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SEPTORIA LEAF SPOT OF *PRUNUS VIRGINIANA* SEEDLINGS - BITTERROOT NATIVE GROWERS NURSERY, HAMILTON, MONTANA

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During the 1992 growing season, young, container-grown chokecherry (*Prunus virginiana* L.) seedlings being produced at the Bitterroot Native Growers Nursery in Hamilton, Montana appeared infected with leaf spot pathogen(s). Affected seedlings had discrete black to deep violet-colored lesions that were evident on both upper and lower leaf surfaces (figure 1). Within the center of most lesions, cream to slightly pink-colored sporodochia were present (see arrow on figure 1). Seedlings infected longer had coalesced lesions with necrosis spreading through leaf petioles into the stem. Microscopic examinations of spores within sporodochia revealed elongate, needle-shaped spores, usually with several septations (figure 2). These spores and associated fruiting structures indicated that the fungus was a species of *Septoria*.

Growers indicated that necrotic lesions first developed shortly after seedling emergence. In some cases, lesions were present on cotyledons. Superficially, symptoms looked like those produced by *Coccomyces hiemalis* Higg. (=*Phloeosporella padi* (Lib.) von Arx; teleomorph = *Blumeriella jaapii* (Rehm) von Arx)(Hepting 1971), cause of the common "shot hole" disease. This disease has previously been reported in the Inland Northwest, particularly on bareroot stock (James and Watkins 1986). However, as *C. hiemalis*-caused lesions become older, affected host tissues typically fall out, leaving the appearance of holes. In advanced stages, with extensive foliar necrosis and colonization of petiole and stem tissues, the "shot hole" disease and disease caused by *Septoria* may appear similar (James and Watkins 1986). The proper way of differentiating the two is by examination of conidia produced within developing lesions.

Septoria spp. cause a wide range of plant diseases on many different types of plants. Leaf spots are particularly common on broadleaved species. One of the most important pathogens in this genus is *S. musiva* Peck, cause of leaf spot and canker of *Populus* spp. (Filer and others 1971; Ostry and others 1989). Other examples of pathogens in this genus include *S. alnifolia* Ell. & Ev., cause of foliage blight of white alder seedlings in California nurseries (Frankel 1990), *S. nodorum* Berk., a common leaf pathogen of wheat and barley (Karjalainen and Lounatmaa 1986), *S. elaegni* (Chev.) Desm., causing leaf spot of Russian-olive seedlings in nurseries (Lorenzini and others 1984), and *S. albopunctata* Cke., which severely affects greenhouse-grown *Vaccinium* spp. (Demaree and Wilcox 1947). The species causing disease on *P. virginiana* seedlings is unknown. A discussion of the taxonomic characteristics of the genus *Septoria* and problems assigning particular taxa to leaf spot pathogens in this genus are discussed in the Appendix.

Septoria spp. overwinter on infected fallen leaves (Ostry 1987). Ascospores of the teleomorphic (=sexual) stage (*Mycosphaerella*) are released from leaf debris beginning in April and continuing through the end of May (Waterman 1954). Primary leaf spots caused by ascospores are first observed by the end of May (Ostry 1987). Symptoms are first observed on leaves about the time they have reached their maximum size. The time from bud bread until development of visible symptoms of *Septoria* infection was approximately 6 weeks in outdoor poplar plantations in Minnesota (Ostry 1987).

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Figure 1--Upper surface of *Prunus virginiana* leaf infected with *Septoria* spp. with discrete necrotic lesions. Mature pycnidia are formed in the center of lesions. Cream to pink-colored sporodochia are formed at the apex of pycnidia (arrow).

