

ROOT DETERIORATION OF CONTAINERIZED
WESTERN WHITE PINE SEEDLINGS
PLUM CREEK NURSERY, PABLO, MONTANA

R. L. James
Plant Pathologist

Cooperative Forestry and Pest Management
USDA Forest Service
Northern Region
Missoula, Montana

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Western white pine (*Pinus monticola* Dougl.) seedlings grown in containers at the Plum Creek Nursery, Pablo, Montana are removed from their styroblock containers in mid-December for cold storage prior to planting in the spring. Several seedlings of the 1986 crop lacked a well-formed root system at the time of lifting. Affected seedlings lacked many lateral roots; lateral roots present often lacked healthy epidermal tissues. Many affected seedlings also had basal swellings that occurred just below the soil line (figure 1). These swellings appeared as nodules on the main stem. Unfortunately, affected seedlings lacked foliar symptoms indicative of disease, although many were slightly dwarfed.



Figure 1. Western white pine seedling from the Plum Creek Nursery with basal swellings and nodulation. Also, lateral roots were not well developed and often lacked epidermal tissues (arrow).

Growers were concerned that the root deterioration might be caused by pathogenic fungi. Therefore, samples of affected seedlings were transported to the laboratory for analysis. Two sets of isolations were made from white pine seedlings from the "Dorena" seedlot. The first set consisted of eight seedlings, root tips from which were placed on 2 percent water agar and a medium selective for Fusarium spp. (Komada 1975). In this first isolation set, none of the root tips were surface sterilized. Results from this first set of isolations yielded extensive bacterial contamination on water agar plates and widespread Trichoderma colonization on Komada plates. However, on the latter media, there was one isolation of Fusarium oxysporum and several yielding a species of Cylindrocarpon.

A second set of isolations was conducted to quantify occurrence of associated fungi from different locations on affected seedlings. Root systems of the seven seedlings selected for these isolations were photographed and divided into three portions: (1) fine root tips with attached epidermal tissues (if present), (2) stele tissues from lateral roots which lacked an epidermis, and (3) tissue from the main stem just below the ground line where swellings were evident. All root tissues were surface sterilized in 10 percent aqueous sodium hypochlorite (bleach) for 30 seconds and rinsed in distilled water prior to placing on agar media. Two types of selective agar media were used for isolations: Komada's medium (for Fusarium) and a medium for isolating species of Pythium (V-8 juice agar amended with pimaricin, rifamycin, amoxicillin, and PCNB). Komada plates were incubated under cool fluorescent light at about 22°C for 7 days; Pythium plates were incubated in the dark for 3 days.

Isolation results are summarized in table 1. An analysis of variance revealed that there were significant differences ($P=0.01$) in colonization of root tips, stele, and basal swellings among the various organisms isolated.

Cylindrocarpon sp. was most commonly associated with root tips and basal swellings. Pythium spp. and Fusarium oxysporum were isolated much less frequently from both tissues. Root stele tissues were often not colonized by any organisms. Cylindrocarpon was consistently isolated from root tip and basal swelling tissues, often at the exclusion of other organisms (figure 2). On media selective for Pythium, Cylindrocarpon was also isolated frequently, sometimes in association with Pythium (figure 3). Statistical tests (Tukey's) revealed that isolations of Cylindrocarpon sp. from root tips and basal swellings were significantly greater ($P=0.05$) than isolations of other organisms. Therefore, it is concluded that this organism is likely the major cause of white pine seedling root deterioration.

Table 1.--Occurrence of microorganisms on roots and basal stem tissues of western white pine seedlings at the Plum Creek Nursery, Pablo, Montana.

Organisms	Percentage colonization ¹		
	Root tips	Stele	Basal stem
<u>Cylindrocarpon</u>	69.1 A	50.0 B	97.1 A
<u>Pythium</u>	9.1 BC	0 B	1.4 BC
<u>Fusarium</u>	3.6 BC	0 B	0 C
<u>Trichoderma</u>	18.2 BC	16.0 B	1.4 BC
<u>Penicillium</u>	7.3 BC	13.3 B	0 C
Bacteria	29.1 B	1.0 B	10.0 B
None	0 C	63.3 A	0 C

¹ Means followed by the same capital letter are not significantly different (P=0.05) using Tukey's comparison test. Percentages underwent arc-sin transformations for statistical analysis.

Close examination of the isolate of Cylindrocarpon frequently obtained from these seedlings revealed characteristics similar to those for C. didymum (Hartig) Wollenw. described by Booth (1966). Colonies were at first white to peach colored, but became beige to purple-brown with age (figure 4). Conidia were oval to cylindrical with rounded ends, usually fairly straight or slightly curved, with 0 or 1 septations. Conidiophores were long and slender, sometimes branched, with cylindrical terminal phialides. Cultures also contained terminal or intercalary chlamydospores which were at first hyaline but became brown with age. This species has been described as occurring in the rhizosphere of several plants (Andrews and Clouston 1937; Kurbis 1937) and as a pathogen of pine seedlings (Houten 1939). However, it has not been described previously as a pathogen of conifer seedlings in the northern Rocky Mountains, although another species of Cylindrocarpon (C. tenue Bugn.) has been associated with diseases of containerized Engelmann spruce seedlings in the region (James and Gilligan 1985). In order to verify the role of C. didymum as a pathogen of conifer seedlings, pathogenicity tests will be required. However, at the present time it appears that this organism is largely responsible for the root deterioration disease complex of white pine seedlings at the Plum Creek Nursery.

