, with 70% of these commodities traded domestically and internationally making these industries potential pathways for pest introductions. Recent pest detections support this concern. In 2015, foliage from lilacs (*Syringa vulgaris*) grown in a Marion County nursery showed a reduction in leaf size, leaf deformation, ring spots, and line patterns. Total nucleic acids analysis of six symptomatic plants revealed inserts that showed >99% homology with the published sequence for *Lilac Ring Mottle Virus* (LiRMV, GenBank Accession: U17391)(S. Scott, Clemson University). This is the first report of LiRMV in the US.

Three seedborne diseases, light leaf spot (LLS, *Cylindrosporium concentricum*), white leaf spot (WLS, *Pseudocercosporella capsulae*), and black leg (*Phoma lingam*), were identified causing an outbreak in crucifer crops grown in western Oregon in 2014. Pure cultures for morphological identification were obtained by transferring single spores for each fungal pathogen from symptomatic tissue onto quarter-strength potato dextrose agar amended with streptomycin; sporulating structures produced on diseased tissue placed into moisture chambers were also examined microscopically. Species identification was verified using PCR and DNA sequencing. LLS was identified on *Brassica* crops and weeds growing in six counties. This is the first report of LLS in the USA. WLS was identified on *Brassica* spp. growing in five counties. This is the first report of WLS on seed crops in Oregon. Blackleg was identified infecting *Brassica* spp. and *Rorippa curvisiliqua* growing in seven counties.

These detections reinforce concerns about nursery plants and seeds as pathways for pests.

Lilac Ring Mottle Virus

Lilac ring mottle virus (LiRMV) (Van Der Meer et al., 1976), an isometric RNA virus within the Family *Bromovirus* and genus *Ilarvirus* (Scott and Ge 1995, Scott and Zimmerman 2008), was discovered infecting lilac 'President Grevy' and 'Krasavitsa Moskvy' plants in a Marion County nursery in 2015. Symptoms observed on the plants included leaf deformation, reduction in leaf size, ring spots, and line patterns (Fig. 1).

LiRMV presence in the lilacs was verified by extracting total nucleic acids (TNA) from symptomatic leaf tissues (Hughes and Galau 1988) and then synthesizing cDNA using the downstream primer 5' GAGACCGAAGTCTTCTTCC 3' (2pmol/ reaction) and Superscript III Reverse Transcriptase. Movement protein-specific primers (downstream: 5' GAGACCGAAGTCTTCTTCC 3' and upstream: 5' CCACGTGCTTCTCACCC 3') were used to amplify a 650 bp amplicon from the cDNA through PCR using a 55°C melting temperature (Scott and Zimmerman 2008). The 650 bp amplicons from seven lilac samples were cloned using the pGem®-T Easy Vector, selected by blue-white screening, and then sequenced using the primer M13F. Six of the seven contained inserts that showed >99% homology with the published sequence for LiRMV (GenBank Accession: U17391).

Samples received by the OSU Plant Clinic were examined using transmission electron microscopy using two different methods. In the first, the leaf tissue was cut into 1 mm strips and put in fixative overnight. The following day the tissue was crushed to express sap, which was then collected on a copper grid, stained, and examined. The second method was a simple expression of sap from unfixed tissue which was then stained. Both preparations were examined using a FEI Titan 80-200 TEM/STEM electron microscope. No virus particles were observed in either preparation. These results are consistent with previous reports that LiRMV is difficult to characterize using electron microscopy (Van der Meer et al, 1976) and suggests that molecular analysis is a more reliable means of diagnosis.

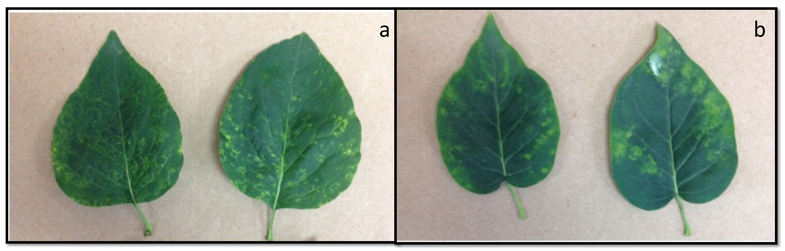

Fig. 1. Lilac ring mottle virus symptoms on lilac cultivars President Grevy (a) and Krasavitsa Moskvy (b).

