ROOT DISEASE OF ENGELMANN SPRUCE SEEDLINGS UNIVERSITY OF MONTANA, MISSOULA

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Nursery Disease Notes #98 January 1990 During the spring of 1989, bareroot 2-O Engelmann spruce (**Picea engelmanni** Parry) seedlings from the USDA Forest Service Lucky Peak Nursery (Boise, Idaho) were being used by Linda Gillilan of the University of Montana for research purposes. These seedlings, which were lifted from the nursery in the early spring, were from a high elevation seed source. About 1000 seedlings were potted during early May into new milk carton containers using commercially prepared peat-vermiculite growing media commonly used for production of conifer seedlings (W. R. Grace and Co.). Shortly after potting, many seedlings began to display root disease symptoms, i. e., chlorosis and necrosis of foliage followed by general overall wilting of the crown. Symptoms developed progressively in most seedlings, and after about two months, nearly 80% of the potted seedlings had died.

Roots of most diseased seedlings were completely necrotic with epidermal tissues becoming easily detached. There was no evidence of actively growing root tips on most seedlings. Cambial necrosis causing girdling of the entire stem was common at the root collar. Necrosis began in the roots and progressed into the root collar region.

Two sets of seedlings in various stages of decline were analyzed for association of potentially pathogenic fungi with diseased roots. Root systems of selected seedlings were washed thoroughly under running tap water to remove adhering particles of growing media. Pieces of root about 3-5 mm in length were aseptically severed from root systems. Pieces were collected from over the entire root system, i. e., intercalary pieces as well as root tips. Root pieces were surface sterilized in a 10% bleach solution (0.525% aqueous sodium hypochlorite) for 1 minute and rinsed with sterile distilled water. In the first isolation set, 10 root pieces from each of 5 seedlings were placed on an agar medium selective for **Fusarium** spp. and related root disease fungi (Komada 1975). In the second isolation set, 20 root pieces from each of 7 seedlings were sampled. Ten were placed on Komada's medium and 10 placed on a selective medium used for isolating **Pythium** spp. and other water mold fungi (V-8 juice agar amended with pimaricin). All plates were incubated at about 25°C. Those with V-8 juice agar were incubated in the dark for 3 days, whereas those with Komada's medium were incubated under diurnal cycles of cool fluorescent light for 7-10 days. Fungi emerging from root pieces were examined under the microscope (200-450X) and representative examples transferred to potato dextrose and carnation leaf agars to facilitate identification (Domsch and others 1980; Nelson and others 1983).

Roots of all sampled seedlings were colonized with Fusarium oxysporum Schlect. table 1). Likewise, all seedlings assayed (set 2) had roots thoroughly colonized with Pythium spp. The two most commonly encountered Pythium species were: P. irregulare Busiman and P. irregulare Trow. Other potential root pathogens isolated from seedling roots included F. sambucinum Fuckel, F. acuminatum Ell. & Ev., and Cylindrocarpon didymum (Hartig)Wollenw. Fusarium sambucinum was isolated at relatively high levels.

Fusarium oxysporum is a common root pathogen of conifer seedlings (Bloomberg 1971), including Engelmann spruce (James and Gilligan 1985). It may often be an aggressive pathogen, causing the type of disease symptoms seen on these spruce seedlings (James and others 1989). Pythium spp. are also potential

Table 1. Color	nization of Engelmar	in spruce seedling	roots with	selected fungi1
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Seedling No.

Percentage Colonization by Selected Fungi²

Set 1	FOXY	FSAM	FACU	CYL	TRI	PYTH
1	90	0	0	20	30	-
2 3	70	10	30	0	0	
	70	40	0	0	0	1 87
4	90	20	0	0	0	
5	60	0	10	0	10	1. /
Average	76	14	8	4	8	
Set 2						
1	80	80	0	0	20	80
2	100	80	0	0	0	90
3	100	70	0	0	10	80
4	100	0	10	0	0	80
2 3 4 5 6	100	0	20	0 0 0	0	90
	100	40	0	0	20	80
7	70	60	0	0	0	90
Average	93	51	4	0	7	84
Overall Average	86	36	6	2	8	84

¹ Ten root pieces from each seedling were assayed on Komada's medium (set 1). For set 2, ten root pieces were assayed on each of two media: Komada's and V-8 juice agar amended with pimaricin.

³ Pythium spp. detected on V-8 juice agar amended with pimaricin for set 2 only. Species included primarily P. irregulare and P. ultimum.

pathogens of spruce seedlings (Anonymous 1960; Grand 1985), which are most important in poorly drained portions of seedbeds where higher moisture levels may persist (James 1982).

² FOXY = Fusarium oxysporum; FSAM = Fusarium sambucinum; FACU = Fusarium acuminatum; Cyl = Cylindrocarpon didymum; TRI = Trichoderma spp.; PYTH = Pythium spp.

