Association of Mortality of Recently Planted Seedlings and Established Saplings in Red Pine Plantations with Sphaeropsis Collar Rot

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ABSTRACT

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Established saplings and recently planted seedlings frequently died during 1991 and 1992, respectively, in red pine (*Pinus resinosa*) plantations in Wisconsin. Mortality of seedlings in 12 plantations ranged from 14 to 95%. Mortality of saplings in 16 plantations ranged from 0 to 30%. Symptoms included blackened cortical tissue and dark staining of the underlying xylem in the lower stems and root collars. Pycnidia of *Sphaeropsis sapinea* were observed in these areas on 17 to 97% of dead seedlings. Pycnidia of the pathogen were observed on the lower stems or root collars of 67 to 87% of dead saplings examined in four plantations. These epidemics appear to be related to substantial rainfall deficits in each year, compared to 30-year averages. These observations comprise the first association of Sphaeropsis collar rot with high frequencies of red pine mortality in these situations.

Additional keywords: *Diplodia pinea,* drought, water stress

Among the most common and widely distributed diseases of conifers is shoot blight and canker, caused by *Sphaeropsis sapinea* (Fr.:Fr.) Dyko & Sutton in Sutton (syn. *Diplodia pinea* (Desmaz.) J. Kickx fil.). This fungus is a pathogen of *Abies*, *Cedrus*, *Juniperus*, *Picea*, and *Pseudotsuga* spp., and more than 30 species of *Pinus* (14–16,26,30,32). The disease occurs worldwide and has been associated with significant economic damage in exotic pine plantations in New Zealand, Australia, and South Africa (8,9,13,31,35,36). It also occurs generally in the central and eastern United States, where severe damage has occurred. Affected species include introduced Austrian pine (*Pinus nigra* Arnold) and Scots pine (*P. sylvestris* L.) (24,25), and native red pine (*P. resinosa* Aiton) and jack pine (*P. banksiana* Lamb.) (19,20,22,23).

Collar rot of red pine seedlings in Wisconsin's nurseries was first observed in 1979 (23,27,28). Symptoms that developed on seedlings killed during their third growing season appeared similar to those observed by Crandall (12) in 1938 on red pine seedlings in a Maryland nursery. Shoots and needles initially appeared chlorotic, then desiccated. Shoots were fully expanded and had set terminal bud, and needles were three-fourths elongated. Black stained, resin-soaked wood in the lower stems and root collars was revealed by removal of necrotic bark tissue in which pycnidia of *S. sapinea* were found. However, compared to seedling mortality from shoot blight (which has exceeded 30%), nursery losses from collar rot were very low (less than 1%) (23).

Although a single instance of a similar disease of 5- or 6-year-old eastern white pines (*P. strobus* L.) was reported over 50 years ago (12), Sphaeropsis collar rot has not been described as a problem of red pine after outplanting of seedlings. However, established saplings and recently planted seedlings frequently died during the summers of 1991 and 1992, respectively, in red pine plantations in Wisconsin. Preliminary observations indicated that lower stems and root collars had been colonized by *S. sapinea*. Surveys were undertaken to quantify the frequency of mortality, confirm the association of *S. sapinea* with this damage, and describe the symptoms of collar rot on these trees.

MATERIALS AND METHODS

Recently planted seedlings. Observations were made and seedlings were collected in September and October 1992 in Barron, Dunn, Eau Claire, and St. Croix counties in west central Wisconsin. Twelve sites were selected for survey based on reports of rapid mortality of seedlings and do not represent a random selection. All sites had been planted in April or May of that year with 2- or 3-year-old bare-root stock that had been produced at a single nursery. Soil types included sands and loamy sands characterized as well- or excessively well-drained. The percentage of seedlings that had died was determined by counting and recording the condition of seedlings in transects across each plantation. At least 300 seedlings were counted at each site except one, where seedlings were not counted but mortality was estimated by a cooperator as low.

Dead seedlings (21 to 115 per plantation) were arbitrarily selected for further examination. Seedlings were uprooted and placed in plastic bags, brought to the laboratory, and stored at 4°C. The lower stems and root collars were inspected for the presence of pycnidia using a dissecting microscope, and *S. sapinea* was identified by examining conidia.