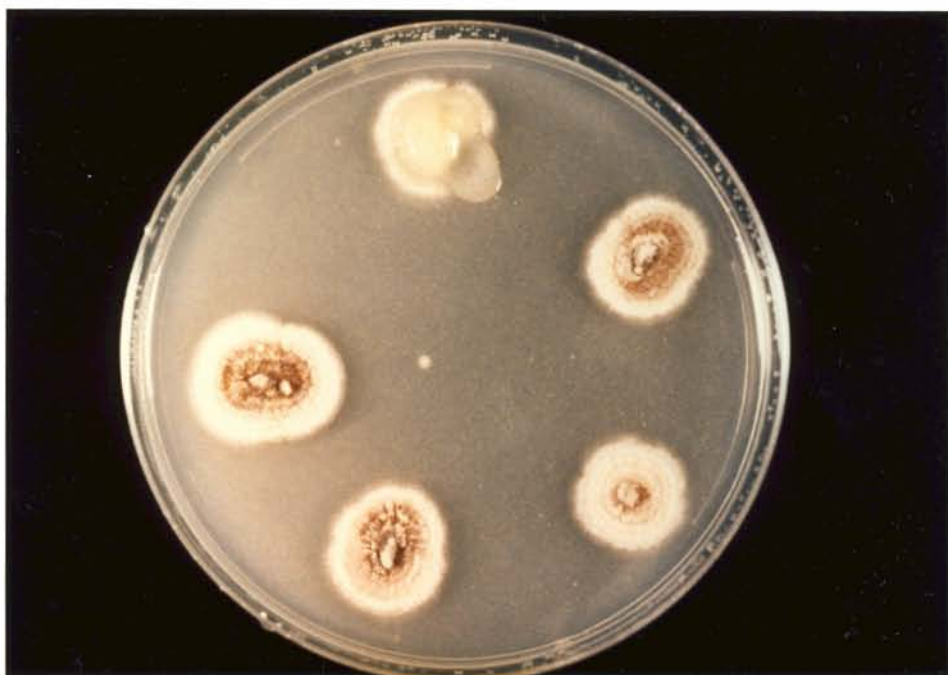


Figure 2. Isolation of Cylindrocarpon sp. on Komada's medium after 7 days' incubation. All colonies are Cylindrocarpon except the top one which is contaminated with bacteria.

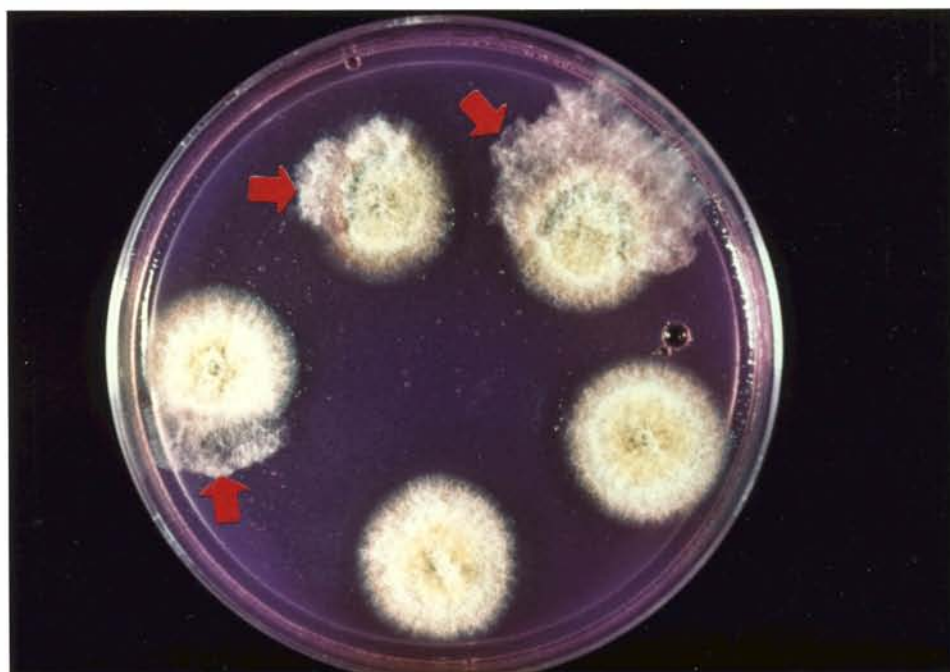


Figure 3. Isolation of Cylindrocarpon sp. and Pythium sp. on selective Pythium medium. All colonies contain Cylindrocarpon; the three with fluffy white mycelia (arrows) also contain Pythium.



Figure 4. Fourteen-day-old culture of C. didymum grown on potato dextrose agar. Colony was at first white but turned beige to purple-brown with age.

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