Crucifer diseases

Three seedborne diseases, light leaf spot (LLS, *Cylindrosporium concentricum*), white leaf spot (WLS, *Pseudocercosporella capsulae*), and blackleg (*Phoma lingam*), were identified in an outbreak in crucifer seed crops grown in western Oregon during the spring of 2014. Pure cultures for morphological identification were obtained by transferring single spores for each fungal pathogen from symptomatic tissue onto quarter-strength potato dextrose agar amended with streptomycin; sporulating structures produced on diseased tissue placed into moisture chambers were also examined microscopically. Species identification was verified using PCR and DNA sequencing. USDA APHIS also confirmed the identity of the LLS agent.

Light leaf spot

LLS (Fig. 2) was identified on *Brassica* crops and weeds growing in 24 locations across six counties. Leaf symptoms begin as discolored patches, which develop into irregular brown lesions with cracked centers, sometimes turning black. Leaf veins around the infected areas sometimes take on an olive-brown discoloration prior to lesion development. When leaf lesions coalesce, the leaf is killed. Infected leaves may exhibit distortions. Fall-infected plants can remain asymptomatic until the following spring although plant stunting may be present. Stem lesions are elongated brownish streaks with grayish-brownish-black margins and are usually superficial. Pods infected early appear distorted whereas pods infected later are healthy appearing but have white spore masses. This is the first report of LLS in the USA.

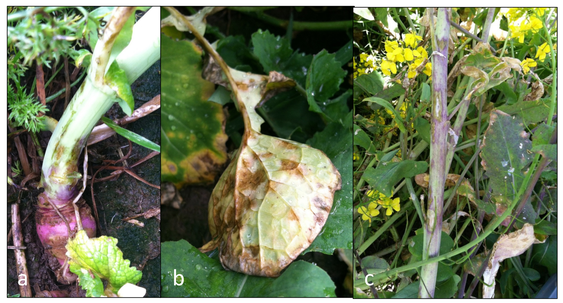

Fig. 2. Light leaf spot on turnip for seed; symptoms on the lower stem (a), leaf (b), and flower stalk (c).

White leaf spot

WLS (Fig. 3) was identified on *Brassica* spp. growing in 13 locations in five counties. Tan, irregular or roundish spots develop on leaves, and then become ashy-gray to white with a brownish margin, sometimes a yellowish halo. White conidia can be observed on the underside of the leaf spots. Leaf spots become dark brown as leaves age, with the center of older spots falling out. Lesions may coalesce when disease is severe resulting in defoliation. Superficial stem lesions are elongated and initially brown, turning ashy-gray to white with a brownish margin and numerous dark pseudothecia; there is a distinct boundary between diseased and healthy tissue. Pod infections start as small brown spots that expand and turn grayish-white with numerous pseudothecia. This is the first report of WLS on crucifer seed crops in Oregon.

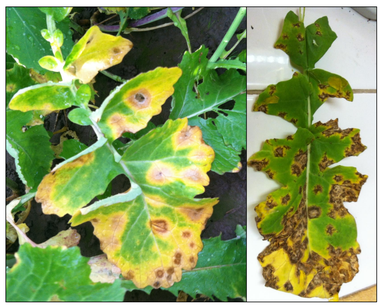

Fig. 3. Symptoms of white leaf spot on the leaves of turnip for seed.

Blackleg

Blackleg (Fig. 4) was identified infecting *Brassica* spp. and *Rorippa curvisiliqua* growing in 43 locations across seven counties; oil, specialty, and vegetable seed crops were severely affected. Pale, irregular spots develop on leaves, stems or petioles, later becoming ashy gray with scattered black pycnidia. Leaf lesions are apt to fall out with rain. Stem cankers are elongated with purple borders and can form near the soil line, extending below the soil surface to cause black rot of the lower stem and roots.

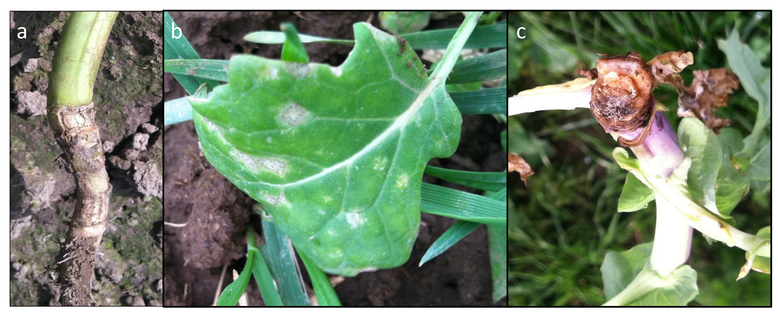

Fig. 4. Blackleg symptoms; stem canker (a) and leaf spots (b) on canola, and dry rot (c) on turnip for seed.