Monthly 1992 rainfall records were obtained for nine weather stations in the vicinity of the study area (2). Monthly rainfall (mean for all stations) was calculated for April through September. These means were compared with 30-year historical averages (21), and the percentage surplus or deficit for each month was calculated.

Established saplings. Symptoms were observed during the 1991 growing season, and the mortality that occurred that year was quantified in 16 red pine plantations in November. Plantations were located in Adams and Wood counties in central Wisconsin, from which reports of mortality were received and do not represent a random selection. Soil types included well- or excessively well-drained sands. Time since planting of bare-root seedlings varied from 2 to 8 years. The sources of seedlings usually could not be identified but might have included several different nurseries in Wisconsin or neighboring states. Live and dead trees were counted in circular 0.004-ha plots that were approximately evenly distributed throughout each plantation. Plots varied in number from eight to 32, depending on the plantation size (16 to 56 ha). The relationship between the number of trees per hectare and percent mortality was examined using linear regression.

In April 1992, 15 dead trees in each of four stands, selected on the basis of relatively high 1991 mortality, were examined. These trees were located at intervals of approximately 20 m along a transect across each plantation. A segment approximately

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15 cm long from the end of each of three shoots (the terminal and two laterals) were collected. Two additional segments approximately 15 cm long were removed from each sampled tree, one from the midstem (i.e., at approximately half the sapling height) and the other from the lower stem and root collar area. Samples were individually bagged, brought to the laboratory, and stored at 4°C. These samples were inspected for the presence of pycnidia using a dissecting microscope, and *S. sapinea* was identified by examining conidia.

Monthly 1991 rainfall records were obtained for three weather stations in the vicinity of the study area (1). Monthly rainfall (mean for all three stations) was calculated for April through September. These means were compared with 30-year historical averages for the same months (21), and the percent surplus or deficit for each month was calculated.

RESULTS

Recently planted seedlings. Many dead seedlings appeared to have died early in the growing season, because little or no shoot growth had occurred. Needles of dead seedlings were pale gray-green, orange, or brown, and desiccated. Bark of the lower stems and root collars was darkly discolored and necrotic, but mechanical damage was not apparent. Removal of this bark revealed symptoms of collar rot, including blackened cortical tissues and dark gray staining of the underlying xylem. Neither the cortex nor the sapwood appeared to be resin-soaked.

Mortality in these plantations and the occurrence of *S. sapinea* pycnidia on seedlings varied among sites (Table 1). From 14 to 95% of the seedlings were dead. The pathogen was identified on from 17 to 97% of the dead seedlings. However, *S. sapinea* pycnidia were present on >60% of the dead seedlings from 10 sites and on >80% of the dead seedlings from five sites.

Precipitation records indicated substantial deviations from historical averages (Table 2). Mean rainfall was lower than usual during May, June, and August. In May, the mean deficit was 59%, and at one station, the deficit was 79%. Even though the mean for all sites indicated more than average rainfall during July, deficits of up to 40% occurred at some stations.

Established saplings. Symptoms that occurred during 1991 were first observed in midsummer after shoot elongation. Foliage turned off-color and wilted, with entire crowns often becoming symptomatic and dying within a few weeks. Neither mechanical damage nor external symptoms were apparent on the lower stems or root collars of these trees. Removal of bark from these locations, however, revealed blackened cortical tissues and dark gray staining of the underlying xylem. However, unlike a root disease of loblolly pine (*P. taeda* L.) and slash pine (*P. elliottii* Engelm.) associated with *S. sapinea* in South Africa (34), extensive colonization of roots was not noted. Also, neither cortex nor sapwood appeared to be resin-soaked.

Stocking at the beginning of the growing season (determined in fall as the number of residual live trees plus the number that had died) varied from 741 to 2,443 per ha (approximately 40 to 98% of the desired 1,976 to 2,470 trees per ha). The number of remaining living red pine saplings varied from 516 to 2,443 per ha (26 to 98% of the desired number). Average mortality for all locations during 1991 was approximately 10%, but it varied from 0% to as high as 30% in one plantation. However, there did not appear to be any strong relationship between previous mortality, indicated by low stocking (before the sapling death that was quantified in this survey), and the percentage of mortality during 1991 (R^2 = 12.4, $P = 0.18$).