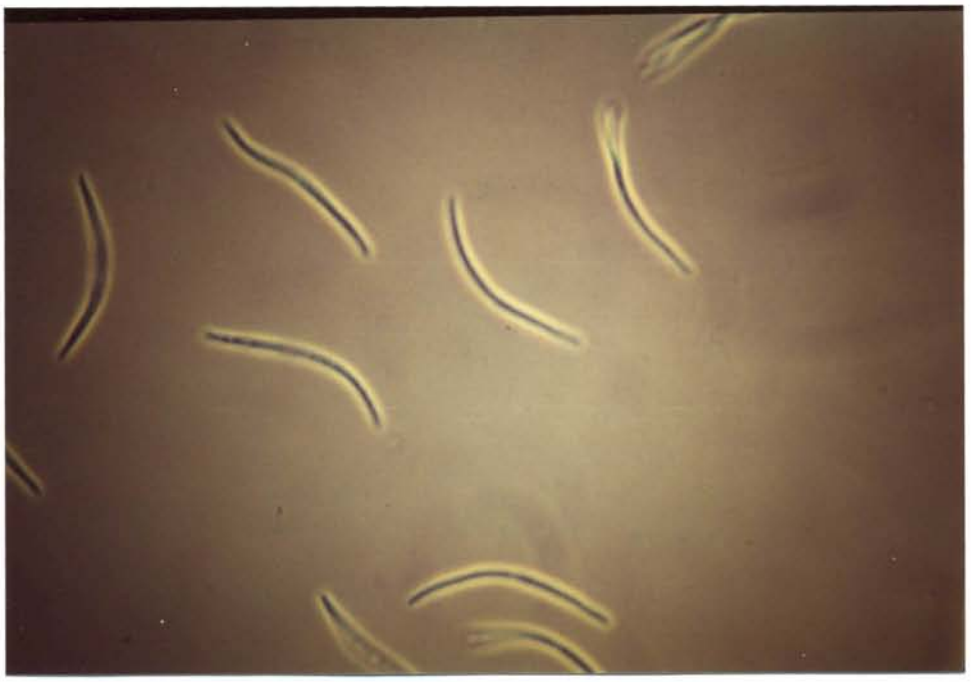

Figure 2--Elongate, narrow, multi-septate condia of *Septoria* spp. produced within sporodochia from lesions on infected *Prunus virginiana* leaves.

These fungi produce asexual fruiting bodies called pycnidia within lesions. Mature conidia are formed in pycnidia from 1 to 2 weeks after leaf spots develop (Ostry 1987). Conidia are disseminated within water to new infection sites (Palmer and others 1980). Spore masses are sticky and are exuded as tendrils from fruiting bodies (Waterman 1956). The peak period of conidial release for *Septoria* spp. begins at the end of July and extends through the middle of August (Ostry 1987). During periods of prolonged moisture on the surface of foliage, spore germination and host infection occurs (Ostry 1987). Penetration of spore germ tubes into host tissues takes place directly through the periclinal walls of epidermal cells into the cell lumen. Most subsequent colonization is intercellular (Karjalainen and Lounatmaa 1986). Penetration may be by mechanical force and/or enzymatic hydrolysis. As fungal colonization increases, expanding zones of necrosis occurs. Cell death extends through the entire leaf cross section; pycnidia form during the latter part of fungal development. Premature leaf abscission on trees severely affected by *Septoria* can occur by the end of June in temperate areas (Ostry 1987). When environmental conditions are conducive, particularly during periods of prolonged moisture, several disease cycles are possible during the growing season (Ostry 1987). Rapid buildup of inoculum and spread throughout the nursery is possible. Significant growth reduction can occur on infected nursery stock (Lorenzini and others 1984).

Control of leaf spot diseases in nurseries, including those caused by *Septoria* spp., are best obtained using standard chemical fungicides (Palmer and others 1980). These chemicals should be applied during periods of high seedling susceptibility when environmental conditions are conducive for disease buildup. Within greenhouses, moist conditions may be the rule throughout much of the growing season. However, when temperatures increase in the summer, leaf spot diseases usually become less serious. Ensuring proper drying of foliage between irrigation events will help reduce susceptibility to leaf spots. Sanitation by periodic removal of seedlings with disease symptoms will help reduce levels of inoculum within greenhouses (Demaree and Wilcox 1947). It is also important that infected foliage from previous crops or other infected plants be removed from within and near greenhouses to reduce primary inoculum (ascospores).

Several fungicides have been recommended for control of Septoria leaf spot. These include benomyl, chlorothalonil, captafol, mancozeb and copper hydroxide (Ostry 1987). Benomyl was the most effective fungicide controlling *Septoria* in poplar nurseries (Ostry 1987), but it is no longer being manufactured. Fortunately, some substitutes with the same or similar modes of action are available. One of these is thiophanate (Cleary's 3336®). Copper hydroxide (Kocide®) is also effective for control of this disease, although this and other chemicals may not be registered for use in Montana. Fungicide applications should be made 2 or 3 times per month beginning at leaf flush and continuing until temperatures become consistently warm (Ostry 1987).
If possible, several different fungicides should be used in rotation to control this and other foliage-type diseases. Many foliar pathogens can readily develop resistance to chemical fungicides, particularly if one chemical is continually applied for prolonged periods (Staub 1991). Selection pressures for developing resistance are minimized if several different chemicals are used, particularly if they have different modes of action.

Septoria spp. have been implicated previously as being seed-borne (Neergaard 1977). These fungi may reside on or within seed of many different types of plants. They are easily isolated from infected seed on standard agar media and if found at high levels, growers should consider treating affected seed with standard surface sterilants such as bleach (aqueous sodium hypochlorite) or hydrogen peroxide.

Future problems with Septoria leaf spot at the nursery should be minimized with careful screening of susceptible crops for disease symptoms, periodic removal of diseased seedlings and other sanitation measures, seed treatment if necessary, and properly timed fungicide treatments.