Fusarium sambucinum and **F. acuminatum** may have both been involved in spruce root disease. Previous investigations (James and Gilligan 1984; James and others 1988, 1989) have shown that **F. sambucinum** is usually not very aggressive on conifer seedlings. However, certain strains of **F. acuminatum** may be quite pathogenic (James and others 1988, 1989). It is also possible that both these species were secondary colonizers of roots previously attacked by **F. oxysporum** or **Pythium** spp.

Cylindrocarpon didymum was isolated from only one spruce seedling (table 1). This species has previously been found associated with conifer seedling diseases (James 1988), but pathogenicity tests to evaluate its potential role in etiology of diseases have not been done.

Levels of **Trichoderma** spp., common saprophytic competitors or antagonists of pathogens (Papavizas 1985), were quite low on sampled roots (table 1). These fungi often occupy the same niches within roots that pathogens such as **Fusarium** colonize. When high levels of **Fusarium** are detected on roots, correspondingly lower levels of **Trichoderma** are often found (James and others 1987).

In conclusion, results from this evaluation indicate that Engelmann spruce seedling mortality at the University of Montana was most likely due to root disease caused by **F. oxysporum** and **Pythlum** spp. Both groups of pathogens may have been involved and it is possible that they acted synergistically to cause rapid disease shortly after seedlings were potted. It is possible that most pathogenic inoculum was carried on stock from the nursery because of the rapidity with which disease symptoms developed. However, **F. oxysporum** has previously been detected at high levels on conifer seedlings grown in greenhouses at the University of Montana (James 1987). Therefore, high resident populations of this fungus may be common within these greenhouses.

LITERATURE CITED

- Anonymous. 1960. Index of plant diseases in the United States. USDA Handbook No. 165. Washington, D. C. 531p.
- Bloomberg, W. J. 1971. Diseases of Douglas-fir seedlings caused by **Fusarium oxysporum**. Phytopathology 61:467-470.

Domsch, K. H., W. Gams and T. H.-Anderson. 1980. Compendium of soil fungi. Academic Press, London. 859p.

Gerlach, W. and H. Nirenberg. 1982. The genus Fusarium - a pictorial atlas. Paul Parey, Berlin. 406 p.

- Grand, L. F. 1985. North Carolina plant disease index. North Carolina Agr. Res. Serv. Tech Bull. 240. 157p.
- James, R. L. 1982. Pythium root disease of Douglas-fir and grand fir seedlings at the Coeur d'Alene Nursery, Idaho. USDA Forest Service, Northern Region. Rept. 82-10. 10p.
- James, R. L. 1987. Fusarium oxysporum on roots of containerized western larch seedlings, University of Montana. USDA Forest Service, Northern Region. Nursery Dis. Notes #46. 3p.

- James, R. L. 1988. Diseases of conifer seedlings associated with **CylIndrocarpon** species: a review. USDA Forest Service, Northern Region. Nursery Dis. Notes # 76. 14p.
- James, R. L., R. K. Dumroese, C. J. Gilligan and D. L. Wenny. 1989. Pathogenicity of Fusarium isolates from Douglas-fir seed and container-grown seedlings. Idaho Forest, Wildlife and Range Exp. Sta., Bull. No. 52. 10p.
- James, R. L., R. K. Dumroese and D. L. Wenny. 1988. Fusarium diseases of containerized conifer seedlings in northern Rocky Mountain nurseries: infection, symptom production and pathogenicity of associated fusaria. Phytopathology 78(12):1533.
- James, R. L., R. K. Dumroese, D. L. Wenny, J. F. Myers and C. J. Gilligan. 1987. Epidemiology of *Fusarium* on containerized Douglas-fir seedlings. 1. Seed and seedling infection, symptom production, and disease progression. USDA Forest Service, Northern Region. Rept. 87-13. 22p.
- James, R. L. and C. J. Gilligan. 1984. Studies of *Fusarium* associated with containerized conifer seedling diseases: pathogenicity tests of isolates from the Alpine Nursery, Kalispell, Montana. USDA Forest Service, Northern Region. Rept. 84-14. 29p.
- James, R. L. and C. J. Gilligan. 1985. Containerized Engelmann spruce seedling disease at the USDA Forest Service Nursery, Coeur d'Alene, Idaho. USDA Forest Service, Northern Region. Rept. 85-17. 15p.
- Komada, H. 1975. Development of a selective medium for quantitative isolation of *Fusarium oxyspc.rum* from natural soil. Rev. Plant Prot. Res. 8:114-125.
- Nelson, P. E., T. A. Toussoun and W. F. O. Marasas. 1983. *Fusarium* species: an illustrated manual for identification. The Pennsylvania State University Press, University Park. 193p.
- Papavizas, G. C. 1985. Trichoderma and Gliocladium: biology, ecology, and potential for biocontrol. Ann. Rev. Phytopathol. 23:23-54.