Pycnidia of *S. sapinea* were observed on the lower stems or root collars of 67 to 87% of dead saplings examined in the four sampled plantations. However, occurrence of pycnidia varied according to the part of the tree examined (Table 3). Pycnidia were observed on the majority of midstem segments from each of the four plantations, but were observed even more frequently on lower stems or root collars. In contrast, *S. sapinea* pycnidia were rarely observed on dead shoots.

Precipitation records indicated periods of relative drought, compared to historical averages (Table 4). The mean rainfall for June through September was approximately 35% below normal, but records of daily precipitation indicated even more severe deficits during portions of that period. For example, following the severe deficit (63%) in June, drought continued with no appreciable rain until July 17. Then, most of the rain in July fell within a

Table 1. Occurrence of *Sphaeropsis sapinea* pycnidia on the lower stems or root collars of recently planted red pine seedlings that died during 1992 in west central Wisconsin

County	Seedling age	Mortality[a] (%)	Pycnidia present[b] (%)
Barron	3	74	17
Barron	3	95	19
Barron	2	14	76
Barron	3	36	92
Barron	3	55	93
Dunn	3	40	74
Dunn	2	low	61
Dunn	2	50	76
Eau Claire	2	50	88
Eau Claire	2	50	97
Eau Claire	2	50	89
St. Croix	2	75	67

[a] The percentages of seedlings that died were determined by counting and recording the condition of at least 300 seedlings in transects across plantations.
[b] Between 21 and 115 dead seedlings from each plantation were examined.

Table 2. Historical average precipitation (mm) and rainfall during spring and summer 1992 in west central Wisconsin where mortality of recently planted red pine seedlings occurred[a]

Month	Historical average	1992 rain	Percent surplus (+) or deficit (–), (range)
April	69	91	+32, (–5 to +76)
May	92	38	–59, (–79 to –33)
June	109	86	–21, (–44 to +11)
July	99	113	+14, (–40 to +37)
August	108	62	–43, (–65 to +10)
September	100	109	+9, (–32 to +43)

[a] Data are averages (and ranges) from nine monitoring stations in the study area.

Table 3. Occurrence of *Sphaeropsis sapinea* pycnidia on established red pine saplings[a] that died during 1991 in four central Wisconsin plantations

Stand	Years since planting	No. with pycnidia/no. examined: Shoots	Midstem	Lower stem or root collar
A	2	0/45	9/15	11/15
B	2	5/45	8/15	10/15
C	7	0/45	9/15	12/15
D	7	0/45	9/15	13/15

[a] Segments approximately 15 cm long of the terminal and two lateral shoots, midstem, and lower stem and root collar of each of 15 trees per plantation were collected and examined in April 1992. Trees were located at intervals of approximately 20 m along a transect across each plantation.

week, and drought resumed in August and September.

DISCUSSION

These observations comprise the first association of Sphaeropsis collar rot with high frequencies of red pine mortality. These epidemics were striking, not just because of the manner of death, but because of the rapidity with which it occurred, and for differences in symptoms from those described previously for red pine. A number of factors for which information was unavailable or was not collected, such as handling of nursery stock, site preparation and planting methods, and other site factors, could have influenced the development of collar rot. However, past descriptions of damage caused by this pathogen on other hosts, and the rainfall deficits during these epidemics, offer explanations for most of what was observed. The uniformity of occurrence of the pathogen on seedlings at some sites and the apparent location of initial infection on both seedlings and saplings also should prompt a reevaluation of the nature of the *S. sapinea*–red pine relationship.

The appearance of the dead seedlings differed somewhat from the previous description of collar rot and its effects on red pine nursery seedlings. Seedlings observed in this survey died with little or no new growth and without noticeable resinosis of the affected tissues. In contrast, Palmer and Nicholls (23) indicated that collar rot in nurseries occurred on seedlings with fully expanded shoots with needles that were three-fourths elongated. They also noted that black-streaked wood at the root collar was resin-soaked. The earlier death and lack of resinosis of these seedlings may reflect an immediate and relatively intense water stress resulting from transplanting and from the deficit in May rainfall.

