NEEDLETIP DIEBACK OF CONTAINERIZED DOUGLAS-FIR SEEDLINGS AT THE USDA FOREST SERIVCE NURSERY, COEUR D'ALENE, IDAHO: ASSOCIATED ROOT FUNGI

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February 1986

Nursey Discuse Notes No. 27

ABSTRACT

Isolations were made from the root systems and soil mixes of three containerized Douglas-fir seedlings displaying typical needletip dieback symptoms often associated with infection by <u>Fusarium</u>. <u>Fusarium oxysporum</u> was recovered from roots and both organic and mineral components of soil mixes of two of the three seedlings. The fungus was most abundant on root tips. <u>Trichoderma</u> and <u>Penicillium</u> were common colonizers of the soil mix and roots of the one seedling without <u>Fusarium</u>.

INTRODUCTION

Needletip dieback of containerized Douglas-fir (<u>Pseudotsuga menziesii</u> (Mirb.) Franco) seedlings has been associated with root infection by species of <u>Fusarium</u> (James 1984a; James 1984b). However, quantification of root infection as related to foliage symptom expression remains to be elucidated. Also, importance of the mineral and organic components of soil mixes in harboring <u>Fusarium</u> inoculum is unknown. Therefore, investigations were undertaken to determine locations of <u>Fusarium</u> infections on roots and extent of soil mix colonization for seedlings displaying typical needletip dieback symptoms.

METHODS

Three seedlings with typical needletip dieback symptoms (figures 1-3) were randomly selected from a greenhouse at the USDA Forest Service Nursery, Coeur d'Alene, Idaho. Seedlings were carefully removed from their containers and their root systems with attached soil particles were placed in beakers of distilled water. Roots were agitated to remove loose soil particles. The soil-water solution was filtered through two layers of cheesecloth to remove large organic particles. The organic component was resuspended in distilled water; samples of this component were randomly selected and placed on a selective medium for <u>Fusarium</u> spp. (Komada 1975). Portions of the mineral component of the soil mix which had passed through the cheesecloth were also placed on the selective medium. Amounts of soil placed on the selective media varied among the samples.

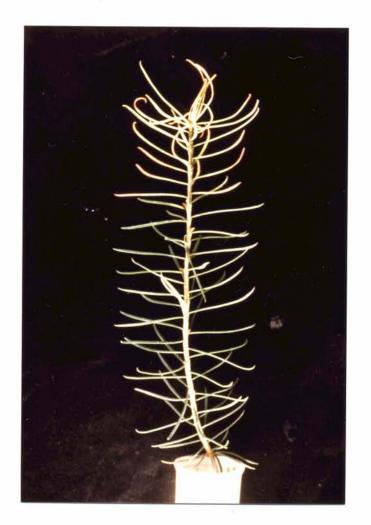


Figure 1.--Containerized Douglas-fir seedling (no. 1) with needletip dieback symptoms from the USDA Forest Service Nursery, Coeur d'Alene, Idaho.



Figure 2.--Containerized Douglas-fir seedling (no. 2) with needletip dieback symptoms from the USDA Forest Service Nursery, Coeur d'Alene, Idaho.



Figure 3.--Containerized Douglas-fir seedling (no. 3) with needletip dieback symptoms from the USDA Forest Service Nursery, Coeur d'Alene, Idaho.

Root systems of each seedling were washed thoroughly under tap water to remove loose soil particles. Fifteen lateral roots were randomly selected and cut from the main taproot on one side of the root system. Root tips about 0.5 cm in length were cut from selected roots and aseptically placed on the selective medium. Also, a piece from the top of each selected root (where it joined the main taproot) was placed on the selective medium. The other side of each root system was surface sterilized in a solution of 4.0 percent bleach (0.21 percent aqueous sodium hypochlorite) for 4 minutes and washed thoroughly to remove excess bleach.

Roots were selected and samples taken as described for the nonsterilized portion.

All plates were incubated for 7 days at about 24° C under a 12-hour diurnal light-darkness regime. The number of root or organic soil mix component pieces infected with <u>Fusarium</u>, <u>Trichoderma</u>, and <u>Penicillium</u> were counted to determine percentage infection by each fungus. Colonization of the mineral soil mix component by each fungus was classified as extensive (E), moderate (M), slight (S), or absent (A).

RESULTS AND DISCUSSION

Fusarium oxysporum Schlect. was isolated from the roots and soil mix of seedlings 2 and 3, but not from no. 1 (table 1). No other <u>Fusarium</u> species was isolated. <u>Fusarium oxysporum</u> colonized both the organic and mineral components of soil mixes; the fungus was more common on root tips than farther up roots (where they join the main taproot). Differences in recovery of the fungus from sterilized and nonsterilized roots were not apparent. This fungus is a common colonizer of conifer seedling roots (James 1985); however, its ability to incite disease varies. <u>Fusarium oxysporum</u> is readily found on healthy plants (Armstrong and Armstrong 1948; Bloomberg 1966) and conditions required for the fungus to become pathogenic on conifer seedlings are largely unknown. Factors of host stress, such as withholding water and nutrients during hardening off and bud set, may be important in container operations (James 1985).

Table 1.--Occurrence of <u>Fusarium oxysporum</u> on roots and within the soil mix of containerized Douglas-fir seedlings at the USDA Forest Service Nursery, Coeur d'Alene, Idaho.

	Soil mix					
	Organic component	Mineral ² component	<u>Nonsteriliz</u> Tips	ed roots Joints	<u>Sterili</u> Tips	zed roots Joints
Seedling 1	0	Â	0	0	0	0
Seedling 2	100	E	100	100	100	92
Seedling 3	69	E	100	14.	100	21

¹Figures in table are percent of root or organic pieces colonized by <u>F. oxysporum</u>.

³Joints - where sampled root met main taproot.

Two examples of common saprophytic colonizers of conifer roots, <u>Trichoderma</u> and <u>Pénicillium</u>, were found on the roots of each seedling (table 2). Colonization was greatest on seedling no. 1, on which <u>Fusarium</u> was not recovered. Since <u>Trichoderma</u> spp. are common antagonists and often mycoparasites of pathogenic fungi (Bell et al. 1982), it is possible that <u>Fusarium</u> was initially responsible for tip dieback symptoms of seedling no. 1, but later replaced by <u>Trichoderma</u>. Where <u>Fusarium</u> was in great abundance; e.g., root tips of seedlings 2 and 3, <u>Trichoderma</u> and <u>Penicillium</u> were not recovered.

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Table 2.--Occurrence of Trichoderma and/or Penicillium spp. on roots and within the soil mix of containerized Douglas-fir seedlings at the USDA Forest Service Nursery, Coeur d'Alene, Idaho.

	Soil mix				124	10. <u>-1</u>
	Organic component	Mineral ² component	<u>Nonsteri</u> Tips	lized roots Joints	<u>Sterili</u> Tips	zed roots Joints
Seedling 1	100	A	100	100	100	100
Seedling 2	6	S	0	0	0	8
Seedling 3	62	м	0	86	0	79

¹Figures in table are percent of root or organic pieces colonized by F. oxysporum.

²Level of colonization: E = extensive, M = moderate, S = slight, A = absent.

³Joints - where sampled root met main taproot.

In conclusion, it appears that F. oxysporum was probably responsible for causing needletip dieback symptoms in the containerized Douglas-fir seedlings sampled. By the time symptoms were evident, the fungus had already colonized much of the root system and adjacent soil mix within the containers.

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