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APPENDIX

Taxonomic Considerations in the genus *Septoria*

Unfortunately, the demarcation between the genus *Septoria* Sacc. and related genera is not very clear (Constantinescu 1984). *Septoria* is most closely related to the genus *Phloeospora* Wallr. and these two genera may be treated separately (Sutton 1980) or considered congeneric (Jorstad 1965; von Arx 1983). There are more than 2000 described taxa within the genus *Septoria* (Sutton 1980) and there has been no effort to evolve a practical system for identification and taxonomy for the genus. The genus is in great need of revision (Sutton 1980), but until that work is done most workers accept a fairly broad generic concept. A general description of fungi that fall within this genus includes: fungi producing pycnidial conidiomata; conidiogenous cells sympodulae and/or annellides; holoblastic conidial ontogeny; hyaline septate conidia provided with unthickened scars (Constantinescu 1984; Farr 1991).

Species of *Septoria* are delimited on the basis of condiogenesis and condial shape, width, and septation (Constantinescu 1984). Although condiogenesis is used extensively as a taxonomic character for anamorphic fungi, *Septoria* spp. may show several different types of conidiogenesis (Constantinescu 1984). Therefore, affinity to certain host genera or species has been the main character used in delimiting *Septoria* species (Farr 1991). As a result, morphologically indistinguishable fungi are give specific rank solely because they have been found on a particular host (Constantinescu 1984).

Three species of *Septoria* are reported to occur on the genus *Prunus* (Farr and others 1989). These are *S. pruni* Pk., *S. purpureo-cincta* Ell. & Ev., and *S. ravenelii* Pk. Because of the uncertainty of affinities of certain taxa in this genus, it is impossible to determine if these really represent different species or if their designation as taxa are based solely on their occurrence on specific hosts. Biological information of these organisms is lacking. Therefore, an understanding of disease epidemiology must be obtained from members of the genus which have been studied most extensively.

One of the most extensively researched species is *S. musiva*. This common pathogen of poplars has been considered similar to another described species, *S. populi* Desm., and may be a form of this species (Waterman 1954). Pycnidia occur on either or both leaf surfaces and are quite distinct. They have either protruding ostioles or ostioles that open widely, exposing a spore-producing layer similar to acervuli (Waterman 1954). The pycnidia produced within leaf spots and those on young stem cankers closely resemble each other. Condia are hyaline, straight or curved, and have 1-4 septa (Palmer and others 1980). When produced en masse, condia are exuded in pink spore tendrils. The teleomorph of *S. musiva* is *Mycosphaerella populorum* Thompson. Perithecia develop abundantly on fallen, overwintered leaves (Waterman 1954). These perithecia contain fasiculate asci with 8 hyaline, biseriate, 1-septate ascospores (Luley and others 1987). These ascospores are slightly constricted at the septum and are discharged forcibly from perithecia. Cultures produced from ascospores grow readily on agar media and produce pycnidia and conidia typical of *S. musiva*.

Another recently-described species is *S. elaegni*. This species produces numerous pycnidia, scattered over leaf lesions, which are brown to dark brown, usually epiphyllous, immersed in host tissue, becoming erumpent, subepidermal, separate, unilocular, and globose to subglobose (Lorenzini and others 1984). One important characteristic of the pycnidia of this species is the very wide ostiolar opening when conidia are mature. Conidiogenous cells for this species are sessile, occasionally on short, septate condiophores, ampulliform or lagniform, producing conidia at the apex from a narrow neck. Conidia are hyaline, flexuous, slightly tapering towards the apex, with their basal end blunt or rounded. They are of variable length with 0-6 septa (Lorenzini and others 1984). This species grows quite slowly on artificial media with greatest radial growth rate occurring at 20°C. Sporulation in culture is scarce in the dark but abundant under continuous light. Conidia germinate from one or more cells simultaneously (Lorenzini and others 1984).

For comparative purposes, one more species is described. This was designated *S. albopunctata* Cke. and occurs on species of *Vaccinium* (blueberries). Well-developed pycnidia are produced only on the upper leaf surface within lesions; upon maturity the ostiole breaks through the epidermis (Demaree and Wilcox 1947). One to 5 pycnidia are produced within each lesion. Conidia are hyaline, straight or curved, 5-11 septate, often with a long, attenuated apical segment. The species grows rapidly in culture on cornmeal agar, producing an greenish grey aerial mycelium. Pycnidia of the same type as those formed on leaves develop on the agar surface (Demaree and Wilcox 1947).

In comparing these descriptions with characteristics of the organism found on chokecherry seedlings from Bitterroot Native Growers Nursery, it appears that that fungus should be placed in the genus *Septoria*, at least as the genus is currently described (Sutton 1980). Since specific descriptions of *Septoria* spp. on the genus *Prunus* are unavailable, it is impossible to assign a species epithet to this fungus at the current time. Several pycnidia were formed within discrete lesions and were restricted to the upper leaf surfaces. Ostiolar openings were quite large and masses of conidia accumulated at these openings. Conidia were fairly long, filiform, and usually multi-septate (figure 2). Although pathogenicity tests were not conducted, consistent association of this fungus with leaf lesions indicates that the likely cause of the disease was a *Septoria* species.