The rapid mortality of saplings also varied from a previously described progressive deterioration of red pines affected by Sphaeropsis shoot blight and canker. Nicholls and Ostry (19) studied an epidemic in Wisconsin and Minnesota during and following a drought that began in 1976. Red pines deteriorated over time, as indicated by increasing numbers of trees exhibiting scattered dead branches, dieback, and eventual death. Production of resin-soaked stem cankers bounded by callus were common. Symptoms of collar rot and the resulting rapid crown death were not described. Again, their occurrence on saplings examined during this more recent survey, and lack of the host responses (resinosis and callus), might be due to differences in the intensity and duration of water stress to which trees were subjected.

Sources of physiological stress have been associated with damage by *S. sapinea* to many hosts (17,19,20,22,30,32,35). Enhanced invasion of wounded, inoculated, water-stressed stems of Austrian, Scots, and Japanese black pine (*P. thunbergiana* Franco) (3,33), cypress (*Cupressus sempervirens* L.) (17), and Monterey pine (*P. radiata* D. Don) (11) has been demonstrated in pot trials. This effect also has been observed for red pine in studies conducted with both potted seedlings and saplings growing in plantations (5,6).

Water stress has been suggested as a key factor in the development of rapid crown wilt and death resulting from pine stem colonization by *S. sapinea* in other parts of the world. The pathogen caused dieback of major portions or entire crowns of *P. radiata* in New Zealand and Chile, accompanied by characteristic sapwood staining lower in the stems of affected trees (11). The downward growth of *S. sapinea* from the point of establishment was extremely limited, with the extent of crown wilting dependent on how low on the stem initial infection occurred. Restriction of the downward spread of the fungus, but mortality of trees initially infected low on the stem, also was described by Birch (4). If a similar limitation of downward extension of the pathogen occurs in red pines, the observation of staining and pycnidia on the lower stems and root collars would indicate that initial infection occurred there. The relative scarcity of the pathogen on sampled shoots of saplings also suggests that these were not the usual location for initiation of disease in these trees.

The high frequency of occurrence of *S. sapinea* in most collections of dead, recently planted seedlings, and the apparent location of initial infection in lower stems and root collars of seedlings and saplings, indicate the need for additional research on the survival and infection biology of this pathogen. Rapid development of symptoms has been observed after infection by *S. sapinea* (7,9). The pathogen directly penetrates young, nonsuberized stems (10) and enters through stomates of expanding needles (7), in addition to exploiting wounds on woody stems. Symptom development was not followed in seedlings and saplings killed in the epidemics described in this paper, however, and wounds were not apparent in bark of their lower stems and root collars.

The recent confirmation that *S. sapinea* persists on or in the stems of asymptomatic red pine nursery seedlings suggests another possible manner of disease development (29). As described by Mussell (18), latency involves long, intimate contact between pathogen and host, an absence of gross symptoms of disease, and host-induced quiescence of the pathogen. Activity of pathogen and symptom development are proposed to be triggered by changes in host physiology. The manner of persistence of *S. sapinea* and the potential for host condition, including water stress, to stimulate a release from latency and development of Sphaeropsis collar rot are subjects of continuing research.

Table 4. Historical average precipitation (mm) and rainfall during spring and summer 1991 in central Wisconsin where mortality of established red pine saplings occurred[a]

Month	Historical average	1991 rain	Percent surplus (+) or deficit (–), (range)
April	69	111	+61, (+47 to +79)
May	85	136	+60, (+7 to +99)
June	97	36	–63, (–68 to –56)
July[b]	99	100	+1, (–7 to +8)
August	97	49	–48, (–55 to –41)
September	105	74	–30, (–41 to –22)

[a] Data are averages (and ranges) from three monitoring stations in the study area.

[b] Drought in June continued with no appreciable rain until July 17, and then most of the rain in July fell within a week.

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